Studies on the Aetiology and Prophylaxis of Hypomagnesaemia in Ruminant Animals.

A Thesis submitted to the University of Glasgow
for the Degree of Doctor of Philosophy by N. S. Ritchie, B.Sc.,
1 9 6 6.

Summary.

The history and occurrence of hypomagnesaemia and hypomagnesaemic tetany in ruminant animals has been reviewed with reference to literature reports of previous investigations.

A study was made over three consecutive spring seasons on the effect of potassium fertilisation of pasture on the development of hypomagnesaemia in lacta ewes. No effect was found in the first two years but where the potassium fertiliser was applied for the third successive year, significant depressions in the mean plasma magnesium concentration of ewes grazing thereon were found.

Studies were also made into the effects of age, breed and number of suckled lambs on the development of hypomagnesaemia in sheep. Age was found to have little influence. The suckling of either single or twin lambs was found to have no effect in one experiment but in a second trial, a greater incidence of hypomagnesaemia was demonstrated in ewes with twin lambs. Differences were found between two breeds of sheep in their relative susceptibili to hypomagnesaemia. Highly significant correlations were demonstrated between the plasma magnesium concentrations of individual ewes in consecutive years.

By collation of existing literature reports, it was demonstrated that

80 - 90% of reported clinical cases in cattle and sheep had a concomitant hypomagnesaemia and hypocalcaemia. The hypocalcaemia was shown to develop rapidly over the 24 hours preceding the appearance of clinical signs.

An experiment is described where an attempt was made to reproduce this situation of hypomagnesaemia in combination with hypocalcaemia. No clinical cases were recorded.

Studies were made with sheep to assess the value of the existing prophylactic measures which are recommended for hypomagnesaemic tetany. The daily administration of magnesium salts, containing 4 g. magnesium, to sheep was found to produce only a small temporary response in the plasma magnesium concentration and this response was not maintained over a 24 hour period. A pasture application of 5 cwts of calcined magnesite per acre produced significant increases in the plasma magnesium concentration of grazing sheep. There was no response from the use of only 0.5 cwts/acre, when applied as a fertiliser in the winter, but this quantity, when applied as a pasture dust, increased the plasma magnesium concentration of sheep grazing thereon.

As a possible new method of prophylaxis, a rumen pellet, containing a metal magnesium/aluminium alloy, was developed. This pellet was shown to corrode in the rumeno-reticular sac at a regular rate over a period of six to eight weeks. Seven trials were carried out on six farms to test the efficiency of a treatment of magnesium rumen pellets as a prophylactic measure for hypomagnesaemic tetany in cattle, sheep and calves. In a total of 181 animals given this treatment, there were no clinical cases of tetany, whereas

in a similar number of comparable control animals, there were six cases of hypomagnesaemic tetany. A response to treatment in the mean plasma magnesium levels was also demonstrated in five of the seven trials.

STUDIES ON THE AETIOLOGY AND PROPHYLAXIS OF HYPOMAGNESAEMIA IN RUMINANT ANIMALS.

A thesis submitted to University of Glasgow

for the degree of

DOCTOR OF PHILOSOPHY

in the Faculty of Medicine.

by

N.S. RITCHIE B.Sc.

Dept. of Animal Husbandry &

Veterinary Preventive Medicine,

Glasgow University Veterinary

Hospital.

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CONTENTS

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I also gratefully acknowledge the provision by Pfizers Ltd. of a research grant to cover the costs of further developmental work with magnesium rumen pellets in 1965, as described in the latter part of Section IV. Magnesium Elektron Ltd. are also to be thanked for their manufacture of the magnesium rumen pellets used in the field trials.

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Preface.

The experimental work of this thesis is largely concerned with studies on hypomagnesaemic tetany in sheep. The choice of this species as the experimental animal for especial study was based on several reasons. Firstly, although considerable attention has been placed on the problem of hypomagnesaemic tetany in cattle in the past thirty years, comparatively little work has been carried out on sheep. Results from cattle experiments have been assumed to apply equally well to sheep. Part of this present work has been to investigate to what extent factors known to be involved in the development of hypomagnesaemic tetany in cattle are implicated in the same problem in sheep. Furthermore, sheep in many ways form a better subject for study in that being less costly to purchase, house and feed, larger numbers can be employed in experiments. This, therefore, permits superior experimental design and better statistical treatment of the results, thereby reducing the risk of fortuitous inaccuracies leading to wrong conclusions. A group of sheep can also be regarded as relatively uniform in that a flock can be obtained which is of a similar age, breed and size and in the Spring at the same stage of lactation. This can be contrasted with a herd of cattle which normally consists of cows of differing ages at various stages of lactation and often of dissimilar size, age and occasionally breed. Less variation can thus be expected in the response of a group of sheep to a particular treatment than would be found in cattle.

In view of the great attention which has been paid to the subject of hypomagnesaemia over the last thirty years, it would require more space than is available here to review exhaustively all the work reported on magnesium nutrition

and deficiency in farm animals. It has been rather the author's intention in this thesis to give as comprehensive a review as possible as an introduction with particular reference to the main investigations carried out on each point. There have been several excellent reviews on this subject in recent years, e.g. Blaxter & McGill (1956); Wilson (1960); Rook & Storry (1962); and Wilson (1964).

Hypomagnesaemic tetany has been known under a variety of names such as "grass staggers", "grass tetany", "lactation tetany", "Herefords disease", and "nutritional tetany". However, it was felt that none of these names was sufficiently precise and the name hypomagnesaemic tetany has therefore been used throughout this work.

S E C T I O N I

(a) A review of the literature concerning the magnesium nutrition of ruminant animals and in particular, of past work pertaining to hypomagnesaemia and hypomagnesaemic tetany.

(a) Historic background.

(b) Incidence of hypomagnesaemic tetany.

(c) Clinical and biochemical signs of hypomagnesaemic tetany.

(d) General biochemistry of magnesium nutrition.

(e) Magnesium absorption, excretion and Homeostasis.

(f) Magnesium requirements in ruminant animals.

(g) Magnesium in foods and pasture.

(h) The aetiology of hypomagnesaemic tetany.

(b) Chemical Methods.

Hypomagnesaemic tetany can be described briefly as a condition of ruminant animals where the animal collapses in tetanic convulsions and where death frequently ensues. The only biochemical abnormality which has consistently been found is the presence in the blood serum of an abnormally low concentration of magnesium. It can be considered to be mainly a problem of the grazing animal, although it is also found under certain circumstances in indoor fed animals.

Hypomagnesaemic tetany was initially recognised and reported as merely the clinical entity of "grass staggers" in the 1920's, since the connection between this clinical condition and the low serum magnesium was not discovered until later. One of the first mentions of "staggers" is made by Calderwood (1925) in connection with the differential diagnosis between it and acute lead poisoning. His description of the symptoms suggest that he was dealing with true hypomagnesaemic tetany, but at the time he ascribed the cause to an intestinal toxaemia. Begg (1939) in a reported discussion on a paper by Green (1939) recalls that in 1917, he attended many cases of what was undoubtedly hypomagnesaemic tetany in cattle which had been turned out suddenly onto young Spring pasture, resulting from a rapid improvement in the weather. These first reports related purely to dairy cattle and reports of a similar disorder in beef cattle and in sheep were not made until later.

Sjollema (1928, 1930 a.b.) and Sjollema & Seekles (1929) were responsible for establishing the relationship between hypomagnesaemic tetany and blood serum magnesium levels. In a series of papers they described the occurrence, nature and treatment of the condition. Even at this early stage, these authors

suggested that the increasing number of cases which were being recorded in Holland seemed to be correlated with the intensification of farming methods, and in particular with heavy fertiliser treatments. From analyses of blood samples from fifty-five cows suffering from hypomagnesaemic tetany, they showed that the mean serum magnesium level was only 0.45 mg Mg/100 ml as compared with 1.6 - 2.0 mg Mg/100 ml for normal animals. Thus the basic biochemical change was established.

In Great Britain, Dryerre was working independently on the same problem and in 1932 (Dryerre 1932) published findings which were mainly in agreement with Sjollema & Seekles, i.e. that "lactation tetany" (as it was sometimes referred to in Great Britain at that time) was due to a magnesium deficiency in the blood serum. In forty-two cases of lactation tetany from different parts of the country, the average magnesium was 0.39 mg Mg/100 ml blood serum. A point of difference, which will be considered in more detail later, that existed between the work of Sjollema and Dryerre, was that the Dutch workers found a high degree of concomitant hypocalcaemia, whereas Dryerre found in the majority of cases, an uncomplicated hypomagnesaemia. Discussion on Dryerre's paper indicated that the incidence of this "new" condition seemed to be increasing. These initial publications were followed by others (e.g. Blakemore & Stewart, 1933; Allcroft & Green, 1934; Adamson, 1936; Powell, 1937; Allcroft & Green, 1938) reporting the occurrence of hypomagnesaemic tetany in various areas. These reports suggested an increasing incidence, although this "increase" would be in part due to a new awareness on the part of veterinary practitioners to the presence of this disorder. Early reports of hypomagnesaemic tetany occurring in other countries were made by

Hopkirk, Marshall & Blake (1933) in New Zealand and by Metzger (1936) from America. In general, all these early papers described cases of hypomagnesaemic tetany occurring in lactating dairy cows when turned out from indoor winter feeding onto Spring pasture, with most cases appearing within the first two to four weeks of grazing.

Beef Cattle.

Although the earlier workers with cattle were mainly concerned with the disorder in dairy cows at Spring pasture, it was soon recognised that the same condition could affect dairy cows at other seasons (though not so markedly) and also that beef cows were susceptible to the disorder. With beef cows, as first noted by Lothian (1931), cases are more liable to occur in the autumn, winter or early spring, rather than the late spring when the flush of grass appears. This was confirmed by Allcroft & Green (1938) who demonstrated that Hereford breeding cows had characteristically low blood magnesium during the winter months with the lowest levels in December and January. This seasonal variation in serum magnesium levels, quite apart from hypomagnesaemic tetany, in herds of cattle kept outside, was emphasised by Allcroft (1947 c) where, over a four year period he demonstrated the correlation between this variation and temperature and weather conditions. Later work (Barker, 1948; Inglis et al., 1954) again confirmed the existence of seasonal hypomagnesaemia. This variation was found to be reflected in the appearance of clinical cases of tetany coinciding with the periods when the serum magnesium was at its lowest (Allcroft 1947 c). However as pointed out earlier by Allcroft & Green (1938) the presence of a low serum magnesium concentration does

not necessarily mean that clinical tetany will ensue. They describe levels as low as 0.50 mg/100 ml in December with no hypomagnesaemic tetany occurring.

Sheep.

The earliest recorded case of hypomagnesaemic tetany in sheep is often quoted to be that of Blumer et al (1939) but although this paper refers to "grass staggers", the author is actually discussing a hypermagnesaemia and hypocalcaemia, which is therefore more truly parturient paresis, and certainly not hypomagnesaemic tetany. Thus the earliest record of hypomagnesaemic tetany in sheep does not seem to be until 1951, when Stewart in the Annual Report of the Animal Diseases Research Association states, "In 1951, low magnesium tetany appeared suddenly in sheep. Up till that year we had never come across this condition in sheep. It occurred within one month after lambing..... Analysis showed low blood magnesium".

An interesting remark however is to be found in a much earlier paper by Greig (1929) on the nature of lambing sickness where he mentions the occurrence of cases of "delayed lambing sickness in ewes with lambs at foot or in advanced lactation, especially if grazed on strong pasture". From the timing and the circumstances of occurrence, this may have been hypomagnesaemia and not lambing sickness as suggested by the author.

A short mention of hypomagnesaemic tetany in sheep is made by Green (1939), although no details are given.

Barrentine & Morrison (1953) reported outbreaks of grass tetany in sheep grazing oat forage, occurring over the previous five years. This marks the appearance of hypomagnesaemic tetany in America in 1948.

These reports were followed in 1955 by further observations (O'Moore, 1955; Pook, 1955; Penny & Arnold, 1955;) of outbreaks of hypomagnesaemic tetany in flocks of ewes with lambs at foot. In all cases, diagnosis was confirmed by analysis which showed the serum magnesium to be low. An interesting secondary point to be found in two of these references is that there was a concomitant hypocalcaemia in the clinically affected ewes. This point will be dealt with in more detail later. The same observation is made by Allcroft (1956) where it is stated in reference to sheep that "there is usually a concomitant hypocalcaemia". Newsom (1958) does not mention hypomagnesaemia in the second edition of his standard book "Sheep diseases", although in the section dealing with hypocalcaemia, the description of symptoms and the conditions under which it occurs are rather more those which are typical of hypomagnesaemic tetany, and there seems to have been, even then, some confusion between these two disorders. Further reports of outbreaks of hypomagnesaemic tetany in sheep have been given more recently by Herd & Peebles (1962) and L'Estrange & Axford (1964 a).

Calves.

Duncan, Huffman & Robinson (1935) first showed that the tetany to which calves succumbed when fed for long periods on a ration entirely of milk was hypomagnesaemic in origin. Outbreaks of tetany in calves had in fact been investigated and reported by Sjollema (1935) but although he found serum magnesium levels below the normal range, he concluded that there was no connection between the tetany and the hypomagnesaemia. Blaxter & Sharman (1955) emphasised the extent to which this was a problem in natural calf rearing. They gave an incidence of 5 per cent of clinical cases in suckled calves on beef rearing farms in Invernesshire.

As found with adult ruminants, the development of clinical cases was in every case accompanied by low serum magnesium values. The converse was not necessarily true in that there were various reports (e.g. Parr & Allcroft, 1953; Parr, 1957; Peers & Armour, 1958) quoting hypomagnesaemia in milk fed calves with no development of tetany, or alternatively the presence of hypomagnesaemia for long periods before clinical tetany developed.

Incidence of hypomagnesaemic tetany in adult Ruminants.

In general, hypomagnesaemic tetany seems to be a problem found only in areas of temperate climate with good rainfall, conducive to the production of rich pasture grazing. The regions affected, as far as is known at the present time, are therefore the Northern European countries including Britain, France, Holland, Germany and the Scandinavian countries, New Zealand, Australia and parts of North America.

Incidence from year to year.

It is well known, as was first pointed out by Green (1939) and later amplified by Allcroft (1947 a), that the incidence of this metabolic disorder varies from year to year, there being some years when very few cases occur and others when there is a sudden spate of outbreaks. This variation almost certainly must be due to climate differences from year to year influencing either the animal itself or alternatively the pasture on which the animal is subsisting either as fresh grass or, more uncommonly, in a conserved form. The precise factors believed to be involved in this will be dealt with in more detail later.

Incidence as affected by season.

Superimposed upon this yearly variation, it is found, that for a given year there is a seasonal variation in the incidence of hypomagnesaemic tetany. This was investigated for cattle by Allcroft (1947 a.) who found that there was a peak incidence of clinical cases in the month of April in the four years of 1937-1941, followed by a sharp decline during the summer months when there were very few cases. There was then a secondary peak of incidence in the autumn months of October-November with other cases continuing to be reported throughout the winter. These incidence peaks of reported cases coincided with the times when the serum magnesium levels were lowest in the experimental herds which were being sampled by Allcroft throughout this period. Many other reports confirm this pattern of incidence, with the greatest number of cases appearing in the spring, a decline during the summer and a smaller outbreak of cases in the autumn and early winter. This pattern appears to be a reflection of firstly dairy herds being put to grass in the spring after the period of winter feeding. This is the season when these cows are most likely to be affected by an acute form of hypomagnesaemia leading possibly to clinical hypomagnesaemic tetany. There are in some years a few cases in these dairy herds in the autumn, associated with the autumn flush of grass (Rook & Storry, 1962). Outbreaks of hypomagnesaemic tetany in the autumn and early winter are normally found in out-wintered beef herds grazing poor pasture with little or no supplementary food. Such cases are usually associated with a chronic form of hypomagnesaemia which has developed over a long period and this is widely believed to be attributable to the low plane of nutrition on which these animals are maintained (Inglis, Weipers & Marr, 1954), and on the severe climatic

conditions to which these animals are exposed (Allcroft & Green, 1938; Allcroft, 1947 c). The pattern of incidence within a given region will depend of course on the type of farming practiced, in that in some areas, winter hypomagnesaemia may be a far greater problem than spring hypomagnesaemia purely because of the larger number of beef herds kept in that district. There has however been a tendency as reported by White (1953) for the autumn and winter hypomagnesaemic tetany to become more prevalent, whereas in the earlier years, the appearance of hypomagnesaemic tetany in the spring was considered to be the major problem. Although incidence of tetany in sheep shows the same tendency to vary from year to year, it occurs mainly in the spring season, generally two to four weeks after lambing. It seems to be associated not only with the spring flush of grass, particularly where the ewes are moved onto improved grassland, but also with the stage of lactation in that cases seldom occur directly after parturition nor do they occur in the later stages of lactation.

Degree of incidence.

Various attempts have been made in the last ten years to assess the annual incidence of hypomagnesaemic tetany in cattle in the various countries where it constitutes a problem. In the 1930's, no surveys were carried out, but some reports (e.g. Sjollema 1930 a; Broersma 1933) give the opinion that the number of animals affected was increasing and several workers suggested an association between this and the increasing intensification of pasture management and animal production.

In a survey on disease incidence in dairy herds in the South of England,

Withers (1955) found the annual incidence of hypomagnesaemic tetany to be 0.1% in the year 1950-1951, and 0.5% in the year 1951-1952. The survey covered 3,500 and 5,000 dairy cows in the respective years.

A wider survey entitled Disease, Wastage and Husbandry in the British Dairy Herd (Ministry of Agriculture 1959) was carried out in 1957-1958, covering over a 1,000 herds which contained a total of 31,000 cows. The overall incidence of hypomagnesaemic tetany in this sample was 0.48%, and the condition was found in 35% of herds with over 60 cows. This figure of 0.48% incidence can be compared with an incidence of 3.54% for milk fever. On the other hand, only 3% of the cows affected by milk fever died whereas 30% of the animals affected by hypomagnesaemic tetany died. Estimates of the total casualties over the whole country from hypomagnesaemic tetany and milk fever are given as 4,300 and 4,400 respectively. Thus to quote the survey "This high mortality rate associated with grass tetany places this disease equal in economic importance to milk fever".

Butler (1963) conducted a survey of 64 dairy herds in South Scotland and an overall incidence figure of 1.1% is given for hypomagnesaemic tetany, the cases occurring in 26% of the herds studied. Some 50% of the herds were receiving supplementary magnesium in some form.

Similar surveys have been conducted in Holland where the incidence of the condition seems to be higher than in Britain. t'Hart (1956) and Seekles & Boogaert (1955) estimated that between 1-2% of the dairy cows were affected annually by hypomagnesaemic tetany. Surveys in other countries e.g. America (Horvath, 1959); Ireland (Anon, 1962) and France (Larvor et al., 1961) have shown similar figures for annual incidence between 0.5 - 2%. A high

incidence of 10% was reported by Peters (1960) in the Schleswig - Holstein region of Germany. All these surveys covered only dairy herds and it should be noted that these incidence figures were found despite the prophylactic measures which would undoubtedly be in force on many of the farms surveyed i.e. incidence would probably have been higher if no preventive measures had been taken.

Incidence of hypomagnesaemic tetany among beef cattle in Britain has not been measured. In a limited survey Anon (1959) gives a figure of 5% in 8 beef herds in England, but the sample was biased toward herds with a known problem and was therefore not random. Horvath (1959) gives a figure of 1.4% of beef suckling cows in certain counties of West Virginia as being stricken annually with tetany. White (1953) makes the statement that in the last few years the beef herds seem to have suffered almost more than the dairy herd.

Incidence among sheep in Great Britain is also an unknown quantity, but Watt (1960) states that in 298 outbreaks of sudden death in sheep in East Scotland, 12% of the outbreaks were attributed to hypomagnesaemic tetany and a further 5% to combined hypomagnesaemia and hypocalcaemia. Watt pointed out in his paper that the figures refer to "outbreaks" and in the case of hypomagnesaemic tetany, one "outbreak" may have involved the death of 20 ewes. Stewart (1954) gave the general statement that the incidence among sheep was increasing.

The incidence of hypomagnesaemic tetany in calves was found by Blaxter & Sharman (1955) to be low but they found that between 5-7% of the calves surveyed developed hypomagnesaemia. The cases were confined to calves on milk diets with little extra feeding.

Incidence as affected by age, milk yield and species.

In a statistical study of hypomagnesaemia, Allcroft (1947 a) found the maximum number of cases in cattle to occur at the third and fourth calvings and he comments on "the association of hypomagnesaemia with the earlier calvings". Blaxter & McGill (1956), however, pointed out that Allcroft, in making this statement had taken no account of the age structure of the cow population in the country. Therefore, these authors recalculated Allcroft's data and showed the relative susceptibility of a cow to hypomagnesaemic tetany rose continuously with age. Old cows, which had had more than 6 calves, were 14 times more likely to develop the condition than were heifers in their first lactation. Cows in their third and fourth lactation were 6 - 10 times more susceptible.

The position is probably similar for sheep in that old ewes are widely believed to be more susceptible to hypomagnesaemic tetany than gimmers, although recorded experimental evidence of this is lacking. Many farmers, however, indicate that they may graze their gimmers with lambs on certain lowland pastures in safety, whereas older cast ewes with lambs are extremely liable to take "grass staggers". An incidental point in this connection is that gimmers may have only a single lamb or may be left with only a single, whereas older ewes generally have a high proportion of twin lambs and consequently higher milk yields. A high milk yield creates a demand for more magnesium and it could be this factor which causes a higher incidence among older ewes and not age per se. This possibility, however, would seem to be discounted by the evidence that, in cattle, there would seem to be no correlation between the incidence of hypomagnesaemic tetany cases and milk yields (Bartlett et al., 1957), despite the fact that a higher milk yield must

create an increased demand for magnesium. No breed differences have been established in either cattle or sheep in their susceptibility, apart from a report by Leech et al (1960) of a high incidence in Ayrshire cattle, but this observation was not intended to be comparative

Clinical Symptoms of hypomagnesaemic tetany.

The symptoms of hypomagnesaemic tetany as seen in dairy and beef cows, sheep and calves are similar and all may be covered by the one description.

One of the first descriptions given of the characteristic symptoms is that of Syollema (1930 a).

Many papers e.g. Dryerre (1932); Blakemore & Stewart (1933); Blaxter, Rook & McDonald (1954); McAleese & Forbes (1959); and Rook & Rowland (1962) have since described the syndrome in greater or lesser detail.

The clinical signs appear as being either acute or subacute. In the acute form, the onset of the condition can be very rapid in that a seemingly healthy animal will suddenly collapse in convulsive fits and unless curative treatment is supplied, it may die within fifteen to thirty minutes. The main symptom is one of hyperexcitability of the neuro-muscular system. This varies in degree from tremors in the superficial muscles to opisthotonus where the spinal column is arched and the head drawn back. The limb muscles usually show tetanic contractions and retraction of the upper eyelids gives the appearance of exophthalmos. Other symptoms are gnashing of the teeth, frothing at the mouth, and increase in the respiratory rate, and generally an elevated body temperature. This period of violent activity is often followed by a comatose condition prior to

death, or in some cases the convulsive and comatose conditions alternate several times during the course of the attack.

The subacute or chronic form of hypomagnesaemic tetany is essentially similar to the acute form and it differs only in the degree to which the various symptoms are shown. The main symptom is again hyperexcitability of the neuromuscular system, but in these subacute cases this is only evidenced by muscular twitching and tremors. The affected animal usually reacts nervously to an external stimulus such as touch or sound. In many cases it also walks with an exaggerated gait and it may appear to have an exaggerated view of obstacles in front of it e.g. it may take a flying leap over the dung channel. Such cases may continue to exhibit these mild symptoms for many days with either eventual recovery or alternatively death. In some cases the subacute condition is merely a prelude to the acute form of the disorder.

Biochemical Findings.

Over the years of study into this problem the only constant abnormality which has been found to exist in cases of hypomagnesaemic tetany in the various species is a lowered level of magnesium in the blood serum or plasma. The normal level of plasma magnesium is given as being between 1.8 - 3.2 mg/100 ml by Allcroft & Green (1934) but Allcroft (1947 a) places the lower limit at 1.7 mg/100 ml. This lower level of "normality" of 1.7 - 1.8 mg/100 ml is significant since it lies close to the renal threshold value for magnesium (Wilson 1960). Values below this level can properly be referred to as hypomagnesaemic.

In cases of hypomagnesaemic tetany, levels of 0.2 - 1.0 mg/100 ml plasma magnesium have been found by many workers e.g. cattle -: Sjollema & Seekles(1929),

Sjollema (1930a), Lothian (1931), Dryerre (1932), Blakemore & Stewart (1932-33), Nicholson & Shearer (1938), Allcroft (1947 a,b) and Marr (1958). Similarly for sheep, several workers have demonstrated the existence of low plasma magnesium values within the range 0.30 - 1.20 in cases of hypomagnesaemic tetany e.g. Penny & Arnold (1955), Inglis, Weipers & Pearce (1959), McAleese & Forbes (1959), Herd & Peebles (1962), and L'Estrange & Axford (1964 a,b). There are also many reports of cases of tetany in calves where analysis of the blood plasma established the presence of low magnesium values in the range 0.5 - 1.0 mg/100 ml e.g. Duncan, Huffman & Robinson (1935), Moine (1936), Peers & Armour (1958), Blaxter & Sharman (1955), Smith (1957,1958) and Todd & Rankin (1959).

There are also reports in the literature of low plasma magnesium concentrations leading to the typical symptoms of magnesium deficiency in other species such as horses (Green et al. 1935), cat (Lawler 1963), dogs (Kruse, Orent & McCollum 1932) and rats (McAleese & Forbes 1961). However, consideration of these non-ruminant species is outside the compass of this present work, particularly since the occurence of magnesium deficiency in other species is often one of artificial induction in feeding experiments and not of natural occurrence as is the case with ruminant animals.

Various studies have been conducted in past years where measurements have been made on experimental animals on a wide range of plasma or serum constituents and on other tissues in order to establish whether any abnormality other than a a low blood magnesium concentration is to be found consistently in cases of hypomagnesaemia. The classic work of Sjollema (1930a), which investigated 55 cases of clinical tetany, gave analyses, besides plasma magnesium, of plasma

calcium, both total and ionic, phosphorus, potassium, sodium and chloride. He also recorded analyses of urine samples from these tetany cases for magnesium, calcium, phosphorus and albumins. Apart from the consistent low serum magnesium, the serum calcium was also found to be low in the majority of the samples, the mean level being 6.65 mg/100 ml. Urinary magnesium was also reported as being low in most of the samples obtained from the clinical hypomagnesaemic tetany cases. The other constituents examined for were found to be either normal or having no consistent abnormal trend.

Dryerre (1932) made an even more detailed investigation of the blood samples from the hypomagnesaemic tetany cases in cattle which he investigated. Analyses were made for serum magnesium, calcium, sodium, potassium, inorganic and organic phosphorus, chloride, glucose, creatinine, haemoglobin, fibrinogen, albumin, globulin, amino-nitrogen, and non-protein nitrogen. He found only the serum magnesium to be abnormal in that the mean concentration was 0.39 (range 0.1-1.16) mg Mg/100 ml. His report differed from that of Sjollema in that serum calcium was normal (mean of 42 samples - 9.3 mg Ca/100 ml) with only two values below 8.0 mg/100cml. Dryerre's results, however, included an unspecified number of samples from unaffected animals in affected herds. His results may not therefore be strictly comparable with those of Sjollema.

Similar studies have been made more recently by Todd & Thomson (1960) and Storry (1961b) in hypomagnesaemic cows. These workers, in addition to analysing for serum magnesium, calcium, sodium, potassium and chloride, added to the growing list by giving figures for carbon dioxide combining power and red blood cell sodium and potassium. Neither investigation found any consistent deviation from the normal in any constituent other than serum magnesium, which was found to be low. Both of these conclusions, however,

could be criticised on the grounds that they were experimenting with hypomagnesaemic but clinically healthy cows and not dealing with actual hypomagnesaemic tetany cases as were Sjollema and, to a certain extent, Dryerre. The possible truth of this criticism is illustrated to some extent in Storry's report where he gives an account of three cows which developed clinical hypomagnesaemic tetany in a field adjacent to the experimental area. In these cows, analyses of the blood serum showed that all three animals had abnormally low levels of both serum magnesium and calcium.

Albuminuria has been associated with grass tetany by two investigators (Sjollema, 1930 a; Mershon & Custer, 1958) but it is difficult to judge the importance of these observations since albuminuria may be a common feature in cattle when grazing protein rich herbage.

It is therefore accepted by most workers in this field that low serum magnesium is the only consistent biochemical finding in cases of hypomagnesaemic tetany. The question of how important low serum calcium is in the development of tetany still remains open. It is one of the aims of this thesis to investigate this point more thoroughly and further discussion on the matter will be reserved until a later section (Section III).

General Biochemistry & Physiology of Magnesium.

The animal body contains about 0.04-0.05% by weight of magnesium of which roughly 59% is in the skeleton, 40% is in the cells of the soft tissues and the remaining 1% is in the extracellular fluids. Thus a 1000 lbs. adult bovine would contain approximately 200 g. of magnesium of which approximately 120 g.

would be skeletal, 80 g. would be in the tissues, and extracellular fluids would contain only 2 g. magnesium.

Intracellular fluid magnesium.

Magnesium is primarily an intracellular ion, in that although it occurs in the extracellular fluid and the skeleton, it is inside the cell that magnesium fulfills many of its functions as an indispensible element. Harris (1956) stated that the highest concentrations of magnesium occurred in the heart and skeletal muscle, but Wilson (1960) recalculated the data on a basis of intracellular water rather than wet tissue weight, and thereby showed most body tissues to have a similar value of about 30 milli-equivalents of magnesium per litre of intracellular water. (i.e. 37 mg Mg/100 ml).

The concentration of magnesium in the extracellular fluid (E.C.F.) is much lower than inside the cell. In most species and in ruminants in particular it is about 2 meg./litre (2.5 mg/100 ml). Thus a large concentration gradient between extracellular and intracellular magnesium is maintained but the mechanism concerned in the maintenance of this gradient is as yet unknown. It may be that only a small proportion of the intracellular magnesium is ionic, in physio-chemical equilibrium with the ionic E.C.F. magnesium, the remainder inside the cell being bound to protein molecules (Wilson, 1960). Evidence for this hypothesis is given by Brandt et al (1958) and McIntyre (1959) who showed, by the use of isotope tracer studies that a small proportion of the intracellular magnesium is exchangeable. The alternative would be that this level of intracellular magnesium is maintained by an active process. Contrasting results have been obtained with regard to any

changes which may occur in the concentration of intracellular magnesium during the development of hypomagnesaemic tetany. Watchorn & McCance (1937) and Blaxter et al. (1954) found no change in the concentration whereas MacIntyre & Davidson (1958) found a fall in the level of magnesium in muscle during the development of magnesium deficiency in rats.

Extracellular fluid magnesium (E.C.F.)

As stated above, the concentration of magnesium in the E.C.F. is normally about 2.5 mgs/100 ml. The E.C.F. is composed of both the plasma and the interstitial fluid, the latter being an ultrafiltrate of the former, and also having a lower concentration of magnesium because of the lower protein content of interstitial fluid. In the study of magnesium nutrition, samples are normally taken from the plasma since it is the only accessible fluid, but the presumption is made that the concentration of magnesium in plasma and interstitial fluid is similar, since they are to some extent the one component.

A proportion of the plasma magnesium is ultrafiltrable, being predominately ionic, and it is this ionic fraction that is believed to be the physiologically active form. Values of from 65 to 80 per cent of the total plasma magnesium have been given for this ultrafiltrable fraction (Blaxter, Cowlishaw & Rook, 1960; Walser, 1961). The fraction which is not ultrafiltrable is bound reversibly to plasma albumin and globulin (Prasad et al. 1950) and this binding can be modified by pH and by the concentration of other ions, such as calcium, which compete for binding sites (Carr, 1955).

Bone Magnesium.

The presence of magnesium in bone is the result of hetero-ionic exchange process between the bone mineral surface and the extracellular fluid at the crystal surface, and magnesium ions thus replace a calcium ion in the apatite crystal lattice. This process is reversible in that if the concentration of magnesium in the surrounding fluid is low, then a calcium ion can replace the magnesium ion which then joins the ionic magnesium fraction of the E.C.F. Blaxter, (1956); Smith, (1959 b). The bone crystal surface, therefore, acts as a resevoir of magnesium to be drawn upon at a time when the concentration of magnesium ions in the E.C.F. falls because of an increased demand from the tissues for growth, increased lactation, or alternatively, reduction in dietary magnesium intake. Blaxter (1955) and Blaxter & Sharman (1955) showed that the skeletons of young calves reared on magnesium deficient diets could lose 60% of their total magnesium content by this mechanism.

On the other hand, this ability of the skeleton to act as a labile source of magnesium in this manner is largely a function of the young animal and decreases with age. In adults, a very large part of the skeleton is metabolically inert, due to the progressive diminution of its blood supply. Recrystallisation of bone mineral also takes place slowly as the animal ages and this tends to further reduce the exchangability of the magnesium in the bone crystal. The adult animal is consequently less able to withdraw bone reserves of magnesium in time of dietary deficiency or increased demand and magnesium homeostasis is thus impaired. This may well form an explanation of the much greater incidence of hypomagnesaemic tetany in older animals and it is, in fact, found that in adult ruminants dying from tetany,

the magnesium content of bones is normal (Cunningham, 1936). The amount of magnesium necessary to maintain a normal serum magnesium concentration would admittedly be small and its removal from bone would probably not be detectable by bone analysis, but conversely the very fact than an adult animal in times of dire necessity is unable to remove rapidly from the bone reserves the relatively minute quantity of 100 mg of magnesium required to raise the serum level by 0.5 mg/100 ml, would suggest that this depot is not rapidly available. It is believed to be slowly exchangeable.

Biochemical Functions of Magnesium.

Magnesium is an important activator of many enzymes and its role as such has been fully reviewed by Lehninger (1950).

Many enzymes require the presence of metallic ions to enable them to carry out their specific functions. On the removal of these metallic ions by dialysis, the enzyme becomes inactive, but on the readdition of these metallic ions, activity is fully restored to the enzyme. These metallic ion "activators" are believed to act by enabling the enzyme to link up with the substrate via the formation of an enzyme - metal - substrate complex. In some enzyme systems, there is a requirement for a specific metal while for other enzymes, any one of many metals will suit.

Magnesium is one of these metallic ions which will catalyse enzyme reactions, in some cases being the only ion which will confer activity to the system and in other cases being one of a number of alternative ions, which can activate a particular enzyme. This function seems to be the basic biochemical role of magnesium in living tissues, and in the animal body, the two main spheres of

magnesium activity are:-

(a) enzymes concerned in carbohydrate metabolism and

(b) enzymes involved in the nervous system.

(a) One of the important reactions in carbohydrate metabolism for which magnesium is a necessary cofactor is in the oxidative decarboxylation of pyruvic acid to acetylcoenzyme A. Any interference with this step would lead to an accumulation of pyruvic acid. A further example in carbohydrate metabolism where magnesium is a specific cofactor is in the oxidative decarboxylation of α-Ketoglutaric acid to form succinic acid as part of the Kreb's cycle and in the absence of magnesium, this reaction would not take place, thereby interrupting Kreb's cycle which in turn would mean a build up of intermediary metabolic compounds and a lack of 'high energy' phosphate bonds, essential for normal metabolic processes. This interference with normal carbohydrate metabolism which would result from severe magnesium deficiency in the tissues, can possibly explain the nutritive failure where animals fail to thrive and to gain weight which is a feature in some species under magnesium deficiency e.g. in dogs (Kruse, Orent & McCollum, 1932).

(b) The second important sphere of activity of magnesium as an enzyme-cofactor is in the nervous system. The site of action is believed to be at the neuromuscular junction (Hoff, Smith & Winkler, 1940) although there is evidence to suggest that it also has a direct effect on the central nervous system (Bryant, Leymann & Knoefel, 1939). Del Castillo & Engbaek (1953) demonstrated that addition of magnesium to a frog muscle preparation diminishes the end plate potential and reduces the amount of acetylcholine released by the motor nerve

endings. This depressant action of magnesium is antagonised by calcium (Malorny & Ohnesorge, 1951). The explanation of this observation would seem to be that magnesium is an essential cofactor for the activity of the enzymes which synthesise and hydrolyse acetylcholine. Acetylcholine is the "transmitting agent" which, when released, acts upon a receptor to trigger off, indirectly, muscular contractions, and therefore rapid destruction of this transmitting agent is of prime importance in the question of muscular control and consequently the accelerating influence of magnesium will be considerable. According to Rook & Storry (1962) in their review of magnesium nutrition, a reduction of extracellular magnesium would

(1) increase the release of acetylcholine by the motor nerve endings

(2) increase the sensitivity of the end plate to liberated acetylcholine

(3) decrease the hydrolysis of liberated acetylcholine, with consequent tendency to delay of recovery from stimulation

(4) increase the sensitivity of the muscle membrane to the electrical impulse from the nerve, causing a greater "twitch"

(5) inhibit the relaxing factor of muscle.

This involvement of magnesium in the transmission of impulses at the neuro-muscular junction is the possible explanation of the experimental fact that in magnesium deficiency in all species, the main symptom is derangement of the nervous system, manifest in convulsions which may prove fatal. It would also explain the converse condition where excess magnesium given intravenously produces anaesthesia and eventual death from respiratory failure. The depressant action of magnesium is largely antagonised by calcium.

Magnesium Absorption, Excretion and Homeostasis.

Absorption.

Based on studies with sheep using the isotope 28 Mg., Field (1960,1961) concluded that absorption of dietary magnesium occurred mainly from the middle third of the small intestine. Other reports, on the whole, confirm this finding both in the sheep (Care & van t'Klooster, 1964) and also in other species e.g. in rabbits (Aikawa, 1959). Stewart & Moodie (1956) however, concluded that in sheep absorption of magnesium could take place from any part of the gut, although the main site was still the duodenum and the ilium. This technique, however, was criticised by Field (1960) and Rook & Storry (1962) in that large quantities of magnesium salts in the gut were used, this not being strictly comparable to practical feeding. On the question of absorption of magnesium from the colon, Smith (1959 b, 1962) demonstrated magnesium absorption in young calves from both the small and the large intestine during the first 3 - 4 weeks of life, but thereafter only from the small intestine. This may explain the observed decrease in availability of dietary magnesium in older milk fed calves as compared with younger animals (Smith 1957, Smith 1959 c). There is still doubt as to whether the mechanism involved in the absorption of magnesium from the intestine is purely a passive ion transport across the gut wall, or alternatively an active process. As pointed out by Storry (1961a) passive diffusion of magnesium into the blood stream could theoretically be expected. On the other hand there is considerable evidence that

in some species at least, an active process is involved since magnesium and calcium absorbtion have been shown to be inversely related suggesting the presence of a common transport mechanism e.g. Alcock & MacIntyre (1962) in rats, Care & Van t'Klooster (1964) in sheep. Phillipson and Storry (1962) with sheep and Smith (1962) with calves reported that in these species no such relationship existed.

Excretion.

There are three pathways of excretion for magnesium from the animal body, namely, urinary, faecal, endogenous losses. In the lactating animal there is additional loss via the milk. Quantitatively, urinary excretion is variable and to some extent controllable, dependant on the magnesium status of the body, whereas endogenous faecal losses and excretion via the milk continue at a given rate, irrespective of the magnesium status of the animal. Urinary excretion appears to be controlled by a filtration - reabsorbtion mechanism in which magnesium behaves as a threshold substance appearing in urine only when the magnesium load filtered by the glomeruli exceeds that reabsorbed by the tubules (Wilson, 1960). Using values of 2 mg/minute and 100 ml/minute for the tubular reabsorption rate and the glomerular filtration rate respectively, he calculated that the threshold concentration in the plasma is about 2 mg/100 ml. This agrees closely with direct measurements in cattle which give a figure of 1.8 mg /100 ml. Above this value, Storry & Rook (1962) found the concentration of plasma magnesium and the urinary excretion rate to be linearly related. Rook, Balch & Line (1958) found a similar rectilinear relationship in cows, but placed the renal threshold value at not greater than

2.15 mg/100 ml. This relationship is clearly shown by Rook & Balch (1958) in an experiment where dairy cows were put to grass in the spring, thereby causing a fall in plasma magnesium value into the hypomagnesaemic range ($\leq$ 1.8 mg/100 ml). Urinary excretion of magnesium in these animals immediately dropped to zero.

Endogenous magnesium is excreted into the gastro-intestinal tract as a constituent of mainly the saliva and the gastric juice, but also the bile and pancreatic juice (Field, 1960). Faecal loss of this endogenous material represents a failure on the animal's part to reabsorb this magnesium during passage of the material along the gastro-intestinal tract. Field (1960) suggested that there is little reabsorption of this material and that the daily endogenous faecal loss is therefore close to the figure of 60 - 200 mg/sheep which he gives for total daily endogenous magnesium output. It is obviously of importance to know the extent of this loss of body magnesium and consequently many measurements by various methods have been made. The results of these measurements for cows vary from 1.5 mg/Kg B.W./day (Simesen et al, 1962) to 3-5 mg/Kg B.W./day (Blaxter & McGill, 1956). In calves the various estimations give a similar range to those given above for cows, with the exception that Smith (1959 a) found the low value of 0.5 mg/Kg B.W./day for young calves of 2-5 weeks, increasing to 2.2 - 4.0 mg/Kg B.W./day once the calves were given access to fibrous food. He attributed this increase in endogenous faecal loss to an increase in salivary secretion.

Magnesium excretion via the milk can be considerable in the cow. Milk contains an average 12 mg. magnesium/100 ml. (Blaxter & McGill, 1956; Robertson et al 1960) i.e. an animal giving 4 galls/day would lose 2.5 g. magnesium/day. Furthermore, several workers have shown that if hypomagnesaemia develops in cattle,

there is no fall in the magnesium concentration of the milk (Cunningham,1930-37; Robertson et al, 1960; Rook, 1962).

Magnesium Homeostasis.

The regulation and the maintenance of the normal magnesium concentration in the plasma fluid (i.e. in the extra cellular fluid) is generally believed to be a balance between intake from the digestive tract and renal excretion. Thus an animal would depend on a sufficiently high continuous absorption of dietary magnesium to balance the unavoidable losses and, on the other hand, renal excretion would serve to remove any excess which is absorbed (above the renal threshold value of approximately 2 mg Mg/100 ml plasma). Magnesium reserves in bone are also believed to act as a buffer in this homeostatic mechanism, although the ability of the skeleton to play this part seems to decrease with age. Many attempts have been made to discover a possible involvement of endocrinal control in magnesium homeostasis, but so far no one gland has been shown to have any great effect on the maintenance of normal plasma magnesium, although there is evidence of some interrelation with the thyroids (Care & Keynes., 1964) and the adrenal cortex (Hanna & MacIntyre, 1960).

Availability of dietary magnesium.

Since magnesium homeostasis would seem to be a balance between absorption and excretion, deficiency will depend very much on factors which affect either of these functions. Thus magnesium nutrition is going to depend on not only the amount of magnesium in the diet, but also on the availability of this dietary magnesium.

Most of the measurements made show that the availability of dietary magnesium is low. Blaxter & Rook (1954) estimated the availability of the magnesium of typical winter rations fed to dairy cows to be 30 - 50%. Rook, Balch & Line (1958) give a lower figure of 23 - 34% for similar diets fed to dairy cattle. Both of these figures were based on "apparent availability" where no attempt was made to allow for endogenous faecal magnesium. Field, McCallum & Butler (1958) estimated the true availability of magnesium from fresh spring grass as 13% and 26% for two sheep. Care (1960 a) found similar true availability values of 10 and 24% for two sheep. In further experiments Field (1960) reported availability figures of 3,7,8 and 12% for magnesium in grass nuts. His results showed not only the low % availability but also the great individual variation between animals.

Magnesium requirements for maintenance and production.

Blaxter & McGill (1956) calculated the minimal net daily requirements for dairy animals of varying weights. Assuming the availability of dietary magnesium to be 33%, they thereby calculated daily actual requirements as shown in Table I.

Table I.

Calculated minimal Mg requirements of 1200 lbs cow.

Milk yield (galls).	Mg Requirement (g/day).	
	Net.	Actual.
0	1.80	5.40
1	2.41	7.23
2	3.02	9.06
3	3.64	10.92
4	4.25	12.75
5	4.86	14.58

This would mean for example that a dairy cow giving four galls. of milk eating 28 lbs. of dry matter per day would have its magnesium requirement met by the diet containing 0.1% Mg. The limited balance trials which have been carried out would seem to confirm that these calculations are in the right order. In particular the work of Drierem et al (1949) demonstrated a correlation between the magnesium content of the diet and the degree of hypomagnesaemia produced. So much, however, depends on whether the assumption of 33% availability for dietary magnesium is correct. According to the work of Field with sheep quoted earlier, %

availability may in fact be much lower than this figure, and this would lead consequently to a much higher figure for actual daily requirement.

Various attempts have been made to detect a drop in availability of dietary magnesium during the change-over from indoor to grass feeding, and there is some evidence to show that the magnesium of young grass cut early in the season, particularly when rich in nitrogen or potash, is of lower than average availability (Van der Horst,(1960); Kemp et al,(1961); Rook & Campling, (1962)). However, Field et al. (1958) found the same % availability for various spring herbage samples irrespective of whether they had come from "tetany producing" or "normal" pastures.

Magnesium in foods and pasture.

Within the context of studies on hypomagnesaemia and hypomagnesaemic tetany in ruminant animals, consideration of magnesium in foods can be confined purely to that of pasture grass and its by-products. Although cereal and protein concentrate foods do have varying contents of magnesium, the normal indoor diets of cattle and sheep usually have comparatively high levels i.e. above 0.20% Mg, and animals given these diets seldom if ever develop hypomagnesaemia. Indeed, the converse is normally true in that extremely artificial indoor diets have to be devised to reproduce hypomagnesaemia experimentally. For example, Rook (1963) made use of constituents such as wheat straw, paper pulp, flaked maize, maize starch, maize gluten, casein, sugar, coconut oil and groundnut oil to produce hypomagnesaemia in a lactating cow. The factors which influence the magnesium content of pasture can be broadly delineated as follows:-

(a) the relative amounts of grass, clovers, and herbs

(b) in a grass sward, the species of the grasses

(c) a seasonal variation from spring to summer

(d) the magnesium status and fertility of the soil

(e) the fertilisation of the sward, particularly with nitrogenous, potassic, and magnesium fertilisers.

As a general rule, grasses have a lower magnesium content than clovers, and herbs have a higher content than both grasses and clovers. An example of this difference is given by Thomson (1960) as grasses 0.24% Mg, legumes 0.69% Mg. and herbs 0.75% Mg. This fact is reflected in the incidence of hypomagnesaemic tetany occurring chiefly on pure grass swards.

Todd (1961 a) gives figures for the difference in magnesium content of a representative list of various grass strains and species, at different seasons, as shown in table 2.

Table 2.

Grass Strain and species differences in magnesium content (Todd 1961 a).

Grass Species.	% Mg in D.M.	
	Apr.-May.	June-Aug.
S23 ryegrass	0.13	0.19
Italian ryegrass	0.15	0.20
Cocksfoot	0.15	0.23
Timothy	0.10	0.14
Meadow grass	0.12	0.19

This demonstrates the variability among species, to the extent of some being at least 50% higher in magnesium content than other species, and also the much lower values that all grass swards have in spring. Consistently higher values are found in June - July. Similar values are given by Wolton (1960) both for species difference and seasonal variation. Wolton gives a more complete picture of these seasonal changes with initially higher values in March, followed by a fall during April and May with a return to higher values in June and July. This variation may be partly due to differences in temperature, since Dijkshoorn & t'Hart (1957) found that the magnesium content of grass grown at a temperature of 40°F was 30% lower than a comparable sward grown at 70°F. This would be in agreement with the peak seasonal incidence of hypomagnesaemia occurring in the early spring. Rook & Wood (1960) gives 0.09 - 0.16% Mg as a normal range for spring pasture samples. This was based on 62 samples taken from various experimental sites.

There seems to be no direct correlation between the incidence of hypomagnesaemic tetany and the magnesium content of herbage being consumed. (Blakemore & Stewart, 1933; Hopkirk, Marshall & Blake, 1933; Nicholson & Shearer, 1938). Komp et al (1960), however, in a survey of 822 cows amongst which 23 showed clinical signs of tetany, found that no cases occurred where the magnesium content of the herbage was above 0.20%. Cases did occur in cattle grazing herbage with contents of from 0.075 - 0.19%Mg, but equally there were many cows grazing herbage with 0.10 - 0.15 which maintained normal plasma magnesium concentrations. It would probably be reasonable to suggest that there may be a correlation in that the lower the herbage magnesium content then the greater

will be the risk of clinical tetany. Since the variation in susceptability of individual aminals is so great, correlation is difficult to demonstrate except at the extreme ends of the scale i.e. at a hypothetical magnesium zero content when case incidence would be high and above 0.20% herbage magnesium where case incidence is zero. Another factor which has to be remembered is the variable extent to which each animal's system can maintain magnesium homeostasis by using body reserves when the diet is low in magnesium.

It is sometimes stated that there is no connection between soil magnesium content and the occurrence of hypomagnesaemic tetany but as Thompson (1960) points out although no obvious correlation will be found, there must be some inter-relationship since clinical cases are not found on soils, such as the magnesium limestone soils in Durham, which are high in magnesium. Similarly at the opposite end of the scale, light soils deficient in magnesium will produce herbage with low magnesium contents. Therefore soil magnesium will have a bearing on the incidence of tetany in so far as it has a correlation with plant magnesium. It is in this connection that magnesium fertiliser applications are of importance. If a soil treatment (with magnesium salts) causes a sufficiently large increase in soil exchangeable magnesium to raise the magnesium uptake by the plant to the point where the herbage magnesium content is above 0.20%, then such a treatment will prevent hypomagnesaemic tetany.

The use of nitrogenous and potassic fertilisers influence the magnesium content of grass in that application of nitrogen alone will increase the magnesium content of grass, (e.g. Walsh & Clark, 1945; Neales, 1956; Wolton, 1960; Hemingway, 1961; and Hunt et al, 1964) and the application of potassium salts alone will decrease the magnesium content of grass (e.g. Watkins, 1957; Kemp, 1960; Wolton, 1960; Hemingway, 1961; and Hunt et al; 1964). However, the

situation is more complex in that these interactions are true for a pure grass sward, but when a pasture field is considered, other factors are then involved. If the pasture is a grass/clover mixture, as is often the case, then nitrogen fertilisation will tend to encourage grass growth with consequent suppression of clover, so that although nitrogen increases the magnesium content of the grass, the reduced proportion of clover (which has a higher magnesium content) may mean that the sward has a lower magnesium content, (e.g. Wolton, 1960). Conversely, potassium fertilisation may encourage clover growth and therefore although it reduces the magnesium content of grass, the end result may be a pasture with a higher magnesium content. However, clover in many swards may not be expected to make an important contribution to the dry matter content of spring pastures.

Grass swards under normal commercial conditions, frequently have very low magnesium contents. For example, Blaxter & McGill (1956) quote 0.08% Mg content for spring herbage, and Thomas et al (1955) give 0.06 - 0.15% Mg as the range for hay crops taken from the Cockle Park plots. These figures should be considered in conjunction with the estimates of Blaxter & McGill (1956) quoted earlier that dairy cows would maintain normal serum magnesium on a diet containing 0.1% Mg, but that below this figure, there is a danger of hypomagnesaemia.

The employment of magnesium salt fertilisers to increase the magnesium content of herbage has been the subject of many investigations. Table 3 details the results of several workers, giving comparative figures for magnesium treated and untreated pastures.

Table 3. % Mg. in spring herbage treated with magnesium fertilisers.

Author	Mg applied (lbs/acre)	Material	% Mg in herbage Untreated.	% Mg in herbage Treated.	% increase.
Wolton(1960)	12	$MgSO_4$	—	—	2
	24	$MgSO_4$	—	—	5
Hemingway(1961)	22	$MgSO_4$	0.10	0.11	10
Birch & Wolton(1961)	30	MgO	0.14	0.17	21
Wolton(1960)	54	$MgSO_4$	—	—	10
Smyth et al(1958)	325	$MgSO_4$	0.16	0.24	50
Parr & Allcroft(1957)	600	MgO(10cwt)	0.16	0.24	73
Birch & Wolton(1961)	600	MgO(10cwt)	0.14	0.25	79
Bartlett et al(1954)	1500	MgO(25cwt)	0.15	0.24	60
Bartlett et al(1954)	1500	MgO(25cwt)	0.17	0.34	100

These results, obtained at various experimental sites, would suggest that there is a more or less proportional increase in herbage magnesium content for increasing amounts of magnesium fertiliser applied and it is only above the level of 300 lbs Mg/acre (= 5 cwts calcined magnesite/acre) that the pasture is consistently above the "safety" level of 0.20% Mg. Although there is no supporting evidence in the above table, it has been found (Collins 1960) that the response to magnesium fertilisers is partly dependant on the type of soil and soil pH. Acid soils respond well and alkaline or heavier soils respond poorly.

To give a quantitative assessment to some of the factors discussed above, t'Hart (1960) estimated that if a pure grass sward, growing at 65°F, with medium

potash and no magnesium fertilisation, is taken as standard at 0.20%

magnesium, then:-		change in % Mg.
	temperature 20°F lower	- 0.05
	200 lbs K_2O extra/acre	- 0.02
	200 lbs MgO extra/acre	+ 0.05
	10% clover	+ 0.03

The above discussion of the effect of fertilisation on the magnesium content of grass deals purely with the effect of such treatments on the sward alone without reference to the animal grazing the pasture. There is a great deal of information on the effect of these fertiliser treatments, via the grass, on the incidence of hypomagnesaemia in cattle.

An association betweem fertiliser use and "grass" tetany was suspected as long ago as 1930 by Sjollema who commented on the possibility of hypomagnesaemia being related to intensive grassland management. Since then many attempts have been made to measure scientifically the degree to which they may be related. t'Hart (1960) gives a summary of the results of five studies in this field (Table 4).

Table 4. Influence of N and K fertilisation on serum magnesium concentrations and incidence of hypomagnesaemic tetany in dairy cows (t'Hart, 1960).

	Fertiliser treatment.			
	Control.	N.	K.	NK.
	serum magnesium (mg/100 ml)			
Bartlett et al(1954)	1.2-1.9	0.4-0.6	0.6-1.3	—
Smyth et al(1958)	1.50	1.52	1.50	0.70
Hvidsten et al(1959)	—	2.54	—	1.93
Kemp(1958b)	2.18	1.00	0.58	0.53
Kemp(1959)	—	2.55	—	1.10
	number of tetany cases			
Bartlett et al(1954)	0	6	1	—
Smyth et al(1958)	0	0	0	2
Hvidsten et al(1959)	—	0	—	2
Kemp(1958b)	0	0	4	2
Kemp(1959)	—	0	—	2

These examples would suggest that there is indeed an association, particularly when nitrogen and potassium are used in conjunction. One comment which may be made at this stage is the unrealistically heavy dressings which were employed in some of these experiments. However more detailed discussion of this aspect will be made in the experimental section Part I.

There is no published information of the extent to which the incidence of hypomagnesaemic tetany in sheep is influenced by fertiliser treatment of the pasture, and therefore the experimental work described in Section II was designed

with a view to providing information on this subject.

The effect of magnesium fertilisation on reducing the incidence of tetany will also be the subject of a later section, and therefore discussion of this aspect will be reserved until Section IV, which deals with the prevention of hypomagnesaemic tetany.

Theories on the aetiology of hypomagnesaemic tetany.

At the outset, it can be said that at the present time the exact cause of hypomagnesaemic tetany in adult cattle and sheep remains unknown. Many underlying factors which exacerbate the incidence have been described and prophylactic measures are known, but this does little to clarify the basic nutritional, physiological or biochemical dysfunction which must occur.

Most theories put forward to explain this metabolic disorder are based on it being a simple or conditioned nutritional deficiency, (using the term "nutritional" in its broadest sense of the net gain to an animal from the diet). The alternative possibility of dysfunction in some endocrinal or physiological mechanism within the body, apart from the gastrointestinal tract, has been suggested by Green (1939), Allcroft (1947 c), and Bartlett et al (1954) but this approach has received little encouragement (Wilson, 1960).

There is the complication of there being both an acute form, usually in the spring and a chronic form of this disorder, usually in the winter months, and therefore they may have different aetiologies.

There is considerable evidence to indicate that hypomagnesaemic tetany in milk-fed calves is a simple uncomplicated dietary deficiency of magnesium. (Blaxter & Sharman, 1955, Smith, 1957, Smith, 1958). Cow's milk contains an average 0.013% magnesium. (=0.1% on D.M. basis). Blaxter & Sharman (1955) reasoned, by calculation, that when milk is the sole diet, it would supply insufficient magnesium to meet the demands for maintenance and body growth. The calf, however, differs from adult ruminants in being able to utilise bone magnesium reserves to maintain a normal plasma magnesium concentration and it is therefore only after severe depletion of these reserves (up to 60% loss) that hypomagnesaemia and tetany

develop. (Blaxter, 1955). This may take up to five months (Smith, 1961). This process can be accelerated to produce tetany in 8 - 9 weeks by the use of milk treated with an ion exchange resin to remove 50% of the magnesium (Todd & Rankin, 1959). The fact that the progressive development of hypomagnesaemia can be accelerated by reducing the magnesium content of the diet is strong evidence for the deficiency being of simple dietary origin. It was further shown by Smith (1961 b) that calves eating fibre in the form of wood shavings develop tetany at an earlier age than muzzled calves, and also suffer a greater net decrease in magnesium absorption. He attributed this decrease in apparent availability to the increased salivary secretion and hence increased endogenous magnesium excretion of calves eating fibre. The development of chronic hypomagnesaemia and hypomagnesaemic tetany in adult cattle during the winter months was attributed by Allcroft (1947 c) to abnormal function of some postulated endocrinal control of magnesium which was in turn influenced by the cold, wet conditions which he found to be associated with the minimal blood magnesium levels. Inglis et al. (1964), however, did not find this association and these authors believed the condition to be rather of dietary origin, since supplementary feeding restored serum magnesium to normal levels. Inglis et al. (1959) gave support to this theory by demonstrating with sheep that hypomagnesaemia could be induced by fasting for a short period. Christian & Williams (1960) and Manktelow & Olivant (1960) described similar results with fasted sheep. The general view now seems to be that hypomagnesaemic tetany in the winter months occurs mainly in outwintered beef cows with only poor grazing and little supplementary food on which to subsist. This undernutrition, coupled with a comparatively high demand for magnesium for

lactation, causes a slow fall in serum magnesium levels over the winter (e.g. Muth & Haag, 1945; White, 1960; Mershon & Custer, 1956; Rook, 1961). Since the fall takes place over a period of several weeks there is the opportunity for the slowly available bone reserves to contribute towards magnesium homeostasis. It is significant in this context that a single injection treatment of such clinical cases only provides a temporary cure, and the serum magnesium quickly returns to a low level. (White, 1960). This can be compared with the permanent cure which is often affected by a single injection of an appropriate amount of magnesium solution in clinical cases on spring pasture. With regard to bone reserves, it is also significant that Allcroft (1960) reports of finding a slightly lower than usual bone magnesium concentration in a few cases where chronic hypomagnesaemia had been present for several months. It is generally agreed that the acute case of hypomagnesaemic tetany occurring in the spring is one of 'nutritional' origin (Ender et al. 1948; Head & Rook, 1955; Wilson, 1958;) but many theories have been put forward to explain the exact nature of the deficiency. Few workers accept that it is an absolute dietary deficiency of magnesium, since it has been shown that pastures on which tetany occurs have the same magnesium content as many pastures on which animals remain healthy. (Stewart 1951). Workers in this field have therefore looked for factors which may interfere with the availability of dietary magnesium in pasture, or alternatively, postulated a reduction in the absorption rate. One theory which has been given attention is the possible incrimination of the high potassium and/or the high protein content of spring grass. Head & Rook(1955,1957) working with cattle and Christian & Williams (1960) working with sheep were unable to prove a definite association between a

high protein intake or a high rumen ammonia concentration and hypomagnesaemia. Similarly attempts to induce hypomagnesaemia by dosing with potassium salts have been unsuccessful (e.g. Pearson et al. 1949; Odell et al. 1952). There is, nevertheless, the close association discussed earlier between incidence of hypomagnesaemic tetany and pasture heavily fertilised with nitrogenous and potassic fertilisers. These pastures usually have high potassium and protein levels but as Blaxter & McGill (1956) point out association does not imply causation

Kemp & t'Hart (1957) by studying a large number of pasture samples have expressed the joint importance of high potassium and low magnesium contents in grass by the ratio $\frac{K}{Mg+Ca}$ (in milli-equivalents). Their results indicated that when this ratio was greater than 2.2, there was an enhanced risk on that pasture of cattle developing hypomagnesaemic tetany. Conversely when the ratio was below 2.2, the risk was negligible. However other workers (e.g. Horst & Hendriks, 1958; Rook & Wood, 1960; Seekles & Hendriks, 1963; Seekles, 1964) have tended to doubt the significance of this ratio. Brouwer (1952), in a similar fashion, discussed the importance of 'earth alkalinity' (Ca+Mg+P) whilst Endor et al. (1957) believed a high sulphur content of the pasture to have an effect, in that nitrogen applied as ammonium sulphate to the pasture induced tetany and nitrate fertilisers did not. Bartlett et al. (1954) and Simesen (1959) similarly found that ammonium fertilisers had an exacerbating effect (as compared with nitrates) on the development of hypomagnesaemia and Simesen suggested this may be due to precipation of magnesium as magnesium ammonium phosphate in the presence of the high rumen ammonia. Paterson & Crichton (1960) found that the use of dietary supplements of sodium salts markedly reduced the incidence of hypomagnesaemic tetany

(and incidentally raised the milk yield) in dairy cows. They postulated that the deleterious influence of potassic fertilisers on tetany was perhaps due to the depression in pasture sodium content caused by the application of potassium. Another precipitation theory is that of Roberts & Yudkin (1960) who postulated the precipitation of insoluble magnesium phytate in the rumen as a cause of magnesium deficiency disorders in grazing animals.

Rook & Balch (1958) found a sharp drop in both the magnesium intake and its apparent availability when grass diets were fed to dairy cows in place of indoor concentrate diets, and they therefore proposed that hypomagnesaemia was a dietary deficiency due to a reduced intake and a lowered availability. These results were confirmed by Kemp et al.(1961).

This latter theory would seem a reasonable explanation particularly since, as pointed out by Wilson (1960), any drop in the absorbtion rate of dietary magnesium would also affect the reabsorption of endogenous magnesium and could lead to a direct loss of extra cellular magnesium. Blaxter & McGill (1956) also make the comment that milk yields generally increase when the change to a grass diet occurs. There is thus the possibility of the coincidental superimposition of several factors adversely affecting magnesium nutrition, when a dairy animal is put to pasture i.e.

(a) reduced magnesium content of diet

(b) possibly reduced dietary intake of dry matter

(c) reduced availability of dietary magnesium

(d) increased loss of endogenous magnesium due to (c)

(e) possibly increased endogenous magnesium output because of increased salivary output stimulated by type of diet

(f) increased magnesium requirement for milk yield.

Bone reserves of magnesium cannot be drawn upon rapidly in the adult animal (Blaxter & McGill 1956) to cover the sudden demand an abrupt dietary change imposes. The aetiology of hypomagnesaemic tetany in sheep has not been investigated closely, although the conditions under which it arises are similar to those for cattle, and it would therefore seem likely that the origins of the disorders are similar. There is, of course, no dietary change from indoor feeding in the spring in the case of sheep, but they often have a pasture change and it is well known that purely changing a flock of sheep from one pasture to another is sufficient to precipitate an attack of hypomagnesaemic tetany. It is the author's experience that if sheep have been on a particular pasture for a period of more than two to three weeks, then a move to another pasture is in most cases accompanied by a drop in plasma magnesium concentrations.

Aims of the present work.

Three main aspects of hypomagnesaemic tetany were chosen as being of particular interest for further study.

(1) Very little attention has been paid to the development of hypomagnesaemia and tetany in sheep. It would be of particular interest to know whether the close association found in cattle between fertilisation of pasture and incidence would also be found with sheep. Observations have also been made on the effect of the age, number of lambs and the breed of the ewe, on the development of hypomagnesaemia in the spring.

(2) The possible implication of serum calcium concentrations in the development of hypomagnesaemic tetany has received surprisingly little attention, despite the fact that the majority of cases recorded in the literature have a concomitant low concentration of plasma calcium. This merited further investigation.

(3) The present methods of prevention are not foolproof and further studies into the comparative usefulness of the present methods have been made, together with an attempt to elucidate in more detail their action. The possibility of a new method of prevention by means of a long acting magnesium containing rumen pellet has been investigated in detail.

Chemical Methods.

Details of the precise procedures used for the various estimations are given in Appendix I. Apart from a more detailed consideration of serum magnesium analysis, only the type of method used and the principles involved will be given here, together with any modifications which were made.

BLOOD PLASMA ANALYSIS.

Calcium and magnesium in plasma.

It was apparent at the commencement of the work described in this thesis that the bulk of the chemical analytical work in this study would be for calcium and magnesium estimations in blood sera. Because of the large number of animals likely to be involved and the frequency of the sampling dates, it was also apparent that the analytical method adopted would have to be capable of being used for large numbers of samples. Because of the seasonal nature of the condition being studied, most of the field experimental work was carried out during a concentrated period in the spring months and, as the work proceeded, there were many occasions when 200 - 300 blood samples were being taken every week. Therefore a rapid but accurate analytical method was necessary. The question of accuracy was obviously of importance and, when choosing an analytical method, it must be kept in mind that some methods may be capable of great accuracy in the hands of a skilled research worker where large numbers of samples are not being handled. Such a method may lose its precision when executed on a large scale by various members of the staff of the average chemical laboratory. Thus a suitable analytical method as well as being rapid and accurate must have a degree of what one might term "robustness" or reliability for routine purposes.

The classical methods of estimating calcium and magnesium concentrations in blood sera are that of Kramer & Tisdall (1921) for calcium and that of Fiske & Subbarow (1925) for magnesium. The calcium is precipitated as the oxalate and titrated with potassium permanganate and from the filtrate of this procedure magnesium is precipitated as magnesium ammonium phosphate, on which phosphate is determined colorimetrically. This is a lengthy procedure as it includes many manipulations of the material. It can be used satisfactorily as a batch process and will yield accurate results when rigorous attention is paid to procedural detail (Walker & Moodie, 1961). In contrast, Holtz & Seekles (1952) quote the case of serum samples analysed in different laboratories for calcium being inaccurate in 70% of the cases, the extreme variation ranging from one third to twice the correct calcium concentration.

There have been many methods described as alternatives to this classical procedure. Two which are widely used are the Titan Yellow method of Cornfield & Pollard (1950) (suitably modified) and an E.D.T.A. titration method of which there are many variations (e.g. Smith, 1955; Friedman & Robin, 1955; Levine & Cummings, 1956). Both these methods have the advantage of relative simplicity, rapidity and reliability. The Titan yellow method involves prior precipitation of the plasma protein followed by a final colorimetric measurement of the red Titan Yellow-magnesium complex developed in alkaline solution. The E.D.T.A. method, on the other hand, relies on the ability of the organic acid ethylene - diamine - tetra - acetic acid to complex cations such as magnesium and calcium in solution. By adjustment of the pH and the use of suitable indicators sensitive to the presence of calcium and magnesium, two separate determinations can be made

(a) a direct titration of 1 ml serum with E.D.T.A. at between pH10-11 using eriochrome black T indicator to give total bivalent ions (which, in serum, is virtually calcium plus magnesium) (b) a direct titration of 1 ml serum with E.D.T.A. at pH13-14 using murexide indicator to assess calcium alone.

These two titration figures are subtracted to give a titration figure for magnesium concentration. The main modification made in this work as compared with previously described procedures was the use of a hot solution for the first titration. This considerably speeds up the reaction rate which otherwise tended to be extremely slow. Both titrations were made on an E.E.L. titrator unit coupled to a galvanometer, thus giving a direct alteration in reading with colour change.

The E.D.T.A. method has the advantage of being capable of determining calcium and magnesium in more or less the one procedure without the need for serum pretreatment. Therefore, providing the accuracy and reliability were comparable to the alternative methods, it would be the most suitable one for use in the studies which were envisaged.

Ten samples were analysed for magnesium by both the E.D.T.A. and the Titan Yellow methods and for calcium by both the oxalate and the E.D.T.A. methods. The results of these comparative analyses are given in Table 5.

Table 5. Plasma Mg and Ca concentrations (mg/100 ml).

Analytical	Magnesium.		Calcium.	
Method.	Titan Yellow.	E.D.T.A.	E.D.T.A.	Oxalate.
sample No. 1.	2.80	2.92	10.73	10.94
2.	3.45	3.28	9.27	9.33
3.	2.95	3.19	10.24	10.10
4.	2.70	2.91	10.08	9.85
5.	2.70	2.60	9.60	9.71
6.	2.50	2.46	11.21	10.65
7.	2.75	2.61	10.32	10.54
8.	2.45	2.43	9.35	8.92
9.	3.45	3.24	11.77	12.18
10.	2.65	2.56	10.56	10.44
mean	2.84	2.82	10.31	10.20

Duplicate samples were also carried out on twelve serum samples by the E.D.T.A. titration method. The results are given in Table 6.

Table 6. Duplicated analyses of 12 plasma samples for magnesium.

sample No.	plasma Mg. (mg/100ml).	
1.	1.85	1.90
2.	1.80	1.90
3.	1.60	1.60
4.	1.65	1.50
5.	1.50	1.45
6.	2.25	2.30
7.	1.75	1.70
8.	1.40	1.50
9.	1.70	1.70
10.	1.60	1.60
11.	0.90	0.95
12.	0.60	0.55
mean	1.55	1.55

To test the recovery of added magnesium 1ml of a standard solution, containing 2 mg Mg/100ml, was added to samples no. 10 and 12 given in table 6. The results were as given in Table 7.

Table 7. Recovery of added magnesium in serum.

		Mg mg/100 ml.			
sample no.	plasma alone.	added.	found.	recovered.	% recovery.
10.	1.60	2.00	3.65	2.05	102.5
12.	0.57	2.00	2.60	2.03	101.5

From the data given in Tables 5, 6 and 7, for comparisons, duplication of results and recoveries, use of E.D.T.A. would seem an accurate and reliable analytical method.

It was, however, important to know how this method would compare with others in large scale use. As a further check, therefore, the samples from experimental work conducted over the period September, 1960 - July, 1961 were subjected to analysis by both the E.D.T.A. titration method and the Titan Yellow colorimetric method. For one series of 743 samples a Titan Yellow method using gum ghatti as a colloid stabiliser was employed and in a second series of 103 samples, polyvinyl alcohol was used as the colloid stabiliser. Table 8(a) gives the extent of agreement for the first series and Table 8(b) for the second series.

Table 8.

	(a)			(b)		
	No. of samples	% of samples		No. of samples	% of samples	
agreement within 0.00-0.1mg/100ml	308	41%		81	50	
" " 0.11-0.2 " "	227	31%	90%	51	31	96%
" " 0.21-0.3 " "	135	18%		24	15	
" " 0.31-0.4 " "	33	5%		6	3	
samples disagreeing more than 0.41 " "	35	5%		1	1	

Any analyses which disagreed by more than 0.3 mg Mg/100ml were repeated. It was found that the E.D.T.A. method was wrong in 35% of the samples repeated, the Titan Yellow method in 51% and both methods were wrong in 15% of the occasions. There was found to be no positive bias from either method in that the overall mean values were the same.

The Titan Yellow method employing gum ghatti gave poorer comparative results than the second method using polyvinyl alcohol and, therefore, taking the second series in (b) as the more accurate comparison, the results indicate that 96% of the samples agreed to within 0.3 mg Mg/100 ml. i.e. less than ±.15 mg/100 ml from the mean of the two figures. One point not brought out here is the possibility of the Titan Yellow method giving large batch errors where every result in a particular batch is depressed or alternatively raised. The E.D.T.A. method, though capable of giving occasional individual errors, is not subject to these 'batch' errors.

It would seem, in conclusion, that although both methods agree reasonably closely with each other, on balance the E.D.T.A. method gave slightly more accurate results. It has the added advantages, as previously mentioned, of requiring no plasmapretreatment and of simultaneously estimating plasma calcium. The E.D.T.A. method was therefore used throughout this programme of work for the analysis of calcium and magnesium.

Plant Material.

Preparation of samples.

Fresh grass samples were dried overnight in a forced draught oven at 90°C. The dried grass was then ground in a Christie and Norris laboratory mill and these ground samples were used in the following analytical procedures. Analyses were made on grass samples for magnesium, calcium, potassium, sodium, phosphorus and manganese. One gram of dried ground material was dry ashed and made up to 100 ml solution. Aliquots of this were used for the following procedures:-

Magnesium. The method of Cornfield & Pollard (1950) was employed. It is very similar in design and identical in principle to that described for serum magnesium analysis by the Titan Yellow complex method. A compensating solution containing calcium, phosphorus and aluminium is used to mask the interfering effect of these ions, and a mannitol solution is added to mask the effect of manganese.

Calcium. The procedure by Hemingway (1956) was used. The principle is based on a final flame photometric measurement of calcium in the plant solution after the removal of phosphate interference by a cation exchange resin.

Sodium and Potassium. were estimated on direct dilutions of the plant ash solution using the E.E.L. flame photometer.

Phosphorus was determined colorimetrically by a modification of the classical method of Fiske & Subbarow (1925) which relies on the blue colour developed by ammonium phospho-molybdate in the presence of a reducing agent.

Manganese was estimated in the plant ash solution, after removal of chloride which interferes, by oxidation of manganese present in acidified solution to potassium

permanganate. The colour developed can be measured photometrically.

Those methods of plant analysis described are all accepted standard methods of analysis known to give reliable and consistent results.

Section II. Experimental Work Part I.

Hypomagnesaemia and Hypomagnesaemic Tetany

in Ewes as influenced by

(a) pasture fertilisation

(b) number of lambs suckled

(c) age of the ewe

(d) breed of the ewe

These factors will first be reviewed in relation to the experiments which were carried out.

Factors involved in the development of hypomagnesaemia and hypomagnesaemic tetany in sheep have received little attention from research workers in comparison to the vast volume of work carried out in cattle. After the first report of hypomagnesaemic tetany by Stewart (1951), the subsequent papers of Barrentine & Morrison (1953), O'Moore (1955), Penny & Arnold (1955) and Pook (1955) were purely reports of naturally occurring outbreaks of tetany in sheep. It is noteworthy that three of these authors (Barrentine & Morrison, Penny & Arnold and Pook) comment particularly on the outbreaks occurring immediately after the ewes had been moved onto recently established ley pasture. In the report of O'Moore (1955), the sheep were also being rotationally grazed on similar leys. Furthermore, Pook (1955) reported that the outbreak was halted by moving the sheep back to older grass pastures. Watt (1960), considering cases of sudden death in sheep, concluded that hypomagnesaemic tetany cases were much more common on young highly manured pastures. If there is indeed an association between the development of tetany and movement of a flock onto young pastures, it would have to be established whether the cause was purely the movement of the flock involving a change of diet, or alternatively being due to the fresh pasture or, again, to the particular fertiliser treatment of the pasture. Hughes & Kershaw (1958) and Michael (1953) reported on metabolic disorders associated with the movement of hill sheep from hill ground onto lowland pasture, prior to lambing. Investigation showed the presence in the affected ewes of abnormally low serum calcium values and a normal or slightly lowered serum magnesium. This type of outbreak must therefore be considered as distinct from hypomagnesaemic tetany both on the grounds of the different

blood picture presented and on the timing of the outbreaks. Michael (1961), over a period of three spring seasons, investigated the magnesium status, as measured by blood samples, of sheep with three week old lambs running on either improved pasture or unfertilised permanent pasture. He concluded that the mean serum magnesium of sheep on the improved ground was significantly lower than that of the other group, although the mean serum magnesium concentrations never fell below 1.70 mg/100 ml. Individual ewes had lower values and on the improved pasture one case of hypomagnesaemic tetany out of a group of twenty-one ewes occurred in the first year. In the second year, two sudden deaths occurred in a group of nineteen ewes on the improved pasture. Hughes (1958) also stressed the importance of movement of ewes onto young green pasture or cereal crops. Hemingway et al. (1960), as a result of some preliminary work on the relationship between pasture fertilisation and hypomagnesaemia in ewes, found no direct effect attributable to the use of nitrogenous and potassic fertilisers either alone or in combination. The mean plasma magnesium concentrations of all groups seldom fell below the normal value of 2.0 mg Mg/100 ml. What was apparent from this paper was the great variation between individuals in plasma magnesium concentrations on the same treatment. Where only small numbers of experimental animals are employed, the real effects of a variety of treatments may be concealed by large errors introduced by individual variations. Further work would therefore need to employ sufficient animals in replicated treatments to yield any statistically valid results.

The use of high rates of nitrogen and potassium fertilisers on grassland, inevitably results in a higher intake of potassium and nitrogen by the grazing sheep. To investigate whether either or both of these elements per se would

influence plasma magnesium values, several workers have given increased nitrogen and potassium intakes to sheep in conjunction with blood magnesium measurements.

Pearson et al. (1949), Odell et al. (1952) and Eaton & Avampato (1952) gave varying quantities of potassium chloride or potassium bicarbonate to sheep on indoor diets and failed to find any reduction in blood magnesium. In contrast, Kunkel et al. (1953) added 5% potassium as potassium bicarbonate to the diet of adult sheep and plasma magnesium was significantly reduced from 2.68 to 1.78% mg Mg/100 ml. Feed intake was found to be reduced on this high potassium diet. Christian & Williams (1960) found that additions of ammonia to the diet of sheep as urea or ammonium acetate had no effect on serum magnesium levels.

At the time the work to be described was initiated, this was the extent of the published information on the effect of pasture improvement on hypomagnesaemia and tetany in sheep, and it was felt that it would be of value to attempt to clarify the extent to which the type of pasture might be involved, and in particular the influence of fertiliser treatment on the magnesium status of the ewe, since many workers in experiments with cattle have incriminated potassic fertilisers either alone or in combination with nitrogen as causal agents in hypomagnesaemia in that species.

For example, Bartlett et al. (1954;1957) applied 4 cwt of potassium sulphate/acre and found no hypomagnesaemia in cattle in the first year. In the second year of the experiment (when the fertiliser application was repeated) a hormone treatment was used to suppress the increased clover growth, and three out of four cows had plasma magnesium levels below 1.0 mg/100 ml. Smyth et al. (1958) found that neither 3 cwts of muriate of potash/acre or 5.5 cwt of nitrochalk/acre

influenced the plasma magnesium levels of dairy cows but a combination of these two treatments induced severe hypomagnesaemia (mean 0.7 mg Mg/100 ml.) and hypomagnesaemic tetany in two of the three cows on this pasture. Kemp (1958 a) used potassic fertiliser applications as follows:- 270 lbs K_2O/acre in spring and 135 lbs K_2O/acre in the autumn of 1956, and in the following year 180 lbs K_2O/acre was given followed by 90 lbs in the autumn. Clover was suppressed by a hormone weed killer. There were no cases of tetany in the first year but in the second year, several cows on the high potassium plot were affected. Hvidsten et al. (1959) treated herbage with 160-187 lbs K_2O/acre in each of three successive years, in combination with high nitrogen application. There were no cases of tetany in the first year, and one case in each of the next two years on the high potassium plot as well as lowered blood magnesium concentrations. In contrast Storry (1961 b) reported no increased incidence of hypomagnesaemia in cattle grazing plots fertilised with 7 cwt of muriate of potash and 10 cwts of sodium nitrate or 7 cwts of ammonium sulphate.

To some extent, it is difficult to evaluate much of this work because of the unrealistically high fertilisation rates which were used. Furthermore potassium fertilisation normally encourages clover growth in a sward and because of the higher magnesium content of clover it would thereby increase the magnesium content of the sward. In at least two of the reports (Bartlett et al., 1954; 1957; Kemp, 1958 a) where potassic fertilisation was shown to induce hypomagnesaemic tetany, the enhanced clover growth resulting from potassium application was deliberately suppressed, by a hormone weed killer. This interference with the normal course of events introduces a bias on the part of the investigators and

to some extent invalidates the result as far as a direct potassium effect is concerned. This would, of course, not apply if the pasture was initially a pure grass sward. Despite these criticisms, there is sufficient evidence to show that potassium and nitrogen fertilisation of grassland do tend to increase the risk of hypomagnesaemic tetany occurring in cattle. This tendency may be severe when excessively high rates of fertiliser use are employed. The influence of more reasonable rates of use is more uncertain.

In Experiment I of this present work, moderate and high applications of potassic fertiliser were used in combination with moderate applications of nitrogen. The particular rates employed were thosewhich might reasonably be used under practical farm conditions. The experiment also employed sufficient experimental animals and treatment replication to allow proper statistical examination of any possible fertiliser effects.

Another facet of the previous studies with cattle on the effect of potassium fertilisation is that in several experiments, no adverse effect attributable to the use of high rates of potassium and nitrogen fertiliser usage was recorded in the first year. However, hypomagnesaemia and hypomagnesaemic tetany were found in the second year when the treatments had been repeated. This possibility, coupled with the varying incidence of hypomagnesaemic tetany which can be found in different years, made it desirable to repeat the fertiliser treatments used in Experiment I in succeeding years. Consequently, Experiments 2 and 3 deal partly with the effect on the development of hypomagnesaemia in the ewe of continued potassium fertiliserusage in grassland for a second and third year respectively.

Number of suckling lambs.

A complicating factor in studies on the development of hypomagnesaemia in ewes may be the influence of the number of lambs which are being suckled by the ewe.

Barrentine & Morrison (1953), reporting on grass tetany outbreaks in ewes grazing oat forage, comment on ewes with twin lambs being more susceptible than those with singles. Ewes with no lambs were unaffected. Penny & Arnold (1955) described an outbreak of clinical tetany in a flock of eighty where all three affected ewes had twin lambs.

Again in the report of Pook (1955) the eleven ewes lost from a flock of two hundred and eighty ewes from hypomagnesaemic tetany all had twin lambs. Hughes (1958) quotes an unpublished experiment of Williams where, in a flock of ewes which had been grazing wheat followed by a long fast, the eighteen ewes which died from hypomagnesaemic tetany all had twin lambs. Herd and Peebles (1962) reporting on grass tetany in sheep in Australia comment on the greater incidence in ewes with twin lambs.

There is therefore considerable evidence to show that in these field outbreaks, ewes with twin lambs at foot are much more susceptible than ewes with single lambs which in turn are more susceptible than ewes with no lambs. Indeed there are no reports of hypomagnesaemic tetany in non-lactating ewes under natural grazing conditions. Experimental evidence to confirm these clinical reports, however, is lacking. Inglis et al. (1959) found that ewes with triplets had considerably lower plasma magnesium levels than other groups with fewer lambs, but on the other hand, they found little difference between groups of sheep with

none, one and twin lambs. Hemingway et al. (1960) whose experiment on pasture fertilisation treatments included equal numbers of ewes with lambs and ewes with no lambs, found no difference in mean plasma magnesium levels between these two groups as a whole, but they did find a lower mean plasma magnesium in the old ewes with lambs as compared with old ewes with no lambs.

The main differences between a ewe with twin lambs as compared with single lamb ewes would be a greater demand for nutrients during pregnancy and presumably a higher milk yield after pregnancy. Inglis et al. (1959) found non-breeding ewes to have higher mean serum magnesium levels. With regard to the milk yields, it is of interest to consider this in conjunction with the timing of those various reported outbreaks of hypomangesaemic tetany in relation to lambing date.

Author.	interval after lambing before cases developed (weeks).
Barrentine & Morrison (1953)	3
O'Moore (1955)	2
Pook (1955)	2
Penny & Arnold (1955)	4
Michael (1961)	4
Herd & Peebles (1962)	2-6

It will be observed that all the above reported cases therefore appear within the close range of 2-6 weeks after lambing which, as Spedding (1965) points out, is the time of peak lactation for ewes.

It may be, then, that the susceptibility of a ewe to hypomagnesaemic tetany is roughly proportional to its milk yield. This would not, however, agree with the findings of published work in cattle. (Blaxter & McGill 1956). It was felt that

this point was worthy of further investigation in Experiment I.

Age of the ewe.

There is little information in the literature concerning the relationship of age of the ewe to the development of hypomagnesaemic tetany. In the outbreak described by Pook (1955) seven of the affected ewes were 2-years-old, two were 3-years-old and a further two were over 4 years old. O'Moore (1955) reporting on a similar outbreak in Ireland states that all affected ewes, apart from one gimmer, were 3 years old. In contrast, Hemingway et al. (1960) found that 5-year-old ewes had lower plasma magnesium values (mean 1.70 mg Mg/100 ml.) over a two month spring period than 2 year old gimmers whose mean plasma level was 2.22 mg/100 ml.

Care must be taken in comparing these contrasting results since Pook (1955) and O'Moore (1955) were both reporting outbreaks of hypomagnesaemic tetany whereas Hemingway et al. (1960) were dealing only with hypomagnesaemia. These are certainly not synonomous but the presumption is that the greater the degree of hypomagnesaemia then the greater will be the risk of clinical tetany ensuing. Whether this presumption is correct is difficult to say but in the absence of clinical cases appearing in an experiment, then plasma magnesium concentrations are, at the present time, the only sensitive indicator which can be measured of the magnesium status of the animal.

Susceptibility of cattle to hypomagnesaemic tetany has been shown to increase with age (Blaxter & McGill 1956) and it would be surprising if cattle and sheep differed in this respect, particularly since the increasing risk of hypomagnesaemic tetany with advancing age in cattle is presumed to depend largely on the

decreasing mobility of magnesium in the skeleton. It is reasonable to expect that the same basic physiological mechanism might operate in sheep.

Further study of the effect of age in sheep in relation to hypomagnesaemia therefore seemed to be desirable and part of Experiment I was designed to allow the influence of this factor to be closely studied.

Breed of the ewe.

There have been no reports in the literature of any particular breed susceptibility in relation to the development of hypomagnesaemic tetany. The various outbreaks for which details have been published have occurred in a variety of breeds, for example Border Leicester (Pook 1955); Cheviot (O'Moore 1955); Welsh Mountain (L'Estrange & Axford 1964 a); Half-Bred (Penny & Arnold 1955; Herd & Peebles 1962); and Scottish Blackface (Inglis et al. 1959).

The consideration of these isolated outbreaks, of course, cannot be in any way comparative and it was therefore felt that it would be useful to investigate the possible existence of any difference in breed susceptibility with regard to tetany. Since hypomagnesaemic tetany can be a big problem of lowland farming as distinct from hillland, Cheviot ewes and Half-Bred ewes were chosen for this comparison as being the two most popular lowland breeds in the West of Scotland.

Experiments 1, 2 and 3 were concerned with investigations made on the same experimental site at Cochno Farm, Hardgate, Dumbartonshire, during the spring seasons of 1961, 1962 and 1963 respectively. The site was laid out in 1961 as a series of eighteen plots in a randomized block arrangement of six replicates of three pasture treatments (see figure 1). The plots were 0.4 acre in size and provided grazing for four ewes per plot over the experimental periods. The potassium fertiliser treatments given in the first year were repeated, apart from some slight modifications, in the second and third years.

In Experiment 1 and 3 data were also collected on the effect of the age of the ewe and the number of lambs suckled by the ewe on the development of hypomagnesaemia. In experiments 2 and 3, the influence of breed differences was investigated. Since some of the sheep involved in Experiment 1 were used again in Experiments 2 and 3, it was possible to make a comparison on the development of hypomagnesaemia in the same sheep between two and three successive years. Each of these three experiments also included a study on the prophylactic effect of one of three different methods of magnesium supplementation to the ewe.

Since Experiments 1, 2 and 3 are interrelated, after dealing with each one separately, a brief general conclusion will be given at the end to collate and compare the results from all three years.

Experiment 1.

Experimental Design.

Seventy-two pregnant Cheviot sheep, thirty-six of which were gimmers and thirty-six of which were 5-year-old ewes, were purchased in early February 1961

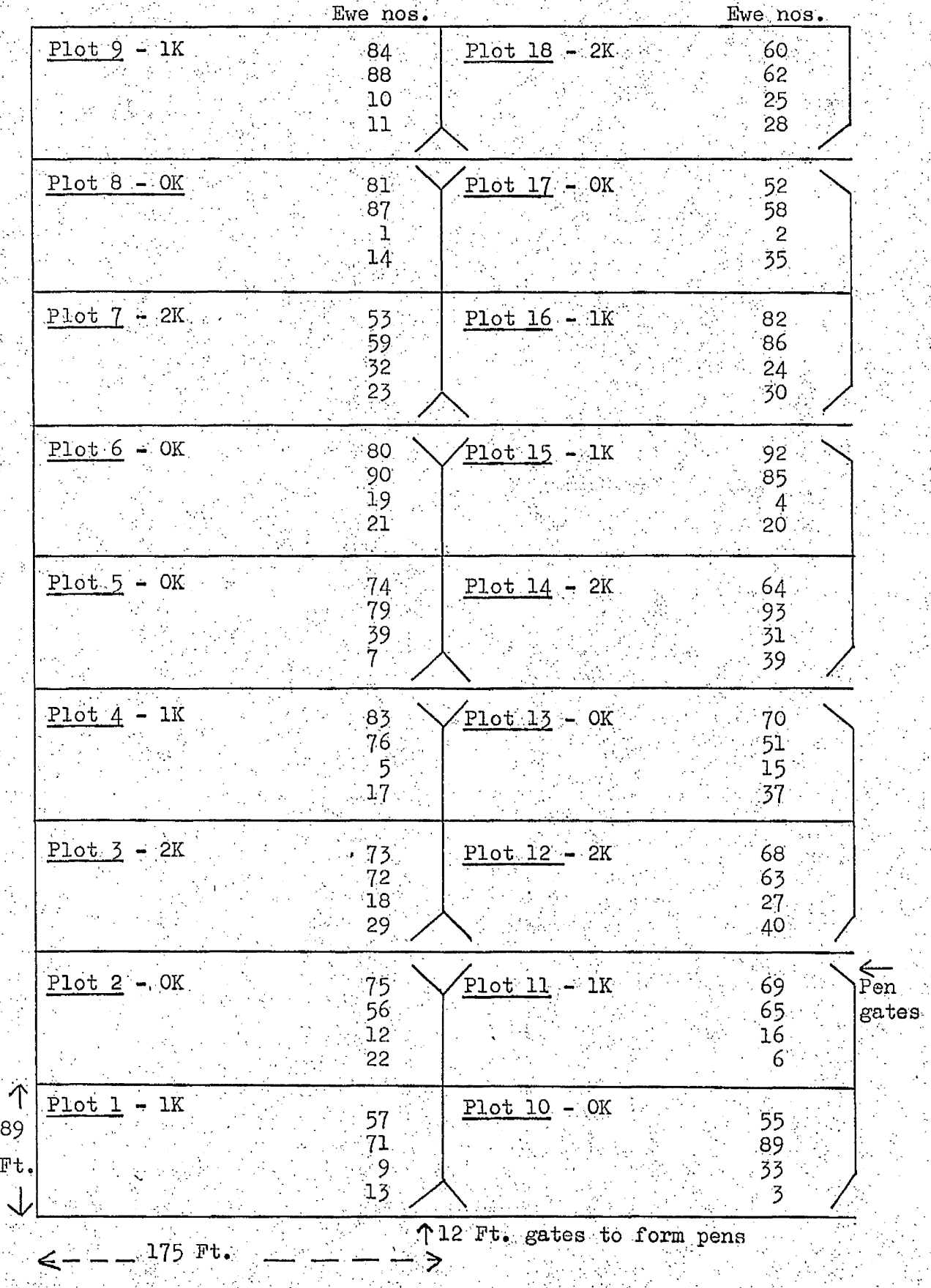
and kept on twenty-five acres of permanent grass. From 14th March onwards they received supplementary concentrate feed at the rate of 1 lb. per head per day, although older ewes tended to eat rather more than the gimmers. This concentrate food contained 0.24% Mg. Lambing commenced on 6th April. Over this preliminary period, blood samples were taken from the flock on seven occasions between 21st February and 19th April. On 19th April, the sheep were transferred to the experimental plots and supplementary feeding was stopped on that day. Forty-one of the sheep had lambed by that day and lambing was complete by the end of April. The exact lambing date and the number of lambs born to each ewe are given in Appendix 2a. Twenty-five of the ewes had twin lambs, the remaining forty-seven having a single lamb.

The sward of the experimental area was a two-year old ley pasture and consisted mainly of ryegrass with some cocksfoot. There was no clover. A uniform dressing of 3 cwts/acre of nitrochalk (21%N) was applied to the whole area on 2nd March.

There were eighteen individual experimental plots of 0.4 acres arranged in six blocks of three plots. Three potassium treatments 0, 1, and 2 cwts per acre of muriate of potash (60% K_2O) were applied at random to the three plots in each block on 2nd March. Two old ewes and two young ewes were allocated at random to each plot. There were therefore twenty-four sheep grazing each of the three fertiliser treatments, half of each group being old ewes and half being young ewes. A plan of the experimental area is shown on Figure 1, giving the plot dimensions, the layout of the pens and the potassium fertiliser treatment given to each plot. The ear tag numbers of the ewes allocated to each plot are also given. Any number within the series 51-93 represents a five-year-old Cheviot

Fig. 1 Layout of experimental plots at Cochno farm (Experiment 1).

0K - No potassium fertiliser applied.
1K - 1 cwt muriate of potash/acre applied.
2K - 2 cwt muriate of potash/acre applied.



ewe and numbers within the series 1-40 represent the young two-year-old gimmers. The distribution of ewes with twins and ewes with single lambs among the experimental plots was random.

A prophylactic treatment was also superimposed on this experimental design. This consisted of two magnesium heavy pellets given to one old ewe and one gimmer in each plot. Therefore, a total of thirty-six ewes, eighteen from each age group received this treatment, the remaining thirty-six ewes acting as controls. Consideration of this factor will be omitted at this stage and dealt with in Section IV. Since the ewes given the magnesium treatment were spread equally among the different fertiliser treatments and between the age groups, consideration of the mean values of these latter factors will not be affected by the magnesium treatment.

Blood samples were taken from all the sheep on seven occasions during the course of the experiment (19th April - 29th May). Herbage samples were taken on four occasions from each plot between mid April and late May. These samples were collected by plucking the grass from at least twenty-five points chosen at random in the plot, to give a bulk sample of about 200 gms fresh wt of grass. There was a good growth of grass before the sheep were transferred to the plots and the grazing could therefore be described as lush spring pasture. Grass growth after the sheep were moved to the experimental area kept pace with the grazing rate of the flock, and consequently at no time were any of the ewes on bare pasture, although all plots were well grazed down by the end of the experiment. A further herbage sample was taken from each plot in mid August. The herbage samples were analysed for magnesium calcium, sodium, potassium,

phosphorus and manganese.

Results.

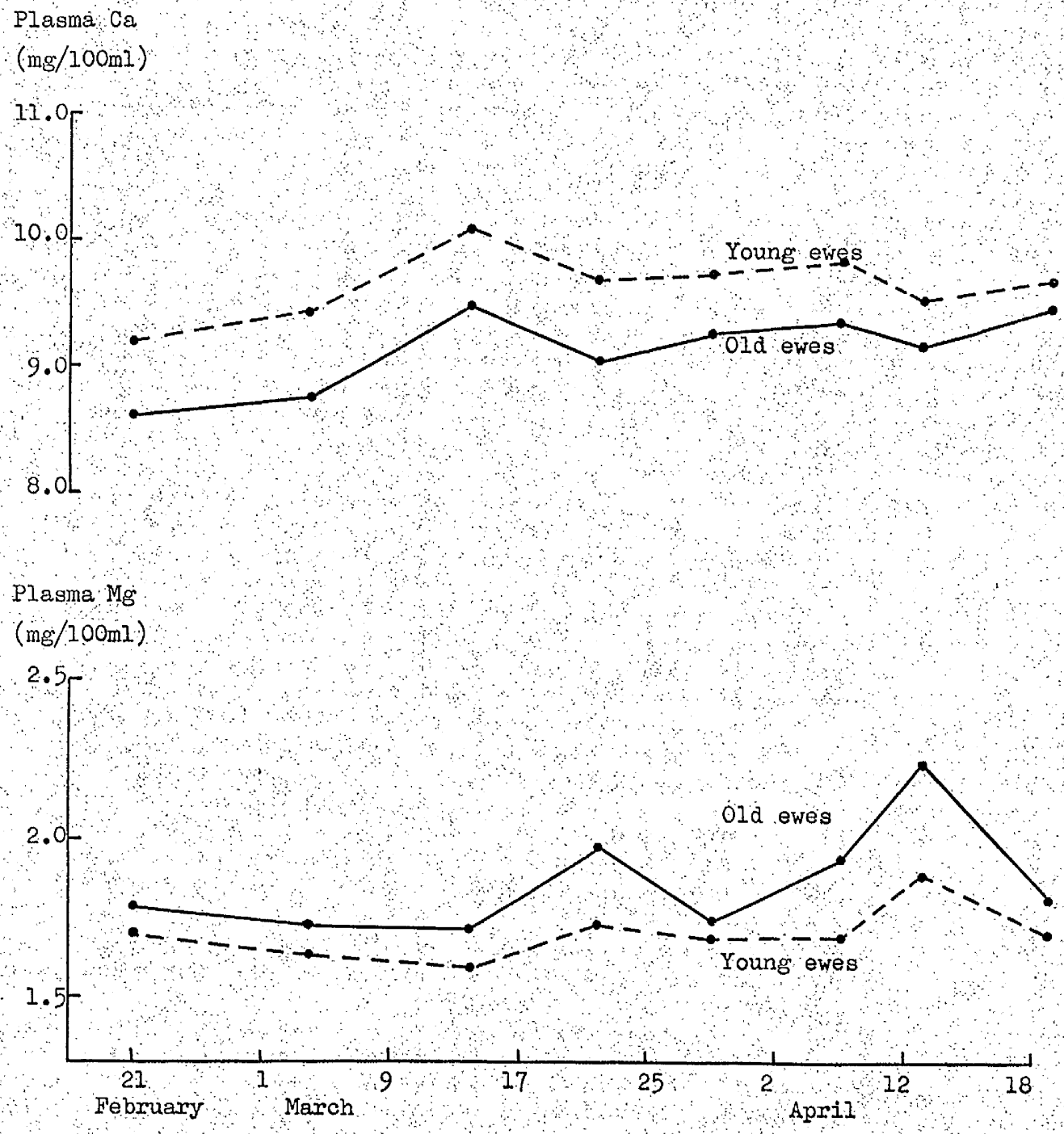
(a) Preliminary period on permanent pasture with supplementary feeding.

Figure 2 shows the mean plasma calcium and magnesium concentrations for the groups of thirty-six old and thirty-six young ewes during the period (21st February - 19th April) of grazing permanent pasture prior to transferrance to the experimental area. The individual plasma magnesium and calcium values for each ewe on each sampling date are given in Appendix 2b. There was little change in mean plasma magnesium levels over this period, but the old ewes had levels of around 1.8 mg/100 ml which were consistently about 0.15 mg/100 ml above that of the gimmers. Also the mean plasma magnesium level of the old ewes rose more sharply after the beginning of April. This may perhaps have been due to their higher intake of supplementary food as noted above. Plasma calcium values increased slightly for both groups over this period from an average of 8.9 mg/100 ml to 9.5 mg/100 ml and the young ewes had levels which were consistently about 0.5 mg Ca/100 ml higher than the old ewes.

Three of the young ewes and five of the old ewes had plasma magnesium values below 1.0 mg/100 ml on at least one sampling occasion prior to 19th April. There were no abnormally low plasma calcium values during this period.

Re-examination of the data for the individual plasma calcium and magnesium values for the sheep which were subsequently allocated to plots given different amounts of muriate of potash, showed that they had closely similar mean values throughout this preliminary period.

Fig 2. Mean plasma calcium and magnesium concentrations of 36 old and 36 young ewes prior to the experimental grazing period of Experiment 1.



Changes during the grazing of the fertilised plots.

Figure 3a shows the mean plasma magnesium and calcium concentrations over the experimental period of 19th April to 29th May for the three groups of twenty-four sheep grazing the 0, 1 and 2 cwts of muriate of potash/acre treatments. Figure 3b gives mean values rearranged into the two groups of thirty-six old and thirty-six young ewes. The individual plasma calcium and magnesium values of the seventy-two sheep and the appropriate mean values for the seven sampling occasions over this experimental period are given in Appendix 2c.

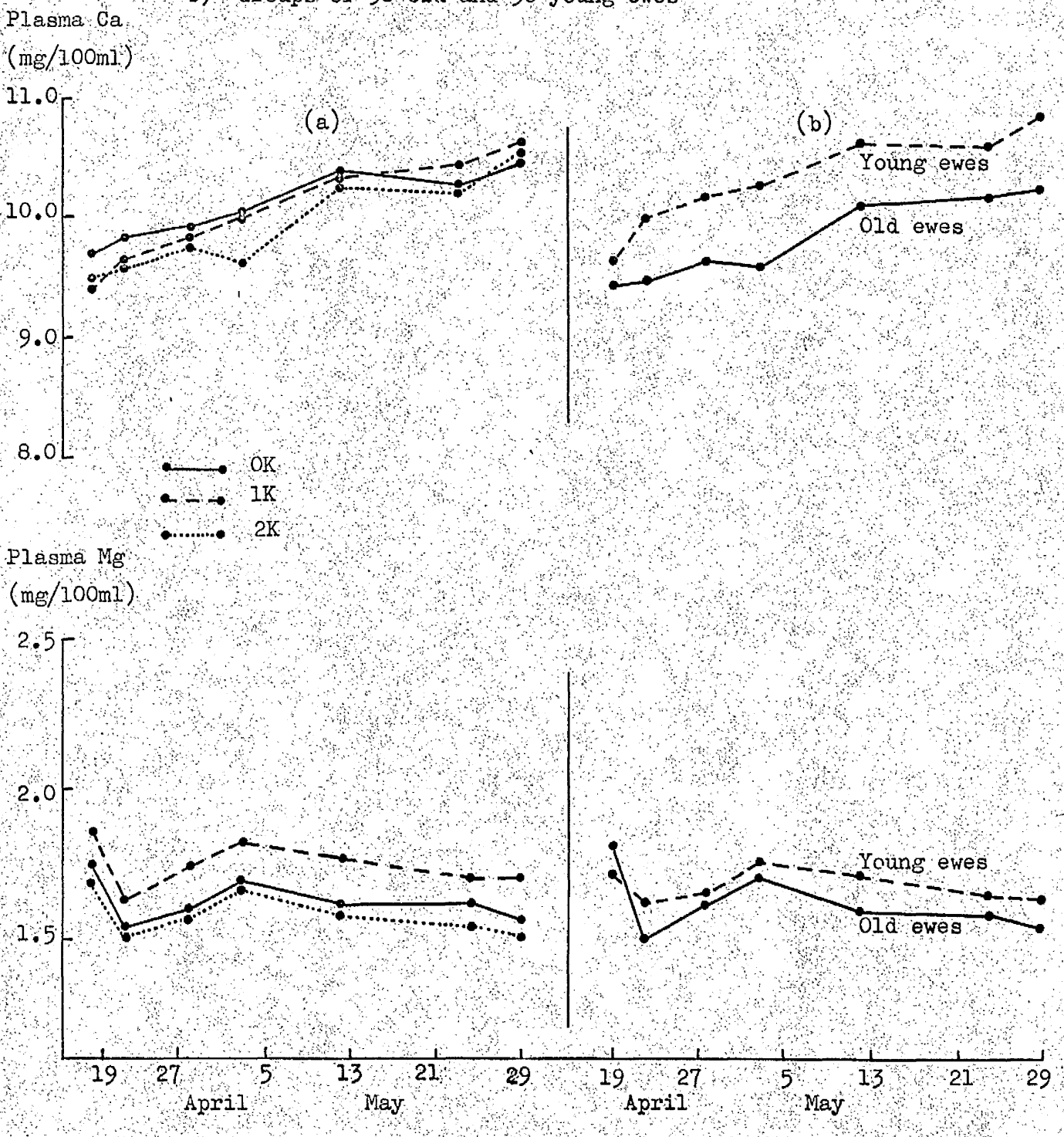
After a small initial fall over the first three days, from 1.76 mg/100 ml to 1.56 mg/100 ml, the mean plasma magnesium concentrations of all groups remained substantially constant over the remainder of the period of the experiment, irrespective of treatment. It should be recorded that the flock had been moved in preparation for the trial to pasture adjacent to the plots one day before the experiment started on 19th April. In retrospect, it might have been better if this move had not been made since it had the effect of allowing a fall in the mean plasma magnesium level of the flock before the exact time of pasture change as evidenced by a comparison between the mean plasma magnesium values on the 13th and 19th April. The initial fall in plasma magnesium over the first three days was therefore less severe than it otherwise might have been.

The mean figures for plasma magnesium conceal the fact that between 19th April and 29th May, a total of sixteen sheep had values below 1.0 mg/100 ml on at least one sampling occasion. This level is commonly considered to be subnormal and below this figure clinical cases of tetany may be encountered. These sixteen sheep included the eight animals which had values below 1.0 mg/100 ml in the

Fig. 3 Mean plasma calcium and magnesium concentrations of 72 lactating ewes at grass (Experiment 1). Arranged as -

a) 3 groups of 24 ewes on plots given 0,1 and 2 cwts of muriate of potash/acre.

b) Groups of 36 old and 36 young ewes



preliminary period. These low plasma magnesium animals were evenly distributed among the three fertiliser treatments and between the two age groups.

There was one case of clinical tetany. This occurred on 26th April and the sheep (no. 70) was an old ewe with twins grazing a K0 plot. The ewe was found recumbent but responded within a few minutes to an injection of magnesium sulphate. It again showed symptoms thirty minutes later and death ensued. Subsequent analysis of a blood sample taken prior to administration of magnesium showed it to contain 0.46 mg Mg/100 ml and 4.0 mg Ca/100 ml. These concentrations of plasma magnesium and calcium on earlier dates were:-

Table 9. Plasma magnesium and calcium concentrations of ewe No. 70.

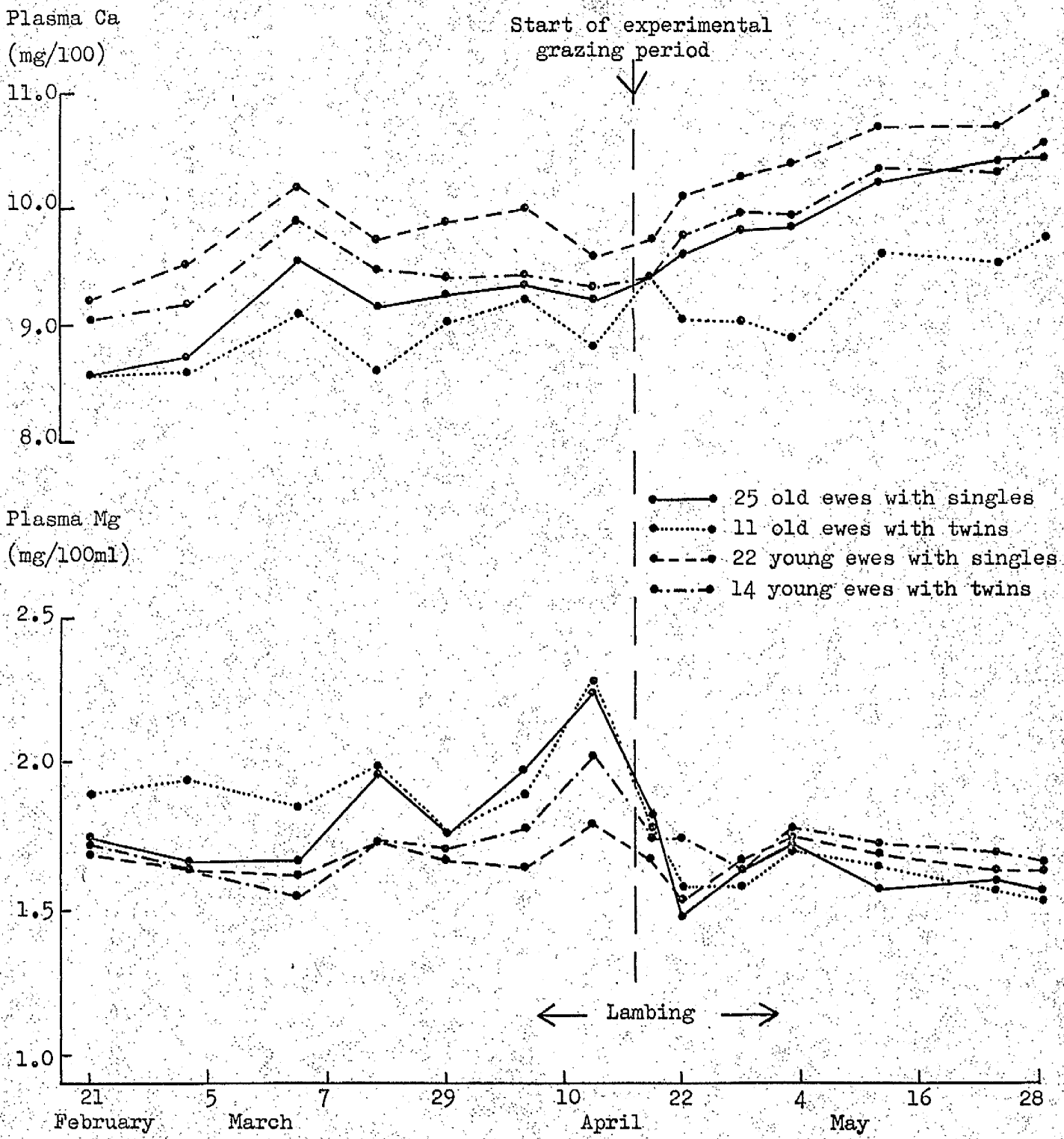
	13th April	19th April	22nd April
ewe No. 70:- Mg(mg/100 ml)	2.7	0.48	0.45
Ca(mg/100 ml)	7.8	9.5	6.8

The ewe lambed on 22nd April.

The mean plasma calcium concentrations of all groups increased steadily over the experimental period from about 9.5 mg/100 ml on 19th April to about 10.5 mg/100 ml on 29th May. There was no apparent effect attributable to fertiliser treatment but the old ewes continued to have a lower mean plasma calcium level than the young ewes.

The analytical data can be rearranged into groups of sheep with twin lambs and sheep with single lambs. The mean values of these groups are given in Appendix 2(d) along with mean values for a further subdivision of these groups into old ewes with single and twin lambs and young ewes with single and twin lambs. These latter means are shown graphically in Fig. 4.

Fig. 4 Mean plasma calcium and magnesium concentrations of 72 lactating ewes at grass (Experiment 1). Arranged according to age and number of lambs.



Statistical Examination of the plasma calcium and magnesium results.

Statistical examination of the results to assess the effects of the various treatments involved the fitting of regression lines for individual sheep to different phases of the experiment. In the first instance, regressions were fitted for blood samples taken between 14th March and 19th April, during which period the sheep were grazing permanent pasture and receiving supplementary food. This enabled 'smoothed' estimates to be made for the plasma calcium and magnesium values of each ewe on 19th April, the date of transfer to the fertilised plots. In a similar manner, regressions were fitted for the period 28th April to 29th May when the sheep were grazing the differently treated plots. The values for 22nd April were omitted as the sheep were likely to be in an unsettled condition. This second set of regressions allowed "smoothed" values for plasma calcium and magnesium to be estimated for 28th April. It was thus possible to assess the varying falls in plasma calcium and magnesium for the separate groups of sheep during the first 9 days of grazing the fertiliser plots. Table 10 details the mean falls in plasma magnesium levels over this period together with the various standard errors. There were clearly no significant differences for any of the treatments other than the fact that old ewes experienced significantly greater falls in plasma magnesium concentration over this transitional period than did young ewes.

Table 10 also shows the comparable data for the mean increases in plasma calcium levels over the same period. There were no statistical differences between treatments.

Table 10. Mean changes in plasma magnesium and calcium over a 9 day period 19-28 April together with relevant standard errors.

(a) Magnesium - mean falls.	K0	K1	K2	Mean
Young sheep	0.11	0.06	0.08	0.08
Old sheep	0.39	0.29	0.37	0.35

standard errors for:- K0, K1, K2 marginal means = 0.113
vertical comparisons within table = 0.153
all other comparisons within table = 0.156
age means (young V old) = 0.062

(b) Calcium - mean increases.	K0	K1	K2	Mean
Young sheep	0.40	1.05	0.44	0.63
Old sheep	0.47	0.26	0.16	0.30

standard errors for:- K0, K1, K2 marginal means = 0.246
comparisons within table = 0.347
age means (young V old) = 0.142

Herbage mineral contents.

The results of the analysis of the individual herbage samples for magnesium, calcium, potassium, sodium, phosphorus and manganese are given in Appendix 2(e). The mean levels of these minerals as influenced by the fertiliser treatments are given in Table 11. The results are averaged for all six replicate plots.

Table 11. Mineral contents of herbage samples from Cochno plots 1961.

Mean of six replicates (results on dry matter basis).

Element	Fertiliser Treatment	16th Apr.	29th Apr.	9th May	17th May	17th Aug.
Mg%	K0	.17	.15	.15	.17	.16
	K1	.16	.15	.15	.17	.15
	K2	.16	.14	.14	.16	.14
Ca%	K0	.65	.59	.40	.46	.63
	K1	.66	.56	.42	.50	.64
	K2	.65	.53	.40	.49	.66
K%	K0	2.8	2.7	2.9	2.9	2.9
	K1	3.2	3.2	3.3	3.2	3.0
	K2	3.3	3.3	3.6	3.5	2.9
Na%	K0	.24	.31	.33	.33	.23
	K1	.16	.23	.26	.26	.18
	K2	.16	.23	.25	.19	.13
P%	K0	.40	.39	.34	.37	.37
	K1	.41	.39	.35	.33	.35
	K2	.41	.40	.39	.39	.35
Mn(ppm)	K0	105	109	120	142	105
	K1	109	108	126	138	109
	K2	105	109	115	131	106

The magnesium content of the herbage from all three treatments, declined from about 0.17% Mg in mid-April to about 0.15% Mg in late April and early May. This mean level rose again to 0.17% at the end of May. The small treatment differences were not statistically significant.

Herbage potassium contents were increased by muriate of potash. Where no potassium was applied, the mean potassium content was 2.8-2.9%. Both 1 and 2 cwts per acre of muriate of potash increased the level significantly ($p = 0.01$) to 3.2 and 3.5%K respectively by mid-May. Samples taken at the end of August showed no treatment differences.

Sodium concentrations increased on all treatments from mid-April to late May. Both rates of application of muriate of potash significantly reduced ($p = 0.01$) the sodium concentration of the herbage. From mid-April to mid-May the sodium levels rose from 0.16 to 0.25% Na compared with an increase of from 0.24 to 0.38% Na where no potassium was applied.

Muriate of potash treatment did not affect the herbage calcium, phosphorus or manganese. There was a slight drop in calcium and phosphorus content and a slight rise in manganese content over the first three sampling dates. These changes are not considered to be of any importance.

Discussion.

Although the mean plasma magnesium levels of the groups in this experiment fell below 1.8 mg/100 ml, which is often taken as the lower limit of the normal range, the degree of hypomagnesaemia involved can only be described as "slight" although several individuals in various groups did develop a severe hypomagnesaemia (below 1.0 mg/100 ml).

Examination of the regression coefficients of the individual sheep over the period 28th April to 29th May showed that the plasma magnesium levels of all the groups of sheep were effectively constant at about 1.60-1.70 mg/100 ml and that there were no treatment differences. Over the same period, the plasma calcium values of all sheep increased steadily from a mean of 9.90 to 10.53 mg/100 ml but again there were no treatment differences in the rate of this increase.

At the rates of application used (1 and 2 cwts per acre) muriate of potash did not depress plasma magnesium levels significantly nor did it affect the plasma calcium concentrations. As far as the effect of these applications on the herbage is concerned, the 2 cwt per acre treatment reduced the mean herbage magnesium content by an average of 0.01% Mg over the 6 week period of grazing but this slight difference was not statistically significant. The effect of both fertiliser treatments of enhancing the potassium content and depressing the sodium content of the herbage are well known effects (e.g. Hemingway 1961) and were to be expected.

There was little difference between the ewes of different age groups. One point brought out in the analysis of the data was that the old ewes experienced greater falls in plasma magnesium than did the young ewes. It was mentioned earlier however that during the preliminary period of supplementary feeding, the old ewes consumed greater quantities and perhaps thereby had more elevated plasma magnesium levels (Fig.2). In consequence, they had a greater potential for falls in these levels when supplementary feeding ceased and a change in pasture made. Their actual mean plasma magnesium level on 28th April was the same as for the young ewes (Fig.3) and the two groups remained similar throughout the remainder

of the experimental period.

Plasma calcium was not affected by any of the fertiliser treatments but over both the preliminary and experimental periods, old ewes consistently had significantly lower levels than the younger ewes.

The effect of age on plasma calcium is also apparent in considering the old and young sheep as groups with single and twin lambs. (Fig.4). After parturition the old ewes with twins had considerably lower mean plasma calcium concentrations than comparable sheep with single lambs. This difference was statistically significant. The same effect to a lesser degree can be seen in the mean plasma calcium values of the young ewes with twins or single lambs. The depressive effects on plasma calcium of twins and increased age were additive. There was no consistent difference for groups with twin or single lambs in mean plasma magnesium concentrations as can be seen from Fig. 4. This finding is contrary to previous results (Hemingway et al. 1960).

With regard to the results for plasma calcium concentrations, it is of interest to record that the ewe which developed hypomagnesaemic tetany had the only abnormally low blood calcium concentration recorded during the experiment, while there were many ewes with equally low plasma magnesium concentrations. Reference to the four samples taken from this ewe over the 14 days prior to the appearance of clinical symptoms (Table 9) show that the low plasma magnesium concentration of 0.46 mg/100 ml was present for the previous seven days. It was the plasma calcium which progressively fell in the few days before the onset of tetany. It is of particular interest to consider this in relation to the similar low plasma calcium concentrations found in ewes succumbing to hypomagnesaemic tetany by O'Moore (1955), Penny & Arnold (1955) and Pook (1955).

It is also curious to note that those factors of increased age and number of lambs which, might have been expected to adversely affect plasma magnesium levels, were found in this experiment to have no measurable influence on plasma magnesium, but they were found to depress the plasma calcium concentrations.

These observations suggest that plasma calcium concentrations may have a more important part to play in the development of the clinical onset of hypomagnesaemic tetany than is at present suspected. This question will form the subject of a later section (Experimental work. Part 2).

The results from this experiment also demonstrate the necessity for having adequate numbers of sheep in investigations of this nature. The individual variation between these ewes was considerable. Based on the standard errors found here, for young and old sheep, a comparison on the basis of four animals in each group (e.g. four young sheep versus four old ewes) would require the fall in mean plasma magnesium levels over the period 19-28th April to differ by more than 0.54 mg/100 ml to be significantly different ($p = 0.05$). Similarly, groups of as many as sixteen animals would be needed to detect differences of 0.25 mg/100 ml. The use of two groups of thirty-six animals in this experiment was sufficient to detect statistically a difference of 0.18 mg/100 ml between the old ewes and the gimmers.

One further point which can be brought out from the data is that there was a highly significant correlation ($r = 0.63$) between the plasma magnesium levels of all sheep on 14th March and on 29th May. Thus ewes with initially high or low values tended to have similar respective levels at the end of the experiment. One feature of this was the presence of several sheep with abnormally low serum

magnesium levels which remained between 0.5 and 1.0 mg/100 ml, throughout the whole experiment and yet showed no outward symptoms of hypomagnesaemic tetany e.g. Nos. 3, 17, 32, 67, 73 in Appendix 2(b). On the other hand, it is difficult to say whether these ewes with consistently low plasma magnesium concentrations are any more likely to develop hypomagnesaemic tetany than ewes with more normal levels, since, in complete contrast to these ewes, the one sheep (no. 70) which did succumb to hypomagnesaemic tetany maintained a normal plasma magnesium until seven days prior to death.

There was no similar correlation between the plasma calcium levels on 14th March and 29th May for the individual sheep.

Experiment 2.

Experiment 2 was designed to study the effect of applications repeated in a second year of the fertiliser treatments given to experimental pastures in Experiment 1. It has previously been found by Bartlett et al. (1954; 1957), Kemp (1958a) and Hvidsten et al. (1959) that in similar experiments with cattle, high rates of potassium fertiliser usage (3-5 cwts/acre) did not induce clinical tetany in the first year of application. They did find, however, that when the treatments were repeated on the same pasture for a second year, the plasma magnesium concentrations of cows grazing these pastures were depressed and cases of hypomagnesaemic tetany occurred.

The 48 sheep used in Experiment 2 were of two different breeds, twenty-four being Cheviot ewes and twenty-four being Half-Bred ewes. This allowed a study to be made of the relative susceptibility of these breeds to hypomagnesaemia. A prophylactic treatment of dietary magnesium, given to half of the sheep of each breed was again superimposed upon the experimental design. The employment in Experiment 2 of some of the same sheep as were used in Experiment 1 enabled comparisons to be made on the plasma magnesium levels of the same sheep in two successive spring seasons.

Experimental Design.

The experimental flock of sheep consisted of twenty-four Cheviot ewes and twenty-four Half-Bred ewes. Both groups were 3 years old and were having their second crop of lambs. The Cheviot ewes were some of the sheep used in the Experiment 1 in 1961. Prior to and during lambing, the flock was grazing on old permanent pasture (0.15% Mg in the dry matter), and they were at that time receiving supplementary food (0.25% Mg content in the dry matter). Blood samples were

taken on 9th and 16th March 1962 before lambing commenced. Lambing took place in the middle fortnight of April. All the Half-Bred ewes had twin lambs apart from a few that had triplets. The Cheviot ewes had either single or twin lambs.

On 14th May, the ewes were transferred with their month-old lambs to the experimental plots. These plots were the same areas as used in Experiment 1, except that since the number of sheep available in 1962 was limited to 48 ewes, only 12 of the plots were used instead of the total of 18. The sward was still composed mainly of perennial ryegrass with some cocksfoot and no clover. All the plots had been given a dressing of 3 cwt/acre of Nitro-Chalk (21%N) on 24th March. The randomised block layout used in 1961 was retained except that there were four complete replicates of the three fertiliser treatments in place of original six blocks. There were therefore twelve individual experimental plots of 0.4 acre arranged in the four blocks of three plots. Within the blocks, the same potash fertiliser treatment was given to each plot as had been given in 1961. Thus 0, 1 and 2 cwts of muriate of potash were applied on 20th March to the appropriate plots in each block. Two Cheviot ewes and two Half-Bred ewes were allocated at random to each plot.

The plots used are shown in Fig.5, together with the repeated treatment and the ear tag number of each sheep allocated at random to the individual plots. Ewes with numbers in the series 2-40 represent Cheviot ewes and ewes with numbers in the series 41-77 represent Half-Bred ewes.

Superimposed upon this design there was a prophylactic treatment with a dietary magnesium supplement. This took the form of a daily drench of 6.64 g of magnesium oxide (supplying 4.0 g of magnesium) given to one Half-Bred ewe and

Fig. 5 Arrangement of fertiliser treatments and sheep in experimental plots at Cochno Farm (Experiment 2)

0K - No potassium fertiliser used.
1K - 1 cwt muriate of potash/acre.
2K - 2 cwt muriate of potash/acre.

	Ewe nos.		Ewe nos.
Plot 9 - 1K	58 51 37 25	Plot 18 - 2K	48 67 10 40
Plot 8 - 0K	54 60 2 15	Plot 17 - 0K	59 52 13 39
Plot 7 - 2K	63 44 32 34	Plot 16 - 1K	64 77 6 7
Plot 6 - 2K	42 53 29 11	Plot 15 - 1K	72 70 12 36
Plot 5 - 0K	46 49 3 21	Plot 14 - 2K	45 41 31 17
Plot 4 - 1K	56 43 20 22	Plot 13 - 0K	55 57 19 5
Plot 3 not used		Plot 12 not used	
Plot 2 not used		Plot 11 not used	
Plot 1 not used		Plot 10 not used	

one Cheviot ewe in each plot at 10 a.m. each morning. Consideration of this factor will be omitted meantime and will be described fully in Section 4.

Blood samples were taken on 14th May when the sheep were allocated to the separate plots and subsequently on the 16th, 19th, 23rd and 24th May. These samples were always obtained just prior to drenching at 10 a.m.

Herbage samples were taken from each plot on 18th May, and after drying and grinding, magnesium, calcium, potassium and sodium were determined.

Experiment 2 commenced 25 days later in the season than the comparable trial in 1961. This late start was entirely due to cold wet weather during April 1962 which delayed the growth of the pasture. When the sheep were finally put onto the experimental plots on 14th May, there was sufficient but not plentiful grazing, and the experiment was terminated after 10 days mainly because of shortage of grazing in the plots. This compares with the 40 days grazing provided for four sheep by each plot in 1961. It was, however, felt that the most critical time to examine closely in this type of experiment was, in any case, the week to 10 day period immediately following the move to the experimental area. Reference to the results of 1961 shows that there was an initial decline in mean plasma magnesium values after the grazing change, followed by a recovery over the following ten days and thereafter mean plasma magnesium levels remained more or less constant.

Results.

Preliminary period of grazing in March 1962.

The analytical results for the plasma magnesium and calcium concentrations of the 48 sheep on the 9th and 16th March are given in Appendix 2(f) together

with the mean values for the twenty-four Cheviot and twenty-four Half-Bred ewes. This is also shown graphically in the first part of Fig.6(a). On these dates which would be approximately one month before lambing, the mean plasma magnesium concentrations of both breeds were within the normal range of 1.8-2.5 mg/100 ml, although the group of Half-Bred ewes had significantly higher values ($p = 0.001$) on both dates than the Cheviot ewes. Only one ewe (Cheviot No. 3) had a plasma magnesium value below 1.00 mg/100 ml, its concentration being 0.95 mg/100 ml on both sampling dates.

The mean plasma calcium concentrations of both the Cheviots and the Half-Breds tended to rise over this preliminary period (Fig. 6(a)). The mean value for the twenty-four Half-Bred ewes increased from 8.88 mg Ca/100 ml on 9th March to 9.33 mg/100 ml on the 16th March, whereas the mean value for the 24 Cheviot ewes increased from 8.95 to 10.06 mg/100 ml over the same period.

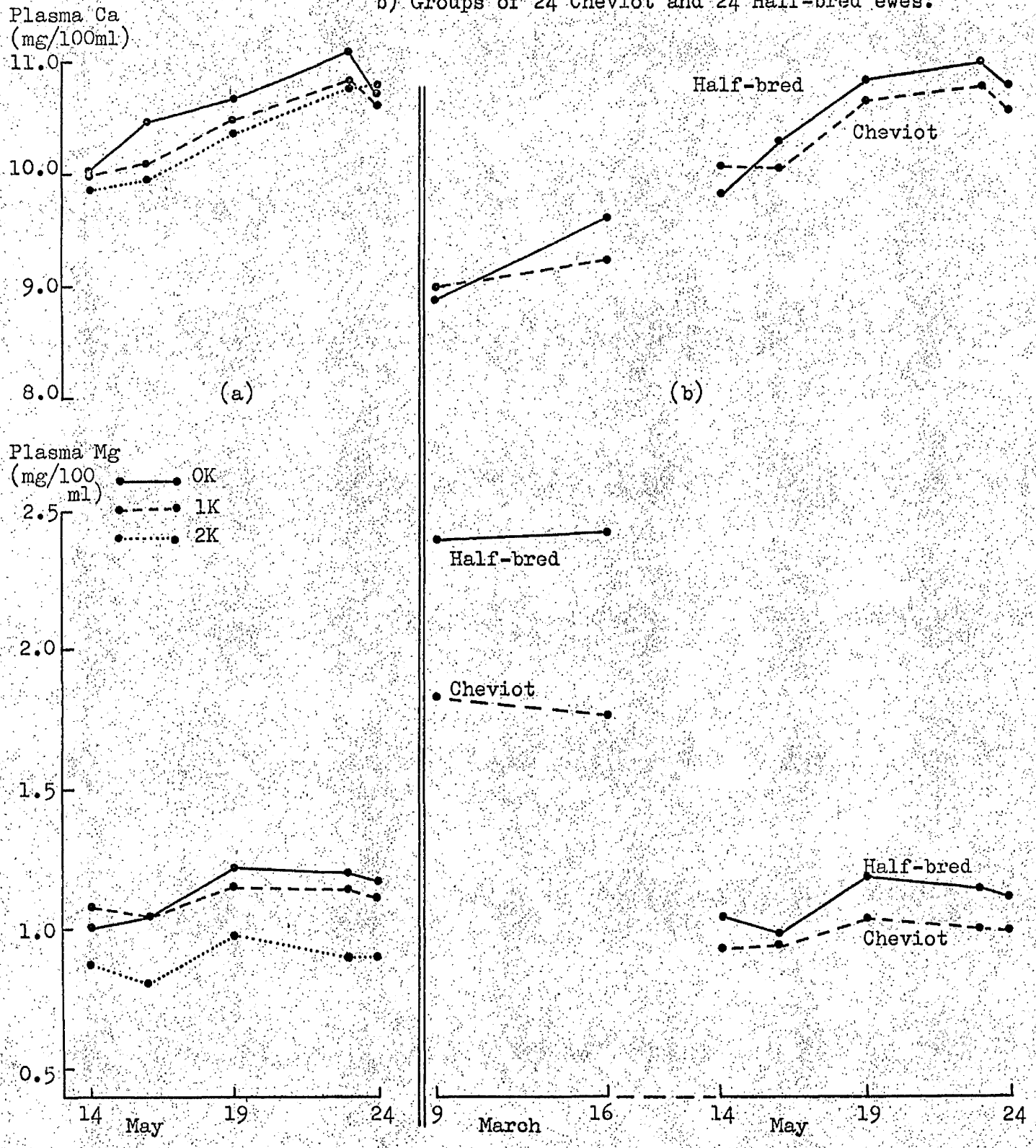
When the data were rearranged, it was apparent that the three groups of ewes which subsequently grazed plots with different potassium treatments had similar mean plasma calcium and magnesium concentrations over this preliminary period. (Fig. 6(b).

<u>Changes in plasma calcium and magnesium concentrations during the experimental period of plot grazing, 14th - 24th May, 1962.</u>

Appendix 2(g) details the analytical results for the plasma calcium and magnesium concentrations of each ewe on the 5 sampling occasions over this period, together with the mean values for each of the groups of treatment. The results are also shown graphically on Fig. 6.

It is apparent from the results for the sample taken on 14th May immediately

Fig. 6 Mean plasma calcium and magnesium concentrations of 48 lactating ewes at grass arranged as a) 3 groups of 16 ewes grazing plots given 0, 1 and 2 cwts muriate of potash/acre.
b) Groups of 24 Cheviot and 24 Half-bred ewes.



prior to the commencement of the various treatments, that considerable changes in plasma magnesium levels had taken place at some time between the previous sampling date on 16th March and this sample of 14th May. The mean plasma magnesium concentrations of the Half-Bred and Cheviot ewes on this latter date were 1.04 and 0.93 mg/100 ml respectively. Some 42% of the Half-Bred ewes and 54% of the Cheviots were below the level of 1.0 mg Mg/100 ml, and 5 of the 48 sheep were below the critically low level of 0.5 mg Mg/100 ml. Therefore a high degree of severe hypomagnesaemia was present in the ewes before the start of the experimental grazing period.

On transference to the manured plots, there were only very slight falls in the mean plasma magnesium concentrations (Fig. 6(b). These were completely non-significant. The mean levels of the Half-Bred and Cheviot sheep were 0.98 and 0.95 mg Mg/100 ml and this entailed 46% and 58% respectively of these sheep being below the level of 1.00 mg Mg/100 ml. Thereafter, for the following three samples taken on 19th, 23rd and 24th May, the mean plasma magnesium values remained substantially constant at these low levels. The ewes of both breeds maintained similar mean plasma magnesium concentrations throughout the whole period.

With regard to the effect of the fertilisation of the sward with potassium, it was apparent that animals grazing the plots given 2 cwt muriate of potash/acre were maintaining lower mean plasma magnesium values throughout the experimental period as compared with the 0K and 1K treatments. However this group started the experiment with a lower mean value, entirely due to chance, and the difference between the groups on later sampling dates was for the most part a maintenance of this initial difference.

Mean plasma calcium values of the separate groups of ewes rose steadily over the experimental period of 14th - 23rd May, with a slight drop in mean values on the 24th May (Fig. 6). The mean values for the Cheviot ewes were 10.00 on 14th May and 10.55 on 24th May. For the Half-Bred ewes the values on these dates were 9.83 and 10.82 mg Ca/100 ml respectively. The mean values of the groups grazing on the three fertiliser treatments all increased in a similar manner with little difference between the means on any sampling date, except that sheep grazing plots given no potassium fertiliser rose to a mean level of 10.5 mg Ca/100 ml on the 16th May, which was 0.5 mg above the other groups. On subsequent sampling dates this difference between the groups was less marked.

Statistical evaluation of the results.

Statistical evaluation of the effects of potassic fertiliser applications and the breed of the ewe on the plasma calcium and magnesium concentrations of the ewe was made by a similar procedure to that used in Experiment 1. Linear Regression lines were fitted for the blood samples for each ewe taken over the period 14-24th May. This enabled the increases in plasma magnesium and calcium over this period to be determined. Table 12 details this data for plasma magnesium and calcium. There was no evidence of any treatment differences in respect of plasma magnesium levels. There is perhaps a trend to be seen in Table 12 for the effect of the three fertiliser treatments on the twenty-four Cheviot ewes. There was a progressively smaller increase in plasma magnesium levels with increasing rate of usage of potassium fertiliser on the pasture, although this trend was not statistically significant. Apart from these linear regressions, consideration of the mean plasma magnesium values of the three groups

gives roughly the same impression. On the first day of plot grazing the mean plasma magnesium values of the Cheviot ewes grazing the 0, 1 cwt and 2 cwt of muriate of potash/acre plots were 0.95, 0.96 and 0.86 mg/100 ml respectively. Ten days later, these mean levels were 1.15, 0.97 and 0.83 mg/100 ml respectively.

Table 12. Increase (as determined by statistically smoothed values) in plasma calcium and magnesium over the 10 day period 14-24th May with relevant standard errors.

(a) Magnesium. (in mg/100 ml).

	K0	K1	K2	Mean
Half-Bred ewes	.20	.11	.10	.14
Cheviot ewes	.17	.05	-.01	.07

Standard errors of K0, K1, K2 marginal means = .072

Vertical comparisons within table = .085

Other comparisons within table = .095

Breed means H.B. V Cheviot = .035

(b) Calcium

	K0	K1	K2	Mean
Half-Bred ewes	.95	.98	1.14	1.03
Cheviot ewes	.61	.58	.84	.68

Standard errors of K0, K1, K2 marginal means = .15

Vertical comparisons within table = .20

Other comparisons within table = .21

Breed means H.B. V Cheviot = .06

In so far as plasma calcium concentrations were concerned (Table 12b), the only effect in evidence was the relatively minor one of the group of Half-Bred ewes showing a greater increase than the Cheviot ewes in plasma calcium over this period.

Herbage mineral content.

Table 13 gives the mean magnesium, calcium, potassium and sodium contents of the herbage samples from the four replicate plots of each potassium fertiliser treatment on 18th May. The individual results are to be found in Appendix 2(h).

Table 13. Magnesium, calcium, potassium and sodium contents of grazed herbage (dry matter) from experimental plots on 18th May, 1962.

(mean of four replicates).

Muriate of potash * (cwts/acre)	% Mg.	% Ca.	% K.	% Na.
0	.170	.57	2.23	0.50
1	.156	.46	2.79	0.37
2	.133	.48	3.01	0.30

* also applied in the previous year.

Muriate of potash applied at the rate of 2 cwts/acre significantly ($P = 0.05$) reduced herbage magnesium. The application of 1 cwt/acre had no significant effect on herbage magnesium, but from the mean figures given in Table 13, it is apparent that the trend is progressively downwards with increasing rates of potassium fertiliser treatment. Both the 1 and 2 cwts/acre rates of

application of potash caused significant reductions ($P = 0.01$) in the herbage sodium and calcium concentrations. Both rates greatly increased (significant at $P = 0.01$) the herbage potassium concentrations.

Discussion.

The results obtained in Experiment 2 differed in two obvious respects from those obtained in the previous year (Experiment 1). Firstly when Experiment 2 began on May, 14th, the flock already had a high degree of hypomagnesaemia to the extent that almost 50% of the animals had plasma magnesium concentrations below 1.0 mg/100 ml. This is in contrast to the more normal levels of 1.70 - 1.80 mg Mg/100 ml. which were found at the beginning of the first year experiment. Furthermore, many of the same Cheviot sheep were used in each year. The 22 Cheviot ewes which were present in both 1961 and 1962 had contrasting mean plasma magnesium levels at the start of the plot grazings of 1.71 mg/100 ml on the 19th April in 1961 and 0.93 mg Mg/100 ml on the 14th May 1962.

A comparison based on calendar date where the two closest dates are 12th May 1961 and 14th May 1962 gives the same mean figures of 1.71 and 0.93 mg Mg/100 ml respectively. The second important difference between the two years was the almost complete absence in 1962 of any fall in plasma magnesium values on transferring the flock to the experimental pasture, whereas in 1961 there had been a sharp drop in mean levels over the first three days on the experimental plots. It could, of course, be that in 1961, the mean plasma magnesium levels were at such a low level at the start of the experiment, there was little room for further falls in these levels. The reason for these low mean levels is unknown and the actual time when the fall in mean levels took place is also

unknown. It must have occurred at some time between the prelambing samples taken in March when the plasma magnesium levels were within the normal range and the start of the experiment in May. It may have occurred at parturition in the middle of April, or possibly during the subsequent month when the ewes were lactating. The fact still remains however that the sequence of events, the pastures grazed and the sheep sampled were exactly the same in 1961 and 1962 and yet the magnesium status of the ewes as measured by blood samples was completely different. One must therefore conclude that some unknown factor of season or weather influenced the development of hypomagnesaemia to a greater extent than any factor which was being studied in this experiment. The obvious seasonal effect to comment on was the delay in pasture growth in 1962 due to poor spring weather. —The paradox to this is that despite there being over 50% of the sheep having a plasma magnesium concentration below 1.00 mg/100 ml at some time during the experiment, no cases of clinical tetany were recorded.

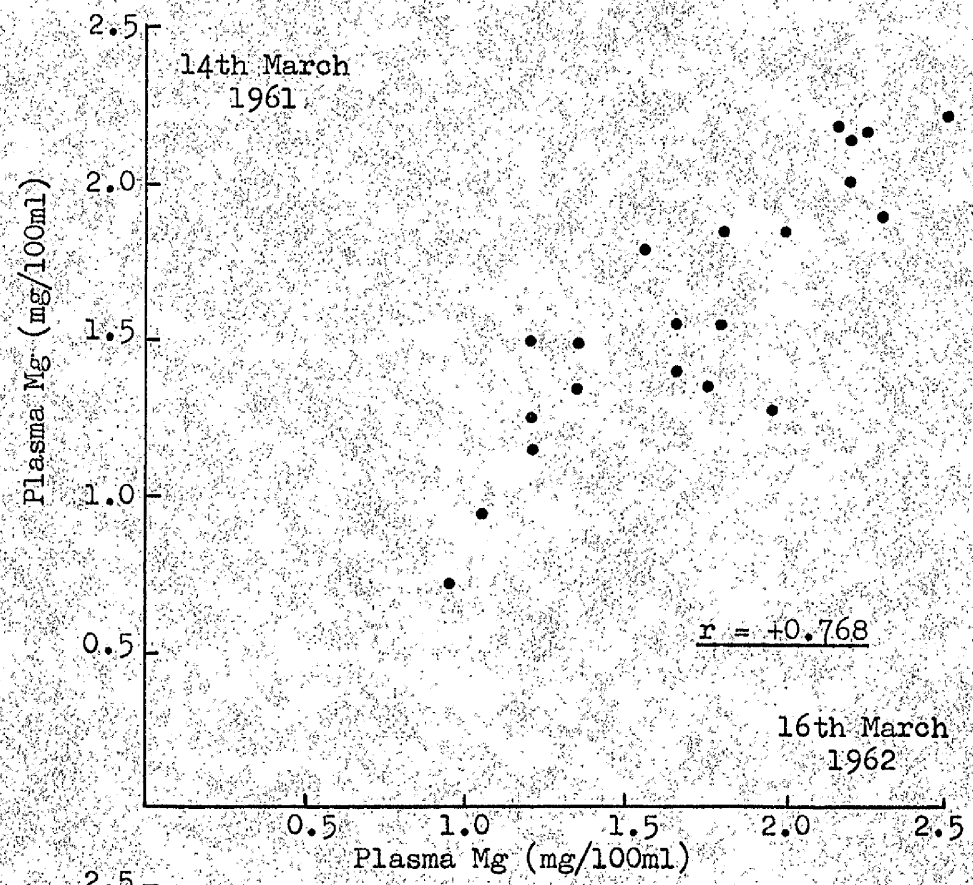
The continued application of 2 cwt of muriate of potash over two spring seasons caused a significant reduction in herbage magnesium as compared to the slight non-significant reduction in herbage magnesium produced in the first year of this treatment. The effect of 1 cwt of muriate of potash on herbage magnesium was proportionally less and it was a non-significant reduction. This effect on the herbage was not reflected by any significant depression in plasma magnesium concentrations attributable to potassium fertiliser application to the pasture. There was, however, a non-significant downward trend to be seen in the mean plasma magnesium levels of the Cheviot ewes which was in proportion to the rate of potassium fertiliser usage.

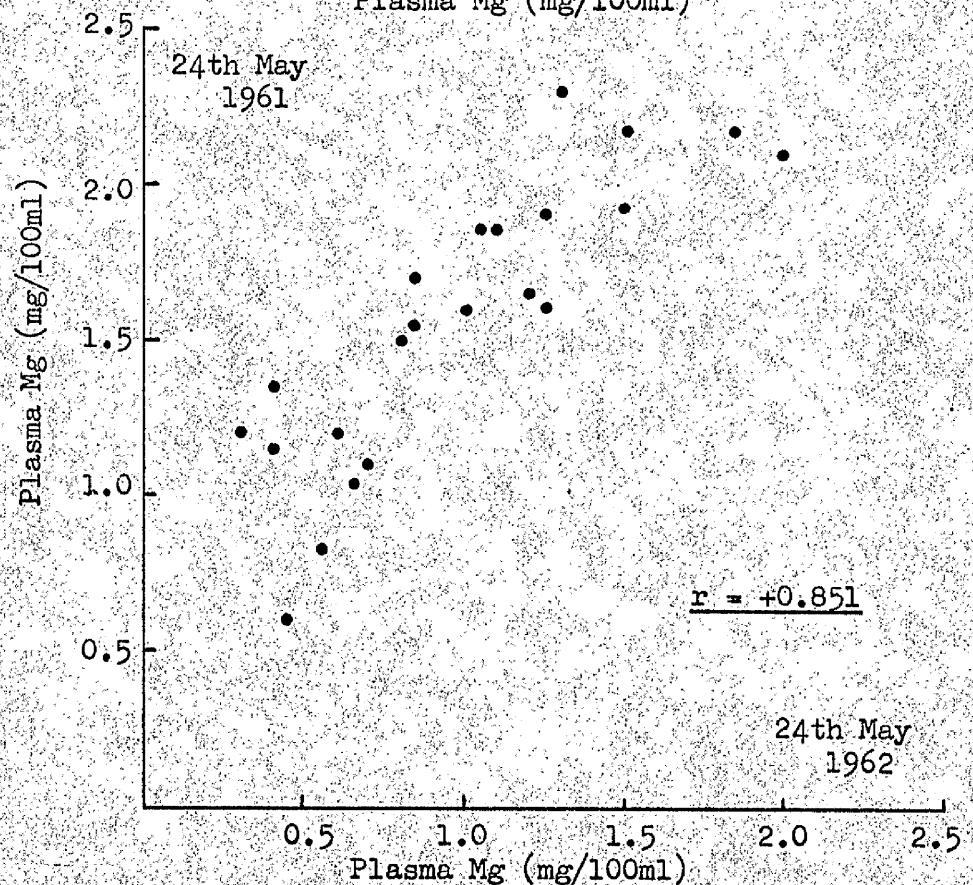
Although the Cheviot ewes tended to have lower plasma calcium and magnesium values than the Half-Bred ewes, these differences were only slight, and the degree of hypomagnesaemia found on May 14th in each breed was almost equally severe. There would, therefore, seem to be little difference between these two breeds under the conditions of this experiment in their susceptibility to hypomagnesaemia. This would not preclude the possibility of there being a difference in their susceptibility to hypomagnesaemic tetany since there may be more involved in the condition other than low plasma magnesium concentrations.

One interesting aspect of the results is the high degree of correlation that can be established between the plasma magnesium concentrations of the twenty-four Cheviot ewes in either March or May in 1962, and the levels in the same period of 1961. Fig. 7 shows the individual plasma magnesium concentrations of twenty-two of the Cheviot ewes on 16th March 1962, plotted against the corresponding individual values on 14th March 1961. It also shows a similar plot for the individuals on the 24th May in 1961 and 1962. The correlation coefficient for the samples in March 1961 and 1962 was +0.768 and for the samples taken after lambing in May 1961 and 1962, it was +0.851. These coefficients were highly significant.

Similar correlations can also be calculated for these ewes between different dates in the same year. The implication of these correlations (and this can be seen most strikingly in Fig. 7) is that any individual ewe which has a low plasma magnesium concentration at any stage in the year will tend to have a low plasma magnesium concentration at other times in that year and also in subsequent seasons. Similarly a ewe found to have high concentrations will tend always to

Fig 7 Correlations between the plasma magnesium concentrations of 22 Cheviot ewes for dates before and after lambing in 1961 and 1962.





have a high plasma magnesium level. On the other hand, the mean plasma magnesium concentrations of these Cheviot ewes was much lower in the May samples in 1962 than in 1961 (1.00 and 1.65 mg/100 ml respectively). But the individual values were each depressed to approximately the same degree, and therefore the sheep with the highest values in 1961 still had the highest values in 1962 and similarly for the sheep with the lowest values.

Experiment 3.

Experiment 3 was the continuation in 1963 for a third year of some of the fertiliser treatments given in 1961 and 1962 to the experimental plots at Cochno Farm. The experimental design was altered to include several magnesium pasture treatments and this made it impossible to retain all three rates of potassium fertiliser application. It was therefore decided to repeat the pasture treatments of 0 and 2 cwts of muriate of potash per acre replicated three times on plots which had received these rates of fertiliser over the last two years. The remainder of the plots were given dressings of magnesium fertiliser, to investigate the effectiveness of this form of preventive treatment. The results of this part of the experiment will be omitted meantime and dealt with in Section IV.

The experimental design was similar to experiments 1 and 2, in that the ewes were transferred to the fertilised plots in the spring. Two breeds of sheep were used and these were again Half-Bred and Cheviot. Because of lack of grazing, this part of the experiment was terminated after 9 days, but after regrowth of the plot pastures had taken place in June a flock of Blackface ewes were transferred to the plots and run as a second part to experiment 3.

Experimental Design.

The experimental flock of sheep in the first part of this experiment consisted of thirty Cheviot ewes and thirty Half-Bred ewes. Both groups were four years old and were having their third crop of lambs. Twenty of the Cheviots and twenty one of the Half-Bred ewes involved in this experiment were present in the flock of sheep used in Experiment 2 in 1962. Twenty seven of the Cheviot

ewes had also previously been used in Experiment 1 in 1961. Prior to and during lambing, the flock was grazing on old permanent pasture. All the Half-Bred ewes and most of the Cheviots had twin lambs. Lambing took place during the middle fortnight in April.

On 15th May, the ewes were transferred with their month old lambs to the experimental plot pastures. These plots were fifteen of the 0.4 acre plots as used in Experiments 1 and 2. Fig. 8 details the fertiliser treatments given to each plot. A general dressing of three cwts of Nitro chalk (21% N) per acre was applied to every plot on 19th March. Three of the plots which had been given 2 cwts of muriate of potash per acre in each of the two preceding years, were given this fertiliser treatment for a third year on 1st March. Three plots which had been given no potassium fertilisation for the past two years were again retained as controls. The remaining nine plots were given some form of magnesium fertiliser treatment. Two Cheviot ewes and two Half-Bred ewes were allocated at random to each plot on 15th May. Figure 8 gives the ear tag numbers of the ewes thus allocated. The ewes with numbers within the series 2-40 represent Cheviot ewes and those in the series 41-81 are Half-Bred ewes. Blood samples were taken from every ewe immediately prior to transference to the experimental pasture on 15th May and again on 17th, 20th and 23rd May. These samples were analysed for plasma calcium and magnesium. The grazing period was of necessity short (9 days) due to lack of grazing but it was again felt that the critical time to examine in this type of experiment was the seven to ten day period following the pasture change. The late start of the experiment in the middle of May was again due to a cold spring causing a delay

Fig. 8 Layout of experimental plots at Cochno Farm (Experiment 3a).
OK - No potassium fertiliser applied.
2K - 2 cwts muriate of potash/acre.
High Mg - 5 cwts calcined magnesite/acre.
Low Mg - 0.5 cwts calcined magnesite/acre.

	Ewe nos.		Ewe nos.
Plot 9 - High Mg	2 23 63 65	Plot 18 - High Mg	9 10 51 68
Plot 8 - OK	3 33 71 77	Plot 17 - OK	5 29 56 79
Plot 7 - 2K	25 32 55 67	Plot 16 - Low Mg (spring)	26 27 57 70
Plot 6 - 2K	17 38 41 45	Plot 15 - High Mg	11 36 52 81
Plot 5 - OK	30 37 53 74	Plot 14 - 2K	6 12 42 60
Plot 4 - Low Mg (spring)	7 22 49 54	Plot 13 - Low Mg (spring)	13 28 64 69
Plot 3 - Low Mg (winter)	15 19 43 72	Plot 12 - Low Mg (winter)	21 34 44 59
Plot 2 - not used		Plot 11 - Low Mg (winter)	16 40 75 80
Plot 1 - not used		Plot 10 - not used	

in pasture growth.

The second part of this experiment involved a flock of twenty four Blackface ewes. Prior to the period of experimental grazing in the plots, these ewes had been maintained indoors on a low calcium diet for the previous six months. Lambing had taken place indoors about the end of March. Since all the ewes had been receiving the same treatment and diet, the results from the grazing experiment when examined on a comparative basis, should not be influenced by this pre-experimental treatment. There were fourteen ewes with single lambs and ten ewes with twins.

These Blackface ewes, together with their three-month-old lambs, were transferred to the experimental plots on 11th June, four ewes being allocated at random to each of six plots. The particular plots used and the ear tag numbers of the sheep grazing in each plot are given in Fig. 9. Eight ewes were therefore grazing on plots given a third annual treatment of 2 cwts muriate of potash/acre and eight were grazing plots given no potassium fertiliser. The remaining eight were allocated to plots given a dressing of magnesium fertiliser. The distribution of ewes with twin lambs was approximately even, there being three or four ewes with twins on each of the three treatments.

Blood samples were taken from all the ewes on the 11th, 12th, 14th, 17th and 21st June and again on the 18th July when the lambs were weaned. These were analysed for plasma magnesium and calcium.

Herbage samples were taken from each plot on 13th and 22nd May and again on 11th June. Analyses were made on these samples for magnesium, calcium and sodium content.

Fig. 9 Layout of experimental plots at Cochno Farm (Experiment 3b)

0K - No potassium fertiliser applied. Blackface ewes
2K - 2 cwts muriate of potash/acre.
High Mg - 5 cwts calcined magnesite/acre.

	Ewe nos.		Ewe nos.
Plot 9 - High Mg	77 82 91 92	Plot 18 - High Mg	68 74 81 88
Plot 8 - 0K	85 87 97 99	Plot 17 - 0K	66 71 76 86
Plot 7 - 2K	67 73 94 96	Plot 16 -	not used
Plot 6 - 2K	80 83 84 93	Plot 15	not used
Plot 5 -	not used	Plot 14	not used
Plot 4	not used	Plot 13	not used
Plot 3	not used	Plot 12	not used
Plot 2	not used	Plot 11	not used
Plot 1	not used	Plot 10	not used

Results. Part (a).

Plasma calcium and magnesium concentrations during the experimental period of plot grazing 15th - 23rd May.

The individual plasma calcium and magnesium concentrations of the Cheviot and Half-Bred ewes on the four sampling dates over this period are given in Appendix 2(i), together with the relevant mean values. The mean levels for the groups of twelve ewes grazing on the plots given 0 and 2 cwts muriate of potash per acre and the mean levels for the 30 ewes of each breed are shown graphically in Fig. 10.

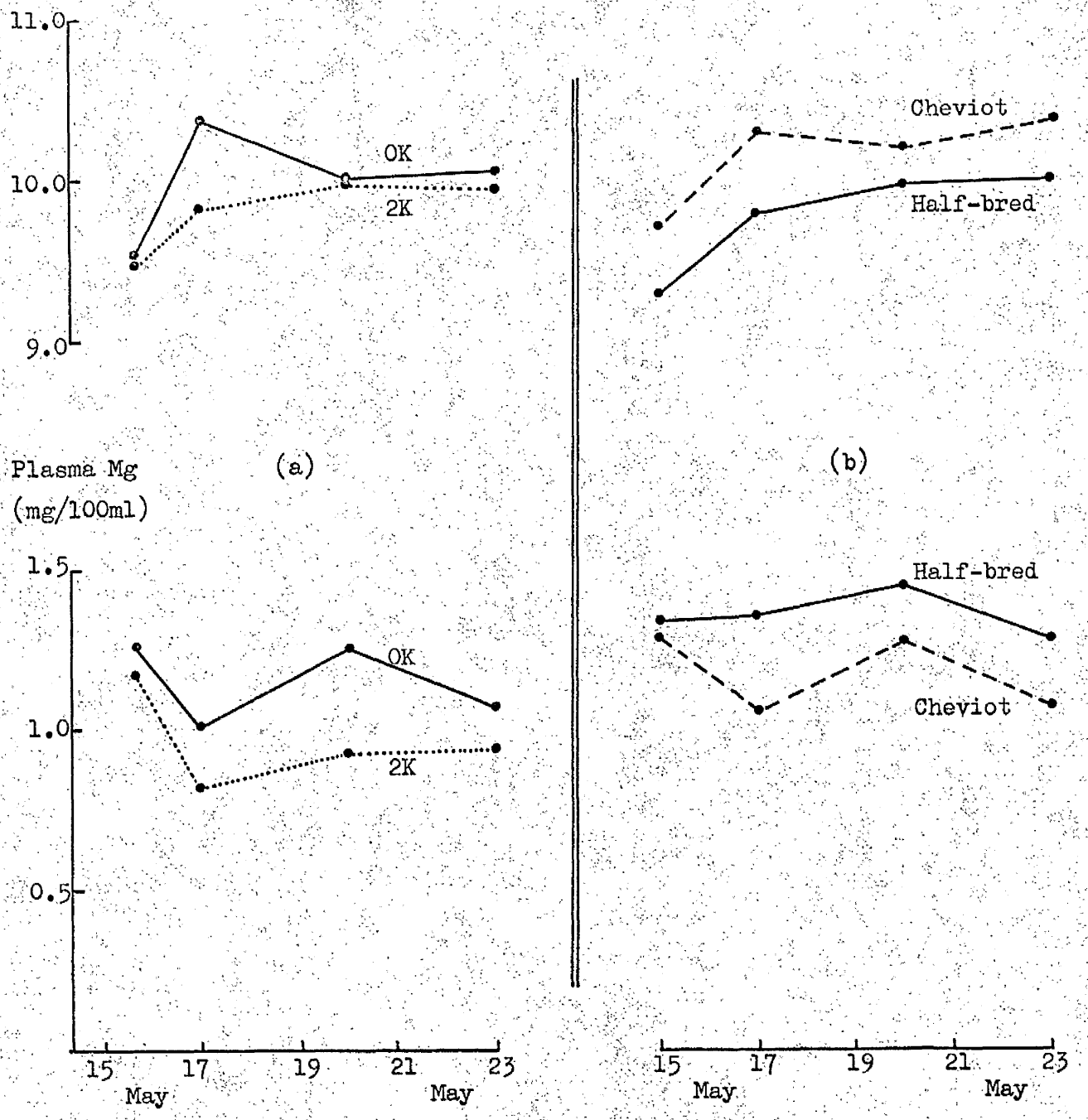
The mean plasma magnesium levels of both the Cheviot and Half-Bred ewes were in the hypomagnesaemic range (1.29 and 1.34 mg/100 ml respectively) at the start of the experiment. (Fig. 10(b). This entailed 22% of the individual plasma magnesium concentrations being below the level of 1.00 mg/100 ml. After two days of grazing in the experimental plots, mean plasma magnesium concentrations of the thirty Cheviot ewes had fallen to 1.05 mg/100 ml and 40% of those ewes had levels below 1.00 mg Mg/100 ml. In contrast the Half-Bred ewes experienced no fall in mean plasma magnesium over this two day period, and 27% had plasma magnesium levels below 1.00 mg/100 ml. This difference between the breeds on this date was statistically significant ($P = 0.02$). On subsequent sampling dates on the 20th and 23rd May, the mean plasma magnesium concentrations were in approximately the same range. That of the Cheviot ewes remained below that of the Half-Bred ewes, but the difference was no longer significant.

The mean plasma calcium concentration of the Cheviot ewes rose over the 9 day experimental period from 9.74 to 10.38 mg/100 ml. This level was

Fig. 10 Mean plasma calcium and magnesium concentrations of lactating ewes at grass (Experiment 3a). Grouped as-:

a) Two groups of 12 ewes on plots given 0 and 2 cwts muriate of potash/acre.

b) Groups of 30 Cheviot and 30 Half-bred ewes.



consistently 0.2–0.4 mg Ca/100 ml above that of the Half-Bred ewes, although this difference was not significant.

The mean plasma magnesium concentrations of the two groups of twelve sheep grazing on plots given nil (0K) and 2 cwt muriate of potash/acre (2K) were respectively 1.23 and 1.16 mg/100 ml at the start of the experiment. (Fig. 10(a). After two days grazing these levels had fallen to 1.00 and 0.80 mg/100 ml. This difference widened on the next sampling date three days later when the mean levels of the two groups were 1.23 and 0.93 mg Mg/100 ml, respectively. Statistical examination of the results for this date show that the twelve sheep grazing the potassium fertilised plots had significantly ($P = 0.02$) lower plasma magnesium values than the twelve sheep on the plots given no potassium. On the following sampling date on 23rd May, the difference between the groups was less marked, and was no longer significant. These groups of twelve sheep on each treatment were composed of 6 Cheviot ewes and 6 Half-Bred ewes in each group. It is of interest to examine the effect of these potassium pasture treatments on each breed, in particular on the Cheviot ewes. Table 14 details the mean plasma magnesium concentrations of the six Cheviot ewes on each potassium treatment on the four sampling occasions of experiment 3.

Table 14. Mean plasma magnesium concentrations of two groups of six Cheviot ewes at Cochno 1963.

fertiliser treatment *	(plasma Mg (mg/100 ml).			
	15/5	17/5	20/5	23/5.
No potassium	1.37	1.08	1.17	0.94
2 cwt muriate potash/acre	1.13	0.54	0.81	0.78

* Repeated each year for 3 years.

It indicates that when the Cheviot ewes are considered separately, on pasture given 2 cwts muriate of potash/acre for three consecutive years, the six ewes experienced a more severe reduction in plasma magnesium concentration than similar ewes on control pastures. The mean levels after three days grazing were 0.54 and 1.08 mg Mg/100 ml respectively. This entailed all six ewes on the treated pasture having individual levels below 1.00 mg/100 ml, whereas only two of the six ewes on the untreated pasture were below this level, and this difference between the groups was clearly significant ($P = 0.01$) on this date. On subsequent sampling dates, there was a rise in the mean plasma magnesium concentration of this group on the treated pastures, and although there was still a difference of 0.20-0.30 mg Mg/100 ml between the two groups, this was no longer statistically significant.

The mean plasma calcium concentrations of the groups on differently treated pastures were very similar on all sampling occasions and they both rose from 9.50 to 10.0 mg Ca/100 ml over the 10 days of the experiment. The exception was on the 17th March, three days after transfer to the plots, when the mean plasma calcium level of the group on the untreated pasture was 10.4 mg/100 ml as compared with 9.83 mg Ca/100 ml for the group on treated pasture.

Results (Part b).

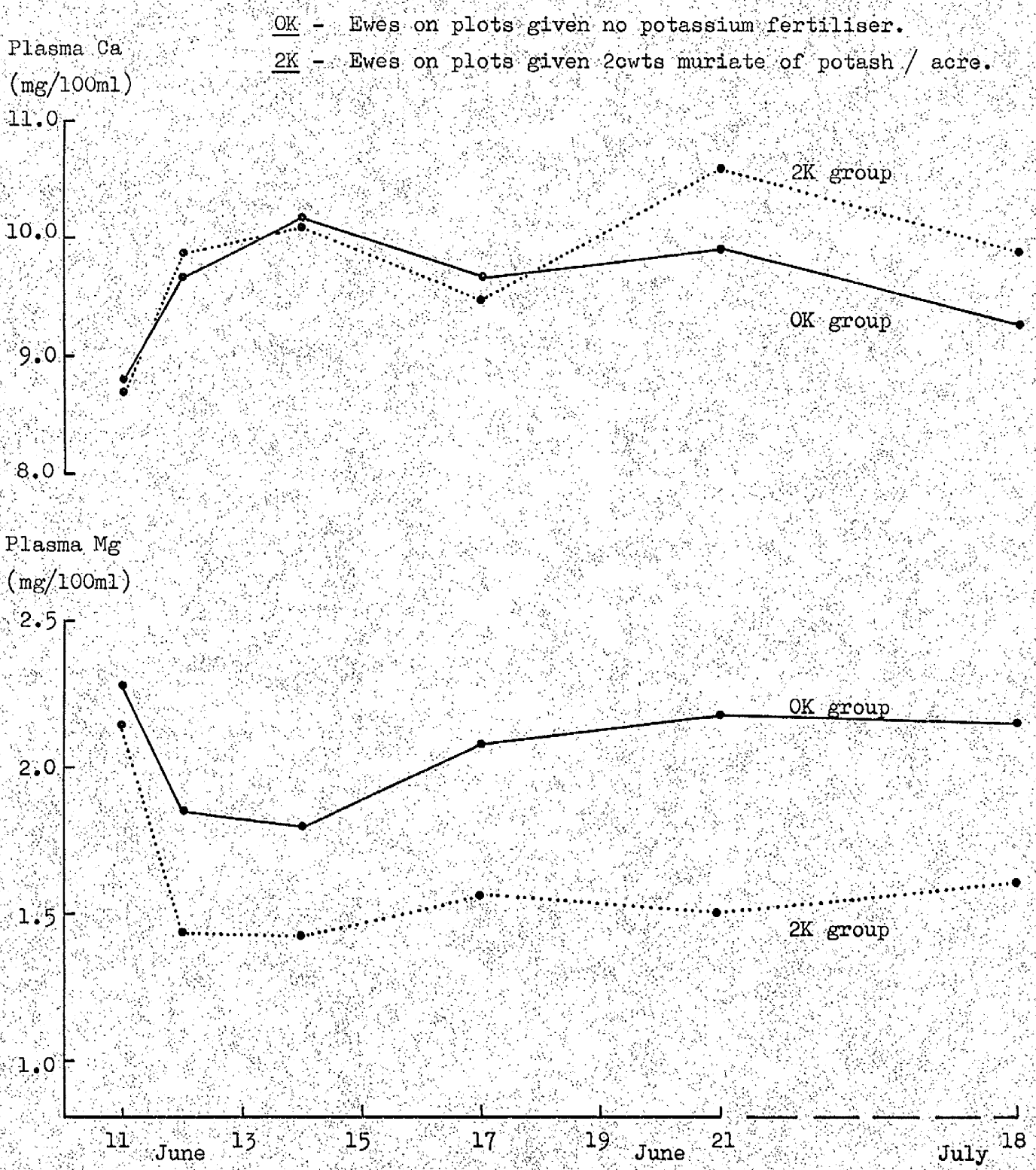
Plasma calcium and magnesium concentrations of 24 Blackface ewes during the experimental period of plot grazing 11th June - 18th July.

The individual plasma calcium and magnesium concentrations of the twenty-four Blackface ewes on six sampling occasions over this period are given in Appendix 2(j), together with the mean levels of the groups grazing on differently

treated pastures. These mean plasma levels are also shown graphically in Fig.11. Once again the results for the groups grazing on pasture given a magnesium fertiliser treatment are omitted meantime (see later Section IV).

The mean plasma magnesium levels of both groups were within the normal range on the day of transfer from indoor feeding to outdoor grazing. The mean levels of both groups suffered a fall over the following twenty four hours. The group on the untreated pasture fell to 1.85 mg Mg/100 ml, but this level was not significantly lower than the previous sample. In contrast the group grazing the pasture which had received dressings of potassium fertiliser suffered a significant fall ($P = 0.02$) in plasma magnesium concentration to 1.43 mg/100 ml. The mean level of this group on the treated pasture remained considerably lower than that of the control group on every sampling date thereafter, the difference between the groups varying from 0.37 to 0.68 mg Mg/100 ml. Statistical examination of these results showed that the groups on treated pasture were significantly lower ($P = 0.01$) on the fifth sampling occasion on 21st June. Although there was no significant reduction on each of the sampling dates, 12th, 14th and 17th June considered singly, when the data for these three dates were combined, the plasma magnesium concentrations of the group of the treated pasture was again found to be significantly lower ($P = 0.01$) than the comparable group on the pasture given no potassium treatment. (Combining the data for several dates is justified when, as in this case, a difference between groups is consistently maintained over several sample dates). On the later sampling date in July when the lambs were being weaned off the ewes, this difference between the groups was still apparent.

Fig. 11 Mean plasma calcium and magnesium concentrations of two groups of 8 Blackface ewes at grass. (Experiment 3b)



The mean plasma calcium concentrations of both groups rose rapidly from 8.5 mg to 9.75 mg/100 ml over the first twenty four hours of the experiment. This rise would be due to the change from the indoor low calcium diet to pasture which had an adequate calcium content. Thereafter the levels of both groups remained reasonably constant at this level of 9.5 - 10.0 mg Ca/100 ml and there was no apparent difference between the two groups of ewes on different pastures.

Herbage mineral contents.

The analytical results for herbage magnesium, calcium, potassium and sodium concentrations in the grass samples from each of the three replicates of each fertiliser treatment are given in Appendix 2(k), together with the treatment means. The treatment means for the plots given 2 cwt muriate of potash/acre and those given no potassium are also detailed in Table 15.

Table 15. Mean mineral content of plots at Cochno 1963. (mean of 3 replicates).

	% Mg.			% Ca.			% K.			% Na.		
	13/5	22/5	11/6	13/5	22/5	11/6	13/5	22/5	11/6	13/5	22/5	11/6
no potassium fertiliser.	.193	.192	.136	.82	.55	.66	2.4	2.4	2.2	.53	.34	.43
2 cwts muriate of potash/acre.	.154	.116	.110	.66	.45	.53	3.6	2.9	3.2	.25	.21	.23

The application of 2 cwts of muriate of potash for three consecutive years caused a significant change ($P = 0.05$) in the herbage content of all four minerals for which analyses were carried out. There were significant reductions in herbage magnesium, calcium and sodium and a significant increase in the herbage potassium, all being attributable to the application of potassium fertilisers. The difference between

the herbage magnesium content of the two treatments on the 22nd May is particularly noticeable, in that the levels were 0.192 and 0.116% Mg for the untreated and treated pastures respectively. This represents a 40% reduction in magnesium content associated with the repeated application of potassium fertilisers.

Discussion.

Although the two parts of this experiment were carried out with separate flocks of sheep running on the same plot pastures at different periods, the results obtained are very similar in certain aspects. Consequently these two sets of results can conveniently be discussed jointly.

As shown in Figs. 10 and 11, the mean plasma magnesium concentrations of both flocks of ewes tended to be depressed by repeated heavy pasture applications of potassium fertiliser (2 cwts muriate of potash). Statistical examination showed that on one sampling date in each case, this reduction was significant when compared with a similar group of sheep on pasture given no potassium. There were also statistical differences found between the groups on other sampling dates when the data for the different dates were combined, and also when the Cheviot ewes were considered separately from the combined Cheviot-Half-Bred flock.

The fact that two separate flocks of ewes reacted in a similar fashion when subjected at different periods to the same experimental treatments, adds weight to the conclusion that repeated potassium fertilisation on pasture depressed the plasma magnesium concentrations of lactating ewes grazing this pasture under the conditions of this experiment. It must be stressed however that the rate of

potassium fertilisation (2 cwts muriate of potash/acre) used in this experiment is in excess of rates which would normally be in use on pasture. A dressing of 1 cwt muriate of potash would normally be considered as more than adequate for the potassium fertilisation of herbage used for spring grazing. The other important factor in this experiment is that this effect was only found after the third annual application on the same pasture. Despite this effect on plasma magnesium concentrations attributableto potassium fertilisation, there were no cases of hypomagnesaemic tetany recorded in this experiment. This was notwithstanding the high proportion of ewes found to have plasma magnesium concentrationsbelow 1.00 mg/100 ml.

It is also of interest to relate this association found between plasma magnesium concentrations and pasture fertilisation, to the herbage mineral contents. Potassium fertilisation of the grass for a third consecutive year was found in this experiment to depress severely (by 40%) the herbage magnesium content. Sheep on such a pasture must necessarily suffer a proportionate reduction in their dietary magnesium intake, and under many circumstances, this must increase the susceptibility of sheep to hypomagnesaemia and hypomagnesaemic tetany. It is postulated, therefore, that this direct effect of potassium fertilisation, in causing a reduction in the magnesium content of pasture, may be the means whereby potassium fertilisation exerts a depressive effect on the plasma magnesium concentrations of sheep grazing the pasture. It would therefore be unnecessary to postulate the existence in heavily fertilised grass of some deleterious component which adversely affects plasma magnesium concentration. Various components which have been suspected as playing this role are high

herbage potassium, high herbage protein levels and low herbage sodium contents. All these "abnormalities" are natural consequences of heavy potassium fertilisation, and as Wilson (1960) reasoned "association does not imply causation".

A breed comparison can be made between the Cheviot ewes and the Half-Bred ewes employed in the first part of this 1963 experiment. The results obtained were similar to those found in Experiment 2 in 1962 where a breed comparison was made with the same sheep used in this current experiment. Thus, in both years, the Cheviot ewes consistently had lower mean plasma magnesium levels than the Half-Bred ewes, and in both years there was consequently a greater proportion of Cheviot ewes with plasma magnesium concentrations below the critical level of 1.00 mg/100 ml. In this experiment, however, this effect was more pronounced and the difference between the groups was such that on one sampling date (17th May), the Cheviot ewes were significantly ($P = 0.02$) lower in plasma magnesium concentration. There would, therefore, seem to have been a greater susceptibility in the Cheviot ewes in this experiment to the development of hypomagnesaemia and thereby possibly to hypomagnesaemic tetany.

Since there was an even distribution of the numbers of Blackface ewes with single and twin lambs amongst the various treatments in the second part of this experiment, it was possible to examine the results for any effect that the number of lambs, being suckled by the ewe, might have on the plasma magnesium concentration of the ewe.

The data given in Appendix 2(j) for the individual plasma calcium and magnesium concentrations can be rearranged into results for a group of fourteen ewes with single lambs and a group of ten ewes with twin lambs. The number of lambs with each ewe is also given in Appendix 2(j). The mean plasma magnesium and calcium levels of these groups is given in Table 16.

Table 16. Mean plasma magnesium and calcium concentrations of groups of Blackface ewes with single and twin lambs. Cochno 1963.

(mg/100 ml.)

Plasma Constit.	Group.	11/6	12/6	14/6	17/6	21/6	18/7
Mg	14 ewes with single lambs	2.32	1.85	1.89	2.12	2.14	2.14
	10 ewes with twin lambs	2.09	1.61	1.57	1.69	1.80	1.69
Ca	14 ewes with single lambs	8.71	9.69	9.43	9.51	10.15	9.42
	10 ewes with twin lambs	8.70	9.66	10.09	9.50	10.04	9.70

The mean plasma magnesium concentration of ewes with twins fell more sharply on transference to grass and they remained at a lower level of 1.57-1.69 mg/100ml. on most subsequent sampling dates. Ewes with single lambs had more normal mean levels in the range 1.85-2.14 mg Mg/100 ml. There was no significant difference between the groups on any one date but since the difference between the groups was maintained consistently over every sampling date, it was again justifiable to examine statistically the combined data for several dates. On this basis,

over the period 14th - 21st June, the ewes with twin lambs had significantly ($P = 0.01$) lower magnesium concentrations than ewes with single lambs. This depressive effect of twin lambs on plasma magnesium is in contrast to the absence of any such effect in Experiment 1 where groups of ewes with twin and single lambs were also compared. It is, however, a finding which is more in agreement with the greater incidence in field outbreaks of hypomagnesaemic tetany in ewes with twin lambs as against ewes with single lambs. (Barrentine & Morrison 1953; Penny & Arnold 1955; Pook 1955; Hughes 1958; and Herd & Peebles 1962).

The mean plasma calcium concentration of ewes with single lambs and ewes with twin lambs were very similar. (Table 16).

The use of the same flock of Cheviot sheep for the third year in succession allowed correlations to be drawn between the plasma magnesium levels of the same sheep in the two successive years 1962 and 1963 (Fig. 12) and also over a period of three years from 1961 and 1963. The correlation between the plasma magnesium levels of the Half-Bred ewes in 1962 and 1963 was also examined. (Fig.13).

On purely visual examination, the plasma magnesium values of the Cheviot ewes in different years is obviously closely correlated. In mathematical terms, the correlation coefficients are 0.846 and 0.762 for the plasma magnesium levels in 1963 v 1962 and 1963 v 1961 respectively. These coefficients are statistically significant at greater than $P = 0.001$. This is in close agreement with the excellent correlation found in Experiment 2, between the years 1961 and 1962. This implies that each Cheviot ewe in the period 1960-1963 always had a similar plasma magnesium concentration in the month of May in each year. There were,

Fig. 12 Correlations between the plasma magnesium concentrations of individual Cheviot ewes in the years 1961, 1962, and 1963 on the 23rd- 24th May.

(concentrations in mg Mg /100ml)

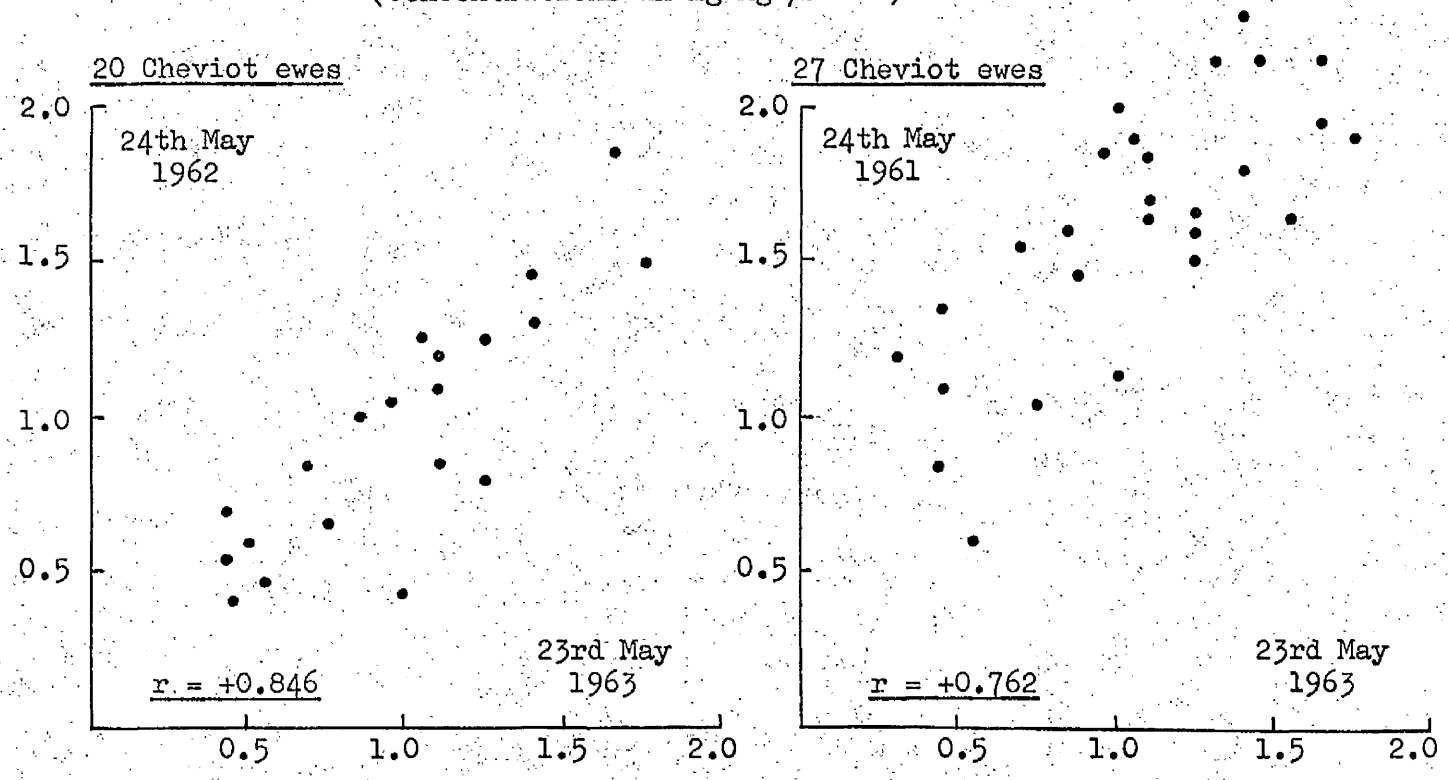
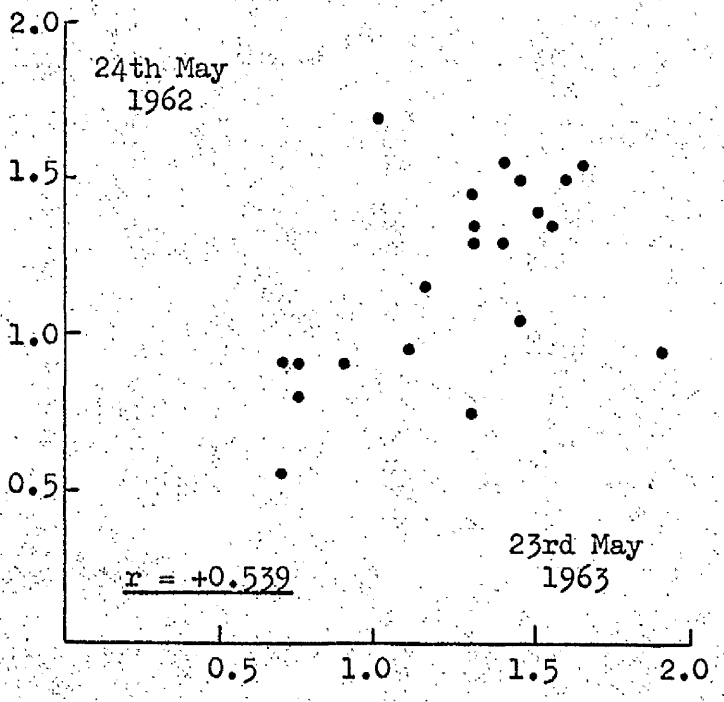


Fig. 13 Correlation between the plasma magnesium concentrations of 21 Half-bred ewes on the 24th May 1962 and 24th May 1963.



therefore, a number of ewes in this flock whose concentration of plasma magnesium was consistently below the accepted "normal" levels in the period after lambing. Correlations were found in Experiment 2 between the plasma magnesium values of these Cheviot ewes in different months of the same year. Combining these two findings of close correlations being found at the same date in different years and at different dates in the same year, a more general conclusion would be that there existed in this flock of Cheviot ewes a number of sheep whose plasma magnesium concentration was possibly always below the accepted "normal" levels. The reason for this may be genetic. It would, however, be doubtful as to whether these sheep are any more susceptible to hypomagnesaemic tetany than their more "normal" neighbours in a flock.

Conclusions - Experiments 1, 2 and 3.

(a) pasture fertilisation.

The fertiliser treatments studied in these experiments were the application of medium dressings of nitrogen (3 cwts nitro chalk/acre) in combination with 0, 1 cwt (adequate) and 2 cwts (excessive) of muriate of potash/acre. The use of these potassium pasture dressings had no effect on the plasma magnesium concentrations of sheep in the first year of application, and there was only a very slight non-significant tendency in the second year for a depression in plasma magnesium concentration to result from the use of these treatments. Only in the third year of fertiliser application was there a significant reduction in the plasma magnesium values of ewes grazing the pasture given the 2 cwt rate of muriate of potash. This was found with two separate flocks grazing the pasture at different periods in this third year.

There would, therefore, seem to be a progressive tendency over the years for the use of an excessive amount of potassium fertiliser to increase the incidence of hypomagnesaemia, but only after continued application every year was the effect measurable. Even then the increase in the degree of hypomagnesaemia was not great.

It was felt that this effect on the magnesium status of the animal can be directly related to the effect that these potassium fertilisers had on the herbage magnesium. Table 17 details overall mean herbage magnesium contents in each year for each treatment. Where samples were taken on several dates in the one year, these have been averaged.

Table 17. Mean herbage magnesium concentrations.

	% Mg (on D.M. basis).		
Pasture treatment	1961	1962	1963
no potassium fertiliser	0.160	0.170	0.174
1 cwt muriate of potash/acre	0.156	0.156	—
2 cwt muriate of potash/acre	0.148	0.133	0.127

These figures show that herbage magnesium is depressed progressively by both increasing rates of usage and increasing numbers of applications of potassium fertiliser. As earlier discussed, this reduction in herbage magnesium with the consequent reduction in dietary magnesium intake by the grazing animal may be sufficient to explain the observed increase in hypomagnesaemia in sheep grazing these pastures.

Since the completion of the work described in Experiments 1, 2 and 3, the work with sheep of L'Estrange & Ayford (1964a) and Black & Richards (1965) has been published. Both of these reports dealt with experiments similar to the work described here, and results of these reports are entirely in agreement with the conclusions reached in this work. Both groups of workers found a slight decrease in the plasma magnesium concentration of ewes, attributable to the combined application of combined nitrogen and potassium fertilisers on the pasture. There was, however, no increase in the incidence of hypomagnesaemic tetany. They further found decreases in herbage magnesium similar to those described here.

There was only one case of hypomagnesaemic tetany during the course of Experiments 1, 2 and 3. This almost complete absence of the clinical form of the disorder entails the conclusions reached in these experiments being made entirely on a basis of the incidence of hypomagnesaemia. How far such conclusions can be applied to the incidence of clinical tetany is difficult to determine. However, based on our knowledge as it stands at the present time, the presumption is that any factor which increases the incidence of hypomagnesaemia, predisposes the animal to hypomagnesaemic tetany.

(b) Number of lambs suckled by the ewe.

Contrasting results were obtained in these experiments on the extent to which the number of lambs being suckled by the ewe affects the magnesium status of the dam. A comparison of ewes with single lambs and ewes with twin lambs in Experiment 1 showed no difference between these groups as far as their plasma magnesium levels were concerned. Yet a similar comparison in Experiment 3

suggested that ewes with twin lambs tended to have lower plasma magnesium concentrations. Any effect which the number of lambs being suckled might have is therefore not a consistent one. However, the findings of Experiment 3 are more in accord with the reports on field outbreaks where a high proportion of ewes affected by hypomagnesaemic tetany had twin lambs. The suckling of two lambs as compared with one by a ewe would naturally affect other physiological functions in the ewe, and it may be that some other parameter, not measured in this experiment, has a bearing on the development of clinical tetany, quite apart from a low plasma magnesium concentration.

(c) Age of the ewe.

In Experiment 1, where the effect of the age of the ewe on the development of hypomagnesaemia was studied, no difference was detected between two-year-old gimmers and five-year-old cast ewes with regard to mean plasma magnesium concentrations. The old ewes did, however, have lower plasma calcium values and this may have a bearing on a greater susceptibility to hypomagnesaemic tetany in old ewes. The one case of hypomagnesaemic tetany which did occur was in an old ewe, and analysis later showed it to have had, immediately prior to death, a low plasma calcium and low plasma magnesium concentration.

(d) Breed of the ewe.

In Experiments 2 and 3, a comparison was made between the mean plasma magnesium levels of Cheviot ewes and Half-Bred ewes. In both experiments Cheviot ewes consistently had lower levels than Half-Bred ewes, and this was particularly noticeable in the greater percentage in each year of Cheviot ewes with plasma levels below 1.00 mg Mg/100 ml. Only in Experiment 3 was there a

statistical difference between the groups. This would therefore suggest that, under the conditions of these experiments, Cheviot ewes were more susceptible to severe hypomagnesaemia than were Half-Bred ewes. In the absence of clinical tetany, however, no definite conclusion can be given on their relative susceptibility to hypomagnesaemic tetany.

(e) Variations in individual plasma magnesium concentrations.

Very close correlations were found in the flock of Cheviot ewes used in these three experiments between the individual plasma magnesium values on

(a) different dates in the same year

(b) the same date in two succeeding years

(c) the same date in two non-consecutive years.

Thus, although the mean plasma magnesium levels varied widely on various sampling dates over the three year period, the same ewes generally always had the higher values and conversely those with the lower values were usually the same ewes.

This finding may have great importance in that there is a possibility of ewes having an inherent susceptibility to hypomagnesaemia and thereby to tetany.

(f) Movement of a flock onto new pasture.

It is noteworthy that in the four flocks put onto the plot pastures in these three experiments, three of the four flocks suffered a fall in plasma magnesium levels within one to three days of being transferred to the new grazing ground. In every experiment this also represented the lowest mean level of plasma magnesium recorded over the experimental periods. This observation would be in accord with reports of field outbreaks where the cases of hypomagnesaemic tetany normally occur within a few days of the flock being transferred to

new pasture.

SECTION III.

Experimental work part 2.

The Possible Importance of Hypocalcaemia in the Development of Hypomagnesaemic Tetany.

(a) A comprehensive review and analysis of existing reports

(b) Experimental - Experiment 4.

Hypocalcaemia and hypomagnesaemic tetany in cattle.

As discussed earlier in the introduction (Section I) to this thesis, one of the complicating issues in the study of hypomagnesaemic tetany is the fact that there are many reports of ruminant animals experiencing severe hypomagnesaemia (below 0.5 mg Mg/100 ml) and yet remaining in good health. Animals which are affected clinically may have plasma magnesium concentrations which are no lower than those of animals which do not succumb to hypomagnesaemic tetany. These reports for cattle include those of Allcroft & Green (1938), Muth & Haag (1945), Brierem et al. (1949), Bartlett et al. (1954), Bartlett et al (1957), Ender et al. (1957), Simeson (1957), Kemp (1958a), Line et al. (1958), Rook & Balch (1958), Kemp et al. (1960), Birch & Wolton (1961), Storry (1961b) and McConaghy et al. (1963).

It is thus evident that severe hypomagnesaemia is not necessarily the only factor involved in the precipitation of clinical tetany. Several workers, e.g. Sjollema (1930a), Dryerre (1932), Todd & Thomson (1960)and Storry (1961b), have looked for further abnormalities in blood constituents which might influence the precipitation of clinical symptoms. In addition to the consistent finding of low plasma magnesium levels, only Sjollema (1930a) found any further abnormality, in that he also found a low mean plasma calcium concentration of 6.65 mg/100 ml in the 55 cases of hypomagnesaemic tetany he studied. Todd & Thomson (1960) and Storry (1961b) were, however, working with hypomagnesaemic cows which remained clinically normal, and their finding need not necessarily apply to hypomagnesaemic tetany. It remains unclear as to how many of the 42 blood samples analysed by Dryerre came from clinical cases, since he mentions

that "some came from cows in affected herds, but themselves showing no clinical symptoms."

Nolan & Hull (1941) in a report on twelve hypomagnesaemic tetany cases in cattle drew particular attention to the low plasma calcium concentrations found in these animals. The mean values for the twelve affected animals were 6.63 mg Ca and 0.90 mg Mg/100 ml plasma. There were five individuals with plasma calcium values below 6.0 mg/100 ml, five between 6.0 and 8.0 and only two animals above 8.0 mg Ca/100 ml. Allcroft (1947a,b) reported the high incidence of 76% of animals having hypomagnesaemia (< 1.7 mg/100 ml) which had a concomitant hypocalcaemia (< 8.4 mg Ca/100 ml). Seekles and Hendriks (1963) gave the opinion that "a decrease of plasma calcium in addition to hypomagnesaemia is essential for the development of clinical grass tetany." Marshak (1958) took the extreme view of questioning the concept that low plasma magnesium causes tetany and he suggested that hypocalcaemia was the primary cause.

To place the possible role of hypocalcaemia in tetany on a more quantitive basis rather than purely on opinions given in separate papers, a survey was carried out of the literature dealing with cases of clinical tetany in cattle for which both Ca and Mg concentrations in the plasma have been quoted. Table 18 summarizes these findings. Many other reports concerning clinical hypomagnesaemic tetany cannot be quoted in this context as plasma calcium analyses were not given.

Table 18. Plasma calcium and magnesium concentrations in 202 cases of clinical tetany in cattle.

Reference.	No. of cases	Plasma Ca (mg/100 ml)		Plasma Mg (mg/100 ml)	
		Mean	Range	Mean	Range
		Individual values given			
Sjollema(1930a)	55	6.65	3.90–9.50	0.45	0.16–1.16
Allcroft & Green(1934)	18	7.30	4.60–10.90	0.64	0.35–1.15
Metzger(1936)	1	7.50	—	0.15	—
Nicholson & Shearer(1938)	2	6.50	5.70–7.30	0.75	0.52–0.99
Nolan & Hull(1941)	12	6.63	4.90–8.35	0.90	0.10–2.46
Breirem et al.(1949)	3	6.10	5.60–6.90	0.46	0.32–0.55
McBarron(1952)	2	4.10	3.70–4.40	1.45	1.30–1.60
Inglis,Weipers & Marr(1954)	1	11.8	—	0.58	—
Ender et al.(1957)	2	6.30	5.10–7.50	0.30	0.20–0.40
Simesen(1957)	3	5.23	4.90–5.40	0.70	0.66–0.75
Weighton(1957)	1	6.30	—	0.60	—
Mershon & Custer(1958)	20	6.30	4.20–9.80	0.70	0.10–1.70
Hughes & Cornelius(1960)	2	6.50	6.00–7.00	0.63	0.60–0.67
Storry(1961b)	3	6.36	6.24–6.44	0.73	0.57–0.81
Rook(1963)	1	6.80	—	0.19	—
Mean for 126 cases		6.49		0.58	
		Individual values not given			
Hopkirk,Marshall & Blake(1933)	35	7.00	4.50–6.90	0.81	0.30–1.50
Weighton(1942)	13	7.40	—	1.00	—
Ender et al.(1948)	3	6.65	—	0.71	—
Bartlett et al.(1954)	6	—	6.80–8.00	—	<0.60
Bartlett et al.(1957)	1	≥8.00	—	<0.60	—
Smyth,Conway & Walsh(1958)	2	>8.00	—	0.70	—
Butler(1963)	11	5.97	2.91–8.11	1.27	0.58–1.75
Mean for 76 cases		6.95		0.87	
Overall mean for 202 cases		6.66		0.69	

It is obvious from Table 18 that some degree of hypocalcaemia has been in most cases a constant feature in reported outbreaks of clinical tetany. For a total of 126 cases reported in the literature where individual values were given, the mean plasma calcium and magnesium concentrations were 6.49 mg/100 ml and 0.58 mg/100 ml respectively. Sjollema (1930a) in his report on 55 cases of clinical tetany, found eighteen of the 55 affected cows to have a plasma calcium concentration below 6.0 mg/100 ml. A further twenty-two had values between 6.0 - 8.0 mg Ca/100 ml, and the remaining fifteen had values above 8.0 mg/100 ml. Another of the earliest papers on this subject quoted in Table 18 is that of Hopkirk et al. (1933), who did not present individual values but gave the mean plasma concentrations of 7.0 mg Ca/100 ml (range 4.5-9.6) and 0.81 mg Mg/100 ml for a total of thirty-five clinical cases of hypomagnesaemic tetany. These authors also gave the comparative figures for nine healthy cows on an affected farm. The mean calcium concentration for these nine was 9.2 mg/100 ml with no individuals below 8.0 mg/100 ml. Another early paper given in Table 18 is that of Allcroft & Green (1934) who presented information on eighteen cases of clinical tetany. The mean plasma calcium and magnesium concentrations were 7.30 and 0.64 mg/100 ml respectively. Only five of the eighteen cows had plasma calcium values above 8.0 mg/100 ml. Eight had values between 6.0 and 8.0 and the remaining five were below 6.0 mg/100 ml. The earlier work of Dryerre (1932) has been omitted from Table 18 since it is uncertain what proportion of the 42 "cases" quoted by him are from affected animals.

Following on these early reports, other workers have confirmed over the years, a generally occurring condition of hypocalcaemia accompanying

hypomagnesaemia in clinical tetany. Nolan & Hull (1941) dealing with twelve cases have already been mentioned. Ender et al. (1948) gave only mean values for a group of eight clinically affected cows. These were 6.65 mg Ca/100 ml and 0.71 mg Mg/100 ml plasma. Mershon & Custor (1958) in dealing with twenty cases of tetany, found only four animals to have plasma calcium levels above 8.0 mg Ca/100 ml, and eleven had values between 6.0-8.0 mg Ca/100 ml, with the remaining five cows being below 6.0. Butler (1963) recorded only the mean plasma values and the range for eleven cows affected by tetany. The mean plasma calcium concentration was 5.97 mg/100 ml (range 2.91-8.81) and the plasma magnesium was 1.27 mg/100 ml. (range 0.58-1.75).

Some of the papers quoted in Table 18 gave not only the plasma calcium and magnesium concentrations of cows with hypomagnesaemic tetany but also the analytical figures for unaffected animals in the same herd. These results are of interest in that they form a comparative basis on which to judge the relative importance of hypocalcaemia in tetany cases. Some of these reports recognised the importance of the reduced plasma calcium concentrations found in affected animals, and this was considered to be the prime feature in distinguishing an affected from an unaffected animal. For example, Bartlett et al. (1954) recorded a total of eighteen cows with plasma magnesium concentrations at or below 0.6 mg/100 ml of which five showed clinical signs of hypomagnesaemic tetany and two died suddenly. The plasma calcium concentrations of these eighteen cows was not recorded in detail, but it was regarded as noteworthy in the report that those cows which subsequently developed tetany had plasma calcium values in the range 6.8-8.0 mg/100 ml on samples taken 2-3 days before clinical symptoms

or death occurred. They also commented on that fact that where low plasma magnesium values were recorded in combination with plasma calcium values within the more normal range of 8.0-10.9 mg/100 ml, no clinical symptoms developed. Simesen (1957) reported three clinical cases for which the ranges of plasma calcium and magnesium values were 4.9-5.4 and 0.66-0.75 mg/100 ml respectively. In contrast three other unaffected animals had low plasma magnesium values below 1.0 mg/100 ml but normal plasma calcium values in the range 9.5-9.8 mg/100 ml. Similarly Hughes & Cornelius (1960) found that the two cows which developed tetany in a herd of sixteen had plasma calcium values of 7.0 and 6.0 mg/100 ml combined with 0.60 and 0.70 mg Mg/100 ml respectively. A further five unaffected animals in the same herd had equally low magnesium values of 0.52-0.75 mg/100 ml associated with normal plasma calcium levels in the range 9.0-10.0 mg/100 ml. Finally, Storry (1961b) reported no cases of clinical tetany in an experiment in which individual plasma magnesium values fell as low as 0.6-0.7 mg/100 ml but calcium concentrations remained in the normal range. In contrast, however, there were three cases of clinical tetany in cattle on an adjacent field at the same time. For these three animals, the plasma calcium ranged from 6.24-6.44 mg/100 ml, combined with plasma magnesium levels of 0.57-0.81 mg/100 ml.

It would therefore seem that considerable suspicion must fall on the plasma calcium level of a hypomangesaemic animal as being involved in the ultimate precipitation of clinical symptoms of hypomagnesaemic tetany. The results quoted in the first part of Table 18 are for 126 clinical cases in cattle for which individual figures are given in the original report. These have been summarised in Table 19 as percentage distributions within particular concentration

ranges of plasma calcium and magnesium.

Table 19. Percentage distribution of plasma calcium and magnesium concentrations of 126 cows with clinical tetany.

Plasma Ca (mg/100 ml)	Plasma Mg (mg/100 ml). 0.0-0.5	0.5-1.0	1.0-1.75	% of samples within each Ca concentration range.
< 4.0-6.0	18.8	12.6	4.7	36.1
6.0-8.0	24.5	18.8	1.6	44.9
> 8.0	8.7	8.7	1.6	19.0
% of samples within each Mg concentration range.	52.0	40.1	7.9	100.0

This table indicates that of these 126 cases reported in the literature, 81% were associated with plasma calcium concentrations below 8.0 mg/100 ml. With regard to magnesium, 92.1% of the cases were reported as having plasma magnesium values below 1.0 mg/100 ml.

In conclusion then, it would seem that in cattle, a large majority of the reported cases of hypomagnesaemic tetany had a concomitant hypocalcaemia, but this would not seem to be an invariable feature in that 19% of the cases were reported with plasma calcium concentrations above 8.0 mg/100 ml. However, it could equally be said from these figures that severe hypomagnesaemia is also not necessarily a constant feature in hypomagnesaemic tetany since 7.9% of the reported cases had a plasma magnesium above 1.00 mg/100 ml. In one case Nolan & Hull (1941), a figure of 2.46 mg Mg/100 ml is given for a case of hypomagnesaemic tetany.

Hypocalcaemia and Hypomagnesaemic Tetany in Sheep.

There are many reports in the literature, as discussed earlier in the introduction to this thesis, of sheep having severe hypomagnesaemia (below 1.0 mg/100 ml) without showing signs of hypomagnesaemic tetany. Section 2 of this thesis included results given for two separate flocks of twenty-four lactating ewes where the mean plasma magnesium levels were 0.99 and 1.13 mg/100 ml. This entailed over 50% of these ewes having values below 1.00 mg Mg/100 ml, with many values in the range 0.25–0.50 mg/100 ml for long periods, and yet no clinical signs of tetany appeared. A recent report of Hemingway et al. (1965) described a flock of forty-four lactating ewes with a mean plasma magnesium of 0.75 mg/100 ml of which only four had clinical tetany. Twenty of the ewes had plasma magnesium concentrations below 0.5 mg/100 ml on at least one sampling occasion and showed no signs of tetany. Other reports giving low plasma magnesium values in clinically normal lactating ewes include those of Inglis et al. (1959), Michael (1961), Owen & Sinclair (1961), Herd & Peebles (1962) and L'Estrange & Axford (1964a,b).

Similar to the situation described for cattle, hypocalcaemia, in association with hypomagnesaemia seems to be a common, indeed almost invariable, finding in the reported cases of hypomagnesaemic tetany. Table 20 details all the reports that could be found on cases of hypomagnesaemic tetany in sheep where the individual plasma calcium and magnesium values are given.

Table 20. Plasma calcium and magnesium concentrations in twenty-two cases of clinical tetany in sheep.

Reference.	No. of cases	Plasma Ca (mg/100 ml)		Plasma Mg (mg/100 ml)	
		Mean	Range	Mean	Range
O'Moore (1955)	2	6.00	—	1.20	—
Penny & Arnold (1955)	3	3.66	3.20–4.20	0.73	0.70–0.80
Inglis et al. (1959)	1	3.40	—	1.07	—
McAleese & Forbes (1959)	5*	6.42	6.40–6.50	0.51	0.40–0.60
Michael (1961)	1	2.80	—	1.60	—
Herd & Peebles (1962)	1	7.00	—	0.70	—
Ritchie et al. (1962)	1*	9.80	—	0.73	—
Hemingway & Ritchie (1963)	1	4.00	—	0.46	—
Hemingway et al. (1965)	4	5.13	3.79–5.89	0.40	0.25–0.50
L'Estrange & Axford (1964a)	2	4.85	3.70–6.00	0.36	0.20–0.52
L'Estrange & Axford (1964b)	1	4.16	—	0.58	—
Mean for 22 cases		5.31		0.66	

* Non-lactating sheep fed on low-Mg diets.

With the exception of a plasma calcium concentration of 9.8 mg/100 ml recorded by Ritchie et al. (1962) for a non-lactating ewe on a magnesium deficient diet, every reported case of hypomagnesaemic tetany was associated with a plasma calcium concentration below 6.5 mg/100 ml. Plasma magnesium concentrations were below 0.8 mg/100 ml for the majority of these affected sheep, but there were five sheep with a value above 1.00 mg/100 ml. It would therefore seem that on a percentage basis, with sheep, hypocalcaemia is a more consistent finding than severe hypomagnesaemia in cases of hypomagnesaemic tetany.

Some of these reports included comparisons between affected and non-affected ewes in the same flock. For example, Herd & Peebles (1962) contrasted a single clinical case of tetany in a lactating ewe which had plasma concentrations of 0.7 mg Mg/100 ml and 7.0 mg Ca/100 ml, with mean values for four healthy ewes in the same flock of 0.77 mg Mg/100 ml and 9.25 mg Ca/100 ml. The four clinical cases of tetany described by Hemingway et al. (1965) had a combined hypocalcaemia (mean 5.13 mg/100 ml) and hypomagnesaemia (mean 0.40 mg/100 ml). There were a further twenty ewes with plasma magnesium values below 0.5 mg/100 ml with no clinical signs of tetany. There were also four ewes in which tetany did not develop with plasma calcium concentrations below 7.0mg/100 ml associated with plasma magnesium values of below 0.65 mg/100 ml. Hemingway & Ritchie (1963) found, in a total of 389 ewes sampled in springtime at pasture, only two low plasma calcium values. These were associated with the only two cases of clinical tetany recorded.

It would therefore seem that a combined hypocalcaemia (below 7.0 mg/100 ml) and hypomagnesaemia (below 0.7 mg/100 ml) must therefore be considered to confer

a very high degree of risk of clinical tetany in sheep such as would not occur as a result of this degree of hypomagnesaemia alone.

Hypocalcaemia and hypomagnesaemic tetany in calves.

Similar to the situation described for cattle and sheep, hypocalcaemia has commonly been reported as an associated factor in the development of hypomagnesaemic tetany in milk-fed calves. For example, Parr & Allcroft (1953) reported that for eight calves with hypomagnesaemic tetany, the mean plasma calcium and magnesium concentrations were 6.2 and 0.6 mg Mg/100 ml. In some of these calves the plasma magnesium values had been as low as 0.4 mg/100 ml in the days prior to tetany but symptoms did not appear until the plasma calcium level fell. Much earlier Huffman & Robinson (1926) reported an associated hypocalcaemia in hypomagnesaemic calves, but later Duncan et al. (1935) and Huffman & Duncan (1936) considered the hypomagnesaemic tetany of calves to be unassociated with low plasma calcium values. Todd & Rankin (1959) also recorded hypomagnesaemia without hypocalcaemia (above 9.0 mg/100 ml) in clinical tetany in young calves. On the other hand, Parr (1957) and Smith (1957, 1958, 1961a) have reported combined low plasma calcium and magnesium concentrations occurring in hypomagnesaemia tetany cases in young calves.

Changes in plasma calcium and magnesium during the development of clinical tetany.

The findings presented above by extraction from published evidence refer to plasma concentrations at the onset of or immediately prior to the development of hypomagnesaemic tetany. The conclusions were that in the majority of cases, both cattle and sheep exhibit low plasma calcium and magnesium values at this time. It would obviously be of great interest to determine at what stage these

abnormalities develop. It is however difficult to obtain evidence regarding the plasma calcium and magnesium concentrations of animals on the few days prior to the onset of symptoms since it is impossible to foretell on which day a particular animal will develop clinical symptoms. Dealing firstly with sheep, there are four reports in the literature of experiments where blood samples had been taken from ewes within the four days prior to the onset of tetany. The results of these four experiments are given in Table 21, arranged according to the day on which the samples were taken, in relation to the day on which clinical symptoms of hypomagnesaemic tetany appeared.

Table 21. Changes in plasma calcium and magnesium concentrations during the development of clinical tetany in eight lactating ewes.

Reference.	Plasma Ca (mg/100 ml) Days before tetany					Plasma Mg(mg/100 ml) Days before tetany				
	4	3	2	1	0	4	3	2	1	0
Hemingway & Ritchie(1963)	6.80				4.00	0.45				0.46
Hemingway et al. (1965)		8.63	8.23		5.89		0.80	0.50		0.50
		7.98	7.02		3.79		0.40	0.30		0.25
	8.55	7.82		8.79	5.48	0.65	0.50		0.50	0.45
			9 .35		5.48			0.50		0.40
L'Estrange & Axford (1964a)	8.20	9.40	10.20	8.20	8.70	0.60	0.65	0.60	0.50	0.52
L'Estrange & Axford (1964b)	10.50	11.50	9.80	6.80	6.00	0.32	0.32	0.45	0.30	0.20
			5.50	6.00	4.16			0.58	0.56	0.58
Mean	8.51	8.69	8.35	7.45	4.81	0.63	0.63	0.49	0.46	0.42

The results have been arranged for all eight cases on each of the four days before tetany. These mean values for calcium and magnesium, rather than the individuals, give a clear picture of events leading up to the precipitation of the clinical attack. The striking feature disclosed by these results is the existence of low plasma magnesium values for several days before the onset of tetany. The mean fall over the four days was only from 0.63-0.42 mg/100 ml. In contrast, however, the mean plasma calcium concentration remained relatively high (8.51-8.35 mg/100 ml) until two days prior to tetany. There was a fall on the day before tetany to 7.45 mg Ca/100 ml. It was, however, only in the final twenty-four hours before tetany developed that there was a drastic fall in the plasma calcium concentration to a mean level of 4.81 when the symptoms appeared.

Comparable data for cattle are difficult to obtain, but a similar situation as that described for sheep is given by Ender et al. (1948) in a report on eight cases of hypomagnesaemic tetany in cattle. Two to five days before the onset of clinical symptoms, the mean plasma calcium and magnesium values of these cows were 9.9 and 0.72 mg/100 ml respectively, but when tetany developed, these mean values had changed to 6.65 mg Ca and 0.71 mg Mg/100 ml. There had therefore been no change in the plasma magnesium, but the plasma calcium concentration had fallen drastically.

Parr (1957) reported on a case of hypomagnesaemic tetany in a calf which had a plasma magnesium concentration of around 0.4 mg/100 ml for a 3 week period prior to the appearance of symptoms. The plasma calcium was 9.2 mg/100 ml when the calf was 9 weeks of age, 9.1 mg/100 ml at 10 weeks, 7.4 at 11 weeks and finally 6.0 mg/100 ml at 12 weeks when tetany occurred.

Effect of calcium and magnesium supplements.

An intriguing consequence of the development of hypocalcaemia in animals with severe hypomagnesaemia, is the effect that dietary supplements of calcium or magnesium have on the plasma constituents. McAleese & Forbes (1959) working with lambs fed indoors on a magnesium deficient diet, recorded that as plasma magnesium concentrations were reduced to 0.9-1.2 mg/100 ml, plasma calcium levels also fell rapidly to as low as 6.5 mg/100 ml. Additional dietary calcium supplements (and/or vitamin D) had no influence on either the plasma calcium and magnesium concentrations. Similarly, L'Estrange & Axford (1964b) found that lactating ewes on a low magnesium diet developed low calcium and magnesium plasma concentrations despite the fact that the diet was adequately supplemented with calcium. They did find however that giving dietary supplements of magnesium produced not only an immediate rise in plasma magnesium levels, but also the calcium concentration returned to normal within a few days. These authors considered that it was the low plasma magnesium that was primarily responsible for inducing the low plasma calcium concentration. Parr (1957) demonstrated a similar response in a calf with low plasma calcium and magnesium concentrations. Eight grams of magnesium (given orally as the carbonate) to the calf restored both the calcium and the magnesium plasma concentrations to the normal range within six days. He had previously been unable to raise the low plasma calcium level of a magnesium depleted calf by administering supplements of dietary calcium quite apart from the fact that a milk diet is rich in calcium in the first place. The diet in this experiment was adequately supplemented with Vitamin D. Smith (1961a) describes similar results for magnesium deficient

calves, by feeding magnesium supplements. Smith (1957,1958) had earlier shown that the severe hypocalcaemia (but not the hypomagnesaemia) in milk fed calves could be alleviated by high dietary levels of Vitamin D.

In conclusion, from the evidence which has been extracted from the literature on this subject, it is clear that the majority of cases of hypomagnesaemic tetany in cattle, sheep and milk fed calves exhibit a combined hypocalcaemia and hypomagnesaemia. In many circumstances, these ruminant animals seem to be able to experience severe hypomagnesaemia for long periods without signs of clinical tetany. There is good evidence to suggest that the actual onset of clinical signs of tetany is in some way associated with a rapid fall in plasma calcium concentration superimposed on an existing state of hypomagnesaemia. It may be that severe hypomagnesaemia in some way interferes with plasma calcium metabolism, as has been suggested by Blaxter & Sharman (1955), Newman & Newman (1958) and Smith (1961a). Support for this suggestion is given by the fact that alleviation of the hypomagnesaemia by dietary supplements of magnesium also alleviates the hypocalcaemia. Vitamin D would seem to be a complicating factor, although Vitamin D deficiency was presumably not present in these experiments. Where Vitamin D was successful in alleviating the hypocalcaemia of hypomagnesaemic animals, the level of supplementation used was well in excess of normal dietary requirements.

Experiment 4.

The inference from the foregoing evidence is that hypocalcaemia may act as a "trigger" mechanism in the development of clinical symptoms of tetany in hypomagnesaemic ruminant animals. It may, in fact, be one of several "trigger" mechanisms, any of which may induce an attack. Based on this hypothesis, Experiment 4 was an attempt to demonstrate, in sheep, a greater susceptibility towards hypomagnesaemic tetany in animals which had been subjected to an experimentally induced combined hypomagnesaemia and hypocalcaemia. Susceptibility was to be measured relative to animals with hypomagnesaemia alone.

The experimental induction of a combined hypocalcaemia/hypomagnesaemia is, in principle, difficult to achieve. Experimental diets may be devised which are deficient in calcium and equally diets can be constructed which are low in magnesium, but no practical diet can be produced which is deficient in both elements, or at any rate not in a quantity sufficient to nutritionally maintain a group of sheep.

A practical approach to the problem was considered to be an initial induction of deficiency of calcium followed by the superimposition of rapid induction of deficiency of magnesium.

It has been demonstrated in the past that the response of lactating ewes (or cattle) to a sudden introduction to lush spring pasture is normally a rapid drop in the plasma magnesium concentration, often into the severe hypomagnesaemic range, where hypomagnesaemic tetany may develop. The intention was to make use of this response in Experiment 4 in that pregnant ewes would be maintained indoors during pregnancy and parturition on a low calcium diet to induce hypocalcaemia.

Subsequently in the spring, they would be turned out suddenly onto good pasture, in the expectation that hypomagnesaemia would rapidly develop while the ewes were still in a hypocalcaemic condition. A comparable group would be given the same indoor diet supplemented with calcium, and on transference to pasture, they might be expected to develop hypomagnesaemia but with normal plasma calcium concentrations. Thus if hypocalcaemia were a feature in the actual precipitation of tetany, a higher incidence of clinical cases should be found in the former group.

Experimental design.

The experimental flock of sheep consisted of twenty-seven 4 year old Cheviot ewes. These sheep were housed at the end of November 1961 and for a preliminary period of 18 days, they were fed a normal hay and concentrate ration to allow them to become accustomed to indoor feeding and housing. After this preliminary period, the experimental ration was introduced gradually after 16th Dec. and the ewes remained on this ration for the following five months throughout pregnancy and parturition. This experimental ration was composed of 1 lbs. timothy hay per head per day and 1.3 lbs. of a concentrate mixture per head per day. The concentrate ration was increased to 1.5 lbs. per head per day from 10th February 1962. The concentrate was a mixture of 6.2 parts of maize to 1 part of decorticated groundnut meal, plus .77% salt added. These foods were chosen as being low in calcium, and timothy hay was similarly chosen as being the lowest in calcium of several batches of hay available.

Nineteen of the ewes received this diet with no further additions and the

remaining group of eight ewes were given this diet plus a supplement of 7.5 gms. calcium carbonate per head per day. This supplement was equivalent to an additional 3 gms. of calcium per day and was calculated to be sufficient, in addition to the diet, to give them an adequate daily calcium supply. This group of eight ewes were therefore considered as the control group of 'normal' ewes.

Lambing took place over the period 17th March to the 10th April. Appendix 3(a) details the date of lambing of each ewe, the number of lambs born and the number of lambs surviving after a period of 4-5 weeks. 55% of the ewes gave birth to twin lambs, but only 26% of the ewes were suckling twin lambs on 4th May.

Over this period of indoor feeding, blood samples were taken from each ewe on 19 occasions. Over the first three months of the feeding period, the blood sampling was carried out at roughly 14 day intervals, but during and after lambing the ewes were sampled more frequently at 3-7 day intervals. Analyses were made on these samples for calcium and magnesium. The feeding stuffs used were also analysed for magnesium and calcium contents.

On the 7th May, 1962, the indoor feeding was stopped and both groups of ewes were transferred to good pasture grass, where they grazed until the termination of the experiment 15 days later. Blood samples were taken from each ewe over this grazing period on the 8th, 9th, 11th, 15th and 22nd May.

Results.

Ca content of the indoor diet.

The calcium and magnesium content of the feeding stuffs used in this experiment are given in Table 22.

Table 22. Calcium and magnesium contents of dietary constituents.

feeding stuff.	amount fed daily for first 56 days. (g)	Ca% (on air dry basis)	Mg%
hay	453	.22	.09
maize	509	.016	.05
groundnut	82	.09	.31
salt	4.5	---	---

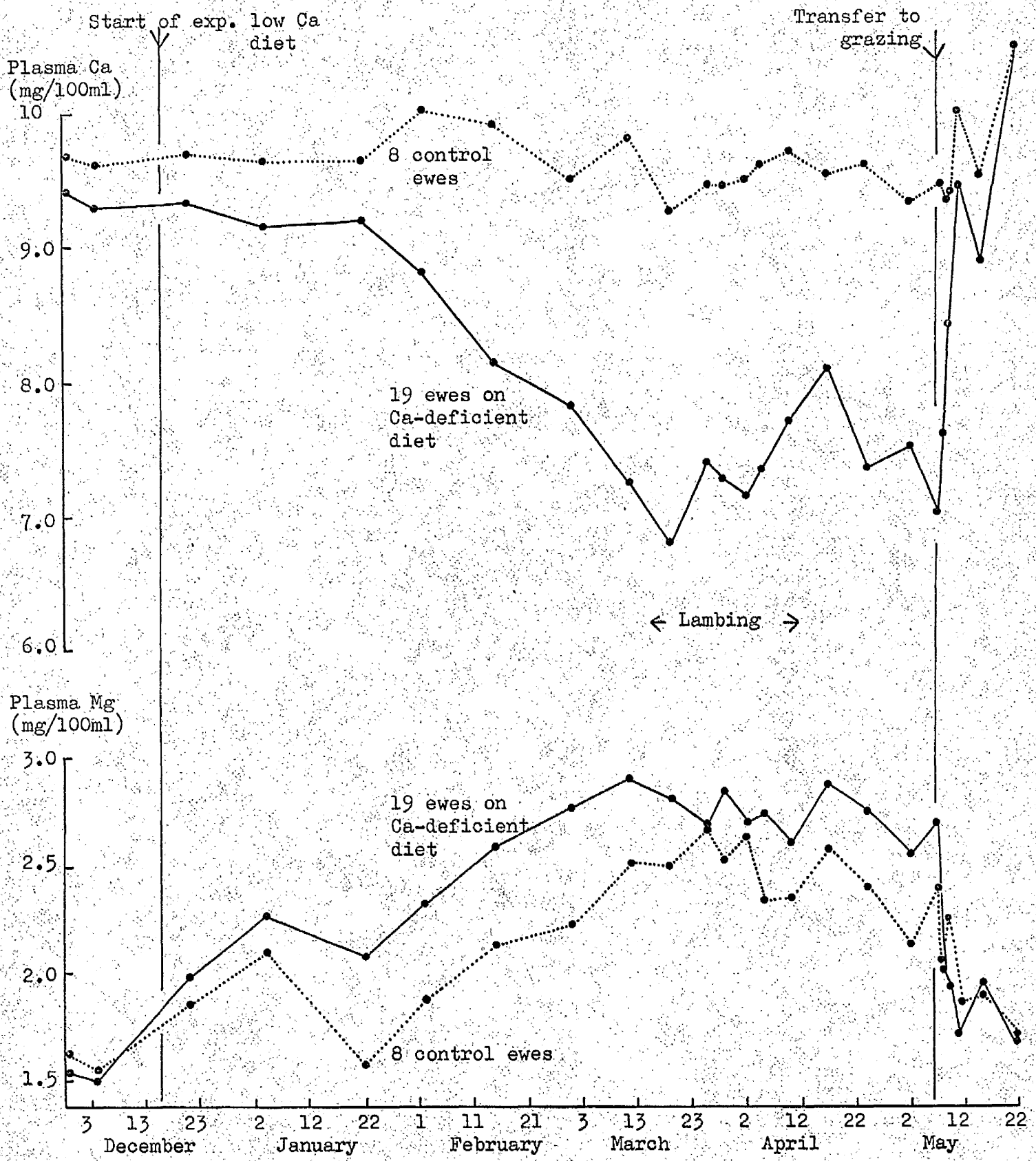
Fed at the rate given, these foods supplied daily to each ewe approximately 1.1 g. calcium and 0.9 g. magnesium. The group receiving the 7.5 g. calcium carbonate daily supplement would therefore receive approximately 4.1 g. calcium and 0.9 g. magnesium per head per day.

Plasma analyses.

The plasma calcium and magnesium concentrations of each ewe in the group fed the low calcium diet are given in Appendix 3(b) for the twenty-five sampling occasions of the experiment. The mean values for the group are also given. The corresponding values for the control group over the same period are given in Appendix 3(c). The mean plasma magnesium and calcium levels of both groups are also shown graphically in Fig. 14.

At the commencement of the experiment over the preliminary 18 day period, both groups had similar mean plasma calcium values in the range 9.3-9.7 mg/100 ml. After the introduction of the experimental rations on 16th December, the group of 19 ewes on the deficient diet maintained a normal mean plasma

Fig. 14 Mean plasma calcium and magnesium concentrations of Cheviot ewes on a diet deficient in calcium (Experiment 4).



calcium level for the first 39 days up to the 22nd January, but thereafter the mean level of this group fell progressively over the following seven weeks to a minimum mean level on the 19th March of 6.8 mg Ca/100 ml. The range of the individual concentrations on this date was from 4.46-8.69 mg Ca/100 ml. Throughout this period the control group given the calcium supplement maintained a mean calcium concentration within the normal range, varying from 9.26-10.02 mg Ca/100 ml.

After lambing commenced on the 17th March, there was a sudden rise in the mean plasma calcium level of the unsupplemented group, as evidenced by the sample taken on the 26th March. This rise continued erratically over the next five sampling occasions until the mean level of this group was above 8.0 mg Ca/100 ml on the 21st April, which would represent fourteen days after the average lambing date. The trend was then reversed, and the mean calcium level again fell erratically and on the 7th May when the indoor feeding was terminated, the mean of the unsupplemented group was 7.02 mg Ca/100 ml. One ewe in the group given no supplement developed milk fever and died on the 27th March, two days after lambing. It had a plasma calcium value of 4.62 on the previous sampling date. Plasma magnesium was normal. During and after lambing, the mean plasma calcium of the group given calcium supplement remained constant at about 9.5 mg/100 ml, and at the end of the indoor feeding period, the mean level was 9.46 mg/100 ml. On every sampling occasion after the 2nd February, there was a highly significant difference between the plasma calcium levels of the two groups.

The mean plasma magnesium concentrations of the two groups were initially

very similar within the range 1.5-1.6 mg/100 ml at the start of the experiment. After the introduction of the experimental rations the mean levels of both groups rose fairly steadily over the winter months until just prior to lambing, the means on the 12th March being 2.51 and 2.90 mg Mg/100 ml for the supplemented and unsupplemented groups respectively. The greater increase in mean plasma magnesium shown by the unsupplemented group just fails to be significantly greater than the increase shown by the control group. Thereafter the mean plasma magnesium level of both groups displayed erratic changes over the lambing period, and on the last day of the indoor feeding on the 7th May, the mean levels were 2.69 and 2.43 mg/100 ml for the unsupplemented and supplemented groups respectively.

On going to pasture on 7th May, there were sudden changes in both plasma calcium and magnesium levels. The mean plasma magnesium level of both groups fell rapidly within the first twenty-four hours of pasture grazingto about 2.00 mg/100 ml. In the following fourteen days, further falls occurred and on the 22nd May, when the experiment was terminated, the group previously given no calcium supplement had a mean plasma level of 1.68 mg Mg/100 ml, as compared with the similar level of 1.71 mg Mg/100 ml for the group previously given calcium supplements. Only one ewe (No. 87 in the unsupplemented group) had a plasma concentration below 1.00 mg Mg/100ml on this date.

The mean plasma calcium concentration of the unsupplemented group was rapidly restored to the normal level of 9.5 mg/100 ml within four days of the ewes going to pasture. Both groups rose over the following 11 days to similar mean plasma levels of 10.45 mg Ca/100 ml on the final day of the

experiment on 22nd May.

There were no cases of hypomagnesaemic tetany in either group over this grazing period.

Discussion.

The severe fall in plasma calcium concentration suffered by the group of ewes given no supplement demonstrates the insufficiency of the 1.1 g. calcium supplied by the experimental ration. A recent estimate of the calcium requirements of pregnant ewes is given by the Agricultural Research Council (1965) as 4.5 g./day in the second month of pregnancy, rising to 7.6 g./ calcium/day at the end of pregnancy. These figures are for an average sized lowland ewe with one foetus. The diet given in this experiment therefore supplies less than a quarter of the estimated calcium requirements. The ewes in this group presumably withdrew calcium from their skeletal reserves over this period, but after the initial 40 days of the experiment the rate of withdrawal was insufficient to maintain the plasma calcium concentration within the normal range. It is of interest to note that the calcium intake of the group given a supplement of 7.5g calcium carbonate would still be below the recommended requirements given by the A.R.C. (1965), but presumably if there was a slight deficit, this was amply met in their case by a limited withdrawal of skeletal reserves.

The aim of the first part of this experiment was therefore achieved in that on the 7th May when the ewes were put to pasture with their month-old lambs, one group of ewes were markedly hypocalcaemic (mean 7.02 mg Ca/100 ml) whereas a group which had been comparably maintained, had normal plasma calcium

concentrations. These contrasting calcium concentrations were associated with normal mean magnesium levels in both groups. On going to pasture, however, the falls in the plasma magnesium levels were not as severe as might have been expected and the lowest mean values recorded of 1.68 and 1.71 mg Mg/ 100 ml for the two groups on 22nd May are well above the level at which hypomagnesaemic tetany might be expected to appear.

Coupled with this non-attainment of severe hypomagnesaemia, the hypocalcaemia which existed prior to the transfer to pasture was rapidly corrected within two - four days. Therefore by the time a slight degree of hypomagnesaemia had developed, the existing hypocalcaemia had disappeared. The aim of the second part of the experiment which had been to induce hypomagnesaemia in hypocalcaemic ewes, was therefore not achieved. The hypothesis that a low plasma calcium concentration might play an important part in the precipitation of the clinical symptoms of hypomagnesaemic tetany remains unproven.

Despite the final outcome, some interesting information emerges from the results obtained during the indoor period when the ewes were on experimental rations. It can be seen from Fig. 14 that hypocalcaemia developed progressively over the pre-lambing period 22nd January - 19th March in ewes fed on a calcium deficient diet. This was followed, however, by a rise in the mean plasma calcium level, continuing erratically over the period 19th March - 17th April, which is coincident with the period over which lambing occurred. After the 17th April the mean plasma calcium level of the group again fell erratically over the next 20 days. The diet and the consequent dietary calcium supply did not vary over this period. Therefore, since these unexpected fluctuations in the

mean plasma calcium level of the unsupplemented group coincided with the period over which lambing was spread, it was suspected that there was some direct association between these two factors. However the long period of 24 days over which lambing is spread tends to obscure any true association that may be present between the time of parturition and plasma calcium concentrations.

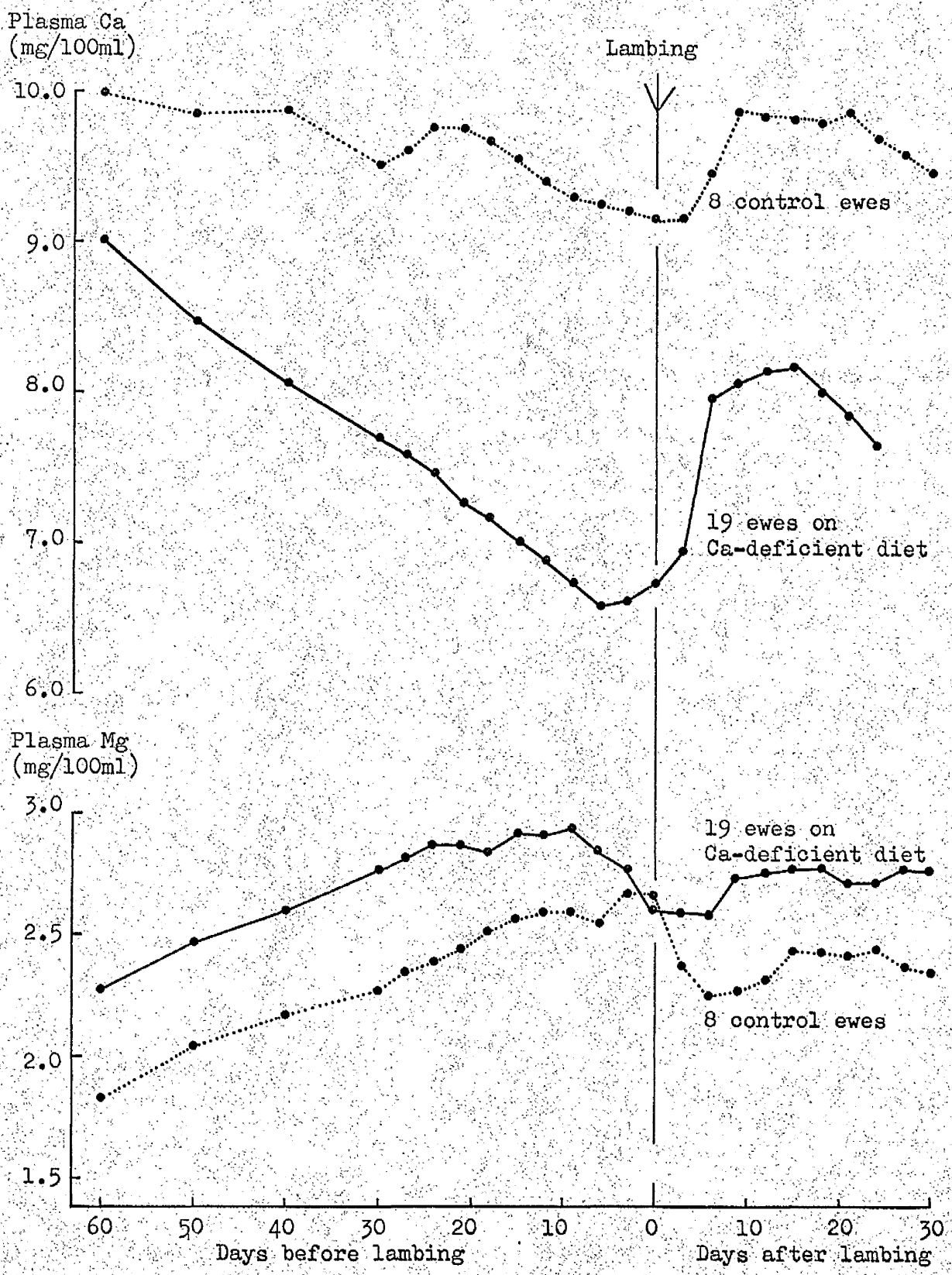
To clarify this situation, the results for the individual plasma calcium and magnesium levels were rearranged according to the lambing date of the individual, and not, as previously, according to calendar date. Lambing date is therefore designated as day 0, and blood samples taken for example 4 days before lambing and seven days after lambing would then represent day - 4 and day+7 respectively. These rearranged results were then interpolated to give an estimated plasma calcium and magnesium concentration for each ewe on every third day during the 30 days prior to lambing and on every third day for the 30 days after lambing. Interpolated figures were also worked out for days representing 60, 50 and 40 days before lambing. These individual figures were then arranged for the group of 19 unsupplemented ewes and for the group of 8 ewes given calcium supplements, and these mean interpolated plasma calcium and magnesium concentrations are given in Table 23.

Table 23. Mean plasma magnesium and calcium concentrations of Cheviot ewes, rearranged on a basis of lambing date.

Days before lambing.	19 ewes given no supplement.		8 ewes given cal. supplement.		Days after lambing	19 ewes given no supplement		8 ewes given cal. supplement	
	Ca.	Mg.	Ca.	Mg.		Ca.	Mg.	Ca.	Mg.
-60	9.03	2.27	10.00	1.83	0	6.73	2.60	9.18	2.66
-50	8.48	2.47	9.85	2.05	3	7.35	2.61	9.17	2.37
-40	8.08	2.60	9.85	2.17	6	7.96	2.59	9.46	2.25
-30	7.68	2.77	9.00	2.28	9	8.05	2.74	9.85	2.27
-27	7.57	2.82	9.61	2.34	12	8.14	2.77	9.85	2.32
-24	7.45	2.88	9.75	2.39	15	8.16	2.78	9.80	2.44
-21	7.28	2.88	9.78	2.44	18	8.00	2.79	9.80	2.44
-18	7.18	2.85	9.67	2.52	21	7.84	2.72	9.85	2.42
-15	7.02	2.92	9.55	2.57	24	7.65	2.72	9.69	2.43
-12	6.88	2.92	9.40	2.60	27	7.40	2.77	9.59	2.39
-9	6.74	2.95	9.32	2.59	30	7.24	2.77	9.46	2.34
-6	6.56	2.85	9.26	2.56					
-3	6.59	2.78	9.24	2.68					

The same data are shown graphically in Fig. 15. This demonstrates more precisely than the original data, the true sequence of events around the time of parturition. The group of 19 ewes given no calcium supplements suffered a

Fig. 15 Mean plasma calcium and magnesium concentrations of 2 groups of Cheviot ewes rearranged according to lambing date.



continuous steady fall in mean plasma calcium concentration from 60 days before lambing to within 3-6 days before lambing, when the lowest mean level of 6.60 mg Ca/100 ml was reached. It is of interest to note that this mean level is considerably lower than the lowest level recorded on any calendar date. On day 0, the date of lambing, the mean plasma calcium of this group started to rise and over the six days after lambing, a very rapid rise occurred in the mean level to 8.0mg Ca/100 ml, followed by further small increases up to 15 days post partum when a maximum of 8.16 mg Ca/100 ml was attained. Thereafter the mean level fell smoothly, until, 30 days post partum it had again fallen to the low level of 7.24 mg Ca/100 ml. Although the control group of 8 ewes given daily calcium supplements maintained a mean plasma calcium level within the normal range, over this period, as shown in Fig. 15, it tends to show to a much smaller degree similar changes to those found in the unsupplemented group. For example it shows a slight decrease in mean plasma calcium level over the 60 day period prior to lambing, followed by a rapid rise in the mean level immediately after parturition, and finally a further slight fall in mean plasma calcium level 20 days post partum.

The mean plasma magnesium concentration of the unsupplemented group varied more or less inversely to the mean plasma calcium levels, in that when the calcium level was falling, the plasma magnesium level was rising and vice-versa. Thus from 60 days before lambing to within 9 days of lambing, the mean plasma magnesium level of this group rose progressively and evenly from 2.27 mg/100 ml to an unusually high mean value of 2.95 mg/100 ml. From within 9 days of lambing to the day of parturition, the mean plasma magnesium level fell to

2.60 mg/100 ml. After lambing, the mean level remained at this level for 6 days, rose slightly over the following 10 days and remained then around a mean of 2.77 mg/100 ml. The group of eight ewes given calcium supplements showed very similar changes in mean plasma magnesium to those described above, except that their mean level was consistently 0.5 mg Mg/100 ml lower than that of the unsupplemented group. They also showed little fall in mean level before parturition, but rather, they suffered a fall over the six days after parturition.

This evidence presented in Fig. 15 (and Table 22) is hard to explain. Referring to the A.R.C. (1965) recommendations for the daily calcium requirements of sheep, a figure of 4.5 - 7.6 gms. Ca/day was given for pregnant ewes.

Estimates for the calcium requirements of lactating ewes are given as 14 g. Ca/day for the first month of lactation, falling to 7.8 g. Ca/day in the 4th month of lactation. These figures for pregnancy and lactation are both based on a 50 Kg. sheep with a single lamb. The requirements of the lactating ewe for calcium are therefore approximately twice as great as the requirements for pregnancy. The picture presented by Fig. 15, however, is one of hypocalcaemia developing progressively throughout pregnancy, and yet a rapid rise in plasma calcium levels during the first 20 days of lactation, when the calcium requirements of the ewe are quoted as being twice as great as in pregnancy. The calcium dietary intake did not change over this period. It may, however, be that the calcium requirement for lactation is initially much lower, when the lambs are newly born and taking only small amounts of milk. If, on the other hand, the calcium requirements are as suggested by the A.R.C. (1965), some stimulus, operating only after parturition, must have increased the internal

release of skeletal reserves of calcium or alternately caused a more efficient gut absorption of ingested calcium. The main regulator of calcium release from the bone is the parathyroid gland, and a drop in serum calcium concentrations is considered to be the direct stimulus for an increase in parathyroid activity. Yet, over the 60 days prior to parturition in this experiment, the group of unsupplemented ewes were unable to maintain normal plasma calcium concentrations and it is therefore presumed that because of the low serum calcium concentration, parathyroid activity is at its maximum. After parturition, when the mean plasma calcium of the group rises at a time when its calcium requirements are doubled, the inference must be that some other stimulus has influenced the plasma calcium concentration either directly or indirectly. Three possible modes of action would be, a direct stimulus to release further skeletal reserves of calcium, a stimulus on the parathyroid increasing its activity, or possibly as mentioned above a stimulus for more efficient gut absorption of calcium. The nature of such a stimulus is not known, but one obvious possibility is the change in circulating oestrogens which occurs at parturition in sheep. For example the high level of circulating oestrogens during pregnancy would tend to encourage growth of bone tissue by virtue of its protein anabolic action, and when the level of circulating oestrogens falls at parturition, catabolic activity may then increase.

Two further inferences can be drawn from Fig. 15, both in connection with the time of occurrence of milk fever cases and hypomagnesaemic tetany cases. It is recognised that the majority of milk fever cases in ewes occur immediately prior to or at the time of parturition. The graph of the plasma calcium level of hypocalcaemic ewes in Fig. 15 gives the lowest mean level immediately prior to parturition, which would be the time of greatest susceptibility to

milk fever. This agrees with clinical observations in practice.

Hypomagnesaemic tetany in sheep is normally encountered 2 - 4 weeks after lambing. Reference to Fig. 15 shows this to be the time when the plasma calcium levels of both groups of ewes in this experiment began to fall again. It may therefore be that, if hypocalcaemia was involved in the precipitation of clinical hypomagnesaemic tetany, the tendency for the serum calcium to rise post partum would protect ewes from tetany for the first 14 - 20 days after lambing, and only when the serum calcium level falls again, 14 - 20 days after lambing, would hypomagnesaemic tetany be liable to occur.

Section IV.

Experimental Work Part 3.

The Prevention of Hypomagnesaemic Tetany in Ruminant Animals.

Present methods of prophylaxis.

(a) Introduction - A review of past experiments into prophylaxis.

(b) Experiments 5 & 6 - The use of daily dietary magnesium supplements for sheep.

(c) Experiment 7 - The use of pasture dressings of magnesium fertiliser for sheep.

Rumen heavy pellet therapy as a method of prophylaxis.

(a) Experiment 8 - The use of magnesium oxide rumen pellets.

(b) Experiment 9 - The development of a heavy rumen pellet containing metallic magnesium.

(c) Experiments 10 - 12 - Trials in cattle of a metal magnesium rumen pellet.

(d) Experiments 13 - 15 - Trials in sheep of a metal magnesium rumen pellet.

Most prophylactic measures recommended for hypomagnesaemic tetany are based on the provision of supplementary dietary magnesium in one form or another. The three principal methods used at present are (a) the inclusion of magnesium salts, normally in a palatable concentrate food (b) the provision of magnesium salt "licks" which the animal is expected to take voluntarily (c) the spreading of magnesium fertilisers on the pasture. There is also the possibility of dosing the animal by hand with a magnesium salt solution, but this is in commercial practice a laborious method to adopt.

A certain measure of control can be achieved by suitable husbandry techniques. For example it is beneficial to make the changeover for dairy cows from indoor feeding to pasture grazing as gradual as possible to eliminate the sudden dietary change involved. It may help to avoid initially, young heavily fertilised pastures until the herd is accustomed to the grass diet. Some farmers take the negative attitude that if they consider fertilisation of pasture to be a causative agent for hypomagnesaemic tetany, they then avoid using more than a bare minimum of fertilisers on grassland. This approach is perhaps to be deprecated since it constitutes a barrier to the intensification of grassland production. For beef cows, Inglis et al. (1954) demonstrated the advantage of feeding hay as a prophylactic measure, and Allcroft (1947c) reported a similar effect by feeding hay and cabbages. Pook (1955) describes the prevention of tetany in sheep by confining their grazings to older leys.

There is now considerable evidence to indicate that supplementary dietary magnesium will alleviate hypomagnesaemia and hypomagnesaemic tetany. There is, however, a lack of certainty as to the degree of protection afforded and also

the quantity necessary to afford protection. Cunningham (1934) first reported that the serum magnesium concentration of cattle and sheep could be increased by the oral administration of magnesium compounds. In the following year, Blakemore & Stewart (1935) confirmed that 31g. magnesium oxide (= 18g. magnesium)/ head/day had a preventive effect on the occurrence of seasonal hypomagnesaemia in beef cattle. Allcroft & Green (1938) working with similar animals, found 45g. MgO/day (= 27g. magnesium)to have only a slight beneficial effect on the low serum magnesium levels found in December. Allcroft (1947c) therefore increased the quantity used to 160g. MgO/day (= 96g. magnesium) which alleviated the fall in serum magnesium but did not succeed in maintaining 'normal' values. In the following years many workers adopted a standard quantity of 2ozs. of calcined magnesite/head/day (= approx. 30g. magnesium since calcined magnesite is normally 87% MgO). For example Allcroft (1954), Jevnaker (1955), Seekles & Boogaerdt (1955, 1956) and Line et al, (1958) demonstrated the prophylactic value of this 2 ozs. quantity in dairy cow herds. Even although this quantity is large in relation to the dietary requirements of the dairy cow, there are many reports of clinical tetany cases occurring when the supplement was in use. For example Seekles & Boogaerdt (1956) in a large scale survey of over 10,000 cows, found a 2.8%incidence of tetany in cows given no supplement, and a 0.8% incidence in cows given 50 g. MgO/day. Thus although the incidence was reduced, it was not eliminated. Another example is that of Allcroft(1953) who recorded the failure of 2 ozs. calcined magnesite to maintain normal plasma magnesium and its failure to prevent deaths from hypomagnesaemic tetany. The possible use

of supplementary magnesium oxide in quantities smaller than the standard "2 ozs" has seldom been investigated. O'Moore (1955) reported that an intake of 12 g. magnesium daily will considerably reduce the incidence of hypomagnesaemic tetany in dairy herds. Presumably the fact that a supplement of 2 ozs. calcined magnesite does not afford 100% protection in cattle has engendered the belief that any less a quantity would inevitably provide even less protection. It should be said however that there is no evidence to suggest that smaller quantities would have any less satisfactory effect or indeed that 100 g. supplementary magnesium oxide would afford any greater degree of protection. Care (1960a) points out that it is important to avoid an excess intake since quantities over 6 ozs. calcined magnesite/day for cattle will cause scouring. Irrespective of the quantity of calcined magnesite to be given, it is essential that it be given daily since there is no carry over of a protective effect from day to day nor is there any possibility of magnesium in excess of requirement being stored in the animal body (Allcroft, 1960). The inclusion of 2 ozs. calcined magnesite does tend to reduce the palatability of food, particularly when cows go to pasture, when they may refuse to eat supplemented concentrates (Allcroft 1947c). Allcroft & Littlejohn (1961) found that supplemented sugar beet pulp was more readily eaten when dampened, but if the food was to be fed dry, a granular form of calcined magnesite mixed with the food was much preferred by cattle. In their experiments, 90% of the concentrates supplemented with 2 ozs. of powdered calcined magnesite was rejected by cattle.

This 2 ozs. quantity of magnesite which is equivalent roughly to 30 g. of magnesium is in excess of the total calculated dietary requirements for cattle.

Blaxter & McGill (1956) quoted 12.75 gms. magnesium as being the daily requirement for a 1200 lbs dairy cow giving 4 gallons of milk/day. A more recent estimate given by the A.R.C. (1965) is a daily requirement of 20.1 gms magnesium for an 1100 lbs cow giving 4.4 galls of milk/day. The A.R.C. (1965) further suggest that any individual variation or interfering factor such as a change of diet or low dietary availability, is amply covered for by allowing a margin of safety of a further 2 g. magnesium/day in addition to the daily requirement of 20.1 g/day. This would give a total requirement figure of 22 g. magnesium/day for a cow as quoted. The results of Rook (1961, 1963) would place the requirement below this figure. He found that an experimental ration had to contain less than 2.0 g. of magnesium before it caused the development of hypomagnesaemia in a lactating cow (milk yield not given). Daily supplements of 5 g. MgO, prevented further falls and 10 g. MgO, restored plasma magnesium levels to normal. The National Research Council (American) recommendations (1957, 1958a,b) are also of a lower order. They give a figure of 6 g.Mg. as being adequate for a 500 kg. cow, irrespective of circumstances such as lactation etc. This would therefore suggest that a dietary supplement of 30 gms. magnesium, in addition to magnesium that would already be present in a diet, is an unnecessarily high level of supplementation.

The addition of supplementary magnesium salts to concentrate mixtures for sheep is also a recommended procedure, (Allcroft, 1960). The quantities used are generally in the order of $\frac{1}{4}$-$\frac{1}{2}$ ozs calcined magnesite/head/day, but there is no experimental evidence to confirm its effectiveness in practice. This range of quantities has probably been derived from consideration of the body size of sheep relative to cattle. Allcroft (1960) found that ewes, which readily ate

a concentrate supplemented with this quantity of calcined magnesite, scoured severely. It was one of the aims of the work to be described in this Section to test experimentally the effectiveness of ¼ ozs of calcined magnesite in controlling hypomagnesaemia in lactating ewes at grass. Pook (1955) has reported the failure of calcined magnesite in amounts up to 2 ozs, mixed with oats and the failure of mineral licks to control an outbreak of hypomagnesaemic tetany in sheep.

An alternative method of directly supplementing the diet of grazing cattle or sheep is the application to the pasture of 20 - 50 lbs MgO/acre as a dust which adheres to the herbage. This is applied a few days before grazing commences and the cattle thereby inevitably consume some calcined magnesite as they graze the pasture, although the effect lasts only for the initial grazing. This method has been demonstrated by Grund (1961) and Todd (1961b,1964) to be effective in raising the plasma magnesium concentrations of lactating cattle at pasture in the spring. It also has the particular advantages of overcoming any refusal on the part of the animal to eat supplements, and ensuring a continuous intake of magnesium.

The main alternative method of prevention of hypomagnesaemic tetany in use at the present time is the application of magnesium fertilisers to pasture land. Magnesium applied in this way becomes incorporated in the soil, thus raising the soil exchangeable magnesium. Grass plants growing on such a soil thereby take up a greater quantity of magnesium, and the higher herbage magnesium content in

turn means a higher dietary magnesium intake for an animal grazing this herbage. The effect of such treatments on the magnesium content of the herbage was discussed earlier in Section I. Reference back to Table 3 given on page 39 will show the response of herbage magnesium to increasing rates of application of magnesium fertiliser. These results, based on the work of several investigators would suggest that there is a more or less proportional increase in herbage magnesium content for increasing amounts of fertiliser applied. It is only when applications were above 300 lbs magnesium/acre (= 5 cwts magnesium oxide/acre) that the pasture magnesium content is consistently above the level of 0.20% Mg.

The effect of these rates of pasture applications on the plasma magnesium concentrations of grazing lactating cows was also examined in some of the investigations quoted in Table 3. For example the biological effects of applications of 0.5, 6, 9, 10 and 25 cwts were studied by Birch & Wolton (1961); Smyth et al. (1958); Birch & Wolton (1961); Parr & Allcroft (1957) and Bartlett et al. (1954) respectively. In every case complete protection against hypomagnesaemic tetany was afforded and very little hypomagnesaemia was recorded. In all these experiments, these results as quoted were found in comparison with control groups which developed severe hypomagnesaemia and in some cases where hypomagnesaemic tetany occurred in the control group. This would therefore seem to be an effective prophylactic measure on suitable sites, although the lack of response on certain soils (Collins 1960) may entail a poor biological protective effect in some cases, particularly where a lower rate of application (0.5 cwts/acre) is being used. The only advantage of applying the high rates of 10 and 25

cwts. calcined magnesite/acre is that they have a persistent effect over several years (Bartlett et al., 1954; Parr & Allcroft, 1957).

The one disadvantage of this method of control for hypomagnesaemic tetany is its unsuitability on farms where the cattle are grazing extensively over a large acreage of ground. It is economically and practically impossible to treat more than a small acreage with calcined magnesite. An alternative method of applying magnesium fertiliser which obviates this difficulty is the use, on soils which have a definite lime requirement, of magnesian limestone in place of ordinary lime. This is economically possible since a dressing of lime would be applied in the normal course of events. Reith (1954) and Stewart & Reith (1956) demonstrated an increase in herbage magnesium content as a result of dressing pasture with 2 tons of magnesian limestone/acre. Parr & Allcroft (1957) investigated the use of 2.5 tons of magnesian limestone/acre and found it less effective than dressings of 25 cwts calcined magnesite/acre. Herbage magnesium contents increased only 20% in the first year as a result of this treatment, and slight hypomagnesaemia still occurred. Ogg (1960) reported the extensive and successful use of such dressings of magnesian limestone in commercial practice.

There are no reports of the use, effective or otherwise, of pasture magnesium fertilisation in the control of hypomagnesaemia and tetany in sheep, and an experiment on this aspect was therefore included in the work to be described in this section.

Experimental.

It is evident from the past work reviewed above that although the various methods of control and prevention of hypomagnesaemic tetany have received considerable attention in experimental work with cattle, no comparable work has been described for sheep. This is despite the fact that in commercial practice, these methods of control are widely applied in sheep husbandry.

It is therefore proposed in this Section to describe investigations carried out on the effectiveness of the present prophylactic measures for hypomagnesaemic tetany in sheep. The methods chosen for study were (a) the direct administration of magnesium oxide as a dietary supplement (b) the use of pasture applications of magnesium oxide applied either as a fertiliser at high and low rates of use or as a pasture dust.

Even where such methods of control can be shown to be effective, there are many circumstances where they are difficult to use in practice. It was therefore decided to investigate the possibility of devising a new method of magnesium supplementation for both sheep and cattle, which would be easier to use under commercial farm conditions. This involved the design of a magnesium-containing rumen pellet which could be administered to the animal by a oesophageal balling gun. It was designed to dissolve slowly in the rumen thereby supplying, continuously and regularly, supplementary magnesium to the animal. The active life of such a pellet should be sufficiently long to protect the animal over a period of risk (e.g. a spring grazing season).

Experiments 5 & 6.

Experiments 5 and 6 were planned to investigate the prophylactic effect in the control of hypomagnesaemia in sheep of dietary supplements of magnesium oxide given as a daily drench. Although this type of supplement would normally be given mixed with supplementary food, daily drenching was employed in these experiments since it ensured that each ewe received a known amount every day and also that each ewe received the same quantity. Supplementation via concentrates is an uncertain method to use experimentally since some ewes inevitably eat more than others, whereas other ewes may refuse supplementary food completely.

Since both experiments 5 and 6 investigated the same problem and gave very comparable results, they will be considered jointly in this Section.

Experiment 5.

In Section I of this thesis, Experiment I described the effect on the plasma magnesium concentrations in sheep of varying rates of potassium fertilisation on grassland. It was reported in that experiment that great individual variation occurred in the plasma magnesium concentrations of individual ewes and consequently, many individuals had plasma magnesium values below 1.00 mg/100 ml. When Experiment I was completed, these hypomagnesaemic individuals were therefore chosen for further study on dietary magnesium supplementation, since they were considered to be those with the greatest capacity to show a response to magnesium supplements.

Experimental Design.

On the 29th May, 1961, on the termination of Experiment I, fourteen Cheviot ewes were selected from the total of seventy-two ewes which had been used. The selection was made on the basis that all fourteen of these ewes had had plasma magnesium values of below 1.0 mg/100 ml at some time after lambing, compared with a mean value of about 1.7 mg Mg/100 ml for the other fifty-eight ewes. At the start of this experiment, the fourteen selected ewes had a mean plasma magnesium level of 1.08 mg/100 ml, the range being from 0.54 to 1.68 mg/100 ml. Lambing had taken place 6-8 weeks previously.

On the 29th May the ewes were transferred to a 4 acre paddock. The herbage consisted mainly of ryegrass, cocksfoot and wild white clover and analysis showed it to contain 0.13% Mg. It had received 3 cwt/acre of Nitro-chalk (21%N) in mid-March and had previously been grazed by young cattle.

The ewes were divided at random into two groups of seven which continued to graze together. One group acted as a control with no magnesium supplement. The other group of seven ewes received a daily drench of 1.66 g. magnesium oxide (= 1.0 g Mg) on the 1st-5th June. This was followed by a daily drench of 6.64 g. magnesium oxide (= 4.0 g. Mg) for the next five days. This same group was later given further daily drenches of 4 g magnesium in the form of magnesium nitrate solution on the 21st - 23rd June. Drenching was invariably carried out at 10 a.m.

Blood samples were taken over this period, 29th May - 23rd June, at frequent intervals of 1 - 3 days. Sampling was carried out in the morning immediately before the daily drench was administered. In consequence, blood samples were taken 24 hours after the previous drench. On the 7th June, blood samples

were also taken at 2 p.m. and 8 p.m. to determine the variation in the plasma magnesium levels within a 24 hour period.

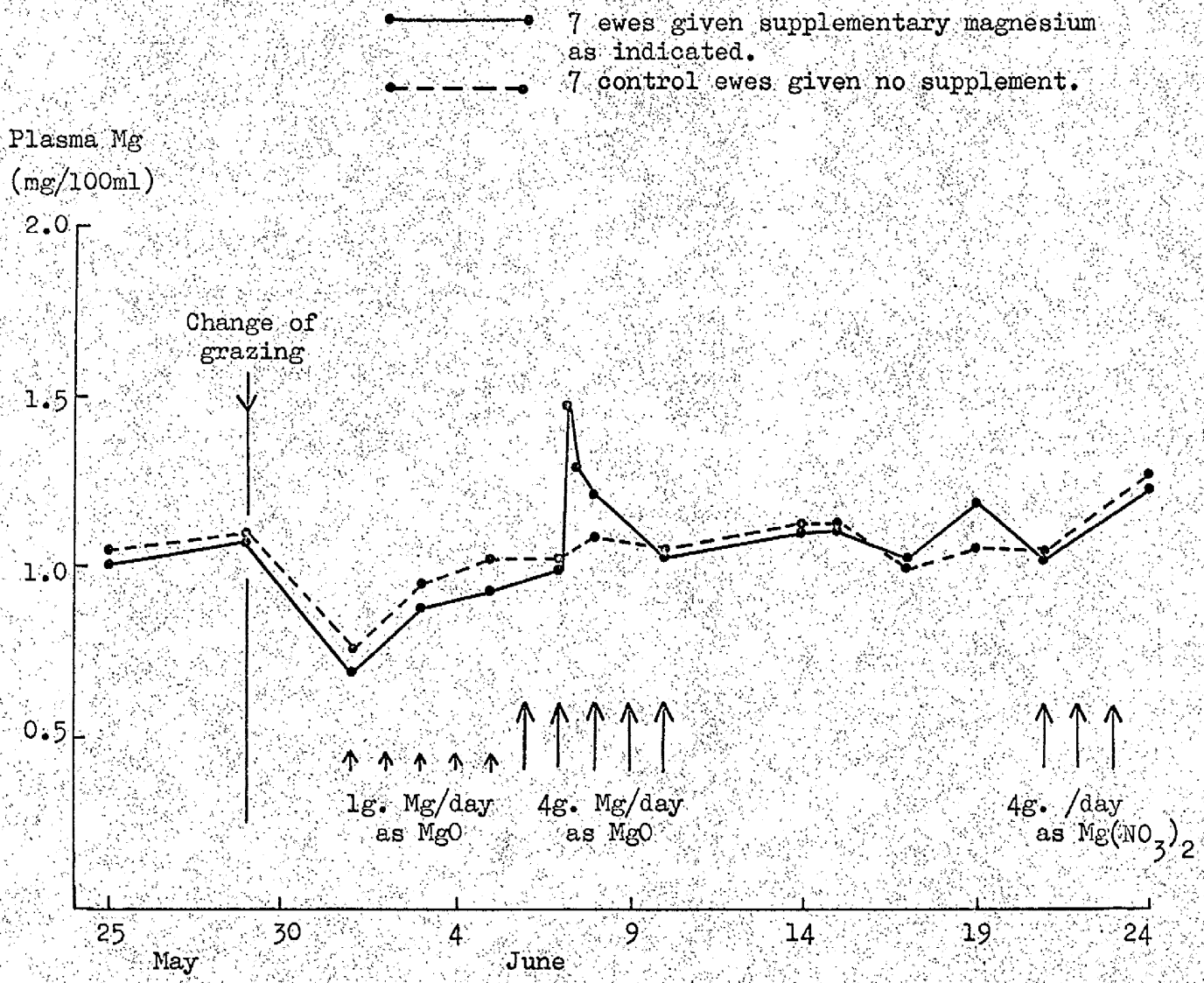
Results.

The individual plasma magnesium concentrations of each ewe on the fifteen sampling occasions over the experimental period are given in Appendix 4(a) together with the mean values of the two groups of seven ewes. The mean plasma magnesium values are also illustrated in Fig. 16. There was a marked fall in the mean plasma magnesium values of both groups when the ewes were transferred to the new grazing. The overall mean value of the fourteen ewes fell from 1.08 mg Mg/100 ml on 29th May to 0.72 mg/100 ml on 1st June. On the 1st June, individual values as low as 0.25, 0.30 and 0.43 mg/100 ml were recorded. There were no cases of hypomagnesaemic tetany. The plasma calcium values of all the ewes remained within the normal range of 9.5-11.5 mg Ca/100 ml over the whole experimental period.

It can be seen from Fig. 16 that there was no response in plasma magnesium values to drenches of up to 4 g magnesium given every day as measured by blood samples taken twenty-four hours after administration of supplementary magnesium. The mean plasma magnesium levels of both the drenched and control groups remained within the range of about 0.9-1.2 mg Mg/100 ml over the whole experimental period.

On the other hand, blood samples taken within a period of 24 hours after drenching on 7th June did show some elevation in plasma magnesium concentration. The mean level of the drenched group rose from 0.99 to 1.48 mg/100 ml for samples

Fig. 16 Mean plasma magnesium concentrations of two groups of 7 lactating ewes at grass (Experiment 5).



taken 4 hours after drenching, but had fallen to 1.29 after a further 6 hours. The uniformity in the individual responses to drenching as measured by the 2 p.m. sample was remarkable. The seven individuals rose by the following amounts:- 0.36, 0.40, 0.47, 0.49, 0.50, 0.51 and 0.72 mg Mg/100 ml. (mean increase 0.49 $\pm$ 0.115 mg Mg/100 ml).

Discussion.

The results of this experiment will be discussed in conjunction with the results from the following Experiment 6.

Experiment 6.

Although Experiment 5 demonstrates clearly that there was no measurable response in plasma magnesium concentrations to daily magnesium drenches, it must be remembered that the sheep used were selected from a larger group by virtue of their low plasma magnesium values. Before giving a definite conclusion, it was therefore thought to be desirable to repeat this experiment using a group of unselected sheep. Thereby any bias on the part of the investigator would be removed.

In consequence, the daily administration of $\frac{1}{4}$ oz magnesium oxide drenches to a group of sheep was included as part of the experimental design of the larger Experiment 2 carried out in 1962.

Experimental Design.

The experimental design of Experiment 2 has already been given in detail in Section II of this thesis (page 87). Briefly, it involved 48 lactating ewes, 24 of which were Cheviot ewes and 24 of which were Half-Bred ewes. They grazed on 12 experimental plots, which were fertilised with 0, 1 and 2 cwts of muriate of potash/acre. Two Cheviot and two Half-Bred ewes were allocated to each plot. The effects of these factors have been discussed previously in Experiment 2.

In addition to this, a daily drench of 6.64 g magnesium oxide (= 4 g Mg.) was given to one Cheviot ewe and one Half-Bred ewe in each plot. There were therefore 24 ewes given a drench and 24 control ewes, each group having equal numbers of the two breeds and having equal numbers grazing the differently fertilised herbage. Drenching was carried out daily at 10 a.m. from 14th May

when the ewes were transferred to the experimental plots until 24th May when the experiment was terminated.

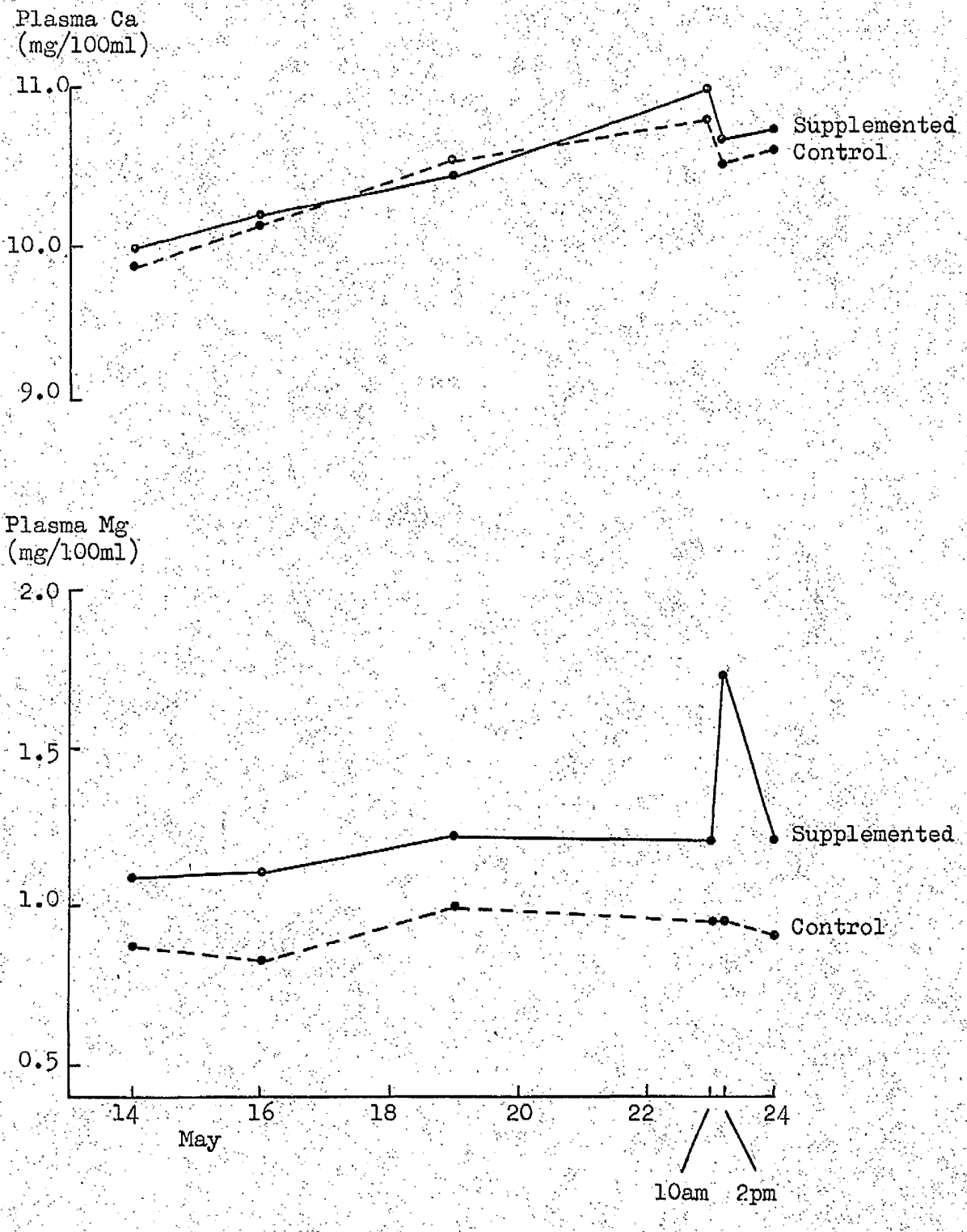
Blood samples were taken on the 14, 16, 19, 23, and 24th May. These samples were always obtained just prior to drenching at 10 a.m. On the 23rd May, an additional blood sample was taken at 2 p.m., 4 hours after the administration of the magnesium oxide drench.

Results.

The individual plasma calcium and magnesium concentrations of the ewes on each sampling date are to be found in Appendix 2(g). This Appendix, previously mentioned in Experiment 2, includes the mean values of the drenched and unsupplemented groups for plasma calcium and magnesium. These mean values are also shown graphically in Fig. 17. There was no fall in the mean plasma magnesium level of either group on transference to the new pasture. There was however an initial difference between the two groups of 0.22 mg Mg/100 ml which was entirely fortuitous due to the grouping involved. The mean levels of the two groups were very low at the start of the experiment, the respective mean levels being 0.87 mg Mg/100 ml for the control group and 1.09 mg Mg/100 ml for the supplemented group. Despite the daily drenching routine, the mean plasma magnesium levels of both groups changed very little over the eleven day experimental period, as shown by Fig. 17. The small difference which is apparent between the groups was merely a maintenance of the difference which was fortuitously present initially. There was therefore no response in plasma magnesium values to daily drenches of 4 g magnesium as measured by blood samples

Fig. 17 Mean plasma calcium and magnesium concentrations of 2 groups of 24 lactating ewes at grass (Experiment 6).

Supplemented group given 6.64g. MgO per day as a drench.



taken twenty-four hours after the previous drench.

There was, however, a response to the magnesium drench, in plasma magnesium concentrations measured four hours after the administration of the supplement. The mean level of the group given the drench rose from 1.21 mg Mg/100 ml at 10 a.m. on the 23rd May to 1.74 mg Mg/100 ml at 2 p.m. on the same day. By 10 a.m. on the following morning, the mean level had fallen again to 1.21 mg Mg/100 ml. The comparable samples taken from the control group showed no change over this 24 hour period, the mean levels being 0.95, 0.96 and 0.90 mg Mg/100 ml for the 10 a.m., 2 p.m. and 10 a.m. samples respectively. There was again a striking uniformity in the responses of the individual ewes to the drenching as measured by the 2 p.m. sample. The mean increase in plasma magnesium level was 0.53 $\pm$ 0.148 mg Mg/100 ml. The range in increase was from 0.35 - 0.95 mg Mg/100 ml but for seventeen of the twenty-four individuals it was within the narrow range 0.40-0.60 mg Mg/100 ml.

Discussion - Experiments 5 and 6.

Experiment 5 demonstrated that neither a soluble form of 4g magnesium given as the nitrate nor an insoluble supplement of either 1g or 4g of magnesium given as the oxide increased the plasma magnesium values of seven selected lactating ewes as measured by blood samples taken 24 hours after the administration of the supplement. These ewes were intentionally selected as being hypomagnesaemic. Experiment 6 confirmed this finding in that 4g supplementary magnesium, administered as a magnesium oxide drench failed to increase the plasma magnesium values of a larger group of twenty-four unselected lactating ewes at grass.

The mean plasma magnesium value of the group given the drench was in each experiment at a hypomagnesaemic level, and there was therefore in each case, considerable capacity for some improvement in the mean level.

There was however a temporary response in both experiments in that when blood samples were taken only 4 hours after the drench had been administered, there was a uniform rise in plasma magnesium values of around 0.5 mg Mg/100 ml. This response had largely disappeared after a further 6 hours in Experiment 5 where samples were taken at 8 p.m. in the evening.

It is therefore apparent that only a trivial proportion of the 4g. magnesium supplement administered per day is being utilised by the animal. It is not clear why, if calcined magnesite increases plasma magnesium levels over the short time interval of about four hours, it should not do so over a much longer period, since the magnesium supplement was being administered each day and, in consequence, should be present continuously for absorption. A rise of 0.5 mg Mg/100 ml in the plasma magnesium concentration of a 50 Kg sheep requires the absorption of only 10 mg. magnesium (Maguire & Wilson 1961). This would represent 0.0025% of the 4g magnesium administered. It is unknown whether this lack of response is due to non-absorption of the magnesium in the first instance, or alternatively due to absorption coupled with rapid excretion. The fact that the mean magnesium concentration of the sheep concerned is considerably below the threshold value of around 2.00 mg/100 ml would suggest that non-absorption is the more likely cause. On the other hand, if this were the case, it is not clear why there should be even an initial temporary rise in the plasma magnesium levels. One possibility which would fit the observed facts would be that the drench,

a water suspension of magnesium oxide, was carried directly to the omasum/abomasum, and not into the rumen of the sheep. The magnesium in the drench would then be presented for absorption within 4 hours of administration, and after 10 hours it would be beyond the small intestine, the area in the gut where magnesium is absorbed.

It is, however, difficult to visualise this sequence of events occurring in all the thirty one sheep involved in Experiments 5 and 6 on every day when a drench was given, without some intermixture of the supplementary magnesium salt with the rumen contents. Once the drench liquid does become mixed with rumen liquor, it should be presented continuously for absorption as the rumen material is passed on for digestion. Therefore, in these circumstances, either a continuous response would be expected or alternatively none at all, but not, as the results of these experiments show, a temporary response after 4 hours.

It is known (Allcroft 1960) that supplements of magnesium salts must be given <u>daily</u> to provide some degree of protection against hypomagnesaemia in ruminants. The results of Experiments 5 and 6 would suggest that <u>daily</u> supplementation is not sufficiently frequent to ensure complete protection. Thus, increasing the frequency of administration might increase the degree of prophylaxis provided. It may, in fact, be the case that the <u>frequency</u> of intake of supplementary magnesium each day, is more important than the absolute amount administered. The results quoted here could for these reasons explain the failures noted by Seekles & Doogaerdt (1956) and Allcroft (1953) where daily supplements of 2 ozs magnesium oxide for cattle did not completely prevent the incidence of hypomagnesaemic tetany.

In complete contrast to the results given for Experiment 5 and 6, the author found in a small subsequent experiment that non-lactating ewes maintained on an indoor diet exhibited a large response in plasma magnesium concentration when a daily drench of 6.64g. magnesium was given. One group of sixteen pregnant Blackface ewes, fed indoors, was given this daily drench for 6 days and a similar group of sixteen ewes was run as a control. Blood samples were taken on three occasions over the six days. Table 24 gives the mean plasma magnesium levels of the two groups on these three occasions.

Table 24. Mean plasma magnesium levels of two groups of sixteen ewes maintained indoors. (Samples taken 24 hours after previous MgO drench).

(concentrations in mg/100 ml).

	day 0.	day 3.	day 6.
16 ewes given 4g. Mg daily drench	2.30	2.98	3.00
16 ewes given no supplement	2.10	2.03	2.01

It is therefore evident that ewes maintained on an indoor diet were capable of a sustained response in plasma magnesium concentration to the daily administration of magnesium supplements. This was found despite the fact that plasma magnesium levels were at a normal level initially. The mean levels after drenching were therefore well above the renal threshhold level, and they could be described as hypermagnesaemic.

It would therefore seem that lactating ewes on spring pasture react in an entirely different way to magnesium supplementation of the diet as compared with pregnant ewes kept indoors. In indoor sheep, there was a direct response in

blood magnesium levels to supplementation, but at pasture, the response was slight and temporary. The reason for this complete contrast in reaction to the magnesium drenching routine is unknown. There is, however, the practical implication that the currently recommended practice of providing magnesium supplements once a day is perhaps at fault. A greater degree of protection against hypomagnesaemic tetany might be achieved by ensuring that the supplement is eaten or administered at least twice if not three times per day.

Experiment 7.

Since lactating ewes were found to respond so poorly to a conventional magnesium drenching routine (Experiments 5 and 6), it was felt desirable to investigate the effectiveness of the alternative methods of prophylaxis which are recommended for hypomagnesaemic tetany in sheep. The main alternative methods are the uses in various forms of magnesium fertiliser dressings on the pasture. The ones chosen for study were

(a) The widely recommended fertiliser dressing of 5 cwts magnesium oxide/acre applied to the land in winter.

(b) A light dressing of 56 lbs magnesium oxide/acre applied in the winter as a fertiliser. This small quantity per acre has previously been found by Birch & Wolton (1961) to be very effective in preventing hypomagnesaemia in cattle.

(c) A light dressing of 56 lbs magnesium oxide/acre applied not strictly as a fertiliser dressing, but rather as a pasture dust applied to the grassland immediately prior to the grazing of the pasture by the sheep. This has previously been found to prevent hypomagnesaemia in cattle by Todd (1961a, 1964).

Experiment 7 was designed to investigate the effect of these three pasture treatments on the plasma magnesium concentrations of lactating ewes grazing such pastures. It was run in conjunction with Experiment 3, which dealt with the effect of continued potassium fertilisation of grassland and possible breed differences in sheep with regard to their susceptibility to hypomagnesaemia. The results for Experiment 3 have already been detailed in Section II. Experiment 7 made use of experimental grass plots adjacent to those used in Experiment 3, and the experimental sheep involved were 24 of the Cheviot ewes and 24 of the

Half-Bred ewes as used in the breed comparison of Experiment 3. The group of twelve ewes grazing plots which were fertilised with 3 cwts of Nitro-chalk/acre served as the control group for both Experiments 3 and 7.

Experimental Design.

The experimental flock of sheep consisted of 24 Cheviot ewes and 24 Half-Bred ewes. Both groups of ewes were four year old and were having their third crop of lambs. Prior to and during lambing, the flock was grazing on old permanent pasture. All the Half-Bred and most of the Cheviot ewes had twin lambs. Lambing took place during the middle fortnight in April. On the 15th May the ewes were transferred with their month old lambs to the experimental plot pastures.

The twelve experimental plot pastures were located at Cochno Farm, Duntocher, Dunbartonshire. Each plot, 0.4 acres in size, received a general dressing of 3 cwts Nitro-chalk (21%N) on the 19th March, 1962. Three plots received no further fertiliser treatment. Three plots were given a dressing of 5 cwts magnesium oxide/acre on the 1st March and a further three plots were given a dressing of 0.5 cwts magnesium oxide/acre on the same day. The remaining three plots received a pasture dust of 0.5 cwt magnesium oxide/acre on the 14th May, one day prior to being grazed by the sheep.

Fig. 8 given on Page 105 in connection with Experiment 3 details the arrangement of the plots and the fertiliser treatment given to each. Two Cheviot ewes and two Half-Bred ewes were allocated at random to each plot and Fig. 8 also gives the ear tag numbers of the ewes allocated in this way.

Blood samples were taken from each ewe immediately prior to transference to the experimental plots and again on the 17th, 20th and 23rd May. These samples were analysed for plasma calcium and magnesium. The grazing period was of necessity short (9 days) due to the poor herbage regrowth on the pasture.

After the regrowth of the sward by the middle of June, a second group of experimental sheep, a flock of sixteen lactating Blackface ewes, were transferred to the experimental plots. Four sheep/plot were allocated at random on June 11th to each of two control plots and two of the plots treated with 5 cwts MgO/acre. The particular plots used and the ear tag numbers of the sheep allocated to them are to be found in Fig. 9 on page 107 . Blood samples were taken from this flock of sheep on the 11th June, immediately prior to the experimental period, and on 4 subsequent occasions during June. A later sample was taken on 18th July when the lambs were weaned and the experiment terminated. Further details concerning this experimental flock will be found in Experiment 3(b), since the same control group of eight ewes and a further group of eight Blackface ewes on plots given potassium fertilisers were involved in Experiment 3(b).

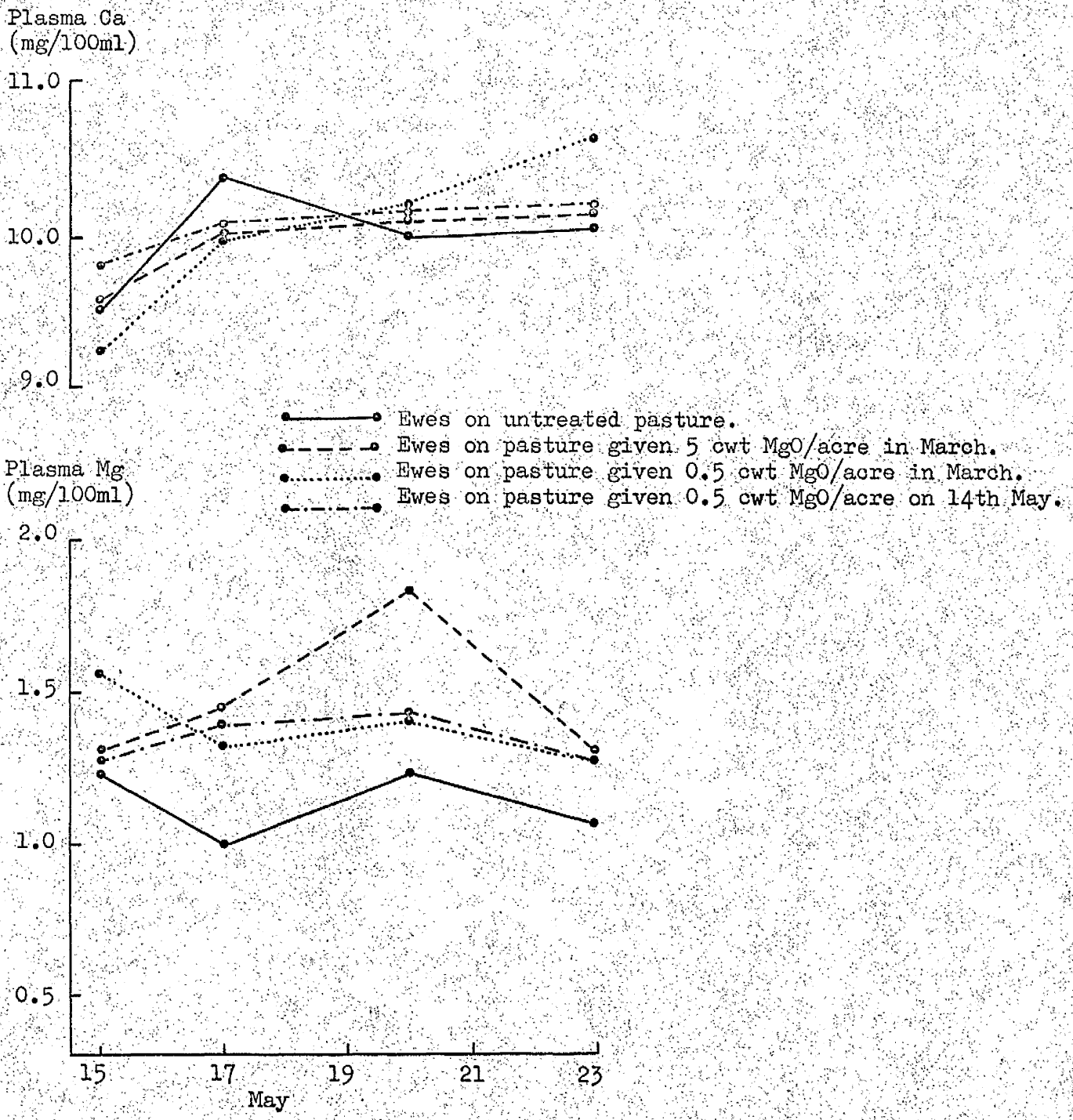
Herbage samples were taken from each plot on the 13th and 22nd May and again on the 11th June. An extra sample was taken on 15th May from the three plots given a pasture dust of magnesium oxide. Analyses were carried out on these samples for magnesium, calcium, potassium and sodium content.

Results.

Flock a. The individual plasma calcium and magnesium concentrations of the 24 Cheviot ewes and the 24 Half-Bred ewes on the four sampling dates in May are given in Appendix 2(i), together with the mean values for each group. The mean plasma magnesium and calcium levels of each group over the experimental period are also shown graphically on Fig. 18.

At the start of the experiment on 15th May, three of the groups of twelve ewes had similar mean plasma magnesium concentrations within the low range of 1.23-1.31 mg Mg/100 ml. The fourth group, grazing the plots given a small magnesium winter dressing had a higher initial level, by chance, of 1.57 mg Mg/100 ml. Both this group and the control group experienced a fall of 0.23-0.24 mg/100 ml in mean plasma magnesium level over the first two days of plot grazing. On the following sampling date on the 20th May, the mean level of the control group rose to 1.23 mg Mg/100 ml, but three days later it had again fallen to 1.07 mg/100 ml. The group of sheep on plots given low magnesium winter dressings behaved similarly apart from maintaining the pre-treatment difference and on no sampling date was it significantly higher than the control group. In contrast, the other two groups experienced no fall in plasma magnesium concentrations when introduced to the experimental pastures. The mean levels rose over the first five days of grazing to 1.33 mg Mg/100 ml for the group grazing plots given high magnesium winter dressings and 1.43 mg Mg/100 ml for the group grazing plots given a pasture magnesium dust. The mean levels of both these groups were significantly ($P = 0.05$) higher than the plasma magnesium level of control group on the 17th May, but on 20th May, only the group grazing the plots

Fig. 18 Mean plasma calcium and magnesium concentrations of 4 groups of 12 lactating ewes on experimental plot pastures (Experiment 7).



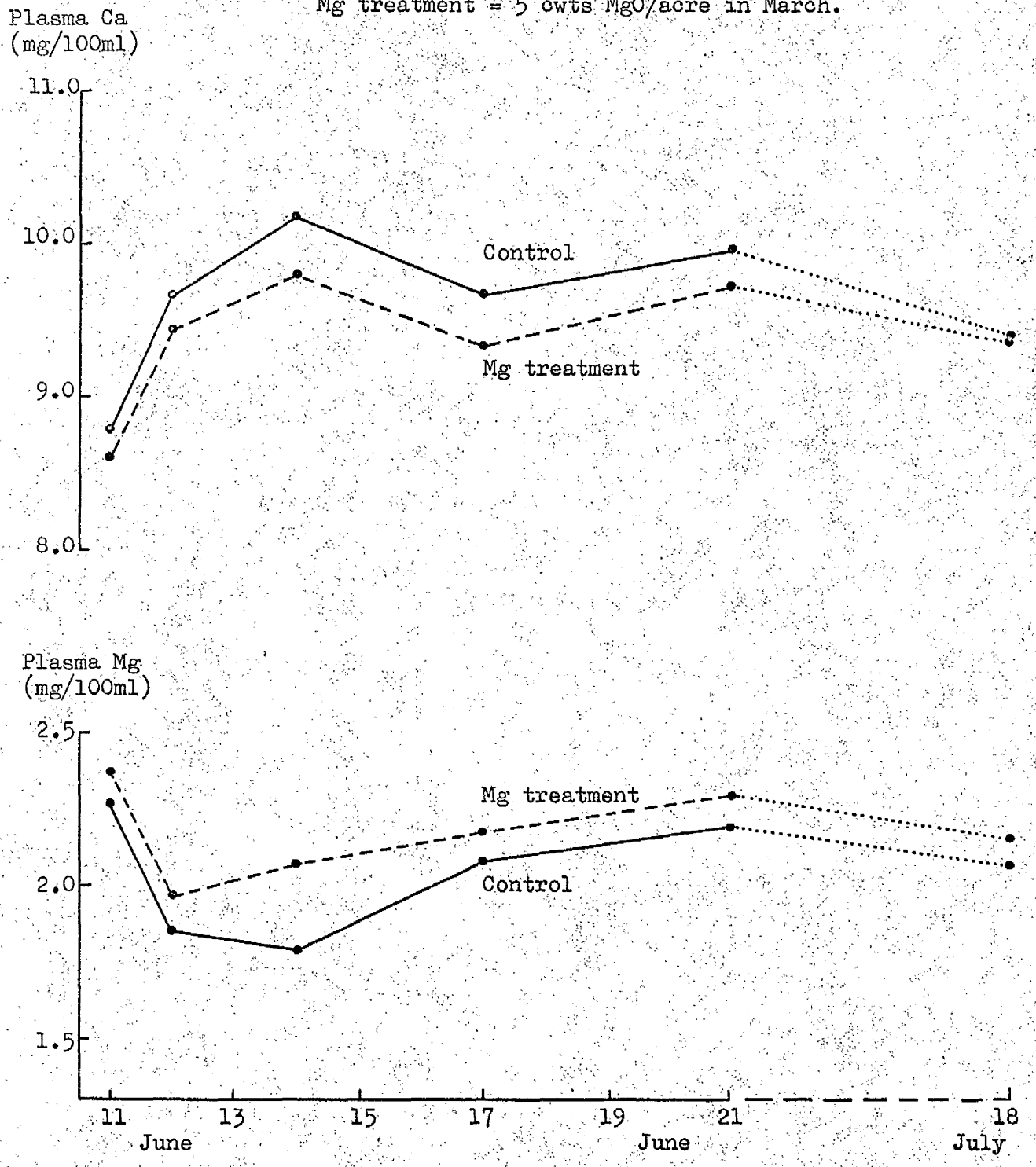
given a high magnesium winter dressing remained significantly higher in mean plasma magnesium level as compared with the control group. By the last sampling day on the 23rd May, the mean plasma magnesium of both these groups had fallen back to their original pre-treatment levels of 1.28 and 1.31 mg Mg/100 ml respectively, and no significant difference was apparent between any of the groups. The mean plasma calcium levels of all groups rose over the experimental period from an average of around 9.5 mg Ca/100 ml to 10.2 mg Ca/100 ml. There were no consistent differences to be noted between the groups in respect of plasma calcium concentrations.

Flock b. The mean plasma calcium and magnesium concentrations of the 16 Blackface ewes on the 5 sampling occasions in June and on the 18th July are given in Appendix 2(j) together with the mean levels of the two groups of eight. The mean levels are also shown graphically on Fig. 19. The mean plasma magnesium levels of both groups were initially at a normal level of 2.27-2.38 mg/100 ml. Only slight falls in the mean levels occurred during the 24 hours after the ewes were transferred to the experimental grazing area. The control group fell to 1.85 mg Mg/100 ml and the group on the magnesium treated pasture to 1.97 mg Mg/100 ml. Apart from the control group having a mean level of 1.79 mg Mg/100 ml after a further two days, the mean plasma magnesium levels of both groups were above 2.00 mg Mg/100 ml on every subsequent sampling date. There were only small differences between the groups and on no date were these significant.

The mean plasma calcium values of the two groups of eight Blackface ewes rose sharply from around 8.75 to around 10.0 mg Ca/100 ml when the ewes were transferred to the experimental pasture. The mean levels of both groups

Fig. 19 Mean plasma calcium and magnesium concentrations of 2 groups of 8 Blackface ewes on experimental plot pastures (Experiment 7).

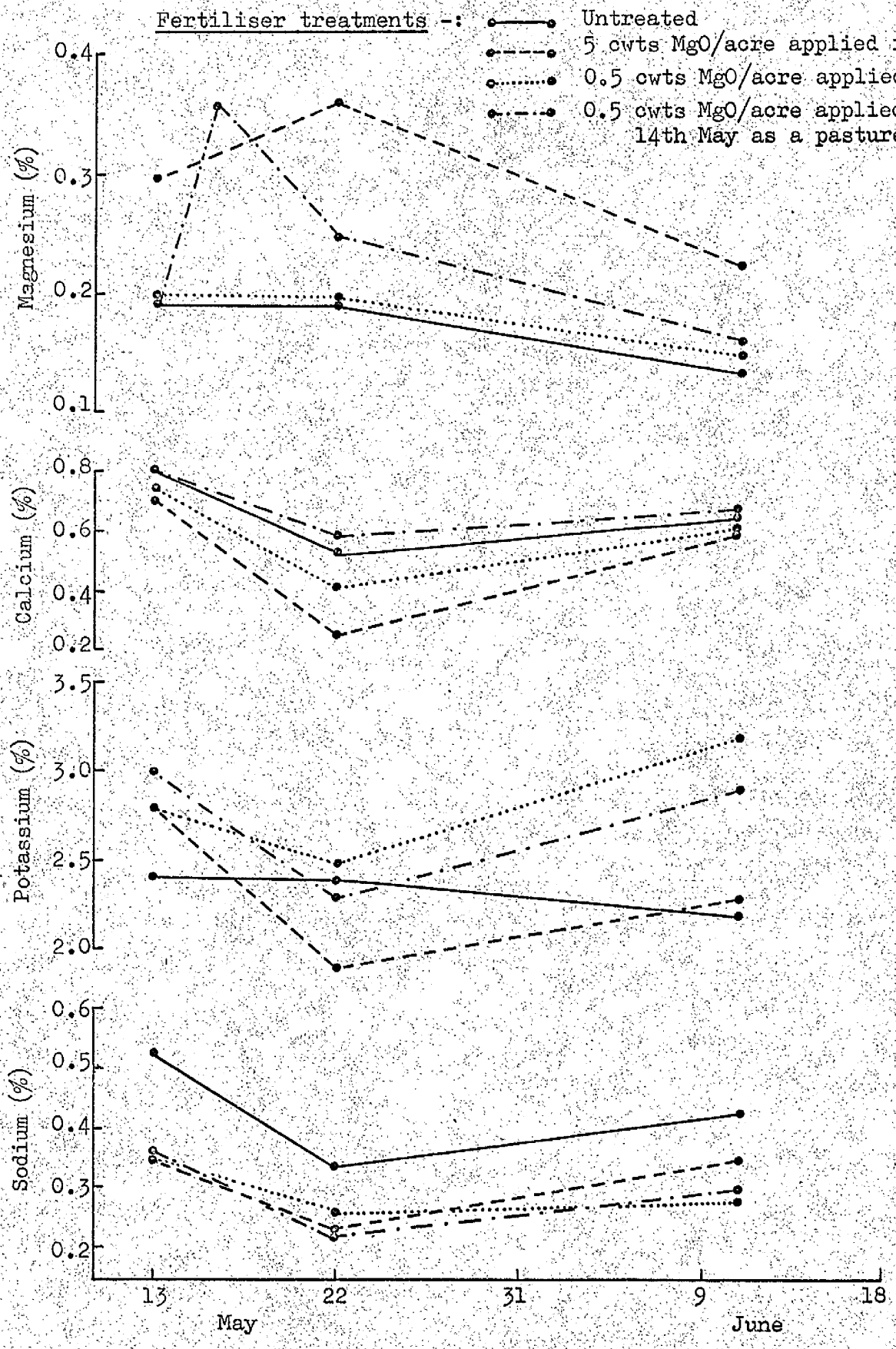
Mg treatment = 5 cwts MgO/acre in March.



remained in the range 9.5-10.0 thereafter. This sudden rise was due to the alleviation of the slight degree of hypocalcaemia that existed before the experiment commenced. This hypocalcaemic condition arose from the flock being maintained previously on a low calcium diet, but as discussed in Experiment 3(b), all the ewes had been treated similarly and consequently this pre-treatment should not affect the comparative results of this experiment. The control group maintained slightly higher mean plasma calcium values than the group on magnesium treated pasture on each sampling date, but at no time were these slight differences significant.

Herbage Analyses. The individual results and the mean values for herbage magnesium, calcium, sodium and potassium content of the three replicate plots of each treatment on the three herbage sampling occasions are given in Appendix 2(k). The mean levels are also shown graphically in Fig.20. The mean herbage magnesium content of the control plots was 0.19% on both sampling dates in May and fell to 0.14% in June. The first obvious comparison to make is, where plots received a dressing of 5 cwts magnesium oxide/acre, the mean herbage magnesium was raised by 0.08-0.17% over the whole experimental period, and it did not fall below 0.22 on any sampling date. On each sampling date, these herbage magnesium levels were significantly higher than those of the control plots. The mean herbage magnesium of the two pasture treatments of 0.5 cwts magnesium oxide/acre were similar to that of the control group on the first sampling occasion. Where the dressing had been given as a fertiliser in the winter, the mean level remained similar to that of the control group on the two

Fig. 20 Herbage mineral contents of experimental plot pastures - means of three replicate plots (Experiment 7).



subsequent sampling occasions and at no time was there any indication of an increase in herbage magnesium attributable to this 0.5 cwts MgO/acre treatment given in the winter. However, for the plots where this small dressing was applied as a pasture dust immediately prior to the grazing of the plots, the apparent herbage magnesium content was raised considerably (and significantly) as evidenced by the grass samples taken 24 hours after the application of the dust. The third sample taken on 22nd May demonstrates the temporary benefit of such a treatment since by this date, 8 days after its application, there was no significant difference between the mean herbage magnesium content of the control and treated plots.

As regards the herbage calcium content, there was little sign of any treatment differences between the mean levels, apart from a depression in herbage calcium on the 22nd May, which was almost significantly attributable to the application of the high rate of magnesium fertilisation. On the other two sampling dates no difference was evident. A slight depression in herbage calcium would be expected when high rates of magnesium fertiliser are applied, since there will be competition between these two divalent ions for uptake by the plant. There were non-consistent differences found between the plot treatments in herbage sodium and potassium. These can not necessarily be attributed to the magnesium treatments applied in this experiment, since the herbage sodium and potassium would be influenced by the past treatment of these plot areas, which had in previous years been given dressings of potassium fertilisers.

Discussion.

The basis of these pasture treatments as preventive measures relies completely on their ability to raise the magnesium content of the herbage and thereby increase the animal's dietary intake of magnesium. Where no increase in the herbage magnesium can be demonstrated, it would be most unlikely that any increase in plasma magnesium levels would be found. This is borne out by the results of this investigation, in that the measured effect of these various pasture treatments on the plasma magnesium concentration of the animal was roughly proportional to the effect of that treatment on the herbage. Where a small dressing of 0.5 cwts magnesium oxide was applied in the winter, there was neither a measurable increase in herbage magnesium content nor in the mean plasma magnesium level of the 12 ewes grazing the herbage. In contrast, where the higher rate of application of 5 cwts magnesium oxide/acre was used, there was a significant rise in the herbage magnesium content to above 0.20% coupled with a significant rise in the plasma magnesium concentration of the sheep on those plots. Assuming that a lactating ewe might consume around 3-4 lbs D.M. per day, there would be an increase of only 1.5-2.0g. Mg in the dietary magnesium intake for a sheep on a treated pasture (.30% Mg content) as compared with a sheep on the control pasture (0.19% Mg content). There was, however, a fall in the mean plasma magnesium level of this group on the last sampling date (23rd May), but this may have been associated with the shortage of grazing which was apparent at the end of the experiment.

On the plots where the small dressing of 0.5 cwts magnesium oxide/acre was spread as a pasture dust, there was a temporary rise in the herbage magnesium

content as evidenced in the sample taken on the 15th May. The mean plasma magnesium content of the twelve sheep on these plots was significantly higher only on the 17th May, which was the first sampling occasion after the sheep were in the plots, but not on subsequent dates. Thus the effect on the sheep was also temporary.

It is interesting to note that although increases in plasma magnesium levels were found in response to treatment, at no time were the mean levels, of the groups concerned, within the accepted 'normal range' for plasma magnesium. It may be that lactating ewes at spring pasture "normally" have lower concentrations than this accepted standard.

It is also interesting to compare the significant response obtained from magnesium pasture treatments with the lack of response obtained in Experiments 5 and 6 from a daily magnesium drench. This contrast is in spite of the pasture treatment supplying the grazing sheep with an additional intake of only 1.5-2.0 g. whereas daily drenching supplied up to 4g. of supplementary magnesium. It was suggested earlier that the lack of response to drenching was partly due to the infrequency of drenching (at 24 hourly intervals), and it was concluded that a more frequent intake would be more likely to produce a sustained response in plasma magnesium concentration in sheep. The intake of additional magnesium via a pasture magnesium treatment represents a more or less continuous intake, and it may therefore be that the effectiveness of this prophylactic method as demonstrated here, is due to this factor of continuity. It would also explain these contrasting results obtained in these experiments for the two different methods of supplying additional dietary magnesium to sheep.

The results obtained in the second part of the experiment with the flock of 16 Blackface ewes add little further information. Only the use of the high rate of magnesium fertilisation was investigated. On no occasion was there a statistical difference between the mean plasma magnesium levels of the treated and the control groups. On the other hand the mean levels of both groups were on most occasions above 2.0 mg Mg/100 ml and at these normal levels, an additional supply of dietary magnesium would not be expected to give any large increment in plasma magnesium concentration.

Rumen magnesium pellets.

The findings of Experiment 5, 6 and 7 were that 4g. of magnesium given as a daily drench did not increase plasma magnesium levels over a twenty-four hour period, whereas approximately 1.5-2.0 gms of magnesium received by sheep as an increase in herbage magnesium content did increase the blood levels. This paradox suggested that continuity of intake was possibly more important than the actual quantity of supplement given.

This evidence, obtained in 1962 and 1963, justified to some extent the possibilities of a new approach (first made in 1961) to the problem of prophylaxis for hypomagnesaemic tetany by means of the rumen pellet principle. Supplementary magnesium provided by this means would represent a continuous supply to the animal.

The principle of rumen pellet therapy was originally derived from the well-known fact that heavy foreign bodies such as metallic objects or stones, when swallowed by a ruminant animal, will be retained permanently in the rumeno-reticular sac. Objects such as nails may cause damagedue to traumatic reticulitis, but smoother objects such as stones can remain indefinately without causing any ill effects in the animal. It is possible, then, to devise a soluble heavy pellet to be administered orally, which will be retained by the ruminant animal and which will dissolve slowly over a given period of time. Depending on the constituents used in the manufacture, such a pellet can be made to provide the animal with a continuous supply of one particular dietary component over a period. This principle has already been successfully applied to the alleviation of cobalt deficiency in areas where 'pining' in cattle and sheep is a problem

(Dewey et al. 1958), but it is potentially applicable to the prevention of a number of dietary deficiency conditions provided of course that a suitable heavy pellet containing the necessary mineral elements or other component can be devised to dissolve at a suitable rate inside the rumen.

The provision of supplementary magnesium to cattle and sheep is one example where rumen pellet therapy might successfully be applied. Such an approach, if successful, would have the particular advantage of providing the animal with a continuous supply of additional magnesium as discussed above, but it would also have several advantages over the present conventional methods of prevention.

It would firstly have the advantage of being a single dose treatment, providing protection for a given period of time, say 5-8 weeks, whereas the present dietary supplements must be given every day during the period of risk. In addition, these daily supplements, because of their unpalatability, are usually fed mixed with a concentrate food which may perhaps not otherwise be necessary. The use of a magnesium rumen pellet would also ensure that every animal received a supplement. This certainty of treatment would have definite advantages over the free access systems of feeding mineral supplements which are relatively uncertain.

The alternative present method of prevention by applying magnesium fertilisers to the ground suffers from the disadvantage of being inapplicable to extensive grazing areas and on certain non-responsive soils e.g. alkaline areas. Under some circumstances, there are no feasible methods of prevention at present, due to the inaccessability of the grazing area for either pasture treatment or

dietary supplementation with magnesium to be undertaken. There is also considerable evidence to show that none of the present methods of control is completely reliable for the prevention of hypomagnesaemic tetany in cattle and sheep, and it may be that a magnesium rumen pellet, if successfully developed, might prove to be a more effective measure.

A possible difficulty to the development of an effective magnesium rumen pellet was likely to be the large quantity of supplementary magnesium needed daily in the diet of cattle and sheep. A rumen pellet is ideally suited for the provision of small quantities of trace elements such as cobalt which is necessary only in quantities of 0.1-1.0 mg per day. In the case of magnesium, however, the quantities required are much greater than these micro-amounts and magnesium can more properly be classed as a 'macro-element'. An even greater problem is the uncertainty which exists as to what daily quantity of magnesium supplement for cattle and sheep is necessary to provide protection against an outbreak of hypomagnesaemic tetany.

At the outset, it was felt that the present recommendations of 2 ozs MgO/day for cattle and $\frac{1}{4}$-$\frac{1}{2}$ oz MgO/day for sheep were out of proportion to the actual requirements. There is no scientific evidence to justify the use of these large quantities as the minimum prophylactic dose.

On a basis of the total requirements for magnesium, the recent estimates of the A.R.C. (1965) are, for a 50 Kg. sheep in peak lactation, a daily magnesium requirement of 2.58 gms. The N.R.C. (America) recommendations (1957, 1958a,b) place the requirement at less than half this figure, at 1.2 g. magnesium/day. The diet of the grazing animal will make a variable contribution towards the

magnesium requirements, dependant upon the magnesium content of the herbage. Rook & Wood (1960) give 0.09-0.16% Mg as the range for spring pasture samples, and Kemp et al. (1960) give a range of 0.08-0.32% Mg for 290 herbage samples taken in the course of experimental work on hypomagnesaemic tetany. Of these 290 samples only one was below 0.10% Mg. Therefore the daily magnesium intake of a lactating 50 Kg. ewe, consuming 3-4 lbs dry matter, at grass would seldom drop below 1.8-2.0g. Mg. Consequently the maximum deficit which would be found between intake and requirement (as estimated by the A.R.C. (1965)) would be in the region of 0.6-0.8g. Mg per day.

A similar calculation for a 500 Kg lactating cow grazing herbage with 0.09-0.10% Mg content gives the minimum dietary magnesium intake as 13.5-15g./day from 33.3lbs Herbage D.M. The A.R.C. (1965) estimate of daily requirement for a 500 Kg cow giving 4.4 gallons of milk/day is 20.1g. Mg. The maximum daily deficit on this basis would therefore be 5.0-6.5g. Mg/day.

These calculated figures of course depend on the following assumptions

(a) the estimate taken in the A.R.C. recommendations of 20% as the minimum availability

(b) both the cattle and sheep consuming an adequate bulk of dry matter as grass.

These assumptions may or may not be correct, but in the absence of alternative estimates for the quantities of magnesium supplements necessary, the figures as calculated above at least give a basis on which further work can be carried out. This work may then confirm or disprove the validity of the calculations.

There is also the important question of whether it is either necessary or possible to prevent completely the occurrence of hypomagnesaemia in order to

prevent the appearance of hypomagnesaemic tetany. The evidence available at present would suggest that hypomagnesaemic tetany is liable to occur only when the plasma magnesium concentration falls below the low level of 0.8-1.0 mg/100 ml. A reasonable degree of hypomagnesaemia may be present in cattle and sheep without any consequent risk of tetany. It may therefore be found that a small supplement of dietary magnesium may not prevent the development of hypomagnesaemia but it may, by virtue of raising the plasma magnesium level above the extreme low values, prevent the appearance of tetany. To eliminate completely the development of any degree of hypomagnesaemia might require a much larger supplement of dietary magnesium.

It may be postulated, therefore, that under most circumstances the maximum deficit in dietary magnesium which lactating sheep at grass are likely to encounter is in the order of 0.6-0.8g. Mg/day and for lactating cattle on pasture the figure suggested is in the order of 5.0-6.5g. Mg/day. By extension of this postulation, if a rumen pellet treatment was devised to supply magnesium in quantities approaching these calculated figures of 5.0-6.5g.Mg/day and 0.6-0.8g. Mg/day respectively for cattle and sheep, then such a treatment might have a reasonable chance of providing protection against hypomagnesaemic tetany in these species.

Experiment 8 describes a trial carried out with sheep to test the effectiveness of a rumen pellet based on magnesium oxide. The pellet treatment as used in this trial supplied magnesium in quantities considerably below those suggested above.

Experiment 9 describes the development of a rumen pellet designed to supply

magnesium in much larger quantities to both cattle and sheep and as far as was possible designed to meet the calculated figures given above. These pellets made use of magnesium metal as the main ingredient.

Experiments 10 - 15 deal with experiments designed to test the effectiveness of magnesium metal rumen heavy pellets as a prophylactic treatment against hypomagnesaemic tetany under field conditions.

Experiment 8.

An opportunity arose in 1961 to carry out a trial to test the effectiveness of a magnesium oxide rumen heavy pellet in preventing hypomagnesaemia and hypomagnesaemic tetany in sheep. This pellet was an experimental formulation designed and manufactured by Aspro-Nicolas Ltd. In a previous experiment with pellets of the same design, Hemingway et al. (1961) found a treatment of either two or four pellets per sheep supplying 150 or 300 mg Mg/day to be successful in preventing the development of hypomagnesaemia in non-pregnant non-lactating sheep maintained on a low magnesium diet over a six week period. Sheep maintained on the same diet but given no pellet treatment had a mean plasma magnesium level over this period of 1.09-1.20 mg/100 ml. This encouraging result, found under experimental conditions in indoor fed sheep, justified the extension of this work to an experiment under more practical conditions with lactating sheep at grass.

Experimental Design.

Heavy Pellets. The pellets used in this experiment were cylindrical in shape, with the two ends slightly rounded (Fig. 21). The overall length was 1.75 inches and the diameter was 0.75 inches. They were made in a mould by compressing a mixture of 45% magnesium oxide, 40% iron powder, 2% starch and 13% water. The iron powder was present merely as a weighting medium, and the starch acted as a binder for the mixture. A central steel core (1" in length $\frac{3}{8}$" diameter) was also included to give the pellet an overall density of 4.0g/cc which was at that time thought necessary to prevent regurgitation of the

Fig. 21 Magnesium oxide rumen pellets after removal from the rumeno-reticular sac of sheep by rumenotomy

Original pellet	Central steel core	Pellets recovered from sheep after 14 days	40 days	50 days

pellet. Each pellet therefore weighed 40g. of which 0-0.5g. was magnesium. Previous tests where pellets were recovered some time after administration indicated that the pellets dissolved at a fairly regular daily rate of 74±12 mg magnesium per day. At this release rate, the pellets could be expected to last for 80 days.

Experimental sheep. The sheep flock employed in this trial was the flock of 72 Cheviot ewes which were used in Experiment I of this thesis. Experiment I was concerned with the effect on plasma magnesium levels of three different rates of potassium fertiliser usage and of two different age groups in the same flock. The fertiliser treatments were applied to 0.4 acres plots, each treatment being replicated six times. There were therefore eighteen plots to each of which two young ewes and two old ewes were allocated. In addition to these factors of fertiliser use and age effect, a pellet treatment was included in the experimental design by administering two magnesium pellets to one old ewe and to one young ewe in each plot. The pellets were easily administered by means of an oseophageal balling gun. A total of 36 ewes therefore received the pellet treatment and 36 ewes were given none. Those ewes given pellets were distributed equally among the three fertiliser treatments and also between the two age groups.

The pellets were administered to the ewes on the 19th April 1961 when the entire flock was transferred to the experimental plots. Blood samples were taken on that date and on six subsequent occasions until the termination of the experiment on the 29th May. Analyses were carried out on these samples for

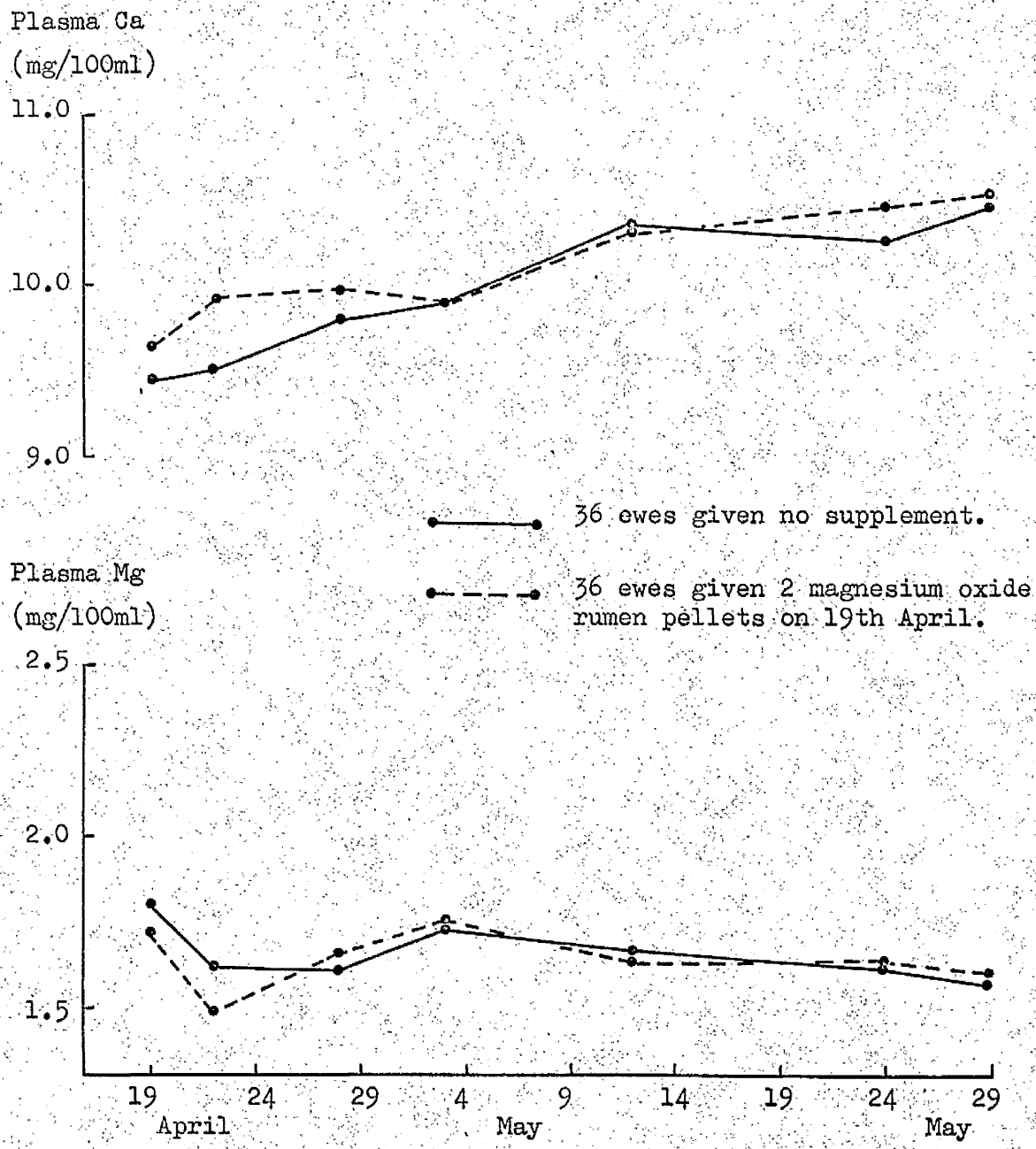
plasma calcium and magnesium concentration.

Results.

The plasma calcium and magnesium concentration of the 72 individual sheep on the seven sampling occasions over the period 19th April-29th May are to be found in Appendix 2(c), together with the mean values for the two groups of 36 treated and 36 untreated ewes. These mean values are also shown graphically on Fig. 22. There were initial falls in the mean plasma magnesium values of both groups over the first three days of the experiment from 1.72-1.80 mg Mg/100 ml on the 19th April to a level of 1.49-1.62 mg Mg/100 ml on the 22nd April. Thereafter both groups maintained similar mean levels which remained substantially constant at around 1.65 mg Mg/100 ml until the end of the experiment on 29th May. At no time was there any significant difference between the groups. The mean plasma calcium values of the two groups were similar over the 40 days of the experiment and there was a general rise in the level of both groups over this period from around 9.5 mg Ca/100 ml to 10.5 mg Ca/100 ml.

One case of clinical tetany occurred in the group given no magnesium pellets. Some of the heavy pellets were recovered by rumenotomy at the end of the experiment and they were found to be dissolving satisfactorily at the expected rate of 70-80 mg Mg/day.

Fig. 22 Mean plasma calcium and magnesium concentrations of 2 groups of 36 Cheviot ewes at grass (Experiment 8).



Discussion.

Two magnesium heavy pellets supplying a total of 150 mg magnesium per day to each ewe had no effect on the mean plasma magnesium levels of the 36 treated ewes as evidenced by a comparison with a similar group given no pellets. Furthermore, the treatment did not prevent the development of severe hypomagnesaemia since there were equal numbers of ewes in both groups which had plasma magnesium values below 1.0 mg/100 ml. The occurrence of one case of hypomagnesaemic tetany can be ignored since pure chance can decide in which of the two groups a single case might occur.

The result was in marked contrast to the positive results which were obtained from the use of an identical pellet treatment in sheep on a low magnesium diet (Hemingway et al. 1961), where highly significant increases in plasma magnesium concentrations were demonstrated. The sheep used in that experiment, however, were non-pregnant non-lactating ewes and would therefore have a much lower requirement for magnesium than the lactating ewes used in this present experiment. In addition to this, Rook (1961) in an experiment where a low magnesium diet was fed to cattle, found that the use of concentrate foods which were low in magnesium seemed to improve the availability of dietary magnesium. Consequently, experiments carried out with low magnesium diets indoors may give misleading results and the conclusions thereby reached may not be applicable when the diet is one of grass. Nevertheless the positive results obtained from a pellet treatment in the earlier work with sheep on a low magnesium diet would suggest that the basic principle of using a magnesium rumen pellet to alleviate hypomagnesaemia is sound.

It can only be concluded from this present experiment that the 0.15 g of magnesium supplied per day by the pellet treatment as used here, was insufficient supplementary magnesium to make any material difference to the magnesium status of the ewe as measured by plasma magnesium analysis. It should be said however that these pellets were purely an empirical attempt at devising a suitable magnesium pellet and the decision to give two pellets/ewe as the treatment level was based on a trial and error system rather than any scientific reasoning.

Experiment 9. The Development of a Rumen Pellet Containing Metal Magnesium.

The conclusion from experiment 8 was that two magnesium oxide rumen pellets supplied insufficient supplementary magnesium to the ewe to prevent hypomagnesaemia, but it was pointed out that the principle of applying bullet therapy to the prevention of this condition was probably basically sound, since earlier work with sheep indoors had given positive results. It was also argued that the main reason for the failure of this pellet treatment in Experiment 7 as compared with the earlier work on indoor sheep was the increased magnesium requirement of ewes when they are at peak lactation.

It could therefore be argued that for a pellet treatment to have any measurable effect on the plasma magnesium concentration of ewes with lambs, a much higher daily release rate of magnesium would be required from the pellet. The possible deficit in dietary mangesium of a lactating ewe at grass was discussed in the introduction to these experiments and the tentative figure put forward was a maximum deficit of 0.6-0.8g. magnesium/day for a 50 Kg. ewe at peak lactation grazing herbage with the low magnesium content of 0.09-0.10% Mg. The comparative figure for an adult cow was tentatively given as 5-6.5g. magnesium/day. If a pellet treatment was designed to supply the animal with daily quantities approaching these amounts, it would have a more realistic relationship with the actual requirements. It would therefore be more likely to have a measurable effect on the plasma magnesium concentration and thereby prevent severe hypomagnesaemia and possibly hypomagnesaemic tetany.

The object of Experiment 9 was therefore to devise a magnesium pellet treatment which would supply sheep with daily supplements of 0.6-0.8g. Mg/day

and which would supply cattle with daily supplements of 5 - 6.5g. Mg/day.

To continue to use the magnesium oxide pellets employed in Experiment 8 would have required the administration of eight or nine pellets per sheep in order to provide a daily supplement of 0.6-0.8g. magnesium/day. This dosage rate was obviously impractical. On the other hand if the design of the magnesium oxide pellet had been altered to provide a higher daily release rate, this would in turn have cut down the period of time for which the pellet remained active, since the pellet only contained 6.5g. magnesium. The need was therefore for a pellet which contained a greater quantity of magnesium and released a greater quantity of magnesium per day. The oxide is the magnesium salt with the greatest proportion (60%) of magnesium by weight, and the only material which contains a greater proportion of magnesium is magnesium metal itself which apart from being 100% magnesium by weight, is a relatively dense material (specific gravity 1.74) and it therefore contains considerably more magnesium both per unit weight and per unit volume. For example, the magnesium oxide pellet as used in Experiment 8 contained 6.5g. magnesium, and a pellet of similar size composed of magnesium metal would contain 22.0g. magnesium. Even if a steel core was included in a metal magnesium pellet to bring the density up to that of the oxide pellet i.e. 4.0g./c.c., a pellet of this size would still contain 14.5g. magnesium. Magnesium metal was for this reason the obvious material to employ in future attempts to devise a heavy rumen pellet with a high magnesium release rate compatable with a long period of activity.

Experiment 9 was therefore concerned with the development of a rumen pellet,

based on magnesium metal as the major ingredient, which would dissolve in the liquor of the rumeno-reticular sac of cattle and sheep, and supply magnesium at a regular daily rate to the animal. To supply the large amount of magnesium required, the release rate would have to be as high as possible, subject to the need for a reasonably long period of pellet activity. There were other factors which had to be borne in mind, such as the need for good retention of the pellet in the animal, and the possible size of the pellets. The description of this experiment will ignore in the meantime the ultimate use of these pellets as a prophylactic treatment for hypomagnesaemic tetany, which will be the subject matter of Experiments 10-15.

Experimental.

There were two alternative ways of incorporating magnesium metal in a rumen pellet. Either a powdered form of the metal could be employed to make a compressed pellet similar to the magnesium oxide pellet described in Experiment 8, or alternatively a solid cylinder of metal could be made by casting.

Preliminary tests were carried out with compressed pellets of magnesium metal powder using material of varying particle size, different pressures for compression and with various mixtures of metal components such as magnesium powder mixed with iron powder or copper powder. The results were poor due to a very rapid dissolution of the pellet in the animal over two or three days and a tendency for the pellet to flake in layers. The release rate of magnesium from such pellets was erratic, unpredictable and excessively fast, and this

approach to the problem was therefore not pursued. The alternative of employing solid magnesium metal in a pellet design was believed to be the more promising method to investigate.

Experimental pellet formulations were tested for cattle by placing them directly into the reticulum of a cow fitted with a rumen cannula. In this way, experimental material could be removed at regular intervals for examination, and subsequently replaced for a further trial period. Attempts were made at one stage to devise an 'in vitro' test by suspending the pellet in a volume of rumen liquor obtained from a fistulated cow, or alternatively suspended in one of several salt solutions such as normal saline, dilute hydrochloric acid, or sodium acetate. No 'in vitro' test was found which could satisfactorily simulate the 'in vivo' conditions within the reticulum as far as pellet dissolution was concerned. A fistulated cow was therefore employed in all future tests with cattle metal magnesium pellets.

Trials with metal pellets for sheep were mainly carried out by administering experimental pellets to a group of sheep and slaughtering them at staggered intervals to recover the pellet residues. Attempts were made at one stage to design a rumen fistula for sheep which would be big enough to allow experimental material to be placed into and recovered from the reticulum by hand. This entailed using cannulae with an internal diameter of $3\frac{1}{2}$ inches, and the most suitable design was therefore the rumen cannula described by Ash (1957). These attempts met with only limited success and some trials with pellet material were carried out in sheep fistulated in this way. The cannula,

however, tended to be so large in relation to the size of the rumen, that normal rumen function was greatly impaired and the results obtained for pellet dissolution in these sheep were therefore abnormal and misleading. Furthermore, since the cannulae were manufactured from rigid material (in this case nylon), they tended to catch on pen walls and feeding troughs, and consequently did not remain securely fitted in the animal for more than a few months on average. Most tests with experimental rumen pellets for sheep were therefore carried out in whole sheep where eventual recovery of the pellet residues was possible only on slaughter.

The first test carried out with solid metal was where cylinders of pure magnesium metal (Analar quality), 2 inches in length and 0.5 inches in diameter were administered to two sheep and also placed in a fistulated cow. These pellets were weighted with a central steel core which was completely enclosed by the magnesium. In neither cattle nor sheep was there any visible signs of corrosion when these pellets were recovered after 21 days. (Fig. 23). The loss of weight as determined by weighing before administration and after recovery was only fractional, being in the order of 5-10 mg. magnesium per day i.e. 100-200 mg. over the 21 day period. It was concluded that pure magnesium metal did not dissolve in the rumen liquor of cattle or sheep.

The second test in this investigation was the administration to sheep of cylinders of pure magnesium metal again 2 inches in length and 0.5 inches in diameter, which were weighted with steel bolts threaded through the centre of the cylinder and exposed at each end (Fig. 24). Two of these experimental

Fig. 23 Pure magnesium rods recovered from the rumeno-reticular sac of a) a sheep and b) a fistulated cow, both after a period of 21 days.

(a) (b)

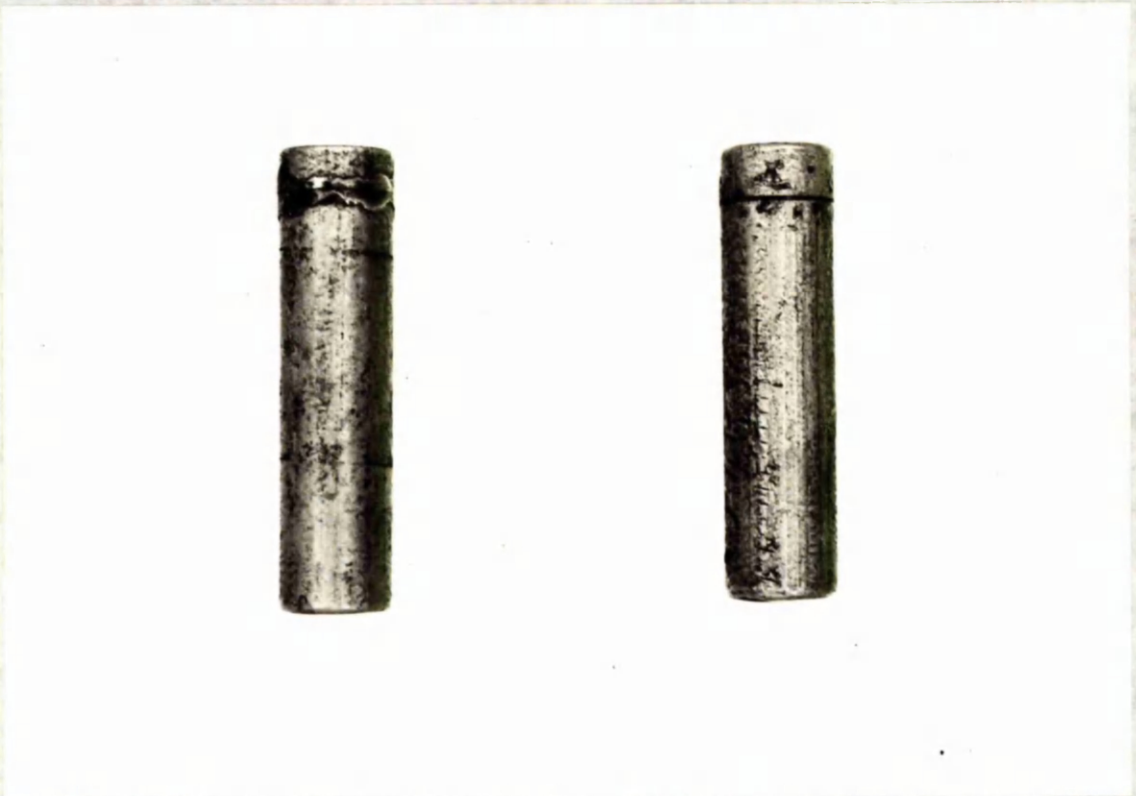
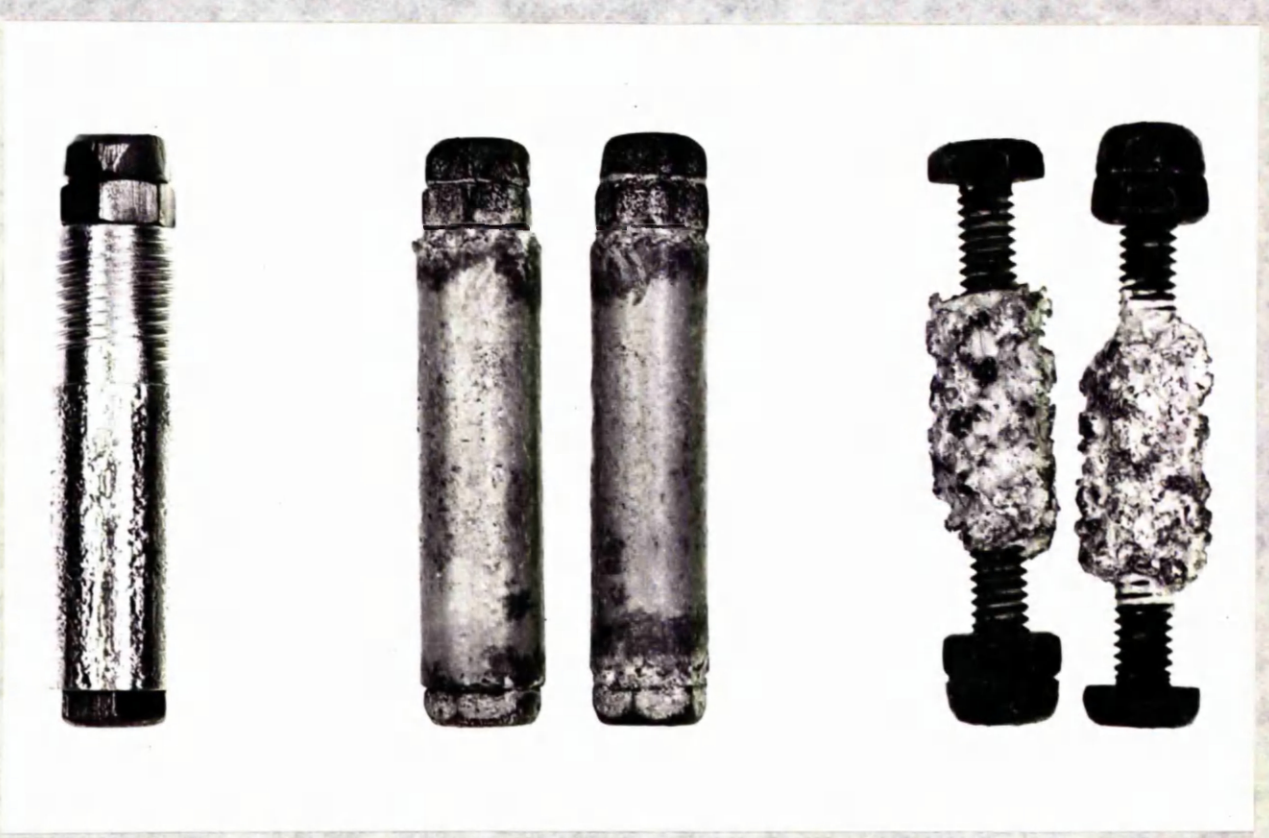


Fig. 24 Pure magnesium rods in contact with steel bolts. Tested in the rumeno-reticular sac of sheep at grass.

Original rod | Removed after 16 days. | Removed after 42 days.



pellets were administered orally to each of four sheep. One animal was slaughtered after 16 days and the remaining three were slaughtered 42 days after the administration of the pellets. (Fig. 24). Each pellet was weighed before administration and after recovery. Table 25 details the weight loss of each pellet.

Table 25. Weight loss of rumen pellets made of pure magnesium in contact with iron bolts. Administered to sheep at grass.

Sheep no.	Pellet no.	no. of days of test.	original weight (g)	final weight (g)	total weight loss(g)	average daily weight loss (g)
16	1	16	30.02	29.70	0.32	0.020
	2	16	30.58	30.40	0.18	0.011
20	3	42	30.18	24.40	5.78	0.138
	4	42	30.40	25.40	5.00	0.119
33	5	42	30.13	23.90	6.23	0.148
	6	42	30.51	23.60	6.91	0.164
34	7	42	30.14	24.40	5.74	0.137
	8	42	30.50	24.70	5.80	0.138

Since the iron bolts remained unaffected by their immersion in rumen liquor, the weight losses given in Table 25 were wholly from the magnesium cylinder. It is obvious from Fig. 24 and Table 25 that these experimental pellets did

dissolve in the rumen of a sheep. It is particularly noticeable from Fig. 24 that material was dissolved mainly from the two ends of the cylinder where it made contact with the steel bolt. Thecorrosion was therefore presumably due to an electrolytic action at the point where those two metals of different potential were in contact. Since magnesium is the more electro-negative metal, it dissolves rather than the iron. Table 25 gives figures for <u>average</u> daily release rate in the last column, but it is obvious that the release of material was not regular throughout the 42 days, since the first two pellets which were recovered after 16 days show very little weight loss. The pattern of release rate was therefore one of very slow corrosion at first, taking place purely at the two ends as shown in Fig. 24, with a gradual acceleration in corrosion as pits developed in the metal. This is an undesirable corrosion pattern since any potential magnesium rumen pellet would have to release magnesium at the same rate throughout its active life to be of any use in practice. Furthermore, as can be seen in Fig. 24, the pellet residue is a pitted object with jagged edges which may represent a potential danger to the animal. It would also be unpractical to leave behind a residue of a steel core in the animal. However this test did demonstrate that magnesium metal could be made to dissolve by being in contact with another metal, under the particular conditions of the rumen of a sheep.

An alternative method of combining solid magnesium with a second metal in a pellet is to have the two metals present in an alloy form with the magnesium preferably being the predominant metal. It is not possible to employ steel or iron for this purpose, since iron can only alloy with magnesium to the very

small extent of 0.08% by weight. There are however a wide range of other metals which alloy with magnesium in any proportion. One proviso that had to be borne in mind when alloys were considered as potential material for magnesium pellets was that any metal used should be non-toxic to ruminant animals in the quantities in which it was included in the pellet.

Many magnesium alloys were screened for their suitability as material for a magnesium rumen pellet. These tests were all carried out in a fistulated cow where the material under test was removed every two or three days for examination. The properties which were looked for in these experimental formulations were

(a) an ability to corrode and release magnesium at a reasonably regular rate throughout a test period

(b) that the corrosion should begin within a day or two of placing the test material in the cow and

(c) that corrosive activity would take place over the complete surface of the pellet thereby producing at all stages a pellet residue which was relatively smooth with no sharp edges such as might injure the animal.

The term 'corrosion' is used intentionally since these are metal materials and they do not dissolve as do salts nor is material abraded from the surface.

Magnesium alloys which were screened for activity in this way, included alloys of magnesium with copper, nickel, cobalt, zinc, calcium, lithium, manganese, sodium, tin and silver. These were made with varying proportions of magnesium from 50-99.5% and corresponding proportions of the second metal from 0.5-50%. For example tests were carried out on magnesium alloyed with 1% Cu. or

alternatively 1% or 50% Ni. Other examples of magnesium alloys tested were with 1,2,5 or 10% of zinc or calcium. Alloys with three constituent metals were also tried, for example a magnesium alloy with 10% zinc and 1% copper.

To make these experimental pellets the various ingredients were weighed out in the correct proportions, placed in an iron smelting pot, and heated in a gas furnace until molten. A suitable flux was used to prevent ignition of the material. After stirring the contents to ensure proper mixing, the molten metal was poured into a tapered cylindrical iron mould with an internal diameter of 1 inch. After cooling, the resultant casting was removed and cut into suitable lengths which were smoothed off by lathe turning.

None of the magnesium alloys using the elements listed above was found to be a suitable material for a rumen pellet as measured by the criterions given previously. Under test, they were either completely non-reactive and therefore released no magnesium or alternatively they corroded by developing large pits on the surface, which produced a very slow corrosion rate at first, but as the pits enlarged, the corrosion rate accelerated until the test material had completely dissolved. Two examples of this undesirable corrosion pattern are shown in Fig. 25, which displays the pitted nature of the half worn pellets. On Fig. 26, the weight losses of these two pellets have been plotted over the test period of 18 days, showing the accelerating nature of the daily release rate of magnesium from these pellets.

As a result of these tests, one class of alloy was found which behaved in a different fashion to those described above. This was a range of magnesium alloys containing between 8 - 20% by weight of aluminium. Any magnesium alloy

Fig. 25 Two typical examples of the unsuitable type of corrosion found with many magnesium alloys when placed in the rumen (Experiment 9).

a) 1% nickel / 99% magnesium b) 1% copper / 99% magnesium

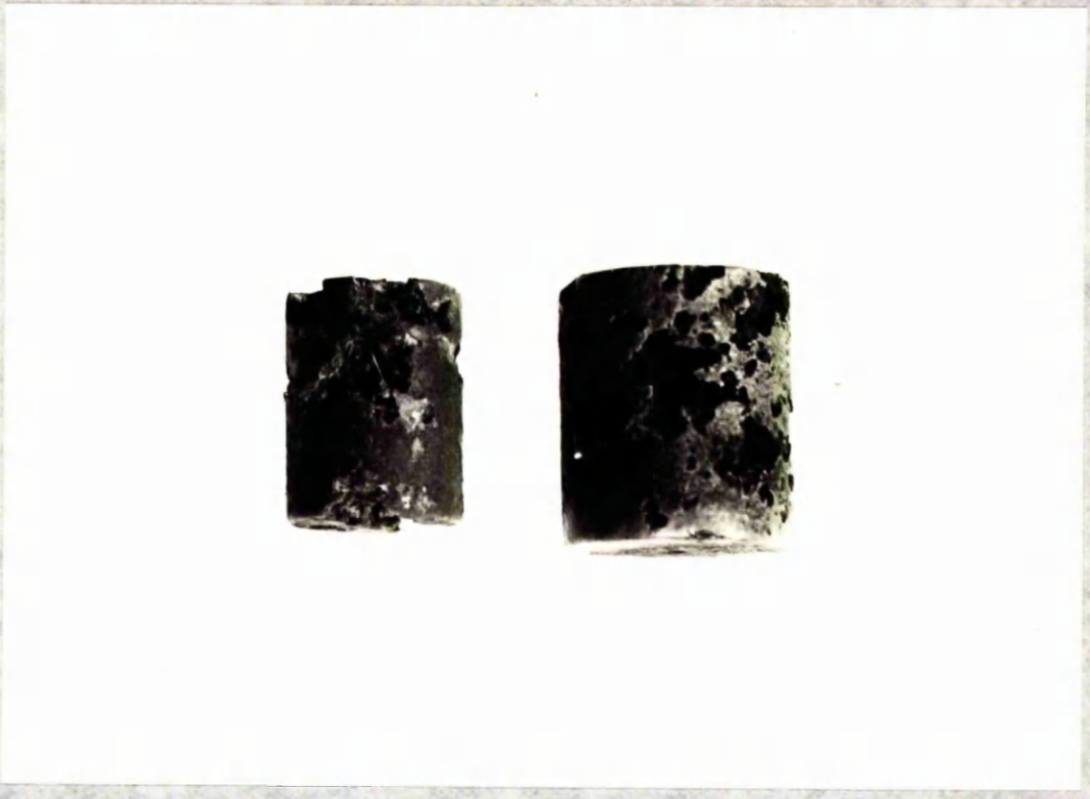
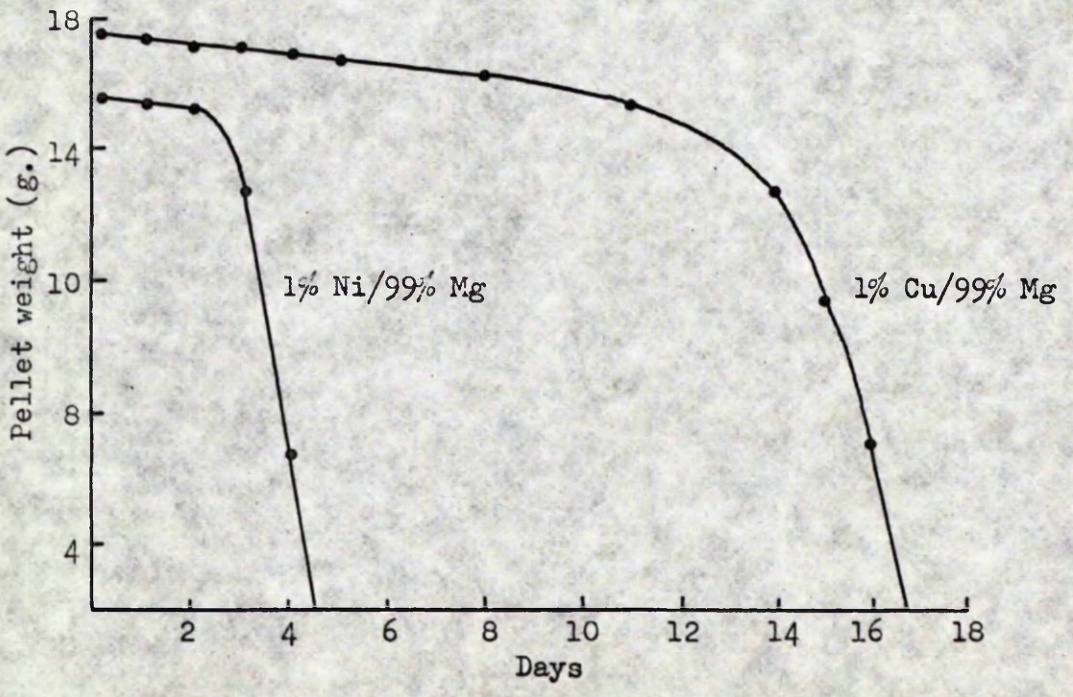


Fig. 26 Weight losses of the alloys shown above when tested in the rumen of a fistulated cow.

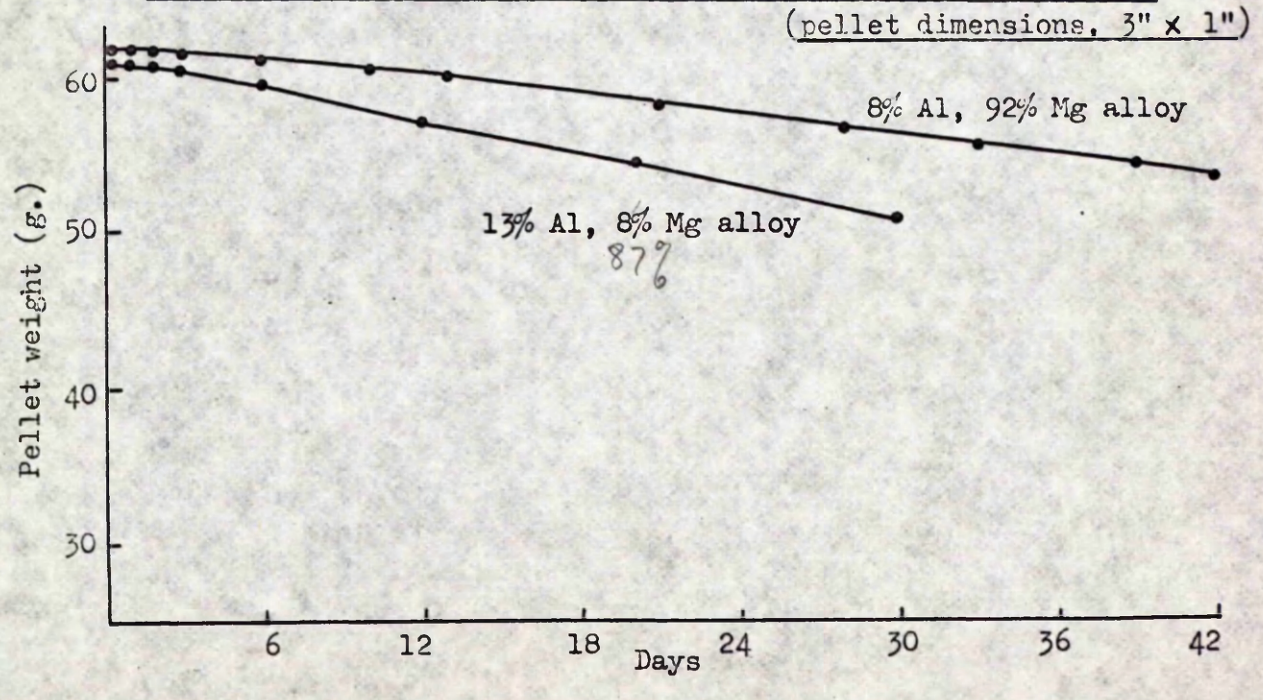


containing aluminium within these proportions was found to corrode evenly over the complete surface as shown in Fig. 27. This type of corrosion produced a regular release of material from the experimental pellet, and Fig. 28 shows the regular slow weight loss of two typical alloys of this class over a test period of 30-42 days. Corrosion in these alloys was evident on the pellet surface after 24 hours, but it can be seen from Fig. 28 that it was only after three days that the corrosion rate reached its maximum. Thereafter for the remainder of the test period, the corrosion rate remained constant. For the 3rd - 42nd day, the 8% aluminium alloy pellet lost weight at the rate of 0.154g./day. Since the alloy is 92% magnesium by weight, this represents a constant release of 0.141g. Mg/day. Similarly the 13% aluminium alloy pellet released 0.316g. Mg/day. Both these pellets were 3 inches in length and 1 inch in diameter, and contained between 53-57g. magnesium. If the release rate of material from the pellets remained constant until the pellets were completely dissolved, the 13% Al/87% Mg alloy pellet would theoretically have lasted for 168 days and the 8% Al pellet for 403 days.

These magnesium aluminium alloys therefore have the desirable properties of regularity of release of material, a fairly rapid commencement of activity, and a smooth pellet residue at any stage during the pellet's life. On the other hand the release rate, although regular, is not particularly high and the length of time for which the pellet will last is unnecessarily long.

Fig. 27 Original and partly worn rumen pellets composed of magnesium/aluminium alloy, removed from the rumen of a fistulated cow at varying periods of 10 - 80 days (original dimensions - 3ins. x 1ins.).

Fig. 28 Weight losses of rumen pellets composed ofmagnesium/aluminium alloy when tested in the rumeno-reticular sac of a fistulated cow.



The corrosion rate and the length of time for which the pellet will last are inversely proportional. Also, since corrosion takes place on the surface of the pellet, the corrosion rate will also be directly proportional to the surface area of the pellet and consequently it will depend on the pellet dimensions. Therefore these three variables of release rate, length of life and pellet size are all interrelated. It would be convenient to discuss at this stage the implications of this interrelationship and decide, if possible, on the most suitable figures to fix these variables at.

First of all the dimensions of the pellets can be arbitrarily decided on, since there is clearly a maximum size of pellet which can be administered to cattle and sheep. The shape is in some ways immaterial, but a cylindrical shape is a convenient one for administration to the animal, as well as having no dangerous sharp corners. It is also a shape which has a large surface area in relation to its volume and weight, which is an important point since a high corrosion rate is dependant on a large surface area. In practice, the 'effective' surface area is much greater than the 'apparent' surface area, due to the development on the pellet surface of numerous small pits which increase the exposed surface area. (see Fig. 27). For sheep, a suitable size of cylindrical pellet is suggested as being 1.75" in length and 0.75" in diameter. Such a pellet has been found in practice to be easily administered with an oesophageal balling gun although it it a size which is probably approaching the maximum which can be swallowed by a small sheep. For cattle a suitable size of cylindrical pellet is suggested as being 3 inches long and 1 inch in diameter. This is not the maximum size which could be administered to cattle,

but it was thought that any larger a pellet might prove difficult to handle from the point of view of the stockman. Assuming that pellets of these dimensionswere entirely composed of a magnesium/aluminium alloy within the range 8-20% Al(approximate density 1.84g./c.c.), the sheep rumen pellet would weigh 23g. of which 18-21g. would be magnesium, and the cattle rumen pellet would weigh 70g. of which 56-64 g. would be magnesium. This ignores in the meantime any weighting material which may have to be added to increase the overall density of the pellet.

The length of time for which the pellet should last can also be arbitrarily decided upon. One of the main advantages of any magnesium rumen pellet should be its ability to provide protection to the animal against hypomagnesaemic tetany over the complete period during which the animals may be at risk. In the case of sheep, this would be a period from lambing time to about six to eight weeks after parturition, which would cover in most cases the months of April and May. This would therefore be a period of about 50 days. For dairy cattle, the main period of risk is that immediately following the turn-out to grass in springtime, extending into May and June, which would again be a danger period of about 50-60 days. The period of risk for beef cattle is much longer, since hypomagnesaemic tetany may occur at any time from late autumn through to the springtime. It is not envisaged that a single treatment with rumen pellets could last over this long period. The minimum period over which pellets should last for sheep and dairy cattle is therefore considered to be about 50-55 days.

Of the three variables, pellet size, length of life and daily release

rate, which were listed above, both the pellet size and the length of life have now been arbitrarily fixed. The daily maximum release rate is thereby also fixed. For cattle, if a pellet of size 3" x 1" which contains 56-64g. magnesium is to have an active life of 50-55 days, the maximum possible release rate lies between 1-1.2g. Mg/day. Similarly a sheep pellet of size 1.75" x 0.75" containing 18-21g. magnesium must have a release rate no higher than 0.30-0.40g. Mg/day if it is to last for 50-55 days.

There are two further factors which may modify these calculations. One is the possibility to be discussed later, that some weighting material may have to be added to the pellet to increase its density in order to improve the retention of the pellet in the rumen. This would have the effect of reducing slightly the amount of magnesium in the pellet. For example the inclusion of sufficient iron particles in the pellet to raise the density to 2.70g./c.c. only reduces the magnesium content of a cattle pellet from 63g. to 55g. The second modifying feature is that as the pellet corrodes, so the size and the surface area of the pellet reduce. If the release rate of this type of pellet is a function of the surface area, the daily magnesium release will therefore tend to fall towards the end of the pellet's life.

Using the range of magnesium/aluminium alloys of between 8-20% Al by weight as a basis for further work, the aim of this experiment was now to increase the magnesium release rate of the cattle rumen pellet to around 1.0 - 1.2g. Mg/day and for the sheep rumen pellet to around 0.30-0.40g. Mg/day. The daily release rates of the 8% Al and the 13% Al magnesium alloy cattle pellets shown in Fig. 27 were 0.14g. Mg/day and 0.32g. Mg/day.

The next stage in this investigation was the finding that if a small quantity of a third metal such as copper, nickel or zinc was added to a magnesium/aluminium alloy, the corrosion rate of a rumen pellet made from this alloy was increased greatly as compared with the magnesium/aluminium alloy alone. Furthermore, these pellets still exhibited regularity in their release rate over a long period, remained relatively smooth and non-pitted, and started to corrode within 24 hours of administration to the cow. Fig. 29 shows the weight loss over a period of 45 days of an alloy composed of 8% Al; 3% Cu; 89% Mg. For comparison the graph also shows the weight loss of the 8% Al; 92% Mg alloy which was already given on Fig. 28. The copper containing alloy lost an average of 0.61g./day which represents a daily release rate of 0.54g. Mg/day, while the similar alloy with no copper had an average daily release of only 0.14g. Mg/day.

A wide range of these magnesium alloys containing aluminium in the range 8-20% plus a small quantity of a third metal were tested in cattle. Various metals were used for this third element e.g. mainly nickel, zinc, copper and cobalt but manganese, tin and calcium were also tried. The number of permutations for the formulation of an alloy containing magnesium, aluminium and any third metal is limitless, but an attempt was made to test at least a comprehensive selection of the formulations possible. The results of preceding tests were also used as a guide on deciding which new formulations to try in further trials.

Fig. 29 Weight losses of two magnesium/aluminium alloy pellets when tested in the rumeno-reticular sac of a fistulated cow.

(pellet dimensions, 3" x 1")

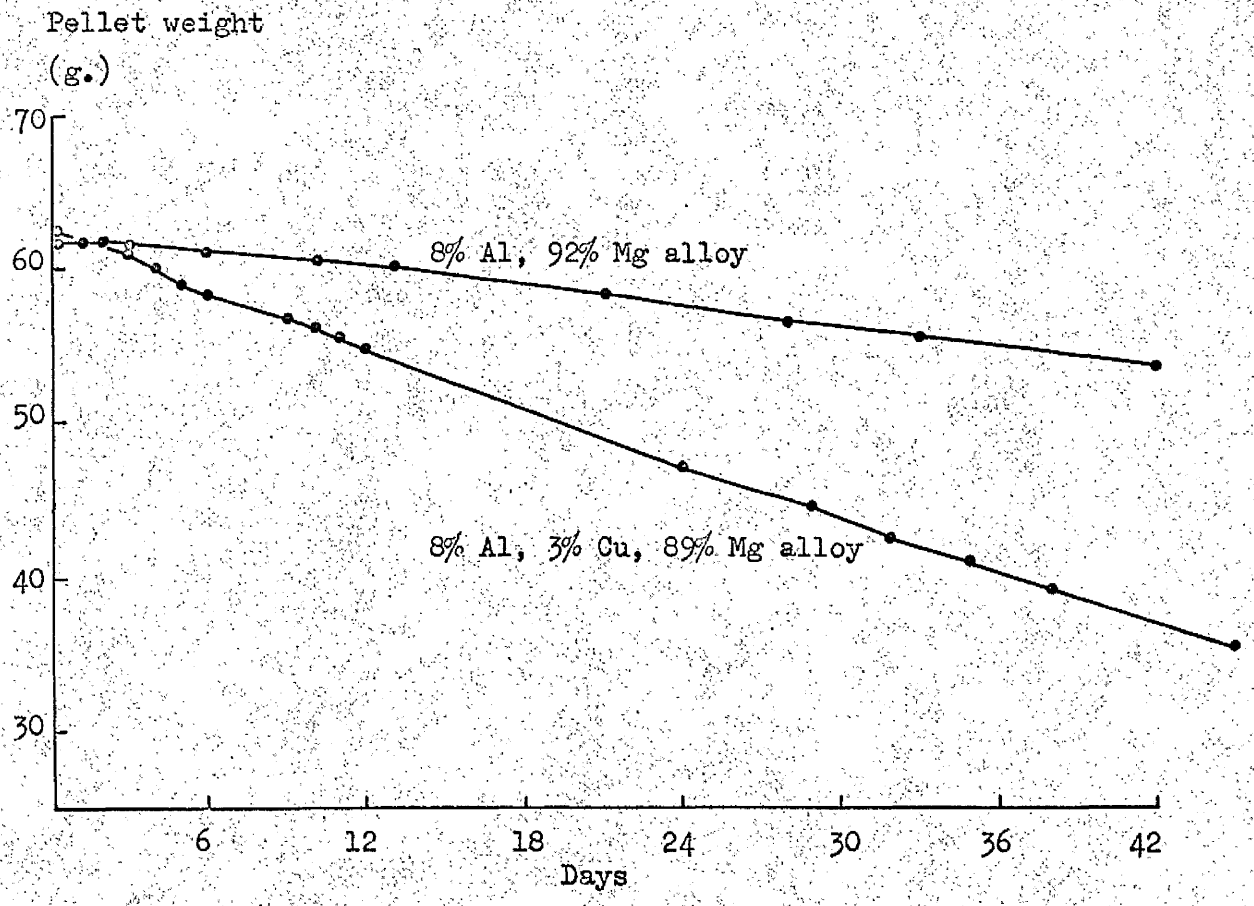


Table 26 lists a representative selection of the alloys which were manufactured and tested, together with a summary of the results. The size and the resultant 'apparent' surface of the pellet used are given in the table because in some of the earlier trials a smaller size of pellet was used with the result that the daily release rates are also smaller. Therefore for comparative purposes the daily magnesium release rate is also given per square cm. of surface area. This figure should be the same for a given formulation irrespective of the size of the pellet. For a cattle rumen pellet of size 3" x 1" to release 1g. Mg/day, this daily release rate would be 14 mg. Mg/sq.cm. pellet surface area/day. For a sheep rumen pellet of size 1.75" x 0.75" to release 0.30g. Mg/day, this value would have to be 9.5 mg Mg/sq.cm./day. It can be seen from Table 26 that the daily magnesium release rates from the various formulations of rumen pellet are variable. Since the tests were carried out with single experimental animals at different times of the year, the results should not be considered as being strictly comparative, but nevertheless some broad conclusions can be drawn as follows:-

(a) Alloys tested with below 8% Al or above 20% were unsuitable.

(b) The ability of a third element to increase the release rate varied according to which element was used and in what quantity. Generally it was found that nickel was more "active" than copper which was more "active" than zinc.

(c) At low aluminium contents (8%) a higher percentage of third element (e.g. 3% Cu) could satisfactorily be used, whereas at higher aluminium contents (13%) only a lower percentage of the third element (e.g. 0.5% Cu) could satisfactorily

Table 26. Experimental results for various magnesium alloy pellets tested in cattle and sheep.

Bullet Formulation (%) (remaining % is Mg.)	Animal No.	Diet.	Size in cms. Dia.	Lgth.	Initial Surface area. (sq.cms)	No. of days on test.	Ave. daily release rate. (g.Mg)	Ave. daily release per sq. cms. (mg.Mg)	Dissolution Characteristics.
(a) TESTS WITH CATTLE									
5Al,5Zn	Fist.	indoors	2.5	7.6	69.5	-	-	-	Rough unsuitable
7Al,0.5Ni	Fist.	grass	2.9	2.6	36.0		accelerating unsuitable		
8Al,0.1Co	Fist.	indoors	2.5	6.4	60.0	28	0.23	3.8	Good.
8Al,3Cu	Fist.	indoors	2.5	7.6	69.5	20	0.37	5.3	Good.
8Al,3Cu,1Co	Fist.	indoors	2.5	8.1	73.4	34	0.52	7.0	Good.
	6 spring grs.		2.5	7.6	69.5	21	0.93	13.4	Good.
	6 spring grs.		2.5	7.3	67.1	21	0.98	14.6	Good.
	5 spring grs.		2.5	8.0	72.6	36	0.81	11.2	Good.
8Al,5Cu,1Co	Fist.	indoors	2.5	7.4	67.9	30	accelerating unsuitable.		
10Al,0.5Ni	Fist.	indoors	0.95	4.2	30.7	16	0.13	4.4	Good.
10Al,2Cu	Fist.	indoors	2.5	7.7	70.2	36	0.31	4.4	Good.
10Al,5Zn	Fist.	indoors	2.5	7.2	66.3	28	0.23	3.5	Good.
12Al,2Cu	Fist.	indoors	2.5	7.6	69.5	18	0.60	8.6	Good.
12Al,2Cu	Various.	grass	2.5	7.6	69.5	av20	0.74	10.6	Good.
13Al,0.5Ni	Fist.	Spr.Grs.	2.7	2.4	31.8	10	0.46	14.4	Good.
13Al,0.5Ni	Various.	indoors	2.5	7.6	69.5	av46	0.60	8.6	Good.
13Al,1Ni	Fist.	indoors	2.9	2.4	33.4	11	0.23	7.6	Good.
13Al,1Ni	Fist.	spr.grs.	2.8	2.3	30.0	10	0.39	11.7	Good.
13Al,5Cu	Fist.	grass	2.8	2.6	35.0	9	0.28	8.0	Good.
13Al,2Cu	Fist.	grass	2.8	2.6	35.0	10	accelerating unsuitable.		
15Al,0.5Ni	Fist.	grass	2.7	2.7	34.3	15	0.21	6.1	Good.
15Al,1Ni	Fist.	grass	2.8	2.5	34.0	10	accelerating unsuitable.		
15Al,1Cu	Fist.	grass	3.0	2.6	38.6	9	0.34	8.8	Good.
15Al,3Zn	Fist.	indoors	2.5	7.6	69.6	37	0.42	6.0	Good.
20Al,5Ni	Fist.	grass	2.9	2.7	35.0	15	0.24	6.8	Reasonable.
20Al,0.5Cu	Fist.	grass	2.9	2.7	35.0	15	0.24	6.8	Reasonable.
25Al,0.5Ni	Fist.	grass	2.9	2.7	35.0	6	accelerating unsuitable.		
25Al,0.5Cu	Fist.	grass	2.9	2.7	35.0		jagged unsuitable.		
(b) TESTS WITH SHEEP									
5Al,5Zn	253	grass	0.8	1.8	35.0	32	.167	4.8	Reasonable.
8Al,1Co	253	grass	0.8	1.8	35.0	32	.183	5.2	Good.
8Al,3Cu	631	grass	0.8	1.8	35.0	7	.379	10.8	Badly pitted.
8Al,3Cu,1Co	Various	indoors			35.0	8	.700	20.0	Badly pitted.
8Al,5Cu,1Co	253	grass	0.8	1.8	35.0	32	accelerating unsuitable.		
10Al,5Zn	253	grass	0.8	1.8	35.0	32	.323	9.2	Pitted.
12Al,2Cu	(257	indoors	0.8	1.8	35.0	26	.324	9.2	Good.
	(948	grass	0.8	1.8	35.0	4	.424	12.1	Good.
13Al,0.5Ni	668	indoors	0.8	1.8	35.0	28	.424	12.1.	Badly pitted.
13Al,0.5Cu	685	indoors	0.8	1.8	35.0	28	.262	7.5	Reasonable.
13Al,2Cu	688	indoors	0.8	1.8	35.0	28	.262	7.5	Good.
15Al,0.5Ni	677	grass	0.8	1.8	35.0	28	.294	8.4	Good.
15Al,1Ni	699	grass	0.8	1.8	35.0	28	.703	20.1	Poor.
15Al,0.5Cu	689	grass	0.8	1.8	35.0	28	.215	6.1	Good.
15Al,3Zn	(257	grass				26	.370	10.6	Good.
	(253	grass				32	.296	8.5	Good.
15Al,5Zn	253	grass				32	.282	8.1	Good.

be used.

(d) The same alloy behaved differently in cattle and sheep and generally in sheep there was a greater tendency for some formulations to give undesirable corrosion characteristics, such as irregular pitting and accelerating release rates. Nickel was in every case too "reactive" for inclusion in sheep pellets.

(e) In every case where the same formulation of alloy pellet was tested in an animal indoors followed by a period of pasture grazing, the daily release of magnesium from the pellet was 60-80% higher when the animal was at grass. Since any future application of these pellets in practice would be in animals at grass, it is the release rate on pasture that was important.

(f) Daily release rates in tests with grazing animals were always higher in the springtime.

(g) In cases where pellets of the same formulation were tested in a fistulated cow and in a normal cow, where the pellet was recovered on slaughter, the daily release rate of the pellet was always higher in the normal cow. This suggests that in the fistulated animal, rumination is in some way slightly abnormal

From the tests with cattle, four particular formulations appeared to be promising.

These were (in percentages by weight) (a) 89Mg, 8Al, 3Cu, 0.1Co
(b) 86Mg, 12Al, 2Cu
(c) 86.5Mg, 13Al, 0.5Ni.
(d) 86.0Mg, 13Al, 1.0Ni.

From tests with sheep, two particular formulations appeared to be promising. These were (in percentages by weight) (a) 86Mg, 12Al, 2Cu
(b) 82Mg, 15Al, 3Zn

All of these formulations were found to release magnesium at a daily rate of above 10.5 mg/sq.cm. of surface area which would represent a release rate of over 0.70g. magnesium/day from a cattle pellet and 0.35g. magnesium/day from a sheep pellet. Only the formulation of 86% Mg/12% Al/2% Cu gave good results in both cattle and sheep.

Pellet Retention in the animal.

It was obviously important that any magnesium alloy rumen pellet would have to be well retained by the animal to be of any value in practice. As far as known, pellet losses are always by regurgitation and not by being passed down the intestinal tract. The earlier work on cobalt rumen pellets (Dewey et al. 1958) had suggested that a density of 4.00g./c.c. was essential for a pellet to be well retained by cattle and sheep. The magnesium oxide pellets, used in the work of Hemingway et al.(1961) had a density of 4.00g./c.c. and 90% were retained in sheep fed indoors.

Although this density of 4.00g./c.c. may be necessary when the pellet is small as in the case of cobalt rumen pellets, it was soon realised in the earlier

stages of the experiments on magnesium alloy pellets that retention was a function of both the density and the size of the pellet. Because of their initial length, the magnesium alloy pellets used in these experiments were still over an inch long when almost completely dissolved. Consequently, during the numerous tests carried out on various formulations of magnesium alloy pellets, both unweighted and cored pellets were used to determine whether it would be necessary to increase the density by incorporating some weighting material in the pellet in order to improve the retention rate. Table 27 summarises the collected results of these tests from the point of view of retention. The density of an unweighted magnesium alloy pellet is 1.84g./c.c.

Table 27. **Retention rate of magnesium alloy pellets by cattle and sheep.**

Average period of trial:- for cattle - 31 days, for sheep - 26 days.

(Pellets weighted by increasing amounts of iron)

	density:-	1.84		2.42		2.70	
	diet	indoor	grass	indoor	grass	indoor	grass
cattle	number of pellets recovered / pellets administered	46/47	9/16	11/11	6/6	116/116	48/50
	% retention	98%	56%	100%	100%	100%	96%

	density:-	1.84		2.25		2.50		2.90	
	diet	indoor	grass	indoor	grass	indoor	grass	indoor	grass
sheep	number of pellets recovered / pellets administered	24/33	21/51	36/40	—	38/40	—	65/65	127/154
	% retention	72%	41%	90%	—	95%	—	100%	83%

Separate results are given in Table 27 for tests on indoor and outdoor diets. It is immediately obvious that the retention rate is poorer when the experimental animals are at pasture as compared with an indoor diet of hay and concentrates. This is particularly so for the unweighted pellets of density 1.84g./c.c. It is also obvious from Table 27 that the retention of alloy pellets is invariably better in cattle than in sheep.

With cattle, the retention rate of unweighted pellets when the test was carried out indoors was almost 100% (the one pellet missing was almost certainly all dissolved and not regurgitated) whereas the same pellets were poorly retained (56%) at pasture. Increasing the density of the cattle pellet to 2.42g./c.c gave retention figures of 100% under all dietary conditions. It was unfortunate that the misleading result of 100% retention of uncored pellets in cattle on hay and concentrate diets led to uncored pellets being used in a series of experiments which were conducted in dairy cattle in the following spring season. After the cattle went to grass, it was then found that the unweighted pellets were being rejected. In subsequent trials with cattle, rumen pellets with a density of 2.70g./c.c. were thereafter used.

With sheep, Table 27 shows that where trials were conducted indoors, the retention rate increased from 72% to 100% as the density of the pellets increased from 1.84 to 2.00g./c.c. At grass the retention of uncored pellets was very poor (41%). For heavier pellets, the retention rate at pasture was 83%, although it is felt that this figure is slightly lower than it should be, due to the strong possibility that some of the pellets dissolved completely before the end of the experiment, rather than were lost by regurgitation. This retention

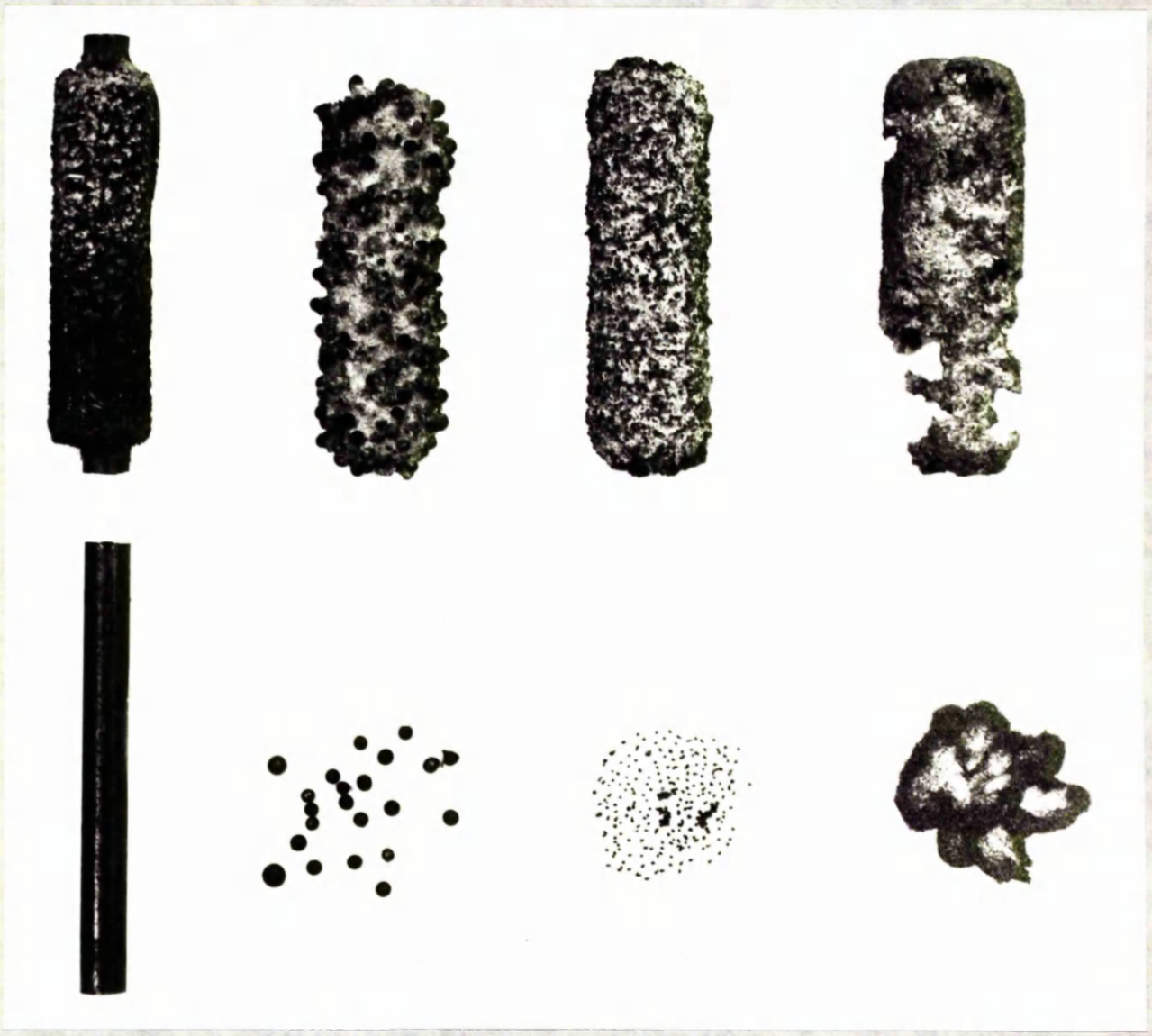
rate of 83% for pellets of density 2.90g./c.c. compares favourably with the 90% retention found for magnesium oxide pellets of density 4.00g./c.c. by Hemingway et al. (1961) in indoor fed sheep. It would therefore appear that there would be no benefit to be derived from increasing the density to 4g./c.c. since even at this high figure, retention is not 100%. In all subsequent experiments with sheep, pellets of density 2.90g./c.c. were used.

Iron was found to be the most suitable material to incorporate as a weighting ingredient in magnesium alloy pellets, since it has a high density (7.8g./c.c.) and is therefore only needed in small volumes, plus the fact that it is non-toxic to animals. For experimental purposes iron was initially incorporated as a rod, forming a central core to the pellet (Fig.30), but this was thought to be an unpractical measure for future use, since it meant that the steel core would be left in the animal as a permanent residue after the magnesium alloy had completely dissolved. The iron present in the pellet would have to be in a 'disposable' form and therefore be present as small discrete particles which could be passed on by the animal. Fig. 30 shows three types of weighted pellets which were devised to achieve this aim. Example (b) has particles of large size iron shot (diameter $^{3}/_{32}$"-$\frac{1}{8}$") distributed evenly throughout the pellet. This type of pellet gave reasonably regular release rates but the part-worn pellet tended to be very rough on the surface. Example (d) of Fig.30 was a uniform mixture of magnesium alloy and fine iron powder. The inclusion of iron powder in this fashion completely changed the corrosion characteristics of the magnesium alloy, and it resulted in a deeply pitted pellet with an erratic, rapid release rate. Example (c) was the most suitable type tested. It was a uniform

Fig. 30 Four methods of including iron in a magnesium rumen pellet to increase the overall density of the pellet.

Weighting material -:

central steel pin	iron shot of diam. $\frac{1}{8}$"	iron shot of diam. 1/32"	fine iron powder



mixture of alloy and small iron shot (diameter $^{1}/_{32}$"). As can be seen from Figs. 30,31, the resultant pellet corroded smoothly and further tests showed that it gave a regular daily release rate similar to the release rate of an uncored pellet of the same formulation. It was not possible to make this type of uniform mixture of iron shot and magnesium alloy without specialised equipment and techniques and consequently these pellets were prepared by a foundry firm specialising in magnesium products. The cattle pellets are composed of 61% magnesium alloy by weight (87% by volume) and 39% iron by weight (13% by volume) which gives an overall density of 2.70g./c.c. and a total weigh of 80-85g. The sheep pellets which have a higher density of 2.90g./c.c. weight 34-36g. and are composed of 49% by weight magnesium alloy (80% by volume) and 51% by weight iron shot (20% by volume). Thus the inclusion of the weighting material reduces the magnesium alloy content of the pellet by only 13-20% depending on the density required.

The most suitable formulation of magnesium alloy for rumen pellets was finally chosen as being composed of 12%Al/2%Cu/86%Mg. A list was given earlier (page 230) of formulations which behaved well in corrosion tests in cattle and sheep, and this was the only alloy formulation which behaved well in both species. Partly for this reason, and partly for other technical reasons, it therefore seemed the most suitable to use in future trials. Fig. 31 shows a sample of a cattle pellet and a sheep pellet in their final form together with examples of partly corroded pellets. Table 28 gives the complete specifications of these pellets.

Fig. 31 The final form of magnesium alloy rumen pellet as developed in Experiment 9 for use in cattle and sheep.
Examples of original and partly worn pellets
Specifications as given in Table 28.

Table 28. Specifications of magnesium alloy rumen pellets for cattle and sheep.

	Cattle pellet.	Sheep pellet.
Length (inches)	3"	1.75"
Diameter (ins)	1"	0.75"
Composition (by wt.)	61% alloy 39% iron shot	49% alloy 51% iron shot
(by volume)	87% alloy 13% iron shot	80% alloy 20% iron shot
Alloy composition	86%Mg, 12%Al, 2% Cu.	86% Mg, 12% Al, 2% Cu.
Iron shot size (ins)	1/32"	1/32"
Overall density (g./c.c.)	2.70	2.90
Total wt. (g.)	80-85	34-36
Total Magnesium content (g)	43-45	15-16
Average magnesium release rate (g/day)	0.8-0.9	0.25-0.35
Expected pellet life	50-60 days	50-60 days

As evidence for the average release rates quoted in Table 28, for both cattle and sheep pellets, Table 29 lists the release rates found for a number of pellets made to these specifications and given to experimental animals at pasture, where pellet recovery was made at slaughter.

Table 29. Average daily magnesium release rates of rumen pellets manufactured to the specifications listed in Table 28.

Cattle pellets.			Sheep pellets.		
Cow No.	Test period (days)	Average daily Mg. release rate (mg/day)	Sheep No.	Test period (days)	Average daily Mg. release rate (mg/day)
Vera	15	855	791	10	235
		706		17	247
Matilda	17	605		23	235
		600	794	10	266
Wye	22	979		17	243
		753		23	291
Violet	22	863	0715	27	331
		764	738	29	325
A283	41	852	745	29	269
		870	808	29	400
Mean release for 10 pellets	23 days	785 Mg mg/day	Mean release for 10 pellets	21 days	284 285 mg Mg/day
		SD 122.3			53.5
		CV 15.6%			19.9%

The results given in Table 29 were for tests carried out in September-October 1965 and past experience (Table 26) has tended to show that release rates on spring grass were always higher than at later seasons in the year. Consequently the release rates of these pellets tested in Table 29 may be higher in the spring. One point which is brought out by Table 29 is that the daily release rates of sheep pellets are more variable than those of cattle pellets. With sheep the variation was from 235-400 mg magnesium released per day (mean 335 mg/day). Previous tests with other formulations gave the same wide variability in sheep. With cattle the daily release rate was more consistent in that for the 10 pellets tested, the variation between individual pellets in 5 different animals was from 600-979 mg. magnesium released per day. (mean 785 mg Mg/day).

Measurement of the magnesium loss of these weighted pellets could not be done by merely weighing the pellet before administration and after recovery, since the weight lost by the pellet includes the weight of some iron shot in addition to alloy. An estimate of the alloy loss (and thereby the magnesium loss) can be arrived at by measuring the pellet density before and after recovery. The density is a direct function of the proportions of alloy to iron in the pellet and if the exact densities of the mixture and the two ingredients, alloy and iron are known, then the alloy content of the pellet can be calculated from the following equation.

$$\text{alloy content}(g) = \frac{100}{0.4163D} - 30.56$$

where density of iron shot = 7.8g./c.c.
" density of magnesium alloy = 1.84g/c.c.
" density of pellet = D.g/c.c.

The pellet density was measured by taking the weight of the pellet in air and the weight suspended in water, from which the pellet volume can be calculated. By combining these two steps, the alloy content was calculated from the following equation

alloy content(g) = 2.4021W - 0.3056M where M = wt. in g. of pellet in air
W = wt. in g. of pellet suspended in water.

One further test which it was thought advisable to carry out was on the question of whether the small iron shot particles were excreted by the animal when the surrounding alloy was dissolved away. Consequently two sheep were put into metabolic cages which allowed the faeces to be collected daily for examination. The sheep were given a normal ration of hay and concentrates over the 16 day experimental period. A gelatin capsule containing a known weight of approximately 35g. of loose iron shot was administered orally to each sheep on day 0 (7th December 1965). This represents the quantity of iron shot which would be left as a residue from two sheep rumen pellets. For the following 15 days, the total faeces of each sheep were removed daily and examined for excreted particles of iron shot. This was done by breaking down the faeces pellets in a blender, pouring the resulting liquid into a plastic bucket at the bottom of which was placed a strong magnet, and decanting the supernatant liquid. Any iron shot particles present were retained by the magnet. It had earlier been found that a quantitative recovery (100%) of iron shot particles, mixed in faeces, was achieved by this method. At the end of the 15 day period, the sheep were slaughtered and the rumen contents examined for

iron shot. Table 30 details the weight of iron shot which was recovered from the faeces of the two sheep .

Table 30. Iron shot (diameter 1/32") excretion in two sheep.

	ewe no. A 436		ewe no. A 433	
day 0-:	37.040g. iron shot administered.		34.983g. iron shot administered	
day	shot excreted in faeces(g.)	cumulative total.(g.)	shot excreted in faeces(g.)	cumulative total(g.)
1	.156	0.156	.055	.055
2	8.689	8.845	.140	.195
3	9.761	18.606	.924	1.119
4	1.331	19.937	1.974	3.093
5	4.928	24.865	0.170	3.263
6	0.243	25.108	0.743	4.006
7	0.501	25.609	5.018	9.024
8	0.156	25.765	0.183	9.211
9	0.026	25.791	0.059	9.266
10	0.016	25.807	0.003	9.269
11	0.042	25.849	0.017	9.286
12	0.130	25.979	0.052	9.338
13	0.024	26.003	0.166	9.504
14	0.097	26.100	0.092	9.596
15	0.097	26.197	0.423	10.019
cage cleanings	0.145	26.342	6.159	16.178
.. % of shot administered which was recovered in the faeces		71.1%		46.2%
residual iron shot found in the rumen (g.)		7.543		15.383
.. total shot recovered (g.)		33.885		31.561
% " " "		91.5%		90.5%

These results demonstrated that iron shot particles of 1/32" diameter were gradually eliminated by sheep in the faeces. Over the 15 day period for which the faeces were collected, one sheep eliminated 71% of the 37g. of shot which was originally present, and the second sheep eliminated 46% of the 35g. originally present. Presumably the remainder of the shot would have been excreted gradually had the experimental period been extended. On slaughtering the sheep, some iron shot was found to be still present in the rumeno-reticular sac and, altogether, some 90-91% of the iron shot which was originally administered was accounted for.

Conclusion.

The aim of this experiment was to devise a rumen pellet treatment capable of supplying at about 0.6g. magnesium/day to sheep and about 5g. magnesium/day to cattle and in each case lasting for a period of 50-55 days. The result of the experiment was the development of a rumen pellet for sheep, releasing an average of 0.30g. magnesium/day and a rumen pellet for cattle releasing an average of 0.90g. magnesium/day. It is therefore proposed that a suitable treatment for sheep would be 2 magnesium alloy rumen pellets thus supplying an average of 0.60g. magnesium/day over a period of 50 days. Similarly a suitable treatment for cattle would be 4 magnesium alloy rumen pellets which would supply an average of 3.6g. magnesium/day over a period of 50-60 days with the possibility that the release rate would be higher at spring grass. This quantity for cattle is slightly below that which was aimed at, but a treatment of four pellets was considered as a maximum number which could be given in

practice. Further experimentation should indicate whether the 3.6g. Mg/day supplied by this treatment is sufficient to give a prophylactic effect against hypomagnesaemic tetany.

The effectiveness of these rumen pellets for cattle and sheep in preventing the development of hypomagnesaemia and hypomagnesaemic tetany forms the subject matter of Experiments 10-15.

Experiments 10-12.

The Use of Magnesium Rumen Pellets in Cattle at Grass.

Experiments 10-12 were designed to test under practical farming conditions the efficiency of magnesium alloy rumen pellets to prevent the development of hypomagnesaemia and hypomagnesaemic tetany in cattle. Experiments 10 and 11 were conducted in dairy herds and Experiment 12 in a beef herd. The site for these trials were initially chosen as farms on which there was a past history of hypomagnesaemic tetany outbreaks in the herd. In each case, half of the herd was given a four pellet treatment, and it was hoped that there would be a sufficiently high incidence of hypomagnesaemic tetany during the experiments to demonstrate the effectiveness or otherwise of the rumen pellet treatment. Blood samples were also taken regularly to determine the effect of the treatment on the plasma magnesium levels.

At the time these experiments were carried out (spring season 1965), it was believed, on the basis of the indoor trials quoted in Table 27, that unweighted pellets were satisfactorily retained by cattle, and uncored pellets were therefore used on all three sites. The alloy formulation was in each case 85% magnesium, 12% aluminium, 3% copper and the size 3" in length and 1" in diameter. Unfortunately it was subsequently found (see results of Experiments 11 and 12) that the retention of these pellets by the animal was not 100%.

The conclusions which can be drawn from the results of these experiments with cattle will be given in conjunction with those from the Experiments 13-15 on sheep rumen pellets at the end of this section.

Experiment 10.

Site - Hairmyres Farm, East Kilbride. There was a past history of severe outbreaks of hypomagnesaemic tetany on this farm, averaging out at 6-7 cases per year in a herd of 90 dairy cows. These outbreaks occurred despite the use of a conventional preventive treatment of 2 ozs calcined magnesite mixed with the concentrate feed.

Experimental Design.

The complete herd of 90 milking cows on this farm was involved in this experiment. On the 27th April, 1965, three days prior to the herd being put to pasture, 45 of the cows were given a treatment of four magnesium rumen pellets, and the remaining 45 cows were left as controls. Randomisation of these two groups was achieved by treating every second cow coming through the milking parlour. For the duration of the experiment, no other form of magnesium supplement was given to the herd. Blood samples were taken from ten cows, selected at random, in each group on the day the pellets were administered and from the same twenty cows on three subsequent occasions on the 11th and 21st May and 1st June. The pellets were unweighted (density 1.84g./c.c.)of size 3" in length and 1" in diameter and were composed of 85% Mg., 12% Al., 3% Cu.

Results.

Clinical cases:- Two cows in the control group were reported as cases of hypomagnesaemic tetany. Blood samples taken by the Veterinary Practitioner

at the time of treatment gave the following results on plasma analyses.

	Plasma Mg. (mg/100 ml)	Plasma Ca (mg/100 ml)
Cow No. 56	0.67	4.03
Cow No. 70	1.00	10.16

Cow No. 56 showed the typical clinical signs of this disorder, whereas cow No. 70 was merely excitable and nervous, and these signs were taken as an early indication of hypomagnesaemic tetany. Veterinary treatment was successful in both cases. There were no clinical cases reported in the group given the rumen pellet treatment.

Plasma Analyses:- The plasma magnesium concentrations of the 10 cows of each group which were sampled on four occasions are given in Table 31 together with the mean levels for the two groups. Only the mean plasma calcium values are given in Table 31 since the individual plasma calcium values were all within the normal range, apart from that of the clinical case (cow No. 56) as quoted above. The results are also shown graphically on Fig. 32 with the mean plasma magnesium levels drawn and the individual results plotted for each group to give a visual comparison of the results.

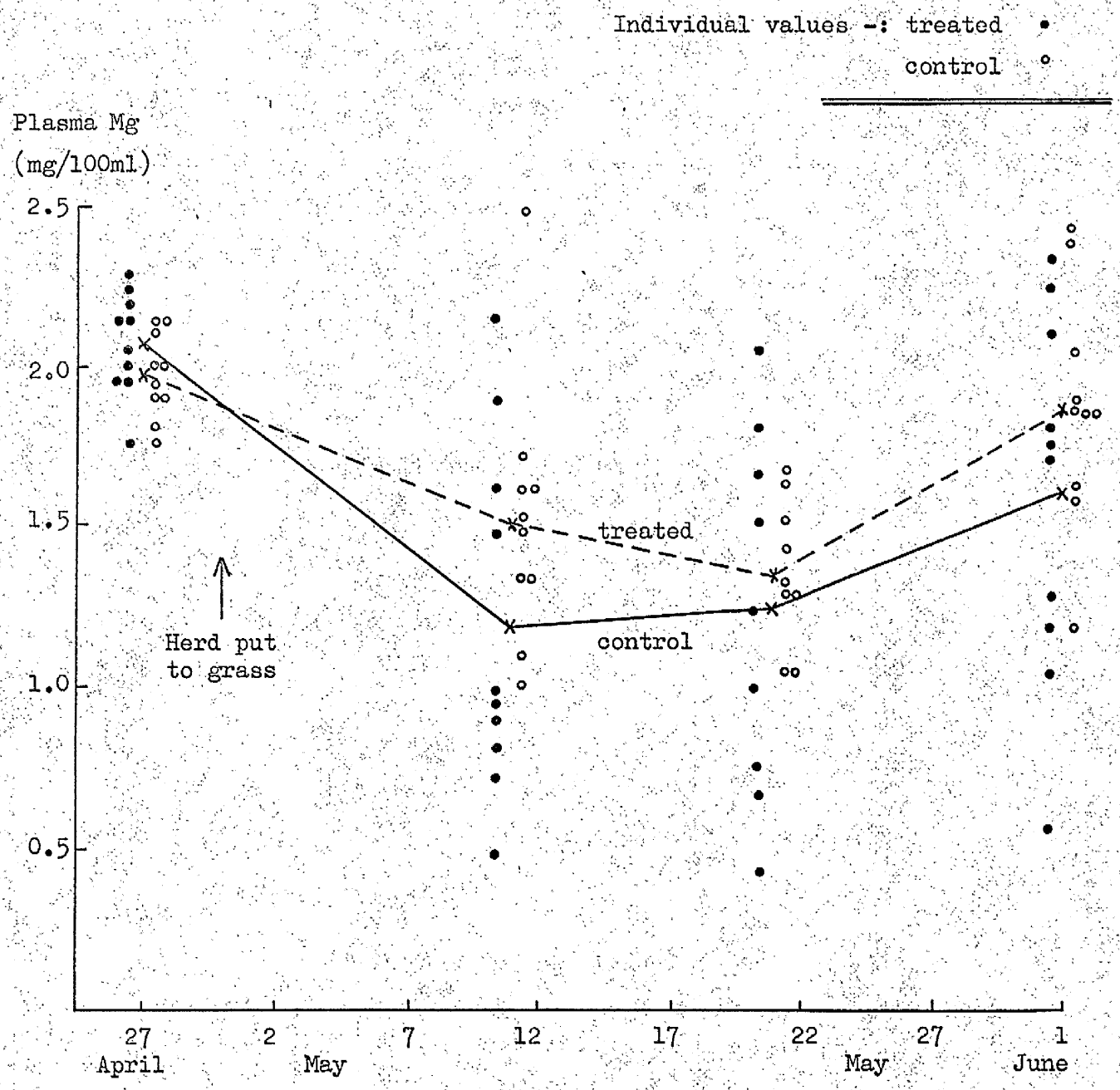
Rejected pellets:- One pellet was found lying in the cattle court. One of the treated cows was slaughtered on 17th June, which was 51 days after the administration of the treatment. No pellets were found in the rumeno-reticular sac of this cow. Since the farm was run on a milking parlour system and the cows were not kept in a byre, pellets may have been rejected without being found either in the collecting yard or in the field.

Table 31. Individual plasma magnesium concentrations and mean plasma magnesium and calcium levels of two groups of 10 milking cows at grass.

(mg/100 ml)

Treated group (4 magnesium pellets)					Control group.				
Cow No.	27/4	11/5	21/5	1/6	Cow No.	27/4	11/5	21/5	1/6
98	2.10	1.33	N.S.	2.43	56	1.76	0.90	0.76	0.57
4	1.95	1.48	1.43	1.86	8	2.19	0.81	0.43	1.05
20	2.00	1.10	1.33	1.19	105	1.95	0.48	1.24	1.81
54	2.14	1.52	1.29	1.86	5	214	0.95	1.00	1.29
87	1.76	1.33	1.62	1.86	102	1.95	0.71	1.67	1.76
42	2.00	1.00	1.05	1.62	36	2.05	1.00	1.52	1.71
33	1.81	2.48	1.29	2.38	66	2.29	1.90	0.67	1.19
12	1.90	1.71	1.52	2.05	49	2.24	1.48	N.S.	2.10
84	2.14	1.62	1.67	1.90	40	2.14	2.14	2.05	2.33
107	1.90	1.62	1.05	1.48	43	2.00	1.62	1.81	2.24
MEAN	1.97	1.51	1.34	1.86	MEAN	2.07	1.19	1.24	1.60
Mean Plasma Calcium levels	9.83	10.19	10.48	10.57	Mean Plasma Calcium levels	9.63	10.31	10.23	10.79

Fig. 32 Individual and mean plasma magnesium concentrations of 2 groups of 10 milking cows at grass, one group having been given 4 magnesium rumen pellets on 27th April (Experiment 10).



Discussion.

Of the two cases of tetany reported in the control group, one was definitely a true hypomagnesaemic tetany condition based on the clinical symptoms and confirmed by the low plasma magnesium and calcium concentrations found in the sample taken prior to treatment. Considerable doubt existed as to whether the second case was in fact "staggers", since the analysis of blood taken at the time showed normal plasma magnesium and calcium concentrations. Two cases in the group of 45 animals represented a 4.5% incidence. The 'high' incidence of clinical cases, predicted by the farmer did not occur, presumably due to seasonal factors.

There was a measurable response to the treatment as indicated by analysis of the plasma samples for magnesium. Fig. 32 shows that after the herd was put to grass, the mean plasma magnesium level of the 10 treated cows which were sampled fell to only 1.51 mg/100 ml as compared with the control group mean level of 1.19 mg/100 ml. This difference between the groups still existed on subsequent sampling dates. Rather more important, however, than the actual mean levels was the trend shown by the individual plasma magnesium concentrations. If the individual results are considered for the 11th May, 5 of the 10 sampled cows in the control group (i.e. 50%) had plasma magnesium levels below 1.00 mg/100 ml. In contrast, none of the treated group were below 1.00 mg/100 ml on this date, nor on either of the two subsequent sampling dates. On the 21st May, three of the control animals were below this level of 1.00 mg/100 ml and on the last sample taken on the 1st June, one individual still had a plasma magnesium concentration of below 1.00 mg/100 ml.

This finding is considered to be of importance since it is generally accepted that the level of 1.00 mg/100 ml is a critical level below which there is a real risk of hypomagnesaemic tetany occurring. Consequently, consideration of the mean figures alone tends to hide the fact that at one point, 50% of the untreated animals were what is considered to be at potential risk, whereas none of the individuals in the treated group were at any time in this position.

The statistical treatment of the results was made difficult by the large spread of individual values within each group e.g. on the 11th May, the individual values of the control group varied from 0.48–2.14 mg/100 ml. Consequently, considering the results as a whole, there was no statistical difference between the two groups in plasma magnesium level, due to the large standard error introduced by the wide spread of individual values. If, however, the six animals in each group with the lowest values on the 11th May are considered, then the spread of values is reduced by the consequent elimination of the higher values. In Table 31, these six animals of each group are listed at the top of the table. This is believed to be a justifiable procedure since animals withhigh plasma magnesium concentrations are not at risk from staggers and it is therefore the animals with the lower values which are of particular interest.

Considered on this basis, there was a highly significant difference ($P = 0.01$) between the six animals with the lowest values in each group on the 11th May. In contrast, there was no significant difference between the same groups of six animals at the start of the experiment. The mean plasma magnesium levels of these two groups of six were:-

	27/4	11/5	21/5	1/6
treated	1.93	1.28	1.34	1.79
control	2.01	0.81	1.09	1.36

The occurrence of one clinical case (and one doubtful) in this control group is not sufficient to give definite evidence on the ability of a pellet treatment to prevent hypomagnesaemic tetany, but the plasma magnesium data do suggest that the rumen pellet treatment prevented the development of severe hypomagnesaemia. The plasma magnesium values were therefore maintained at a level above that at which hypomagnesaemic tetany might be expected to occur.

Experiment 11.

Site : Basket Farm, High Blantyre. This was a farm which also had a previous history of hypomagnesaemic tetany, and two or three cases usually occurred in each spring season immediately after the herd was put to pasture. There had been no attempts made in the past, however, to prevent the condition by feeding magnesium supplements.

Experimental Design.

The design of this experiment was very similar to that of the preceding Experiment No. 10. The complete herd of 62 cows were involved in the trial and on 13th May, a treatment of four magnesium rumen pellets was given to 31 cows, with the remaining 31 cows being left as a control group. Randomisation of the two groups was achieved by giving the treatment to every second cow in the sequence in which they stood in the byre. The herd was put to pasture two days after this treatment was given. The pellets used were of the same design as those used in Experiment 10. Blood samples were taken from 16 animals in each group on the day the treatment was given on the 13th May, and from the same total of 32 animals on three subsequent occasions at weekly intervals thereafter. At no time during the trial was any other magnesium supplement offered to the animals.

Results.

Clinical cases:- Three cows, all in the control group, were reported by the Veterinary Practioner as being cases of hypomagnesaemic tetany. These were:

(1) Cow No. 151. developed clinical signs on three occasions on 17th and 21st May and on 9th June. The animal died on the third occasion. It had plasma magnesium and calcium concentrations of 0.14 and 10.0 mg/100 ml respectively on 20th May which was one day prior to the second attack.

(2) Cow No. 146 developed obvious signs of mild tetany on the 26th May. For example it held its head back, walked with an exaggerated gait and showed signs of extreme nervousness to stimuli such as sound and touch. When last sampled on 20th May, this animal had the extremely low plasma magnesium value of 0.10 mg/100 ml and a plasma calcium value of 10.24 mg/100 ml.

(3) Cow No. 125 was reported as being nervous and excited on the 20th May, but no other symptoms were recorded. On that date the plasma magnesium concentration was 0.62 mg/100 ml and the calcium level was 9.84 mg/100 ml.

No cases or signs of hypomagnesaemic tetany were reported in the treated group.

Plasma Analyses : Table 32 details the individual and mean plasma magnesium concentrations for the two groups of 16 sampled cows on the four sampling dates. Since there were no individual plasma calcium concentrations recorded outwith the normal range, only the mean calcium levels are given.

Table 32. Plasma magnesium concentrations of two groups of sixteen milking cows at grass (Basket Farm - 1965).

Plasma magnesium values (mgms/100 ml)

4 pellet treatment					CONTROL				
Cow No.	13/5	20/5	27/5	3/6	Cow No.	13/5	20/5	27/5	3/6
83	1.76	1.38	1.81	1.90	151	1.93	0.14	0.24	0.52
61	1.95	1.05	2.19	1.71	124	2.10	0.62	0.95	1.71
62	1.86	1.33	1.43	1.62	125	2.33	0.76	0.62	1.43
63	1.90	1.52	1.52	1.71	126	1.86	0.38	0.90	1.62
64	1.90	1.71	2.10	1.95	130	1.52	0.19	0.33	1.81
65	2.10	0.90	0.86	2.24	146	1.81	0.10	0.62	1.48
66	1.81	1.76	1.71	1.71	134	2.10	0.90	1.38	2.10
67	1.57	1.48	0.62	1.62	121	2.14	1.24	1.29	1.52
68	2.43	1.67	2.05	2.05	122	1.95	1.24	1.57	2.43
69	1.76	1.14	0.76	2.00	123	1.57	1.52	1.76	2.29
70	2.00	1.81	1.71	2.10	127	2.29	2.24	1.67	2.57
71	2.05	1.86	1.57	2.24	128	2.29	1.48	1.24	1.90
72	2.00	1.62	1.57	1.76	129	1.62	1.38	1.24	1.81
73	1.67	1.14	1.76	1.81	131	1.67	1.29	1.38	1.57
74	1.90	1.76	1.57	2.24	132	1.90	1.14	1.76	1.81
75	1.57	1.05	1.29	1.86	133	1.81	1.71	1.67	1.90
MEAN	1.89	1.45	1.53	1.91	MEAN	1.93	1.02	1.16	1.78

Mean Plasma Calcium Values.	13/5/65	20/5/65	27/5/65	3/6/65
Treated	9.70	10.01	10.04	10.09
Control	9.46	9.88	10.08	10.31

The results are also shown graphically on Fig. 33 with the mean plasma magnesium levels of the two groups drawn over the four week period, together with the individual values plotted on each date.

Rejected Pellets: A number of regurgitated pellets were found by the farmer during the course of the experiment. The majority of these were found in the feeding troughs in the byre after milking time, and therefore the date of rejection was known exactly. The final weight of these rejected pellets was determined and although the initial weight of each individual pellet was not known exactly, the number of rejected pellets was sufficiently large to allow the average weight of a similar batch of unworn pellets to be used in computing an average magnesium daily release rate for these rejected pellets. Table 33 lists the weight of these pellets, the number of days for which it was retained, and the number of the cow from which the pellet was recovered.

Fig. 33 Individual and mean plasma magnesium concentrations of 2 groups of 16 milking cows at grass, one group having been given 4 magnesium rumen pellets on 13th May (Experiment 11).

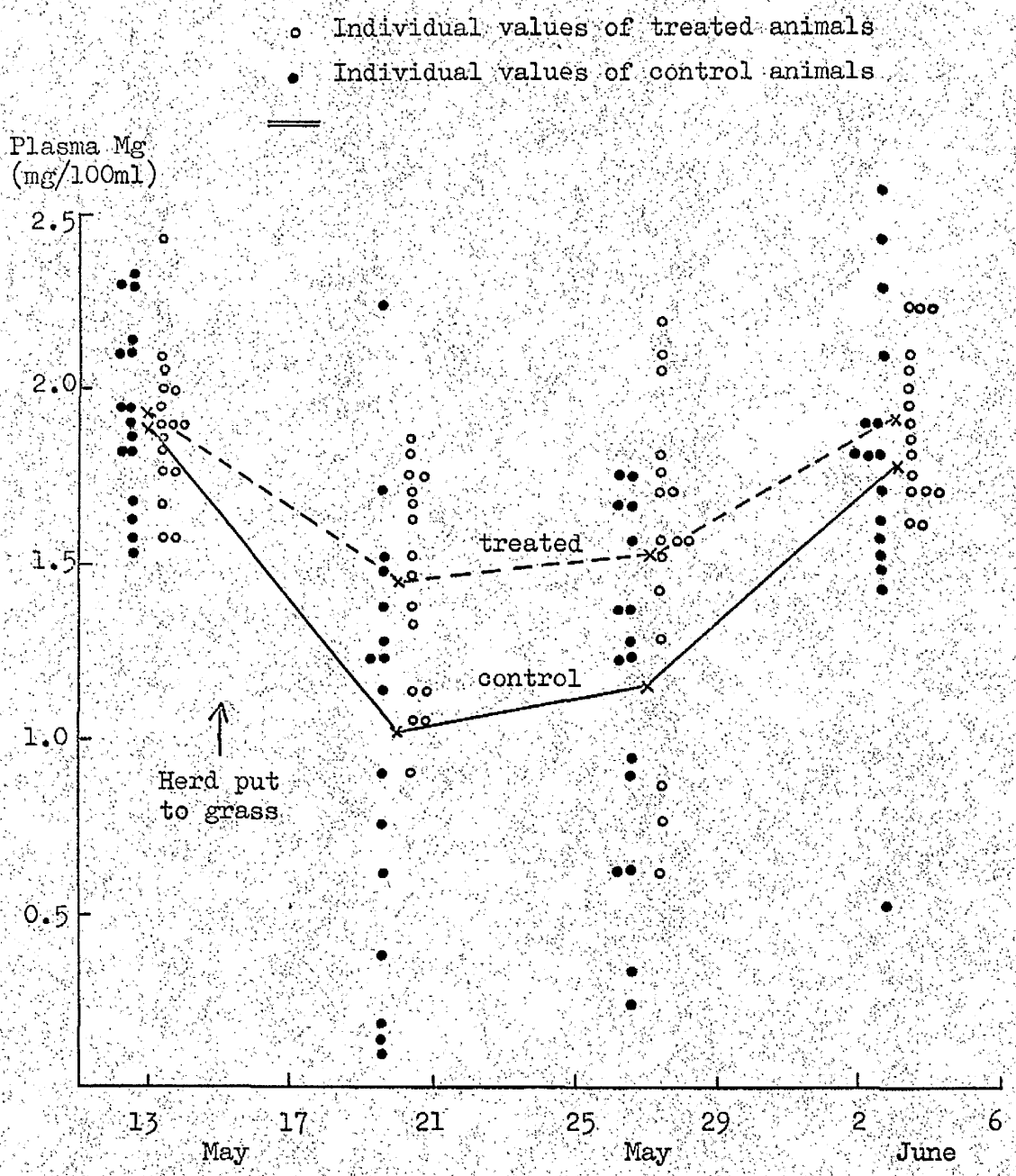


Table 33. Weight of regurgitated pellets and the date of rejection from cattle involved in Experiment 11.

Cow No.	Approx. or exact period of retention (days).	Final Pellet Weight.(g.)	Initial Pellet Weight.(g.)
? in field	2-7	51.229	
? in field	2-7	55.241	
? in field	4-5	52.783	
? in field	4-5	51.356	
? in field	4-5	51.200	mean wt. of
89	7	53.799	60 unworn
87	8	50.550	pellets:-
88	8	51.229	59.857
77	11	47.759	
88	12	44.668	range:-
66	13	49.153	57.8-63.8
66	13	46.906	
?	13	45.932	
80	13	46.253	
75	15	39.491	
88	16	38.898	
Total for 16 pellets	151 days	776.447	
Mean	9.5 days	48.527	59.857

Mean weight loss of 16 bullets = 11.332 g. over 9.5 days.
= 1.193 g. alloy/day.

average supplied by each pellet = 1.01 g. magnesium/day.

The data of Table 33 confirms that the release rate of the pellets was very close to the forecasted figure of 1.00 g. magnesium/day/pellet.

The regurgitation of 16 pellets over the first 16 days of the experiment represents a 13% rejection rate i.e. 87% of the pellets administered were possibly retained. Over the following 20 days, a further 14 pellets were found, mainly in the pasture field. Since the exact date of rejection was not known, no estimate of the daily release rate can be made for these 20 pellets.

A total of 30 pellets were therefore known to have been rejected within 36 days of their administration. This represents a minimum rejection rate of 24% i.e. maximum retention percentage of 76%. This figure is, however, based on rejected pellets which were found, and since there would certainly be more which were regurgitated and not picked up, the retention rate was undoubtedly lower than the 76% quoted here.

Discussion.

The two definite cases of clinical tetany which were reported in the untreated group of 31 cows represented a 6.5% incidence. The third case which was reported in this group has been taken as doubtful, although it had a sufficiently low plasma magnesium concentration (0.62 mg/100 ml) to have been on the verge of succumbing to this condition. It is noteworthy that no clinical cases were reported in the group given the rumen pellet treatment.

The results for the plasma analyses again show a definite response in the mean plasma magnesium levels to the use of a magnesium pellet treatment. The pattern of the results closely followed that found in Experiment 10, in that

there was a sharp fall in the plasma magnesium values with five days of the cows being put to grass. However, whereas the control group (of 16 sampled cows) fell to a mean level of 1.02 mg Mg/100 ml, the treated group maintained a higher level of 1.45 mg Mg/100 ml. This difference between the groups was statistically significant ($P = 0.02$). On the following sampling date on 27th May, the mean plasma magnesium level of the treated group rose to 1.53 mg/100 ml and this level was still significantly higher ($P = 0.05$) than the control group which had a mean level of 1.16 mg/100 ml. By the last sampling date on 3rd June, the plasma magnesium of both groups had risen to a higher mean level at which a pellet effect would no longer be expected to be found. There was therefore no significant difference between the groups on this date.

Quite apart from the statistical significance of the difference between the groups as a whole, it is again important to consider the trend shown by the individual values, particularly with regard to those animals with plasma magnesium concentrations below the level of 1.00 mg/100 ml. On the second sampling occasion which was five days after the cows went to grass, 7 out of the 16 individuals in the control group had plasma magnesium values below this critically low level. This represented 44% of the group, whereas only 1 cow in the treated group was just below this figure. Similarly, on the 27th May, there were 6 cows in the control group in this category and one cow still below this level on the last sampling date on 6th June.

In comparison there were 3 cows in the treated group on 27th May with values below 1.00 mg Mg/100 ml, but none were below this level on 3rd June.

There was therefore a significant response to the magnesium treatment as measured by the mean plasma magnesium levels, by the number of animals, below the level of 1.00 mg Mg/100 ml, and possibly by the incidence of cases of hypomagnesaemic tetany. Moreover, this response was obtained despite the high number of pellet rejections which was known to have occurred. It may be that the unexpected increase on the 27th May of the number of treated animals with plasma concentrations below 1.00 mg Mg/100 ml can be explained by the increasing number of pellet rejections by that date. Pellet rejection was undoubtedly a serious problem in this experiment and the result indicated that in future work, it would be essential to increase the density of the pellet to above 2.70g./c.c. by weighting and thereby improve the retention rate.

It can be concluded from this experiment that a treatment of four magnesium rumen pellets probably reduced the incidence of hypomagnesaemic tetany, in this dairy herd. This was achieved not necessarily by eliminating completely the development of hypomagnesaemia, but rather by preventing the appearance of the severely low plasma magnesium concentrations at which tetany is liable to occur.

Experiment No. 12.

Site - Killoch Farm, Barrhead. The beef cattle herd on this farm had suffered the loss of one or two cows from hypomagnesaemic tetany in earlier years.

In the two years prior to 1965, magnesium supplemented cake had successfully been employed to prevent completely the appearance of this condition. There was, however, a problem of losses amongst the beef calves from hypomagnesaemic tetany, and two or three had been lostannually over the previous four years. This was particularly disturbing to the farmer since there were no preventive measures that could be applied to suckled calves.

Experimental Design. A herd of 22 beef cows was made available for this experiment, of which 16 cows were suckling calves of varying age and 6 were due to calve within the following 25 days. The cows had been grazing over the winter on an extensive tract of reasonably good hill grazing, and supplementary concentrate feeding was given daily. Prior to the experiment, these concentrates were supplemented with magnesium oxide but for the duration of this trial, unsupplemented cake (0.29%Mg) was fed at the rate of 4 lbs/head/day.

The herd was divided at random into two groups of 11 cows and on the 26th March, 1965, 4 magnesium rumen pellets were administered to one group while the second group was left untreated as controls. Of the sixteen calves which were available, four were excluded from the trial as being too young, and the remaining twelve calves were divided into two groups at random, six being given two magnesium rumen pellets on the 26th March and six being left untreated as control calves.

The pellets given to the cows were of the same design and formulation as

those used in Experiments 10 and 11. The calf rumen pellets were of the same formulation viz. 85% Mg, 12% Al, 3% Cu but a smaller size of 2" length and 0.9" diameter was used.

Blood samples were taken from all the cattle and calves at the start of the experiment on 26th March, and again on 6th April and 4th May. A further sample was taken from the calves on 18th May.

Results.

Clinical Cases : One calf (R110) in the control group displayed the typical symptoms of hypomagnesaemic tetany on the 4th May, during the time the herd was being blood sampled on that date. The calf died before any remedial treatment could be administered. On the previous sampling date on the 16th April, it had a plasma magnesium value of 0.38 mg/100 ml and a calcium value of 9.51 mg/100 ml.

There were no clinical cases in the treated group of calves, nor were there any cases in either the treated or the control groups of cows.

Plasma Analyses: The individual and mean plasma concentrations of the 22 cows and 12 calves are given in Table 34 together with the mean plasma calcium levels. Only the mean levels are given for plasma calcium, since there were no individuals outside the normal range apart from one control calf, (R.122) which had a plasma calcium concentration of 7.48, 7.05 and 8.38 mg/100 ml on the first three sampling dates respectively.

Table 34. Individual and mean plasma magnesium and mean plasma calcium concentrations of 22 hill cows and 12 suckling calves at grass on Killoch Farm.

(Concentrations in mg/100 ml.)

COWS	TREATED (4 Mg.Pellets)				CONTROL				
Cow-No.	26/3	6/4	16/4	4/5	Cow No.	26/3	6/4	16/4	4/5
MR23	1.86	1.41	1.33	0.86	R114	1.76	1.66	1.52	1.38
R15	1.52	1.37	1.48	1.14	R16	1.67	1.51	1.57	1.67
M123	1.38	0.93	0.71	0.24	2824	1.52	1.07	0.71	0.33
M31	1.86	1.46	1.52	1.19	3500	1.48	1.12	0.81	0.57
ML46	1.33	1.12	1.10	0.57	M122	1.52	1.32	1.19	0.52
R71	1.76	1.76	1.71	2.48	R77	1.52	0.73	1.33	0.81
R118	1.33	0.98	0.57	1.10	R79	1.57	1.46	1.43	0.86
R119	1.52	1.02	1.24	0.48	R117	1.57	1.37	1.67	0.67
R17	1.62	1.46	1.67	1.14	R8	1.81	1.51	1.76	1.33
2951	N.S.	0.98	1.05	0.57	R18	N.S.	1.41	1.19	1.57
R115	1.92	1.27	1.52	1.43	R116	1.52	0.68	0.80	0.33
MEAN	1.61	1.20	1.26	1.02		1.59	1.20	1.27	0.91
MEAN CALCIUM	10.00	9.61	9.68	9.39		9.94	9.43	9.67	9.63

CALVES.	TREATED (2 Mg pellets)					CONTROL				
Calf No.	26/3	6/4	16/4	4/5	18/5	Calf No.	26/3	6/4	16/4	4/5	18/5
R120	1.67	1.27	1.43	1.05	0.67	R121	1.14	1.02	0.81	0.90	1.05
R123	0.76	0.78	0.67	0.81	0.52	R122	0.67	0.39	0.48	0.19	0.24
R124	1.10	1.71	1.62	1.29	N.S.	R125	1.00	0.34	0.33	0.52	1.14
R126	1.33	1.41	1.43	0.05	1.19	R128	0.95	0.58	0.38	0.71	0.29
R127	1.29	1.61	1.57	1.33	1.10	R110	0.81	0.49	0.38	died	staggers
R129	0.81	1.56	1.58	0.71	0.86	R130	1.61	1.61	1.81	1.57	1.14
MEAN	1.16	1.39	1.35	1.02	0.87	MEAN	1.03	0.74	0.70	0.78	0.77
MEAN CALCIUM	10.62	10.96	10.05	10.74	10.44	MEAN CALCIUM	10.53	9.95	10.20	10.43	10.31

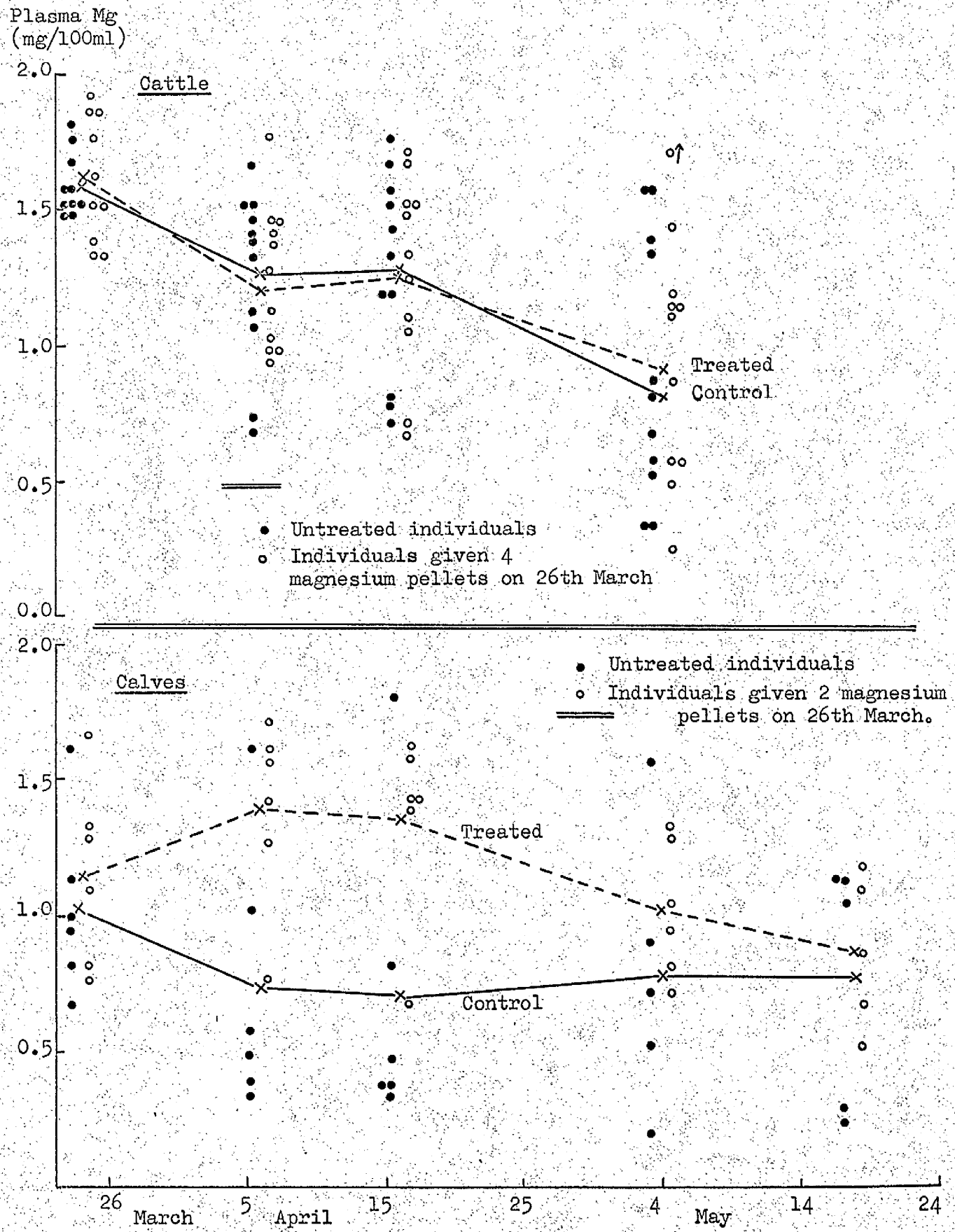
The mean plasma magnesium levels of the cows and calves are also shown graphically on Fig. 34 with the individuals plotted on each date to give a give a visual comparison between the groups.

Bullet Rejection: Over the 39 days of the experiment, 4 pellets regurgitated by cows and one pellet from a calf were found in the field. If five were the maximum number lost, this would then represent a 90% retention of the 56 pellets administered, but under the extensive nature of the hill grazings in this experiment, there would undoubtedly be other regurgitated pellets which were not found. Cow M.123 was slaughtered on the 5th May (=40 days after pellet administration) and no pellet residues were found in the rumeno-reticular sac. All four pellets must therefore have been regurgitated at some earlier date or alternatively have dissolved completely within the 40 days. Complete dissolution would be unlikely since the five pellets which were found on the ground were evenly worn to the degree that would be expected if the release rate was around 1g. Mg/day.

Discussion.

Cows : Since no clinical cases occurred, no comment can be made on any possible effect that the pellet treatment may have had on the incidence of hypomagnesaemic tetany. However, in contrast to the positive results obtained in Experiments 10 and 11, with dairy cattle, there was no evidence in this beef cow herd of any response to a magnesium pellet treatment as measured by plasma magnesium analyses. There was no mean difference in plasma magnesium

Fig. 34 Individual and mean plasma magnesium concentrations of 2 groups of 11 beef cows and 2 groups of 6 suckled calves at grass (Experiment 12).



concentration between the treated and control groups on any sampling date throughout the experiment, despite the existence of severe hypomagnesaemia in this herd (Table 34 and Fig. 34). On the last sampling date in particular, the mean plasma magnesium levels of the treated and control groups were 1.02 and 0.01 mg/100 ml respectively.

Study of the individual results showed that there were approximately equal numbers of animals in each group with values below 1.00 mg/100 ml on each sampling date, and the pellet treatment therefore did nothing to prevent the development of severe hypomagnesaemia. In particular, on 18th May, there were 5 treated cows and 7 control cows below this level of 1.00 mg/100 ml.

It may be that many of the pellets administered were regurgitated within a short time and this could be the explanation for the lack of response demonstrated in this experiment. Evidence on this point was given by the slaughter of cow M. 123 which was one of the individuals in the treated group below the level of 1.00 mg/100 ml on the last sampling date. When slaughtered on the next day, no pellet residues were recovered from the rumen.

It could however be equally said that the possible response found in dairy herds in Experiments 10 and 11, were found despite pellet losses which were probably as high as those which may have been suffered in this case. The explanation of the lack of response may therefore lie in the radical difference between an outwintered beef herd and a dairy herd at grass in the spring. The long standing nature of hypomagnesaemia in beef cattle over the winter may be a more difficult condition to reverse than the temporary hypomagnesaemia suffered by dairy cattle in the spring.

Calves. Positive results were obtained in the suckled calves in response to the two magnesium pellet treatment. The one death from hypomagnesaemic tetany which occurred was in the control group. It was encouraging to note the absence of cases in the treated group, although the actual significance of one death as against none in two groups of only six is difficult to assess.

With regard to the plasma analyses, the response obtained was highly significant. Starting with groups which had similar mean plasma magnesium levels of 1.16 and 1.03 mg/100 ml, there was, over the following 11 days, both a fall in the mean level of the control group and a measurable rise in the mean level of the treated group . (Table 34, Fig. 34). This resulted in almost complete separation of the individual values of the two groups into a "high" and a "low" category, with 5 of the 6 control calves below 1.03 mg/100 ml and 5 of the 6 treated calves above 1.24 mg/100 ml. Statistical examination of these results confirmed that the treated group were significantly ($P = 0.05$) higher than the control group.

On the following sampling occasion, on the 16th May, 21 days after treatment, the difference between the groups had increased and 5 of the 6 control calves were then below 0.82 mg/100 ml and 5 of the 6 treated calves were above 1.37 mg/100 ml. This difference was again significant ($P = 0.05$), but apart from the statistical aspect, stress should be laid on the potential danger of the majority of the control calves being below 0.83 mg/100 ml whereas the majority of the treated animals although still exhibiting a slight degree of hypomagnesaemia, were above the level at which tetany is likely to occur.

On sampling 40 days after treatment, the difference between the groups was decreasing and by the 16th May, there was little difference between the groups in mean plasma magnesium. The difference that did exist between the two groups on these two dates was not statistically significant. This decrease in response to the pellet treatment after 40 days may have been due to either the pellets being almost at the end of their active life, or alternatively have been a reflection of an increasing rejection rate of pellets. The former explanation is the more likely, since examination of the individual values on Fig.34 shows that on the 4th May, the lower mean valuewas due to all the individual values being lower than on the previous date.

The fact that the plasma magnesium values of the control group were falling 40 days after treatment endorsed the finding that the elevated concentrations in the treated group on the 6th and 16th May were attributable to a treatment effect. In other words, when after 40 days, the treatment supply of magnesium diminished or failed, the plasma magnesium levels returned to their pre-treatment levels.

Pellet regurgitation was again evident in these calves in that one calf rumen pellet (out of 12 administered) was found. However, since a highly significant difference was found attributable to pellet treatment over 40 days, the majority of the treated calves must have retained at least one pellet. The one treated calf (No.R123) which showed no response to treatment may have failed to retain either of the pellets.

The Use of Magnesium Rumen Pellets in Sheep at Crossa.

Experiments 13-15.

Experiments 13-15 were designed to test, under practical farming conditions, the ability of two magnesium rumen pellets, to prevent the development of hypomagnesaemia and hypomagnesaemic tetany in sheep. Experiments 13 and 14 were conducted on sites picked intentionally as farms on which outbreaks of hypomagnesaemic tetany had occurred previously. Experiment 15 was conducted on the Glasgow University Veterinary Field Station, using the commercial breeding flock. In all the experiments, the two pellet treatment was given to only half the flock and the remainder left as control animals. As in the work with cattle, the two factors which were looked for in response to the treatment were a decrease in the incidence of hypomagnesaemic tetany if any occurred and secondly an increase in plasma magnesium levels.

The pellets used in all three experiments were weighted with iron shot to give a density of 2.9g./c.c. The iron shot which was distributed evenly throughout the pellet was the larger variety with a diameter of $^{3}/_{32}$" - $\frac{1}{8}$" (as shown on Fig. 30). The alloy formulation was 86%Mg, 12%Al, 2%Cu, and the initial size of the pellet was 1.8" length x 0.75" diameter.

The conclusions from these experiments will be discussed jointly with the cattle rumen pellet experiments at the end of this section.

Experiment 13.

Site : Middleton Farm, Newton Mearns. In the week preceding the start of Experiment 13, there had been 4 deaths from hypomagnesaemic tetany out of a flock of 100 ewes on the farm. Since the owner had been farming this land for only one year, nothing was known about the history of hypomagnesaemic tetany in previous years.

Experimental Design.

The sheep involved in experiment 10 were 59 Blackface ewes with 2-3 week old lambs at foot and were part of the larger flock in which deaths had occurred in the previous week. The 59 ewes were grazing on upland pasture with a small daily ration of concentrate feeding provided. On the 1st May, 1965, 30 ewes were given two magnesium rumen pellets and the remaining 29 were left as a control group. The ewes given pellets were picked at random by treating every second ewe as they came through the sheep pens. No other magnesium supplement was made available to the flock during the course of the experiment.

Blood samples were taken from each ewe at the start of the experiment, and again on the 10th and 18th May.

Results.

Clinical cases : Two sheep in the control group were recorded as hypomagnesaemic tetany cases over the experimental period

(a) sheep no. 420 was reported on the 11th May as displaying the typical symptoms of acute hypomagnesaemic tetany and it responded well to an injection of magnesium sulphate solution. On the following day, however, it again went

down with tetany and death ensued. No blood sample was obtained.

(b) sheep no. 417 displayed definite mild symptoms of hypomagnesaemic tetany on the 10th May. It stood apart from the flock, deserted by its lambs, with its head down and generally lethargic. This diagnosis was confirmed by a blood sample (taken prior to treatment) which had a plasma concentration of 0.67 mg Mg/100 ml and 4.75 mg Ca/100 ml. No clinical cases were recorded in the treated group during the trial. Subsequently on the 17th - 24th June, three deaths occurred from what the farmer believed to be hypomagnesaemic tetany in the group which had been treated 50-55 days previously. This is a longer period than the pellets were expected to last

Plasma Analyses. The individual and mean plasma magnesium concentrations of the 59 ewes on the 3 sampling occasions are given in Table 35. For reasons explained in the discussion, the results are arranged in such a way that any ewe which had a plasma magnesium value below 1.50 mg/100 ml at the start of the experiment, is listed at the top of the table and separate mean levels are given for these particular sheep in both groups. The mean levels are also shown graphically in Fig. 35 with the individual values plotted in to give a visual comparison between the groups. The mean levels of the two groups of sheep in the particular category of having initial values below 1.50 mg Mg/100 ml are given in Fig. 35(b).

Pellet rejection. No pellets were found in the field and no treated ewes died or were slaughtered to recover pellet residues. Thus no information was available on the extent to which the pellets were retained. Previous work described in Experiment 9 with pellets of the same design had given a retention figure of 83%. A similar figure would probably apply in this experiment.

Table 35. Individual and mean plasma magnesium concentrations of 59 ewes at grass at Middleton Farm.

(Concentrations in mg/100 ml)

Treated (2Mg pellets)				Control			
Sheep No.	1/5	10/5	18/5	Sheep No.	1/5	10/5	18/5
265	0.52	1.57	1.24	401	0.48	0.81	1.14
256	0.86	2.00	1.43	403	0.95	1.29	1.33
260	0.85	0.95	1.52	407	0.52	0.76	0.29
269	0.81	-	1.81	408	0.62	1.05	1.05
280	0.62	1.05	1.67	412	0.62	1.00	1.62
258	1.24	1.43	2.00	417	0.57	0.67	0.38
259	1.10	1.29	1.29	422	0.43	0.38	0.24
264	1.43	1.86	1.95	423	0.86	6.90	0.90
267	1.29	0.62	1.29	425	0.71	0.62	0.33
268	1.43	1.81	1.67	402	1.14	1.10	1.33
270	1.29	1.33	1.76	404	1.10	0.95	0.81
271	1.14	1.29	1.57	406	1.29	1.19	2.19
273	1.48	1.71	2.10	209	1.00	1.05	1.10
274	1.38	1.19	1.57	410	1.29	1.43	1.62
278	1.43	1.71	1.52	411	1.05	1.29	1.43
279	1.05	1.00	1.33	413	1.10	0.81	1.14
252	1.29	1.43	1.71	415	1.43	1.52	1.24
				416	1.43	1.19	1.43
Mean of 17 initially below 1.5	1.13	1.39	1.61	418	1.33	0.52	0.38
				419	1.00	1.00	0.95
				420	1.14	1.33	DEAD
				421	1.48	2.00	1.24
				424	1.05	1.29	0.52
				427	1.14	1.81	1.76
251	1.95	2.19	2.05				
253	1.57	1.52	1.57				
254	1.71	2.19	1.90				
255	1.81	1.81	1.86				
257	1.95	1.62	-				
261	1.76	1.52	1.24				
262	1.67	1.90	1.43				
263	1.62	1.48	2.05				
266	1.86	1.86	1.81				
272	1.52	1.48	1.43				
275	1.75	1.95	-				
276	1.71	1.43	1.81				
277	2.48	2.38	-				
				Mean of 24 initially below 1.5	0.99	1.08	1.06
				405	1.57	2.29	1.67
				414	1.62	1.14	1.05
				426	1.76	1.38	1.14
				428	1.62	1.57	0.90
				429	1.76	1.86	1.95
Mean for all 30 sheep.	1.42	1.57	1.65	Mean for all 29 sheep.	1.10	1.18	1.11

Fig. 35a Individual and mean plasma magnesium concentrations of 29 control ewes and 30 treated ewes given 2 magnesium rumen pellets on 1st May.

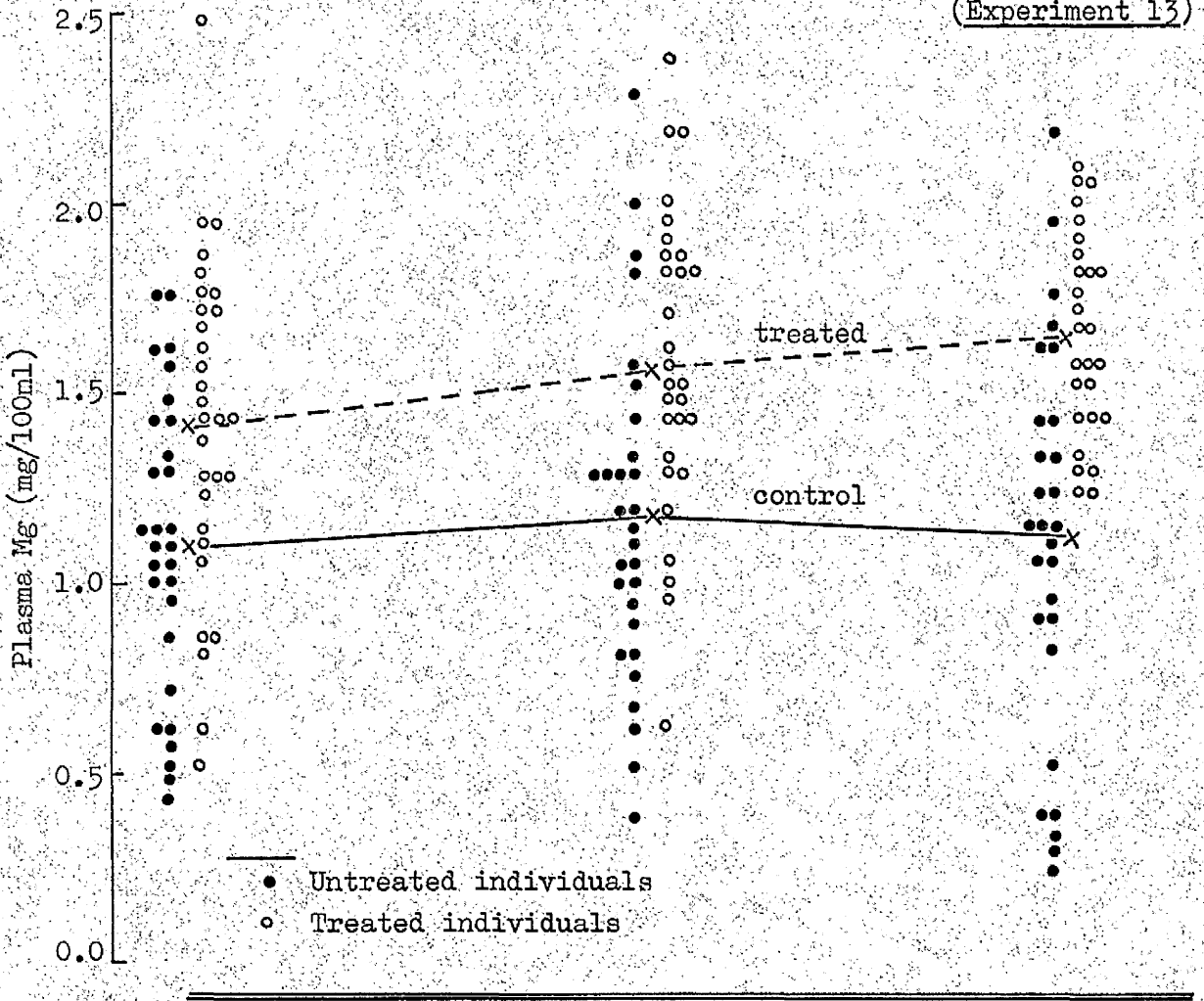
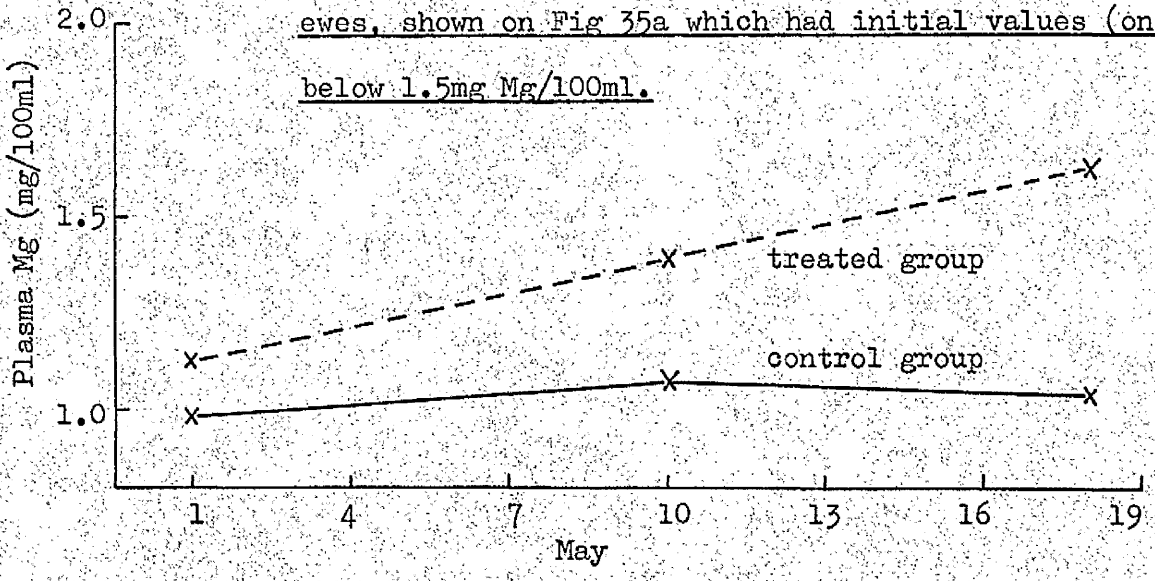


Fig. 35b Mean plasma magnesium concentrations of the 17 treated and 24 control ewes, shown on Fig 35a which had initial values (on 1st May) below 1.5mg Mg/100ml.



Discussion.

The occurrence of two cases of clinical tetany in the control group of 29 sheep represented a 7% incidence of tetany, which was a similar figure to the 4% case incidence which had occurred before the experiment began. It was noteworthy that no cases occurred in the group given the pellet treatment during the actual trial, although the significance of two cases as against none in groups of thirty is not conclusive. The fact that, subsequent to the experimental period, three deaths, believed to be hypomagnesaemic tetany cases, occurred in sheep originally treated with pellets 50-55 days earlier, can be considered as speculative evidence for the efficiency of the pellet treatment. These three potential clinical cases may possibly have received adequate protection with the pellet treatment for 50 days, and it was only after the effect of the treatment had finally disappeared that they once again became susceptible to hypomagnesaemic tetany.

Interpretation of the data on plasma analyses (Table 36) is complicated by two difficulties. Firstly, the wide spread of the individual values on the first sampling date on 1st May (from 0.4-2.46 mg/100 ml), makes statistical analysis difficult because of the large standard errors of the means. Secondly, the fortuitous division of the flock into two groups which had differing pre-treatment plasma magnesium means makes the comparison of the groups on later dates difficult. The control group had an initial mean plasma magnesium level of 1.10 mg/100 ml whereas the treated group was initially at the higher level of 1.42 mg/100 ml. The difference between the groups however increased and by the 18th May, the mean of the treated animals had risen to 1.65 mg Mg/100 ml whereas

the mean for the control group was still 1.11 mg Mg/100 ml. There were also in the treated group a total of five animals with plasma magnesium values below 1.00 mg/100 ml before the treatment was given and none below this figure by the end of the trial. The comparative figures for the group given no treatment were 9 animals below the level at the start and 10 animals below this level at the end of the trial.

An alternative approach to the interpretation of the data is to consider only those animals with initial plasma magnesium values below 1.50 mg/100 ml. This obviates the difficulties by reducing the spread of the individual values and by giving initially similar mean plasma magnesium values for the two groups. It is considered to be justifiable to select the data in this fashion, since only sheep with a degree of hypomagnesaemia (i.e. below 1.5 mg Mg/100 ml) could be expected to exhibit a response to a dietary magnesium supplement. There were 17 treated sheep and 24 control sheep which were in this category of being below 1.5 mg/100 ml and these two groups had reasonably similar mean plasma magnesium levels at the start, of 1.13 and 0.90 mg/100 ml for the 17 treated and 24 control animals respectively.

The mean plasma magnesium level of this treated group of 17 rose continuously over the 18 day experimental period and on the 18th May the mean level was 1.61 mg/100 ml. The control group of 24 animals, on the other hand, did not rise in mean plasma magnesium and on the 18th May, the mean level was 1.06 mg/100 ml (Fig. 35(b) and Table 35).

Considered on this basis, it can be shown statistically that these two groups of 17 and 24 animals were initially of the same population, but after

treatment, there was on both of the subsequent sampling dates a significant difference between the groups (at P = 0.02) on 10th May and P = 0.001 on 18th May). Furthermore, it can also be shown that the mean plasma magnesium levels of the treated group on the 10th and 18th May were significantly higher than the initial pre-treatment level i.e. the magnesium pellet treatment caused a significant increase in plasma magnesium.

It could therefore be concluded that there was a pronounced response in this experiment to a magnesium pellet treatment which effectively raised all treated ewes above the level of 1.20 mg Mg/100 ml within 17 days of treatment administration, whereas 13 control ewes remained below 1.20 mg Mg/100 ml. The response was shown to be statistically significant when ewes with low initial values were considered separately. There was possibly a demonstration of the protective effect of the treatment against tetany in that two cases were recorded in the control group and none in the treated group. Any protection afforded by the pellet treatment had disappeared after 50- days as evidenced by the occurrence on 17-24th June of clinical cases of tetany in three ewes which were originally treated on the 1st May.

Experiment 14.

Site : Broomfield Farm, Stonehouse. This was a lowland farm with a flock of 5-year old cast Blackface ewes grazing on good, lush pasture. Hypomagnesaemic tetany had previously been encountered. The owner was concerned that the particular conditions of having old ewes grazing good heavily fertilised pasture might be conducive to a further outbreak.

Experimental Design.

The experimental design was almost identical to that described for Experiment 13. Two magnesium rumen pellets were administered on 12th April to every second ewe in a flock of 57 Blackface ewes with lambs at foot. This gave a group of 29 treated ewes and one of 28 control ewes. No supplementary food of any kind was being fed. The pellets used were again composed of 86%Mg, 12%Al, 2%Cu alloy mixed with iron shot to give a density of 2.90g./c.c. Three barren ewes, grazing in the same field as the 57 experimental sheep, were each given 2 magnesium pellets of known weight. These ewes were slaughtered at a later date to recover the pellet residues.

Blood samples were taken from all 57 ewes on 12th, 21st and 30th April and on the 7th May.

Results.

Clinical Cases : No hypomagnesaemic tetany occurred in either group.

Blood Analysis : Table 36 details the individual and mean plasma magnesium values of the two groups over the experimental period. The same data is

Table 36. Individual and mean plasma magnesium concentrations of 57 Blackface ewes at grass on Broomfield Farm.

(concentrations in mg/100 ml)

TREATED (2mg pellets)					CONTROL				
Sheep No.	12/4	21/4	30/4	7/5	Sheep No.	12/4	21/4	30/4	7/5
451	1.57	2.00	1.86	2.05	309	1.14	1.02	2.24	1.95
452	1.71	1.41	1.29	1.33	310	1.29	1.22	1.29	0.67
453	1.05	0.88	0.90	0.67	311	1.52	1.56	1.90	1.62
457	1.95	1.71	1.52	1.31	312	2.24	-	1.76	1.71
458	1.24	-	2.05	2.10	313	1.43	1.80	1.57	1.71
459	1.52	1.95	2.10	2.33	314	1.43	1.41	1.14	0.52
460	1.67	2.66	1.90	1.90	315	1.90	1.61	2.19	2.00
461	1.00	2.00	2.00	1.81	316	0.67	1.17	0.76	1.19
462	1.33	-	1.33	0.86	317	1.29	1.17	1.33	0.76
463	2.38	2.44	2.43	2.05	318	1.71	1.46	1.62	1.67
464	1.90	1.90	2.05	1.90	319	0.67	0.29	1.80	1.00
465	1.52	-	2.05	2.10	320	1.71	1.95	2.67	2.10
466	1.76	2.15	2.00	2.24	357	1.95	1.22	1.33	1.81
467	2.05	1.80	1.95	1.90	358	1.05	1.17	0.31	1.38
468	1.67	1.95	2.10	2.33	359	1.24	1.71	1.71	1.38
469	1.14	1.22	1.43	1.33	360	1.80	1.76	1.86	1.90
470	1.52	1.41	2.05	1.86	361	1.24	2.24	1.71	2.19
471	2.19	2.15	2.24	2.05	362	0.67	0.34	0.76	0.57
472	2.24	0.63	1.10	1.33	363	1.57	1.41	1.95	1.76
473	2.19	2.05	2.29	2.00	365	2.29	2.—	-	1.76
474	1.76	2.20	2.43	2.38	754	2.43	0.29	1.05	1.33
475	1.62	0.49	0.95	0.76	755	1.38	1.17	0.86	1.00
476	1.71	1.80	1.95	1.86	756	2.57	1.37	1.38	1.62
477	1.81	1.90	1.95	1.62	757	1.29	-	0.81	0.62
478	1.67	1.61	2.10	1.62	758	1.52	0.93	1.52	1.43
479	2.00	2.00	1.71	1.81	759	2.00	1.66	1.14	1.38
480	2.29	2.05	2.33	2.86	760	2.05	1.61	2.43	3.05
481	1.90	1.95	1.86	2.38	661	2.00	1.90	1.90	1.71
482	2.33	2.34	2.10	1.76					
MEAN	1.75	1.80	1.86	1.83		1.58	1.36	1.54	1.49

repeated graphically in Fig. 36 to give a visual comparison of the results.

Pellet Recovery. No regurgitated pellets were found, but the three barren ewes, given pellets of known weight were slaughtered after 5, 22 and 22 days respectively and two pellets residues were recovered from each. The following release rates were calculated from the initial and final weights:-

	Average magnesium release rate (mg/day).
Ewe no. 1 slaughtered after 5 days	348
	437
Ewe no. 2 slaughtered after 22 days	413
	389
Ewe no. 3 slaughtered after 22 days	522
	480
Average magnesium loss per pellet	437 mg./day.

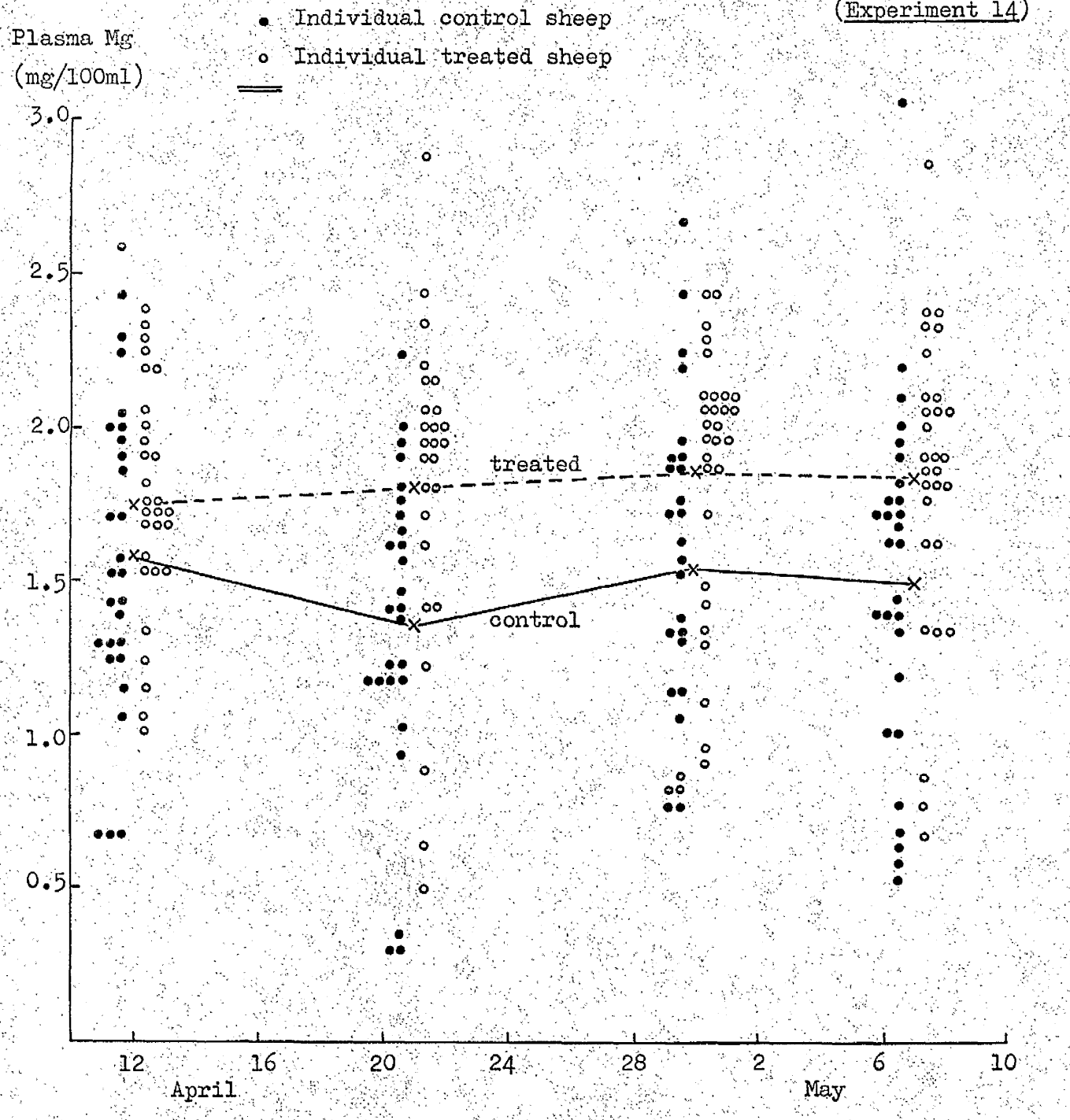
Discussion.

Since no hypomagnesaemic tetany occurred during the course of this experiment, no information was obtained on the efficiency of the pellet treatment as a preventive measure for this condition.

On the basis of the data for the plasma magnesium analyses, a positive response was again recorded as a result of the treatment. The two groups were in this case initially reasonably similar with regard to their mean plasma magnesium levels and to the spread of the individual values. On the 12th April when the treatment was applied the mean levels were 1.58 and 1.75 mg/100 ml for the control and treated groups respectively. On the following sample taken 9 days later, the mean level of the treated animals rose to 1.80 mg Mg/100 ml, whereas the mean level of the control group fell to 1.36 mg/100 ml.

Fig. 36 Individual and mean plasma magnesium concentrations of 28 untreated ewes and 27 ewes given 2 magnesium rumen pellets on 12th April.

(Experiment 14)



It can be seen from Fig. 36 that this difference between the groups was maintained on the following two sampling dates on the 30th April and 7th June. Statistical analysis confirmed that there was no significant difference between the groups immediately prior to treatment, and yet on each sampling occasion thereafter, the treated group were significantly higher in mean plasma magnesium level than the control group. Nine days after treatment the difference was significant at ($P = 0.005$). 18 days and 25 days after treatment the significance level of the difference was $P = 0.02$.

On each sampling occasion after the start of the experiment, there were 8 or 9 animals in the control group which had plasma magnesium values below 1.20 mg/100 ml. Of these 8 or 9 animals, there were 4 or 5 on each date below 1.00 mg Mg/100 ml. In comparison, there were only three treated ewes on each sampling occasion which were below 1.20 mg Mg/100 ml, and also below the level of 1.00 mg Mg/100 ml. The pellet treatment in this experiment therefore markedly reduced the incidence of severe hypomagnesaemia, but it did not eliminate it entirely.

An attempt was made to examine the results from the point of view of those animals with initially low plasma magnesium values, as was done in Experiment 13. In this case, however, the individual values were generally higher than in Experiment 13, and there was an insufficient number of animals with initially low values for them to be considered separately.

The pellets which were recovered from the three slaughtered sheep were corroding evenly, and since the average daily release rates of pellets recovered after 5 and 22 days were similar, it can be assumed that the daily

magnesium release rate was reasonably constant. The rate of wear in the three individuals was also reasonably similar (range 348-522 mg Mg/day). On the basis of the average daily magnesium release rate of 432 mg., the average pellet life in this experiment would be 38 days with a two pellet treatment supplying about 0.8g. magnesium/day over this period. The release rate found was slightly higher and the pellet life consequently slightly shorter than was anticipated from earlier experimental trials with this type of pellet.

It could be concluded from this experiment that a significant increase was found in plasma magnesium levels of lactating ewes in response to the two pellet treatment, although in this case the incidence of severe hypomagnesaemia was not entirely eliminated.

Experiment 15.

Site : Glasgow University Veterinary Field Station, Cochno Farm, Duntocher.

Although hypomagnesaemic tetany has seldom been reported on this farm in previous years, earlier experiments (Section II of this thesis) have frequently found that severe hypomagnesaemia occurs in ewe flocks grazing thereon. It was consequently considered to be a good experimental site for a magnesium pellet treatment.

Experimental Design.

The experimental design was identical to that employed in Experiments 13 and 14. A flock of Cheviot ewes were divided at random on the 27th April into a control group of 31 ewes and a group of 30 ewes which were treated with 2 magnesium rumen pellets. The flock was grazing on good fertilised pasture and no other supplementary food was provided. The pellets used were of the same design as in Experiments 13 and 14 viz. of alloy formulation 86%Mg, 12%Al, 2%Cu and mixed with iron shot to give a density of 2.9g./c.c.

Blood samples were taken from all the ewes on the 27th April and the 8th and 14th of May.

Results.

Clinical : No hypomagnesaemic tetany occurred in either group.

Plasma Analysis. The individual and mean plasma magnesium values of all the ewes on each sampling date are given in Table 37.

Table 37. Individual and mean plasma magnesium values of 61 Cheviot ewes at grass at Cochno Farm.

(concentrations in mg/100 ml.)

TREATED (2 Mg. pellets).					CONTROL				
Ewe No.	27/4	3/5	8/5	14/5	Ewe No.	27/4	3/5	8/5	14/5
373	0.86	1.10	0.90	1.00	539	0.81	0.90	0.86	1.14
376	0.81	0.95	0.90	1.33	541	0.38	0.52	0.52	0.86
388	0.62	0.95	1.00	1.33	543	0.67	0.96	0.95	0.71
389	0.95	0.86	0.71	0.76	545	0.86	0.92	0.90	1.05
392	0.81	1.14	1.19	1.24	546	0.38	0.72	1.14	1.29
383	0.71	1.14	1.33	1.19	547	0.76	0.67	0.67	0.57
377	1.38	1.62	1.67	2.00	549	0.38	0.43	0.38	-
378	1.10	1.00	0.95	0.90	550	0.52	0.90	0.71	0.57
381	1.38	1.33	1.43	1.43	551	0.86	0.71	1.14	1.00
384	1.19	1.05	0.90	0.67	553	0.71	1.14	0.95	0.95
372	1.10	1.38	1.43	1.62	554	0.71	0.86	0.57	0.62
386	1.14	1.33	0.86	0.90	558	0.86	0.76	0.76	0.52
390	1.05	1.05	1.24	1.05	560	0.81	0.52	1.19	1.00
391	1.38	1.48	1.38	1.76	561	0.95	0.76	1.19	1.14
393	1.38	1.00	1.24	1.48	562	0.76	0.81	0.90	1.43
379	1.24	1.38	1.71	1.62	548	0.95	0.95	1.52	0.71
364	1.29	1.24	1.43	1.48	540	1.38	1.19	1.05	0.81
365	1.48	1.00	1.05	1.14	542	1.00	0.62	1.10	1.14
367	1.38	1.67	1.48	1.81	544	1.10	1.24	1.19	1.52
368	1.33	1.57	1.14	1.43	556	1.14	0.95	1.19	1.29
369	1.38	0.95	1.14	1.52	557	1.19	1.33	1.48	1.57
366	0.95	1.81	1.67	1.90	559	1.29	1.43	1.38	1.76
380	0.81	0.81	0.95	1.52	534	1.48	1.57	1.33	1.90
					535	1.38	1.33	1.43	1.81
					536	1.24	1.19	-	1.48
					555	1.19	1.05	0.95	1.71
Mean of those * below1.5	1.12	1.21	1.20	1.35	Mean of those * below1.5	0.91	0.94	1.02	1.14
371	1.90	1.62	1.52	1.90					
374	1.57	1.81	1.43	1.71					
375	1.57	1.62	1.81	1.71	552	2.24	1.62	1.67	1.57
382	1.71	1.33	1.81	1.57	532	1.62	1.62	1.86	1.95
370	1.71	1.86	2.05	2.24	533	1.76	1.57	0.95	2.10
385	1.62	1.29	1.38	1.90	537	1.76	1.62	1.71	1.71
387	1.86	1.90	1.86	2.00	538	1.81	1.57	1.62	1.86
Total Mean	1.25	1.31	1.32	1.47	Total Mean	1.06	1.05	1.11	1.26

* below 1.50 mg Mg/100 ml on the 27/4.

Any individual sheep which had an initial plasma magnesium value below 1.50 mg/100 ml has been entered at the top of Table 37 and the mean levels of the 23 treated ewes and the 26 control ewes which fall into this category are also shown in the Table. The individual and mean plasma magnesium levels of these two groups of 30 treated and 31 untreated ewes are also shown graphically on Fig. 37.

Pellet Rejections. No regurgitated pellets were found on the pasture, nor were any sheep slaughtered to recover the pellet residues. There was therefore no information on pellet wear or retention.

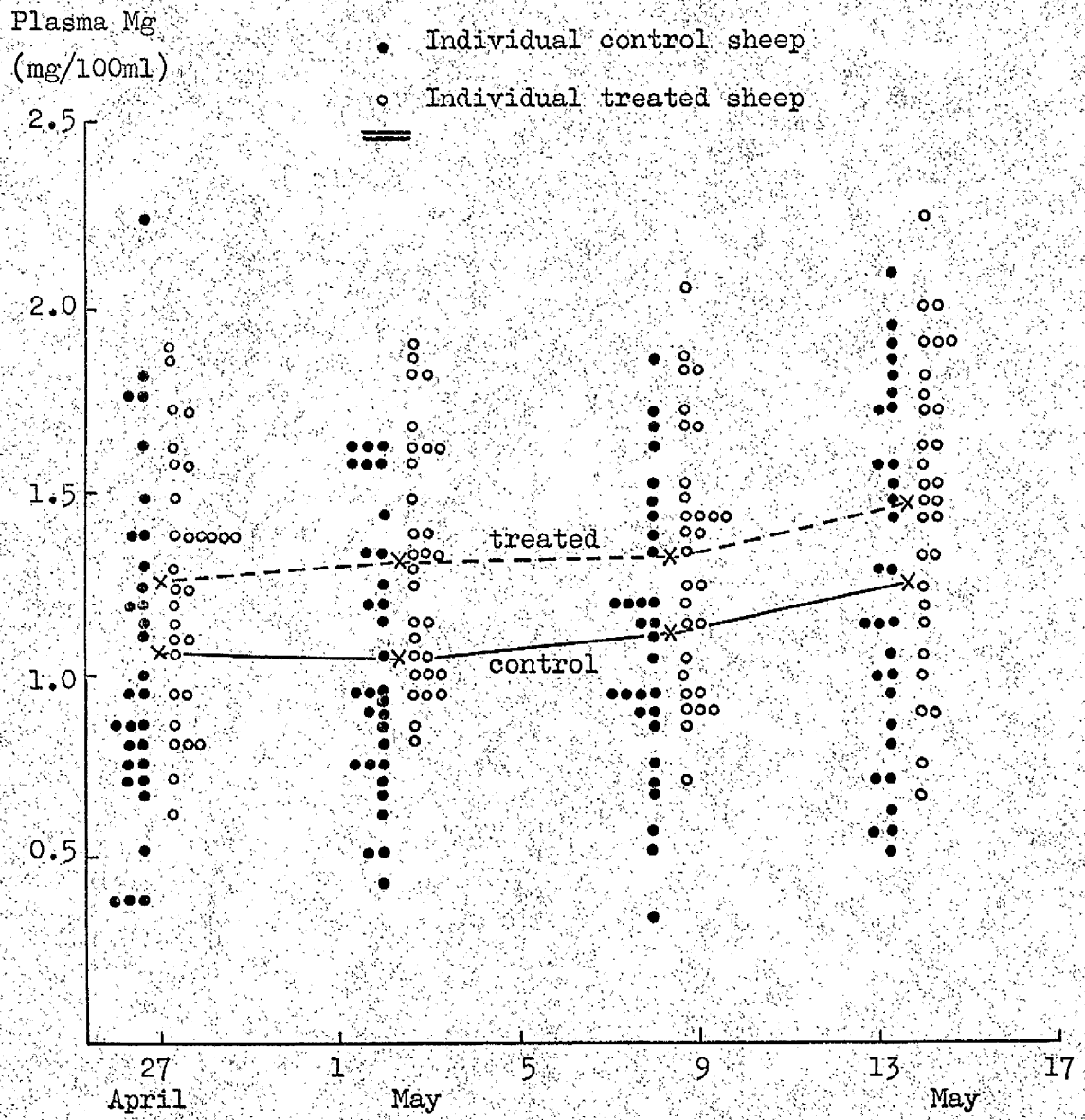
Results.

By chance, the groups, arranged at random, were seriously unequal in the numbers of animals which initially had low plasma magnesium values. The control group had 16 ewes with initial values below 1.0 mg/100 ml, whereas there were only 8 such animals in the group given treatment. This disparity is reflected in the differing initial mean levels of the two groups (Table 37), the mean of the control group being 1.06 mg/100 ml and the group given treatment being 1.25 mg/100 ml. Thus although the treated group maintained a higher mean plasma magnesium level over the 17 day experimental period than did the control group, it is difficult to say to what extent this was due to treatment and how much was due to the pretreatment difference.

In an attempt to equalise the groups and produce more comparable data, those 23 treated and 26 control ewes with initial values below 1.50 mg/100 ml were considered separately as shown in the top half of Table 37. However

Fig. 37 Individual and mean plasma magnesium concentrations of 31 untreated ewes and 30 ewes given 2 magnesium rumen pellets on 27th April.

(Experiment 15)



even on this basis, the results are difficult to interpret since an initial pretreatment difference of 0.21 mg/100 ml still existed between the two groups. A statistically significant difference can be demonstrated to exist between the treated and control groups on samples taken 7 days after treatment, but it can also be shown that there was a statistical difference between the groups before the treatment was given.

However, it is probably doubtful if there was much response to pellet treatment in this experiment, since the mean plasma magnesium concentrations of the treated group was at a sufficiently low level for there to be considerable scope for improvement in the mean. This improvement, had it occurred should have been demonstrable despite the disparity between the groups. The only feature which is suggestive of any treatment effect is that a visual appraisal of the scatter of the individual values plotted on Fig. 37 might indicate that on 3rd May, there was an upward shift of the individuals in the treated group and no such change in the control group. On the other hand the numbers of animals in each group with severe hypomagnesaemia on each date remains relatively constant:-

ewes with a plasma magnesium value below 1.00 mg/100 ml.	27th April	3rd May	8th May	14th May
treated group-:	8	5	7	4
control group-:	16	17	13	9

The conclusion from Experiment 15 must therefore be that there was little evidence of any response in the plasma magnesium concentration of sheep attributable to a magnesium rumen pellet treatment, and the incidence of severe hypomagnesaemia was not eliminated in treated animals. The interpretation of the results of the experiment was, however, marred by the fortituous presence of pretreatment differences in the plasma magnesium concentrations of the two groups.

Conclusions to be drawn from Experiments 10-15.

Over these six experiments, a magnesium rumen pellet treatment was given to 182 animals which included, in the various trials, dairy cattle, beef cows, suckled calves and sheep. A similar total of 181 animals were left with no treatment as control groups in each experiment and as far as was possible, the groups were comparable in all other respects. In no one experiment was there a sufficiently high incidence of hypomagnesaemic tetany in a control group to give positive proof of the ability of a magnesium rumen pellet to prevent the development of this condition. Where only one or two cases develop in a trial of this nature, pure chance can decide in which of two groups these one or two cases may occur. When, however, the same result is found consistently in several experiments, the possibility of the result being entirely due to chance is diminished. The incidence of tetany in these six experiments is collated in Table 38.

Table 38. The incidence of hypomagnesaemic tetany in six experiments on magnesium rumen pellet treatment in cattle and sheep.

Experiment No.	Experimental animals.	Control Group. No. of cases.		Treated Group. No. of cases.
		definite.	doubtful.	
10	2 groups of 45 dairy cows.	1	1	—
11	2 groups of 31 dairy cows.	2	1	—
12	2 groups of 11 beef cows.	-	-	—
12	2 groups of 6 suckled calves.	1	-	—
13	2 groups of 30 sheep.	2	-	—
14	2 groups of 29 sheep.	-	-	—
15	2 groups of 30 sheep.	-	-	—
Total for 2 groups of 182 animals		6	2	—

There was, therefore, as shown in Table 38, a total of 6 definite and two doubtful cases of hypomagnesaemic tetany in the 181 untreated animals concerned in the six experiments. In contrast, there was none in the comparable 182 animals which were given a magnesium pellet treatment. This was a very encouraging result and one which should be approaching the point where it would be very unlikely to be purely due to chance.

The other factor which was examined in each of the Experiments 10-15 was the response of the animal to pellet treatment as measured by the plasma magnesium concentrations of the treated and control groups.

A statistically significant response to treatment was demonstrated in the two experiments involving dairy cows, in the one experiment involving suckled calves, and in two of the three experiments concerned with lactating ewes. No measurable response to the treatment could be found in the one experiment in a beef cow herd, nor in one of the three trials with sheep. In total therefore, a significant response to treatment was demonstrated in 5 out of 7 trials.

Quite apart from consideration of the mean plasma magnesium levels, the plasma analytical data were also examined in each experiment from the point of view of the number of animals in each group with severe hypomagnesaemia i.e. with plasma magnesium concentrations below 1.00 mg/100 ml. This was felt to be an important aspect of the results since for any proposed prophylactic treatment for hypomagnesaemic tetany to be effective, it should be capable of preventing the development of severe hypomagnesaemia. To collate the results of Experiments 10-15 with regard to the incidence of severe hypomagnesaemia,

Table 39 details the number of treated and control animals which fell into this category of having a plasma magnesium value below 1.00 mg/100 ml on each sampling date after treatment.

Table 39. Animals in Experiments 10-15 which had plasma magnesium values below 1.00 mg/100 ml.

Expt. No.	experimental animals.	No. of days after treatment.	Control Group.	Treated Group.
10	2 gps. of 10 dairy cows	14	5	0
		24	3	0
		35	1	0
11	2 gps. of 16 dairy cows	7	7	1
		14	6	3
		21	1	0
12	2 gps. of 11 beef cows	11	2	3
		21	3	2
		39	7	5
12	2 gps. of 6 suckled calves	11	4	1
		21	5	1
		39	5	3
		53	3	3
13	2 gps. of 30 ewes	9	9	2
		17	10	0
14	2 gps. of 30 ewes	9	4	3
		18	5	2
		25	5	3
15	2 gps. of 30 ewes	6	17	5
		13	13	7
		19	9	4
	TOTAL		124 control	48 treated.

Total No. of samples taken = 375 from each group.

∴ % of samples below 1.00 mg Mg/100 ml out of total of 375 samples-: 33.0% 12.8%

From Table 89 it is obvious that there was a high degree of severe hypomagnesaemia in animals given no treatment. There was a total of 124 plasma magnesium samples taken which had a magnesium concentration below 1.00 mg/100 ml. This represents 33.0% of the total 375 samples which were taken from untreated animals. In contrast, there were 48 samples taken from treated animals which were in this category. This represents 12.8% of the total 375 samples taken from animals given a pellet treatment.

Thus the incidence of severe hypomagnesaemia was markedly reduced but it was not eliminated by the rumen pellet treatment. These figures are based however on all the data collected, and in some particular experiments, the comparative figures were better than these overall figures. For example in Experiment 10 the comparative incidences of severe hypomagnesaemia in the untreated and treated groups were 30% and 0% respectively. One factor which will adversely effect the efficiency of pellet treatment as demonstrated in this way, is the extent to which pellets may have been regurgitated in some of these trials. This is particularly so for the experiments dealing with cattle where unweighted pellets were used. In Experiment 11, it was known that at least 24% of the pellets administered were rejected.

It is now known that the retention rate of rumen pellets in cattle can be greatly improved (probably to 100% retention) by increasing the density of the pellet to 2.7 g./c.c. In the case of sheep, the pellets used in these experiments were weighted to give an overall density of 2.90g./c.c. Earlier work (Experiment 9) showed that these weighted pellets are still rejected to a certain degree but that the retention rate was in the order of 83%. Furthermore

the work of Hemingway et al. (1961) where magnesium oxide pellets of density 4.0g./c.c. were used, suggested that further increases in density to the level of 4.0g./c.c. would not improve the retention rate to any great extent. It would therefore seem that a limited amount of pellet regurgitation in sheep is inevitable.

To summarise the results of Experiments 10-15, a prophylactic treatment with metal magnesium rumen pellets given to a total of 182 animals at grass led to:-

(a) the elimination of outbreaks of hypomagnesaemic tetany in treated animals, whereas in 181 comparable control animals there were six confirmed cases of hypomagnesaemic tetany.

(b) the demonstration of statistically significant responses in the mean plasma magnesium levels of the treated animals in five of the seven trials carried out.

(c) a reduction in the development of severe hypomagnesaemia (levels below 1.00 mg Mg/100 ml) in the treated animals. Out of 375 samples taken from treated animals, 12.8% were below 1.00 mg/100 ml, whereas the comparative figure for 375 samples taken from control animals was 33.0%.

(d) these results were obtained despite a certain degree of pellet rejection which was known to have occurred.

These results would therefore suggest that a magnesium rumen pellet treatment may have a definite prophylactic effect for hypomagnesaemic tetany in both cattle and sheep. Although the results demonstrate that the appearance of severe hypomagnesaemia was not entirely eliminated, it was felt that the complete

absence of hypomagnesaemic tetany in treated animals was a particularly encouraging finding. It must also be remembered that the present methods of prophylaxis for hypomagnesaemic tetany are not 100% successful. In particular the survey carried out by Seekles & Boogaerdt (1959) with over 11,000 dairy cows, showed that the incidence of tetany was reduced to 0.8% by feeding 2 ozs. magnesium oxide/day. In comparison, there was an incidence of 2.8% hypomagnesaemic tetany in cows not receiving this supplement. Thus, although the incidence was reduced by over two-thirds, it was by no means eliminated by the daily feeding of this large quantity of 30g.Mg.

Seen in comparison with this type of result from the use of conventional prophylactic methods, the results of these preliminary experiments with magnesium rumen pellets are relatively satisfactory. More conclusive evidence on the effectiveness of such a pellet treatment would have to be obtained before it would be possible to suggest its adoption as a prophylactic measure in practice. These preliminary results, however, should be sufficiently encouraging to merit further trials being carried out on a more extensive basis to investigate the potentialities of this treatment.

The final result may in fact be that 100% protection against hypomagnesaemic tetany is an impossible target to aim for with any form of treatment. It may, on the other hand, be that a rumen pellet treatment, though not 100% effective, could be as good as any of the present conventional methods for prophylaxis. It would however have the definite advantages of being in many cases an easier and more certain form of treatment to administer, and of being in other circumstances a possible form of treatment for animals which at present

are inaccessible for the application of the present methods of prophylaxis against hypomagnesaemic tetany.

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Appendix I

Chemical analysis - laboratory procedures.

Blood plasma - calcium and magnesium.

Reagents.

1. E.D.T.A. solution. 0.760g 'Analar' disodium salt of ethylene-diamine-tetra-acetic acid, dissolved in one litre of distilled water.
2. Murexide indicator. Approximately 0.004g ammonium purpurate dissolved in 200 ml distilled water. Solution in 20 ml cuvette should read 80 on galvanometer scale of E.E.L. Titrator where set at 0 with water.
3. Eriochrome black T Indicator.

   Stock solution. Dissolve 0.4g in 100 ml methyl alcohol. Add 4.0 ml 'analar' conc. ammonia solution.

   Solution used. Add 4 ml of stock solution plus 2 ml ethanolamine to 200 ml distilled water.
4. $^{N}/_{10}$ NaOH solution. Prepare 500 ml fresh by dilution of stock solution of 2N sodium hydroxide.

Procedure.

Pipette 1 ml serum into each of two 20 ml cuvettes.

Add 6 ml NaOH solution and 3 ml murexide indicator to one cuvette.

Add 6 ml boiling distilled water and 3 ml eriochrome indicator to the second cuvette.

Titrate each in the E.E.L. titrator unit with light filters 606 and 607 respectively against E.D.T.A. solution.

The end point is indicated by the galvanometer reading reaching a maximum constant reading.

Blank titrations are run with each indicator using the reagent solutions alone.

Results. Calcium:- The murexide titration minus the blank value x 8.183 = mg Ca/100 ml

Magnesium:- Difference of two titrations minus respective blank x 0.4965 = mg Mg/100 ml.

A standard solution of 10 mg Ca/100 ml and 2 mg Mg/100 ml is normally included as a check.

Plant Material.

Preparation of plant ash solution.

Dry ash. One gram of dried ground grass in a silica crucible at 550°C. Dissolve the resulting ash in dilute hydrochloric acid, evaporate to dryness, redissolve in distilled water and transfer to a 100 ml flask. Make up to the mark.

Plant Magnesium. (Cornfield & Pollard 1950)

Reagents. 1. Compensating solution. 0.96g potassium dihydrogen phosphate 0.88g potassium aluminium sulphate, 13.9g calcium chloride and 2 ml concentrated hydrochloric acid, dissolved in one litre of distilled water.

2. Mannitol solution. 2.5% aqueous solution.

3. Titan yellow solution. 0.05% aqueous solution.

4. 3N Sodium hydroxide solution. 120g 'analar' sodium hydroxide dissolved in distilled water and made up to 1,000 cc.

Procedure.

Pipette an aliquot (generally 2 ml) of plant ash solution into a 25-30 test tube.

Add 1 ml of compensating solution and 1 ml of mannitol solution and make up to 20 ml volume.

Add 2 ml Titan yellow solution and 2 ml of 3N sodium hydroxide

Allow to stand for 10 minutes after shaking well.

Shake to disperse the lake and read immediately in an E.E.L. colorimeter with a 625 green filter (set at zero with distilled water).

Calibrate the procedure by using aliquots of a magnesium standard solution containing 0 - 0.10 mg. magnesium to give a standard graph.

Plant Calcium. (Hemingway 1956)

Cation exchange resin columns. A 5 cms column of Amberlite IR - 120(H) resin, suitably ground is packed in a 35 cm x 7 m.m. tapered open glass tube, activated by washing with 5N nitric acid followed by several washings with distilled water.

Procedure.

Transfer a suitable aliquot (generally 5 ml) of the plant ash solution to the column, and allow to drain. After washing the column through several times with distilled water, the cations are eluted off the column into a 10 ml standard flask

by 5N nitric acid. Make this solution to the mark with 5N nitric acid.

Measure the calcium concentration in this solution in the E.E.L. flame photometer which is set at 0 with 5N nitric acid and at 100 with a solution of 75 p.p.m. calcium, using the calcium filter.

Plant sodium. Dilute 1 ml of the plant ash solution to 10 ml with distilled water, and using the sodium filter, measure the sodium concentration in the E.E.L. flame photometer, set at 0 with distilled water and at 100 with a standard solution containing 10 p.p.m. sodium.

Plant potassium. Dilute 1 ml of the plant ash solution to 100 ml with distilled water and using the potassium filter, measure the potassium concentration in the E.E.L. flame photometer, set at 0 with distilled water and at 100 with a standard solution containing 10 p.p.m. potassium.

Plant phosphorus.

Reagents.

Ammonium molybdate solution. Add 300 ml of dilute sulphuric acid to 100 ml of a 10% solution of ammonium molybdate.

Stannous chloride solution. Dissolve 2.5g stannous chloride (stored in refrigerator) in 27.5 ml concentrated hydrochloric acid. Dilute to 100 ml with distilled water.

Procedure. Dilute 1 ml of the plant ash solution to approximately 70 ml

in a 100 ml standard flask. Add 2 cc of ammonium molybdate solution and 5 drops of stannous chloride solution, and make up to 100 cc. Read after 15 minutes, in the E.E.L. colorimeter with filter ORI, set at zero with distilled water. Construct a standard graph using aliquots of a standard phosphate solution, containing 0 - 0.1 mg. phosphorus.

Plant Manganese. Pipette 50 ml of plant ash solution into an evaporating dish, add 5 cc conc. nitric acid, and evaporate to dryness (to remove chlorides).

Dissolve residue in 50 cc dist. H_2O

Add 2cc phosphoric acid and about 0.2g potassium periodate.

Boil for 5-10 minutes to develop pink colour.

After cooling, make up to 100 ml.

Read in the colorimeter set at 0 with water using filter 624.

Construct a standard graph by using aliquots of a standard manganese solution containing 0.0 - 0.10 mg. manganese.

Appendix 2a. Date of Lambing and number of lambs of Cheviot ewes Spring, 1961

Ewes with twins		Ewes with singles			
Ewe No	Date of Lambing	Ewe No	Date of Lambing	Ewe No	Date of Lambing
55	22/4	75	22/4	15	19/4
70	22/4	74	27/4	22	13/4
50	13/4	81	22/4	7	6/4
89	19/4	52	19/4	3	19/4
57	19/4	79	22/4	37	6/4
69	13/4	87	24/4	5	6/4
71	19/4	51	24/4	10	23/4
65	19/4	58	22/4	4	24/4
80	24/4	76	19/4	24	6/4
53	13/4	88	28/4	18	24/4
12	6/4	85	19/4	19	22/4
36	22/4	86	19/4	32	6/4
33	13/4	72	2/5	27	6/4
2	19/4	90	19/4	13	6/4
14	6/5	59	19/4	17	19/4
35	24/4	63	22/4	6	6/4
9	13/4	62	22/4	30	6/4
16	13/4	1	28/4	82	19/4
20	6/4	29	22/4	92	29/3
31	13/4	21	22/4	84	23/4
25	24/4	40	6/4	83	19/4
23	13/4	28	1/5	73	25/4
39	19/4	60	23/4	68	1/5
11	22/4			64	19/4
93	28/3				

Appendix 2b. Plasma calcium and magnesium values of 72 Cheviot ewes over the preliminary experimental period 21st February to 19th April, 1961.

Ewe No.	Plasma magnesium (mg/100 ml)							Plasma calcium (mg/100 ml)						
	21/2	3/3	14/3	22/3	29/3	6/4	13/4	21/2	3/3	14/3	22/3	29/3	6/4	13/4
1	1.56	1.35	1.24	1.32	1.45	1.44	1.43	9.00	8.80	9.90	9.05	9.80	9.80	9.85
2	1.25	1.50	1.23	1.41	1.08	1.30	1.67	8.55	8.47	9.90	10.20	9.70	9.70	9.50
3	1.35	0.91	0.72	1.04	1.03	0.95	0.86	9.90	10.80	10.65	10.80	10.40	10.80	10.10
4	2.06	1.83	1.95	1.94	1.91	2.00	2.50	9.35	8.80	9.35	9.85	9.35	10. 10	10.00
5	1.66	1.59	2.01	1.78	1.48	1.65	1.88	9.70	9.42	10.50	9.85	9.60	10.35	9.90
6	1.04	1.31	1.50	1.63	1.63	1.70	1.80	9.00	9.42	10.10	9.35	9.60	10.00	8.25
7	2.40	2.31	2.19	2.00	2.05	1.60	1.70	8.00	9.05	9.90	9.20	10.80	10.50	10.00
9	2.15	1.89	1.95	2.05	1.82	1.87	2.00	9.50	9.50	9.90	9.50	9.30	9.90	9.50
10	1.95	2.11	1.84	2.10	1.89	2.20	2.43	9.35	9.70	9.70	9.05	9.85	9.30	9.85
11	1.65	1.59	1.40	1.81	1.64	2.00	2.13	8.90	10.20	10.30	9.85	10.65	9.85	9.20
12	1.56	1.36	1.24	1.69	1.34	1.40	1.56	9.05	9.42	9.90	9.60	9.40	9.60	10.40
13	1.20	1.19	1.16	1.34	1.57	1.19	1.46	8.90	9.50	9.90	10.00	10.40	9.50	9.20
14	1.71	1.31	1.28	1.48	1.43	1.41	2.00	9.10	8.97	10.20	9.35	9.35	9.40	9.80
15	2.18	2.01	2.23	2.30	2.18	2.50	2.58	10.30	9.92	10.35	9.60	10.85	9.20	8.55
16	1.55	1.37	1.54	1.81	1.66	1.50	1.22	9.90	9.42	10.10	9.85	9.05	9.30	9.80
17	1.22	1.43	1.34	1.30	1.30	1.19	1.44	8.50	9.70	10.40	10.10	9.50	10.65	10.00
18	1.70	1.57	1.69	1.43	1.42	1.41	1.90	8.60	8.55	9.40	9.20	9.05	9.30	7.90
19	1.45	1.48	1.54	1.85	1.77	1.68	1.55	9.70	9.60	10.65	10.20	10.10	9.70	10.65
20	1.80	1.96	2.17	2.30	2.30	2.25	2.20	8.55	8.10	9.20	7.90	7.75	7.50	7.25
21	1.75	2.11	1.89	1.95	1.56	1.95	2.00	10.40	9.90	10.10	10.00	10.10	10.30	8.10
22	1.73	1.79	1.28	1.85	1.56	1.46	1.45	9.70	10.40	10.65	10.10	10.40	10.90	10.70
23	2.05	1.94	1.86	1.91	1.90	1.90	1.78	8.10	9.20	9.10	9.10	8.55	8.60	9.85
24	2.19	2.00	1.77	2.10	2.20	2.00	2.10	9.30	9.60	10.65	9.35	9.90	10.80	9.90
25	1.75	1.60	1.50	2.12	2.33	2.40	2.50	9.80	9.40	10.30	9.70	9.40	8.55	8.10
27	2.03	2.23	2.14	2.35	2.15	2.30	2.27	9.85	9.90	10.20	9.70	10.20	9.85	9.85
28	1.35	1.20	1.54	1.48	1.80	1.80	1.91	9.60	9.35	10.40	9.85	10.10	9.50	9.70
29	1.30	1.26	1.88	1.24	1.41	1.10	1.71	7.75	9.05	10.50	10.00	10.20	10.60	10.35
30	2.00	1.70	1.74	1.97	2.00	2.00	2.00	7.80	9.30	10.00	10.10	7.80	8.50	9.35
31	1.40	1.40	1.34	1.47	1.12	1.63	1.68	8.60	8.90	9.30	9.10	9.35	9.10	8.70
32	1.04	0.98	0.94	1.09	1.04	0.90	0.89	9.50	9.85	10.50	10.00	9.90	9.60	9.35
33	1.70	1.45	1.39	1.40	1.58	1.60	2.54	9.05	10.10	10.80	10.10	10.50	10.60	10.35
35	1.34	1.47	1.05	1.17	1.22	1.40	1.80	8.25	8.25	10.00	9.50	9.50	10.10	9.50
36	2.16	1.87	1.55	1.65	1.83	2.15	2.95	9.85	9.50	10.00	10.35	10.20	10.30	9.00
37	2.10	1.70	1.84	2.20	1.81	1.49	2.18	9.00	9.30	10.00	9.50	9.50	10.90	9.70
39	2.20	2.38	2.15	2.10	2.34	2.16	2.03	9.85	9.60	9.90	9.20	9.40	9.70	9.70
40	2.12	2.03	1.57	2.04	1.73	1.67	1.86	9.90	10.10	10.65	9.90	10.40	10 .40	10.50
Mean Young ewes	1.71	1.64	1.60	1.74	1.69	1.70	1.89	9.17	9.42	10.09	9.67	9.72	9.80	9.52

Appendix 2b. Plasma calcium and magnesium values of 72 Cheviot ewes over the preliminary experimental period 21st February to 19th April 1961

Ewe No	Plasma magnesium (mg/100 ml)							Plasma Calcium (mg/100 ml)						
	21/2	3/3	14/3	22/3	29/3	6/4	13/4	21/2	3/3	14/3	22/3	29/3	6/4	13/4
51	1.80	2.00	1.71	2.01	1.66	21.7	2.05	7.50	6.55	9.50	8.80	7.90	8.00	7.50
52	2.22	2.23	1.55	2.40	2.20	2.25	3.12	9.90	9.70	10.50	10.00	9.60	9.00	8.70
53	1.85	1.75	1.79	1.83	1.58	2.00	1.99	8.30	8.10	8.90	8.40	8.90	9.50	9.05
55	1.85	1.82	1.84	1.37	1.46	1.49	1.73	9.60	8.55	8.70	9.20	9.30	9.60	9.10
56	1.58	1.55	1.56	1.84	2.00	2.10	2.20	8.10	8.90	9.40	9.70	9.70	9.70	8.10
57	1.43	1.46	1.59	1.53	1.23	1.58	1.50	7.50	7.75	9.20	8.90	9.05	9.35	7.90
58	1.60	1.31	1.37	1.90	1.66	1.82	2.14	9.20	9.12	9.85	8.60	9.40	9.60	9.60
59	1.70	1.54	1.39	1.69	1.87	2.42	2.72	9.40	9.20	9.80	9.85	9.35	8.50	9.10
60	1.00	0.98	0.87	1.90	1.49	2.30	2.50	7.80	8.10	8.55	7.15	7.75	9.00	8.52
62	2.06	1.96	1.38	1.95	1.82	1.90	2.50	8.25	9.05	9.85	10.20	9.85	10.20	9.35
63	2.20	2.20	2.06	1.82	1.44	1.54	1.42	9.10	9.42	9.70	9.85	10.30	9.85	10.20
64	1.70	1.59	1.92	2.25	1.66	2.00	1.72	7.25	7.00	8.90	8.55	8.55	8.90	8.40
65	2.25	2.68	2.32	2.26	2.28	1.74	2.50	8.55	8.62	9.20	9.30	10.20	9.50	9.90
68	1.85	2.09	2.13	1.96	1.88	2.00	2.10	7.90	8.70	8.25	8.80	9.05	9.35	8.25
69	2.14	2.57	2.13	2.40	1.74	2.40	2.61	8.80	8.50	9.30	7.80	8.70	8.30	8.90
70	1.75	1.78	1.74	1.61	1.61	1.51	2.72	8.10	9.00	8.50	8.40	8.30	9.40	7.80
71	2.00	1.75	1.65	2.10	1.49	1.90	2.46	8.50	8.35	9.80	8.25	8.90	9.00	8.70
72	1.38	1.24	1.68	1.77	1.52	1.80	2.07	8.70	9.30	9.70	9.05	9.60	10.20	10.40
73	1.36	1.35	1.41	1.89	1.60	1.82	2.02	8.40	8.80	9.35	8.60	8.90	8.90	8.90
74	1.55	1.75	1.52	1.79	1.39	1.45	2.30	9.70	8.90	10;65	9.70	9.80	10.10	9.40
75	2.05	0.98	1.84	1.64	1.66	1.80	1.94	8.90	9.40	9.85	9.70	9.35	9.50	10.00
76	1.75	1.57	1.84	2.15	1.56	2.00	2.37	8.50	7.80	9.35	9.05	8.55	9.00	8.40
79	2.15	1.84	2.36	2.30	2.30	2.31	3.10	7.70	8.40	9.30	9.00	9.00	8.70	8.55
80	2.26	2.14	2.12	3.00	2.30	2.37	3.09	9.05	8.40	9.05	8.70	8.30	9.00	7.90
81	1.78	1.81	1.63	2.65	2.30	2.20	2.70	9.00	9.60	10.30	9.10	8.25	9.70	9.60
82	1.95	1.96	1.93	2.05	1.88	2.10	2.49	8.00	8.40	9.50	8.60	9.05	9.60	9.30
83	1.20	1.21	1.22	1.76	1.52	1.93	1.77	8.10	8.70	10.20	9.35	9.20	9.30	9.70
84	1.73	1.77	1.65	1.80	1.95	2.18	2.35	8.40	8.20	8.70	9.20	10.35	9.85	9.80
85	2.30	1.63	1.74	2.40	1.90	2.40	2.94	8.70	9.40	9.60	9.40	9.80	9.20	9.90
86	1.67	2.08	1.68	2.17	1.95	1.72	2.40	9.00	8.60	9.50	9.05	9.50	9.40	8.50
87	1.10	0.75	1.18	1.26	1.01	1.11	1.13	9.50	9.35	10.40	10.10	10.20	10.35	10.50
88	1.75	1.84	1.93	1.86	1.98	2.07	1.90	9.20	9.90	9.30	10.20	9.20	9.50	9.00
89	2.05	2.20	1.99	2.12	2.00	1.90	1.95	7.70	7.90	8.10	5.77	7.07	7.70	8.50
90	1.81	1.70	1.99	2.00	2.15	2.39	2.20	7.90	9.00	9.10	8.40	9.20	9.10	9.50
92	2.00	1.78	1.69	1.91	1.41	1.70	1.77	9.00	9.35	9.85	9.60	10.30	9.70	9.85
93	1.79	1.47	1.61	1.84	1.61	1.96	2.34	10.30	10.60	10.80	10.30	11.15	10.60	11.15
Mean old ewes	1.79	1.73	1.72	1.98	1.75	1.95	2.24	8.60	8.74	9.46	9.02	9.21	9.34	9.11

Appendix 2 c

Mean Plasma Calcium and Magnesium of 72 Sheep on Experimental Plots - Spring, 1961.

DATE: 19.4.61.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	OLD						YOUNG						Plot Means	
	No Pellets			Pellets			No Pellets			Pellets				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
2	75	2.20	9.00	56	2.11	10.10	12	1.04	9.90	22	1.73	10.65	1.77	
5	74	1.63	10.20	79	2.70	8.55	36	2.51	9.30	7	1.86	9.40	2.18	
8	81	2.49	9.80	87	1.14	10.80	1	1.56	9.30	14	1.48	9.85	1.67	
KO 10	55	1.61	9.70	89	1.90	9.00	33	1.59	10.90	3	0.37	10.10	1.37	
13	70	0.48	9.50	51	2.10	7.90	15	1.91	8.55	37	2.02	10.50	1.63	
17	52	1.55	10.65	58	2.35	9.70	2	1.53	9.40	35	1.80	10.00	1.81	
MEAN		1.66	9.81		2.05	9.34		1.69	9.56		1.54	10.08	1.74	9.70
1	57	2.11	8.25	71	2.18	9.40	9	2.00	8.40	13	1.80	8.90	2.02	
4	83	1.61	10.10	76	1.97	8.40	5	1.80	9.85	17	1.00	9.20	1.60	
9	84	2.02	10.10	88	1.60	8.10	10	2.11	9.40	11	1.52	9.20	1.81	
K1 11	69	1.78	9.40	65	1.79	9.60	16	1.59	10.00	6	1.55	10.60	1.68	
15	92	2.36	10.35	85	1.84	10.60	4	2.48	9.40	20	1.90	9.30	2.15	
16	82	1.93	8.70	86	1.80	8.90	24	2.13	10.65	30	1.51	9.35	1.84	
MEAN		1.97	9.48		1.86	9.17		2.02	9.62		1.55	9.43	1.85	9.42
3	73	1.21	9.05	72	1.63	10.65	18	2.07	8.10	29	1.43	9.20	1.59	
6	80	2.21	8.50	90	2.09	10.30	19	1.30	10.60	21	1.68	9.80	1.82	
7	53	1.59	9.70	59	1.00	10.85	32	0.62	10.50	23	2.00	9.70	1.30	
K2 12	68	1.77	9.40	63	1.30	8.25	27	2.36	10.20	40	1.33	10.65	1.69	
14	64	1.90	8.10	93	1.85	10.65	31	1.70	8.40	39	1.79	10.20	1.81	
18	60	1.63	7.80	62	1.59	8.80	25	2.27	7.75	28	2.14	9.80	1.91	
MEAN		1.72	8.89		1.58	9.92		1.72	9.26		1.73	9.89	1.69	9.49
OVERALL MEANS		1.78	9.39		1.83	9.48		1.81	9.48		1.61	9.80	1.75	9.54

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Young Sheep	1.71	9.64	Pellets	1.72	9.64
	Old Sheep	1.81	9.43	No Pellets	1.80	9.44

Appendix 2 c

Mean Plasma Calcium and Magnesium of 72 Sheep on Experimental Plots - Spring, 1961.

DATE: 22.4.61.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	OLD						YOUNG						Plot Means	
	No Pellets			Pellets			No Pellets			Pellets				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
2	75	1.55	10.00	56	1.76	10.08	12	1.17	10.24	22	1.49	10.64	1.49	
5	74	1.23	10.20	79	2.01	9.19	36	1.59	9.92	7	1.65	9.76	1.52	
8	81	1.69	10.60	87	1.62	9.11	1	1.50	10.40	14	1.37	10.32	1.55	
KO 10	55	1.61	8.80	89	1.86	8.87	33	2.10	9.52	3	0.46	10.89	1.51	
13	70	0.45	6.75	51	1.68	8.55	15	2.06	10.08	37	2.00	10.48	1.55	
17	52	2.15	9.05	58	1.03	12.26	2	1.21	10.32	35	1.60	9.52	1.50	
MEAN		1.47	9.23		1.66	9.68		1.61	10.08		1.43	10.27	1.54	9.81
1	57	1.21	8.87	71	1.66	9.76	9	1.90	10.56	13	0.93	10.73	1.43	
4	83	0.95	9.60	76	1.47	8.39	5	1.37	10.64	17	0.94	10.08	1.18	
9	84	1.91	9.03	88	1.37	10.32	10	1.94	9.52	11	1.80	9.60	1.76	
K1 11	69	2.31	9.11	65	1.23	7.90	16	1.27	10.08	6	1.03	10.81	1.46	
15	92	1.86	9.52	85	1.50	10.64	4	2.43	9.68	20	2.14	10.16	1.73	
16	82	1.90	8.87	86	1.79	9.52	24	2.29	9.76	30	1.91	10.16	1.72	
MEAN		1.69	9.17		1.50	9.42		1.87	10.04		1.46	10.26	1.63	9.72
3	73	0.90	9.35	72	1.03	10.73	18	1.95	8.47	29	0.88	10.24	1.19	
6	80	1.80	8.87	90	1.52	10.16	19	1.17	10.24	21	1.52	10.32	1.50	
7	53	1.84	9.11	59	1.25	10.16	32	0.55	9.35	23	2.20	8.95	1.46	
K2 12	68	1.84	9.92	63	0.90	9.27	27	2.50	10.32	40	1.29	10.48	1.63	
14	64	1.69	7.66	93	1.50	11.45	31	1.53	8.39	39	2.53	9.92	1.81	
18	60	1.13	10.10	62	0.92	8.31	25	1.94	9.76	28	1.97	9.84	1.49	
MEAN		1.53	9.17		1.19	10.01		1.61	9.42		1.73	9.96	1.51	9.64
OVERALL MEANS		1.56	9.19		1.45	9.70		1.69	9.85		1.54	10.16	1.56	9.73

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Young Sheep	1.62	10.00	Pellets	1.49	9.93
	Old Sheep	1.50	9.45	No Pellets	1.62	9.52

Appendix 2 c

Mean Plasma Calcium and Magnesium of 72 Sheep on Experimental Plots - Spring, 1961.

DATE: 28.4.61.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	OLD						YOUNG						Plot Means	
	No Pellets			Pellets			No Pellets			Pellets				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
2	75	1.56	10.64	56	2.09	10.73	12	0.91	10.32	22	1.73	11.05	1.57	
5	74	1.93	9.35	79	2.29	9.19	36	1.37	10.48	7	1.97	9.52	1.89	
8	81	1.76	10.56	87	0.96	9.76	1	1.52	9.35	14	1.60	10.48	1.46	
KO 10	55	1.40	9.52	89	1.61	8.79	33	1.93	10.00	3	0.51	11.45	1.38	
13	70	0.40	6.00	51	1.97	9.52	15	2.41	10.00	37	1.90	10.73	1.67	
17	52	1.18	11.61	58	1.85	9.76	2	1.25	10.08	35	1.57	9.11	1.46	
MEAN		1.37	9.61		1.80	9.63		1.57	10.04		1.55	10.39	1.57	9.92
1	57	1.37	8.87	71	1.79	9.27	9	2.06	10.81	13	1.26	10.89	1.62	
4	83	1.00	10.64	76	1.57	9.03	5	1.70	10.08	17	0.97	10.24	1.31	
9	84	1.76	10.48	88	2.17	9.76	10	1.57	9.76	11	1.83	10.00	1.83	
K1 11	69	1.92	9.43	65	0.72	8.55	16	1.60	10.56	6	1.20	11.05	1.36	
15	92	2.00	9.76	85	2.24	9.92	4	2.48	9.60	20	2.27	9.92	2.25	
16	82	2.00	9.68	86	2.00	9.11	24	2.10	10.56	30	2.15	10.40	2.06	
MEAN		1.68	9.81		1.75	9.27		1.92	10.23		1.61	10.42	1.74	9.93
3	73	0.52	8.39	72	2.07	10.08	18	1.76	9.84	29	1.27	9.84	1.41	
6	80	1.79	8.15	90	2.00	10.32	19	1.67	10.64	21	2.14	10.24	1.90	
7	53	1.92	9.60	59	1.01	9.84	32	0.95	10.16	23	1.78	9.43	1.42	
K2 12	68	2.02	10.00	63	1.52	9.68	27	2.52	10.32	40	0.89	10.64	1.74	
14	64	1.76	9.27	93	2.31	10.73	31	1.03	9.68	39	1.60	9.43	1.60	
18	60	0.73	9.19	62	1.15	10.73	25	2.21	9.76	28	1.72	10.32	1.45	
MEAN		1.46	9.10		1.68	10.23		1.69	10.07		1.57	9.98	1.60	9.85
OVERALL MEANS		1.50	9.51		1.74	9.71		1.72	10.11		1.58	10.26	1.64	9.90

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Young Sheep	1.65	10.19	Pellets	1.66	9.99
	Old Sheep	1.62	9.61	No Pellets	1.61	9.81

Appendix 2 c

Mean Plasma Calcium and Magnesium of 72 Sheep on Experimental Plots - Spring, 1961.

DATE: 3. 5. 61.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	OLD						YOUNG						Plot Means	
	No Pellets			Pellets			No Pellets			Pellets				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
KO 2	75	1.54	10.73	56	2.14	10.89	12	1.02	10.16	22	1.83	10.56	1.63	
5	74	2.00	10.28	79	2.35	10.16	36	2.08	11.29	7	2.07	10.32	2.13	
8	81	1.97	10.40	87	0.53	10.40	1	0.97	10.32	14	1.60	9.84	1.27	
10	55	1.41	9.60	89	2.02	9.11	33	2.00	10.56	3	0.57	11.37	1.50	
13	70	0.40	6.00	51	2.23	8.71	15	2.60	10.40	37	2.22	10.40	1.61	
17	52	1.89	10.64	58	1.80	10.48	2	1.32	9.43	35	2.00	9.11	1.75	
MEAN		1.54	9.61		1.85	9.96		1.67	10.36		1.72	10.27	1.69	10.05
K1 1	57	1.47	9.27	71	1.70	9.67	9	2.17	10.48	13	1.22	10.97	1.64	
4	83	1.16	9.84	76	1.70	9.27	5	1.91	10.40	17	1.02	10.16	1.45	
9	84	1.90	10.24	88	1.40	10.32	10	1.70	10.73	11	1.85	10.16	1.71	
11	69	2.19	9.52	65	1.81	9.27	16	1.62	10.64	6	1.07	11.13	1.67	
15	92	2.11	9.35	85	2.61	9.92	4	2.38	10.97	20	2.33	10.24	2.11	
16	82	2.00	9.52	86	2.20	8.06	24	2.30	10.81	30	1.91	10.48	2.10	
MEAN		1.81	9.62		1.90	9.42		2.01	10.67		1.57	10.52	1.82	10.06
K2 3	73	0.42	9.52	72	1.82	10.16	18	1.81	10.24	29	1.62	10.24	1.41	
6	80	2.07	8.23	90	1.67	9.67	19	1.66	10.48	21	1.93	8.39	1.84	
7	53	1.65	9.19	59	1.63	9.92	32	1.04	10.00	23	1.83	9.92	1.54	
12	68	2.31	9.19	63	1.30	9.84	27	2.73	10.64	40	1.74	10.48	2.02	
14	64	1.96	9.27	93	1.83	7.50	31	1.08	10.00	39	1.64	9.03	1.63	
18	60	1.07	9.67	62	1.39	10.48	25	2.35	8.55	28	2.00	9.92	1.70	
MEAN		1.58	9.18		1.61	9.59		1.78	9.99		1.79	9.66	1.69	9.61
OVERALL MEANS		1.64	9.47		1.79	9.66		1.82	10.34		1.69	10.15	1.73	9.90

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Young Sheep	1.76	10.25	Pellets	1.74	9.90
	Old Sheep	1.71	9.56	No Pellets	1.73	9.90

Appendix 2 c

Mean Plasma Calcium and Magnesium of 72 Sheep on Experimental Plots - Spring, 1961.

DATE: 12.5.61.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	OLD						YOUNG						Plot Means	
	No Pellets			Pellets			No Pellets			Pellets				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
KO 2	75	1.41	10.40	56	1.97	10.48	12	1.07	10.56	22	2.00	11.37	1.61	
5	74	1.66	10.81	79	2.10	10.16	36	1.90	11.45	7	2.05	10.48	1.93	
8	81	1.79	10.73	87	0.70	10.89	1	1.30	11.13	14	1.20	10.56	1.25	
10	55	1.50	9.68	89	2.05	9.92	33	1.81	11.37	3	0.56	11.42	1.46	
13	70	0.40	6.00	51	2.09	9.50	15	2.24	10.56	37	1.92	11.05	1.64	
17	52	1.97	10.40	58	1.92	10.24	2	1.33	10.73	35	1.70	9.11	1.73	
MEAN		1.46	9.67		1.80	10.20		1.61	10.97		1.57	10.67	1.61	10.37
K1 1	57	1.66	9.68	71	1.90	9.84	9	2.10	10.40	13	1.40	10.56	1.77	
4	83	1.19	10.32	76	1.52	9.84	5	1.71	10.89	17	1.00	10.89	1.36	
9	84	1.90	10.48	88	1.10	9.92	10	1.58	10.48	11	1.92	10.24	1.63	
11	69	2.24	9.92	65	1.85	10.48	16	1.82	11.13	6	1.15	11.53	1.77	
15	92	1.75	10.16	85	2.05	10.24	4	2.50	10.40	20	2.00	10.24	2.08	
16	82	1.87	10.16	86	1.90	8.87	24	2.32	11.05	30	2.00	10.97	2.02	
MEAN		1.77	10.12		1.72	9.87		2.00	10.73		1.58	10.74	1.77	10.36
K2 3	73	1.07	10.56	72	1.47	10.48	18	1.92	10.16	29	1.88	10.56	1.59	
6	80	1.60	10.08	90	1.74	10.08	19	1.77	10.48	21	1.52	9.03	1.66	
7	53	1.66	10.84	59	1.20	10.97	32	1.04	10.48	23	1.87	9.84	1.44	
12	68	1.54	10.81	63	1.47	11.85	27	2.32	10.89	40	1.30	11.13	1.66	
14	64	1.80	9.92	93	1.29	9.19	31	1.07	9.52	39	2.04	10.08	1.55	
18	60	0.85	10.24	62	1.25	9.11	25	2.30	9.76	28	1.88	10.32	1.57	
MEAN		1.42	10.41		1.40	10.28		1.76	10.22		1.74	10.16	1.58	10.27
OVERALL MEANS		1.55	10.07		1.64	10.11		1.78	10.64		1.63	10.52	1.65	10.33

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Young Sheep	1.71	10.58	Pellets	1.64	10.32
	Old Sheep	1.60	10.09	No Pellets	1.67	10.35

Appendix 2 c

Mean Plasma Calcium and Magnesium of 72 Sheep on Experimental Plots - Spring, 1961.

DATE: 24.5.61.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	OLD						YOUNG						Plot Means	
	No Pellets			Pellets			No Pellets			Pellets				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
2	75	1.83	9.92	56	1.80	10.24	12	1.15	10.56	22	1.70	11.05	1.62	
5	74	1.72	9.84	79	2.30	10.48	36	1.86	10.40	7	2.31	10.73	2.05	
8	81	1.83	10.64	87	0.78	11.45	1	1.46	10.73	14	1.35	10.40	1.36	
KO 10	55	1.51	9.76	89	1.60	9.76	33	1.64	10.24	3	0.60	10.97	1.35	
13	70	0.40	6.00	51	2.16	10.08	15	2.17	10.81	37	1.86	10.56	1.65	
17	52	2.03	10.40	58	1.71	10.48	2	1.50	10.40	35	1.39	10.16	1.66	
MEAN		1.56	9.43		1.72	10.42		1.63	10.52		1.54	10.65	1.61	10.25
1	57	1.35	8.63	71	1.88	10.40	9	1.81	10.56	13	1.10	10.48	1.54	
4	83	1.26	10.40	76	1.50	10.00	5	1.67	10.00	17	1.05	10.56	1.37	
9	84	1.95	11.13	88	1.50	10.40	10	1.54	10.89	11	1.92	11.13	1.73	
K1 11	69	2.07	10.08	65	2.00	10.32	16	1.66	10.89	6	1.35	10.64	1.77	
15	92	2.00	10.48	85	1.90	10.73	4	2.00	10.32	20	2.18	9.92	2.02	
16	82	1.75	10.32	86	1.30	10.48	24	2.15	10.97	30	2.00	10.40	1.80	
MEAN		1.73	10.17		1.68	10.39		1.81	10.61		1.60	10.52	1.70	10.42
3	73	0.91	10.40	72	1.60	10.56	18	1.80	10.73	29	1.20	10.32	1.38	
6	80	1.60	10.73	90	1.70	10.64	19	1.61	11.29	21	1.61	10.97	1.63	
7	53	1.66	9.67	59	1.15	9.92	32	0.83	10.32	23	1.95	9.43	1.40	
K2 12	68	1.40	10.24	63	1.71	10.73	27	2.17	10.81	40	1.47	11.21	1.69	
14	64	1.95	10.24	93	1.47	9.52	31	1.20	9.67	39	2.10	10.16	1.68	
18	60	0.69	10.24	62	1.16	9.92	25	1.91	10.73	28	2.17	11.61	1.48	
MEAN		1.37	10.25		1.47	10.22		1.59	10.59		1.75	10.62	1.54	10.42
OVERALL MEANS		1.55	9.95		1.62	10.34		1.67	10.57			10.59	1.62	10.36

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Young Sheep	1.65	10.58	Pellets	1.63	10.47
	Old Sheep	1.59	10.15	No Pellets	1.61	10.26

Appendix 2 c

Mean Plasma Calcium and Magnesium of 72 Sheep on Experimental Plots - Spring, 1961.

DATE: 29.5.61.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	OLD						YOUNG						Plot Means	
	No Pellets			Pellets			No Pellets			Pellets				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
2	75	1.37	9.92	56	1.84	10.16	12	1.02	10.64	22	1.66	11.37	1.47	
5	74	1.59	10.40	79	1.84	9.92	36	1.87	10.81	7	2.10	10.81	1.85	
8	81	1.87	11.61	87	1.06	11.13	1	1.47	11.13	14	1.52	10.40	1.48	
KO 10	55	1.62	9.52	89	1.49	10.08	33	1.65	10.73	3	0.54	11.05	1.33	
13	70	0.40	6.00	51	2.15	9.50	15	2.02	10.81	37	1.72	10.81	1.57	
17	52	1.90	11.37	58	1.74	10.89	2	1.49	11.29	35	1.48	10.40	1.65	
MEAN		1.46	9.80		1.69	10.28		1.59	10.90		1.50	10.81	1.56	10.45
1	57	1.26	9.52	71	2.17	10.40	9	2.10	10.48	13	1.05	10.89	1.65	
4	83	1.38	10.08	76	1.01	9.76	5	1.55	10.89	17	0.97	10.89	1.23	
9	84	1.73	10.73	88	1.87	10.81	10	1.70	11.21	11	1.72	11.37	1.76	
K1 11	69	1.72	9.92	65	1.98	10.16	16	1.55	10.32	6	1.06	10.73	1.58	
15	92	2.15	9.84	85	2.05	10.89	4	2.20	10.47	20	1.78	10.73	2.05	
16	82	1.95	10.48	86	1.67	10.48	24	2.12	11.53	30	2.20	11.61	1.99	
MEAN		1.70	10.10		1.79	10.42		1.87	10.90		1.46	11.04	1.70	10.61
3	73	1.22	10.16	72	1.34	9.76	18	1.72	10.73	29	1.68	11.05	1.49	
6	80	1.53	10.73	90	1.95	9.92	19	1.55	10.97	21	1.67	10.56	1.68	
7	53	1.75	10.89	59	1.15	10.56	32	0.95	10.81	23	1.90	10.24	1.44	
K2 12	68	1.02	10.64	63	1.40	11.29	27	2.10	11.29	40	1.70	11.29	1.56	
14	64	1.40	10.40	93	1.14	9.92	31	1.15	9.92	39	2.20	10.08	1.47	
18	60	0.72	10.89	62	1.20	10.00	25	1.87	10.81	28	1.92	10.16	1.43	
MEAN		1.27	10.62		1.36	10.24		1.56	10.76		1.85	10.56	1.51	10.54
OVERALL MEANS		1.48	10.17		1.61	10.31		1.67			1.60	10.80	1.59	10.53

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Young Sheep	1.64	10.83	Pellets	1.60	10.56
	Old Sheep	1.55	10.24	No Pellets	1.57	10.51

Appendix 2d. Plasma magnesium and calcium values of 72 Cheviot ewes grouped according to number of lambs suckled and also age

Plasma Constituent	Group	Lambs	Pretreatment period							Experimental period						
			21/2	3/3	14/3	22/3	29/3	6/4	13/4	19/4	22/4	28/4	3/5	12/5	24/5	29/5
Mg	47 ewes	single	1.72	1.64	1.65	1.86	1.72	1.82	2.03	1.75	1.50	1.65	1.73	1.63	1.61	1.58
	25 ewes	twins	1.81	1.67	1.68	1.85	1.72	1.82	2.13	1.77	1.66	1.61	1.74	1.69	1.64	1.61
	22 young ewes	single	1.70	1.64	1.63	1.74	1.68	1.64	1.80	1.67	1.54	1.65	1.74	1.70	1.63	1.63
	14 young ewes	twins	1.73	1.65	1.55	1.74	1.71	1.78	2.03	1.76	1.74	1.64	1.78	1.72	1.69	1.66
	25 old ewes	single	1.75	1.65	1.67	1.97	1.75	1.98	2.25	1.82	1.47	1.64	1.72	1.57	1.59	1.55
	11 old ewes	twins	1.90	1.94	1.85	1.99	1.75	1.90	2.28	1.78	1.57	1.58	1.70	1.65	1.58	1.53
Ca	47 ewes	single	8.90	9.12	9.88	9.47	9.58	9.69	9.43	9.59	9.86	10.06	10.12	10.49	10.56	10.70
	25 ewes	twin s	8.86	8.96	9.59	9.12	9.27	9.35	9.11	9.44	9.47	9.59	9.51	10.04	9.99	10.22
	22 young ewes	single	9.23	9.54	10.20	9.76	9.90	10.02	9.62	9.76	10.13	10.30	10.43	10.72	10.74	10.98
	14 young ewes	twins	9.07	9.22	9.92	9.52	9.44	9.44	9.35	9.45	9.80	10.00	9.96	10.36	10.33	10.58
	25 old ewes	single	8.60	8.73	9.58	9.19	9.28	9.38	9.24	9.43	9.62	9.85	9.84	10.28	10.40	10.45
	11 old ewes	twins	8.59	8.62	9.18	8.61	9.05	9.24	8.82	9.44	9.05	9.06	8.93	9.65	9.55	9.75

Appendix 2(e) Mineral content of herbage samples from Cochno plots Spring 1961

Fertiliser Treatment.	Plot No.	Magnesium (% in D.M.)					Calcium (% in D.M.)				
		18/4	29/4	9/5	17/5	17/8	18/4	29/4	9/5	17/5	17/8
	2	.17	.15	.15	.18	.17	.65	.57	.36	.51	.55
	5	.19	.15	.15	.16	.15	.70	.65	.43	.51	.68
	8	.19	.16	.16	.18	.16	.60	.61	.43	.47	.64
K0	10	.16	.16	.15	.17	.16	.60	.47	.32	.34	.57
	13	.16	.15	.15	.18	.15	.67	.57	.42	.45	.61
	17	.18	.15	.15	.15	.17	.70	.67	.45	.45	.70
K0	MEAN	.17	.15	.15	.17	.16	.65	.59	.40	.46	.63
	1	.16	.15	.16	.18	.16	.65	.49	.34	.45	.64
	4	.16	.15	.16	.18	.16	.66	.55	.36	.49	.61
K1	9	.17	.16	.16	.16	.16	.65	.59	.45	.45	.57
	11	.15	.13	.14	.18	.14	.65	.55	.43	.49	.59
	15	.15	.14	.15	.17	.15	.66	.59	.45	.53	.63
	16	.16	.15	.14	.17	.16	.66	.61	.51	.59	.78
K1	MEAN	.16	.15	.15	.17	.15	.66	.56	.42	.50	.64
	3	.17	.16	.15	.16	.14	.70	.51	.38	.53	.68
	6	.17	.15	.14	.16	.15	.65	.57	.38	.57	.63
K2	7	.18	.15	.15	.17	.15	.65	.53	.40	.49	.76
	12	.16	.14	.14	.17	.15	.60	.49	.45	.49	.70
	14	.15	.13	.13	.15	.14	.65	.49	.38	.40	.64
	18	.16	.13	.14	.16	.14	.66	.59	.38	.45	.55
K2	MEAN	.16	.14	.14	.16	.14	.65	.53	.40	.49	.66

Appendix 2(e) Mineral content of herbage samples from Cochno plots Spring 1961

Fertiliser Treatment.	Plot No.	Potassium (% in D.M.)					Sodium (% in D.M.)				
		18/4	29/4	9/5	17/5	17/8	18/4	29/4	9/5	17/5	17/8
	2	2.8	2.8	3.1	3.1	3.1	.20	.29	.39	.40	.20
	5	2.8	2.5	3.0	3.2	2.2	.26	.36	.41	.31	.32
K0	8	2.8	2.5	3.0	2.9	2.7	.22	.38	.37	.39	.25
	10	3.0	3.3	2.8	3.0	3.5	.20	.24	.28	.36	.15
	13	2.8	2.8	3.0	2.9	3.2	.22	.26	.35	.40	.17
	17	2.6	2.4	2.7	2.5	2.5	.36	.31	.49	.41	.31
K0	MEAN	2.8	2.7	2.9	2.9	2.9	.24	.31	.38	.38	.23
	1	3.3	3.5	3.4	3.3	3.4	.20	.28	.34	.27	.16
	4	3.5	3.3	3.7	3.5	3.2	.15	.26	.27	.30	.18
	9	3.3	3.2	3.4	3.3	3.3	.16	.26	.26	.30	.17
K1	11	3.3	3.2	3.2	3.4	2.8	.13	.15	.23	.27	.13
	15	3.0	3.1	3.1	3.0	3.0	.17	.20	.23	.21	.17
	16	3.0	2.9	3.2	2.9	2.3	.17	.20	.27	.23	.29
K1	MEAN	3.2	3.2	3.3	3.2	3.0	.16	.23	.26	.26	.18
	3	3.0	3.8	3.7	3.5	2.9	.23	.36	.27	.18	.12
	6	3.4	3.2	3.6	3.4	3.2	.13	.22	.27	.17	.13
K2	7	3.3	3.2	3.6	3.9	2.6	.17	.28	.33	.24	.17
	12	3.3	3.5	3.6	3.3	2.8	.16	.14	.20	.19	.15
	14	3.5	3.3	3.5	3.5	2.9	.12	.15	.15	.16	.10
	18	3.4	3.0	3.7	3.4	2.9	.17	.21	.28	.20	.13
K2	MEAN	3.3.	3.3	3.6	3.5	2.9	.16	.23	.25	.19	.13

Appendix 2(e) Mineral Content of herbage samples from Cochno plots Spring 1961

Fertiliser Treatment.	Plot No.	Phosphorus (% in D.M.)					Manganese (p.p.m. in D.M.)				
		18/4	29/4	9/5	17/5	17/8	18/4	29/4	9/5	17/5	17/8
	2	.48	.34	.38	.32	.45	91	110	131	167	132
	5	.43	.41	.38	.41	.32	110	106	122	114	112
K0	8	.34	.38	.37	.33	.35	104	96	91	122	79
	10	.38	.39	.18	.27	.38	123	120	152	185	126
	13	.39	.39	.33	.45	.37	107	111	122	154	88
	17	.36	.41	.42	.46	.34	96	108	104	111	92
K0	MEAN	.40	.39	.34	.37	.37	105	109	120	142	105
	1	.50	.42	.36	.20	.40	130	120	148	177	126
	4	.42	.38	.36	.50	.35	100	101	126	150	93
	9	.34	.39	.33	.41	.35	100	91	102	121	97
K1	11	.42	.38	.35	.33	.30	106	103	123	163	111
	15	.38	.38	.33	.34	.37	109	119	138	115	115
	16	.38	.38	.39	.35	.35	110	113	122	102	112
K1	MEAN	.41	.39	.35	.33	.35	109	108	126	138	109
	3	.39	.40	.38	.27	.38	102	117	121	121	131
	6	.42	.38	.39	.41	.35	113	100	106	122	90
K2	7	.42	.39	.42	.42	.33	110	111	106	127	97
	12	.43	.41	.33	.42	.35	-	115	131	168	108
	14	.38	.39	.41	.36	.37	120	117	128	135	112
	18	.39	.40	.41	.43	.36	80	93	96	115	99
K2	MEAN	.41	.40	.39	.39	.35	105	109	115	131	106

Appendix 2 f. Mean plasma calcium and magnesium concentrations of 24 Half-bred and 24 Cheviot ewes in March, 1962.

(Concentrations in mg/100 ml)

Half-Bred Ewe No.	Mg 9/3	Mg 16/3	Ca 9/3	Ca 16/3	Cheviot Ewe No.	Mg 9/3	Mg 16/3	Ca 9/3	Ca 16/3
41	1.90	2.30	7.52	9.84	2	1.00	1.20	10.08	9.27
42	2.20	2.50	9.27	9.52	3	0.95	0.95	10.00	10.08
43	2.75	2.10	7.98	9.52	5	2.10	2.20	8.95	9.68
44	1.50	1.60	9.84	10.56	6	2.00	1.35	8.47	8.79
45	2.20	1.85	9.60	10.08	7	1.90	2.15	8.79	9.68
46	1.80	2.00	9.60	9.76	10	1.60	1.80	9.52	9.27
48	2.90	2.55	8.55	10.40	11	1.95	1.65	9.60	9.60
49	1.80	2.05	9.03	9.11	12	1.30	1.20	10.00	9.84
51	2.60	1.95	7.98	9.92	13	1.40	1.20	9.92	9.19
52	3.15	3.05	7.90	8.23	15	2.45	2.50	8.63	8.79
53	2.45	2.40	9.03	10.08	17	1.20	1.35	9.19	9.43
54	2.50	2.75	9.19	10.73	19	1.40	1.65	9.19	10.08
55	3.05	2.65	8.31	10.16	20	2.25	2.25	7.98	7.90
56	2.15	2.30	9.43	10.40	21	2.35	2.30	8.63	9.43
57	2.80	2.95	9.03	9.19	22	2.00	1.95	8.71	9.35
58	2.30	2.30	9.11	9.84	25	3.20	2.85	7.17	7.01
59	2.30	2.90	9.35	9.92	29	1.85	1.55	8.85	8.89
60	3.15	3.40	7.66	7.98	31	1.40	1.75	8.31	7.66
63	2.20	2.35	9.35	10.16	32	1.15	1.05	9.43	10.48
64	2.05	2.30	9.03	9.84	34	2.15	2.15	9.27	9.35
67	2.95	2.50	8.06	8.31	36	2.15	1.80	8.47	10.08
70	2.30	2.70	9.03	8.23	37	1.90	2.00	8.71	8.87
72	2.10	2.20	9.19	9.35	39	2.50	2.20	7.66	8.95
77	2.65	2.35	7.98	9.52	40	1.85	1.80	9.52	9.60
Mean 24 Half-Bred Ewes	2.40	2.42	8.88	9.61	Mean 24 Cheviot Ewes.	1.83	1.77	8.95	9.22

Appendix 2(g)

Mean plasma calcium and magnesium of 48 sheep on Experimental plots 1962

DATE: 14.5.62.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.		HALF-BRED EWES						CHEVIOT EWES						Plot Means	
		No Drench			With Drench			No Drench			With Drench				
		Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
KO	5	46	0.50	9.92	49	0.95	9.44	3	0.50	10.24	21	1.10	10.56	0.76	10.04
	8	54	0.95	9.76	60	1.45	10.00	2	0.40	9.27	15	1.65	10.16	1.11	9.80
	13	55	1.10	10.00	57	1.25	9.92	19	0.70	10.48	5	0.80	10.32	0.96	10.18
	17	59	1.30	9.76	52	0.85	9.68	13	0.75	10.16	39	1.80	10.32	1.17	9.98
MEAN			0.96	9.86		1.12	9.76		0.58	10.04		1.33	10.34	1.00	10.00
K1	4	56	1.30	9.92	43	0.80	9.60	20	1.35	9.60	22	0.85	11.21	1.07	10.08
	9	58	0.70	10.16	51	1.70	9.92	37	1.20	9.92	25	1.05	10.08	1.16	10.02
	15	72	1.25	9.92	70	1.10	9.76	12	0.60	9.52	36	1.15	10.48	1.02	9.92
	16	64	1.35	10.08	77	1.10	9.19	6	0.45	9.92	7	1.10	10.32	1.00	9.88
MEAN			1.15	10.02		1.17	9.62		0.90	9.74		1.03	10.52	1.07	9.98
K2	6	42	1.30	9.84	63	1.35	10.48	29	0.65	8.87	11	1.40	10.00	1.17	9.80
	7	43	0.55	10.48	44	0.80	9.92	32	0.35	10.24	34	1.35	11.13	0.76	10.44
	14	45	1.00	9.84	41	1.05	8.79	31	0.85	9.68	17	0.45	10.16	0.83	9.62
	18	48	0.80	10.16	67	0.45	9.27	10	1.00	9.68	40	0.80	9.19	0.76	9.58
MEAN			0.91	10.08		0.91	9.62		0.71	9.62		1.00	10.12	0.88	9.86
OVERALL MEANS			1.01	9.99		1.07	9.66		0.73	9.80		1.13	10.33	0.98	9.94

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Halfbred	1.04	9.83	Drench	1.09	10.00
	Cheviot	0.93	10.06	No Drench	0.87	9.89

Appendix 2(g)

Mean plasma calcium and magnesium of 48 sheep on Experimental plots 1962

DATE: 16.5.62.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.		HALF-BRED EWES						CHEVIOT EWES						Plot Means	
		No Drench			With Drench			No Drench			With Drench				
		Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
KO	5	46	0.60	10.48	49	0.80	10.40	3	0.50	11.29	21	1.30	9.84	0.80	10.50
	8	54	1.00	10.48	60	1.25	10.56	2	0.90	9.52	15	1.90	9.92	1.26	10.12
	13	55	1.15	10.24	57	1.20	10.87	19	0.75	10.48	5	1.00	9.92	1.02	10.38
	17	59	1.00	11.21	52	0.90	11.45	13	0.65	10.73	39	1.70	10.48	1.06	10.96
MEAN			0.93	10.60		1.03	10.82		0.70	10.50		1.47	10.04	1.04	10.49
K1	4	56	1.15	9.92	43	1.25	10.65	20	1.50	9.03	22	0.90	10.00	1.20	9.90
	9	58	0.55	10.89	51	1.60	11.13	37	1.00	10.16	25	0.90	10.89	1.01	10.77
	15	72	1.30	9.52	70	1.25	9.35	12	0.35	10.32	36	1.25	9.60	1.03	9.70
	16	64	1.25	10.56	77	0.70	9.84	6	0.50	10.00	7	1.20	9.68	0.91	10.02
MEAN			1.06	10.22		1.20	10.24		0.83	9.88		1.06	10.04	1.04	10.10
K2	6	42	1.35	9.52	53	1.25	10.08	29	0.60	9.35	11	1.40	9.84	1.15	9.69
	7	43	0.55	10.73	44	0.80	9.84	32	0.35	10.16	34	1.25	11.53	0.73	10.56
	14	45	0.40	9.68	41	0.65	10.08	31	0.75	9.11	17	0.75	10.00	0.63	9.71
	18	48	0.80	10.65	67	0.65	8.79	10	0.85	9.52	40	0.70	10.08	0.75	9.76
MEAN			0.77	10.14		0.83			0.63	9.53		1.02	10.36	0.82	9.94
OVERALL MEANS			0.93	10.32		1.03	10.25		0.73	9.97		1.19	10.15	0.96	10.17

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Halfbred	0.98	10.29	Drench	1.11	10.20
	Cheviot	0.95	10.06	No Drench	0.83	10.15

Appendix 2(g)

Mean plasma calcium and magnesium of 48 sheep on Experimental plots 1962

DATE: 19. 5. 62.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	HALF-BRED EWES						CHEVIOT EWES						Plot Means	
	No Drench			With Drench			No Drench			With Drench				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
5	46	0.90	10.89	49	1.15	9.92	3	0.75	11.37	21	1.15	10.56	0.98	10.69
8	54	1.05	10.56	60	1.35	10.73	2	0.90	10.00	15	2.00	10.16	1.32	10.36
KO 13	55	1.45	10.73	57	1.25	11.45	19	0.90	11.21	5	1.10	10.65	1.17	11.01
17	59	1.30	10.89	52	1.50	10.81	13	0.90	10.65	39	1.80	10.16	1.37	10.63
MEAN		1.17	10.77		1.31	10.73		0.86	10.81		1.51	10.38	1.22	10.67
4	56	1.40	10.81	43	1.65	10.24	20	1.35	10.16	22	0.90	10.65	1.32	10.47
9	58	0.70	10.65	51	1.80	10.97	37	1.20	10.65	25	1.05	10.48	1.18	10.69
K1 15	72	1.40	10.00	70	1.10	10.32	12	0.65	10.08	36	0.95	10.40	1.02	10.20
16	64	1.65	10.56	77	0.70	10.16	6	0.65	10.81	7	1.25	10.81	1.06	10.59
MEAN		1.28	10.51		1.31	10.42		0.96	10.43		1.03	10.59	1.15	10.48
6	42	1.55	10.08	53	1.40	10.24	29	0.60	10.81	11	1.45	9.92	1.25	10.26
7	43	0.80	11.29	44	1.00	10.89	32	0.55	10.24	34	1.55	10.56	0.97	10.75
K2 14	45	0.50	10.16	41	1.10	9.68	31	0.75	9.60	17	0.65	10.24	0.75	9.86
18	48	1.10	11.45	67	0.80	10.40	10	0.95	9.68	40	1.00	10.16	0.96	10.42
MEAN		0.98	10.75		1.07	10.30		0.71	10.08		1.16	10.22	0.98	10.34
OVERALL MEANS		1.15	10.67		1.23	10.48		0.85	10.44		1.24	10.40	1.12	10.50

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Halfbred	1.19	10.58	Drench	1.23	10.44
	Cheviot	1.04	10.42	No Drench	1.00	10.56

Appendix 2(g)

Mean plasma calcium and magnesium of 48 sheep on Experimental plots 1962

DATE: 23. 5. 62. (10 a.m.)

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	HALF-BRED EWES						CHEVIOT EWES						Plot Means	
	No Drench			With Drench			No Drench			With Drench				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
5	46	0.90	10.81	49	1.10	10.81	3	0.45	11.29	21	1.15	10.73	0.90	10.91
8	54	0.90	11.45	60	1.45	11.37	2	1.05	10.73	15	1.90	10.89	1.33	11.11
KO 13	55	1.20	11.61	57	1.45	11.37	19	1.00	10.89	5	1.15	11.05	1.20	11.23
17	59	1.50	11.45	52	1.40	10.81	13	0.75	10.89	39	1.85	11.37	1.38	11.13
MEAN		1.13	11.33		1.35	11.09		0.81	10.95		1.51	11.01	1.20	11.10
4	56	1.65	10.89	43	1.30	11.21	20	1.40	9.84	22	0.75	10.97	1.28	10.73
9	58	0.85	11.61	51	1.20	11.37	37	1.35	10.48	25	1.40	10.97	1.20	11.11
K1 15	72	1.45	10.00	70	1.05	10.89	12	0.40	10.16	36	1.15	11.29	1.01	10.59
16	64	1.75	11.37	77	0.80	10.73	6	0.50	10.73	7	1.30	10.97	1.09	10.95
MEAN		1.43	10.97		1.09	11.05		0.91	10.30		1.15	11.05	1.14	10.84
6	42	1.05	10.97	53	1.40	10.89	29	0.55	11.37	11	1.35	11.13	1.09	11.09
7	43	0.70	10.89	44	1.20	11.05	32	0.35	10.32	34	1.35	11.77	0.90	11.01
K2 14	45	0.50	10.16	41	9.90	10.48	31	0.65	10.08	17	0.65	10.32	0.68	10.26
18	48	1.05	11.45	67	0.85	10.81	10	0.95	10.16	40	0.90	10.81	0.94	10.81
MEAN		0.83	10.87		1.09	10.81		0.63	10.48		1.06	11.01	0.90	10.79
OVERALL MEANS		1.13	11.07		1.18	10.98		0.78	10.58		1.24	11.02	1.08	10.91

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Halfbred	1.15	11.02	Drench	1.21	11.00
	Cheviot	1.01	10.80	No Drench	0.95	10.82

Appendix 2(g)

Mean plasma calcium and magnesium of 48 sheep on Experimental plots 1962

DATE: 23. 5. 62. (2 p.m.)

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	HALF-BRED EWES						CHEVIOT EWES						Plot Means	
	No Drench			With Drench			No Drench			With Drench				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
5	46	0.80	10.32	49	1.70	10.81	3	0.55	11.13	21	1.70	10.48	1.19	10.69
8	54	0.90	10.65	60	2.05	10.73	2	1.05	10.32	15	2.30	10.48	1.58	10.55
K0 13	55	1.15	11.29	57	1.80	10.97	19	1.05	11.37	5	1.60	11.05	1.40	11.17
17	59	1.35	11.05	52	1.75	10.65	13	0.70	10.81	39	2.35	10.97	1.54	10.87
MEAN		1.05	10.83		1.83	10.79		0.84	10.91		1.99	10.75	1.43	10.82
4	56	1.50	10.65	43	1.95	10.73	20	1.35	9.76	22	1.20	11.05	1.50	10.55
9	58	0.75	10.89	51	2.00	10.73	37	1.35	10.24	25	1.85	10.65	1.49	10.63
K1 15	72	1.50	10.00	70	1.65	10.48	12	0.35	10.32	36	1.75	10.97	1.31	10.44
16	64	1.55	10.73	77	1.55	10.16	6	0.45	10.40	7	2.25	10.81	1.45	10.53
MEAN		1.33	10.57		1.79	10.53		0.88	10.18		1.76	10.87	1.44	10.54
6	42	1.70	10.08	53	1.90	10.48	29	0.65	10.40	11	1.75	11.13	1.50	10.52
7	43	0.75	10.48	44	1.55	10.56	32	0.50	10.00	34	1.80	10.97	1.15	10.50
K2 14	45	0.50	10.16	41	1.45	10.32	31	0.60	10.32	17	1.10	10.32	0.91	10.28
18	48	1.05	10.73	67	1.40	10.65	10	0.90	10.40	40	1.35	9.84	1.18	10.41
MEAN		1.00	10.36		1.58	10.50		0.66	10.28		1.50	10.57	1.18	10.43
OVERALL MEANS		1.13	10.59		1.73	10.61		0.79	10.46		1.75	10.73	1.35	10.59

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Halfbred	1.43	10.60	Drench	1.74	10.67
	Cheviot	1.27	10.59	No Drench	0.96	10.52

Appendix 2(g)

Mean plasma calcium and magnesium of 48 sheep on Experimental plots 1962

DATE: 24. 5. 62.

(Concentrations in mg/100 ml)

Plot Treatment and Plot No.	HALF-BRED EWES						CHEVIOT EWES						Plot Means	
	No Drench			With Drench			No Drench			With Drench				
	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Ewe No	Mg	Ca	Mg	Ca
5	46	0.80	10.40	49	0.95	10.65	3	0.45	10.97	21	1.25	11.15	0.86	10.79
8	54	0.95	10.81	60	1.55	10.97	2	0.80	10.48	15	1.85	10.32	1.28	10.65
KO 13	55	1.30	10.65	57	1.45	10.89	19	1.00	10.56	5	1.20	10.56	1.23	10.67
17	59	1.35	11.13	52	1.05	11.13	13	0.70	10.08	39	2.00	10.56	1.27	10.73
MEAN		1.10	10.75		1.25	10.91		0.73	10.52		1.57	10.65	1.17	10.71
4	56	1.50	11.13	43	1.40	10.73	20	1.50	10.00	22	0.85	11.05	1.31	10.73
9	58	0.45	11.13	51	1.70	10.65	37	1.05	10.32	25	1.25	11.05	1.11	10.79
K1 15	72	1.35	10.48	70	1.15	10.48	12	0.40	10.00	36	1.10	10.81	1.00	10.44
16	64	1.50	11.05	77	0.90	10.08	6	0.40	10.40	7	1.30	10.48	1.02	10.50
MEAN		1.20	10.95		1.28	10.49		0.83	10.18		1.12	10.85	1.11	10.62
6	42	1.55	10.73	53	1.30	11.13	29	0.60	10.24	11	1.50	10.81	1.23	10.73
7	43	0.75	11.37	44	0.90	11.29	32	0.55	10.73	34	1.45	11.21	0.91	11.15
K2 14	45	0.55	10.81	41	0.90	10.32	31	0.30	10.65	17	0.65	10.16	0.60	10.49
18	48	1.00	10.89	67	0.80	10.89	10	0.85	9.84	40	0.80	10.73	0.86	10.59
MEAN		0.96	10.95		0.97	10.91		0.57	10.37		1.10	10.73	0.90	10.74
OVERALL MEANS		1.09	10.88		1.17	10.77		0.72	10.36		1.27	10.74	1.06	10.69

COMPOSITE MEANS		Mg	Ca		Mg	Ca
	Halfbred	1.12	10.82	Drench	1.22	10.75
	Cheviot	1.00	10.55	No Drench	0.90	10.62

Appendix 2h. Mineral content of herbage samples from Cochno plots on 18th May, 1962.

(figures quoted as dry matter basis)

Potassium Treatment	Plot No.	Mg%	Ca%	K%	Na%
	5	.168	.64	2.0	.56
	8	.175	.64	2.25	.50
K0	13	.168	.49	2.65	.36
	17	.168	.51	2.00	.56
K0 Mean		.170	.57	2.23	.50
	4	.155	.52	3.2	.35
	9	.183	-	3.0	.35
K1	15	.150	.40	2.95	.35
	16	.138	.47	2.50	.43
K1 Mean		.156	.46	2.79	.37
	6	.125	.53	3.0	.27
	7	.125	.53	3.1	.30
K2	14	.130	.40	3.1	.24
	18	.153	.46	2.85	.37
K2 Mean		.133	.48	3.01	.29

Appendix 2(i) Plasma magnesium concentrations of 30 Cheviot ewes and 30 Half Breds on plots at Cochno Farm - 1963

Treatment	Plot No.	CHEVIOT Ewe No	15/5	17/5	20/5	23/5	HALF-BRED Ewe No	15/5	17/5	20/5	23/5	PLOT MEANS (Both Breeds) 15/5	17/5	20/5	23/5
Low Mg applied winter	3	419	1.10	1.35	1.10	0.85	K12	1.35	1.40	1.70	1.30	1.45	1.49	1.36	1.32
		415	2.15	1.85	1.15	1.65	643	1.20	1.35	1.50	1.50				
	11	440	1.50	1.05	1.45	0.90	K15	2.20	1.85	1.40	1.70	1.61	1.29	1.46	1.27
		416	1.10	1.05	1.55	1.25	K20	1.65	1.20	1.45	1.25				
	12	434	1.60	1.25	1.45	1.40	659	1.70	1.70	1.59	1.55	1.66	1.21	1.38	1.24
		421	1.65	1.00	1.45	1.25	644	1.70	0.90	1.05	0.75				
MEAN			1.52	1.26	1.34	1.22		1.63	1.40	1.65	1.34	1.57	1.33	1.40	1.28
Low Mg applied Spring.	4	422	1.05	1.05	1.20	1.10	649	0.90	2.05	1.00	1.90	1.15	1.44	1.26	1.38
		407	1.50	1.35	1.45	1.40	654	1.15	1.30	1.40	1.10				
	13	413	0.60	0.60	0.85	0.45	K9	1.70	1.95	1.55	1.60	1.38	1.42	1.42	1.24
		428	1.70	1.40	1.35	1.30	664	1.50	1.75	1.95	1.60				
	16	426	1.00	0.95	1.40	0.85	657	1.40	1.25	1.60	1.30	1.31	1.34	1.60	1.19
		427	1.70	1.65	1.60	1.45	K10	1.15	1.50	1.80	1.15				
MEAN			1.26	1.17	1.45	1.09		1.30	1.63	1.55	1.41	1.28	1.40	1.43	1.27
High Mg.	9	423	1.85	1.55	1.95	1.65	K5	1.45	2.05	2.15	1.85	1.22	1.29	1.90	1.51
		402	0.85	0.90	1.35	1.25	663	0.75	0.65	2.15	1.30				
	18	409	1.65	1.45	1.90	1.40	651	1.75	2.15	2.25	1.00	1.65	1.66	2.04	1.00
		410	1.30	1.30	1.75	0.70	K8	1.90	1.75	2.25	0.90				
	15	436	1.15	0.95	1.90	1.10	652	0.80	1.85	1.35	1.45	1.06	1.39	1.54	1.39
		411	1.40	1.15	1.45	1.75	81	0.90	1.60	1.45	1.25				
MEAN			1.37	1.22	1.72	1.31		1.26	1.68	1.93	1.29	1.31	1.45	1.83	1.30
KO	5	430	1.70	1.75	1.60	1.00	653	1.00	0.40	1.15	1.30	1.18	1.05	1.17	1.07
		437	0.65	0.80	0.75	0.95	K14	1.35	1.25	1.20	1.05				
	8	433	1.85	1.25	1.75	1.55	K17	1.20	0.70	1.45	0.90	1.32	0.98	1.40	1.15
		403	0.60	0.55	0.85	0.55	K11	1.65	1.40	1.55	1.60				
	17	429	0.80	1.05	0.90	0.50	656	1.25	1.00	1.55	1.45	1.19	0.99	1.17	0.97
		405	1.35	1.10	1.15	1.10	K19	1.35	0.80	1.10	0.85				
MEAN			1.37	1.08	1.17	0.94		1.26	0.93	1.33	1.19	1.23	1.00	1.23	1.07
K2	6	417	0.80	0.40	0.60	0.75	645	0.70	0.45	0.90	0.70	0.91	0.59	0.90	0.77
		438	1.25	0.70	1.10	0.95	641	0.90	0.80	1.00	0.70				
	7	425	1.75	0.85	1.25	1.05	K7	1.05	0.90	0.95	0.75	1.20	0.90	0.95	0.91
		432	0.65	0.35	0.45	0.45	655	1.35	1.50	1.15	1.40				
	14	412	1.35	0.40	0.80	1.00	660	1.60	1.75	1.20	1.65	1.37	0.99	0.95	1.15
		406	1.00	0.55	0.65	0.45	642	1.55	1.25	1.15	1.40				
MEAN			1.13	0.54	0.81	0.78		1.19	1.11	1.06	1.10	1.16	0.82	0.93	0.94
OVERALL BREED MEANS			1.29	1.05	1.27	1.07		1.34	1.35	1.46	1.27				

Appendix 2 (i) Plasma calcium concentrations of 30 Cheviot ewes and 30 Half-Bred ewes on plots at Cochno Farm - 1963.

Treatment.	Plot No.	CHEVIOT Ewe No.	15/5	17/5	20/5	23/5	HALF-BRED Ewe No.	15/5	17/5	20/5	23/5	PLOT MEANS (Both Breeds) 15/5	17/5	20/5	23/5
Low Mg winter.	3	(419	10.24	10.48	10.56	10.97	K12	9.11	10.00	9.60	10.40)	9.78	10.34	10.18	10.38
		(415	9.84	9.92	10.32	10.24	643	9.92	10.97	10.24	9.92)				
	11	(440	9.84	10.16	10.16	11.69	K15	3.95	6.85	9.76	10.24)	8.50	9.37	10.10	10.70
		(416	10.80	10.48	10.56	11.13	K20	9.43	10.00	9.92	9.76)				
	12	(434	10.08	10.97	11.13	11.45	659	9.68	10.08	10.16	10.48)	9.49	10.26	10.32	10.90
		(421	8.95	10.08	10.00	10.89	644	9.27	9.92	10.08	10.80)				
	MEAN		9.96	10.35	10.46	11.06		8.56	9.64	9.96	10.27	9.26	9.99	10.20	10.66
Low Mg Spring.	4	(422	9.76	10.88	10.32	10.89	649	9.35	9.11	10.64	7.82)	9.82	10.22	10.34	10.06
		(407	9.92	10.40	10.00	10.48	654	10.24	10.48	10.40	11.05)				
	13	(413	10.00	10.80	10.40	10.64	K9	10.48	8.79	9.68	9.68)	10.06	9.79	9.96	10.08
		(428	10.00	9.52	9.92	10.08	664	9.76	10.08	9.84	9.92)				
	16	(426	8.71	9.76	9.52	10.24	657	9.68	10.72	9.84	10.00)	9.55	10.14	10.04	10.39
		(427	10.64	10.24	10.72	11.25	K10	9.19	9.84	10.08	10.08)				
	MEAN		9.84	10.27	10.15	10.60		9.78	9.84	10.08	9.76	9.81	10.05	10.11	10.18
High Mg.	9	(423	9.19	9.92	9.92	9.84	K5	9.60	9.68	10.00	10.08)	9.41	9.81	9.84	10.02
		(402	9.35	9.68	10.00	9.84	663	9.52	9.98	9.43	10.32)				
	18	(409	9.43	10.16	10.56	9.84	651	9.60	10.08	10.40	10.56)	9.64	10.24	10.24	10.22
		(410	9.68	9.84	9.76	9.76	K8	9.84	10.88	10.24	10.72)				
	15	(436	9.68	10.48	10.00	10.97	652	9.92	9.27	10.56	10.00)	9.70	10.04	10.28	10.30
		(411	9.52	10.40	10.40	9.84	81	9.68	10.00	10.16	10.40)				
	MEAN		9.48	10.08	10.11	10.02		9.69	9.98	10.13	10.35	9.58	10.03	10.12	10.18
KO	5	(430	9.52	10.00	10.32	10.80	653	9.92	10.32	10.32	10.16)	9.48	9.81	9.92	9.86
		(437	9.84	10.16	9.76	9.92	K14	8.63	9.01	9.27	8.55)				
	8	(433	10.32	11.13	10.80	10.80	K17	8.79	10.64	8.87	9.92)	9.74	10.72	10.26	10.50
		(403	10.40	10.80	11.13	11.05	K11	9.43	10.32	10.24	10.24)				
	17	(429	9.60	11.45	8.95	9.68	656	9.43	10.32	9.92	10.08)	9.35	10.60	9.84	9.78
		(405	9.92	10.88	10.80	10.24	K19	8.46	9.76	9.68	9.11)				
	MEAN		9.93	10.74	10.29	10.42		9.11	10.06	9.72	9.59	9.52	10.39	10.00	10.05
K2	6	(417	9.19	10.32	10.08	9.68	645	10.40	10.40	9.92	10.56)	9.51	9.71	9.89	9.88
		(438	9.52	9.11	10.16	9.68	641	8.95	9.01	9.43	9.60)				
	7	(425	9.92	10.72	10.40	10.40	K7	8.87	9.11	9.11	10.40)	9.50	9.86	10.04	10.08
		(432	9.35	9.76	10.08	9.52	655	9.84	9.84	10.56	10.00)				
	14	(412	9.52	9.27	9.43	9.27	660	9.01	9.43	11.13	9.06)	9.85	9.91	10.10	9.92
		(406	9.52	11.53	9.92	10.40	642	9.35	9.43	9.92	10.24)				
	MEAN		9.50	10.12	10.01	9.83		9.40	9.54	10.01	10.10	9.45	9.83	10.01	9.96
OVERALL BREED MEANS			9.74	10.31	10.20	10.38		9.31	9.81	9.98	10.03				

Appendix 2(i) Plasma calcium and magnesium concentrations of 24 Blackface ewes at Cochno Farm - June 1963.

Fertiliser Treatment.	Plot No.	Ewe No.	No. of Lambs.	Mg (mg/100 ml)						Ca (mg/100 ml)					
				11/6	12/6	14/6	17/6	21/6	18/7	11/6	12/6	14/6	17/6	21/6	18/7
										9.03	10.80	12.16	10.07	9.75	7.82
	8	85	1	2.50	1.65	2.45	2.20	2.25	2.60	8.87	9.44	9.76	9.27	10.16	10.00
		87	1	2.20	2.05	0.85	2.70	2.30	2.30	8.39	9.84	10.08	9.52	10.88	9.52
		97	1	2.25	1.95	1.85	2.50	2.35	2.25	8.79	9.52	9.84	9.76	9.60	9.35
KO		90	2	2.35	1.95	1.65	2.15	2.00	2.20	8.87	10.32	10.48	8.71	9.84	9.52
	17	66	2	1.55	1.10	1.45	2.15	2.00	1.60	7.98	8.06	8.71	10.64	9.35	8.55
		71	2	2.80	2.60	2.65	1.05	2.35	2.60	9.19	10.24	10.40	9.43	10.00	10.48
		76	1	2.35	1.80	1.60	1.70	2.40	1.85	9.19	9.44	10.08	9.19	10.16	10.08
		86	1	1.70	1.70	1.85	2.15	1.85	1.90	8.81	9.51	9.92	9.49	9.84	9.66
MEAN				2.27	1.85	1.79	2.08	2.19	2.16	8.79	9.68	10.19	9.69	9.97	9.42
	6	80	1	1.35	0.80	0.45	0.55	0.70	0.90	8.87	9.52	9.11	8.95	10.32	10.64
		83	1	2.20	1.40	0.80	1.45	1.50	1.20	8.23	10.56	11.45	9.68	10.97	10.32
		84	1	2.90	1.75	1.45	2.05	2.00	2.60	7.98	9.68	10.97	9.84	10.32	8.46
K2		93	2	1.90	0.90	0.80	1.10	0.80	1.10	9.44	10.16	10.24	9.52	10.56	9.52
	7	67	1	1.55	1.05	2.35	1.00	1.40	1.65	9.19	9.84	9.35	9.68	10.56	9.92
		73	2	2.35	1.50	1.75	2.30	2.20	1.75	8.87	11.29	10.32	8.81	10.08	9.76
		94	1	1.95	1.80	2.25	2.45	2.00	2.50	9.27	10.08	9.84	9.19	10.40	9.60
		96	2	2.90	2.20	1.45	1.70	1.50	1.25	7.90	7.98	11.05	10.08	11.53	10.32
MEAN				2.14	1.43	1.42	1.57	1.51	1.62	8.72	9.89	10.17	9.47	10.59	9.82
	9	77	2	2.10	1.50	1.60	1.90	2.30	2.05	8.47	9.84	11.21	8.87	9.27	9.52
		82	1	2.55	2.50	2.35	2.85	2.40	2.40	9.44	10.48	10.56	9.35	10.88	9.27
		91	2	1.60	1.60	2.15	1.85	2.15	1.80	8.47	8.87	8.14	8.95	9.52	9.35
High		92	1	2.55	2.00	1.85	2.40	2.10	2.20	7.74	7.74	9.52	8.95	9.68	8.63
Mg.	18	68	1	2.60	1.80	2.75	1.80	2.35	1.95	8.47	9.60	8.46	9.68	9.01	8.30
		74	1	3.45	2.45	1.95	2.60	3.15	2.25	8.47	9.27	10.40	8.71	9.19	9.76
		81	2	2.05	2.00	1.80	2.15	2.05	1.65	9.35	11.05	11.77	10.72	10.32	10.48
		88	1	1.75	1.95	2.05	1.90	1.90	2.25	8.55	8.79	9.11	9.68	10.16	9.76
MEAN				2.38	1.97	2.07	2.18	2.30	2.07	8.62	9.45	9.82	9.36	9.75	9.38

Appendix 2(k) Herbage mineral contents of Cochno plots in May and June, 1963.

Fertiliser Treatment.	Plot No.	% Mg.			% Ca			% K			% Na.		
		13/5	22/5	11/6	13/5	22/5	11/6	13/5	22/5	11/6	13/5	22/5	11/6
CONTROL	5	.163	.257	.163	.78	.62	.72	2.4	2.9	3.3	.48	.28	.37
	8	.190	.175	.103	.82	.64	.62	2.9	2.8	2.0	.48	.32	.34
	17	.225	.145	.143	.86	.40	.66	2.0	1.4	1.4	.64	.43	.57
MEAN		.193	.192	.136	.82	.55	.66	2.4	2.4	2.2	.53	.34	.43
POTASSIUM	6	.155	.105	.105	.67	.40	.50	3.6	2.7	3.1	.24	.19	.18
	7	.150	.122	.148	.66	.58	.60	3.6	3.2	3.8	.26	.21	.27
	14	.158	.125	.078	.66	.37	.50	3.6	2.9	2.8	.24	.22	.24
MEAN		.154	.116	.110	.66	.45	.53	3.6	2.9	3.2	.25	.21	.23
LARGE Mg. APPLICATION	9	.285	.315	.238	.70	.40	.60	2.9	2.0	2.4	.33	.21	.36
	15	.332	.430	.190	.70	.22	.59	2.8	2.0	2.3	.34	.21	.22
	18	.270	.340	.240	.72	.16	.68	2.7	1.8	2.3	.38	.27	.46
MEAN		.296	.362	.223	.71	.26	.62	2.8	1.9	2.3	.35	.23	.35
LOW Mg. APPLIED IN WINTER	3	.198	.207	.150	.76	.39	.53	2.8	2.8	3.1	.34	.22	.23
	11	.195	.195	.143	.72	.55	.62	2.8	2.3	3.1	.34	.30	.31
	12	.198	.185	.163	.77	.31	.66	2.8	2.3	3.4	.35	.25	.29
MEAN		.197	.195	.152	.75	.42	.60	2.8	2.5	3.2	.35	.26	.28
LOW Mg. APPLIED IN SPRING	4	.173	.212	.160	.92	.53	.66	3.2	3.0	3.0	.34	.23	.22
	13	.185	.262	.175	.82	.62	.74	3.2	2.1	2.6	.44	.24	.39
	16	.165	.272	.158	.68	.66	.66	2.7	1.9	3.2	.30	.19	.29
MEAN		.175	.249	.164	.81	.60	.69	3.0	2.3	2.9	.36	.22	.30

Extra sample taken from low Spring Mg Plots on 15/5 :-

Plot 4 - 0.32% Mg.
13 - 0.46% Mg.
16 - 0.30% Mg.

MEAN - .36% Mg.

Appendix 3(a) Lambing date and number of lambs born and suckling each ewe in Experiment 4.

Serial No. of Ewe.	Date of Lambing.	No. of Lambs Born.	No. of Lambs on 4.5.62.
CALCIUM SUPPLEMENTED GROUP.			
30	24/3	2	2
40	30/4	2	2
53	17/3	2	1
35	31/3	2	1
56	2/4	2	1
63	1/4	1	1
74	5/4	1	-
90	27/3	2	2
CALCIUM DEFICIENT GROUP.			
33	2/4	2	2
60	30/3	1	1
58	7/4	2	1
82	20/3	2	1
84	18/3	1	1
88	1/4	1	1
92	30/3	1	1
59	10/4	1	1
68	8/4	2	2
69	6/4	1	1
73	10/4	1	-
79	24/3	1	-
87	2/4	2	1
16	21/3	2	2
54	1/4	1	1
64	5/4	1	1
76	2/4	2	-
81	4/4	2	2
93	25/3	2	-

Appendix 3(b) Plasma magnesium concentrations of 19 Cheviot ewes on low calcium diet 1962.

Ewe No.	28/11	4/12	21/12	4/1	22/1	2/2	15/2	1/3	12/3	19/3	26/3	29/3
33	1.50	1.60	1.35	1.60	1.40	1.80	1.50	1.80	2.55	3.10	2.90	2.95
60	1.10	1.10	2.25	2.50	2.20	2.95	3.45	3.70	4.05	3.65	3.20	3.45
58	1.35	1.30	1.85	2.40	2.30	2.15	2.10	2.50	2.60	2.10	2.65	2.70
82	1.80	1.70	2.40	2.80	2.35	2.45	3.10	3.25	3.10	3.10	2.00	2.60
84	1.65	1.50	2.00	2.30	2.45	2.80	2.75	3.00	3.00	2.70	3.50	3.15
88	1.90	1.70	2.30	2.10	1.80	2.35	2.70	2.80	2.95	3.05	2.95	2.85
92	1.95	1.75	1.75	2.10	2.00	2.25	2.35	2.70	2.50	2.55	0.75	2.80
59	1.30	1.60	1.60	2.25	2.15	2.20	2.50	2.40	2.95	2.80	3.95	4.05
68	1.65	1.35	2.30	1.75	2.15	2.65	3.25	2.90	3.35	3.40	3.50	3.40
69	1.80	1.45	2.35	2.55	2.20	2.65	3.45	3.45	3.95	4.25	3.20	3.75
73	1.10	1.25	1.60	2.40	1.05	1.80	2.20	2.40	2.25	2.35	1.65	1.75
79	1.90	2.15	2.75	2.65	2.60	2.60	2.95	3.25	3.00	2.50	3.20	2.60
87	0.95	0.85	1.40	2.00	1.25	2.10	2.70	2.45	3.20	2.75	2.85	2.00
16	1.40	1.15	1.60	1.85	1.85	2.05	2.10	2.20	2.45	2.40	1.75	1.25
54	1.30	1.10	1.65	1.95	2.30	2.10	2.60	2.50	2.70	2.85	3.15	2.85
64	1.75	2.00	2.00	2.15	2.40	2.55	2.20	2.15	2.30	2.50	3.40	3.50
76	1.60	1.80	2.15	2.40	3.10	2.10	2.20	2.30	2.20	2.50	2.15	2.50
81	1.85	1.60	2.15	2.35	2.05	2.70	2.80	3.25	3.10	1.90	2.45	2.90
93	1.35	1.50	2.30	1.95	1.85	2.05	2.25	3.55	2.90	2.85	1.95	Died
MEAN	1.54	1.50	1.99	2.27	2.08	2.33	2.59	2.77	2.90	2.81	2.69	2.84

Ewe No.	2/4	5/4	10/4	17/4	24/4	2/5	7/5	Transferred to good pasture on 7/5	8/5	9/5	11/5	15/5	22/5
33	2.80	3.15	2.50	2.35	2.30	2.75	2.60		1.70	1.35	1.70	1.85	1.60
60	3.10	2.90	2.45	4.05	2.95	2.45	2.75		1.80	1.65	1.40	1.75	1.15
58	2.60	2.40	1.95	2.50	2.40	2.05	2.00		1.60	1.65	1.45	1.90	1.80
82	2.75	2.50	3.15	3.90	2.95	2.90	2.25		1.60	1.90	1.35	2.05	2.00
84	2.95	3.30	2.65	3.05	2.60	2.20	2.25		1.80	1.70	1.95	1.85	1.70
88	2.85	2.40	3.05	2.90	2.80	2.70	2.25		1.85	2.10	1.85	2.20	1.40
92	3.45	2.30	3.20	2.85	3.00	2.35	2.95		2.55	2.25	2.10	2.05	2.15
59	3.05	4.20	3.50	4.05	3.70	3.30	3.50		3.60	3.50	2.00	1.60	1.25
68	3.15	3.40	2.35	2.75	3.15	2.60	3.70		2.55	2.65	2.40	2.45	1.80
69	3.80	3.45	3.65	3.85	3.80	3.60	4.05		2.90	1.70	2.20	2.40	2.35
73	2.30	2.15	2.35	2.25	2.20	1.70	2.05		1.35	1.15	0.95	1.95	1.25
79	2.80	2.80	2.70	2.95	2.25	2.35	2.30		2.15	2.30	1.75	1.95	1.95
87	1.75	2.30	2.30	2.45	2.45	2.75	3.00		1.25	1.60	0.85	1.40	0.75
16	1.55	2.05	1.90	2.05	2.80	2.60	3.05		2.00	1.50	1.60	1.80	2.00
54	2.55	2.10	2.65	3.00	3.20	3.05	3.15		1.90	1.80	1.90	2.05	2.05
64	3.60	3.90	2.40	2.00	2.45	2.05	2.70		1.75	1.90	2.10	2.20	2.10
76	1.00	1.40	1.70	2.00	1.80	1.50	2.15		2.20	2.20	1.55	1.30	1.45
81	2.45	2.20	2.30	2.65	2.95	2.95	1.75		1.60	1.90	1.70	2.30	1.55
93	-	-	-	-	-	-	-						
MEAN	2.69	2.72	2.60	2.87	2.76	2.55	2.69		2.01	1.93	1.71	1.95	1.68

Appendix 3(b) Plasma calcium concentrations of 19 Cheviot ewes on low calcium diet 1962

Ewe No.	20/11	4/12	21/12	4/1	22/1	2/2	15/2	1/3	12/3	19/3	26/3	29/3
33	10.24	10.32	10.40	10.32	10.64	10.48	9.84	9.11	9.35	8.08	7.90	6.72
60	9.03	9.19	9.35	8.95	8.79	8.31	6.77	5.49	6.38	5.88	6.97	5.91
58	9.92	9.76	9.43	9.68	9.60	9.60	8.23	7.10	5.58	7.42	7.66	7.74
82	9.43	9.27	8.95	7.90	9.27	7.82	6.61	5.95	5.41	6.48	9.19	8.71
84	9.76	9.92	9.60	8.63	7.98	7.34	7.09	6.69	5.80	7.13	7.82	8.39
88	9.19	9.35	9.03	8.39	9.19	8.87	7.90	8.31	7.90	7.21	6.80	6.48
92	8.79	8.95	8.87	9.43	9.35	8.23	8.47	8.06	7.02	6.08	7.21	6.80
59	9.68	8.55	10.08	9.84	10.40	9.11	9.92	9.03	9.03	7.90	7.29	7.13
68	9.76	9.35	9.27	9.35	9.68	9.27	8.87	8.95	7.74	6.28	6.48	5.67
69	10.16	9.67	9.52	9.76	8.87	8.71	8.55	9.03	7.98	7.42	8.31	6.24
73	10.00	9.27	9.60	9.11	8.71	9.19	8.95	8.87	7.50	7.66	8.39	8.63
79	9.03	8.79	9.27	8.31	7.80	6.36	5.88	5.89	5.00	4.70	6.64	8.63
87	10.08	10.40	10.24	10.40	10.08	10.16	9.19	8.79	7.18	6.40	6.72	6.80
16	10.24	10.32	10.32	10.00	10.81	9.84	9.48	9.35	9.11	8.47	9.92	9.92
54	10.32	9.84	8.39	8.87	9.43	10.08	9.11	9.48	9.68	8.71	8.71	8.71
64	8.23	7.42	8.06	10.08	8.63	8.23	7.82	8.02	8.63	8.69	7.82	6.64
76	8.47	9.35	9.52	9.27	8.31	8.63	7.23	6.20	5.25	4.86	4.86	5.02
81	9.60	9.76	10.00	9.52	9.92	9.43	8.47	7.50	7.75	7.53	7.21	6.97
93	7.19	7.58	8.06	6.52	7.25	7.98	6.20	6.85	5.33	4.46	4.62	Died
MEAN	9.43	9.32	9.37	9.17	9.20	8.82	8.14	7.83	7.24	6.80	7.39	7.28

Ewe No.	2/4	5/4	10/4	17/4	24/4	2/5	7/5		8/5	9/5	11/5	15/5	22/5
33	7.58	9.19	9.11	9.35	9.23	8.91	8.75		9.56	10.45	10.21	9.27	11.29
60	5.63	6.89	6.40	6.56	7.05	5.83	5.99		6.48	6.97	7.78	7.74	10.56
58	6.36	6.97	7.98	7.50	7.70	7.05	7.29		8.59	8.99	9.88	8.63	10.40
82	8.47	8.55	8.47	6.64	6.08	4.86	5.35		5.83	7.70	10.77	9.27	10.73
84	7.58	6.48	6.80	7.13	6.56	6.56	7.05		7.45	8.10	10.45	9.11	10.32
88	5.75	6.16	7.53	7.50	6.48	6.08	6.08		5.02	5.35	5.83	7.58	10.24
92	6.56	7.21	7.21	8.87	6.56	6.56	6.24		6.64	7.21	8.83	7.90	9.43
59	7.74	6.56	6.97	8.55	7.53	7.37	6.16		8.10	8.99	9.80	9.43	10.43
68	5.31	5.43	7.13	6.97	7.53	7.86	7.13		7.45	8.75	9.48	8.87	11.21
69	6.72	5.02	7.13	7.82	6.32	7.29	7.29		8.42	9.40	9.48	8.87	10.08
73	8.31	7.66	7.82	6.80	8.10	8.91	7.78		8.26	10.37	10.77	8.47	10.40
79	7.58	7.13	6.56	6.32	5.51	7.29	7.61		7.45	7.61	9.07	8.47	10.40
87	8.23	10.08	9.03	10.00	7.86	8.18	7.53		7.29	8.34	10.77	9.76	11.53
16	10.00	10.56	10.48	10.24	8.91	8.90	8.57		9.40	10.94	11.50	9.35	11.45
54	8.95	9.27	9.84	10.08	8.02	7.94	7.13		8.26	8.26	10.21	9.92	10.73
64	6.28	6.72	6.56	8.87	8.42	9.23	7.05		7.61	7.53	8.59	7.74	9.84
76	4.62	5.83	6.24	7.74	6.64	7.10	6.72		7.05	7.13	6.40	10.48	8.71
81	6.97	6.56	7.61	8.39	7.70	8.34	6.80		8.26	9.23	10.13	9.03	10.97
93	-	-	-	-	-	-	-		-	-	-	-	-
MEAN	7.14	7.34	7.71	8.07	7.34	7.50	7.02		7.61	8.40	9.44	8.88	10.48

Transferred to Good Pasture on 7/5

Appendix 3(c) Plasma magnesium concentrations of 8 Cheviot ewes on calcium supplemented indoor diet 1962.

Ewe No.	28/11	4/12	21/12	4/1	22/1	2/2	15/2	1/3	12/3	19/3	26/3	29/3
30	1.65	1.75	1.70	2.55	1.75	2.20	2.30	2.45	2.75	2.70	2.35	2.00
40	1.25	1.20	1.35	2.05	1.50	1.60	1.50	2.15	2.35	2.40	2.70	2.65
53	1.75	1.65	2.30	2.15	1.75	2.05	2.60	2.40	2.45	2.25	2.70	2.25
35	1.30	1.45	1.65	1.65	0.90	1.40	1.85	1.65	2.45	2.40	2.35	2.25
56	1.90	2.05	2.25	1.95	1.50	2.00	2.30	2.30	2.35	2.55	2.35	2.55
63	1.80	1.45	2.10	2.25	2.05	2.10	2.40	2.20	2.55	2.65	2.80	3.50
74	1.50	1.20	1.45	1.55	1.15	1.40	1.70	1.95	2.10	2.15	2.65	2.25
90	1.90	1.75	2.10	2.70	1.95	2.35	2.30	2.65	3.10	2.90	3.50	2.80
MEAN	1.63	1.56	1.86	2.10	1.57	1.88	2.12	2.22	2.51	2.50	2.67	2.53

Ewe No.	2/4	5/4	10/4	17/4	24/4	2/5	7/5		8/5	9/5	11/5	15/5	22/5
30	2.10	2.05	2.30	2.60	2.40	2.15	2.40	Moved to Grazing on 7/5	2.15	2.20	1.70	1.85	1.75
40	2.45	2.55	2.60	2.35	2.35	2.25	1.85		1.60	2.60	1.30	1.80	1.40
53	2.25	2.15	2.60	2.75	2.60	1.65	2.95		2.35	2.45	1.40	1.75	1.85
35	2.80	1.40	1.60	1.80	1.90	1.15	2.10		1.50	2.10	1.75	1.55	1.30
56	3.05	2.40	2.20	2.45	2.15	2.25	2.50		1.95	2.25	2.35	2.05	2.00
63	3.35	3.05	3.05	3.60	2.90	2.95	2.85		2.05	2.20	2.20	2.00	1.85
74	2.25	2.15	1.75	2.20	2.30	2.00	1.95		2.20	1.75	1.90	2.05	2.10
90	2.80	2.90	2.80	2.80	2.65	2.65	2.85		2.55	2.45	2.15	2.10	1.45
MEAN	2.63	2.33	2.36	2.57	2.40	2.13	2.43		2.04	2.25	1.84	1.89	1.71

Appendix 3(a) Plasma calcium concentrations of 8 Cheviot ewes on calcium supplemented indoor diet 1962.

Ewe No.	28/11	4/12	21/12	4/1	22/1	2/2	15/2	1/3	12/3	19/3	26/3	29/3
30	9.68	9.60	9.52	9.68	10.16	9.84	10.00	9.60	9.43	9.67	9.35	9.52
40	10.32	10.24	9.52	10.00	10.00	10.40	10.48	10.24	10.40	9.60	9.76	9.52
53	9.60	9.27	10.00	9.68	9.11	9.92	9.52	9.43	9.11	8.87	9.84	9.68
35	9.35	9.52	9.35	9.52	9.27	9.68	9.68	9.60	9.60	9.11	9.11	9.11
56	9.60	9.27	9.76	10.08	8.95	10.16	10.16	8.39	9.92	9.67	9.52	9.35
63	10.00	10.08	10.24	9.76	9.84	10.32	10.08	10.00	10.48	9.84	10.08	10.32
74	9.43	9.60	9.92	9.52	10.08	10.64	10.48	9.84	10.89	10.08	10.40	10.16
90	9.67	9.35	9.35	8.87	9.60	9.19	9.19	9.03	8.63	7.21	7.66	7.82
MEAN	9.71	9.62	9.71	9.64	9.63	10.02	9.95	9.52	9.81	9.26	9.46	9.43

Ewe No.	2/4	5/4	10/4	17/4	24/4	2/5	7/5		8/5	9/5	11/5	15/5	22/5
30	9.92	10.16	10.16	9.52	8.91	9.48	9.15		9.32	9.23	9.15	8.95	9.84
40	9.68	9.52	9.60	9.27	9.23	9.32	10.53		10.77	10.61	10.50	10.16	11.77
53	9.84	10.24	9.19	9.92	9.32	9.07	9.15		8.18	8.91	10.50	10.08	10.97
35	8.95	9.60	10.00	9.68	9.64	8.99	9.56		9.40	8.91	9.40	8.95	10.48
56	9.03	9.11	9.60	9.92	10.13	10.04	9.96		9.64	9.80	10.13	10.24	10.00
63	10.00	8.87	10.40	8.95	10.53	9.96	9.48		10.04	9.40	10.61	10.61	10.73
74	9.68	10.08	10.00	10.08	10.56	9.88	9.96		9.40	10.29	11.02	9.67	10.00
90	8.87	9.27	8.79	9.11	8.59	8.10	7.94		7.94	8.02	8.75	8.06	10.24
MEAN	9.50	9.61	9.72	9.56	9.61	9.35	9.46		9.33	9.40	10.01	9.52	10.45

Moved to Grazing on 7/5

Appendix 4(a) Plasma magnesium concentrations of two groups of seven lactating ewes at grass at Cochno Farm 1962.

(Experiment 5)

All samples taken around 10 a.m. unless otherwise stated.

Group.	Ewe No.	29/5	1/6	3/6	5/6	7/6	2pm 7/6	8pm 7/6	8/6	10/6	14/6	15/6	17/6	19/6	21/6	23/6
DRENCHED	3	0.54	0.56	0.63	0.48	0.78	1.18	1.36	0.98	0.96	0.73	0.62	0.62	0.69	0.56	0.85
	12	1.02	0.30	0.97	0.85	0.87	1.34	1.07	0.87	0.85	1.26	1.28	1.05	1.18	0.78	1.30
	17	0.97	0.72	1.20	1.12	0.97	1.47	1.33	1.21	1.10	1.32	1.07	1.10	1.44	1.35	1.44
	29	1.68	1.20	0.82	1.33	1.50	1.86	1.65	1.95	1.39	1.66	1.76	1.47	1.62	1.60	1.68
	59	1.15	1.15	0.73	1.09	1.00	1.49	1.31	1.28	0.97	1.18	1.35	0.89	1.45	1.21	1.29
	60	0.72	0.25	0.85	0.59	0.78	1.50	1.00	0.82	0.80	0.57	0.62	0.74	0.71	0.46	0.80
	83	1.38	0.64	0.97	1.07	1.02	1.53	1.28	1.39	1.13	0.98	1.08	1.37	1.24	1.16	1.30
	MEAN	1.07	0.69	0.88	0.93	0.99	1.48	1.29	1.21	1.03	1.10	1.11	1.03	1.19	1.02	1.24
CONTROLS	6	1.06	0.68	1.05	0.81	1.06	-	-	1.00	1.06	0.99	1.02	1.07	1.11	0.99	1.44
	13	1.05	1.04	1.35	1.30	1.23	-	-	1.31	1.16	1.21	1.26	1.03	1.13	1.21	1.34
	31	1.15	1.09	1.25	1.40	1.42	-	-	1.47	1.54	1.46	1.56	1.49	1.60	1.65	1.75
	32	0.95	0.43	0.84	0.81	0.73	-	-	0.92	0.90	0.90	0.86	0.82	0.84	0.90	0.99
	62	1.20	0.74	1.17	1.00	1.23	-	-	1.52	1.10	1.37	1.20	1.08	1.20	1.30	1.30
	73	1.22	0.58	0.50	0.93	0.75	-	-	0.79	0.95	1.04	1.00	0.84	0.87	0.51	1.16
	87	1.06	0.74	0.48	0.89	0.75	-	-	0.59	0.56	0.70	0.95	0.64	0.58	0.66	0.91
	MEAN	1.10	0.76	0.95	1.02	1.02	-	-	1.09	1.04	1.10	1.12	1.00	1.05	1.03	1.27