PRODUCTION AND METABOLISM STUDIES ON BOVINE TRICHOSTRONGYLOSIS

by

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SUMMARY

In this thesis the sequential development of naturally occurring bovine trichostrongylosis in the same animals was studied, for the first time, over a two year period. The species of gastro-intestinal trichostrongyle most frequently present were primarily Ostertagia ostertagi and Cooperia oncophora; a few Trichostrongylus axei were also recorded.

In a control group of 16 calves, overwintered larvae of O. ostertagi and C. oncophora were acquired in the spring of their first grazing season. The eggs resulting from these infections gave rise to another generation of larvae on the pastures by August, which was sufficient in number to cause the type I disease, characterised by loss of appetite, diarrhoea and weight loss. One animal was destroyed in extremis just prior to the type I disease, clinical signs of parasitic bronchitis occurred namely, increased respiratory rate and coughing necessitating treatment with diethylcarbamazine and then levamisole. At housing in October, five calves were slaughtered and the post-mortem worm burdens consisted almost entirely of early 4th stage larvae of O. ostertagi and C. oncophora, which were arrested in development. Following housing in October, the animals were clinically normal during the so-called pre-type II phase, until late February and early March when some reduction in appetite was noted which was followed by clinical diarrhoea and weight loss in April and May in the so called type II disease. During the second grazing season, the animals acquired a solid immunity to C. oncophora and a good immunity to O. ostertagi and this was reflected in the low numbers of eggs in the faeces, relatively low numbers of larvae on the pasture and low worm burdens at final slaughter two months after housing in October.

A second group of 16 calves, which were grazed on immediately adjacent but separate fields, received a sustained release device containing the anthelmintic morantel tartrate, which was introduced by a special dosing gun into the rumen of each calf prior to grazing in the spring of each year. The boli were designed to release the drug over a 90-day period and their introduction prevented the build-up of larval infection on the pasture and the occurrence of the type I and the type II disease. The advantage in liveweight gain alone over the

two year period amounted to a mean of 33 kg over the controls. Furthermore, the introduction of the boli did not interfere with the acquisition of immunity in the second grazing season.

Several biochemical parameters were monitored of which two, serum pepsinogen and gastrin levels, proved particularly interesting. All values of these parameters became markedly elevated when large numbers of parasites were actively developing to the adult stage i.e. during the type I and type II disease, but not during the pre-type II phase when the worm populations consisted mainly of arrested larval stages, or when the animals become immune. The single and combined linear relationship between pepsinogen, gastrin and numbers of developing and adult Ostertagia parasites was very highly significant. Although further information is necessary to define normal bovine plasma gastrin levels and the affecting factors, the results in this study suggest that the evaluation of plasma gastrin could be a useful adjunct to plasma pepsinogen as a combination diagnostic test for ostertagiasis, particularly when pepsinogen values may be only moderately elevated in immune cattle under larval challenge.

Metabolism, production and carcass appraisal studies were made on the control calves and those receiving the morantel bolus at the end of the first grazing, during the pre-type II and type II phases and at final slaughter. For comparative purposes, another group of seven cattle which were left free of trichostrongyle infection by fortnightly anthelmintic therapy and which had also grazed immediately adjacent but separate pasture was included. At slaughter, following the first grazing season, significant differences in the carcass appraisal such as the killing-out percentage, dry matter content and mass of eye muscle of the forerib indicator joint favoured the bolus treated animals.

In the winter housing store period, during/an imposed pair-fed regime where digestion and Nitrogen balance studies were conducted, the bolus treated and clean animals displayed significant superiority over the control cattle in the digestibility of the whole diet dry matter, crude protein and gross energy. Also the nitrogen retention was lowest in the control steers, particularly in the type II phase (which occurred after 17 weeks of housing), where an increased urinary nitrogen excretion was a major contributor. This contrasted with pre-type II findings where increased faecal nitrogen, in the same steers, significantly lowered the apparent crude protein digestibility. This section records the

first reported metabolism studies conducted with cattle experiencing clinical type II ostertagiasis.

Following performance studies conducted during the second grazing and final fattening period indoors, a carcass appraisal was conducted at a modified 'constant-time' slaughter. Significant differences in official grading, several important carcass measurements and indicator rib joint analyses were found between the inferior control cattle and both the clean and the bolus treated steers. Economic appraisal showed there to be a considerable benefit for the bolus treated group compared with the control cattle.

At no stage in the whole experiment was there any evidence of compensatory growth in the cattle which had experienced clinical ostertagiasis.

Finally, in the General Materials and Methods Section of this thesis, is a complete description of the improved metabolism stall, faecal and urine collection systems which were specially developed for this work. The excellent faecal and urine recoveries obtained minimised loss and therefore experimental errors.

LITERATURE REVIEW ON BOVINE TRICHOSTRONGYLOSIS WITH SPECIAL REFERENCE TO OSTERTAGIASIS

It is well recognized that Ostertagia ostertagi is the most important cause of bovine parasitic gastritis in temperate areas of the world.

Although O. ostertagi is the species associated with most of the recorded clinical outbreaks of parasitic gastritis, other members of the genus such as O. bisonis and O. leptospicularis have also been implicated in some instances (Worley and Sharman, 1966; Bissett, 1980; Al Saqur, Armour, Bairden, Dunn and Jennings, 1980). O. ostertagi is commonly called the medium brown stomach worm and, as this name suggests is intermediate in size (up to 1 cm in length) between bovine abomasal parasites of other genera, Haemonchus spp. and Trichostrongylus axei. The latter parasites are more frequently associated with outbreaks of bovine parasitic gastritis in the tropics and sub-tropics respectively.

Life Cycle of O. ostertagi

All Ostertagia species have a direct life-cycle with two distinct phases (see Fig. 1). In the free-living phase, eggs passed in the faeces develop and hatch to become first-stage larvae (L_1) which feed, grow and moult to second stage larvae (L_2). The same process is repeated by L_2 larvae to reach the third and infective stage (L_3). The latter retains the outer sheath of the L_2 and therefore does not feed. It is the most resistant of the free-living stages to adverse climatic conditions. All of this development occurs in the faecal pats and under suitably moist conditions the L_3 then migrate onto the herbage.

The parasitic stage begins after the ingestion of the L_3 with herbage. It exsheaths in the rumen and enters the tubular gastric glands of the abomasum, especially those in the fundic region.

Moulting to the early fourth stage larvae (L_4) takes place by four days post infection, after which larval development usually continues without delay, taking about 10 days to reach the L_5 stage. These grow, and around 18 days from first infection emerge from the

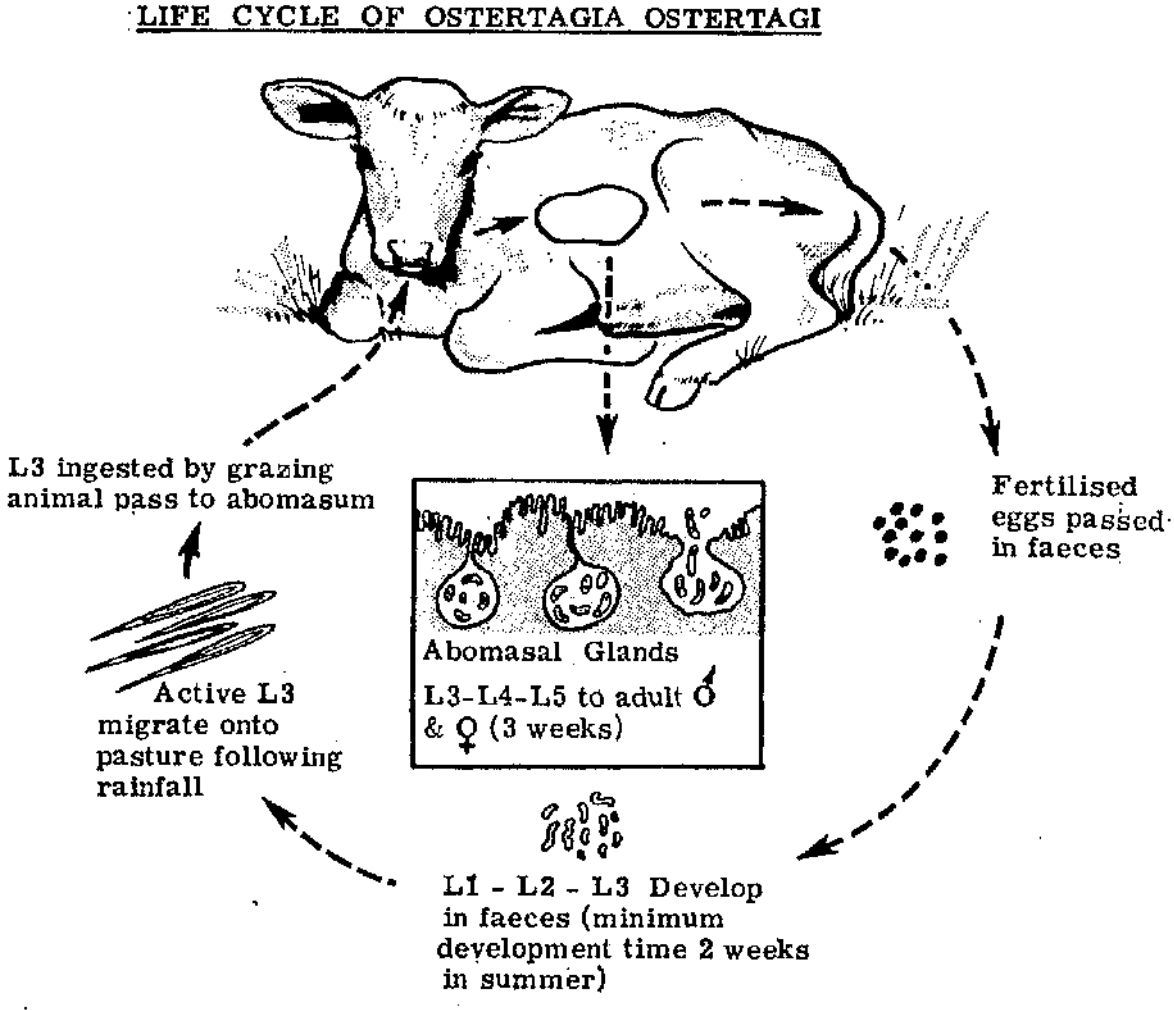
glands onto the surface of the abomasal mucosa to become mature adults.

Another possibility is for the early fourth stage larvae (EL_4) to interrupt their development for several months and then recommence development and maturation. In the Northern Hemisphere this seems to occur primarily in larvae ingested during autumn and winter.

Fertilized adult female worms lay eggs 70-80µm long and 40-50 µm wide, which are passed out in the faeces at the morula stage.

The life cycle is summarised in Fig. 1 below.

Fig. 1



THE CLINICAL DISEASE

At the end of the nineteenth century T.S.Cobbold (1864) produced a significant contribution to the literature in Veterinary Parasitology in Great Britain. He noted that in several "herbivorous animals" a "remarkably numerous" number of nematode parasites existed, and listed those present in "the ox" in a subsequent publication (Cobbold 1873).

The clinical description of the disease was very limited until the beginning of the twentieth century when reports on outbreaks of parasitic gastritis began to appear (Gardener, 1911; Ackert and Muldoon, 1920). Since then, many reports coming from Northern England (Stewart and Crofton, 1941), Southern England (Bruford and Fincham, 1945), Scotland (Martin, Thomas and Urquhart, 1957) and Northern Ireland (Gracey, 1960) have demonstrated the widespread occurrence of the disease. More recent field studies (Anderson, Armour, Jarrett, Jennings, Ritchie and Urquhart, 1965b, Michel, 1969a) have confirmed that Ostertagia parasites are the most important cause of parasitic gastritis of cattle in Britain.

Many reports from all over Europe have also recognised the importance of bovine ostertagiasis. For example in Germany by Burger, Eckert, Wetzel and Michael (1966); in France by Raynaud, Landren and Jolivet, (1971); in Denmark by Henriksen, Jørgensen, Nansen, Serjrsen, Larsen and Klansen, (1975); in the Netherlands by Borgsteed, (1977).

There are also accounts of its importance in countries with a sub-tropical climate provided there is winter rainfall e.g. in Southern Australia (Anderson, 1971), in the USA (Williams and Knox, 1976; Craig, 1979) and in South America in the temperate areas of Argentina (Entrocasso and Steffan, 1980; Steffan, Fiel, Entrocasso, Acuna and Rojas Panelo, 1982).

An important contribution to the understanding of the disease was made by Martin et al (1957) who described outbreaks of what they termed 'atypical parasitic gastritis' in young cattle in Britain which had been housed for several months following their first grazing season. Despite the apparent absence of fresh infection during housing, these animals harboured large worm burdens consisting of mature adults, developing and early fourth stage larvae (EL_4) of O. ostertagi. The authors suggested that the large population of EL_4's in the stomach wall might have accumulated as a result of an immunity

acquired during the grazing season and that they subsequently developed, as immunity waned during the period of non-exposure while the animal was housed.

After this description by Martin et al (1957) a detailed field study of suspected outbreaks of ostertagiasis was carried out in South West Scotland and Northern England by Anderson et al (1965b). As a result of these studies it became apparent that the disease occurred in three phases, two of which are clinically evident, namely the type I and type II disease. The first, type I, was seen in young cattle during the summer, principally between July and October and was characterised by weight loss and profuse green diarrhoea. The type I clinical entity apparently resulted from the rapid acquisition of large numbers of larvae which completed their development to the pathogenic L_5 and adult stages in three to four weeks.

The other phase, pre-type II ostertagiasis, was regarded as being asymptomatic or showing only mild symptoms and preceded the clinical type II disease. At the pre-type II stage the worm population consisted almost entirely of EL_4 stages which were apparently arrested in development and had been acquired as L_3 in the previous autumn. The EL_4 were considered to be relatively non-pathogenic. Much of the data on which this description was based was obtained from examination of the clinically non-infected animals in herds where type II was present.

The clinical type II disease occurred in the late winter or early spring (March to May), usually in cattle housed after their first grazing season but was also seen in cattle which had grazed during the winter. Inappetance and marked loss of weight were typical signs and diarrhoea, which ranged from being intermittant to profuse and prolonged, was present. Sometimes it was difficult to detect the intermittent diarrhoea if the diet was largely dry forage.

Submandibular oedema, moderate anaemia and hypoalbuminaemia were also often seen in type II disease.

The clinical nature of the type II phase was thought to occur when large numbers of the relatively non-pathogenic EL_4 developed synchronously to the pathogenic L_5 and adult stage.

Cases among older cattle are less common but type II outbreaks have been seen in milking cows, predominantly heifers at first calving in New Zealand (Wedderburn, 1970), Canada (Smith and Perreault, 1972) and Scotland (Petrie, Armour and Stevenson, 1984).

In beef cattle two outbreaks from Scotland were described (Selman, Reid, Armour and Jennings, 1976), one in an autumn-calving herd and the other in a spring-calving herd where the clinical biochemical, haematological, parasitological and pathological findings were similar to those characteristic of type II ostertagiasis in immature dairy cattle. In southern temperate environments, type II disease has been seen in a small percentage of animals from two to four years of age, mainly cows calving in the autumn (Hotson, 1967; Anderson, Donald and Waller, 1983).

PATHOGENESIS

STRUCTURAL CHANGES

In general the severity of the damage in ostertagiasis is related to the number of parasites which reach the adult stage within a short period of time and the persistence of these parasites in the abomasum.

If the numbers of larvae accumulate slowly, it allows the animal time to acquire an immunity and to repair and compensate for the damage inflicted by the parasite, a process which is accelerated in older animals.

Following infection with O.ostertagi, white raised, umbilicated nodules develop on the surface of the abomasum. In heavy infections coalescence of nodules occurs giving the appearance of 'Morocco leather'. Hyperaemia and oedema are also usually present in heavy infection. Ackert and Muldoon, (1920) were the first to describe the basic gross lesions which Martin et al (1957) defined in more detail together with the histo-pathological changes in the abomasum of naturally occurring cases of type II disease. They described a thickening and granulation of the fundic mucosa with the pyloric area showing similar, but less severe lesions. Oedema was also noted in the submucosa of the abomasal fold with some hypertrophy in the mucosa. In severe cases, the lymph nodes adjacent to the abomasum were enlarged and hyperplastic. The granulation and the thickening of the mucosa were either due to the presence of nematodes in various different stages of development in the gland lumina, or due to an increase in the length of the fundic gland caused by hyperplasia of the mucous cells which line the gastric pits. This corresponds with the gross description and histopathological lesions later described as type II

ostertagiasis, by Anderson et al (1965,b).

Using experimental infections, Ross (1963) found that repeated infection was necessary to produce the most severe inflamatory lesions in the abomasum with thickening and oedema of the mucosa. The thickening of the mucosa was due to hyperplasia of the mucous cells of the fundic gland but it was noticed that there was also some reduction of the zymogen cell numbers. In further experimental infections carried out by Ross and Dow (1964, 1965) and Ross and Todd (1965) on calves, the findings were similar.

From the result of sequential necropsies of animals infected with fixed larval doses and, in some instances, fitted with abomasal cannulae to collect abomasal secretions, (Ritchie, Anderson, Armour, Jarrett, Jennings and Urquhart 1966; Jennings, Armour, Lawson and Robert, 1966; Murray, Jennings and Armour 1970) it seems that the parasitic part of the life-cycle can be divided into three parts.

The first commences following infection, when the larvae penetrate and develop in the gastric glands. Although there is hypertrophy of the parasitised gland, the lesions are confined to this gland and the mature differentiated secretory cells of the gland, particularly the parietal cells are replaced by undifferentiated ones. At this time there are no significant alterations in the biochemical values in the blood or abomasal fluid nor clinical signs (analogous with the pre-type II phase in the field). The second phase begins with the development of the L_4 to the L_5 stage as the latter emerge from the gastric gland around 18 days post-infection. This phase is marked by a loss of differentiation in the cells lining not only the parasitised glands, but also in those of the surrounding glands. In particular, the HCl producing parietal cells are replaced by rapidly dividing undifferentiated cells. In heavy infection this results in a rise in pH of the abomasal contents, from the normal 2.5 to 7.

From electron microscopy of the lesion it became apparent that in the parietal cells there is a loss of intracellular canaliculi and a decrease in the number of microvilli, elements necessary for the secretion of HCl. An increase in vacuoles in the cytoplasm of the parietal cells with the loss of mitochondria was also reported by Murray et al. (1970). It also became apparent that separation of the plasmalemmata of the zona occludens which seals cell junctions had occurred, and electron-dense protein material was observed lying between the parietal cell junction (Murray, 1969; Murray et al.,

1970). These authors suggested that the separated cell junction facilitated the passage of protein macromolecules through the epithelial sheet as well as electrolytes. Pepsinogen produced by zymogen cells is not converted to pepsin due to the lack of HCl and may leak into the circulation, giving detectably elevated levels.

Although this theory of macromolecules flowing in and out of the epithelial sheet has been generally accepted, Stringfellow and Madden (1979) have challenged its validity. In studies involving experimental infection of calves with 250,000 larvae of O. ostertagi they recorded high levels of plasma pepsinogen, but postulated that the zymogen cells release the pepsinogen directly into the circulation, rather than taking it up from the gastric contents through a damaged mucosa.

The gastro-enteric loss of protein and electrolytes may lead to retention of fluid and development of oedema due to the loss in osmotic and hydrostatic pressure of the blood. Such a change in the ratio of fluid and body solid was described by Halliday, Dalton, Anderson, and Mulligan (1965).

The functional consequences in this phase of heavy Ostertagia infection may therefore be summarised as:

i) Abomasal disfunction due to lack of HCl production, with reduced digestive efficiency.

ii) Diarrhoea: the cause of this is not well known but the loss of protein and electrolytes may be involved. Another possible cause is the increase in bacterial population which occurs when the bacteriostatic effect in the stomach is reduced with an elevation in pH.

iii) Loss of appetite: the reason for this is unclear but hormonal and neural reflexes are known to exert important influences.

The third phase is dominated by the regeneration of the abomasal mucosa accompanied by elimination of the adult worm burden and restoration of normal pH levels, with the gradual return of the parietal and zymogen cells in those areas formerly occupied by undifferentiated cells. At this time a marked increase in immunoglobulin-producing cells in the lamina propia of the abomasum also occurs (Murray et al,1970; Eckert, Burger, Kongismann, Chevalier and Rahman, 1968)

Provided there is no re-infection, diarrhoea ceases, the animal begins to recover its appetite and starts to gain weight again.

Although the pH of the abomasum fluid will return to normal fairly quickly, plasma pepsinogen levels take a considerable time to return to pre-infection levels and in field situations, where animals are continuously exposed, they seldom return to those levels.

PHYSIOLOGICAL CHANGES

Far less is known about the pathophysiological changes caused by helminth infections, including Ostertagia spp., in cattle than in sheep. This is largely due to a scarcity of reported cattle experimentation in this field, most probably owing to the high cost of bovine animals and the considerable attendant difficulties in managing comparatively large ruminant animals in metabolism stalls.

The main features of derangement of normal function with parasitological infection in the host include changes in appetite, nitrogen, mineral and water balance, together with other effects upon blood biochemistry and hormone status.

Despite the lack of work on bovine ostertagiasis, it is appropriate here to consider each of the affected functions in more detail and refer to work with related trichostrongyle in other hosts, particularly sheep.

Appetite

It is generally accepted and recorded that reduced food intake is associated with many of the more commonly occurring nematode infections of both sheep and cattle. In sheep, the abomasal parasite O. circumcincta had a lesser effect upon food intake reduction than the intesinal nematode T. colubriformis. Thus reductions of 6-20% were recorded in sheep infected with O. circumcincta where larval intakes ranged from 1,000-17,000/day (Sykes and Coop, 1977; Coop, Sykes and Angus, 1982; Symons, Steel and Jones, 1981 a), and from 9-56% in cases of intestinal parasitism as a result of a daily larval intake of T. colubriformis ranging from 300-3,000 (Sykes and Coop, 1976; Steel, Symons and Jones, 1980). Experimental infection produced by a single large dose of O. circumcincta larvae in sheep also gave a considerable reduction in voluntary feed intake (Holmes and McLean, 1971).

The appetite loss is often temporary and after some weeks may return to normal, (e.g. Steel et al. 1980; Symons et al. 1981; Coop et al, 1982)

With the increased experimental use of pair-feeding i.e. offering to uninfected control animals the same quantity of food consumed by the infected animal, it was often demonstrated that reduced feed intake per se was one major contributor to impaired production. In general, the magnitude of the reduction is related to the severity of infection

and it seems there is a threshold level of exposure below which there is no significant depression of appetite (Steel, 1978; Symons et al, 1981a).

The cause of the reduction in food intake remains unclear. It is suggested that localised damage occurs in areas which could affect receptors concerned with monitoring tension, changes in digesta content, pH, gut motility and gut hormones, and so account for the decline in food intake.

The gastrointestinal hormone, cholecystokinin (CCK), is known to be one of the factors involved in feeding behaviour. This has been demonstrated in sheep in which the injection of CCK was followed by a decrease in feed intake (Della-Fera and Baile,1979). Since then, Symons and Hennessy (1981) have demonstrated an increase in CCK concentration in sheep infected with T. colubriformis. The rise in CCK was followed by a drop in food consumption but both returned to pre-infection levels four to six days after administration of an anthelmintic to remove the worms.

Another associated feature is a decrease in hydrochloric acid (HCl) production which could produce some delay in abomasal flow, since acidity is a potent stimulus for reticulorumen contraction (Argenzio, 1980) and could also affect the feed intake.

However, until now it has not been possible to give comprehensive reasons for the reduction in food intake and it seems that the interaction of several factors are involved.

Nitrogen Metabolism

Although reduced appetite has a pronounced effect of the nitrogen metabolism of the host, studies with pair-fed sheep have demonstrated that other factors are important in altering the nitrogen economy. (Sykes,1982).

The structural changes created by nematode infection may lead to local alterations in gastrointestinal function in relation to digestion and absorption. However, an impaired digestion of nitrogen in the proximal gastrointestinal tract, such as the abomasum infected with Ostertagia spp., may not necessarily be reflected in an altered faecal nitrogen excretion, if compensatory increases in digestion and absorption occur at sites distal to the lesion (Steel and Symons, 1982).

Digestibility and Nitrogen Retention

Digestibility studies alone, particularly for the crude protein

fraction, fail to give a complete picture of the host nitrogen economy since some proximal non-digested protein could be degraded by bacteria to ammonia in the large intestine, which in turn is converted into urea by the liver. Part of this urea will be transported to the rumen by the saliva and by direct transfer across the rumen wall and some will be excreted in the urine. Nitrogen balance studies help to achieve a better understanding of the overall input-output nitrogen economy of the host.

Both digestive efficiency and nitrogen retention studies have been used to investigate the gross effect of parasite infection in the ruminant.

Early reports showed that the digestibility of the crude protein (CP) fraction of the diet was apparently reduced following nematode infection in sheep (Stewart, 1932/1933; Spedding, 1954; Horak and Clark, 1964). However, these results have only a limited value because a pair-feeding system with uninfected animals was not used.

To properly evaluate factors such as weight gain, feed conversion efficiency , N retention and digestive efficiences, the use of pair-fed control animals is an essential part of the experimental design.

Contradictory results are reported about the adverse effect on digestibility of several different types of parasitism. In infections with Cooperia curticei in sheep, Andrews (1938) found no effect on the digestibility coefficients or apparent absorption of the various components of the feed. With T. colubriformis in sheep, Andrews, Kauffman and Davis (1944); Roseby (1973) and Sykes and Coop (1976) found similar results. But in Roseby's work the nitrogen retention of infected lambs was significantly lower because of increased urinary nitrogen excretion.

In studies with the ovine abomasal parasite O. circumcincta, Parkins, Holmes and Bremner, (1973), using varying levels of experimental infection, found a reduction in crude protein (CP) digestibility when sheep were infected with a single dose of one million larvae and with half a million larvae (50,000 L_3 given on each of 10 consecutive days). The N balance study showed the infected sheep to be in marked negative retention due to an increased urinary N output, particularly in the acute period of marked anorexia.

Sykes and Coop (1977) also working with O. circumcincta gave lambs a daily dose of 4,000 larvae and reported a reduction in the CP digestibility and also a reduced N retention. This however, was due to increased faecal nitrogen, with the urinary N output being similar in

both infected and control animals.

In bovine ostertagiasis, Canale, Valente, Dotta and Balbo (1977) reported no changes in digestibility in calves infected with 250,000 L_3 O. ostertagi. However, the infected animals required nearly 3 times more food to produce 1 kg of body weight than the non-infected animals.

Depression in feed intake, N retention and nutrient digestibility in the third week after infection with about 600,000 mixed larvae of O. ostertagi and C. oncophora was found by Randall and Gibbs (1981). They also reported that in a subclinical infection (60,000 L_3), both N absorption and energy balance were significantly depressed two to three weeks after infection, with an improvement about five weeks post-infection in the heavily infected animals.

Parkins, Bairden and Armour (1982 a,b) studied the subsequent effect of anthelmintic treatment on animals which had acquired a natural infection of O. ostertagi. In the first study young cattle which had developed type I ostertagiasis in August were treated with thiabendazole (Tbz) at housing in October. Performance, digestibility and nitrogen balance studies followed and results were compared with those of parasite-free control animals. The diet of both groups contained an excess (125%) of protein, but not of metabolisable energy (ME) over that recommended for a growth of 0.6 kg per day. There was a marked lag in growth for the first 6 weeks of housing for the Tbz dosed calves; thereafter the growth rates were similar. Nitrogen (N) balance and digestibility studies conducted at intervals over a 17-week period following treatment showed little difference in the digestibility of the diet but an overall significantly greater N retention in the parasite-free calves. Daily water output in both faeces and urine was greater in the Tbz treated calves. Total serum protein was lower but plasma urea concentration was higher in Tbz treated calves.

It was speculated that only a short-term impairment of post absorptive metabolism may have occurred in the Tbz treated calves and that the high protein intake may have reduced the severity of the lowered N balance observed.

In a second experiment, (Parkins et al. 1982 b), the performance of young cattle subjected to different anthelmintic control programmes during their first grazing season (May - October) were compared during the subsequent 20 weeks of winter housing (October - February). One group of calves (group A) were given no anthelmintic treatment and developed type I ostertagiasis in August and were still showing

clinical signs of the disease at housing in October. A second group was given the anthelmintic fenbendazole (Fbz) at 3 and 6 weeks after turnout to grazing in May and this effectively limited the pasture contamination to such a degree that the animals became only lightly infected; they were also treated again at housing to remove the residual infection. The third group (C) were treated with fenbendazole every fortnight and were effectively parasite-free controls.

The diet given to the cattle in the winter period contained sufficient ME but only about 60% of the recommended crude protein to sustain a daily growth rate of 0.5 kg. Growth rates were only 50% of that expected on a basis of ME input with untreated calves (A) being poorest. Water intake and output were also greatest in group A . There were no significant differences in digestibility between groups although group A had the lowest apparent crude protein digestibility. N balance was always lowest in group A, and highest in group C, but group B demonstrated a marked increase in N retention with time after housing. Differences in N retention were largely accounted for by increased urinary N excretion. It was concluded that under conditions of sub-optimal protein intake, nematode infections in the growing calf can markedly affect production even after efficient anthelmintic treatment.

This long recovery effect after treatment in parasitised animals was also reported by Jones (1983) in lambs chronically infected with T. colubriformis. He found that after 5 and 10 weeks of treatment some enzymes such as alkaline phosphatase were lower than in non-infected controls. Although morphological integrity was completely restored, recovery of the full functional capability of the intestinal mucosa appeared to take longer.

The actual physical location of the parasite seems to be important in its pathological consequences, perhaps because, where the parasites are more distally sited, there is less opportunity for the host to compensate in digestion and absorption from the damaged area. With abomasal parasitism in sheep, Symons et al (1981 a) found that infection with 120,000 L_3 O. circumcinta was required to produce an increase in gastroenteric plasma loss of 120 ml/day during the 4th week. The values return to normal at about the 8th week. The liveweight gain was significantly depressed only during the first 4 weeks following infection and a decreased wool growth rate continued until the 12th week.

However, doses of only 30,000 L_3 of T. colubriformis given

weekly were needed to produce losses of about 150 ml/day of gastroenteric plasma until the 12th week of infection with a marked depression in rates of liveweight gain and wool growth,(Steel et al, 1980). Intestinal trichostrongylosis thus produced a greater plasma protein loss into the gut and lasted longer than abomasal parasitism with Ostertagia.

In large intestinal parsitism with Oesophagostomum radiatum, Bremner, (1982) reviewed several experiments in bovines where the animals showed severe pathological consequences of this parasite due to increases in plasma protein loss during the period of exudative enteropathy (Bremner,1969).

Digestibility and N balance studies still do not give a complete picture of what really happens in different parts of the digestive tract with respect to digestion and absorption of protein. Measurement of the movements of digesta nitrogen through the gastrointestinal tract made by Steel (1978) does not support the idea that increases in faecal nitrogen output in ostertagiasis is due to a decrease of protein digestion as suggested by Sykes and Coop (1977) and Symons et al (1981 a). Thus, using animals cannulated in different parts of the digestive tract and infected with 60,000 O. circumcinta larvae weekly he showed that approximately 80% of the non-ammonia nitrogen (NAN) at the pylorus, which was concluded to be endogenous in origin, was apparently digested and absorbed by the small intestine. By comparison, in the non-infected controls, 59% of abomasal NAN disappeared between the pylorus and terminal ileum. The authors concluded, that, in infections (with Ostertagia) regarded as moderate to high in grazing animals, the capacity to digest protein is not impaired. The infected animals are apparently more efficient at reabsorbing endogenous protein from the small intestine than pair-fed, non-infected sheep. If there is an increase in faecal nitrogen output this may therefore represent only a small proportion of the total endogenous protein loss which has escaped digestion in the small and large intestine.

Endogenous Protein Loss

An important contribution to endogenous protein loss comes from the leakage of plasma into the alimentary tract. The elevated gastroenteric plasma loss in ostertagiasis has been reported by Mulligan, Dalton and Anderson (1963) in clinically affected cattle.

This was studied by injecting the cattle with the polymer polyvinyl pyrolidone (PVP) labelled with I^{131} and measuring its loss

in the faeces. A higher proportion of the polymer appeared in the faeces of clinically affected animals than in the faeces of uninfected controls. The half-life of the albumin was shortened in clinical cases of ostertagiasis.

In type II ostertagiasis, hypercatabolism of the albumin, which leads to hypoalbuminaemia was reported by Mulligan, Maclean, Halliday and Dalton (1967) and Halliday, Mulligan and Dalton (1968). In both studies it was suggested that the hypercatabolism was due to abnormal leakage of plasma protein into the gastrointestinal tract.

As mentioned previously, one mechanism responsible for the loss of protein is thought to be related to a mucosal reaction to the presence of the parasite, in which the cells lining the gastric glands, both infected and uninfected, became replaced by rapidly multiplying undifferentiated cells. The rapid turnover of cells with inadequate time to permit the sealing of cell junctions, allows the passage of protein molecules in and out of the epithelial sheet. This was clearly described by Murray (1969) and Murray et al. (1970).

One effect of this change in the permeability of the gastric mucosa is the enhanced passage of pepsinogen molecules, produced by the zymogen cells, into the circulation. Thus, when the pH of the abomasum, and in particular the lumen of the gastric gland, becomes elevated and non-acidic (as in heavy Ostertagia infection) then pepsinogen is not converted to pepsin and leaks into the circulation via the open cell junction (Jennings et al, 1966; Murray et al, 1970).

It is also possible that enhanced leakage of pepsinogen may occur when the permeability of the mucosa is increased due to a hypersensitivity reaction in immune animals. (Armour, Bairden, Duncan, Jennings and Parkins 1979). Nevertheless, elevated plasma pepsinogen levels have proved a useful guide to the diagnosis of ostertagiasis (Anderson et al. 1965 b; Brunsdon, 1971).

Another possible endogenous source of protein loss could be the increase in mucous production and the proliferation of goblet cells at the site of infection observed in ostertagiasis (Armour et al, 1966; Murray et al, 1970). However, there are no quantitative estimations of mucin release in parasitised farm animals and its contribution to endogenous nitrogen loss cannot be precisely assessed.

The functional significance of mucous release is also incompletely understood, but it has been suggested that antibodies entrapped in mucous may be an important factor in acquired resistance to some

nematodes, i.e. Oe.columbianum, (Dobson, 1967) and H. contortus, (Smith, 1977).

Of particular significance is recent work by Lee and Ogilvie (1980) who found that exsheathed larvea of Trichinella spiralis injected intra-duodenally into immune rats, are unable to penetrate the mucous barrier and are rapidly expelled. Similar results were obtained by Miller, Huntley and Dawson, (1981) with Nippostrongylus brasiliensis in rats. The authors also reported an increase of mucus synthesis and number of goblet cells.

Recently Douch, Harrison, Buchanan and Greer, (1983) described an in vitro bioassay of sheep which demonstrated the inhibition of nematode larval migration activity. They found that mucus from the gastrointestinal tract of sheep resistant to nematode infection inhibited larval migration of T. colubriformis by up to 93%, whereas mucus from heavily infected sheep, reared helminth free, did not significantly inhibit larval migration.

These results indicate that the mucus may play a primary role in preventing the establishment of challenge infections in the gut of the immune animal, but this may well represent an increase in protein loss and a cost in energy.

In intestinal parasitism an elevated rate of release of mucosal epithelial cells would contribute to the increased endogenous nitrogen loss (Symons, 1969), which might explain the typical villous atrophy of the infected intestine. Increased turnover and loss into the gut of intestinal epithelial cells occurs in rats infested with Nippostrongylus brasiliensis (Symons, 1965). However, there are no estimates of the magnitude of this cell proliferation and desquamation in ruminants. With both T. colubriformis and T. vitrinus infection of lambs, the elongated crypts contained increased mitotic figures (Coop and Angus,1975; Coop, Angus and Sykes, 1979) and are thus indicative of an increased cell proliferation. Symons (1978) found an increased mitotic rate of intestinal crypt cells in areas distal to the infection, demonstrating that a faster rate of epithelial cell turnover may not be confined solely to the infected region of the gut. He also suggested that the cause may be due either to the passage down the tract of "toxic products" released from the proximally damaged intestine, or from the parasite, or as part of some immune response of the host.

Gastrointestinal Tissue and Skeletal Muscle Synthesis

To date there has been no published study on the effects of O. ostertagi infection on protein deposition in cattle, nor has there been a study on carcass evaluation at slaughter. However, in lambs chronically infected with O. circumcincta Sykes and Coop (1977), Sykes, Coop and Angus (1977) and Coop, Sykes and Angus (1982) demonstrated a significant reduction in the deposition of both carcass fat and protein, at subsequent slaughter. The deposition of muscle was principally depressed by the reduced feed intake but fat and therefore energy deposition was also affected by a reduction in the efficiency of utilisation of metabolisable energy. These changes in muscle protein deposition and fat deposition were much more marked following infections with the intestinal nematode T. colubriformis (Sykes and Coop, 1976)

In a study with acute intestinal trichostrongylosis in guinea-pigs, Symons and Jones (1981, 1983) described the effect of infection on the partition of amino acid utilisation for protein synthesis. They measured the distribution of radioactivity in the organs and tissue following injections of ^{14}C-L-Leucine. The level of radioactivity (which reflected synthesis) was higher in the stomach, small intestine, caecum, large intestine and liver of infected animals than those same areas in the non-infected controls. These increases were not due to anorexia because they did not occur in the control group which was pair-fed with the infected group. The greater deposition of protein in the liver and gut was apparently at the expense of the skeletal muscle.

A similar experiment, in lambs infected with T.colubriformis, (Jones and Symons, 1982), measured the fractional synthesis rate (FSR) of albumin and liver protein. The amount of liver protein synthesised per day was increased by infection but a depression was found in both skeletal muscle and kidney.

Symons, Steel and Jones (1981 b) summarised the sequential events in parasitised animals where initially, enteric losses of protein are counterbalanced by an early increase in the synthesis of albumin by the liver. However, the rate of synthesis eventually declines and further losses lead to hypoalbuminaemia. Both synthesis and catabolism of protein in the muscle are decreased during parasitism but nitrogen is

lost from host tissue because the rate of synthesis is lower than that of catabolism. A large part of the nitrogen mobilised from muscle protein seems to be excreted by the kidney. The net result is a redistribution of amino nitrogen from the muscle, liver and possibly skin to the intestine which decreases the availability for growth and lean tissue production.

Also, in trichostrongylosis of the guinea-pig, growth of muscle in the very young animal may be permanently affected if infection reduces the amount of DNA (i.e. the number of nucleii) and hence the number of muscle fibres that may develop. If this occurs before the potential number has developed, then the muscle will be permanently reduced in size (Symons and Jones, 1972; Symons et al, 1981 b). This interrelation between gastointestinal tissue and skeletal muscle synthesis might have a direct effect on carcass quality.

Changes in Plasma Protein

Plasma albumin concentration is usually depressed in gastro-intestinal nematode infection but globulin concentrations are elevated so making the total plasma protein content relatively constant. However, in heavy infections the reduction in albumin is eventually accompanied by a drop in globulin resulting in an overall hypoproteinaemia. One of the first reports of a hypoproteinaemia in bovine ostertagiasis was that of Martin et al (1957); since then, several workers have reported this finding in natural as well as in artificially infected animals (Anderson et al, 1965 b; Mahrt, Hammond and Miner, 1964; Ross and Todd, 1965). Mulligan et al (1963) first presented evidence , using I^{131}-labelled albumin, that a higher proportion of labelled albumin was present in faeces of animals with clinical symptoms of ostertagiasis than unaffected animals. Similar results were obtained by Neilsen (1966) and Halliday et al (1968) in type II ostertagiasis and from these findings it became clear that bovine ostertagiasis is a true protein-losing gastropathy. Also, in sheep infected with O. circumcincta, Holmes and MacLean (1971) and Yakoob, Holmes, Parkins and Armour (1983a) obtained similar results.

Neilsen (1966) demonstrated that both albumin and immunoglobin were affected. He found that when the animals were studied during a period with clinical symptoms, such as diarrhoea, both the albumin and the immoglobulin turnover rates were greatly accelerated, whereas animals studied during either a quiescent period or during convalescense had normal (or even sub-normal) turnover rates. This is

summarised below.

Effect of clinical status upon metabolism of ^{131}I-albumin and ^{125}I-immunoglobin G in chronic ostertagiasis. The immunoglobin preparation contained both IgG1 and IgG2. (After Nielsen,1966).

Diagnosis	Status	Fractional catabolic rate (%/day)		Plasma half life (days)	
Ostertagiasis	Diarrhoeic	13.8	12.4	10.9	8.0
Ostertagiasis	Quiescent	5.6	5.8	22.0	19.0
Control	Normal	8.5	8.3	20.4	14.7

^{131}I-albumin metabolism in 7 calves with chronic ostertagiasis (type II ostertagiasis). Note difference in fractional catabolic rates and plasma half lives between diarrhoeic animals. (After Nielsen, 1966).

Animal	Clinical State	Total plasma protein g/l	Plasma albumin g/l	Fractional catabolic rate, per cent/day	Plasma Half life, days
1	Diarrhoea	57	12.9	15.5	11.0
2	"	53	20.4	13.8	10.9
3	"	51	19.4	17.6	8.0
4	"	40	8.1	9.9	11.5
5	"	52	14.8	11.0	10.0
6	Quiescent	50	14.8	6.5	20.1
7	"	44	13.9	5.6	22.0
Eight normal controls (mean)		74	28.4	8.2	20.6

The use of radioisotopes to study protein metabolism demonstrated that plasma protein is in a state of dynamic equilibrium between the intravascular and extravascular pools due to a flux of synthesis and catabolism. Parasitic infection affects this balanced relationship and the degree of this change depends upon the number of parasites and the relative proportions of the different developmental stages.

Young animals are usually more affected, but Yakoob, Holmes and Armour (1983 b) also demonstrated in mature sheep, naturally infected

with O. circumcinta at grazing and subsequently experimentally challenged with larvae of the same species, an increase in the albumin fractional catabolic rate of about twice that of animals left unchallenged. The authors suggested that the aetiology of this enteric plasma loss may be associated with an increase in permeability induced by an immune hypersensitivity reaction.

The immune reaction possibly contributes to a rise in plasma globulin coincident with the decline in albumin owing to losses into the gut. The elevated rate of synthesis may be speculated to result in a proportionately greater utilisation of available amino acid for replacement synthesis and a concomitant diversion from other synthetic sites such as muscle cells.

Water Metabolism Changes

Diarrhoea is commonly seen in parasitic infection. It has been demonstrated in experimental Ostertagia infections that the onset of diarrhoea occurs at about the time when the parasites have completed the L_5 larval stage and are developing into adults and reach their greatest severity during the maturation of adult worms (Ritchie et al, 1966; Jennings et al, 1966; Murray, 1970).

The presence of diarrhoea is a clinical sign of importance because it indicates a major disturbance in the regulation of body fluids which can rapidly lead to death. Water and electrolyte balance is known to be regulated by a number of hormones including aldosterone, anti-diuretic hormone and the renin-angiotensin system. Investigations using tritiated water (Halliday et al, 1965) demonstrated that in type II ostertagiasis the total body solid content was higher in non-infected animals compared with those which had acquired the disease naturally, despite the diarrhoea. In a similar study Baker, Black, Anand and Fisk (1965) reported a similar finding in yearling Hereford cattle with a mixed infection of O. ostertagi, T. axei and Cooperia spp.. Parkins et al (1982 b) reported that animals, which had been kept parasite-free by fortnightly anthelmintic treatment at grass, retained less water in the subsequent winter period than untreated animals or those which had been treated routinely three and six weeks after going to grass (and again at housing). Water retention by parasitised animals clearly indicates that the full extent of tissue loss attributable to parasitic infection cannot be measured by changes in body weight alone. This water retention could also severly affect

the carcass quality (watery carcass) thus resulting in severe economic consequences.

Mineral Metabolism and Skeletal Growth Changes

Very little information is available for bovines about mineral metabolism in parasite-infected animals. Waymark and Torbert (1969 a,b) showed the faecal and urinary excretion of calcium (Ca), phosphorus (P) and magnesium (Mg) to be increased in calves infected with Ostertagia spp.. In sheep, Horak and Clark (1964), noticed a sharp decrease in plasma inorganic phosphate in animals infected with O. circumcinta. More recently it has been demonstrated that chronic, but subclinical, abomasal parasitism with O. circumcinta in sheep significantly reduced the growth of the skeleton. Sykes et al (1977) found that deposition of bone minerals (Ca and P) in infected lambs eating ad libitum was only 35% of that of uninfected lambs. This was confirmed by chemical analysis of individual bones which demonstrated a comparable reduction in bone ash deposition. The bone size was also reduced and this, in part, could be attributed to the reduction in food intake since the bones of pair-fed animals were smaller than those of non-infected animals which had no food reduction. Deficiency in energy or protein is also likely to be involved in this process. However, recent data of Wilson and Field (1983) reported that Ostertagia infection had an effect on P secretion into, or absorption from, the digestive tract in sheep without symptoms of the disease. The endogenous Ca excretion was apparently higher in the infected animals than in the control, but it is not clear whether the efficiency of absorbtion was reduced or the action of the worm increased the leakage of Ca through the gastric mucosa. The same experiment compared results obtained from intestinal parasitism with T. colubriformis and found more severe effects upon the absorption of P and Ca from the gut. This data agrees with previous reports by Sykes, Coop and Angus (1975) and Sykes and Coop (1977).

The most probable cause of the poor mineralisation of bone in O. circumcinta infection seems to be the increased protein loss into the gut. The greater effect of T. colubriformis on bone mineralisation is caused by a combined nitrogen and phosphorus effect. It is also suspected that specific hormonal changes in the host may also be involved. Hormones, such as thyroxin, insulin and corticosteroids may be playing a role in parasitised animals (Pritchard, Hennesey and

Griffiths, 1974; Sykes et al, 1975).

Gut Motility and Digesta Flow

Little information is available concerning the effects of abomasal parasitism on gut motility or digesta flow despite its obvious importance in digestive efficiency. Changes in tonus or frequency of gastrointestinal movements may indirectly affect the digestion and absorbtion of food by changing the rate of digesta flow in the alimentary tract. It is interesting that Martin et al (1957) reported that the ruminal movements of cattle suffering from parasitic gastritis were weaker and of a shorter duration than those of uninfected animals. In sheep infected with O. circumcincta, Horak and Clark (1964) found an increased retention time of ingesta in the rumen that could explain the increased crude fibre digestibility observed in similarly infected animals described by Parkins et al (1973).

In sheep infected with 5000 L_3 O. circumcincta per day over an 8 week period, Wilson and Field (1983), reported an increase in the flow of water and dry matter in the abomasum of infected animals compared with pair-fed non-infected controls.

Changes in the electrical activity of the gut wall in sheep infected with T. axei and Chabertia ovina were reported by Bueno, Dorchies and Ruckesbuch (1975). They found a drop of 40-60% in the frequency of reticular and abomasal bulb contraction during a two day period before the onset of diarrhoea. This remained depressed during the two-day diarrhoeic episode and then returned rapidly to normal. In the jejunum the flow was more rapid due to almost continuous irregular spiking activity, instead of the normal, regular spiking of the migrating myoelectric complex.

The control of gut motility seems to be a very complicated system in which mechanical and chemical receptors are involved with different gastrointestinal hormones that may effect different actions in different parts of the digestive tract.

Hormonal Changes

In both type I and type II ostertagiasis, changes in gastric pH, generated by loss of full HCl production, and the increase in plasma pepsinogen have been very well documented during the last 20 years (Armour and Osbourne, 1982). These biochemical changes in

ostertagiasis have recently stimulated renewed interest in the interaction of gut parasites and gut hormones, with the development of radioimmunoassay techniques to study gastrointestinal hormones.

Gastrointestinal hormones influence the exocrine and endocrine secretion of the gut, gut motility, absorbtion and permeability. Some have trophic influences on the gastrointestinal tract and others have been implicated in the regulation of feed intake. The hormones may interact or modify the action of another or alter the sensitivity of a target organ. Changes in cholecystokinin (CCK) levels and its relation with feeding behaviour have already been described. Changes in plasma gastrin levels have also been noted in parasitised animals, principally those with abomasal parasites. Gastrin is a peptide hormone present in relatively high concentration in the mucosa of the pyloric antrum and with lower concentrations present in the duodenal mucosa. It is produced and stored in gastrin (G) cells. Its mode of action is in stimulating secretion of HCl by the parietal cells, gastrointestinal motility and absorption (Gregory, 1974) as well as having trophic effects on the gastrointestinal mucosa (Lankisch, 1980; Wright 1981). In a normal physiological situation, gastrin contributes importantly in the ruminant to the regulation of the volume of HCl secreted by the abomasum according to the quantity and quality of food being ingested. The secretion of gastrin is influenced by the vagus nerve and variations in abomasal pH also produce modifications in gastrin secretion; i.e. lower (more acid) pH causing a reduction, higher pH an increase.

McLeay, Anderson, Bingley and Titchen (1973) carried out a series of elegant experiments in sheep infected with O. circumcincta after an abomasal pouch had been created, which was physically separated from the remainder of the abomasum. They found that the secretory activity of the abomasal fundic pouches, not exposed to the parasite O. circumcincta was maintained, or even increased in its secretory activity when the infected abomasum showed an elevated pH and an increase in the sodium concentration. Ultrastructural studies demonstrated that the parietal cells of the pouches had the appearance of cells subjected to strong secretory stimuli, whereas those of infected areas were similar to gastric mucosa cells exposed to a secretion suppressing agent. The authors postulated two different mechanisms. One suggested a factor to be released from either the parasite, or from the host itself which could inhibit parietal cell

function directly. The other suggested that an increase in circulating levels of gastrin accounted for the increased secretion of HCl. Results from a similar experiment by Anderson, Hansky and Titchen (1976, 1981) suggested that the parasite itself was responsible for the increase in gastrin, since this was detected prior to any increase in the pH of abomasal contents. In reinfected sheep however, there was an increase in gastrin activity without the abomasal pH rising above 4.

Adult parasites appear to have more effect in the promotion of hypergastrinaemia than the larval stages of O. circumcincta because the most marked increase in gastrin happens 11-20 days after infection when adult worms would be expected to be present (Anderson et al, 1981). Increased plasma gastrin was seen before major changes in abomasal pH with the presence of adult parasites. Only later were higher values associated with a decrease in gastric acidity (Anderson et al 1981).

No less important a role would appear to be the trophic action of gastrin as a modulator of cell proliferation in the gut. For example, after administering pentagastrin for 21 days to rats, Crean, Marshall and Rumsey (1969) reported parietal cell hyperplasia and upper duodenal tissue growth stimulation as measured by protein and RNA synthesis (Johnson, Aures and Yuen, 1969). Casteleyn, Dubrasquet and Willems (1977) found a similar effect with gastrin in pigs. In man it is well recognised that gastrin has trophic effects on the stomach and small intestine as well as being a strong stimulant for acid production, (Hansen, Pedersen and Rehfeld 1976). Some interesting results were obtained by Cook, Williams and Litchenberger (1981) where the development of the cat cestode parasite Taenia taeniaeformis in the liver of rats, as Cysticercus fasciolaris, resulted in gross hyperplastic changes in the stomach and small intestine of parabiotic partners. This hyperplasia was presented with or without hypergastrinaemia, but the cause of the changes were unknown. Similarly, Baker and Titchen (1982) published results that clearly show some interaction between the intestine of sheep parasitised with T. colubriformis and dysfunction in the abomasum due to a reduction in HCl production. The authors suggested that this reduction is mediated by the release from the parasitised small intestine of a gastric inhibitory factor which acts systemically. This factor could be another gastrointestinal hormone, such as secretin, which is able to

block the affect of pentagastrin in the development of parietal cells in rats (Stanley, Coalson, Grossman and Johnson, 1972).

Other hormones emanating from endocrine cells in the upper small intestine are known to inhibit gastric secretion. These include gastric inhibitory polypeptide (GIP); vasoactive intestinal polypeptide (VIP), glucagon and somatostatin. The trophic effect of gastrin could thus have an important influence on the ability of the animal to cope with abomasal parasitic infection since its action extends beyond the stomach into the small intestine. This could explain, in part at least, the compensatory digestion and absorption which appear to occur in lower parts of the digestive tract of animals infected with abomasal parasites.

Some reports have described changes in the status of other hormones in the parasitised state. For example, in intestinal parasitism with T.colubriformis, Pritchard et al, (1974) found that clinically affected lambs showed a significant decrease in thyroxin and insulin levels and an increase in corticosteroids. Insulin seemed to be related directly to reduced feed intake because the pair-fed, non-infected animals showed the same decrease in insulin. Sykes et al (1975) obtained similar results in sheep infected with T. colubriformis. The implications of these hormonal changes on protein metabolism are related to the physiological action. Excess or deficiency of thyroxin produces a depressive effect in skeletal mass because of interruption in the function of protein synthesis and degredation (Buttery, 1983).

It is interesting to note that while insulin promotes the uptake of amino acids and protein synthesis in the muscle, cortisol promotes an increased flux of amino acid from the muscle because of its catabolic action (Rannels and Jefferson, 1980). Increased corticoids, then, have a catabolic effect on protein metabolism in muscle and may be associated with lowered growth rates in cattle.

Usually the effect of parasites on productivity, metabolic and physiological responses has been studied in animals infected with a single species of parasite. There is a need for caution when applying the results of such experiments directly to field situations in which animals may acquire more than one genus of parasite. A recent experiment (Steel, Jones and Symons 1982) demonstrated that lambs infected with both O. circumcincta and T. colubriformis had

liveweight gain and wool growth depressed to a substantially greater extent than would be expected from a simple summation of the effects of the individual infection with each parasite. Measurements of gastroenteric plasma loss indicated that the gut damage and extent of the hypoalbuminaemia was greater and more sustained.

EPIDEMIOLOGY

Studies on the epidemiology of bovine ostertagiasis have centred on three main areas, namely, the population dynamics of the free-living larval stages; the acquisition of immunity by cattle; and arrested larval development at the EL_4 stage.

POPULATION DYNAMICS OF THE FREE-LIVING LARVAL STAGES

Michel (1969,b) made a most important development in epidemiological knowledge by demonstrating that the numbers of free-living L_3 of O. ostertagi fluctuate seasonally on the herbage. In Britain, and indeed the Northern Hemisphere, a considerable increase in larval numbers is noticed sometime during the second half of the recognised grazing season, from July onwards, declining during the subsequent winter months, to reach low levels by the following spring, and around the zero mark by the next June.

This seasonal pattern appears to be a constant one and is accounted for by two important facts. Firstly, the increase of L_3 during the latter half of the recognised grazing season originates from infection deposited as eggs on the pasture, in the early grazing period, i.e. spring or early summer, by grazing animals. Thus research by Michel showed that eggs of O. ostertagi deposited in faeces during the late spring and early summer first appear as L_3 sometime during mid-summer; thereafter development of eggs to L_3 slows, with little or no development occuring after September. This would allow one, or at most two, generations of Ostertagia spp. to occur annually with the important contamination period being in the spring or early summer (April, May, June). Previous postulations had allowed for the development of several cycles of L_3, based on an idea that L_3 development proceeds quickly once the mean day/night temperature is greater than $10^{o}C$.

Secondly, once established on the pasture in summer or autumn, L_3 can survive until the following spring and thus infect the next seasons grazing calves. Since Michel's first observations on seasonal fluctuation, there have been many studies on the seasonal development of O. ostertagi. Other European studies confirmed what Michel had recorded (Armour and Osbourne, 1982) whilst other world-wide studies have all demonstrated a seasonality of infection with clearly defined

times of contamination and development of infective larval populations on the pasture with only a limited number of annual cycles.

Temperate areas of the southern hemisphere, such as Australia and Argentina, where there is high rainfall and mild temperatures also provide suitable environments for the transmission of nematodes. Ostertagia spp., Trichostrongylus spp. and Cooperia spp. are the main genus involved in parasitic outbreaks in beef cattle in these areas (Rosa, Lukovich and Niec, 1971; Smeal, Hotson, Mylrea, Jackson, Campbell and Kirkton, 1977; Entrocasso and Steffan, 1980).

Calves are mainly born in winter-spring and weaned at 6-8 months of age, late in the summer-autumn period, they graze under an extensive pastoral system and before weaning there are few possibilities of outbreaks of parasitism due to the low rate of animals per unit area and the dilution effect of the immune adults.

After weaning, the numbers of infective larvae on the herbage increase to levels that could produce a substantial loss of production in winter and spring (Smeal, Fraser and Robinson, 1980). During the winter a severe decrease in grass availability normally occurs, which forces the animals to graze close to the ground and near to faecal pats. This coincides with the highest nutritional requirements of the young animal because of it s rapid growth and the need to cope with low temperatures and bad weather conditions. Studies carried out by Young and Anderson (1981) and de Chanet, Dixon and Barker (1981,a) in Australia, and Entrocasso and Steffan (1980) and Steffan et al (1981) in Argentina have demonstrated the epidemiological significance of the high pasture contamination in autumn and winter.

A sharp decrease in L_3 on the pasture occurs from late spring and there are negligible numbers present in summer. However, Young and Anderson (1981) have shown that infective larvae arising from eggs deposited in late spring may survive in faecal pats over the summer and subsequently make a substantial contribution to available larvae on the pasture in the following autumn and winter.

By observing the effects of temperature and moisture on the free-living larval development in Scotland, Gettingby, Bairden, Armour and Benitez-Usher (1979) were able to develop a prediction formula for both timing and magnitude of the seasonal increase of L_3 numbers, allowing a more precise determination as to when control measures should be applied to avoid heavy infections.

Recent reports have also shown that Ostertagia L_3 could

increase on pastures, such as aftermath grazing, which had not been contaminated during the crucial early grazing season (Armour, Al Saqur, Bairden, Duncan and Urquhart, 1980). The results of studies by Al Saqur, Armour, Bairden and Gettinby (1982) suggest a possible reason why the appearance of maverick populations of L_3 might occur on ungrazed pastures. Soil samples were taken from a previously heavily infected pasture which was then left ungrazed for 18 months. Analysis of the samples over a 24 month period showed that infective larvae were present down to a depth of 15 cm declining in numbers during the summer. At 5 cm depth the number of infective larvae was equivalent to that recorded nine months previously. Variations also occured in the numbers of L_3 on the herbage of the ungrazed pastures, comparable with the changes recorded in the larvae numbers of the upper soil layers. Although little is known of how the larvae migrate in the soil or on the herbage, Oakley (1981) has recently recovered viable bovine lungworm larvae from earthworms, allowing a possible link to be made between larvae infection and earthworm activity. The extent of this phenomenon is unknown. It does, however, represent a possible problem to the effective functioning of control measures.

ARRESTED LARVAL DEVELOPMENT

Arrested larval development (or inhibited development or hypobiosis) may be defined as the temporary cessation of development of a nematode at a precise point in its early parasitic life, this being the EL_4 stage in Ostertagia spp., and should be distinguished from slowing of growth or stunting of worms in which parasites in various stages of development are present. Michel (1974) defined three acceptable criteria for determining the occurence of true larval arrest:-

(i) the finding of a large number of larvae at precisely the same stage in grazing animals where there has been no recent uptake of infective larvae;

(ii) the same finding in animals withheld from possible infection for a time longer than that required to reach the stage of arrest;

(iii) the occurence of a bimodal size distribution in worm population from hosts not exposed to a corresponding pattern of infection.

The phenomenon of arrested larva development was not considered to be of great epidemiological importance until Martin et al. (1957) published an article on outbreaks of atypical parasitic gastritis in housed cattle. The authors suggested that the disease was caused by the maturation of worms which had been ingested as L_3 during the previous grazing season and then arrested in their development in the EL_4 stage, presumably because of an immunity acquired by the host as a result of exposure to larval challenge during the grazing season. This theory of arrest due to host immunity persisted for years, and has remained an acceptable proposition in older cattle. However, the results of experiments by Anderson, Armour, Jennings, Ritchie and Urquhart (1965 a) suggested that host immunity was not the major factor in inducing the arrested development of O. ostertagi larvae. In an epidemiological study carried out on two farms under identical conditions over a two-year period, 10 helminth-naive 'permanent' calves (3 months old) were grazed on known contaminated pasture from May to October, remaining on pasture unless the onset of clinical disease required their removal and necropsy. Groups of two helminth-naive "tracer" calves were grazed throughout the experiment season at fortnightly intervals, to monitor the seasonal fluctuations and availability of O. ostertagi L_3. Post-mortem burdens of O. ostertagi in both permanent and tracer calves gave unexpected results in comparison to previous field observations.

Thus, analysis of the burdens of the permanents showed that, if the worm burdens of the calves necropsied before and after October 1st were compared, there was a marked increase in the percentage of EL_4 stages in those necropsied after October 1st. A similar seasonal trend occurred in the proportion of EL_4 stages present in the tracer calves. The authors suggested that physiological changes in either the host or parasite during the autumn were responsible for the arrested development of larvae.

A second experiment was carried out in the following year in which the periods grazed by the tracers were varied to one, 14 and 28 days. This was to overcome the possible criticism that the increase in numbers of arrested EL_4 larvae in the autumn tracers might have been due to an exaggerated antigenic stimulus from the ingestion of high numbers of larvae on the pasture. The results showed that the proportions of arrested larvae were similar in all the tracers and therefore independent of the length of time grazed and presumably of

the level of larval challenge.

Subsequently, several attempts were made to induce arrested larval development experimentally by subjecting L_3 of O. ostertagi to the environmental conditions of a European late autumn and inoculating these larvae to helminth-naive calves. The isolate of L_3 used in these experiments had been experimentally passaged in calves on numerous occasions since it was first isolated at the Ministry of Agriculture laboratories in Weybridge, England. The experiments were repeated using a fresh field isolate obtained from calves in the Glasgow area. The results of 'autumn-conditioning' these larvae and inoculating them to naive calves clearly show that a large proportion of 'conditioned larvae' became arrested in development. Simple storage at 4°C produces the same effect and if cold, or autumn-conditioned, larvae are then subjected to spring conditions they appear to have lost the inclination to become arrested in development.

The final experiments in the Glasgow studies were designed to assess if the host had any role in inducing seasonal larval arrest or stimulating the eventual development of these arrested larvae. Freshly harvested L_3 of a recent field isolate or those conditioned for several weeks at 4°C were administered to calves kept in large animal climatic chambers simulating a spring environment; only the calves given the cold-conditioned larvae had burdens containing significant proportions of arrested larvae and the proportions of arrested larvae were similar to those present in calves kept under normal winter conditions at the Glasgow University farm. These results emphasised the relationship between the larval condition prior to ingestion by the calf and the proportion of larvae which became arrested in development.

The maturation of arrested larvae was studied in calves, in which large numbers of arrested EL_4 larvae had been produced, following the inoculation of cold-conditioned larvae from a recently isolated field strain and the removal of susceptible adults by an anthelmintic. Serial slaughter of such calves showed that the majority of larvae did not start to develop again until four months later (Armour and Bruce, 1974). This experiment was completed during the winter and when it was repeated in the spring the period of maturation was again four months, although the rate of maturation was faster. The spontaneous and synchronous development of the arrested larvae in the Glasgow experiments, together with the ability of temperature variations to induce arrest, prompted the Glasgow group to suggest that the whole

phenomenon of larval arrest and maturation in O. ostertagi resembles diapause in insects, particularly as the ultrastructural appearance of the larvae indicated a reduction in secretory and neuromuscular activity. Other studies in the Northern Hemisphere have similarly shown that arrest of O. ostertagi ingested in autumn grazing is a common occurence (see review by Armour and Ogbourne, 1982).

Michel and his colleagues have also been able to confirm the Glasgow findings in relation to the induction of arrested development by prior exposure of O. ostertagi L_3 to cold-conditioning (Michel, 1974). One area of dissent exists, namely in relation to factors affecting maturation of arrested larvae. In the experiments of Armour and Bruce (1974) EL_4 matured after four months in a spontaneous and synchronous fashion, Michel and his group were unable to confirm these findings using a Glasgow isolate in both field and experimental conditions (Michel, Lancaster and Hong, 1976,a,b). To the contrary, they consider that there is a regular turnover of Ostertagia populations in the host and that maturation is most definitely not spontaneous or synchronous. They do, however, concede that a higher proportion matures in the spring. Other than the obvious geographical and climatic influences it is not known what causes the differences between the Glasgow and Weybridge results.

In other areas of the world arrested O. ostertagi EL_4 accumulate at different seasons. In the Southern Hemisphere, in temperate areas of Australia, they accumulate in spring (Smeal et al, 1977). In New Zealand, the autumn shows the highest levels (Brunsdon, 1972), and in southern parts of the United States EL_4 larvae of O. ostertagi are most common in late winter and early spring (Williams, 1980; Craig, 1980). In these countries cold-conditioning has failed to induce larval arrest, which is not surprising as the onset of the phenomenom occurs principally in spring or under warmer and drier conditions.

The finding of Smeal et al (1980) that isolates of O. ostertagi from different areas of Australia display a varying propensity to become arrested in development has provided the opportunity to compare such isolates under different environments. Smeal and Donald (1981) made studies of two isolates of O. ostertagi, one from the northern Tablelands of New South Wales and the other from the coastal region. The former was prone to arrested development while the latter showed a lower propensity for arrestment. Transferring the

isolates to the opposite environment produced arrestment in development to a similar degree as their original environment, suggesting that propensity for arrestment is genetically determined and independent of environmental stimulus.

In summary it appears that when arrested development of O. ostertagi occurs during a particular season of the year, it is an heritable trait of the larva itself and may occur in response to various adverse environmental stimuli, one of which, in the Northern Hemisphere, appears to be declining temperature or cold conditions.

HOST IMMUNITY

Three main findings have emerged from studies on the acquisition of immunity by the host in ostertagiasis.

Firstly, both field and experiment studies would suggest that there is no absolute age immunity against O. ostertagi. For example, outbreaks of clinical osteragiasis have been noted in USA and Britain when adult cattle have been moved from areas where the causal parasite does not occur to areas where outbreaks are common (Bailey and Herlich, 1953; Selman et al. 1976). Although experimental studies in USA and Britain have shown a greater resistance in adult cattle to the debilitating effects of ostertagiasis, with slower worm development and lower egg production, considerable numbers of worms do develop with pathological changes being similar to those in young, naive calves (Herlich, 1960; Armour, 1967). Speculation that adult cattle will acquire immunity more rapidly than young stock has, however, not yet been substantiated.

Secondly, young cattle acquire immunity relatively slowly and outbreaks may still occur in animals after three to four months constant exposure. The results of British studies showed that calves at the end of their first full grazing season (May to October), displayed a good immunity to experimental challenge infection (Ross and Dow, 1965; Armour, 1967) but following a winter housing period there was a considerable decrease in the level of immunity by the following April (Armour, 1967). The result of further studies of young cattle over two consecutive summer grazing seasons, with winter housing intervening, showed that there was a markedly lower faecal egg count and worm burden in the cattle in the second grazing season (Armour, Bairden, Duncan, Jennings and Parkins, 1979). However, although young

cattle acquire a good immunity by the second grazing season, sufficient burdens are established to maintain contamination of the pasture, albeit at a reduced level. As might be expected from these results attempts to induce an artificial immunity in young cattle using X-ray attenuated larvae have been unsuccessful (Armour, 1967; Burger, Eckert, Chevalier, Rahman and Konigsmann, 1968).

Thirdly, although a certain state of immunity may be established by the end of the second grazing season, such a state is not necessarily permanent. This is especially so for the heifer cow around calving and during early lactation, as shown by Michel, Lancaster and Hong (1979). Petrie et al (1984) recently described an outbreak of type II ostertagiasis in dairy heifers which calved in November/ December and becoming ill in the following May. One heifer died and one other was sacrificed to confirm the diagnosis of ostertagiasis. Individually affected heifers showed a fall of 700 litres from the predicted yield of milk assessed at 80 days post calving. The authors also found a strong correlation between the fall in milk yield and the elevated serum pepsinogen.

In general, immunity appears to be restored towards the end of the first lactation though low adult worm burdens may remain in older cattle. On occasion where dairy cows have accumulated large numbers of parasites, mainly arrested larvae, the stress of calving may precipitate type II disease (Hotson, 1967; Wedderburn, 1970; Raynaud and Bouchet, 1976 a,b).

CONTROL

The control of ostertagiasis can conveniently be divided into the treatment of existing infections and their prevention or prophylaxis.

TREATMENT

The wide-spectrum anthelmintics currently available for treatment come from four main chemical groupings:

(i) the benzimidazoles and pro-benzimidazoles;
(ii) the tetramisoles;
(iii) the pyrantel group;
(iv) the avermectins.

Most are effective against developing larval stages and adults of

Ostertagia spp. and some are also effective against the arrested larval stages. The efficiency of the group (1) anthelmintics is high but only the less soluble compounds are highly effective against arrested larvae (fenbendazole, oxfendazole and albendazole and febantel). The tetramisoles also possess a high degree of efficiency against the developing larvae and adult parasites but neither these nor the pyrantel group are effective against the arrested larvae stage. Studies with the newly found avermectins, in particular the natural B_{1A} component, ivermectin, have shown a wide spectrum of activity against all stages of Ostertagia, including arrested larvae (Armour, Bairden and Preston, 1980; Williams, Knox, Baumann, Snider, Kimball and Hoerner, 1981).

Although treatment of animals infected with Ostertagia may result in the removal of the worm populations, it does not alleviate the pathological damage already caused and it is now widely accepted that the prevention of infection benefits production more than treatment of already infected stock.

PROPHYLAXIS

Prevention of ostertagiasis is applied mainly to yearling stock in the herd by a variety of methods. The seasonal fluctuation in numbers of L_3 on herbage has provided the basis for the application of the control methods. Thus the Weybridge 'dose and move' method (Michel, 1969), as used in the Northern Hemisphere, relies on two main factors. Firstly, that young cattle grazed in spring rarely acquire sufficient amounts of L_3 to be seriously affected although significant numbers of eggs may be deposited on the pasture within three to four weeks. Secondly, since it takes until mid-July for the newly deposited eggs to mature to L_3, a move early in July to pasture ungrazed by cattle since the previous autumn, should result in only light infection being subsequently acquired. If the move is accompanied by an effective anthelmintic treatment the stock should remain relatively worm-free for the rest of the grazing season (Michel, 1969). Spedding (1969) however, pointed out that although the helminth disease may be eradicated it is not possible to eradicate the helminths themselves.

Rotational grazing of cattle, alternate grazing of cattle with different host species or integrated rotational grazing of different age groups of cattle are also used to combat bovine ostertagiasis. The straightforward rotational system involving solely cattle has proved to

be less effective than set-stocking of cattle on pasture, since the rotation may return the cattle to an area at a time when the L_3 are accumulating, e.g. in July and August (Levine and Clark, 1961; Michel, 1969). Furthermore, the luxuriant herbage cover in rotated pasture encourages larval survival whereas the sparse herbage in set-stocked areas mitigates against survival.

Better control has been achieved by methods which involve either grazing mixed host species together (Arundel and Hamilton, 1975) or alternate grazing of different host species (Barger and Southcott, 1975; Southcott and Barger, 1975; Rutter, 1975). The success of both systems depending on the host specificity of Ostertagia spp.

Improved liveweight gains have also been reported when susceptible dairy calves were rotationally grazed on permanent pastures and followed by replacement heifers in their second or third grazing season (Leaver, 1970; Nagle, Brophy, Caffrey and O'Nuallain, 1980). The calves are only permitted to graze the upper leafy part of the herbage before being moved onto the next paddock. This is to avoid the mass of L_3 concentrated in the lower quartile of the herbage (Crofton, 1954). Much depends on the immunity of the heifers and their previous exposure to the parasite, as age per se does not guarantee a high immunity. Even in heifers which have acquired a good resistance there is evidence, as already stated, that this weakens around first calving (Michel et al., 1979). Dry cows may be used in place of heifers in this system but the availability of such cows may be limited if the ratio of cows to calves in a herd is maintained (Downey and Fallon, 1973).

Permanent housing of animals has been used as an alternative method of preventing ostertagiasis, though it is more widely employed on the continent than in Britain (Borgsteede, 1977). This involves the feeding of harvested grass to the stabled animals. In Borgsteede's trial of a partial zero grazing method, the housing period was limited from April to June by which time any pasture infestation had declined to extremely low levels owing to the mortality of the overwintered L_3; although some infection occurred in the calves grazed after June the subsequent levels of L_3 were insignificant compared with the control pastures.

Finally, although anthelmintic treatment of the young cattle set-stocked on permanent pasture has been shown to be economically beneficial (Cornwell, Jones and Pott, 1973) regular reinfection occurs

and several treatments are necessary to maintain production levels; this is undesirable from the management viewpoint particularly when labour costs are ever increasing. To overcome the first of these problems it has been suggested that treatment should be concentrated in the early part of the grazing season (Pott, Jones and Cornwell, 1974; Armour, 1978; Herd and Heider, 1980). By limiting pasture contamination to a sufficiently low level in the spring and early summer the expected increase in L_3 numbers from mid-July onwards would be considerably reduced and economically significant weight gains acheived.

In an attempt to overcome some of the practical difficulties associated with repeated treatments it was suggested that the provision of continuous anthelmintic medication in feed or water could be beneficial (Downey, O'Shea and Spillane, 1974; Jones, Potts and Cornwell, 1978; Potts, Jones and Cornwell, 1979). However, although it overcame the management problem of regular treatment it has been difficult to regulate individual consumption of the anthelmintic on a daily basis under field conditions. A more attractive development is that of sustained-release devices which are delivered into the rumen via a special drenching gun (Jones, 1981). One such device was designed to provide a sustained release of the anthelmintic morantel tartrate over 90-days and so prevent the establishment of infective larvae in the alimentary tract during this period. Under conditions existing in Western Europe the application of this device in the spring had been shown to limit the acquisition of overwintered O. ostertagi L_3 and so reduce significantly the contamination of pasture for the vital three months of April, May and June with liveweight gain benefits over untreated controls of up to 41 kg being achieved over one grazing season (Jones, 1981; Armour, Bairden, Duncan, Jones and Bliss, 1981; Jacobs, Fox, Walker, Jones and Bliss, 1981; Tharaldsen and Helle, 1982; Helle and Tharaldsen, 1982).

Nevertheless, many beef or dairy/beef cattle are turned out for all or part of a second grazing before they are sold or sent to slaughter. Replacement heifers also undergo a second grazing season before entering the adult herd. The morantel sustained release bolus (MSRB) has been used in limited numbers of cattle in their second grazing season or beyond. In England, Condor, Jones and Bliss (1983) reported the results of seven farms in which the MSRB was used in second year animals and in six of them the difference in body-weight

gain was greater in the MSRB treated group. This difference was significant in three farms. Surprisingly, the largest differences in bodyweight gain (39 kg) over the control group occurred in a farm in which the larvae recoveries from pasture were relatively low (maximum 2000 L_3/kg dry herbage in August) and, by then, an efficient immune response would be expected. It is difficult to comment on that because there is no information on the evolution of the bodyweight during the time of the experiment.

Guldenhaupt and Burger (1983) in Germany, reported the results of five farms in which the MSRB was used in second-season cattle. Significant differences in bodyweight gain was obtained in only one farm. The nematode egg production in the faeces were less in MSRB treated cattle during the period of drug-release from the bolus than in the control group, becoming higher afterwards, but always within low values. The larvae recoveries from the pasture were also similar and low in both groups.

Clearly there is now a wide range of effective anthelmintics for the treatment of ostertagiasis, in countries of the Northern Hemisphere, and several methods for its prevention. Depending upon local epidemiological factors it should be possible to develop a suitable control program for a particular farm's requirements with regard to the system it employs.

On the basis of the same principles as outlined above, systems of controlling ostertagiasis by reserving safe pasture have also been developed in many area of the Southern Hemispere, such as Australia (Brunsdon, 1980; Barger and Southcott, 1975; Smeal, Nicholls, Robinson, Bowler, Webb and Walker, 1981) while programs dependent upon the use of prophylactic anthelmintic medication have been used in Argentina (Niec, Rosa and Lukovich, 1968; Rosa, Niec, Lukovich, Martin Vidal, Monje and Hofer, 1976/77; Entrocasso and Steffan, 1980) and in Brazil (Pinheiro, 1970; Ramos and Ramos, 1978).

GENERAL MATERIALS AND METHODS

1. ANIMALS AND GRAZING PLOT MANAGEMENT.

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3. PATHOLOGICAL AND PARASITOLOGICAL METHODS.
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   Worm burdens at necropsy
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5. DIGESTIBILITY AND NITROGEN RETENTION STUDIES:
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GENERAL MATERIALS AND METHODS

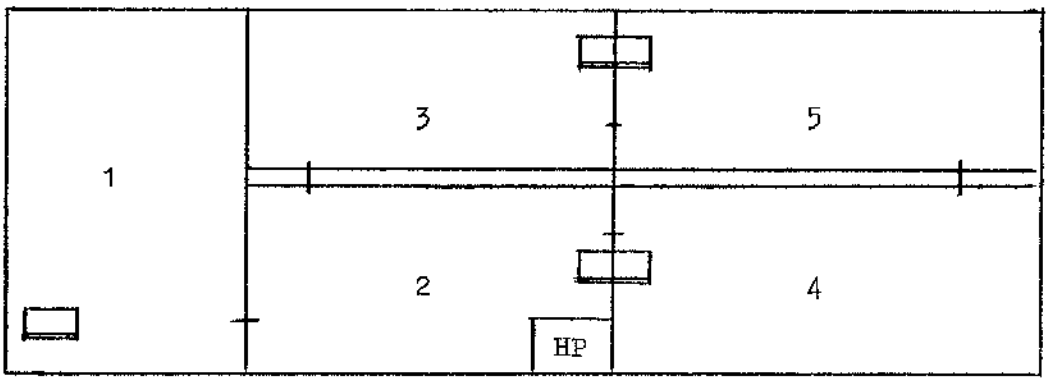
ANIMALS AND GRAZING PLOT MANAGEMENT

The castrated male Friesian calves used in the major two year grazing experiment were purchased from a local livestock dealer/farmer who collected weaned calves together over a short period of time into a uniform group with respect to age and size. Time did not permit the treatment of these calves with lungworm vaccine.

The calves were housed indoors together as one large group and given hay and commercial rearing concentrates (160 g CP/kg) for at least one month before the transference to grass at Glasgow University Veterinary School in May 1982.

The grazing area, which was known to be contaminated with *O. ostertagi* larvae, consisted of one large field of 6.2 ha which was purposely divided into 5 separate plots (see Fig. 2). Four of the plots were designated to be used by the control and MSRB treated calves and were about 1.2 ha each in area. (Plots 2, 3, 4 and 5). A further plot (1) which was to be set-stocked by a smaller clean group of calves, had an effective 1.4 ha of grazing area. Plots 3 and 5 were space separated from plots 2 and 4 by a double fence 1.5 m apart (see Plate 1).

Fig. 2 Stylised diagram of the grass plots used in the major two year grazing and metabolism study with Friesian steers.



HP Handling pen equipped with crush and accurate weighing machine

In the first year (1982) plots 3 and 5 were grazed by MSRB treated calves and plots 2 and 4 by control calves. In the second year, in

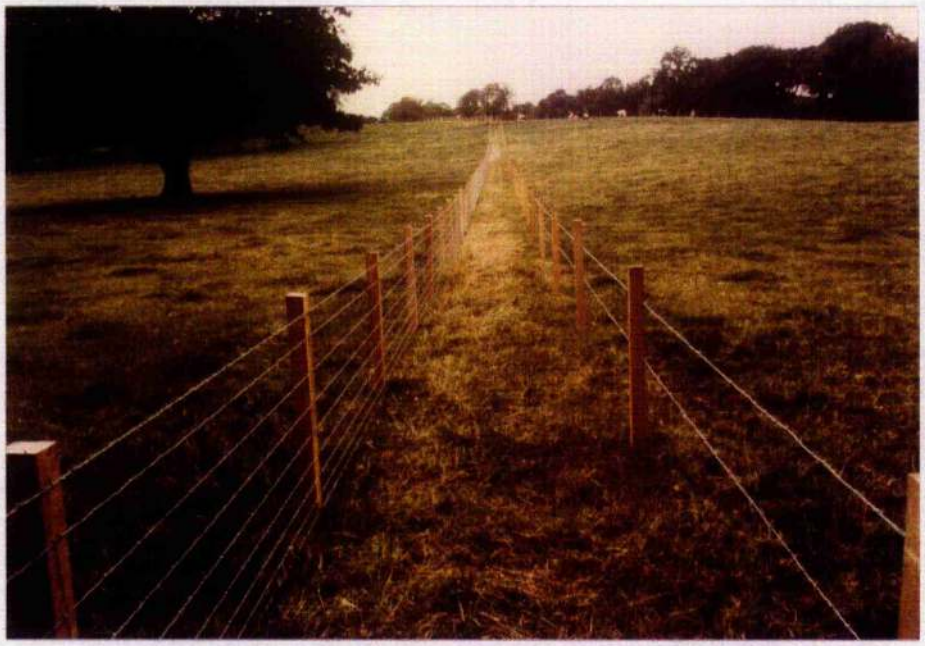


Plate 1 Arrangement of fences to divide paddocks.

Plate 2 Paratect bolus and dosing gun.

order to equalize the larval challenge presented to both groups, paddocks 3 and 4 were allocated to MSRB steers and 2 and 5 to the control animals.

In each of the two grazing seasons a total of 200 kg of fertilizer Nitrogen (N) was applied per hectare of grassland. This consisted of an 'early bite' application in late April of 50 kg/ha followed by three successive 50 kg N administrations during the grazing season. The clean group of cattle was kept in plot 1 during the whole of both seasons whereas cattle in the control and MSRB treated groups were alternated between one of their two allotted paddocks according to the grass availability. On each occasion groups of cattle were moved on the same day. The vacated plots were then fertilized. More precise details of the plots grazed by each group and the dates of transfer between plots are given in the appropriate experimental sections of this thesis.

MORANTEL SUSTAINED RELEASE BOLUS

In the large-scale two year grazing experiment the morantel sustained release bolus (Paratect, Pfizer Inc.) used to treat the MSRB group had the following characteristics.

The basic description was given by Jones (1983) and included a labelled diagram of the device. The bolus (see Plate 2) was comprised of a stainless steel tube (9.1 cm in length and 2.7 cm in diameter) fitted at each end with a sintered polythene disc impregnated with cellulose triacetate hydrogel and a crimped aluminium cap end. The tube was encased (except for the ends) with a heat shrunk polyolefin band. The bolus was filled with a blend of 22.7 g morantel tartrate (equivalent to 13.5 g morantel base), 14.7 g polyethylene glycol 400 and 4.1 g sodium metaphosphate.

The bolus was reported by Jones (1983) to have been designed to release 'approximately 90 mg of base/day for an extended period (at least 60 days).'

Experimental results recorded in the same publication in fact show a mean release rate of 170 mg/d for days 0-30, 75 mg/d for days 30-60 and only 47 mg/d for days 60-90. A residue of morantel of 4.69 g was recorded as still being present in the bolus after 90 days.

PATHOLOGICAL AND PARASITOLOGICAL METHODS

The animals used in the two year grazing study were slaughtered at two different locations. A total of 15 animals were killed after the

first season at grass and following winter period (including one which was destroyed *in extremis* during grazing) for parasitological, pathological and carcass evaluation. This was performed at one of the University of Glasgow Veterinary School's post-mortem facilities. However, at the end of the study, the remaining 24 animals were slaughtered at a nearby commercial abattoir in Paisley in order that the carcasses could be officially graded for potential eligibility for the EEC Beef Premium Scheme. The slaughter, post-slaughter and hanging procedures at both locations were essentially similar. Also the procedures adopted in the collection of body fluids and pathological specimens were the same at both sites.

The cattle were euthanased by captive bolt and immediate exsanguination. After opening the abdomen, the pyloric sphincter was ligatured and the gastro-intestinal tract removed from the body cavity. The large intestine and contents were discarded. The abomasum was removed intact with the duodenum being tied off at the abomasal - duodenal junction. After removal of any fatty surrounds, the abomasum was opened by incision along the greater curvature. A small (c.200 ml) sample of abomasal fluid was immediately collected into a glass jar and stoppered for the earliest possible determination of pH using a Radiometer pH meter type PHM 26c (Electronic Measuring Instruments Ltd., Copenhagen, Denmark). The period of time from collection to measurement of pH was generally no greater than about one hour.

Estimation of worm burdens at necropsy

The abomasal contents were collected into graduated buckets and the volume made up with tap water to a standard 4 litres, except where an unusually large amount of material was present requiring a greater volume of water. Two samples each of 200 ml were withdrawn after thorough mixing, and following the addition of 10 ml of 40% formalin were stored in jars for subsequent microscopic examination. The abomasum was then laid out on a board, cut in half longitudinally and the mucosa from each half scraped off with a sharp post-mortem knife. The mucosal scrapings were digested in three times its own volume of a pepsin-hydrochloric acid mixture (Herlich, 1956) at 42° C for 6 hours. The digested mixture was then made up to 4 l and formalinised 200 ml subsamples withdrawn as described before.

Parasites present in 10 separate 4 ml aliquots were counted and classified as adult male or female, developing fourth or fifth larval

stages or early fourth stage larvae (EL_4) depending on bursal or vulvular development, the presence of a sheath projection and size respectively. For Ostertagia spp. and Cooperia spp. the EL_4 were considered to be arrested in development since this stage is reached in four days and all the animals (excepting the individual killed in extremis at grass) were housed and kept from further infection for at least one week before slaughter.

Mucosal fraction

At post-mortem the gross appearance of the abomasal mucosa was noted and the presence of lesions described and where appropriate photographed.

In order to quantify the relative magnitude of any hypertrophic reaction noted in the abomasal mucosa of the experimental animals it was decided to define the 'mucosal fraction' as the number of grams of scraped mucosal weight per kg of liveweight of the animal. Additionally, the degree of oedema present was given a simple subjective score i.e. light (+), moderate (++) and heavy (+++).

Lungworm recoveries

During the first grazing season where lungworm larvae were known to be present on the herbage, the lungs were processed using a modification of the Inderbitzen (1976) perfusion technique. Essentially the procedure was as follows:

The pericardial sac was incised and reflected to expose the pulmonary artery in which a 2 cm. incision was made. Rubber tubing was introduced into the artery and fixed in situ by double ligatures. The remaining great vessels were tied off and water from a mains supply allowed to enter via the pulmonary artery. Twenty litres of washings were collected from the lungs and concentrated by passing through a 37 u aperture sieve. Parasites recovered in this way were allowed to relax overnight at $4^{o}C$, counted and differentiated on the basis of size and stage of development i.e. size range 1-2 cm and the absence of any definite internal structure (4th larval stages), size range 3-5 cm and a noticeably increasing developmental pattern, particularly in the uterine and bursal regions (5th larval stages), size range 5-10 cm, presence of eggs in females and fully developed bursal regions in males (mature parasites).

Faecal egg counts

Faecal samples were collected either directly from the rectum or from the faeces collection from those individuals housed in metabolism stalls in the winter periods. The samples were examined by a modified McMaster technique (Gordon and Whitlock, 1939). In this method 3 g faeces were homogenised with 42 ml of water and the resultant suspension passed through a 250 micron sieve (Endecotts Test Sieves Ltd., Morden, London). After thorough mixing of the filtrate, 15 ml were withdrawn into each of two flat bottomed centrifuge tubes (capacity 15 ml) and centrifuged at 2000 rpm for two minutes. The supernatants from both tubes were then discarded and the remaining faecal mass broken up by rotary agitation. One tube was then filled to its former level with saturated salt solution and after inverting six times a volume of the suspension, sufficient to fill both chambers, was quickly transferred by pipette to a 'McMaster' slide. The number of eggs under the etched areas of the slide were counted and the result multiplied by 50 to give an estimation of the number of eggs per gram of faeces according to the following calculation:-

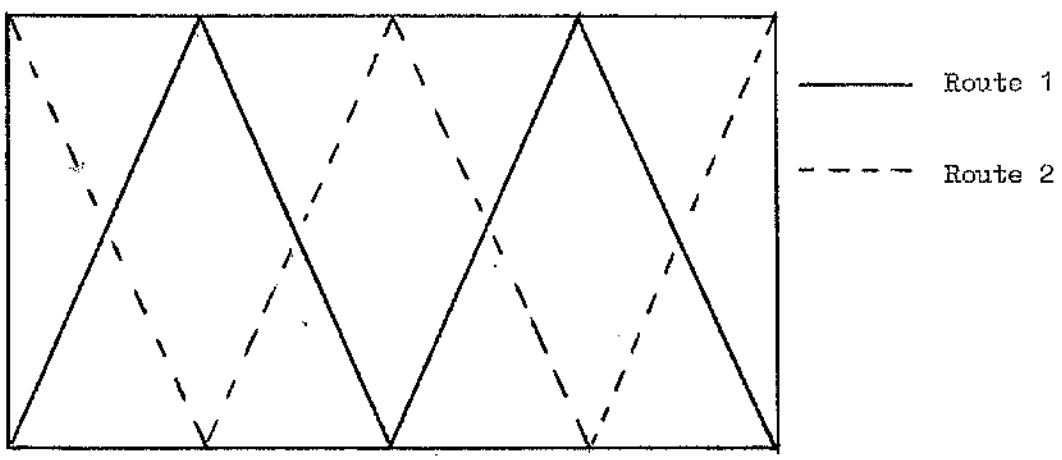
3 g of faeces in 42 ml gives 1 g in 15 ml

Volume under one square equals 0.15 ml

No of eggs seen in two squares x 50 = No of eggs/gram

Analysis of herbage for the presence of trichostrongyle L_3 larvae

Pasture samples were collected by traversing the experimental plots as shown in the following diagram and combining samples taken on routes 1 and 2.



Fifty evenly spaced stops were made along routes 1 and 2 and at each stop four plucks of grass (the amount that could be grasped between thumb and forefinger) were taken giving a total of 400 plucks per plot. The grass was then processed by a method similar to that used by Parfitt (1955).

After weighing, the grass was soaked in 20 litres of warm water plus 5 ml of non-ionic detergent (Lissapol , ICI Ltd., U.K.). Twenty-four hours later the herbage was transferred to 20 litres of warm water and rinsed overnight after which as much water as possible was removed by manual squeezing and the grass spread out on to trays. After drying at 80°C the herbage was weighed again and the dry weight used to calculate the number of larvae present as L_3/kilogram dried herbage (kdh).

The grass washings were allowed to sediment for a minimum of six hours. The supernatant was then drawn off and the sediments bulked and filtered through a double milk filter (Maxa Filters, A. McCaskie, Stirling, Scotland). The retained larvae were recovered using standard Baermann apparatus. A 150 micron aperture sieve supported the milk filter which allowed motile larvae to migrate to the warmer side of the temperature gradient. After twelve hours 200 ml of fluid was withdrawn and reduced (by a process of sedimentation and centrifugation) to a final volume of 10 ml. The larvae in a 1 ml aliquot were then microscopically differentiated and counted. The criteria taken for larval identification were those detailed in Technical Bulletin No. 18 (Ministry of Agriculture, Fisheries and Food 1971) and those used by Keith (1953) i.e. body length, prolongation of the second larval sheath beyond the tail of the third stage larva and the presence of refractile structures.

BLOOD BIOCHEMICAL METHODS

Plasma pepsinogen estimation

Blood samples for pepsinogen estimation (as for all other biochemical analyses) were taken directly from the jugular vein into heparinised vacutainer tubes (Becton Dickinson Ltd.) and centrifuged at 2,000 rpm for 20 minutes after which the clear plasma was carefully removed. The method used was essentially that described by Edwards, Jepson and Wood (1960).

Reaction

Plasma is incubated with bovine serum albumin (BSA) at pH 2 for 24 hours and the phenolic amino acids liberated (tyrosine like) are estimated using Folin-Ciocalteau Reaction. Corrections are made for the normal (i.e. non-incubated) content of tyrosine-like substances and also for the release of these substances from BSA when incubated alone.

Reagents

2% Bovine Serum Albumin (BSA)
2N HCl
4% Trichloracetic Acid (TCA)
N/4 Caustic Soda
Folin-Ciocalteau's Reagent (diluted 1+2 with water)
Stock Standard Tyrosine. 1.812 g tyrosine in 1000 ml N/10 HCL (10 u mols/ml).
Working Standards 10 ml Stock Standard diluted to 1000 ml (2.0 ml contains 0.2 u mols) and 20 ml diluted to 1000 ml (2.0 ml contains 0.4 u mols)

Procedure

Plasma Tests 2.5 ml plasma and 10 ml 2% BSA. Adjusted to pH 2 with 2 N HCl (Approx 0.5 ml) and water added to make total volume 15 ml (2.0 ml H_2O)

BSA Blank 2.5 ml water and 10 ml 2% BSA. Adjusted to pH 2 with 2N HCl (Approx 0.35 ml) and water added to make volume to 15 ml (2.15 ml H_2O).

1. 6.0 ml aliquots of tests were pipetted into universals and incubated at 37°C for 24 hours.
2. 6.0 ml aliquots of BSA blanks were pipetted into another set of universals and the protein precipitated with 10 ml of 4% TCA.
3. Precipitated blanks were allowed to stand for ten minutes and then filtered through a No. 44 Whatman filter paper.
4. Tests were precipitated after incubation with 10 ml 4% TCA then processed as in (3) above.
5. 2 ml of all filtrates were pipetted into suitably labelled flasks containing 20 ml N/4 NaOH.
6. Flasks containing 2 ml of each working standard were set up with 20 ml N/4 NaOH i.e. 2 u mols and 4 u mols tyrosine.
7. A reagent blank containing 2 ml H_2O with 20 ml N/10 NaOH was

set up.

8. 3.0 ml diluted Folin and Ciocalteau's reagent was added to all flasks.
9. After standing for 30 minutes the blue colour was read in a spectrophotometer at a wavelength of 680 mu.

Calculation of results

1. The reagent blank was subtracted from all readings.
2. From tyrosine standards the factor for conversion of all spectrophotometer reading to u mols tyrosine was calculated and all readings converted to u mols tyrosine.
3. If incubated BSA and plasma = A
   and non-incubated BSA and plasma = B
   then A-B = total release of tyrosine on incubation.
4. If incubated BSA alone = C
   and non-incubated BSA alone = D
   then C-D = release of tyrosine from BSA substrate due to incubation alone, i.e. NO PEPSINOGEN.
5. Therefore (A-B) - (C-D) = tyrosine in u mols released on incubation of the equivalent of 0.125 ml serum for 24 hours with substrate.
6. The amount of tyrosine in u mols released per 1000 ml plasma per minute = International Units or x 1000 = milli Units tyrosine, (u mol (5) x 5.56).

Gastrin estimation

Gastrin was determined by radioimmunoassay using a commercially obtained kit (Cambridge Medical Diagnostics, Inc. Maryland, U.S.A.). The protocol for the method does not state specifically if the developed technique is a direct adaptation from any one or two particular published methods but the selected bibliography quotes 27 references and includes both Yalow and Berson (1970) and Stadil and Rehfeld (1971) where these latter authors report on the preparation of ^{125}I-labelled synthetic human gastrin for radioimmunoanalysis. The method used in this thesis is briefly outlined as follows:

^{125}I Gastrin radioimmunoassay

Reagents: ^{125}I Gastrin; CNR-115:

^{125}I-labelled human synthetic gastrin and non-immune rabbit serum. Supplied in lyophilised form. Contains approximately 2.0 u Ci ^{125}I .

Precipitating reagent; CNR-645:

Goat anti rabbit gamma globulin and 2% Polyethylene glycol in borate buffer containing preservative.

Gastrin (human synthetic) standard; CNR-315:

Synthetic human gastrin in Assay Buffer, at the following approximate concentrations: 50, 100, 175, 300, 500 and 1000 pg/ml.

Gastrin controls; CNR-515 and CNR-516:

Human synthetic gastrin in human plasma, plus preservative. Supplied in lyophilised form.

Reconstitution of lyophilised reagents:

The vial was tapped gently to dislodge any large particles that were trapped on the stopper or the side of the vial and then the rubber stopper was carefully removed and placed inverted onto a clean surface. The appropriate reconstitution volume was carefully added to each vial, the stopper replaced and the solid matter allowed to dissolve at room temperature for 20 minutes.

After 20 minutes had elapsed, the vial was inverted gently a few times to wash off any particles that may have adhered to the septum, then the vial was allowed to stand for a few more minutes to achieve a complete solution. After visually checking for complete reconstitution, the vial was gently swirled to obtain a homogeneous solution. It was important to avoid vigorous agitation and foaming.

Specimen collection and storage:

The usual venipuncture technique was followed. Either serum or plasma (collected in EDTA) may be used for the assay. After centrifugation to remove cells, the serum or plasma is frozen at -20°C. (Repeated freezing and thawing should be avoided).

Assay procedure

Optimal results were obtained by strict adherence to this protocol. Careful pipetting was essential. It was recommended to run

the standard level points and samples in duplicate. A standard curve was run with each set of clinical specimens.

The capped reagent bottles and vials were brought to room temperature (18-30°C) before use and immediately refrozen afterwards to minimise deterioration.

A. 12 x 75 mm glass tubes were labelled as follows:
Total counts (TC), tubes 1 and 2
Non-specific binding (NSB), tubes 3 and 4
Maximum binding (B_o), tubes 5 and 6
Standard (A to F), tubes 7 to 18
Control (low and high), tubes 19 to 22
Starting with tube 23, each pair of sample test tubes were numbered consecutively.

B. 200 ul of Assay Buffer was added to the NSB tubes, numbers 3 and 4.

C. 100 ul of Assay Buffer was added to the B_o tubes, numbers 5 and 6.

D. 100 ul ^{125}I gastrin solution was added to all tubes.

E. 100 ul gastrin antiserum was added to all except the TC and NSB tubes.

F. All the tubes were shaken gently within the test tube rack, covered and incubated for 60 minutes at room temperature.

G. After shaking the bottle of precipitating reagent well to ensure an even suspension, 1 ml of reagent was added to all except the TC tubes.

H. The tubes were then put into a vortex mixer and then incubated for 10 minutes at room temperature.

I. The tubes were then centrifuged at 1500 g for 10 minutes.

J. The supernatant of each tube (except the TC tubes) was decanted very carefully to avoid dislodging the pellet and the rim of each tube blotted.

K. The radioactivity in all tubes was counted for at least one minute to remove the counting statistics as a source of variability.

Calculation of results:

(i) Determine the average counts for each set of duplicate assay tubes.

(ii) Subtract the average NSB counts (tubes 3 and 4) from samples to determine the net average counts for each sample.

(iii) Divide the average net B_o counts (tubes 5 and 6) into the average net counts of the standard, control and samples. Multiply this number by 10 to yield the % B/B_o for each sample.

$$\%B/B = \frac{cpm_x - cpm_{NSB}}{cpm_{B_o} - cpm_{NSB}} \times 100$$

(iv) Plot $\%B/B_o$ for each standard vs the standard concentration in pg/ml on semi-log graph paper. The concentration of gastrin in each sample and control may be read directly from this standard curve.

Other blood analyses

Analyses for urea and inorganic phosphate were performed by standard automated techniques (Technicon Auto Analyser II) by which total protein was also determined using a biuret method (Weichselbaum, 1946) and albumin concentration by a modification of the technique of Rodkey (1965).

GENERAL CHEMICAL PROCEDURES

Preparation of feed, faecal and urine samples for anlyses

Representative samples of the diets used were taken for analyses at regular intervals throughout each experimental period. During the winter metabolism studies, the bulked daily 2% faecal sample obtained from each steer during the collection periods were well mixed and suitably sized aliquots retained for analyses. At least 1 kg was taken for immediate drying and for later subsequent analyses to be performed upon this dried material. A further 200 g was taken directly and slurried as described by the Grassland Research Institute (Commonwealth Bureau of Pasture and Field Crops, 1961) in preparation for the determination of nitrogen. A portion of this slurry was dried

immediately and the dry matter value used in calculating the N content per kg of dried faeces.

Dried feed and faecal samples were prepared for any subsequent chemical analyses by grinding suitable quantities through a 2 mm screen in an 8" laboratory hammer mill (Christy and Norris, England).

The bulked acidified daily 2% urine samples from each steer were well mixed by manual agitation and 2x 100 ml aliquots were retained and stored at -20°C for later analysis for N concentration and specific gravity measurement.

Chemical analyses of feed, faeces and urine samples

All the analytical methods used were officially established procedures i.e. MAFF et al, 1973; Fertiliser & Feedingstuffs Regulations 1968, 1976 and 1982.

Dry matter (DM)

The dry matter in food and faecal samples was determined by heating known quantities (200g) in a hot air oven at 80°C for 36 to 48 h until a constant weight was attained.

Organic matter

The organic matter (OM) content was determined by placing a crucible and dried contents in a muffle furnace at 600°C for 4 hours. The consequent loss in weight was recorded as the OM present and where the residue was regarded as ash.

Total nitrogen (N)

Total nitrogen in food and faecal samples was measured by an automated semi-micro Kheldahl technique (Kjell-Foss Automatic 16210). Urine nitrogen content was measured using a micro-Kheldahl method.

Ether extract (EE), crude fibre (CF) and ash

The ether extract, crude fibre and ash contents of food and faeces were determined using standard methods (The Fertiliser and Feedingstuffs Regulations, 1976).

Gross energy (GE)

The gross energy of food and faeces samples was measured using a Gallenkamp automatic adiabatic bomb calorimeter equipped with a digital data systems calorific value microprocessor. Calibration was performed using benzoic acid (Thermochemical Standard, BDH). Both samples and benzoic acid were pelleted using a die operated by an hydraulic press.

DIGESTIBILITY AND N RETENTION STUDIES

Calculation of digestibility coefficients and N retention

"The digestibility of a food is most accurately defined as that proportion which is not excreted in the faeces and which is, therefore, assumed to be absorbed by the animal" (McDonald, Edwards and Greenhalgh, 1981). It is commonly expressed, in terms of dry matter, as a coefficient:-

$$\text{DM Digestibility coefficient} = \frac{\text{amount apparently absorbed}}{\text{amount in feed (DM)}}$$

The coefficients for each constituent of the DM, e.g. CP, CF, EE, etc. can be calculated in the same way.

Although the proportion of food not excreted in the faeces is commonly assumed to be equal to that which is absorbed from the digestive tract, there are objections to this assumption, particularly in the case of ruminants. For example, in ruminants methane arising from the fermentation of carbohydrate is lost by eructation and thus is not absorbed. This results in an over-estimation of the digestible carbohydrate and digestible energy of the feed. Also because of the presence of metabolic faecal nitrogen in faeces, i.e. nitrogen derived from non-dietary sources such as sloughed epithelial cells, enzymes, etc., there is under-estimation of the proportion of nitrogen absorbed by the animal. Ether extractable substances and minerals of metabolic origin are also found in the faeces, again leading to an under-estimation of their absorption. Consequently, the values obtained in digestiblity studies are referred to as 'apparent' digestibility coefficients.

In this thesis, N balance is expressed as g N retained or lost on a daily basis and was calculated using the following equation:-

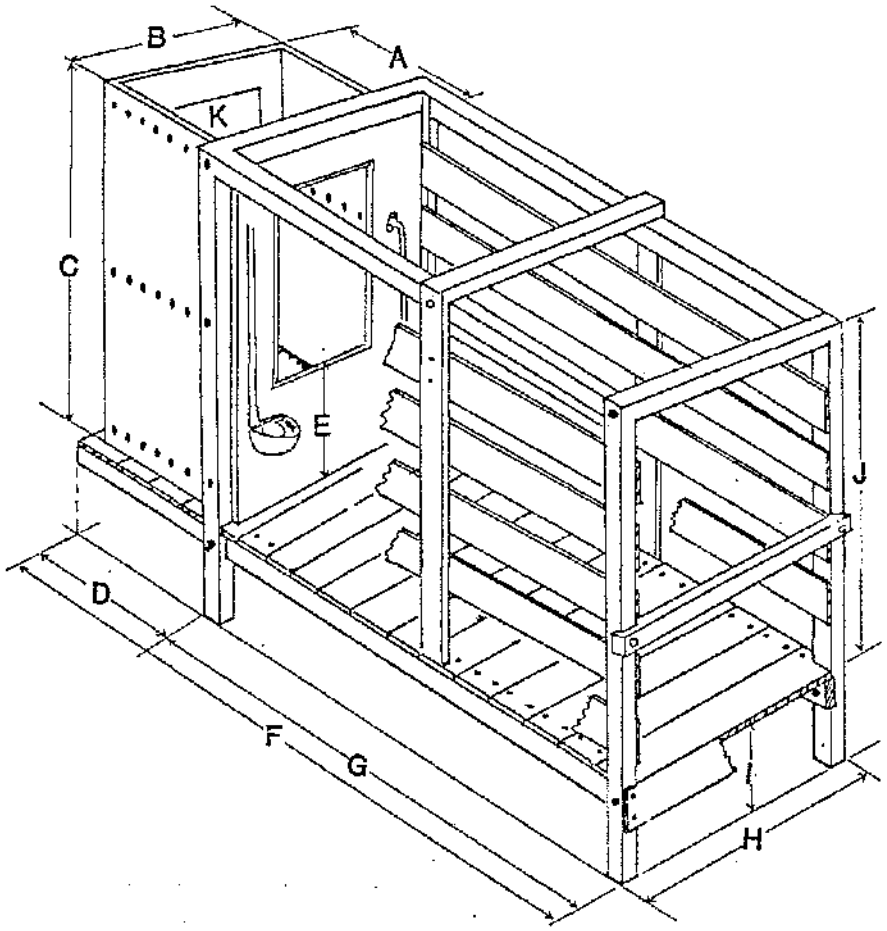
$$\text{N balance (gN)} = \text{gN feed} - (\text{gN faeces} + \text{gN urine})$$

AN IMPROVED TECHNIQUE FOR THE COMPLETE COLLECTION OF FAECES AND URINE FROM HEAVYWEIGHT STEERS

An important initial stage of the metabolism studies reported in this thesis was the design, construction and subsequent modifications

Fig. 3

CATTLE METABOLISM STALL



Key (mm)

A	610
B	760
C	1220
D	610
E	400
F	2440
G	1830
H	915
I	400
J	1380
K	flap

Design by: J.J. Parkins,
Glasgow University
Veterinary School

1981

made to a faeces and urine collection system from tethered male castrate animals weighing up to 600 kg. The specially designed cattle metabolism stall which was used is shown in Fig. 3. The stalls were built from wood sections held together by steel bolts. Precise construction and material details may be obtained from the Department of Animal Husbandry, Glasgow University Veterinary School. Features and modifications made to the stall in final use worth noting here are included in the following description.

METABOLISM STALL DESIGN AND FEATURES

(i) The height I, shown in Fig. 3, was increased when required by placing and securing wooden blocks as 'feet' under the legs in order to more easily accommodate the urine collection vessels (10 l plastic 'jerry-can' type containers). The increased height helped in the drainage of urine (by gravity) from the funnel collector strapped to the animal.

(ii) The floor of the stall was lined with a thick rubber cow-mat. This greatly improved the comfort of the animal.

(iii) The feeding box was adjustable so that it could be moved in or out of the body of the stall in order to accommodate different sizes of cattle. The feeding box was secured with 6" steel bolts at both the top and bottom of the mainframe supports. The front feed flap (K) allowed feeding access from in front of the stall without disturbance to the animal. The edges of the access port to the feed box should be 'tinned' to prevent the possibilty of crib biting. The alkathene water pipe leading to the water bowl attached to the front wall of the feedbox was protected by an outer metal conduit tube in order to prevent the animal from chewing it. A chain slide was fitted to the front wall of the feed box which allowed comfortable movement whilst preventing the animal from turning around in the stall when attached to a neck chain.

(iv) The central cross-piece of the mainframe of the stall was removed for very large and tall animals (ca 600 kg) in order to prevent back-chafing.

(v) One of the horizontal side panels on each side of the stall was removed to facilitate the ease of adjustment of the body harness

and fittings, particularly those of the straps supporting the urine collection funnel.

(vi) The entrance ramps which had been constructed were found, by experience, to be unnecessary as the animals were easily able to step in and out of the stall without them.

(vii) The crates were in continual use for two complete winter periods. Apart from routine daily cleaning and preservation treatment (during the summer when the stalls were temporarily unoccupied) with a commercial wood preservative, very little repair was needed. The feed boxes almost totally prevented spillage of the chopped hay component of the diet given to the cattle. Water spillage was minimal and the cattle generally seemed to be content whilst housed in the stalls.

BODY HARNESS, FAECAL COLLECTION TECHNIQUE AND URINE FUNNEL HISTORY AND DEVELOPMENT

Considerable time and effort was devoted to the development of a highly acceptable system for the complete collection of faeces and urine from large steers which helped to minimise any disturbance and discomfiture to the animals. The stresses caused by the fitting of conventional body harnesses are difficult to measure but have previously been reported as potential sources of error in nutritional and parasitic investigations (e.g. Parkins et al., 1982). Webster, Brockway and Smith (1974) reported a reduction in digestibility when bags were used for faecal collection compared with a polythene chute system and Greenhalgh and Reid (1973) found a reduction of about 9% in dry matter intake when cattle were fitted with faecal bags. Sores, also, are a continual problem with traditional body harness and faecal bag methods of collection. Collection methods which avoid completely the fitting of a faecal bag to a body harness clearly have an attraction such as that developed by Tim Smith at Shinfield (personal communication). However such methods are subject to animal weight limitations and an inherent degree of potential excretary material loss.

Brockway (1979) first described a method for the collection of faeces from steers which utilised a long polythene chute in place of the conventional bag carried by the animal. The increasing weight of faeces collected in the conventional bags contributes greatly to the

causation of sores under the straps of the supporting harness in such systems.

In extensive preliminary experiments conducted using the method of Brockway (1979) the current investigator discovered several problems which made modification to this technique essential. The problems were as follows:

Faeces collected from scouring or semi-scouring steers did not slide freely down the polythene chute but lined the edges and created a 'bottleneck' which resulted in heavy weights of a wet faecal mass accumulating in the chute which ripped the reinforced 'tail hole' of the tubing and also unduly tensioned the supporting harness and ring surround, often dislocating its position. Sometimes the bags became torn and completely detached and in many cases, even with normally constituted faeces, considerable stress and sore points were noted in these steers. This method was abandoned following repeated failures to satisfactorily collect from steers producing semi-soft and soft faeces. The time taken to repair or prepare new tubing satisfactorily reinforced around the tail head area and the tail hole aperture was completely excessive.

The following method (after much trial and error) was finally developed and used in the winter metabolism periods.

(a) A simplified body harness was found to be extremely satisfactory. This was comprised of a front 'chest-strap' and rear 'abdomen-strap', where the chest-strap was made from a cotton woven fire-hose jacket material (4") and the abdomen strap from elasticated upholsterer's seat webbing. These were joined together by two sets of lateral straps. The most important straps were those running 'head to tail' on each side of the backbone extending over the tailhead area of the animal presenting two buckle-ends to which the faecal bag straps were attached. The neck collar (as is often used in complete body harnesses) was found to be unnecessary and was dispensed with. The animal was tethered, quite simply; directly by chain to the fixed chain slide attached to the feedbox of the stall. This body harness was lightweight, quite comfortable and easily adjusted.

Faeces were collected into a large polythene vessel, centrally - hung by chains, and sited immediately below a cow faecal bag which had been cut open at the bottom end (Avon Industrial Polymers Ltd., Wiltshire). Plates 3, 4, and 5 show the collection system in operation. The conventional cow faecal collection bag cut open at the

Plate 3a Collection system for faeces.

Plate 3b Arrangement of bags during periods of non-collection.

Plate 4a Close up of rubber bag with open bottom.

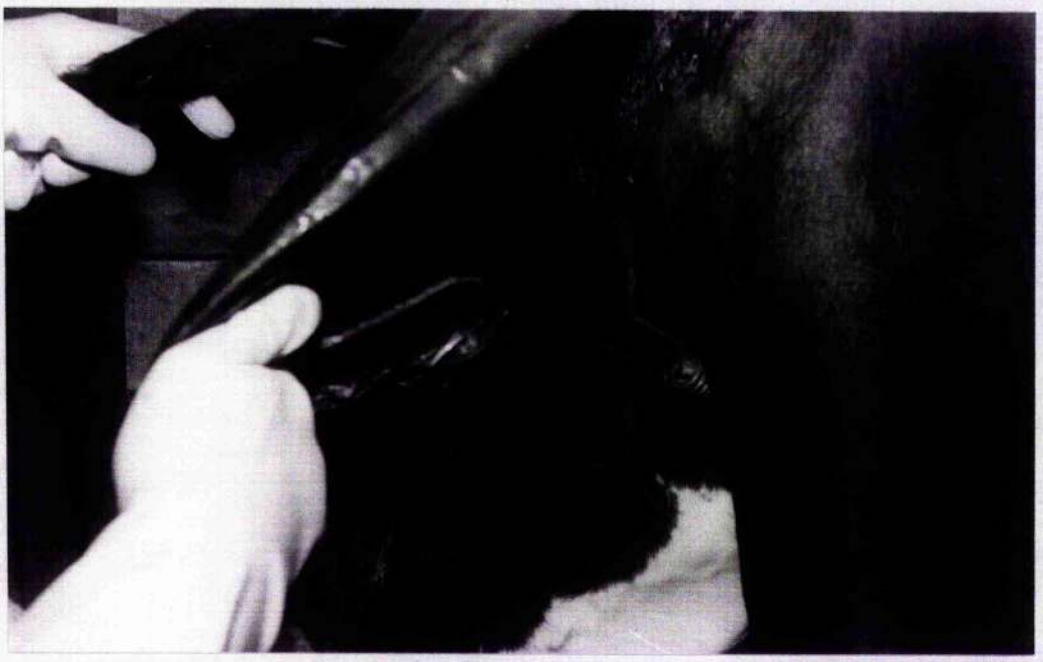
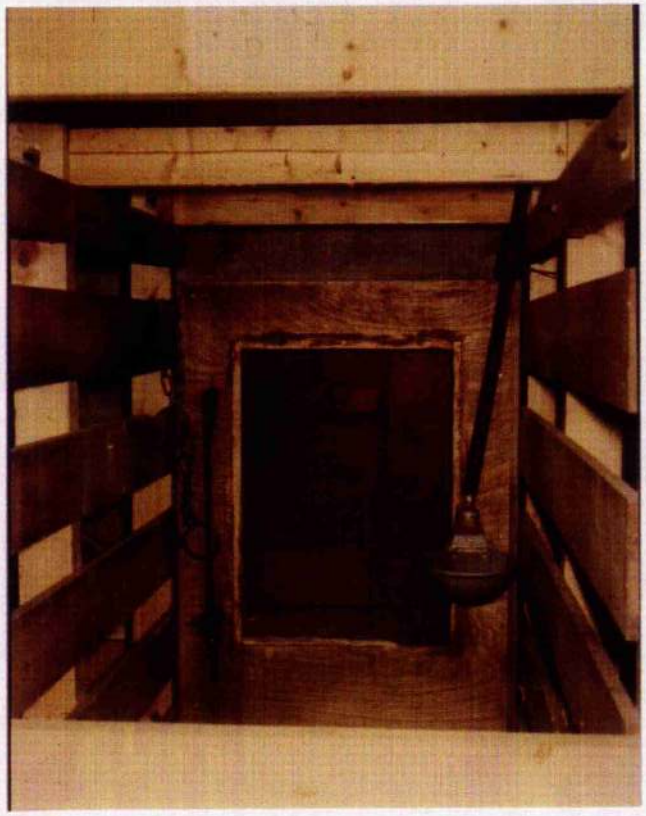
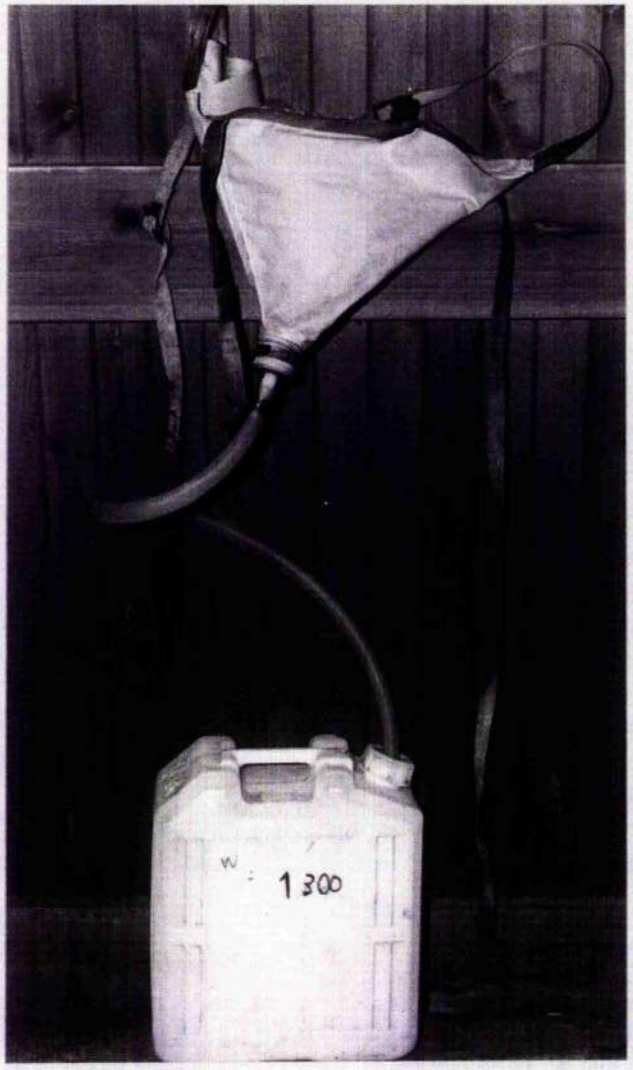


Plate 4b Close up of strap and hook between legs to maintain rubber bag in position.



a)



b)

Plate 5a Interior of metabolism stall.

Plate 5b Funnel for collection of urine by gravity.

bottom thus became a substitute chute for the previously described polythene tube described by Brockway (1979). Its very considerable advantages were that it was substantial in construction and did not need periodical replacing and was directly and properly attached to the body harness without the risk of tearing or dislodgement and was easily cleaned.

An important feature (in modifying a conventional cow bag to act as a faecal chute) was in ensuring as good a 'catch' of faeces as possible. It was found important, to this end, to attach from the body harness, elasticated ropes between the rear legs of the steer directly to the faecal bag at points below the anus in order to ensure a snug fit and almost entirely prevent any faecal loss.

(c) Urine was collected continuously from the steers into a flexible plastic fabric funnel attached to cover the penis of the steer by two light body straps and connected by flexible tube to a previously acidified (12N sulphuric acid, Martin, 1966) polythene vessel sited below the metabolism stall. Very little difficulty was encountered in the successful collection of urine by this elementary but robust method.

Discussion

The essential difference between a conventional harness and bag collection system and the method eventually used in the metabolism work described in this thesis is the conversion of the bag to a faecal transport tube which deposits the faeces into a suspended receptor vessel (plastic dustbin) which can tilt about its support points. There is no accumulating weight of faeces to stress the animal or to shift the harness fixtures so as to disturb the fully efficient collection of faeces.

The system is very strong compared to the relative fragility of the Brockway (1979) polythene tube system, easily maintained and reliable. Lesions caused by harness abrasion were very few. The only sores of note were caused in the early development of the system when the rubber faecal bags were accidentally overtensioned by elastic thigh straps and rubbed the flesh beneath the anus. Faecal losses were minimal and generally resulted from a small faecal leakage at the time of excretion around the tail hole of the faecal bag. In the long term, the cost of the system was economically favourable as the life of the collection apparatus (still in good order after two complete winter

periods of continual use) was considerably greater than the polythene tube system which required continual replacement. From the very important viewpoint of time economy the system was superior in that cleaning and adjustment time was relatively low in comparison to both polythene tube and conventional bag techniques.

Daily routine during metabolism trial work

Feeding The animals were fed 2 x d at 0730 h and 1620 h and given half the daily feed allowance of both hay and concentrate. Any feed refusals were removed and weighed at the feeding at 0730 h each day.

Faecal and urine collection

0800 h. On each day of the 7 day collection period the total faecal output from each steer was weighed and two 2% subsamples taken, bulked and stored at 0-4°C. One sample was taken for dry matter determination and the other for N analysis. Urine output from each steer was collected in acidified polythene vessels. The vessels had been pre-weighed. On each morning the vessel plus urine was weighed. A 2% subsample was taken, bulked and stored in the metabolism house for subsequent analyses.

Cleaning Following feeding, faecal and urine collection, weighing and subsampling the metabolism stalls and building surrounds were washed. (The harnesses were carefully readjusted where necessary and the collection vessels for both urine and faeces refitted immediately after collection to preclude excreta losses during the weighing and subsampling time).

CARCASS EVALUATION AND MEAT ANALYSIS METHODS

MLC Grading at Abattoir

At the end of the main experiment some 24 steers (9 from each of the control and MSRB treated groups and 6 clean control) were slaughtered at Paisley abattoir. As part of the normal commercial procedure the hot carcasses were weighed, inspected and graded by an MLC official who gave an assessment of conformation and fat cover according to the EEC grading scheme. Conformation is scored according to the letters EUROP. Most buyers prefer U+ conformation animals but only about 2% of finished cattle in the UK fall into this category

(Scottish Agricultural Colleges Leaflet, 'Better Beef Marketing', 1984). U and R conformation, which are the most common in Britain, are typically Hereford and Aberdeen Angus crosses. Purebred Friesian may have O and O- conformation at best.

The fatness score is an important assessment. The score ranges from 1 (very thin cover) to 5 (excessive fat cover) with the common scores e.g. 4, having subdivisions ie. 4H and 4L. The grade score is designed to assist dead- market buyers where, naturally enough, different buyers prefer different levels of fat cover. For example, independent UK butchers prefer fatter animals (e.g. fatness 4L) while multiple retailers prefer leaner continental-type (eg. Charolais) crosses with a fatness score of 3. Animals suited for the export market are usually leaner than is generally sought after in Britain (fatness 2) and are typically Limousin, Simmental and Charolais X dairy animals.

However, carcasses failing to achieve a minimum grading of O2 (conformation O and fatness cover 2) are not eligible for the EEC Beef Premium Scheme (BPS). A variable premium system operates in the UK where the government sets a weekly price scale or target price so that when the average national market price of certified fat cattle falls below target price then farmers receive a variable premium equal to the difference between actual market and target price, subject to a maximum of 8.74 p/kg LW (April 1984). (The EEC in Brussels pays 40% and the remaining 60% is carried by the UK Treasury). Currently in UK (April 1984) clawback of variable premiums on exports of beef have been introduced.

The major reason for Friesian and Friesian cross carcasses failing to achieve grading is lack of conformation (Scott, Wholesale Meat Merchant, personal communication).

Carcass Measurements

A simplified assessment of the 39 carcasses obtained in the large two year trial was conducted. There were some differences in technique between the two major slaughter periods.

Detail of the measurements of the skeletal and muscular reference points were taken from the Meat and Livestock Commission (Marketing Department) 'Instructions for assessment, photography, jointing, retail cutting and tissue separation of beef carcasses': a memorandum, 1974 (Kempster, A.J. Personal Communication, 1982).

At the end of the first season grazing in 1982, 11 carcasses (6 from the MSRB treated group and 5 from the control group) were assessed at Glasgow University Veterinary School (1 control calf had been destroyed *in extremis* during the grazing season). Photographs were taken of the intact left side of each animal showing a) lateral view, b) dorsal view and c) a medial view. Each photograph contained a clearly marked 50 cm scale and a card marked with the animal number.

Side weights were recorded for the cold carcass by suspension of the whole side on a spring balance (live weights had been recorded 1 day before slaughter).

A forerib 'indicator' joint was taken from the left side of each set carcass. This standard commercial joint comprised the 7-10th rib and was taken with the rib bones cut as described (MLC, 1974). This joint was also photographed showing the cut 10th rib face (in colour) together with a scale and identification number clearly in the frame.

The carcass and forerib photographic slides were analysed (by reference to the internal markers present in the frame) for various measurements in this year (1982) using a computer-linked image analyser (Magiscan; Joyce Loebl, Vickers Instruments Ltd.) running on a programme written especially for this work (M.G. White, Pharmacology Department, Glasgow University Veterinary School, personal communication, 1982). The important measurements taken from the carcass slides were:

(a) Side length; this being the length of the side, from the middle of the anterior edge of the first rib to the anterior edge of the *symphysis pubis* (SP).

(b) length of hind leg; this being the distance on the hanging side between the most distal point of the remaining tarsals and the anterior edge of the SP.

(c) Thickness of hind leg; distance between anterior edge of SP and the lateral surface of the leg horizontally aligned from the SP.

(d) Width (or depth) of carcass at the 7th rib.

(e) Total length; this being the overall length of the carcass i.e. side length (a) plus the length of hind leg (b).

At the end of the experiment in 1983 the 24 animals were killed at Paisley abattoir where, following grading, weighing and photography of the hot carcass sides the physical measurements described above (a)-(e) were taken directly using a measuring tape. Additionally a further important measurement recorded was that of the circumference of the

hind leg. The computer-linked image analyser was unfortunately unavailable for any further use. Measurements and analyses performed upon the indicator forerib joints were as follows.

(a) The rib was photographed featuring the cut surface in the region of the 10th rib. A ruler was included in one two photographs as an internal marker. In 1982 (following one season at grass) the area of the eye muscle and other linear measures were taken using the previously described image analyser (Joyce Loebl, Vickers Instruments Ltd.,). In 1983 the eye muscle area was determined by an older and more classical method by weighing a cut out portion, which constituted the eye muscle alone, from a life-size monochrome photograph of the joint surface. Several carefully measured 100 cm^2 sections of the same photographic material (10 x 10 cm) had previously been weighed so that the weight of cut-out photograph could be directly related to the area of the eye muscle. Any linear measurements required were taken directly from the life-size photographs.

(b) The rib joints were physically dissected, by hand, into lean, bone and fat tissue. Eye muscle and other lean tissues were further separated from one another. Each dissection took about 20-30 minutes.

(c) Samples of eye muscle tissue were taken from the freshly-cut sample joints for dry matter and crude protein content and (in 1983 alone) water holding capacity. Dry matter content of 3-4 g samples of eye muscle taken in 1982 were determined by an automated Karl Fischer technique and in 1983 by completely freeze-drying the sample using a high-efficiency Edwards Modulyo vacuum freeze drying system.

Crude protein was determined in a 1-2 g sample of fresh tissue by an automated Kjeldahl technique (Kjel-Foss Auto). Water holding capacity % (WHC) was determined following the suggestions of Hamm (1975) where a 3-4 g weighed fresh muscle sample was centrifuged in a Beckmann ultracentrifuge at 50,000 g for 30 minutes. The meat sample was then carefully removed from the centrifuge tube with foreceps, dried with paper tissue and then reweighed to determine liquor loss. The amount of liquor loss expressed as a percentage of the fresh weight of sample was defined as the water holding capacity.

GENERAL INTRODUCTION TO THE EXPERIMENTAL STUDIES

From the literature review it is clear that, compared to sheep, very few studies have been made on the production and metabolic effects of Ostertagia infections in cattle. Where these studies have been carried out, they have examined animals of a particular age group and at a particular stage of the disease; there has been no study in which the same group of cattle, exposed to naturally occurring Ostertagia infection, have been followed from first exposure, at around four months old, to marketing in the case of beef animals, or entering the milking herd in the case of dairy cattle.

In this thesis the results of a study on the production and metabolic effects of naturally acquired Ostertagia on Friesian steers over a two year period are reported, in three groups of cattle. The first group received no special control other than anthelmintic treatment when clinical disease occurred and are referred to as infected controls. The second group received a sustained release bolus containing the anthelmintic morantel tartrate at the beginning of each grazing season, this group is referred to as the MSRB treated group. The third group was kept virtually free of worm infection and is called the clean control group. The experiment can be conveniently divided into three sections and the results are described and discussed within these sections as follows:

Section A covers the period of the first grazing season (May to October 1982) of calves turned out to grass at approximately four months old. Clinical, parasitological and biochemical aspects were monitored and at housing in October 1982, some calves were slaughtered for pathological and carcass evaluation.

Section B covers the period of winter housing in October 1982 to turnout in May 1983 for the second grazing season. Apart from the monitoring of clinical, parasitological, biochemical and performance parameters, a detailed digestibility and nitrogen balance study was made using groups of steers from each group at intervals throughout the winter period.

Section C was originally intended to study the same parameters in the animals during their second grazing season (May to October 1983)

and at slaughter in the autumn. However, for a variety of reasons, they were housed again in the autumn and given a hay plus concentrates ration until slaughter in December 1983 and January 1984. At slaughter a carcass evaluation was undertaken.

A schematic outline of the two year grazing, metabolismand carcass evaluation study is given below.

Age Months	Date	Event	Measurements
0	ca Dec 81	Fr. male calves born Bucket reared	
4-5	May 82	Turned out to first season at grass (140 kg LW) 16 given PARATECT bolus (MSRB) 16 left untreated (Control) 7 treated fortnightly with Fbz (Clean) 6 Control and 6 MSRB killed at housing in October.	LW, epg, plasma pepsinogen, gastrin, blood biochemistry, pasture larval counts, post-mortem pathological and parasitological examination and carcass appraisal.
10	Oct 82	Housed for winter store period, (225-250 kg LW) Target LWG 0.6 kg/d Turned out to grass at 315-360 kg LW	As above, Digestive efficiency and N balance metabolism studies.
15-16	May 83	Second grazing season on same plots. MSRB group given second bolus and clean group treated fortnightly with Fbz.	As in first grazing.
20-	Sept-Jan 84	Housed October. Given concentrate fattening ration.	LW, epg, plasma pepsinogen and gastrin.

Slaughtered at approximately same fat finish in MSRB and clean groups. Control cattle killed in January.	Carcass appraisal.

For ease of presentation the Figures and Tables pertaining to each section are placed in order at the end of the appropriate section. They are labelled A, B or C to indicate the section and then numerically to indicate their order within that section.

SECTION A: PERFORMANCE AND CARCASS STUDIES ON GROUPS OF CALVES EXPOSED TO *O. OSTERTAGI* DURING THE FIRST GRAZING SEASON.

INTRODUCTION

The abomasal nematode Ostertagia ostertagi is recognised as the most pathogenic and economically important gastrointestinal parasite in cattle in the temperate zones of the world (Armour and Ogbourne, 1982). Seasonal fluctuation in the numbers and availability of infective O. ostertagi larvae has been demonstrated by Anderson et al. (1969) and by Michel (1969). Several Western European authors (see review) have confirmed that in spring the pastures are still contaminated by relatively low numbers of infective larvae which survived the winter. Cattle which are infected by these overwintered larvae eventually produce faeces containing fresh parasite eggs which are deposited on the pasture and develop to L_3 resulting in an accumulation of large numbers of L_3 from about July onwards. These larvae persist during autumn and then there is a steady decline in numbers until the following spring.

Epidemiologically the important period of pasture contamination with O. ostertagi eggs is during the spring and early summer. Several experimental attempts have been made to limit contamination in the early part of the grazing season in order to prevent the increase in larval infestations which occur later (Michel, 1969; Michel et al., 1970; Armour, 1978). Some of these studies used repeated anthelmintic treatment in the spring, sometimes combined with a change of grazing during the summer (e.g. Michel and Lancaster, 1970; Oostendorp and Harmsen, 1968; Armour and Urquhart, 1974).

In an attempt to overcome some of the practical difficulties associated with repeated treatments a novel concept was described by Jones (1981) and involved the use of an intraruminal device designed to provide a sustained release of the anthelmintic morantel tartrate over a sixty day period. It was suggested that the bolus should be administered prior to turnout in spring and that the steady release of the drug would prevent contamination of the pasture during the vital spring/early summer period. Results from five English farms showed the 'combined' liveweight gain benefit in bolus treated animals to be 24.5 kg compared with the untreated animals - even allowing for the fact that 28 untreated calves on 3 of the farms had to receive medication during the trial period because of severe clinical gastrointestinal parasitism. The same beneficial trend was shown by Jacobs et al.

(1981), using a bolus which released the drug over 90 days. The animals treated with the bolus immediately before turnout to grass and showed a higher liveweight gain over the grazing season of 19 kg/head.

Using a 60 day bolus in Sweden, Tornquist et al. (1981) reported the results of two trials (conducted on the same farm) where a combined mean liveweight gain advantage of 21.5 kg was observed in bolus treated animals. Tolling et al. (1981) investigated the effect of a morantel sustained release bolus (MSRB), programmed to release the drug over a 90-day period and administered at spring turnout compared to a similar bolus treatment given in the middle of the grazing season. The liveweight gain benefit from the early season treatment was 19.9 kg over that of control animals whereas the mid-season treatment provided only a 10.0 kg liveweight gain advantage.

Armour et al. (1981) in Scotland, in a similarly designed experiment, demonstrated a 24 kg improvement in calves given a 90-day MSRB treatment at spring turnout compared with only a 9 kg advantage over untreated animals when the treatment was delayed until mid-season. Helle and Tharaldsen (1982) in Norway found a 29 kg liveweight gain advantage where calves were given two MSRB treatments, one before turnout to grass and the second given 90 days later. In a second experiment Tharaldsen and Helle (1982) using animals treated only at turnout reported a 44 kg increase in weight gain compared to an untreated control group in which two animals died exhibiting symptoms of severe parasitic gastroenteritis. Later results from Austria, Netherlands, France and Sweden (Prosl, Supperer, Jones, Lockwood and Bliss, 1983; Borgsteed, 1983; Raynaud, Jones, Bliss, Le Stang and Kerboeuf, 1983; Tornquist and Tolling, 1983) have showed similar beneficial results in animals treated with the MSRB at turnout in spring.

Clearly, liveweight gain has been used as an obvious appropriate production parameter in such parasitological investigations. However, further parameters such as changes in body composition obtainable from detailed measurement and analyses of the carcass should provide even more data on the economic effect of gastrointestinal parasitism.

Increased body water was reported by Orraca-Tetteh and Platt (1964) together with a decrease in body fat and protein in rats infected with Nippostrongylus muris. Halliday et al. (1965) studied changes in total body water and solids in cattle infected with Ostertagia spp. In experimental type I infection no differences were

found but in a naturally acquired type II infection total body solids were 21.3% of body weight compared with 37.5% in an uninfected control group.

In an experimental chronic infection of sheep with Trichostrongylus vitrinus, Sykes et al (1979) found a significant reduction in the concentration of protein in the fat-free carcasses of infected animals. This was directly associated with a significant increase in the degree of hydration.

Steel et al. (1980) using sheep infected with T. colubriformis examined the interactive effects between larval exposure levels and production losses, plus physiological and metabolic changes associated with the development of disease. Inter alia they reported that an increased level of infection produced a decrease in the percentage of dressed weight and dry matter of the carcasses.

This current report details results from the first stage of a two-year production and metabolism study with young Friesian calves treated with the MSRB bolus. The result of the first summer grazing period are presented in which some epidemiological, parasitological and pathophysiological aspects are considered.

MATERIALS AND METHODS

Experimental Design

The object in the first summer grazing period was to follow the epidemiology of naturally acquired ostertagiasis in young cattle with especial reference to faecal egg counts, herbage larval numbers, worm burdens, plasma pepsinogen levels, plasma gastrin levels, blood biochemistry, liveweight and carcass characteristics under a split-plot grazing management.

Animals

On May 13th, 1982, 39 Friesian castrated calves aged 14 to 17 weeks, of mean liveweight 140 kg which had been reared under helminth-free conditions were grazed in three groups on adjacent separate grass paddocks known to be contaminated with O. ostertagi larvae.

Each group of calves was balanced with respect to the distribution

and mean of liveweight. The calves were treated as follows: the control group consisted of 16 calves and was allowed to become naturally infected with O. ostertagi and had no intended anthelmintic treatment; the clean group of 7 calves was given a two-weekly anthelmintic treatment with fenbendazole at fortnightly intervals (Panacur; Hoechst) at 7.5 mg/kg liveweight, the first treatment being administered after one day at grass; the MSRB treated group of 16 calves was given a morantel sustained release bolus (MSRB; PARATECT, Pfizer Ltd.) at turnout. Fenbendazole was chosen for the clean group because of its high efficiency against all stages of O.ostertagi (Duncan et al. 1976).

Two weeks after treatment each calf in the MSRB treatment group was scanned with a metal detector (C-scope) in order to confirm the presence of the boluses.

Grazing ended on October 13 for the control and MSRB treated groups and one week earlier for the clean group because of an acute grass shortage in that paddock. The animals were housed indoors thereafter and given a dietary allowance sufficient for maintenance plus a daily liveweight gain of about 0.5 kg in accordance with an accepted normal husbandry practise for such animals. One week after housing six calves (the three heaviest and the three lightest) from the MSRB treated group and five calves (three heaviest, two lightest) from the control group (one light control calf having already been destroyed in extremis whilst still at grass) were slaughtered and examined for the presence of parasites (Ritchie et al. 1966). These animals were taken for slaughter in order that the groups of remaining calves, destined for study during the winter and ensuing spring periods, would be as uniform as possible (within any one group) with respect to liveweight. The animals were maintained indoors for seven days to permit the maturation of larvae beyond four days and so allow the differentiation between arrested and developing larvae (Armour et al. 1979). The carcasses were examined with respect to dressing percentage, linear measurements, 7-10th rib sample joint dissection and chemical analysis following the suggestions of Kempster and Jones (1977) and Kempster (1982, personal communication).

Grazing Paddocks

The total grazing area (situated in one field) comprised 6.2 ha. This was divided into five units, four units of 1.2 ha in area each and

a further area (for the smaller clean group) of 1.4 ha. Grazing plots grazed by the control and MSRB treated calves were separated by a double fence (1.5 m apart). Water was supplied to each plot in drinking troughs. The mean stocking rate over the whole grazing season was about 6 calves/ha. The paddocks were fertilized with a total of 200 kg N/ha in four separate applications each of 50 kg N/ha applied shortly after each completed grazing of the plots. The clean group was set-stocked on the 1.4 ha plot. Each of the control and MSRB treated groups were randomly allocated to two of the four equal split-plots at the beginning of the trial. The control and MSRB treated groups were then alternated between their fixed allotted plots as determined by the availability of the grass. Both groups were moved to their alternate plots on the same day. There were four separate grazings of the alternate plots. Each grazing period lasted about five weeks on average (22 weeks total grazing) although the initial grazing time was longer being nearly seven weeks due to a plentiful grass supply in the early season whereas in mid-season a dry spell curtailed later luxuriant growth in the secondary plots.

Observations

All animals were clinically examined and faecal sampled once a week. The animals were weighed and blood samples collected at two weekly intervals. Herbage samples from each of the five separate plots were collected each week.

Rainfall and temperature data were collected from the meteorological station at nearby Glasgow Airport. Rainfall in mm and minimum temperatures in degrees are shown in Fig. A1.

Parasitological Techniques

Faecal samples were examined for the presence of trichostrongyle eggs by a modified McMaster flotation technique (Gordon and Whitlock, 1939) and also by Baermann extraction for Dictyocaulus viviparus larvae. The numbers of trichostrongyle larvae on the herbage samples were estimated by a similar technique to that of Parfitt (1955).

At post mortem the lungs and gastrointestinal tract were removed and subjected to examination as described by Inderbitzen (1976) and Ritchie et al (1966), respectively.

Pathology

At post-mortem the gross appearance of the abomasal mucosa was noted and the presence of lesions described and, where appropiate, photographed. In addition an attempt to quantify the apparent degree of damage was made by formulating an index based on the weight of abomasal mucosa associated with each kilogram of liveweight.

$$\text{i.e.} \quad \text{Mucosal Fraction} = \frac{\text{Abomasal mucosa weight (g)}}{\text{Liveweight (kg)}}$$

Blood Analyses Methods

Jugular vein blood samples were collected into heparin (Vaccutainer; Becton Dickinson) and the plasma stored at $-20^{o}C$ until subsequent batch analyses. Plasma pepsinogen was determined by the method of Edwards et al (1960) and results expressed as international units of tyrosine (I.u.). Gastrin was measured using the Cambridge Medical Diagnostic, Inc. Gastrin Assay and it is expressed in pico grams of gastrin per millilitre (pg/ml). For this assay five animals from each group were sampled on eight occasions, namely weeks 0, 2, 6, 8, 12, 16, 20, and 22. Other blood analyses were performed on all animals every two weeks. Analyses for urea and phosphate were performed by standard automated techniques (Technicon Auto Analyser II) by which total protein was also determined using a biuret method (Weichselbaum 1946) and albumin concentration by a modification of the technique of Rodkey (1965).

Carcass Measurements

Dressing percentage was estimated with the kidney knob and channeling fat (KKCF) present in the cold carcass and expressed as that percentage of the liveweight of the animal which is presented as a dressed carcass (i.e. less weight of gut contents and gut offal, head, hide, horn, hoof and blood).

Dressed carcass sides were photographed with a marked 50 cm linear reference indicator present in the frame. Linear measurements of carcass sides e.g. total length, side length, leg width and length were made directly from the photographs using slide projection analyses. An indicator joint consisting of the 7-10th ribs inclusively ('forerib') was removed from the left side of each carcass. The joint was photographed featuring the cut 10th rib face. An estimate of the total

area of the eye-muscle was made from these photographs using a computer-linked image analyser programme (Magiscan, Joyce Loebl, Vickers Ltd.) The indicator joint chosen in this study followed a careful consideration of the work of Kempster and Jones (1977) and Kempster (personal communication) where dissection data were used to predict the lean contents of carcass sides from a direct measurement of the lean content of standardised wholesale joints. In summarising the results from 753 steer carcasses Kempster and Jones (1977) commented that "the fore rib probably offered the best compromise between cost and precision" in this respect.

The joint was physically dissected and measurements made for total joint, bone, fat, total lean tissue and eye muscle weights. Samples of eye muscle were taken for the chemical analysis of total water (Karl Fischer, Baird & Tatlock Ltd.) and total nitrogen content by an automated Kjeldahl procedure (Kjel Foss, York, U.K.).

Statistical Analyses

Statistical analyses were performed by computer (ICL 2988; 'Minitab'). Most considerations of group mean value comparisons used the standard two-sample 't' test. A few alternate analyses using the Mann-Whitney two sample rank procedure examined some of the carcass data. (A non-parametric approach may be justified here since the animals which were killed were selected as the three heaviest from each of the groups or the three lightest animals from each group, thus presenting a non-Guassian distribution). Regression analyses were also performed using 'Minitab' (Ryan, Penn State University, 1981).

RESULTS

Grazing

At turnout all the paddocks were similar with respect to grass availability following an initial 'early bite' application of 50 kg N fertilizer/ha. The overall stocking rate was 6.7 calves/ha for both control and MSRB treated groups and somewhat less (5.0) for the set-stocked plot assigned to the clean group. This paddock was affected by adverse weather during the experiment. A prolonged mid-season dry period beginning at the end of July followed by very heavy rains and some flooding in early September resulted in the necessity for some supplementary feeding (2 kg hay/hd/d) for a two week period (weeks 14-16, mid August-September) and again for a further 18

Plate 6a Pasture grazed by controls - August 1982.

Plate 6b Pasture grazed by MSRB group - August 1982

day period before the calves were housed. The paddocks grazed by the control and MSRB treated groups were only affected by the dry period when grass growth was very poor, but no supplementary feeding was given.

The paddocks and animals were inspected daily and it became visually apparent in the later stages of the 22 weeks of grazing that the grass eaten by the MSRB treated group was in short supply (particularly weeks 18-22) whereas a more plentiful supply of growth was still available to the control group (Plate 6). There were three nights of ground frost in the week preceeding housing.

CLEAN GROUP

Clinical Findings

The clean group treated every 14 days with 7.5 mg fenbenazole/kg LW remained apparently clinically normal throughout the experiment.

Liveweight

Mean bodyweights of all three groups are shown in Fig. A2. Up to the end of July (week 12) the overall mean liveweight gain in the clean group was 75 kg (0.92 kg/d) which was regarded as normal. The prolonged dry period however reduced the regrowth availability resulting in a lowered daily liveweight gain even with supplementary hay feeding so that by the time of housing the overall daily liveweight gain had fallen to 0.67 kg/d (see Table A1).

Plasma pepsinogen activity

Plasma pepsinogen levels (I.u. tyrosine) recorded in the three groups of calves are shown graphically in Fig. A3. Values shown for the clean group remained consistently low throughout the experimental period with normal values of about 1.0 I.u.. Mean values (± SE) are shown in Table A2.

Plasma gastrin assay

Mean plasma gastrin levels in the three groups of calves are shown in Fig. A4 and the mean values (±SE) in Table A3. An increase was observed in the mean gastrin levels by the second week from the initial mean value of 148 pg/ml to a mean value of 320 pg/ml. Thereafter a sustained decrease occurred until week 20 when mean levels of 82 pg/ml

were recorded. A small increase then occurred in week 22 to 117 pg/ml.

Faecal egg counts

Mean trichostrongyle egg counts in faeces (epg) are shown in Fig. A5. While occasionally a very low egg count was observed for this group, at all other times no trichostrongyle eggs were detected. Mean egg counts (including the presence of lungworm larvae) are detailed in Table A4.

Pasture larval counts

Figure A6 details the recoveries of O. ostertagi L_3/kg DM from the paddocks grazed by the clean, MSRB treated and control calves. Larval recoveries from the clean group set-stocked paddock were zero except in weeks 3, 4 and 13 when 145, 100 and 536 L_3/kg were recovered respectively. These were not considered to be significant. Larval counts including Cooperia spp., Nematodirus helvetianus and Dictyocaulus viviparus are given in Table A5.

MSRB TREATED GROUP

Clinical findings

The faeces of the calves were normal throughout the experimental period. However, some coughing occurred which was first noted during week 14 (mid-August) coincident with the shortage of grass and by the end of week 15 coughing was observed in 8 of the 16 animals in the group. The group was then treated on three consecutive days with a diethylcarbamazine preparation (Dicarocide 40% w/v, Willows Francis Veterinary) for the treatment of lungworm.

Liveweight

Early in the grazing season when the grass supply was abundant the calves increased in mean liveweight from 140 kg at turnout to 214 kg after 12 weeks grazing (i.e. a daily liveweight gain of about 0.9 kg). However during the subsequent period of grass shortage in the dry period the growth rate declined so that at housing the overall daily gain had declined to 0.68 kg/d, the mean liveweight gain in the last 8 weeks of grazing being only 30 kg. (See Fig. A2 and Table A1).

Plasma Pepsinogen activity

There was a slight rise from 0.9 to 1.5 I.u. by the end of week 4

in this group but thereafter values remained normal at about 1.1 I.u. with a further non-significant return to 1.5 I.u. by week 20 (see Table A2).

Plasma gastrin assay

A similar trend of plasma gastrin levels to those of the clean animals was observed in this group. By the second week the mean values had increased to 465 pg/ml from an initial level of 125 pg/ml and thereafter declined steadily until week 22.

Faecal Egg Counts

Mean trichostrongyle faecal egg counts are given in Table A4 and displayed in Fig. A5. Low levels (3 to 31 epg) were observed from weeks 3 to 14. The mean values rose slightly thereafter (range 16-53) for the remainder of the experiment. During week 16 the presence of D. viviparus larvae was positively confirmed.

Pasture larval counts

Occasionally low numbers of O. ostertagi L_3 were recovered (Fig. A6) in June to August (c. 200/kg DM) with a maximum value of 2,535 L_3/kg DM being recorded at the end of grazing in October when the grass was in very short supply. Examination of Table A5 shows the finding of D. viviparus L_3 in June, July and August the highest count being 306/kg DM in mid-July. The presence of Cooperia spp. L_3 was also recorded with low levels of about 200 L_3/kg DM in August and September the maximum value of 1127/kg DM being recorded during the last week of grazing.

CONTROL GROUP

Clinical Findings

The appetite and general body condition of this group appeared normal, these animals having a good daily growth rate until week 12 of the trial. At this time some animals were noticed to be coughing. Two weeks later 12 of the 16 animals in the group were coughing to some extent and their condition had deteriorated significantly with a noticeable loss of appetite. Additionally and importantly at this time (week 15; at the end of August) some 50% of the animals were exhibiting loose faeces. The presence of lungworm larvae in the faeces and on the

pasture were confirmed and the calves of this group were also treated (but not the clean group), on three consecutive days with a diethylcabamazine preparation as previously described.

One week later (week 16) many of the animals were diarrhoeic and inappetant, one animal (No. 85) displayed such severe clinical signs of parasitic gastroenteritis to warrant early slaughter on 3rd September in week 16. Following careful observation of these animals it was decided in week 18 to treat all of the animals in the control group with a therapeutic dose of levamisole (Nilverm; ICI Ltd.) since this would remove a large proportion of adults and developing larvae but not the inhibited 4th stage O. ostertagi larvae (Reid et al. 1968).

Liveweight

A mean liveweight gain of 74 kg was recorded (Table A1, Fig. A2) from turnout until week 12 of the experiment (average daily liveweight gain 0.9 kg) which was similar to that of clean and MSRB treated groups.

After this time the animals were seen to be coughing and diarrhoeic. Following treatment with diethylcarbamazine and subsequently levamisole the animals failed to improve significantly in performance, gaining only a mean of 11 kg overall from weeks 12 to 22 of the experiment even though an adequate grazing supply was still available. This reduced the overall mean daily liveweight gain (0-22 weeks) in the grazing period to 0.55 kg which was significantly lower ($P < 0.05$) than the mean of about 0.68 kg observed in the clean and the MSRB treated groups.

Plasma Pepsinogen activity

Fig. A3 shows an initial increase in mean plasma pepsinogen to 1.8 Iu tyrosine 4 weeks after turnout to grass. There were no clinical symptoms evident at this time. After week 12 the levels rose dramatically to a maximum mean value of 5.9 in week 16. At this time clinical signs of acute parasitic gastroenteritis were seen in many animals. Following the single treatment with levamisole the pepsinogen value fell to 4.9 by week 20 but sharply increased again to 5.9 at the termination of grazing (Table A2).

Plasma gastrin assay

As in the other two groups an increase was recorded in the second

week of grazing from a mean of 93 to 296 pg/ml although the levels reached were slightly lower. Thereafter a decrease was observed until week 12. A sharp increase occurred in week 16 from a mean of 202 to 1212 pg/ml, simultaneously with the elevation of the plasma pepsinogen levels and the appearance of type I ostertagiasis. A decrease to around 830 pg/ml was observed in week 20 and 22, levels which were still approximately seven times the values given by the clean and MSRB groups.

Faecal Egg Counts

Mean trichostrongyle egg counts in the faeces of the untreated control group are shown graphically in Fig. A5 and numerically in Table A4. An initial peak of egg output was seen in week 6 (24 June) with a mean of 190 epg being recorded. Egg counts then decreased gradually to an average of 34 epg by week 11. A sharp increase was then recorded over the following 5 weeks until a maximum value of 413 epg together with the confirmed presence of D. viviparus larvae was observed in week 16 (1 September). Levamisole treatment was given in week 18 after which a sudden drop in egg count was seen. However by the end of the grazing period the egg output had increased again to a mean of 237 epg.

Pasture Larval Counts

In the first 8 weeks of the trial only low levels of infective O. ostertagi larvae (and some Nematodirus spp.) were recovered from the paddocks, (Fig. A6 and Table A5). After this time a dramatic increase in O. ostertagi infective larval numbers was seen rising to a maximum value of 29,500 L_3/kg dried herbage in week 19 (late September).

Cooperia spp. values also increased sharply in week 12 from 667 to 3,750/kg DM by the end of grazing in week 22. D. viviparus larvae were also recorded from herbage samples in week 5 and again in week 15.

Blood Analyses

Mean analyses for urea, phosphate, total protein and albumin on blood samples collected on six separate occasions during the course of the grazing period are given in Table A6.

Statistical comparisons are shown between the treatment group means for any one occasion.

Results for urea, phosphate and total protein all showed apparent statistical differences between groups on several sampling occasions.

However, the values all lay within an accepted 'normal' range and no particular biological significance can be interpreted from these data.

The mean albumin concentrations in the control group of calves remained fairly constant throughout the grazing period at about 31 g/l. However, from week 14 until the end of the grazing this value was significantly lower than the mean values recorded for the other two groups. Mean albumin concentration tended to increase in the control and MSRB treated groups as the season progressed.

POST-MORTEM EXAMINATION

Bolus Recovery

At the last weighing examination before removal from grass all MSRB treated calves were checked positively by metal-detector for the presence of the bolus. All boli were recovered from the rumen-reticulum of the six MSRB treated calves killed one week after housing.

Pathological Observations

At post mortem classical lesions of ostertagiasis i.e. coalescence of nodules, hyperaemia and oedema were clearly evident in the control animals (Plate 7). In contrast minimal lesions were present in the MSRB treated animals.

As might be expected from the gross pathological changes the mucosal fraction in the control animals ranged from 3.4 to 5.0 and were significantly higher than those recorded in the MSRB treated group whose values ranged from 1.4 to 2.3.

Parasitological Findings

Table A7 details the worm burdens of O. ostertagi, Cooperia spp. and N. helvetianus and D. viviparus of the control and MSRB treated calves necropsied after the grazing period. The pH of abomasal fluid at post-mortem is also recorded. The post-mortem findings in calf 85, a control animal slaughtered in extremis on 3 September are included.

Control calves had a significantly higher pH (5.2) compared with the MSRB treated calves (3.4). Total numbers of O. ostertagi worms were highly significantly greater in the control calves (c. 320,000) compared with those found in the MSRB treated calves (17,500). In the

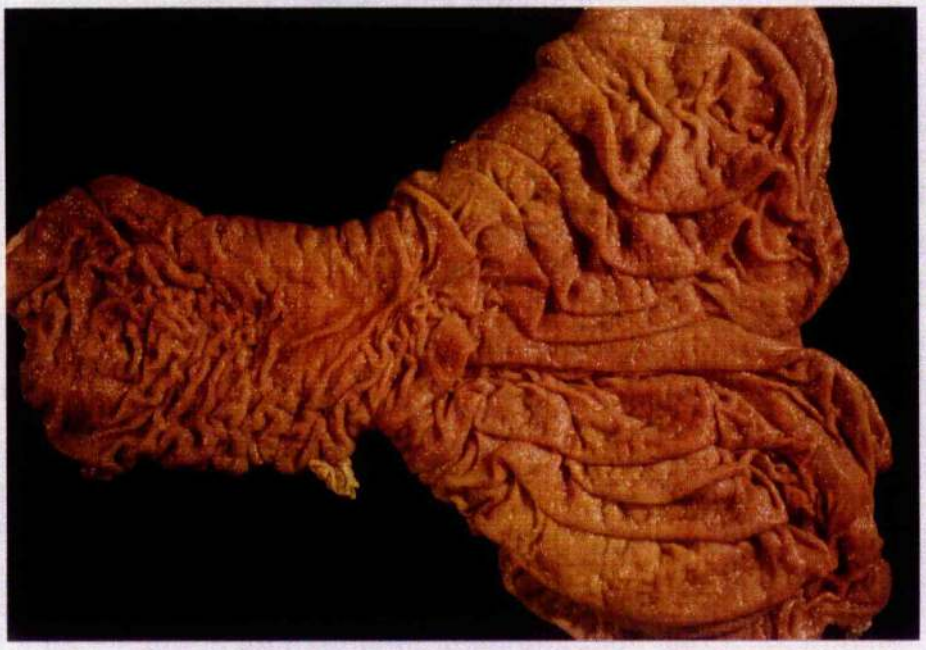


Plate 7a Abomasum from control calf with Type I ostertagiasis showing hyperplasia and oedema.

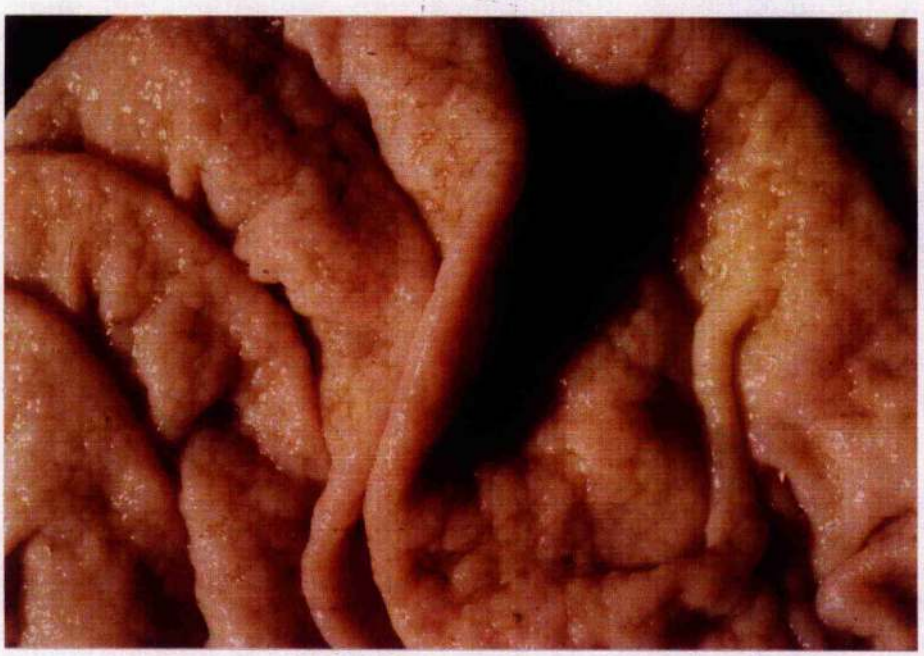


Plate 7b Close up of above abomasum showing nodules, hyperplasia and oedema.

control group 87% of the total were L_4 compared with 70% in the MSRB treated group.

Only small numbers of N. helvetianus were recorded in both control and MSRB treated groups (302 and 85 respectively). At necropsy two of 5 control calves had lungworm present with one calf in 6 MSRB treated calves also having D. viviparus present. The control calf (No. 85) killed in extremis had a total of c.150,000 O. ostertagi in total but not unexpectedly only 3,500 of these were L_4 stage larvae since slaughter was before the normal season of larval arrestation.

Carcass Analyses

As might be expected with 10 month old Friesian steers (230 kg liveweight) after one grazing season the set carcasses were very lean with only minimal fat cover. Linear measurements after photography and picture analyses of the side length, leg length and width, side width at the 7th rib all showed no statistical differences between control and MSRB treated groups when all values are considered, the mean values being very similar. (Table A8). Also linear measurements of the length and depth of the eye muscle at the cut 10th rib of this sample joint showed the mean values of each to be very similar between control and MSRB treated groups).

The dressing or killing out percentage was significantly better (49.4 compared with 46.9) in the MSRB treated calves. A separate statistical consideration compared independently the mean results of the three heavy calves (labelled b in Table A9) in each group with one another and also the mean results from the lightest calves in each group (labelled a in Table A9). No significant difference was seen between the total group means for carcass side length. However when light and heavy calves are compared between groups separately, MSRB treated calves were shown to have significantly greater carcass side lengths ($P < 0.001$). (Plate 8).

The 7-10th forerib joint dissections showed the total fresh weights of the joints to be 2575 g for all MSRB treated calves compared with 1990 g for all control calves (not significant). However consideration of the lighter calves alone (a) showed MSRB treated calves to have highly significantly heavier joints than the control calves. (Table A9) Dissection and weighing of the eye muscles of these joints showed a significantly heavier value for MSRB treated calves (661 g

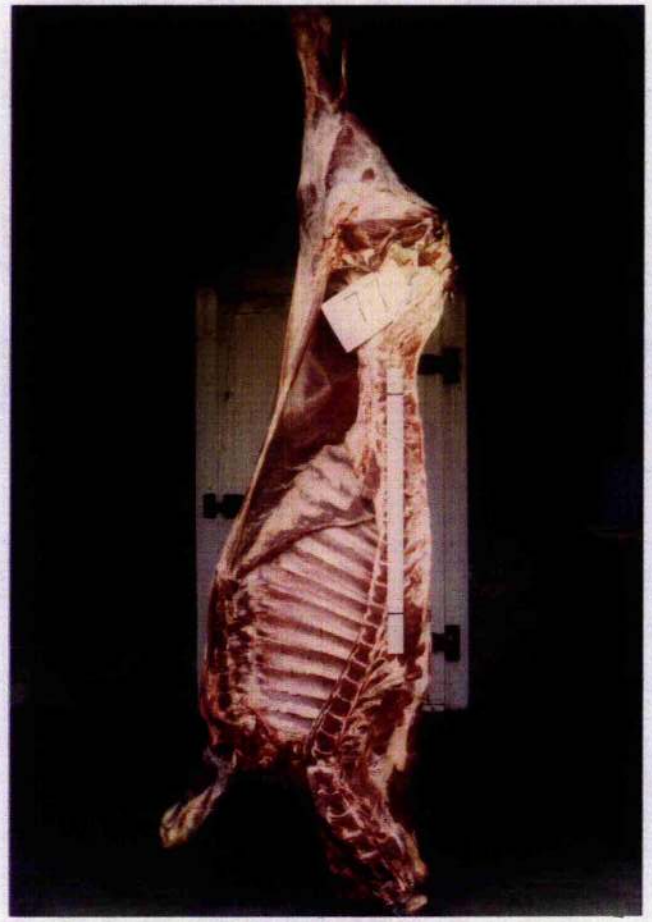


Plate 8a Split carcass from control calf with 50cm marker.

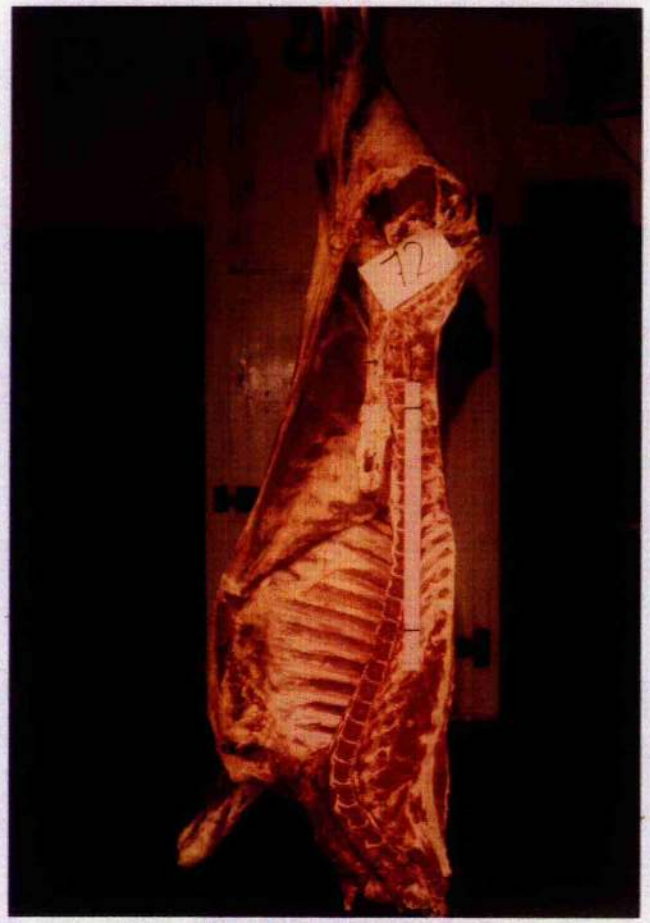


Plate 8b Split carcass from MSRB calf with 50cm marker.

compared with 506 g for the control calves) largely due to the higher weights of eye muscle observed in the 'lighter' calves (Table A10). Measurement of the eye muscle surface area (cm^2) at the cut 10th rib surface showed MSRB treated calves to have a significantly greater muscle area than control calves (55.0 compared with 46.1) again mainly due to the eye muscles of lighter calves in the control group being much smaller than the corresponding 'light' calves in the MSRB treated group.

Fat content of the sample joints in both groups was very small (c. 4.5% of total joint weight) and was not significantly different. Bone content as a percentage of total joint weight was also very similar at about 34%. However, the total weight of lean meat was ($P<0.01$) greater (1621 g) in the MSRB treated group compared with the control group (1190 g) because the lighter MSRB treated animals (a) were very much greater than the control joints.

Indicator 7-10th Rib Joint Eye Muscle Analyses

The weights of the dissected eye muscles from the 7-10th rib indicator joints together with the analyses for dry matter content (g DM/kg FM) and crude protein (g CP/kg DM tissue) are given in Table A10. Also shown are the total weights of dry matter and crude protein of the eye muscles.

The mean dry matter content was significantly greater in the eye muscles of the MSRB treated group (301 g) compared with the mean of the control group (274 g). The mean crude protein content of the dry matter of this tissue was higher in the control group (830 g/kg) compared with the mean of the MSRB treated group (771 g/kg). This is assumed to be as a result of a lower intramuscular fat content in the eye muscle tissue of the control group. Calculated mean total dry matter and crude protein weights of the eye muscles were both significantly greater in the MSRB treated group compared with the control group means.

Correlation studies

Correlation studies examined the linear relationship between plasma pepsinogen and gastrin levels in five calves per group during the summer grazing season when clinical typeI ostertagiasis occurred in the control cattle. A total of 73 plasma samples were assayed for both pepsinogen and gastrin during this period.

The relationship: Gastrin = 126 + 77.7 pepsinogen

(where gastrin is measured in pg/ml and pepsinogen as I.u. tyrosine) had a significant r^2 value of 44.4%.

Individual values for plasma pepsinogen, gastrin, abomasal fluid pH, L_5 + adult O. ostertagi and total worm burdens observed at necropsy are included in Table 11. The necropsy results showed that the mean values for abomasal weight, mucosal fraction, pH, plasma pepsinogen and gastrin and all groupings of larval numbers of O. ostertagi are significantly greater in the control group compared with the MSRB treated animals.

Table A12 details some linear regression equations obtained using the data shown in Table A11. Figures A7 to A10 inclusive are included as examples of the computer printout obtained when plot and predictor regression analyses were performed on this data using the 'Minitab' (Ryan, Penn State University, 1981) statistical package run on a Sirius 2.4 Mbyte microcomputer linked to a Fujitsu 830 printer.

Where predictors are used to calculate plasma pepsinogen values (Equation numbers 1-4 inclusive), a high r^2 value was obtained only when the number of L_{5A} had a high degree of confidence in an r^2 value of 94%. In contrast, regression of pepsinogen and L_4 alone as a predictor gave an r^2 value of only 38% (equation 3), which expectedly improved to 46% when the total worm count (i.e. including L_{5A}) was tested as a single predictor (equation 2). This suggested that it was the numbers of developing and actively metabolising O. ostertagi i.e. the L_5 and adults which related most directly to the pepsinogen values observed and not the numbers of arrested or inhibited L_4 larvae present.

However, relationships with pH (viz. worm burdens, mucosal fractions etc.) are generally clearly curvilinear in nature as is exemplified in Fig. A10 where pepsinogen is directly related to pH. This is equally so for the relationship between gastrin and pH, where a linear analysis shows only a poor relationship (Equation 9). The gastrin values were directly related to pepsinogen values (Equation 10) with an r^2% of 58.0. However, in an analogous manner to the pepsinogen relationships, gastrin also was linearly related to L_{5A} with the highest degree of confidence (r% 69, Equation 7) and only less well with L_4 larvae (r% 43, Equation 8). Multiple regression relating the numbers of L_{5A} to the two predictors in both plasma pepsinogen and gastrin assays showed and extremely high r^2 of 95.4% (Equation 11).

Table A12. Regression analyses performed on data obtained at post mortem on Control and MSRB calves after one season at grass.

Equation form: $Y = b_o + b_1x_1 + b_2x_2 \ldots\ldots$

Key: PP = plasma pepsinogen (I.u. tyrosine)
Gas = gastrin (pg/ml)
L_{5A} = Numbers of L_5 larvae = adult *O. ostertagi* x 10^{-3}
TW = total number of *O. ostertagi* worms x 10^{-3}
Mu = wt g of abomasal mucosa
MuF = g abomasal mucosa/kg LW
pH = pH
L_4 = numbers of L_4 larvae x 10^{-3}
b_o = constant
r^2 = r^2 adjusted for d.f.

Equation No.				r^2%
1	PP	=	1.06 + 0.107 L_{5A}	94.1
2	PP	=	1.86 + 0.0099 TW	46.0
3	PP	=	2.04 + 0.01 L_4	38.5
4	PP	=	−4.11 + 1.80 pH	62.3
5	pH	=	3.38 + 0.038 L_{5A}	60.6
6	pH	=	3.61 + 0.0045 L_4	40.2
7	Gas	=	94.20 + 7.6 L_{5A}	68.9
8	Gas	=	144.00 + 0.88 L_4	43.3
9	Gas	=	−124.00 +92.2 pH	17.7
10	PP	=	0.90 + 0.01 Gas	58.0
11	L_{5A}	=	9.35 + 7.33 PP + 0.024 Gas	95.4

DISCUSSION

The weather pattern in the summer of 1982 was fairly consistent for that of the West of Scotland, with some rain occurring in all bar one month. No rain fell in August and this dry summer period of approximately five weeks extended from late July until the beginning of September. Despite the longer than usual dry period in summer, the pattern of O. ostertagi L_3 population on the herbage grazed by the control animals was similar to that previously reported (Armour and Ogbourne, 1982). Thus, low numbers of L_3, which had survived from the previous year, were present in the spring. Following ingestion of these larvae by the calves, eggs first appeared in the faeces three weeks after turnout and increased steadily over the following months. The larvae resulting from these eggs reached the herbage from mid-July onwards, eventually attaining the peak (of almost 30,000) L_3/kg dried herbage (kdh)) in late September.

Coincident with the increasing levels of O. ostertagi larvae on the pasture, clinical type I ostertagiasis occurred in the control group; this necessitated treatment with the anthelmintic levamisole. Associated with the onset of clinical disease there was a sharp reduction in appetite and a marked elevation in both plasma pepsinogen and plasma gastrin levels. The timing of these events is similar to that described following experimental infection in cattle with O. ostertagi (Murray et al., 1970) and in sheep with O. circumcincta (Anderson, 1976, 1981), in which the gastrin levels were also measured. A numerically lower, but similar, trend in pasture larval numbers of C. oncophora was also noted. The occurrence of N. helvetianus and D. viviparus larvae were more sporadic although lungworm numbers were sufficient to cause clinical parasitic bronchitis, necessitating treatment with diethylcarbamazine citrate. This drug was chosen since it has no effect on gut worms and would not interfere with the epidemiology of those species.

However, the efficacy of this treatment was not absolute since the control animal killed in extremis still had 45 live adult lungworms. Following the subsequent treatment of all the control animals with levamisole, to control the symptoms of parasitic gastro-enteritis, clinical signs of parasitic bronchitis also subsided.

Failure to recover lungworm larvae consistently from the pasture

is almost certainly related to the technique employed for herbage larval recovery. In the observations reported here herbage samples were collected randomly from the pasture plots, but Jorgensen (1980) has shown that lungworm larval recoveries are more consistent when herbage samples are collected preferentially from around faecal pats. In this context it is interesting that the onset of parasitic bronchitis occurred during the dry period when the cattle may have resorted to grazing closer to the dung pats than is normal.

Administration of the MSRB device altered the pattern of infection with gut trichostrongyles but not that of lungworm. Thus the faecal egg counts of the MSRB treated calves never exceeded a mean value of 50 epg. As a result, pasture larval counts remained below 1000 L_3/kdh excepting on two occasions at the end of September and October when 2500 Ostertagia and 1100 Cooperia L_3/kdh were present. These results are in accordance with other results obtained with the MSRB bolus in other parts of Europe (see Introduction). They were also reflected in significantly lower numbers of Ostertagia and Cooperia species being found at post mortem compared to those in the controls, and in less stomach damage as evidenced by low mucosal fractions, and lower plasma pepsinogens and gastrin levels.

Parasitic bronchitis occured in the MSRB treated group two weeks after the known expiry date of the activity of the MSRB bolus and apparently without prior pasture contamination, since lungworm larvae were not detected in the faeces of this group during the active phase of the bolus. The origin of these larvae remains a mystery but it once again demonstrates the ubiquitous nature of this parasite. As has been suggested by Duncan et al, 1979 they may have originated from a reservoir of larval infection in the soil or by windborn transmission via Pilobolus sp. spores from the adjacent pasture.

Also, it should be clearly noted that the MSRB treatment did not completely eliminate trichostrongyle eggs. The viability of these eggs is not known, however their presence in low numbers may be beneficial in promoting pasture larval infections sufficient to provoke an immune response, but not to cause an impaired production.

Finally, it is interesting that despite fortnightly treatment with the wide spectrum anthelmintic fenbendazole (Fbz) the group of calves, designated as clean, acquired sufficient infective larvae to produce sporadic but low faecal egg counts which resulted in a small population

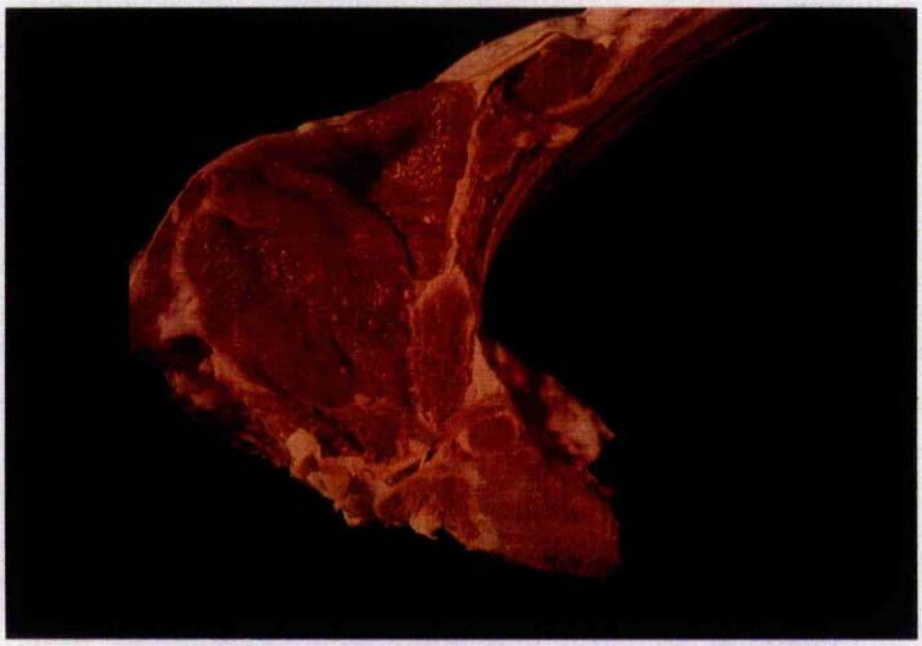


Plate 9a Indicator joint (10th rib) of control calf showing watery muscle.

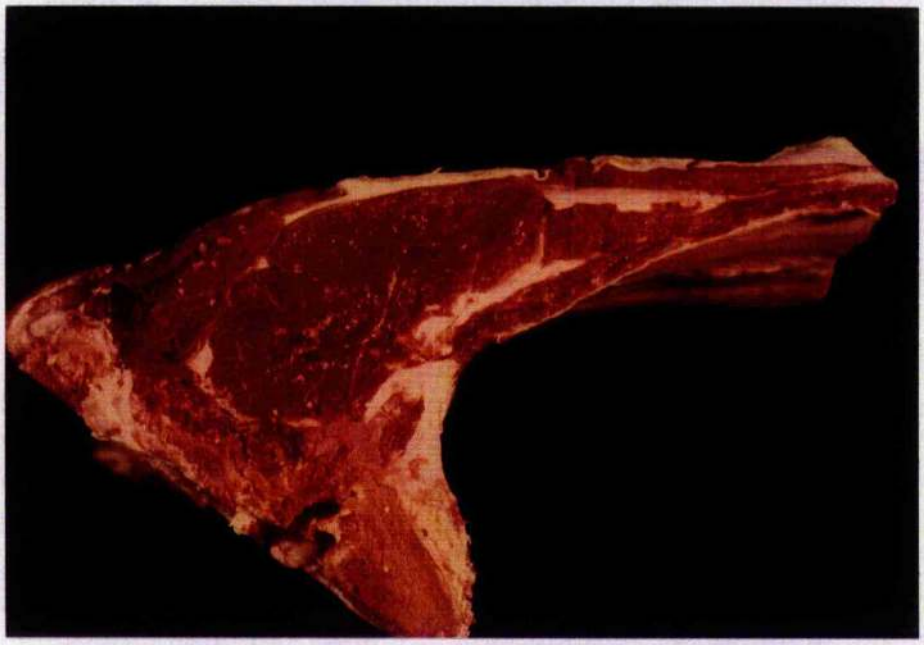


Plate 9b Indicator joint (10th rib) of MSRB calf

of infective larvae on the pasture.

The decrease in appetite was the first clinical sign to be observed and occurred about a week prior to the onset of the diarrhoea. It is unlikely that farmers would notice this sign as they would not be observing the cattle as closely on a daily basis as was done in this experiment. Nevertheless, the loss of appetite was significantly marked to enable a visual distinction to be readily made between the areas grazed by the naturally infected controls and those of the MSRB treated calves; the pasture grazed by the controls retained an ample grass cover compared to the sparse grazing remaining in the pasture grazed by the MSRB treated animals (Plate 6). The marked increase in plasma pepsinogen levels which coincided with the clinical sign of ostertagiasis confirmed the usefulness of this parameter in the diagnosis of ostertagiasis in young grazing cattle. It is generally accepted that individual plasma pepsinogen levels above 3.0 I.u. of tyrosine are indicative of abomasal damage sufficient to cause clinical disease (Armour and Ogbourne, 1982) and this was also true in the current studies.

Gastrin levels were estimated routinely in five cattle in each group, and since the mean pepsinogen levels in these five cattle were similar to those of the entire group and it is therefore reasonable to assume that they were representative of the group. The marked elevation of plasma gastrin which occurred in the control animals, largely coincident with the rise in plasma pepsinogen, suggests that the two events were responding to similar changes in the abomasal mucosa and in particular to the reduction in acid production known to occur in Ostertagia infections. Thus, the excess pepsinogen, not converted to pepsin in the absence of enough acid production, passes into the circulation while the hypergastrinaemia occurs in response to the reduction of acid conditions in the antrum which stimulates secretion by the gastrin-producing G cells. It would appear therefore that the combined elevation of pepsinogen and gastrin could be a good indicator of an 'active lesion of ostertagiasis' related to the presence of developing and adult parasites, a suggestion which is supported by the statistical correlation shown between those parameters in the current studies.

These studies indicated a significant link in the described coexistence of significantly elevated levels of both plasma pepsinogen and gastrin in the cattle clinically affected with the type I

ostertagiasis at grass during this first grazing season. At necropsy the separate correlations of gastrin and pepsinogen values with the L_5A were highly significant but not with L_4 larvae. Even with only limited data available from the 11 post-mortem findings (where there were five clinically type I ostertagiasis affected calves), a highly significant multiple regression relationship was seen where the numbers of L_5 larvae and adults could be predicted from a knowledge of the combination of both gastrin and pepsinogen values. It is interesting that early in the grazing season there was a small increase in gastrin levels in all groups which had decreased to previous levels by mid-summer; at that time there was no similar increase in plasma pepsinogen levels. The reason for this early season rise in plasma gastrin is not known but may reflect a physiological response to the change in diet, from concentrates to grass, rather than a response to a change in abomasal acidity. However, the role of ingested larvae in this process cannot be ignored as following experimental infections of Ostertagia in sheep, Anderson et al. (1981) have reported a moderate increase in plasma gastrin in advance of any major change in abomasal pH associated with the presence of parasites.

It is also interesting that although the onset of clinical signs occurred in the face of an increasing larval challenge from mid-July onwards, the level of pasture larval infections which apparently initially triggered the clinical disease was only 2000 L_3/kdh. From experimental studies (Ritchie et al., 1966; Murray et al., 1970) it is known that it takes a minimum of three weeks before the pathogenic effects of ostertagiasis are experienced and 2000 L_3/kdh was the level three weeks prior to the first clinical signs being noticed. This level is well below that associated with experimental reproduction of the disease (Ritchie et al., 1966) and possibly reflects the response of animals which had been previously sensitised by a low infection, in this case the overwintered larvae ingested in spring. It has been suggested that in older immune ruminants under challenge with Ostertagia larvae (Anderson , 1973; Armour et al., 1979; Yakoob et al., 1983) that the immune reaction response in the abomasal mucosa, is associated with a hypersensitivity reaction, which is reflected in an increased permeability of the mucosa to protein molecules; it is possible that in young grazing cattle, in which a partial immunity has developed following the ingestion of overwintered larvae, there is an accelerated and exaggerated pathological response

in the abomasal mucosa on subsequent exposure to larval challenge in the summer.

The small, but significant reduction, in the killing-out percentage found in the control animals infected with O. ostertagi had also been observed in growing sheep experimentally infected with O. circumcincta (Sykes and Coop, 1977). Reduced dressing percentages were also reported following artificial T. colubriformis infections in sheep (Sykes and Coop, 1976 and Steel et al, 1980).

Differences in killing-out percentages arise from differences in the weight of non-carcass components i.e. head, hide, hoof, blood and gut offals. The relative proportion of these non-carcass components (with respect to the liveweight of the animal) decreases with increasing liveweight. In the study here the animals selected for slaughter were the lightest and heaviest from each of the control and MSRB treated groups. However, the mean liveweights only differed in favour of the MSRB treated group by less than 20 kg and so the significantly better killing-out percentage was mainly due to differences in the weight of the gut and gut contents. Both groups of animals were fed and managed indoors in an exactly similar manner for one week before slaughter, which included a 12 hour fast (or 'shrink'). Although the rumen and gut digesta contents could unfortunately not be weighed at slaughter, visually there were no apparent differences in the digesta volumes and masses.

The hypertrophy of the abomasal mucosa in the control calves was reflected in the much greater weight of this tissue. This has been reported several times (e.g. Anderson et al., 1967; Michel, 1969). Coop et al (1977) also demonstrated a heavier overall abomasal weight in infected sheep infected with O. circumcincta (even compared with the pair-fed group). Symons and Jones (1978, 1981 and 1983) in a number of experiments have shown increases in protein synthesis rates in the gut and liver of parasitised animals together with decreases in skeletal muscle and kidney cortex deposition. Even with the relatively small numbers of slaughter animals available at this stage of the investigation, it could be speculated that the differences in killing-out percentage were mainly attributable to increased total gut mass and decreased skeletal muscle deposition. This view can be supported by the measurements taken from both the carcass and the 7-10th rib indicator joint. In the rib dissection, a significantly lowered mean weight of lean tissue was present in the control animals

compared with the MSRB treated cattle, which represented over 70% of the difference observed in the total weights of the joint. Subsequently, the proportion of lean meat was lowered in this joint in the control cattle. The bone mass in this joint in the infected animal was apparently much less affected since there was no significant difference in the mean weight of bone and where this reduced weight accounted for the remaining 30% of the difference in the total joint weight. Similar results in protein deposition have been found by Sykes and Coop (1977) and Coop, Sykes and Angus (1982) in lambs chronically infected with O. circumcincta.

The linear measurements taken from the carcasses were essentially of the skeletal dimensions, such as length of carcass side, length of leg and width at the 7th rib and were not significantly different with the mean values being similar. In a study on chronic ostertagiasis in artificially infected grazing lambs, Sykes et al (1977) reported severe effects of the infection upon the development of the skeleton, manifested in a reduced bone size and degree of mineralisation. In the present study control animals were acutely affected by type I ostertagiasis and were treated with levamisole to arrest the severity of the disease. It might be argued that such an acute short-term infection, followed by treatment, would not result in any great alteration in skeletal development but could clearly derange muscle protein deposition and affect the gut weight: total bodyweight ratio and so alter the dressing percentage.

Further interesting observations on the analyses of the indicator rib joint centred on the the eye muscle. The apparent difference in skeletal muscle deposition was again seen in significantly reduced eye muscle weight and area in the control animals. The rib joint muscles from the most severely affected control animals had a poorly set texture with a distictively pale and watery appearance compared with the well-set, bright red coloration of all the MSRB treated samples. Analyses of tissue samples taken from the centre of the eye muscle at the 7th rib showed a significant difference in mean water content. The method used was an automated Karl Fischer titration which may have underestimated the water content by a small degree. (In later studies all tissue dry matter estimations were performed using a high efficiency freeze-drying method). Nevertheless, on a comparative basis, the small increase in water content of this muscle apparently contributed to the marked difference in the physical appearance and

texture. Protein analyses of this tissue sample showed a significantly higher content in the control cattle which is speculated to be as a result of lowered intramuscular fat deposition in the joint.

The apparent increase in muscle water content may not have been linked to an increased water retention in the whole body since, although Halliday et al (1965) showed cattle with type II ostertagiasis to have a significant increase in whole body water content (inevitably coupled to a hypoalbuminaemia), cattle with an experimental type I ostertagiasis did not have significantly greater total body water than uninfected control animals.

Some of the increased water content of the control cattle carcasses found here may be associated with the slight degree of hypoalbuminaemia observed from week 16 of grazing, coinciding with the clinical evidence for acute parasitic gastroenteritis. It is known that hypoalbuminaemia often accompanies the acute phase of the infection (Mahrt et al, 1964; Neilsen, 1966; Ross and Todd, 1965 and Dotta et al 1977) and is normally associated with a high rate of albumin removal from the plasma (Mulligan et al., 1963; Yakoob et al., 1983).

From the production point of view all three groups of calves gained a mean of about 75 kg liveweight in the first three months of grazing. During this period the weather and the grass growth pattern was normal with the grass in plentiful supply. Production of about 500 kg of liveweight gain (LWG) per hectare was calculated for both the control and the MSRB treated groups. The production in terms of LWG/ha, for the paddock containing the set-stocked clean group was somewhat less at about 400 kg/ha. This was mainly due to the unavoidable difference in stocking density. However in the second part of the grazing season (weeks 12 to 22) a prolonged five-week dry spell here coincided with the natural mid-season depression in grass growth. The clean and MSRB treated calves clearly were short of grass in this period and their depressed growth rate is a reflection of that shortage. Both of those groups only gained a mean of around 30 kg during this period being less than half the gain observed in the early part of the season. However the control calves in this period became ill with type I ostertagiasis and displayed a loss of appetite followed by diarrhoea. The grass in these plots was not in short supply, in contrast to the MSRB treated group's grazing, owing to this appetite loss. Following treatment with levamisole, grazing appetite appeared

to return to normal but the calves did not show any weight gain compensation. The mean liveweight gain in this period was only 11 kg i.e. only one third of the noted poor performance of the other two groups.

In the grazing season of 22 weeks overall, the poor liveweight gain performance by the control calves is reflected in a total gain which was some 20 kg less than the other groups and resulted in production output of only 530 kg LWG/ha compared with 700 kg LWG/ha in the MSRB treated plots. It is noted however that the grass shortage at the end of the season in both the MSRB treated and the clean groups clearly limited their potential for growth expression and the liveweight differences between these groups and the infected control group could have been even greater than was observed.

Even so, it has been demonstrated, that after one season at grass, liveweight gain alone does not comprehensively express the differences to be found by carcass appraisal and which may have severe economic consequences. Further study, after a second season at grass, on these aspects is described in the following sections of this thesis.

Fig. A1 Temperature and rainfall patterns for the experimental years 1982 - 3 compared with those for 1975 - 79.

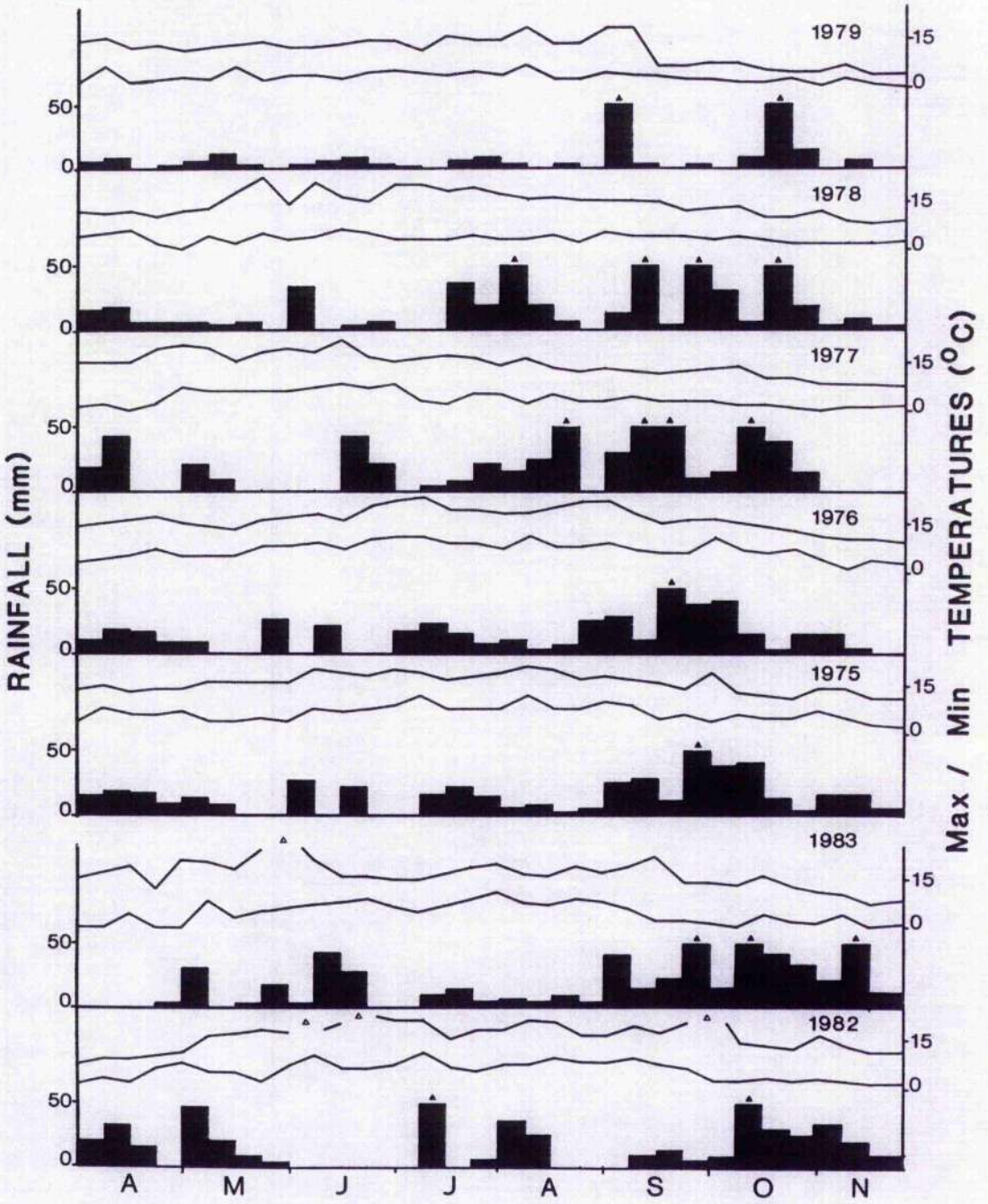

1979
1978
1977
1976
1975
1983
1982
RAINFALL (mm)
Max / Min TEMPERATURES (°C)
50
0
15
0
A
M
J
J
A
S
O
N

Fig. A2. Mean liveweight (kg) of clean, MSRB treated and control calves in the first grazing season.

Mean liveweight (kg) of clean, MSRB treated and control calves in the first grazing season

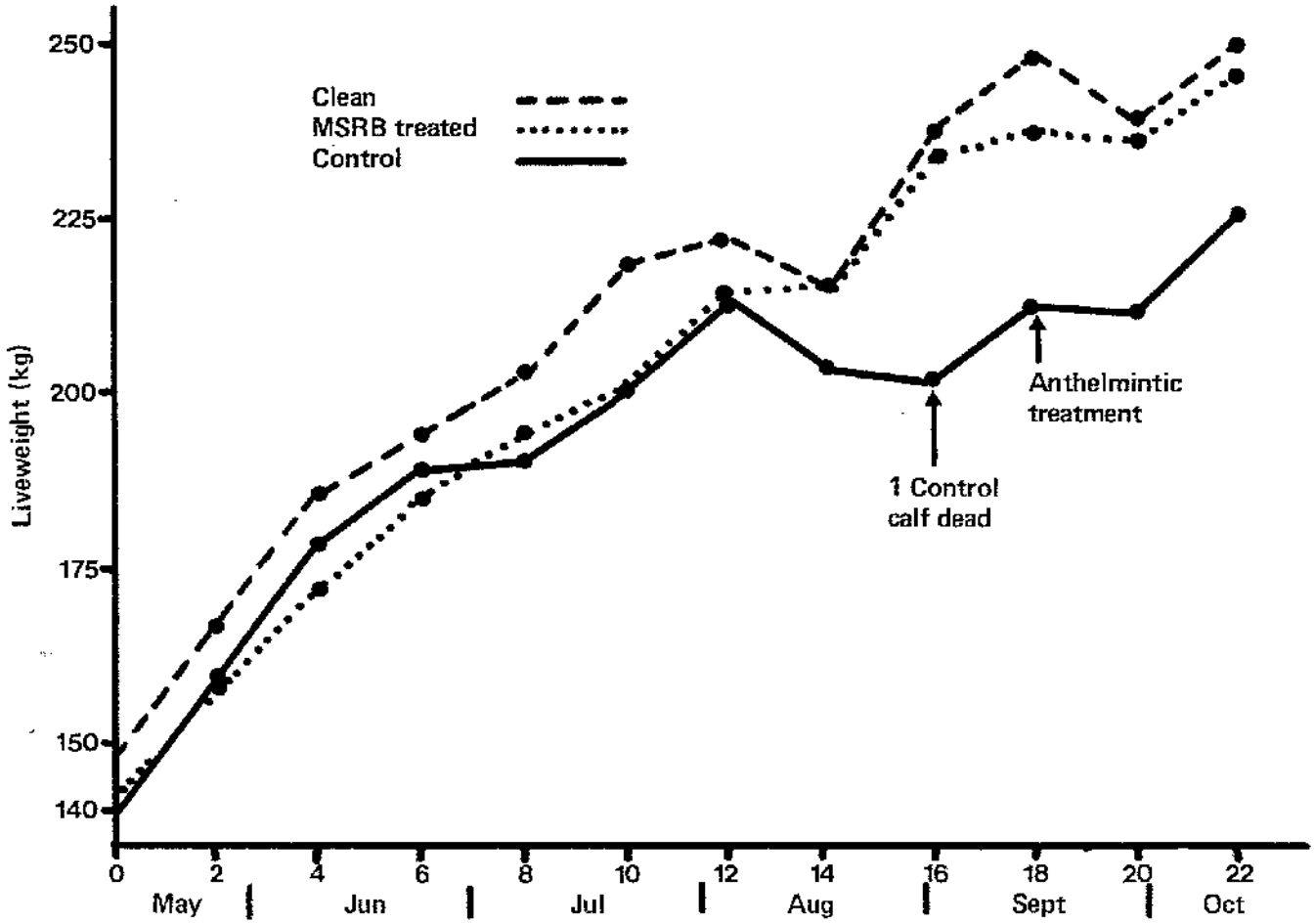


Fig. 3. Mean plasma pepsinogen levels (Iu tyrosine) of clean, MSRB treated and control calves in the first grazing season.

Mean plasma pepsinogen levels (Iu tyrosine) of clean, MSRB treated and control calves in the first grazing season

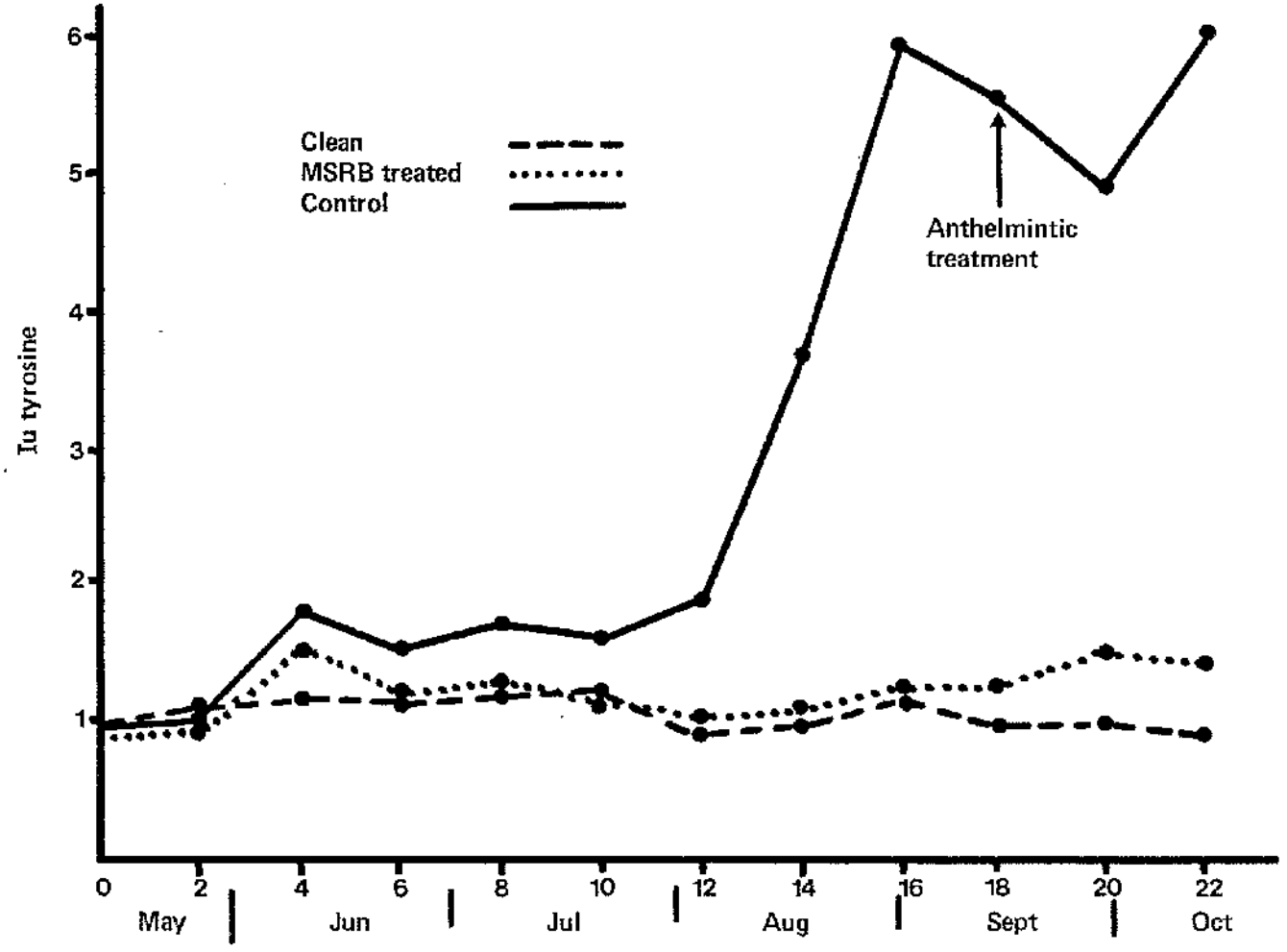


Fig. A4. Mean plasma gastrin levels of clean, MSRB treated and control calves during first grazing season.

MEAN PLASMA GASTRIN LEVELS OF CLEAN, MSRB TREATED AND CONTROL CALVES DURING FIRST GRAZING SEASON

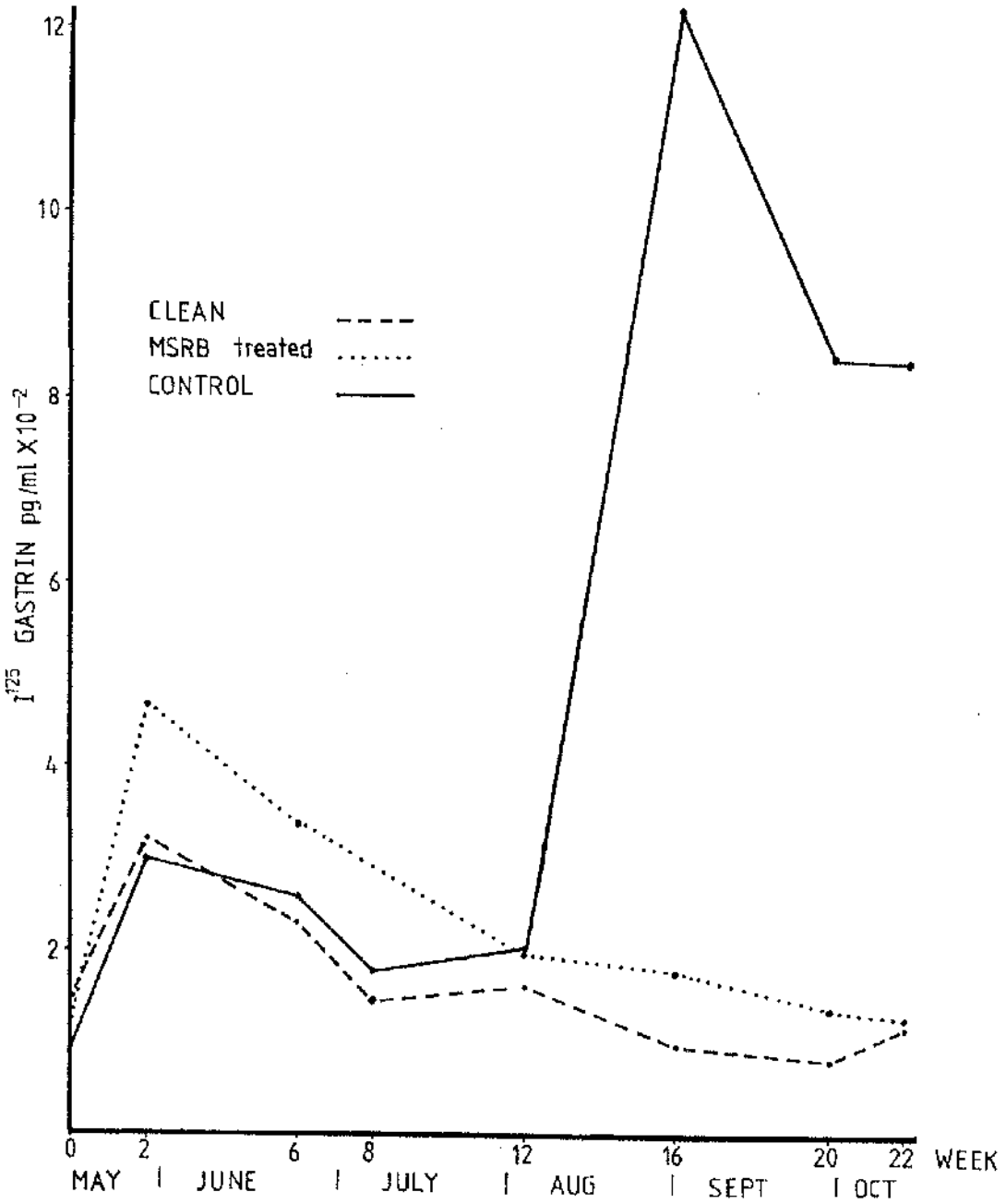


Fig. A5. Mean trichostrongyle egg counts in faeces of clean, MSRB treated and control calves in the first grazing season.

Mean trichostrongyle egg counts in faeces of clean, MSRB treated and control calves in the first grazing season

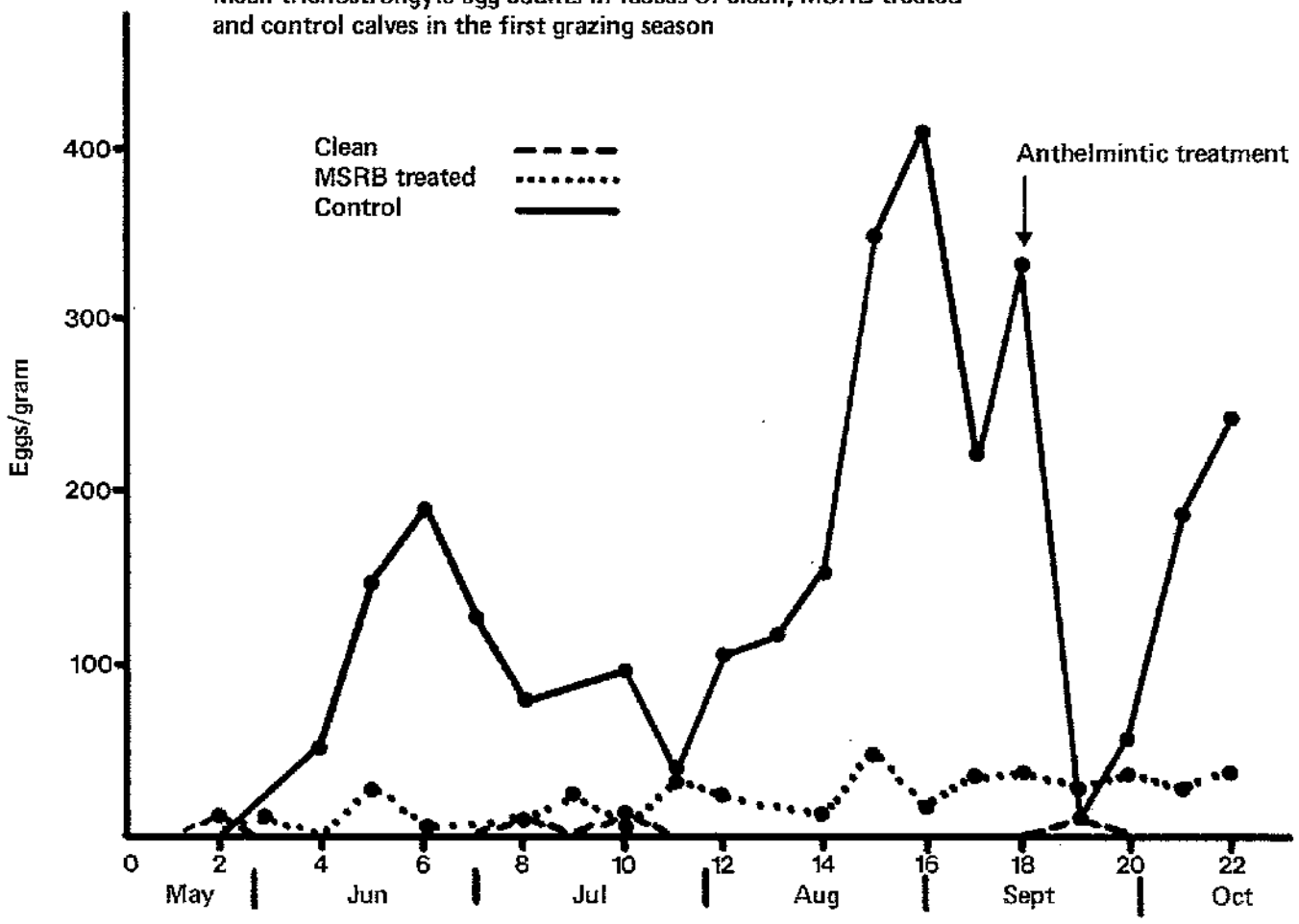


Fig. A6. Recoveries of O.ostertagi L_3 from paddocks grazed by clean, MSRB treated and control calves in the first grazing season.

Recoveries of O.ostertagi L_3 from paddocks grazed by clean, MSRB treated and control calves in the first grazing season

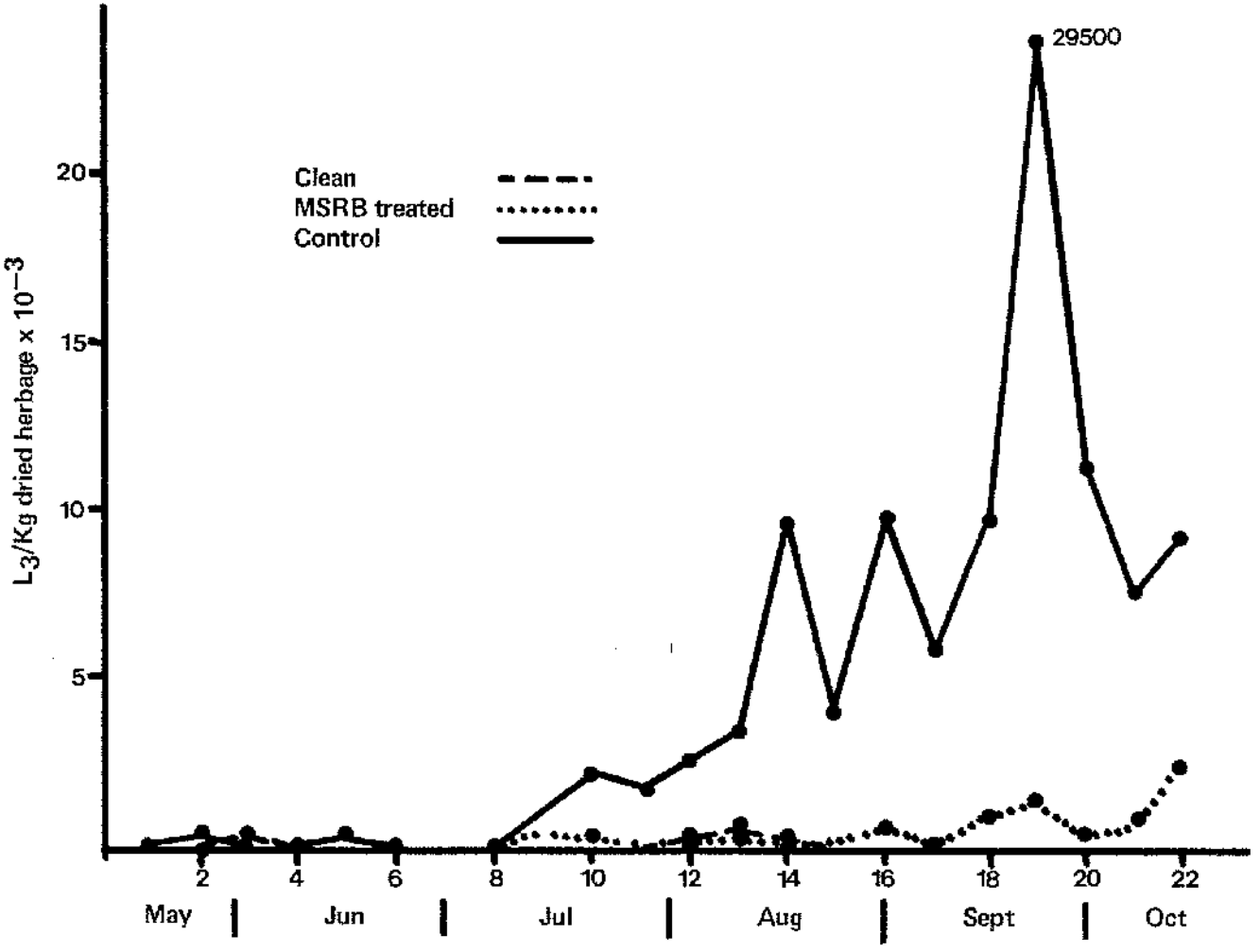
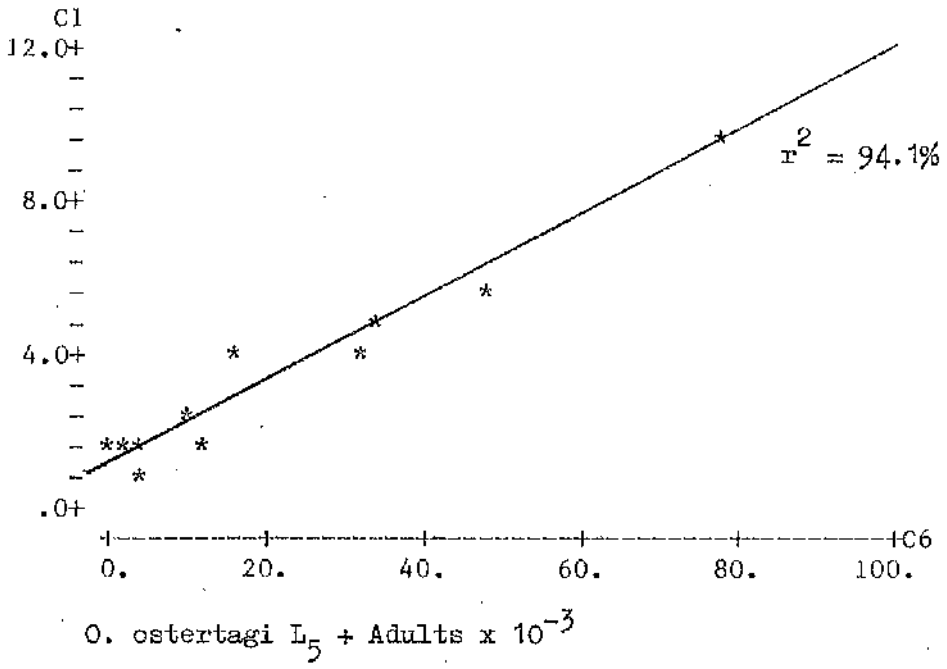


Fig. A7



THE REGRESSION EQUATION IS No. 1
Y = 1.06 + .107 X1

	COLUMN	COEFFICIENT	ST. DEV. OF COEF.	T-RATIO = COEF/S.D.
	--	1.0634	0.2681	3.97
X1	C6	0.106656	0.008414	12.68

THE ST. DEV. OF Y ABOUT REGRESSION LINE IS
S = 0.6479
WITH (11- 2) = 9 DEGREES OF FREEDOM

R-SQUARED = 94.7 PERCENT
R-SQUARED = 94.1 PERCENT, ADJUSTED FOR D.F.

ANALYSIS OF VARIANCE

DUE TO	DF	SS	MS=SS/DF
REGRESSION	1	67.4489	67.4489
RESIDUAL	9	3.7777	0.4197
TOTAL	10	71.2266	

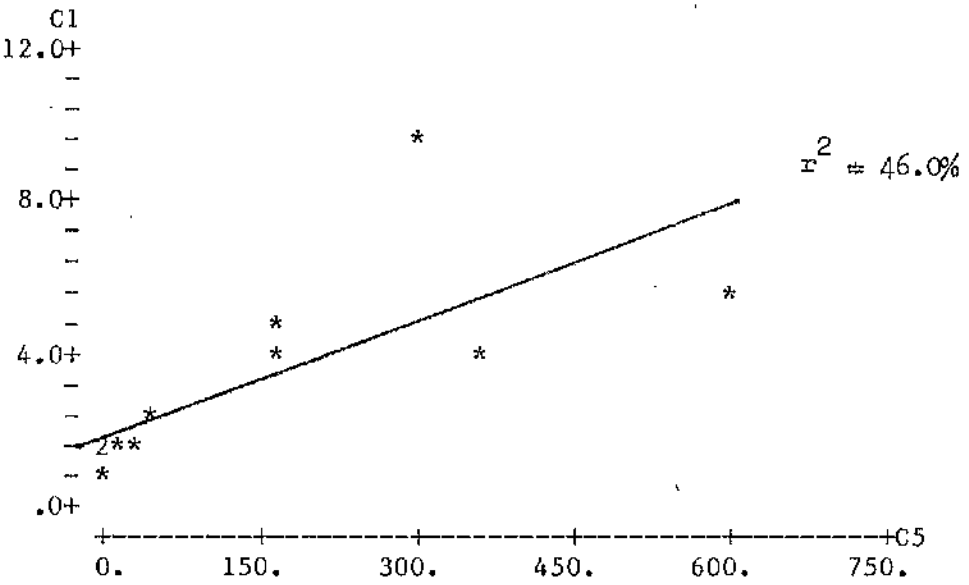
ROW	X1 C6	Y C1	PRED. Y VALUE	ST.DEV. PRED. Y	RESIDUAL	ST.RES.
8	16.2	4.069	2.791	0.201	1.278	2.07RH
11	78.7	9.639	9.457	0.517	0.182	0.47HX

R DENOTES AN OBS. WITH A LARGE ST. RES.
X DENOTES AN OBS. WHOSE X VALUE GIVES IT LARGE INFLUENCE.

DURBIN-WATSON STATISTIC = 2.75

ig. A8

lasma
epsinogen
Iu tyrosine)



Total O. ostertagi worms x 10^{-3}

THE REGRESSION EQUATION IS No. 2
Y = 1.86 + .0099 X1

	COLUMN	COEFFICIENT	ST. DEV. OF COEF.	T-RATIO = COEF/S.D.
	--	1.8593	0.7728	2.41
X1	C5	0.009888	0.003208	3.08

THE ST. DEV. OF Y ABOUT REGRESSION LINE IS
S = 1.962
WITH (11- 2) = 9 DEGREES OF FREEDOM

R-SQUARED = 51.4 PERCENT
R-SQUARED = 46.0 PERCENT, ADJUSTED FOR D.F.

ANALYSIS OF VARIANCE

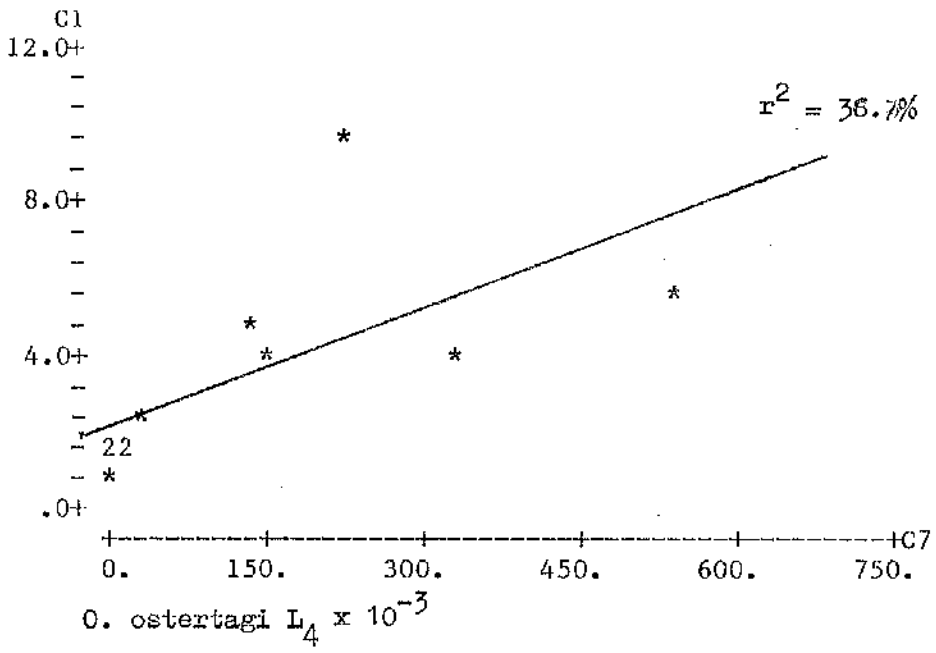
DUE TO	DF	SS	MS=SS/DF
REGRESSION	1	36.578	36.578
RESIDUAL	9	34.648	3.850
TOTAL	10	71.227	

ROW	X1 C5	Y C1	PRED. Y VALUE	ST.DEV. PRED. Y	RESIDUAL	ST.RES.
7	596	5.763	7.748	1.532	-1.985	-1.62HX
11	300	9.639	4.831	0.754	4.808	2.65RH

R DENOTES AN OBS. WITH A LARGE ST. RES.
X DENOTES AN OBS. WHOSE X VALUE GIVES IT LARGE INFLUENCE.

DURBIN-WATSON STATISTIC = .96

Fig. A9



Plasma Pepsinogen (Iu tyrosine)

THE REGRESSION EQUATION IS No.3
Y = 2.06 + .0100 X1

	COLUMN	COEFFICIENT	ST. DEV. OF COEF.	T-RATIO = COEF/S.D.
	--	2.0563	0.8197	2.51
X1	C7	0.010044	0.003850	2.61

THE ST. DEV. OF Y ABOUT REGRESSION LINE IS
S = 2.123
WITH (11- 2) = 9 DEGREES OF FREEDOM

R-SQUARED = 43.1 PERCENT
R-SQUARED = 36.7 PERCENT, ADJUSTED FOR D.F.

ANALYSIS OF VARIANCE

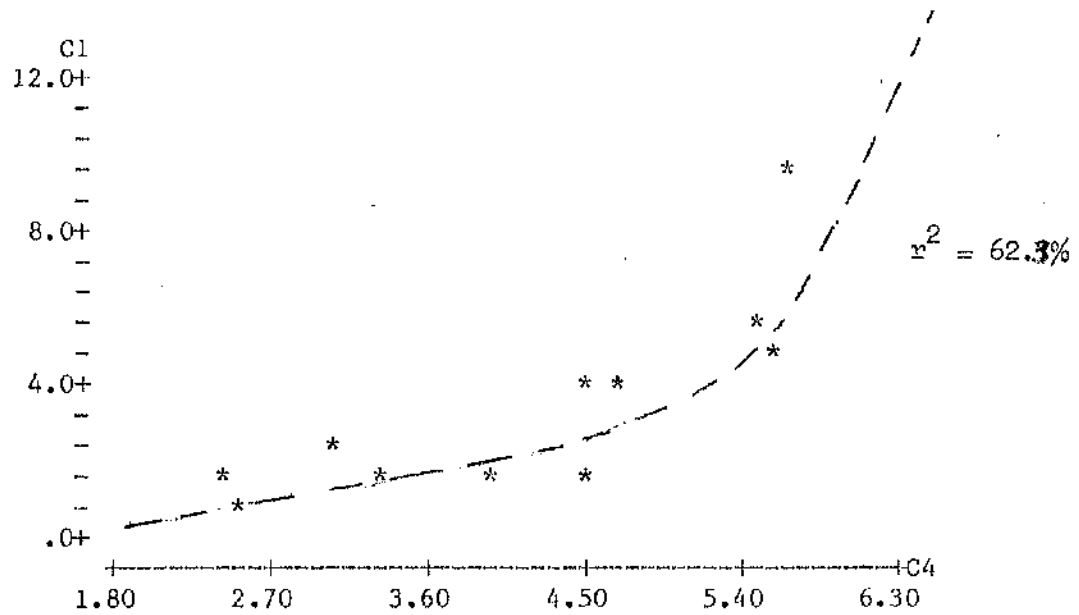
DUE TO	DF	SS	MS=SS/DF
REGRESSION	1	30.667	30.667
RESIDUAL	9	40.560	4.507
TOTAL	10	71.227	

ROW	X1 C7	Y C1	PRED. Y VALUE	ST.DEV. PRED. Y	RESIDUAL	ST.RES.
7	544	5.763	7.516	1.705	-1.753	-1.39HX
11	222	9.639	4.284	0.726	5.355	2.68RH

R DENOTES AN OBS. WITH A LARGE ST. RES.
X DENOTES AN OBS. WHOSE X VALUE GIVES IT LARGE INFLUENCE.

DURBIN-WATSON STATISTIC = .83

Fig. A10



THE REGRESSION EQUATION IS No.4
Y = - 4.11 + 1.80 X1

	COLUMN	COEFFICIENT	ST. DEV. OF COEF.	T-RATIO = COEF/S.D.
	--	-4.109	1.857	-2.21
X1	C4	1.8016	0.4300	4.19

THE ST. DEV. OF Y ABOUT REGRESSION LINE IS
S = 1.638
WITH (11- 2) = 9 DEGREES OF FREEDOM

R-SQUARED = 66.1 PERCENT
R-SQUARED = 62.3 PERCENT, ADJUSTED FOR D.F.

ANALYSIS OF VARIANCE

DUE TO	DF	SS	MS=SS/DF
REGRESSION	1	47.084	47.084
RESIDUAL	9	24.143	2.683
TOTAL	10	71.227	

ROW	X1 C4	Y C1	PRED. Y VALUE	ST.DEV. PRED. Y	RESIDUAL	ST.RES.
11	5.70	9.639	6.160	0.825	3.479	2.46RH

R DENOTES AN OBS. WITH A LARGE ST. RES.

DURBIN-WATSON STATISTIC = 1.79

Table A1 Mean liveweight and average daily liveweight gain (ADLWG) (kg) of Clean, Control and MSRB treated calves during the first grazing period (0 - 22 weeks)

Week	0	2	4	6	8	10	12	14	16	18	20	22	Total Gain	ADL WG
CLEAN	147	168	185	194	202	218	222	214	237	248	238	250	103	0.67
±SE	8.8	9.63	10.6	9	7.41	7.13	8.44	8.43	7.55	7.81	8.32	9.06		
CONTROL	139	158	178	189	190	200	213	203	201	212	211	224	85	0.55
±SE	5.9	5.9	6.24	6.52	7.74	7.95	7.2	7.56	6	6.22	6.9	6.47		
MSRB	140	157	172	185	194	202	214	215	233	237	236	245	105	0.68
±SE	5.15	5.22	6.05	7.19	7.18	6.74	6.9	7.28	7.9	7.24	7.56	7.81		

MAY	JUNE	JULY	AUGUST	SEPTEMBER	OCTOBER

Table A2 Mean Plasma Pepsinogen levels (I.u. tyrosine) of Clean, Control and MSRB treated calves during the first grazing season

Week	0	2	4	6	8	10	12	14	16	18	20	22
Clean	1.0	1.1	1.2	1.1	1.1	1.2	0.9	1.0	1.1	1.0	1.0	0.9
±SE	.05	.08	.08	.07	.08	.08	.05	.05	.08	.08	.08	.03
Control	0.9	1.0	1.8	1.5	1.7	1.6	1.9	3.7	5.9	5.5	4.9	6.0
±SE	.10	.06	.1	.07	.08	.07	.1	.5	.5	.3	.4	.4
MSRB	0.9	0.9	1.5	1.2	1.3	1.1	1.0	1.1	1.2	1.2	1.5	1.4
±SE	.05	.09	.13	.09	.09	.06	.04	.04	.06	.07	.12	.1
	May		June		July		August		September		October	

Table A3. Mean plasma gastrin (pg/ml) concentrations (±SE) determined in Clean, MSRB treated and Control cattle during the first grazing season

Week	Date	Clean	MSRB	Control
0	13. 5.82	148 ±15.5^{a}	125 ± 6.8^{a}	93 ± 8.4^{b}
2	26. 5.82	320 ±14.7^{b}	465 ±26.6^{A}	296 ± 12.1^{b}
6	24. 6.82	230 ±15.1^{b}	336 ±19.7^{a}	259 ± 11.4^{b}
8	7. 7.82	147 ±17.4	145 ± 9.5	178 ± 17.9
12	3. 8.82	161 ±17.1	197 ±24.1	202 ± 26.7
16	1. 9.82	99 ± 8.2^{b}	178 ±18.5^{b}	1212 ±286.3^{a}
20	29. 9.82	82 ± 7.7	137 ±13.4	836 ±351.7*
22	13.10.82	117 ± 5.9^{b}	128 ±18.0^{b}	832 ± 95.4^{A}

a,b $P < 0.05$ A,b $P < 0.001$

* One value underestimated

Table A4 Mean weekly trichostrongyle faecal egg counts of Clean, Control and MSRB treated calves during the first grazing period

Key: LW – Lungworm

P – Present

T – Treated with Levamisole

N – Negative

Week	0	1	2	3	4	5	6	7	8	9	10	11
Clean	0	0	0	0	0	0	0	0	6	0	6	0
Control	0	0	0	25	53	150	189	122	75	81	94	34
MSRB	0	0	0	6	0	28	3	3	9	22	3	31

Week	12	13	14	15	16	17	18	19	20	21	22	23
Clean	0	0	0	$0\frac{LW}{N}$	$0\frac{LW}{N}$	0	0	6	0	0	0	0
Control	103	133	153	350 P	413 P	217	340 T	3	57	177	237	327
MSRB	22	16	13	50 N	16 P	34	34	28	34	27	38	53

Table A5 Larval counts (L/kg DM) on herbage grazed by Clean, Control and MSRB treated calves during the first season at grass

O. o Ostertagia ostertagi

C. o Cooperia oncophora

N. h Nematodirus helvetianus

D. v Dictyocaulus viviparus

	WEEK	-1	0	1	2	3	4	5	6	7	8	9	10	11	12	13
Group																
Clean	O.o.	31	0	100	0	145	100	0	0	0	0	0	0	0	0	526
	C.o.	0	0	0	0	145	0	0	0	0	0	0	0	0	0	0
	N.h.	0	0	0	0	0	0	9	118	0	0	0	0	0	0	0
	D.v.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Control	O.o.	0	0	130	227	0	0	137	0	0	0	1154	2133	1739	2485	3333
	C.o.	0	0	0	0	0	0	137	0	0	0	231	800	145	667	392
	N.h.	0	12	0	227	408	0	137	0	0	0	0	0	0	0	0
	D.v.	0	0	0	0	0	0	137	0	0	0	0	0	0	0	0
MSRB	O.o.	0	100	0	137	0	0	63	139	0	0	306	225	154	0	323
	C.o.	0	0	0	0	0	0	0	0	0	0	0	112	0	0	0
	N.h.	0	0	0	0	0	290	0	0	0	0	0	0	0	0	0
	D.v.	0	0	0	0	0	0	63	0	0	0	306	0	0	0	175
			MAY				JUNE					JULY			AUGUST	

WEEK		14	15	16	17	18	19	20	21	22
Group										
Clean	O.o.	0	0	0	0	0	0	0	0	0
	C.o.	0	0	0	0	0	0	0	0	0
	N.h.	0	0	0	0	0	0	0	0	0
	D.v.	0	0	0	0	0	0	0	0	0
Control	O.o.	9815	3889	9884	5833	9455	29500	11724	7429	9014
	C.o.	2593	1204	1953	1146	2363	3500	1724	1714	3750
	N.h.	185	0	0	0	182	0	0	0	141
	D.v.	0	185	0	0	0	0	0	0	0
MSRB	O.o.	0	202	583	84	941	1290	377	645	2535
	C.o.	0	0	194	0	235	0	0	0	1127
	N.h.	0	0	0	0	0	0	0	0	0
	D.v.	0	0	0	0	0	0	0	0	0
		AUGUST				SEPTEMBER			OCTOBER	

Table A6 Blood biochemical analyses on six occasions from Clean, MSRB treated and Control calves (SE) during the first grazing season

On any one occasion values with different lettered superscripts are significantly different viz.,

a, b $P < 0.05$

A, b $P < 0.01$

A, B $P < 0.001$

Week	4	6	14	16	20	22
Urea mmol/l						
Clean	$5.24^{a} \pm 0.28$	$5.61^{a} \pm 0.19$	$9.13^{A} \pm 0.35$	$7.10^{A} \pm 0.35$	$6.17^{B} \pm 0.25$	$2.33^{B} \pm 0.38$
MSRB	$4.25^{b} \pm 0.23$	$5.49^{a} \pm 0.13$	$6.49^{B} \pm 0.15$	$5.31^{b} \pm 0.11$	$6.37^{B} \pm 0.15$	$5.66^{A} \pm 0.19$
Control	$3.99^{b} \pm 0.22$	$4.72^{b} \pm 0.13$	$6.27^{B} \pm 0.62$	$6.45^{A} \pm 0.24$	$9.19^{A} \pm 0.25$	$5.89^{A} \pm 0.22$
Inorganic phosphate mmol/l						
Clean	2.15 ± 0.12	2.26 ± 0.07	$2.26^{b} \pm 0.09$	2.36 ± 0.13	2.20 ± 0.08	2.54 ± 0.16
MSRB	$2.07^{b} \pm 0.07$	2.36 ± 0.07	$2.63^{a} \pm 0.06$	$2.63^{A} \pm 0.07$	2.39 ± 0.07	2.46 ± 0.05
Control	$2.34^{a} \pm 0.06$	2.40 ± 0.06	$2.61^{a} \pm 0.07$	$2.28^{b} \pm 0.09$	2.68 ± 0.98	2.26 ± 0.07
Total Protein g/l						
Clean	68.7 ± 1.69	69.01 ± 1.69	72.3 ± 1.27	73.01 ± 2.33	69.6 ± 1.79	$68.3^{b} \pm 2.08$
MSRB	68.0 ± 1.15	$70.81^{a} \pm 1.22$	70.2 ± 0.79	68.01 ± 0.95	70.1 ± 0.88	74.91 ± 1.22
Control	67.9 ± 0.83	$67.11^{b} \pm 0.99$	71.4 ± 1.86	72.7 ± 2.05	71.9 ± 1.11	71.5 ± 1.49
Albumin g/l						
Clean	31.4 ± 0.53	$31.7^{b} \pm 0.63$	$36.1^{A} \pm 0.74$	$36.0^{A} \pm 0.73$	$35.3^{A} \pm 0.83$	$31.3^{b} \pm 1.17$
MSRB	$30.3^{a} \pm 0.31$	$33.8^{a} \pm 0.35$	$34.7^{b} \pm 0.44$	$32.4^{c} \pm 0.53$	$36.3^{A} \pm 0.53$	$36.2^{A} \pm 0.54$
Control	$31.3^{b} \pm 0.37$	$31.9^{b} \pm 0.06$	$32.2^{B} \pm 0.52$	$29.0^{B} \pm 0.95$	$30.4^{B} \pm 0.78$	$29.9^{B} \pm 0.99$

Table A7 Individual and mean worm burdens of Control and MSRB treated calves at post-mortem examination after first grazing season

Control vs MSRB * $P \leq 0.05$
** $P \leq 0.01$
*** $P \leq 0.001$

ABOMASUM						SMALL INTESTINE								LUNG
CONTROL	O. ostertagi					Cooperia Spp				N.helvetianus				D.viviparus
No.	L_4	L_5	Adult	Total	pH	L_4	L_5	Adult	Total	L_4	L_5	Adult	Total	
64	547,550	6,050	41,950	595,550	5.5	6,200	0	2,000	8,200	0	0	0	0	0
80	141,400	600	15,600	167,600	4.5	13,650	0	10,200	23,850	150	0	0	150	0
77	331,600	1,000	31,600	364,200	4.7	100	0	0	100	0	0	0	0	5
68	139,000	1,200	31,800	172,000	5.6	150	0	150	300	0	0	0	0	0
89	221,800	4,700	74,000	300,500	5.7	16,800	0	27,450	44,250	150	450	750	1,350	5
Mean	278,270***	2,710**	38,990***	319,970***	5.2*	7,380***	0	7,961***	15,340***	61.8	92	152	302	2/5
SE	67,541	995.5	8,690	70,256	0.2	3,064	0	4,667	7,530	32.2	80	133	235	
CONTROL	(Killed in-extremis 3/9/82)													
85	3,500	23,200	122,000	148,700	0	0	200	9,000	9,200	0	0	0	0	45
MSRB GROUP														
60	19,900	500	11,400	31,800	4.5	0	0	700	700	0	0	0	0	0
62	5,700	0	950	6,650	3.3	500	0	400	900	0	0	0	0	0
93	2,200	0	2,000	4,200	4.0	0	0	0	0	400	0	0	400	0
72	32,000	200	9,000	41,200	3.7	300	0	300	600	0	0	0	0	0
87	2,600	200	3,200	6,000	2.5	100	0	100	200	0	0	100	100	20
84	11,000	0	4,200	15,200	2.4	100	0	300	400	0	0	0	0	0
Mean	12,233	150	5,125	17,508	3.4	168	0	301	467	68	0	19	85	1/6+
SE	4,369	73	1,547	5,770	0.3	72.7	0	90.8	123.8	60.6	0	14.7	59.4	

Table A8 Carcass Measurements on MSRB treated and Control calves killed one week after housing

Calf No.	LW+kg	Dead kg	KO%(i)	1	2	3	4
MSRB							
84a	210	97.6	46.5	101.4	64.4	50.3	36.5
87a	190	96.9	51.0	100.3	60.8	42.6	33.4
93a	210	102.7	48.9	103.1	65.1	47.5	36.5
60b	290	150.0	51.7	113.9	68.2	56.6	41.9
62b	260	128.2	49.3	113.2	62.5	50.2	38.5
72b	310	150.9	48.7	114.8	66.4	55.9	39.3
Mean	245	121	49.4**	107.8	64.6	50.7	37.7
±SE	18.2	9.5	0.68	2.5	2.4	1.9	1.1
Control							
64b	250	117.3	46.9	112.2	67.6	47.2	40.0
68b	270	128.6	47.6	108.3	68.8	51.1	38.4
77b	250	119.5	47.8	107.5	67.5	51.3	37.8
80a	180	83.6	46.4	99.4	63.2	47.2	35.5
89a	180	82.8	46.0	96.4	58.5	45.0	29.2
Mean	226	106.4	46.9	105.1	65.1	48.4	36.2
±SE	17.1	8.6	0.34	2.2	2.1	1.1	1.7

a = light calves
b = heavy calves

+MSRBa> Control a***

1. Side Length cm
2. Leg Length cm
3. 7th Rib Width cm
4. Leg Width (P/S)cm

(i) MSRBa> Control a*
MSRBb> Control b***

* P≤ 0.05
*** P< 0.001

Table A9. Dissection of 7-10th Rib Joint of MSRB treated and Control calves killed after the first grazing season

	Total	Lean		Bone		Fat	
		g	(%)	g	(%)	g	(%)
MSRB							
84a	2381	1512	(63.5)	807	(33.9)	62	(2.6)
87a	2116	1438	(68.0)	677	(32.0)	0	(0.0)
93a	1995	1367	(68.5)	569	(28.5)	60	(3.0)
60b	3324	1829	(55.0)	1166	(35.1)	326	(9.8)
62b	2434	1627	(66.7)	672	(27.6)	139	(5.7)
72b	3202	1960	(61.2)	1226	(38.3)	16	(0.5)
Mean	2575	1621**	(63.8)	853	(32.6)	101	(3.8)
SE±	208	86.2	(1.9)	103	(1.5)	45	(1.3)
Control							
80a	1517	951	(62.7)	464	(30.6)	105	(6.9)
89a	1475	968	(65.6)	463	(31.4)	44	(3.0)
64b	2321	1316	(56.7)	924	(39.8)	81	(3.5)
68b	2648	1493	(56.4)	1014	(38.3)	140	(5.3)
77b	1990	1224	(61.5)	607	(30.5)	159	(8.0)
Mean	1990	1190	(60.9)	694	(34.1)	106	(5.3)
SE±	203	93	(1.6)	104	(1.8)	18	(0.9)

a = light calves

b = heavy calves

MSRB a** Control a

MSRB a* Control a

Table A10, Indicator Rib Joint (7-10th) eye muscle (EM) analyses in MSRB treated and Control calves killed after the first grazing season

Group	No.	Area cm^2	Wt g	gDM/kg	gCP/kgDM	Total g DM	Total g CP
MSRB							
	84a	51.9	635	297	730.6	188.6	137.8
	87a	54.6	532	290	837.9	154.3	129.3
	93a	52.3	601	296	837.8	177.9	149.0
	60b	56.3	720	280	775.0	201.6	156.2
	62b	56.2	648	318	729.5	206.1	150.3
	72b	58.7	829	323	715.2	271.0	193.8
	Mean	55.0*	660.8*	301*	771.0	199.9*	152.7*
	±SE	0.97	38.6	6.2	22.7	14.7	8.3
Control							
	64b	53.9	576	292	777.4	168.2	130.7
	68b	55.0	612	278	777.0	170.1	132.2
	77b	42.0	517	284	823.9	146.8	47.5
	80a	34.0	382	245	918.4	93.6	86.0
	89a	45.2	441	270	855.5	119.7	120.4
	Mean	46.1	505.6	274	830.4*	139.7	103.4
	±SE	3.4	37.9	7.2	26.5	13.1	14.5

* $P < 0.05$ MSRB a > Control a *** a small calves

**$P < 0.01$ MSRB b > Control b * b larger calves

Table A11 Correlation studies conducted with data obtained at slaughter from MSRB treated and Control calves after first grazing season

Relationships between plasma pepsinogen, gastrin, abomasal pH, weight and fraction (g mucosa/kg LW), O. ostertagi L_5 + adult and total worm burdens observed at slaughter

Key: PP Plasma pepsinogen I.u. tyrosine
Mu Weight of abomasal mucosa g
MuF Mu/LW g/kg
Gastrin pg/100 ml

** $P < 0.01$
*** $P < 0.001$

Group	No.	PP	Gastrin	Mu	MuF	pH	x 10^{-3} Total O.o	L_3 + Adult	L_4
MSRB	84	1.3	310	499	2.3	2.4	15.20	4.20	11.00
	60	1.4	140	510	1.7	4.5	31.80	11.90	19.90
	62	1.4	160	507	1.9	3.3	6.65	0.95	5.70
	93	1.6	90	408	1.9	4.0	4.20	2.00	2.20
	72	2.0	100	599	1.9	3.1	41.20	9.20	32.00
	87	1.1	150	270	1.4	2.5	6.00	3.40	2.60
Mean		1.47	158.3	465.5	1.9	3.4	17.50	5.27	12.233
SE ±		0.123	29.6	42.2	0.11	0.3	5.77	1.61	4.37
Control	64	5.8	640	998	4.0	5.5	595.55	48.00	547.55
	80	4.1	160	910	5.0	4.5	167.60	16.20	151.40
	77	3.9	255	874	3.5	4.7	364.20	32.60	331.60
	68	5.2	135	893	3.4	5.6	172.00	33.00	139.00
	89	9.6	720	771	4.3	5.7	300.50	78.70	221.80
Mean		5.69***	382**	889.2***	4.0***	5.2***	319.97***	41.70**	278.27***
SE ±		0.881	110.8	32.5	0.26	0.2	70.25	9.42	67.54

** P 0.01

*** P 0.001

SECTION B: METABOLISM AND GROWTH STUDIES, CONDUCTED DURING WINTER FEEDING, ON TWO GROUPS OF CALVES EXPOSED TO NATURAL INFECTION WITH *O. OSTERTAGI*

INTRODUCTION

Recent studies have reported the effects upon growth, nitrogen (N) balance and diet digestibility in dairy-bred calves during part of the winter housing period immediately following natural infection with *O. ostertagi* during one season at grass.

Calves naturally infected with gastrointestinal nematodes and treated with thiabendazole (Thibenzole: Merck, Sharp and Dohme) showed no real differences in digestive efficiency but an overall significantly lowered N retention compared with a paired group of parasite-free control animals. (Parkins *et al*, 1982a). These calves however were given a relatively high protein diet which arguably may have ameliorated the severity of the after effects of infestation. A further study (Parkins *et al* 1982b) examined, using similar techniques, the effects of *O. ostertagi* infection in first season grazing calves during the following winter housing period on growth, N balance and digestive efficiency. During the grazing period these calves had been left either untreated (A) or had been treated with fenbendazole on three occasions; three and six weeks after going to grass and at housing (B), or every two weeks (C). The overall protein content of the winter diet in this study was only 60% of that recommended to sustain a daily growth rate of 0.5 kg, although the energy intake was adequate. Digestibility of the food was not really affected but N balance was always lowest in group A and highest in group C. Most interestingly, group B demonstrated a marked increase in N retention with time after housing and the authors concluded that, under many conditions of suboptimal protein intake, nematode infection in the growing calf can severly affect production even following a recognized anthelmintic programme of treatment.

This present experimental section describes the growth, N balance and feed digestibility recorded from the dairy calves described in Section A and which had completed one grazing season and were then housed for a complete winter housing period. Clinical ostertagiasis and parasitic bronchitis had occurred in the group of infected controls which necessitated treatment with levamisole. The group given an MSRB bolus gained a mean of 20 kg more than the infected control group and showed no clinical signs of ostertagiasis although parasitic bronchitis also occurred in this group. A clean control group remained virtually free of parasites.

MATERIALS AND METHODS

Winter Feeding, Growth, Digestibility and N Balance Studies

The three groups of animals (10 calves in each of the MSRB and Control groups and 7 in the clean group) were housed in adjacent but separate pens fitted with self-locking individual concentrate feeders and hayracks for group-feeding. The housing period was from October 1982 to May 1983 (29 weeks). All the calves were group-fed hay (mean analysis over the whole winter from 18 determinations was; Dry Matter coefficient (DM) 0.84, 84 g crude protein (CP), 326 g crude fibre (CF), 10 g ether extract (EE), 61 g ash per kg DM with 18.0 MJ Gross Energy (GE)/kg DM) and individually given in two feeds per day a weighed allowance of a commercial concentrate (mean analysis also from 18 determinations was; 0.86 DM, 153 CP, 119 CF, 28 EE, 154 ash/kg DM with 16.8 MJGE/kg DM). The feed allowance from October was 5 kg fresh matter (FM) of hay together with 3 kg FM concentrate allowance which was increased to 4 kg FM/d in February.

The cattle were clinically examined routinely. They were weighed and faecal and blood samples collected at two weekly intervals. Faecal samples were examined for trichostrongyle eggs and blood samples analysed for values of plasma pepsinogen, urea, total protein, albumin and inorganic phosphate. Blood samples were also examined for hormonal gastrin activity levels.

During the 29 weeks of housing a total of nine digestibility and N balance studies were conducted when selected animals were housed in metabolism stalls.

The timetable design showing the identity of the calves which were used in each study period is given in Table B1 . Three matched pairs from each of the groups clean, control and MSRB were studied on three separate occasions over the winter period.

The method of faecal and urinary collection, parasitological, biochemical and chemical analysis techniques used are fully described in the General Materials and Methods Section of this thesis.

Calves used for N balance and digestive efficiency examinations were kept in purpose-built metabolism stalls and individually fed for seven days before the recording period. A quantity of hay, adequate for the requirements of the complete digestion study period, was passed through a 2.5 cm screen (Feedmobile, Mill Feed Services Ltd.,) and kept in a damp-proof storage room.

The chopped hay and concentrate was weighed daily before feeding to the animals in the metabolism stalls. The chopping of the hay helped to make the roughage quality uniform during the experiment and it minimised spillage from the feeders compared with giving long roughage which is known to permit waste and selection in similar accommodation.

In the metabolism study periods, two calves from each of the groups were taken at the same time. The calves in the clean and MSRB treated groups were pair-fed to the control animals in any study period where only those amounts of both hay and concentrate consumed by the control animal was offered to the matched pairs in the other groups on the following day.

Table B1. Timetable for individual steers used during metabolism study periods.

			Calf Number		
Date	Period	Week	Clean	Control	MSRB
16-22.11	I	4	G30,66	8,90	76
30.11-5.12	II	6	88,73	65,7	75,384
14-20.12	III	8	74,9	79,92	67,91
18-23.01	IV	13	G30,66	8,90	76,86
16-21.02	V	17	88	65,7	75,384
3-9.03	VI	19	74,4	79,92	67,91
23-29.03	VII	22	G30,66	8,90	76,86
20-26.04	VIII	26	88,73	65,7	75,384
12-17.05	IX	29	74,9	79,92	67,91

RESULTS

Clinical Observations and Liveweight Changes

On housing the cattle were given hay alone and were gradually introduced, over a two week period, to the full initial allowance of 3 kg of commercial concentrate.

All the animals had a normal appearance and appetite until the 17th week of housing (in February, 1983) at which time the steers in the control group became slow in consuming their concentrate allowance. These animals quickly became dull and many exhibited diarrhoea. Animals in the other two groups remained normal in all respects.

There were two clear clinical states apparent during the winter period. Subsequent consideration of metabolic and biochemical measurements are thus related directly to these two distinctly different clinical periods. Since weeks 0-17 constituted a time when the clinical picture was apparently normal, this is now referred to as the pre-type II phase. The clear symptoms of type II ostertagiasis were evident in the control steers during weeks 17 to 29 inclusive and this time period is now referred to as the type II phase.

The mean liveweights of the three groups of animals recorded during the winter experimental period are shown graphically in Fig B1. and are also given in Table B2.

From the start of the experiment until the clinical onset on type II ostertagiasis in week 17, i.e. the pre-type II period, the calves in the clean group gained an average 68 kg (0.58 kg/d), the calves in the MSRB treated group 69 kg and the control animals 67 kg. So the gross liveweight gains of all three groups were similar during the clinical pre-type II period.

In the following five weeks when clinical type II was first observed in many of the control cattle, a mean gain of about 0.6 kg/d was maintained in all groups. However, from week 22 to the end of the housing period (a total of seven weeks), there was a complete change in the liveweight gain pattern.

The animals in all groups, irrespective of the occurrence or otherwise of clinical type II ostertagiasis, apparently gained no weight at all during this last 7 weeks of housing. However, the general appearance of the animals in the MSRB and clean groups was

satisfactory, compared with the dull, diarrhoeic pot-bellied appearance of the control animals. The cessation of growth coincided with a new batch of commercial feed compound being fed. The possible implication of this feed on the animal performance is discussed later.

PARASITOLOGICAL OBSERVATIONS

Faecal Egg Counts

Mean trichostrongyle faeacal egg counts are detailed in Table B3 and are illustrated in Fig. B2. Positive faecal egg counts were recorded from different animals of the clean group on three separate occasions with a mean egg count of seven epg being recorded in each of weeks 17, 20 and 27. Low levels of egg count were also observed in the MSRB treated calves and ranged from a mean of 70 epg in the first week to nil in week 13. Thereafter, a slight increase to a mean maximum value of 83 epg in week 26 was recorded.

At the beginning of the winter period the mean epg of the control animals was 426 epg, but by week 15 this had fallen to an average of 45 epg. Similar egg outputs to those of the MSRB treated group were recorded for the remainder of the housed period, with a maximum of 140 epg being observed in week 26.

POST MORTEM EXAMINATION

At the end of the winter housing period one animal from each group was slaughtered and post-mortem worm counts carried out. Other measurements taken at this time included the pH of abomasal contents, abomasal mucosal fraction and plasma pepsinogen and gastrin levels. Table B4 details these findings. Only O. ostertagi were recovered from the post-mortem examination of the alimentary tract.

Table B4. Post-mortem examination of one animal from each of the clean, MSRB treated and control groups at the end of the winter period.

Group		Clean	MSRB	Control
O.ostertagi	EL_4	0	400	72,500
	L_5	0	200	10,800
	Adults	0	2600	163,800
	Total	0	3200	247,100
Abomasal pH		3	3.7	6.4
Mucosal fraction[1]		1.5	2.1	7.9
Oedema		-	-	+
Plasma pepsinogen[2]		0.9	1.9	10.5
Plasma gastrin[3]		220	240	2,400

[1] g/kg LW [2] I.u. tyrosine [3] pg/ml

The steer taken for necropsy from the control group (No. 83) was one which had a very poor appearance and appetite. The animal taken from the MSRB treated group also did not appear totally healthy and was selected for parasitological examination. The animal chosen from the clean group had unfortunately sustained some physical injury in an accident when attempting to jump a gate during the last fortnightly weighing routine.

At post-mortem, the clean animal had no worms, a normal pH of 3, a normal abomasal appearance with no oedema, a normal plasma pepsinogen value of 0.9 and a gastrin level of 220. The MSRB treated steer had only a total of 3,200 O. ostertagi, most of these (2,600) being adult. All other values were normal. The control steer, however presented a dramatic contrast with 72,500 EL_4 O. ostertagi larvae in a total of 247,100. The abomasal pH was 6.4 and the appearance was of gross oedema (Plate 10, a,b,c). The plasma pepsinogen was 10.5 I.u. and plasma gastrin 2,400 pg/ml. Most importantly, there was a large perforated ulceration present (see Plate 11a). Also present in the contents of the abomasum was a considerable quantity of what appeared to be small, grey-black, grit-sized stones which weighed in total almost 500g (see Plate 11b). Subsequent investigation showed that these were a limestone mineral purposely included, at a rate of about 5% by weight, in the commercial concentrate. Very few particles of

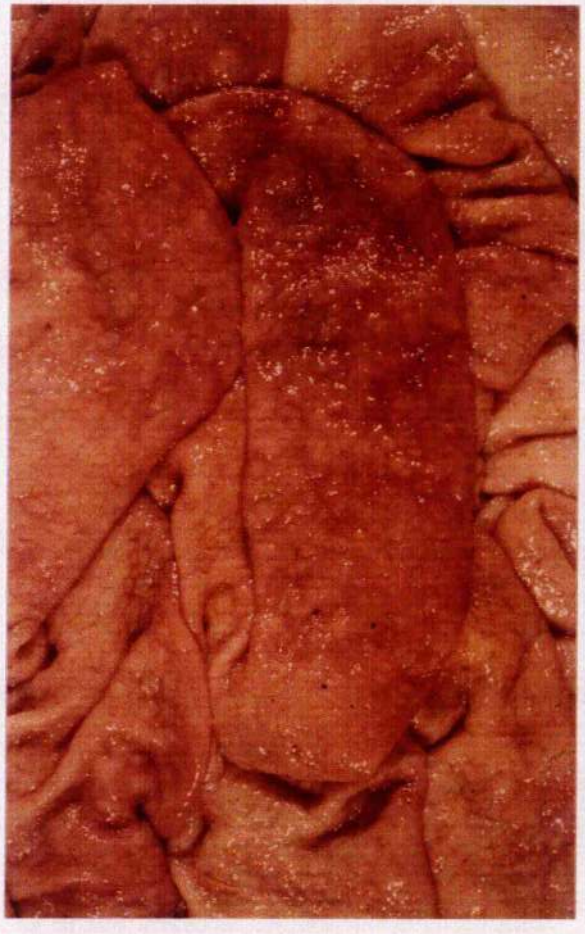


Plate 10a Abomasum from control steer with Type II ostertagiasis.

Plate 10b From steer in MSRB group.

Plate 10c From clean group calf.

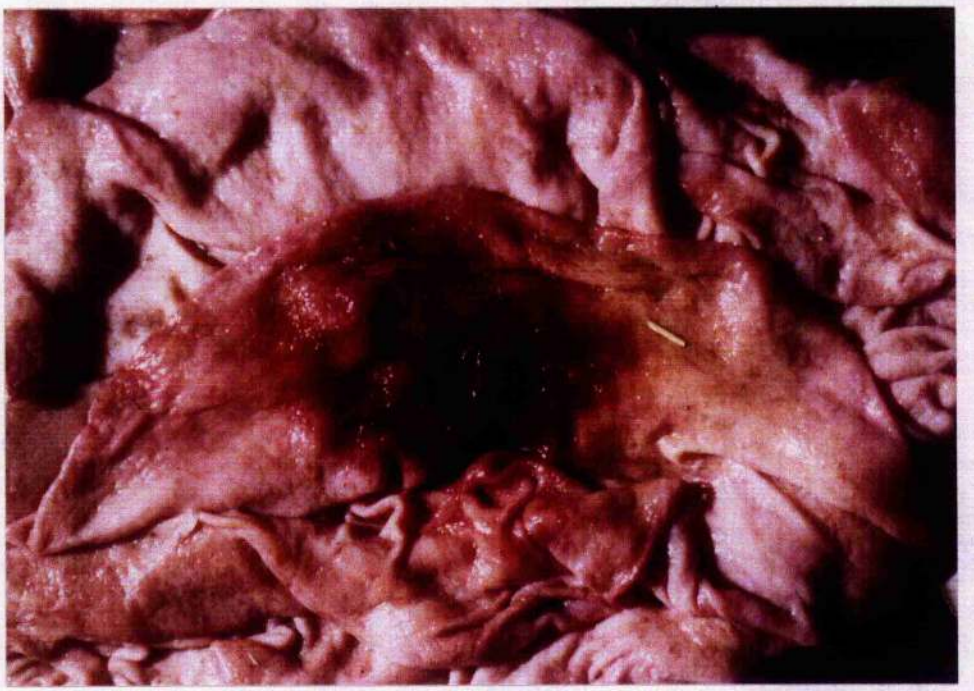


Plate 11a Abomasum from control calf 83 showing gastric ulceration.

Plate 11b Grit collected from abomasum of calf 83.

this supplement were found in the abomasal contents of the other two animals slaughtered at this time. The total ash content of the new batch of feedstuff, which was given from February to May, had an average value of about 170 g/kg DM of which 50 g/kg DM was derived directly from the added limestone grit. Normally such mineral additions are of a fine particle nature.

Further investigations showed however that the limestone grits (obtained by slurrying a quantity of the commercial concentrate cubes with warm water in a plastic bucket and decanting off the food material) dissolved rapidly in a dilute hydrochloric acid solution (at a physiological normal abomasal pH of 3.0).

A discussion on the possible effects of these gritstones upon the steer's performance is presented later.

BIOCHEMICAL ANALYSES

Plasma pepsinogen

Figure B3. shows the pattern of plasma pepsinogen values of the three groups throughout the winter period with mean fortnightly results being detailed in Table B5.

During the entire winter housing period the plasma pepsinogen levels of the clean animals were normal ranging from 0.8 to 1.1 I.u. tyrosine.

Almost normal values were recorded for the MSRB treated calves although in general these were slightly higher than those of the clean group with levels ranging from 1.1 to 1.6 I.u. tyrosine, with the highers values being recorded during the type II period.

The mean pepsinogen levels of the control animals were significantly higher than both of the other groups. A decrease from 6.3 at week 0 to 2.2 I.u tyrosine in week 8 was observed, and remained fairly steady from weeks 8 to 17 (range: 2.2 to 2.9 I.u. tyrosine). An increase in pepsinogen from week 17 onwards was then observed, the maximum value recorded being 7.0 I.u tyrosine in week 29. It was during this period that classical symptoms of type II ostertagiasis was observed in several animals. Since only one animal per group was slaughtered, no correlation with worm burdens were determined.

Plasma Gastrin Levels

After housing the plasma gastrin levels were estimated, in five

animals of each group, on seven occasions, in weeks 0, 8, 13 (the pre-type II phase) and in weeks 17, 24, 27, and 29 during the type II phase. The sampling dates chosen were dictated by the availability of kits and the clinical symptoms of the disease. Mean plasma gastrin levels of the clean, MSRB treated and control animals are shown in Fig. B4. and are also detailed in Table B6.

Following housing a slight increase occurred in the clean and the MSRB treated groups from below a mean 130 to around 200 pg/ml. The levels in the control animals fell from 590 at the beginning to 292 pg/ml in week 13, i.e. during the type II phase. Thereafter the values remained at approximately the same level in the clean and the MSRB treated groups. In contrast a marked and steady increase of plasma gastrin values occurred in the control group after week 13, reaching a maximum mean value of 784 pg/ml in week 27. This increase coincided with a rise in plasma pepsinogen and clinical symptoms of type II ostertagiasis, diarrhoea, loss of appetite and bodyweight reduction in some animals of this group. As the clinical signs abated the mean gastrin levels fell to 573 pg/ml by week 29. Since only one animal per group was slaughtered, it was not possible to make correlations with worm burdens. The correlation between plasma gastrin and pepsinogen determined on the same samples throughout the winter was only 42.6 per cent.

Other Biochemical Analyses

The results of blood analyses performed at times during the winter period are shown in Table B7. The plasma urea concentrations during the winter period were all fairly low and were typical for growing cattle given a hay plus concentrate store ration. Similarly, the inorganic phosphate concentrations were all very similar and were also considered normal. The albumin concentrations, however, were always lower (and generally significantly) in the control cattle compared with the mean values for the animals in the other two groups at all times during the winter. The total protein concentrations however did not always reflect the lowered albumin levels in the control cattle since the clean cattle generally maintained the highest total protein concentration with both the control and MSRB treated cattle having fairly similarly reduced concentrations.

METABOLISM STUDIES

Digestibility studies

A total of 52 individual digestibility and concurrent N balance studies were conducted during the winter period from November 1982 to May 1983.

The results for the digestibility of the whole diet given to individual animals in the clean, MSRB treated and control groups are given in Table B8. Mean values for the digestibility coefficients for each of the feed fractions in the pre-type II and type II phases are summarised in Table B9. Statistical comparisons between group means are also shown in this table. Inspection of the individual values reveals the amount of variation recorded in the digestibility coefficients between different collection periods (and therefore different animals). The mean values during the pre-type II phase for DM and gross energy (GE) apparent digestibility were significantly lower in the control calves (0.54 DM and 0.56 GE) than in either of the other two groups (both with 0.57 for DM and about 0.59 for GE). The apparent digestibility for all other feed fractions was also lower in the control group (but not significantly) than in either of the other groups.

Calculation of the ME supplied and hence the predicted liveweight gain was made on the daily ration of 5 kg hay and 3 kg concentrate FM and using the experimentally determined digestible energy (DE) values during this pre-type II phase. A total input of 6.8 kg DM supplied a determined 71 MJ DE which was calculated (x 0.82) to supply 58 MJ ME where the ration M/D was 8.5 MJ/kg DM. For steers of about 250 kg this was calculated (MAFF *et al*, 1975) to permit a daily liveweight gain of about 0.6 kg per day.

Similarly the ration was calculated (using the experimentally determined digestibility coefficient for crude protein) to supply about 400 g of digestible crude protein (DCP) which is the amount suggested as being that required for maintenance of a 250 kg steer and the provision of 0.7 kg LWG/d (ADAS, 1976). The DM intake of 6.8 kg was close to the maximum appetite limit of 6.6 kg DM suggested by MAFF *et al* (1975).

In the clinical type II phase from weeks 17-29 (i.e. collection periods VI-IX inclusive) the ration allowance was changed a little as the daily concentrate allowance was increased in early February from 3

to 4 kg (FM). During the type II phase, statistically both the OM and GE digestibility were significantly greater in the MSRB group compared with both the clean and control calves. Otherwise there were no differences in apparent digestibility of the other feed fractions.

Using the experimentally determined apparent digestibility coefficients for GE, the daily DE intake was calculated as being about 80 MJ. A total offered input of 7.7 kg DM thus supplied a calculated 65 MJ ME (80 x 0.82) with an M/D = 8.4. For steers of about 350 kg LW (MSRB treated and clean calves from week 22) this was calculated (MAFF et al, 1975) to support a daily LWG of just under 0.5 kg. This ration supplied a determined 470 g DCP per day which was a little in excess of the 405 g suggested by ADAS (1976) as that being required for a DLWG of 0.5 kg by 350 kg animals. A reduction in voluntary intake of say 2 kg of total diet is calculated to reduce this theoretical LWG potential to little over maintenance.

When all the 52 individual digestibility studies are combined and the overall means are compared it can be seen that in the control group, DM, CP and GE apparent digestibilities are significantly lower than those in the MSRB group. Only energy digestibility, however, was significantly lowered in the control group when compared with the clean group.

Nitrogen balance studies

Individual and mean nitrogen balance data for each of the periods I-IX are given in Table B10. This mean data of faecal, urinary and retained nitrogen expressed as a percentage of N input is presented in Table B11. This same data is also presented graphically. Figures B5, B6 and B7, depict faecal, urinary and retained nitrogen as a percentage of input respectively for each of the recording periods I-IX. Further, as for the digestibility data, the mean daily N balance data (gN and % of input) are presented in Table B12 for the pre-type II phase (periods I-V inclusive) and the type II phase (periods VI-IX inclusive) and for the overall experiment (periods I-IX inclusive). This is also presented in a bar chart in Fig. B8.

It should be remembered that in these studies a pair-feeding system was operated whereby the clinically unaffected clean and MSRB treated calves were restricted to the amount of feed consumed by the

control calves i.e. a dual pair-feeding system.

Inspection of the individual data shows there to have been close pair-feeding in all periods excepting one (period VI) where impaired appetite of a control calf (no. 92) was, unfortunately, not able to be matched to it's pairs in the other two groups.

The individual percentage outputs of faecal and urinary nitrogen however were seen to be quite varied from period to period and the mean values shown graphically in Figs. B5, B6 and B7 clearly depict the changes which occurred with time; most especially the more dramatic retention failure which occurred in the type II phase for the control calves.

Examination of the meaned balance data for pre-type II and type II phases together with that for the total winter period shows several interesting comparisons. (TABLE B12) In the pre-type II phase, with a mean daily input of 131 g N, about 32% of this (41 g) was excreted in the urine; there being no difference between groups. Mean faecal N output was however a little higher (but not significantly) in the control calves (66.6 g/d i.e. 51% of input) compared with the other two groups (about 61 g/d i.e. 47% of input). The percentage retentions of 22.1, 21.1 and 18.2 for clean, MSRB and control calves were all quite similar. This is clearly seen also in the bar graph representation of Fig. B8.

In the type II phase the mean N intakes are similar to those of the pre-type II phase even though an additional 1 kg of concentrate is being offered in the ration because the voluntary intake (of hay particularly) was reduced in the control calves and so the pair-fed MSRB treated and clean calves were restricted accordingly.

In this phase again there were no significant differences in faecal outputs (which again represented about 50% of the N input) but there was a significant increase in urinary N output in the control group over the clean group. This occurred mainly in period VI (see Fig. B7.) and contributed to the mean output of 51g N/d representing 42% of the dietary input. This output was not, however, significantly greater than the 38% excretion of the MSRB treated group. The resultant retention (see bar graph Fig. B8) of 7.2% of N input was less than half that of the other two groups and the pattern of this depressed retention is clearly depicted for each collection period in Fig. B7.

When these values are averaged over the whole experiment the clear changes which occurred in the type II phase are reduced and only faecal

N output and retention become significantly different to the corresponding values in both the MSRB treated group and the control group. Urinary N outputs over the whole experiment are very similar, representing about 35% of the dietary N intake.

DISCUSSION

Since Anderson et al (1965b) first classified the disease, several authors have discussed the clinical symptoms and biochemical changes which occurred in the clinical phases of type I and type II ostertagiasis and referred to the asymptomatic and biochemically inert phase of pre-type II (Armour and Ogbourne, 1982). However information about the changes in metabolism during all phases of bovine ostertagiasis are few and mainly related to type I disease produced by artificial infection with only O. ostertagi larvae (Canale et al, 1977) or mixed with C. oncophora (Randall and Gibbs, 1981; Jordan et al, 1977). The first reports about metabolism aspects in animals naturally infected during the grazing season were those of Parkins et al (1982 a,b). These studies were made during the first 17 weeks of the following winter and compared infected animals, with or without anthelmintic treatment at housing with uninfected clean control animals. More information is available on ovine ostertagiasis but, as in the bovine it relates primarily to artificial infections (e.g. Parkins et al, 1973, Sykes and Coop, 1977; Coop et al, 1977; Coop et al, 1982

In this chapter the results discussed pertaining to the whole winter period of 29 weeks and, for ease of analysis, is divided into the asymptomatic pre-type II phase (weeks 0-17) and type II phase (weeks 17-29) associated with the clinical symptoms of the disease.

Historical background

At the end of the grazing period six animals of the MSRB treated group and five of the controls were sacrificed. The animals of the control group, which had suffered type I ostertagiasis and were treated with levamisole five weeks before necropsy, had a total mean burden of 319,970 O. ostertagi. Only 13% of these worms (38,990) were adults, and the remaining 87% were arrested EL_4; the latter burdens are considered sufficient to produce type II disease in the following spring (Anderson et al, 1965 b). Meanwhile, in the MSRB treated

animals the mean total of O.ostertagi was 17,508 (95.4% less than the controls) with 70% being arrested EL_4 (12,233) and the remaining 30% (5,275) being adults and developing stages. These are light infections and the number of arrested EL_4 do not represent enough to be capable of producing type II in the following spring.

The significantly higher values in plasma pepsinogen and gastrin and the lowered serum albumin levels found in the control group compared with those of the clean and the MSRB treated animals, were clearly an expression of the increased abomasal damage in the controls (Jennings et al, 1966; Anderson et al, 1981). During the winter period the amount of food given to the animals was calculated to allow a daily liveweight gain of about 0.6 kg/day (MAFF et al, 1975) and the amount of protein supply was enough for the anticipated weight gains (ADAS, 1976). A novel dual pair-feeding system was used during the whole winter experiment in order to overcome the problems associated with a possible lack in appetite developing in the control cattle.

Pre-type II phase, (0-17 weeks)

From the time of housing there was a sustained reduction in both plasma pepsinogen and gastrin levels in the control animals which coincided with a dramatic decrease in the faecal egg counts. These results probably reflect a steady loss of existing adults and a gradual reduction in the number of new adults occuring from the population of arrested larvae.

A similar decrease of plasma pepsinogen has been described by Jørgensen et al (1978) and Armour et al (1979) at the beginning of the winter period in cattle naturally infected in the previous grazing season. An anthelmintic treatment was given at housing in the latter work. In the clean and MSRB treated groups a slight increase in plasma gastrin levels was observed which could possibly be explained by the change in the type of feeding i.e. from grass to hay plus concentrates. The total weight gains of the animals in the three groups during this phase were very similar, being around 68 kg per head and were similar to those recorded by Parkins et al (1982 a,b) over a similar period of time where also a hay plus a barley based concentrate diet was given.

No lag in liveweight gain occurred following housing and no major differences in bodyweight developed between control and either clean or MSRB treated animals as was observed by Tolling et al (1981). In

their study a mean 20 kg in difference between naturally infected control animals and early season MSRB treated cattle had increased to 36 kg in the six weeks following housing and reached 44 kg by the end of the winter. The feed was available ad lib in this experiment and appeared to be a high M/D diet. Jørgensen et al, (1978) and Tornquist et al, (1981) have also reported some 'compensatory' liveweight gain in infected controls. This occured during the first week of the housed period and may reflect the response of animals which had previously shown a lack of appetite and 'compensation' could be accounted for by a simple increase in gut fill.

During the pre-type II phase in the current experiment the apparent digestibilities of DM and gross energy were significantly less in the control calves than in those of the clean and MSRB treated groups. Other digestibility values were also lower, but not significantly so, in the control animals.

In abomasal parasitism there is some evidence that a reduction in the apparent digestibility of DM occurs e.g. in bovines, Canale et al (1977) and Parkins et al (1982 a), and in ovine ostertagiasis Parkins et al (1973) and Coop et al (1982).

The lower apparent digestibility of the gross energy in the control group was not accompanied by a reduced bodyweight gain. However, calculation showed that the lowered GE digestibility of the control cattle would result in a reduction in the daily ME supply of less than 3 MJ (0.03 GE coefficient reduction x 120 MJGE supplied in the ration x 0.82 factor to convert DE to ME). Over the whole 17-week pre-type II phase this amounted to a calculated reduced ME intake on about 360 MJ (equivalent to about 27 kg barley DM). This could be calculated, for such cattle on the mixed hay/concentrate ration, with an M/D of about 8.5, to have resulted, at most, in a reduction of about 4 kg LW. This calculated weight differential clearly could be outside the limits of experimental error in this study.

Differences in liveweight however do not always represent a straightforward difference in accumulated bone, fat and lean tissue. Differences in both gut fill and total body water content can represent many kilograms of apparent 'liveweight'. Also the differences in carcass composition observed in the necropsy animals at the end of the first grazing season have been fully discussed in Section A of this thesis. The significantly lowered plasma albumin concentrations recorded in the control cattle might have considerably increased water

retention.

A further complication, is the fact that all three groups were offered the same quantities of the mixed hay and concentrate ration. In the pre-type II phase the lower mean liveweight of the control group of about 25-30 kg (compared with the other two groups) meant that this group had a reduced daily ME cost for maintenance of about 2-3 MJ (MAFF et al, 1975), which fortuitously corresponds to the amount of ME apparently 'lost' to the control animals because of the significantly lowered digestibility observed in this phase.

The concurrent N balance data collected in this phase shows a reduced daily N retention for the control calves accounted for entirely by an increased faecal N output. The dietary intakes here in the metabolism studies were very similar in animals from all three treatment groups because of the success of the novel dual pair-feeding system where intake of the control animal governed the intake of the two paired animals. In two experiments conducted with housed calves following natural infection with O. ostertagi during a first season at grass Parkins et al (1982 a,b) reported little differences in the digestibility of diets given to either the parasite exposed or a similar group of clean control calves. However the CP digestibility was generally lower in the infected animals even though they had received a recognised anthelmintic treatment at housing. In the first experiment where a high protein diet was given, the significantly lowered N retention in the infected calves was due to approximately equal increases in both urinary and faecal N outputs. In the second trial, where the dietary protein intake was only 60% of that recommended, the lowered N retention in the infected calves was mainly due to a higher N excretion in the urine. In both experiments the infected cattle had been treated at housing and observations did not extend beyond 18 weeks of housing which corresponds only to the pre-type II phase of this currently described work. The reasons for the reduced digestibilities and N retentions in this current work are not entirely clear but clearly the remaining adult Ostertagia worm burden coupled with a slow maturation of arrested larvae during this period may play a significant role. Certainly other observations on the higher egg counts, increased pepsinogen and gastrin levels and albumin during this phase would indicate that some tissue damage was still present.

Type II phase, (17-29 weeks)

The clinical, structural and biochemical changes observed in type II ostertagiasis are basically the same as those found in type I but usually more extensive (Anderson et al, 1965 b). In both types the severity is affected by the number of young adult and developing parasites and is increased when high numbers of larvae develop synchronously and in a short period of time, as occurs with arrested larvae in type II.

In the current experiment a change was observed, from February onwards, in the normal feeding behaviour of some animals in the control group. Initially this was seen as a considerable delay in the time required to ingest a given quantity of the concentrate food as compared to other animals in the group. Subsequently a reduction in appetite was recorded when the animals were individually fed in the metabolism stalls and a mean 32% decrease in intake of the total feed offered was recorded in those animals of the control group which were clinically affected. However, this reduction was not fully reflected in the liveweight gain of the whole group because the hay, which was refused by some animals in the group pen, was eaten by other members of the same group which were less affected. The lack of appetite in ostertagiasis has been widely reported (Armour et al, 1973; Dargie, 1975) and per se is probably the major contributor to impaired production (Topps, 1983).

Coincidental with the lack of appetite there were sharp increases in plasma pepsinogen and gastrin levels which were maintained until the end of the housing period. But, unlike the situation observed in type I there was only a slight increase in the faecal epg's. Anderson et al (1965 b) also noticed that the epg's of cattle with type II ostertagiasis were frequently low and on some occasions negative. These authors therefore suggested that the plasma pepsinogen values were a better diagnostic aid in the type II disease and reflected the changes occurring in the abomasum such as loss of acidity and increased permeability of the mucosa. Selman et al (1976) agreed with the previous findings of Anderson and his colleagues and suggested that plasma pepsinogen values in excess of 3.0 I.u. of tyrosine were indicative of significant abomasal damage. However, where pepsinogen readings of between 1.5 and 3.0 I.u. were recorded, difficulties arose in interpretation since those levels are achieved in many animals which are constantly exposed to natural infection but without the presence of

large numbers of developing Ostertagia parasites. In the current experiment there was a parallel increase of plasma pepsinogen and plasma gastrin; the latter rose to levels around a mean of 800 pg/ml which is more than four times that of the clean group and three times the initial level of the infected control group. It is possible that the increase in gastrin is a positive response to a loss of acid conditions in the abomasum and, taken in conjunction with the elevated pepsinogen values, indicates that significant populations of developing Ostertagia parasites are present. Since the cattle were under constant dietary conditions at this time it is unlikely that the elevation in gastrin could have occurred for other physiological reasons. The diagnostic value of elevated pepsinogen and gastrin levels as an aid in detecting active lesions of type II ostertagiasis could be considerable since collectively they appear to indicate the need for treatment to remove developing parasites.

Owing to the limited number of available animals and since one principal objective of the experiment was to study the production effects of parasitism and the benefits of the MSRB system as a control over a two year period, it was not originally intended to slaughter any animals during the winter period. However, due to the severity of the type II disease in animal number 83 of the control group, it was decided to sacrifice this animal. For comparison purposes one animal from each of the clean and the MSRB treated group were also slaughtered. In order not to affect the continuity of rotation of the animals in the metabolism stalls, these two animals were selected from the calves which were not utilised in that part of the experiment. The worm burden at necropsy of No. 83 was typical of clinical type II ostertagiasis, namely an Ostertagia burden of 247,100 of which 163,800 (70.3%) were adults; this is a complete reversal of the composition of worm burdens in the calves necropsied in the previous October when only 13% of the burden were adults and developing larvae and 87% were arrested L_4. The high pH of the abomasum content, the mucosal hyperplasia and oedema and the very high plasma pepsinogen and gastrin levels correlate well with the high burden of adult Ostertagia and also conform to the classic picture of type II ostertagiasis. The clean group animal was negative for the presence of parasites and lesions could not be detected at necropsy. The MSRB treated animal had only 3,200 Ostertagia worms and the biochemical parameters were normal for the age of an animal with a previous history

of exposure to internal parasitism, however it might be suggested that a value of 1.9 I.u. of pepsinogen in the plasma of the MSRB treated animal is higher than considered normal by many authorities, but it is the opinion of this author that it is normal for cattle of this age and previous grazing history and level of exposure to Ostertagia. Interestingly, only 100 Cooperia parasites were detected in the control calf although an average of 15,340 were present in the controls necropsied at housing in autumn, and yet again this emphasises the high degree of immunity which develops to this parasite.

During the type II phase the MSRB group recorded higher digestibility coefficients than those of the clean and the control groups, being significant in both gross energy and OM digestibility. Although a pair-feeding system was used, the mean increase in liveweight during this period was 28, 25 and 21 kg in the clean, MSRB treated and control groups respectively. These differences showed that the clean animals were apparently the most efficient at food conversion irrespective of the digestibility data. The MSRB treated animals had an apparently superior digestive efficiency to that of the clean group but the mean liveweight gain was 3 kg less. According to the severity of the clinical symptoms showed by the control group greater differences were expected, however, the possibility of an important retention of water has to be considered due to the finding of a slight carcass oedema in the control animal killed at the end of the winter and the consistently significantly lower albumin levels in the control group. The water retention in type II ostertagiasis has previously been described by Halliday et al (1965).

Due to the lack of comparative information on type II ostertagiasis in cattle and considering this phase of the disease as one of an 'active parasitism' produced by the development of the previously arrested L_4, it is possible to compare the data obtained by artificial infection with Ostertagia spp. to produce type I ostertagiasis in both the bovine and ovine species. Studies with a single massive infection over a short period of time showed no differences in apparent digestibility coefficients in clinical and subclinical artificial infection with O. ostertagi by Canale et al (1977) and Jordan et al (1977). However, Randall and Gibbs (1981) found a decrease in CP and energy digestibility at week three post infection but recoveries were seen at week five after infection with C. oncophora and O. ostertagi; pair-feeding was not used in these

trials.

In ovine ostertagiasis, Parkins et al (1973) found few differences in digestibility values between infected and control animals using one massive dose of O. circumcincta in each of three experiments. However, the values were constantly lower in the infected animals. Reduction in apparent digestibilty of CP was found by Sykes and Coop (1977) in chronically infected growing sheep over a 14 week period. These differences were recorded at the beginning of the trial (weeks 2-3) but the effect gradually becomes less marked with time. Less total body protein deposition was found in the infected animals at the end of the experiment.

In the current experiment in the overall winter period (0-29 weeks) the mean DM and CP apparent digestibilities of the MSRB treated group were significantly higher than those of the control group. The corresponding digestibilities in the clean group did not differ significantly from either group but was higher than the control group. The overall apparent digestibility coefficents of the dietary gross energy in both the MSRB treated and clean groups was significantly higher than in the control animals. The remainder of the feed fraction digestibility values of the control group were consistently lower than the other groups. This higher overall digestibility of the diet obtained by MSRB treated animals is seemingly not reflected in improved liveweight gains. This may be because even moderately parasitised animals might require more energy and amino acids for essential function and so they are diverted from growth. Thus, Jones and Symons (1982) have suggested that the synthesis of albumin and repair of damaged tissues, increased immunoglobulin synthesis and tissue hyperplasia have priority over liveweight gain as tissue deposition when animals are under the challenge of a parasitic infection.

Other lower digestibility coefficients in the control group could be explained by an increase in intestinal motility resulting in reduced transit times through the digestive tract with consequent impaired absorption. However, another possible influence on the transit rate of ingesta could be an alteration in reticulo-rumen motility as described in clinical ostertagiasis by Martin et al (1957) which were found to be weak and of short duration. Bueno et al (1975) reported a reduction in the frequencies of reticular and abomasal bulb contractions with an increase in the flow of ingesta in the intestine of animals infected with the abomasal parasite T. axei and also the

large intestinal parasite C. ovina. Gastrin could probably be involved in this process as Bell et al (1977) reported that a physiological quantity of pentagastrin injected into calves reduced the time of abomasal emptying by affecting gut motility.

The digestibility coefficients of the whole diet can be compared for any one group between the pre-type II and the type II phases of the disease. Generally increased digestibilities for DM, OM and GE are noted in the type II phase and are entirely predictable since the concentrate portion of the ration allowance had been increased from 3 to 4 kg FM in this phase. There was a considerable reduction in the intake of some individual control animals in the metabolism stalls during this phase and the corresponding pairs in both the MSRB treated and the clean groups had been restricted to the infected animals intake of hay and concentrate. A lowered food intake, in some circumstances, may increase the apparent DM, OM and CF digestibilities (see McDonald, Edwards and Greenhalgh, 1981) but this did not occur to the same extent in the clean animals. An excellent short review by Topps (1983) discusses the effect of parasitism on gut function, digestion, absorption, protein and mineral metabolism.

The possible role of the parasites in these animals, perhaps via hormonal or neural stimulus, in initiating a compensatory improved digestibility has to be considered. Steel (1978) in a sophisticated experiment demonstrated that the small intestine is able to compensate the abomasal dysfunction in ovine ostertagiasis, by enhancing digestion and absorption lower down the tract. This mechanism can also explain the results of Sykes and Coop (1977) in sheep chronically infected with artificial infections of O. circumcincta in which a gradual recovery of nitrogen digestibility occurred with time. It is clear that in type II ostertagiasis the N retention is severly affected, careful examination of the N components of the urine should be considered in future experiments.

The reasons for the poor liveweight gain performance of all the steers in the last seven weeks of housing are not immediately clear and may be complex. Certainly as the animals became heavier the available resting area in the metabolism stalls was reduced and may have contributed markedly to an increased stress known to be associated with such experiments. The reduced feed intakes of the control animals, when in the stalls, was shared (because of the paired feeding regime) by the animals of the other two groups. Reductions of liveweight gain

during the metabolism periods would thereforelower the group means. The fixed allowance of 5 kg hay and 4 kg concentrate in that phase was calculated to provide adequate energy and protein for maintenance and the provision of about 0.4-0.5 kg LWG per day. Certainly the presence of some 5% by weight (i.e. 200 g per day) of limestone chips in the concentrate feed might be thought to have had an effect upon normal digestive function but the metabolism studies showed no gross abnormality. In the steer necropsied from the control group the accumulation of the limestone particles was clearly related to the lack of hydrochloric acid production in the abomasum (the pH of the abomasal fluid at post-mortem was 6.4). In the other cattle, normal HCl production (as shown by the normal abomasal pH's) would have dissolved the limestone in time. It might also be speculated that exacerbation of the ulcerous condition occurred as a direct result of the physical abrasion caused by the accumulating limestone particles. In all the control cattle, where HCl production may have been reduced by the occurrence of the type II disease, it may also be speculated that some physical tissue damage may have been caused by the limestone.

From the data of the current experiment it appears that protein metabolism is severely affected by the levels of O. ostertagi infection found in this trial. Also the results perhaps did not fully reflect the real damage the Ostertagia infection produced locally, due to the capability of the small intestine to increase digestion and absorption. The pancreas could also be involved in compensating by enzyme production in CCK as has been described in T. colubriformis infection of lambs by Symons and Hennessy (1981).

In ostertagiasis, hypercatabolism of protein due to an increase in loss of albumin in the stomach has been widely described (Mulligan et al, 1963; Nielsen, 1966;, Halliday et al, 1968; Holmes and McLean, 1971; Yakoob et al, 1983). Reduction in gastric protein digestion is based on the loss of differentiated acid secreting cells and also pepsinogen producing cells which occurred in abomasa infected with Ostertagia parasites (Ritchie et al, 1966; Murray et al, 1970). It appears that in the clinical disease the protein loss exceeds the capacity of the compensation by the small intestine in digestion and absorption and part of this protein is excreted in the faeces. This is expressed in lower apparent digestibility coefficient of CP and reduction in N balance, also shown by N retention (Sykes and Coop, 1977; Symons et al, 1981). Another possibility is that part of the

lost protein can reach the large intestine where bacteria degrade it to NH_3 and, after being absorbed, it is transported to the liver where it is converted to urea and excreted by the kidneys in the urine, or part can be used for re-synthesis of amino acid as is necessary (Vernon and Peaker, 1983). The urea excreted in urine will also affect the N retention by increasing the N output (Parkins et al, 1973; 1982 b). The possibility of compensatory digestion and an increase in absorption by the small intesinc will necessarily depend on the integrity of this tissue. For this reason concurrent infections with abomasal and intestinal helminth infections, could have a retarding effect on compensatory digestion in the small intestine. However, although a concurrent infection with the intestinal parasite C. oncophora was present as shown in data obtained from animals slaughtered in the autumn the development of a strong immunity to C. oncophora and the loss of existing infections during the winter suggest that the alterations recorded in the type II phase were primarily the responsibility of O. ostertagi.

Fig. B1

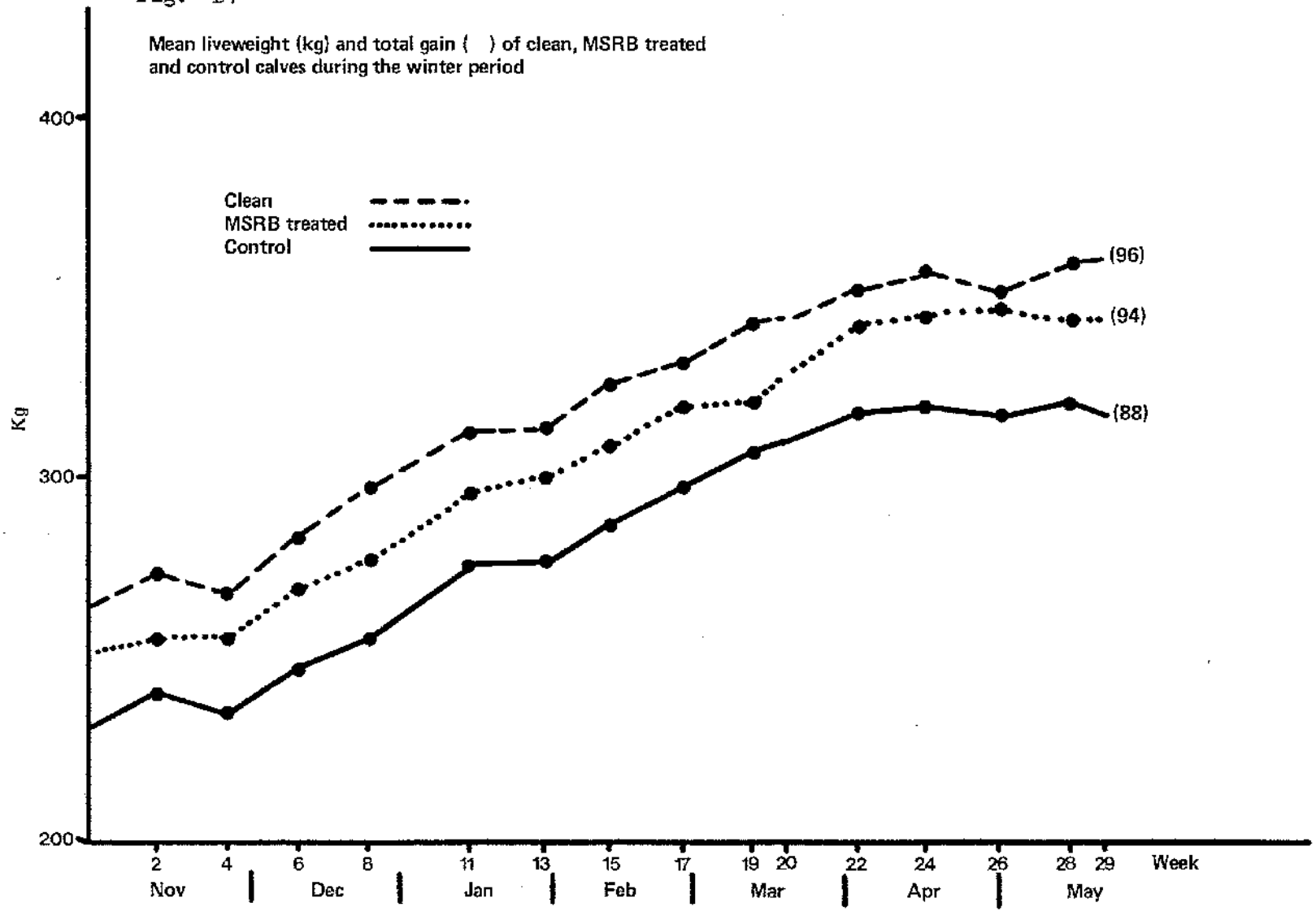

Mean liveweight (kg) and total gain ( ) of clean, MSRB treated and control calves during the winter period
Clean
MSRB treated
Control
400
300
200
Kg
(96)
(94)
(88)
2 4 6 8 11 13 15 17 19 20 22 24 26 28 29 Week
Nov
Dec
Jan
Feb
Mar
Apr
May

Fig. B2

Mean trichostrongyle egg count in faeces of clean, MSRB treated and control calves during the winter period

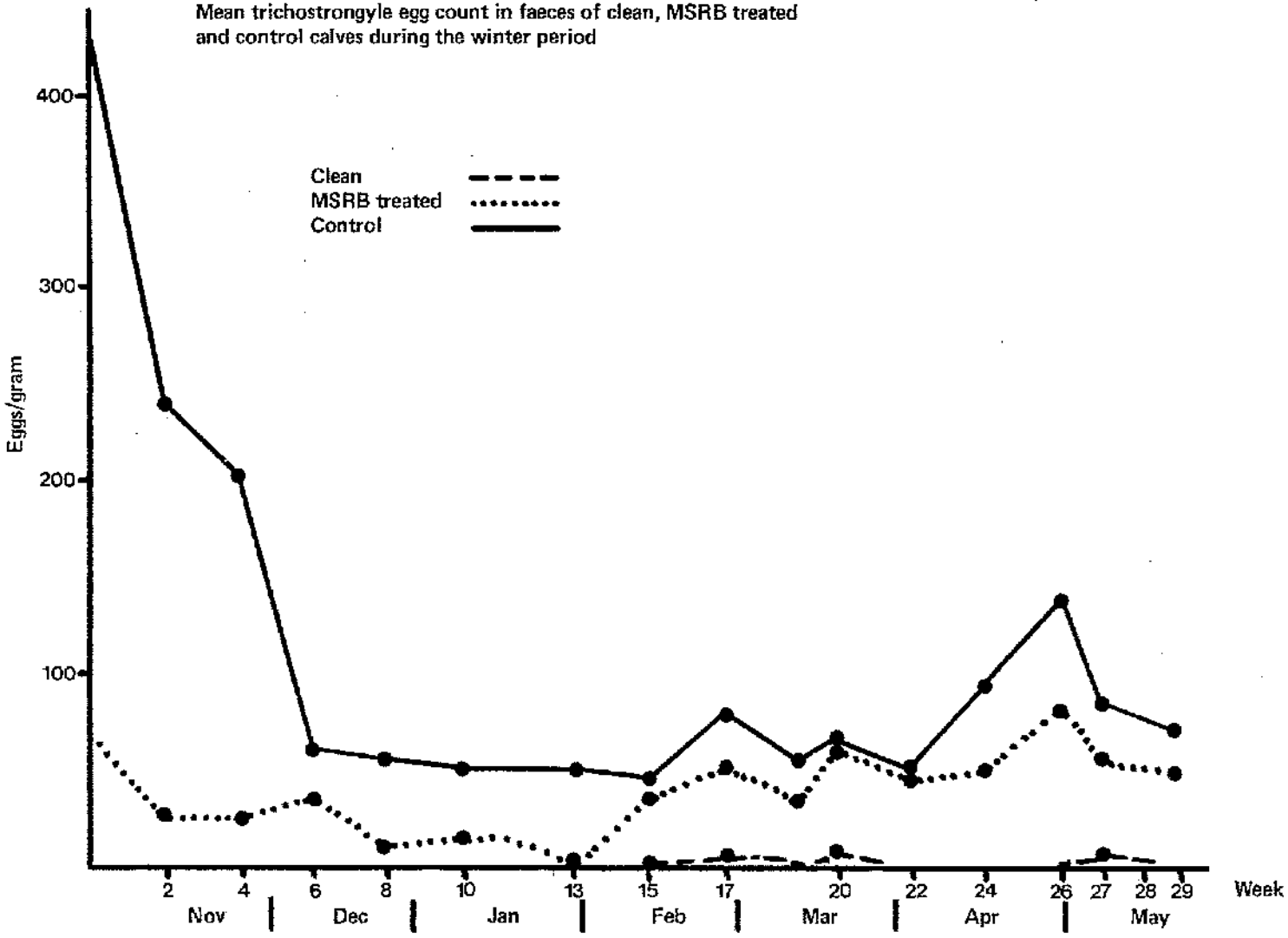


Fig. B3

Mean plasma pepsinogen levels (Iu tyrosine) of clean, MSRB treated and control calves during the winter period

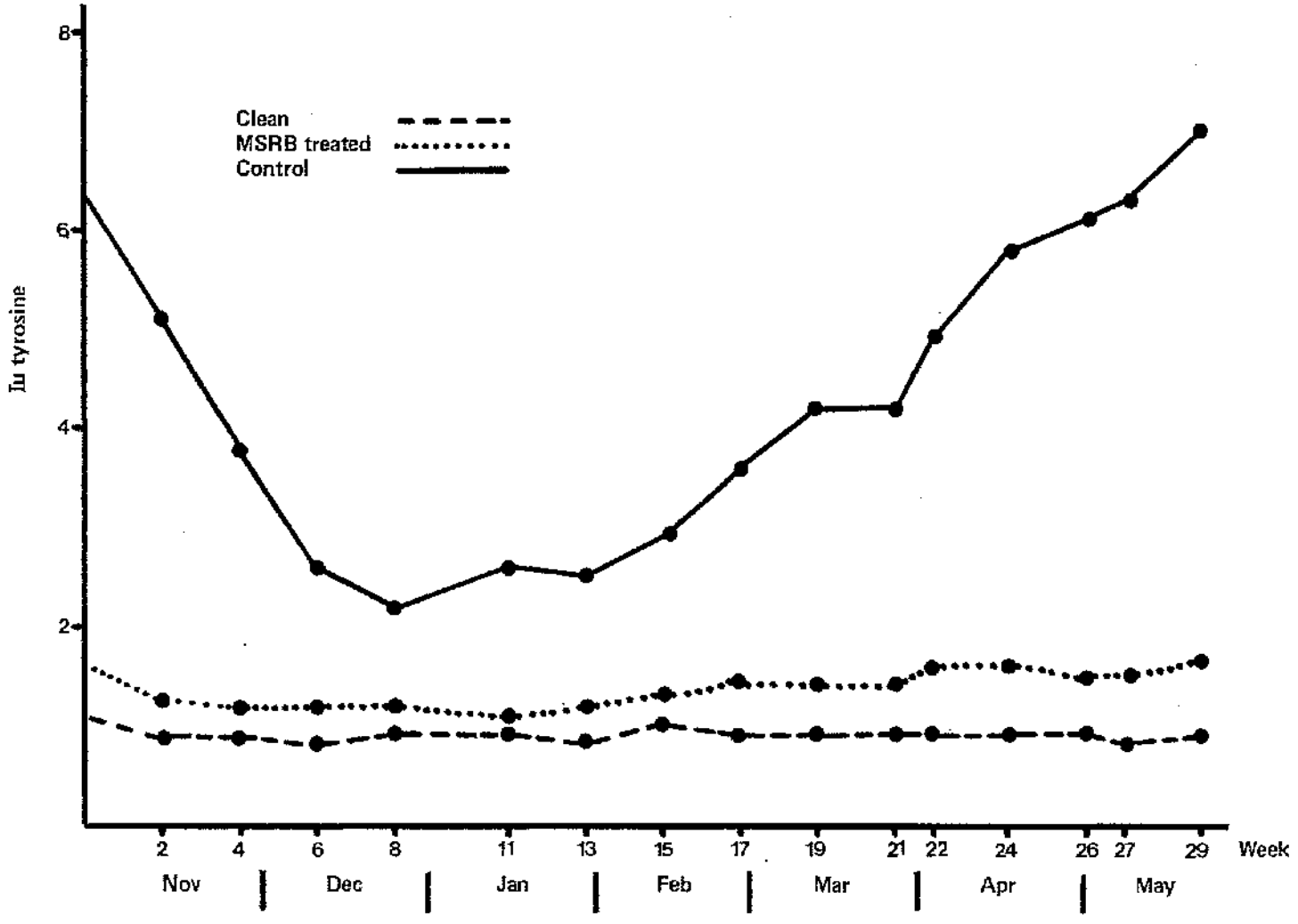


Fig. B4

MEAN PLASMA GASTRIN LEVELS OF CLEAN, MSRB TREATED AND CONTROL CALVES DURING WINTER PERIOD

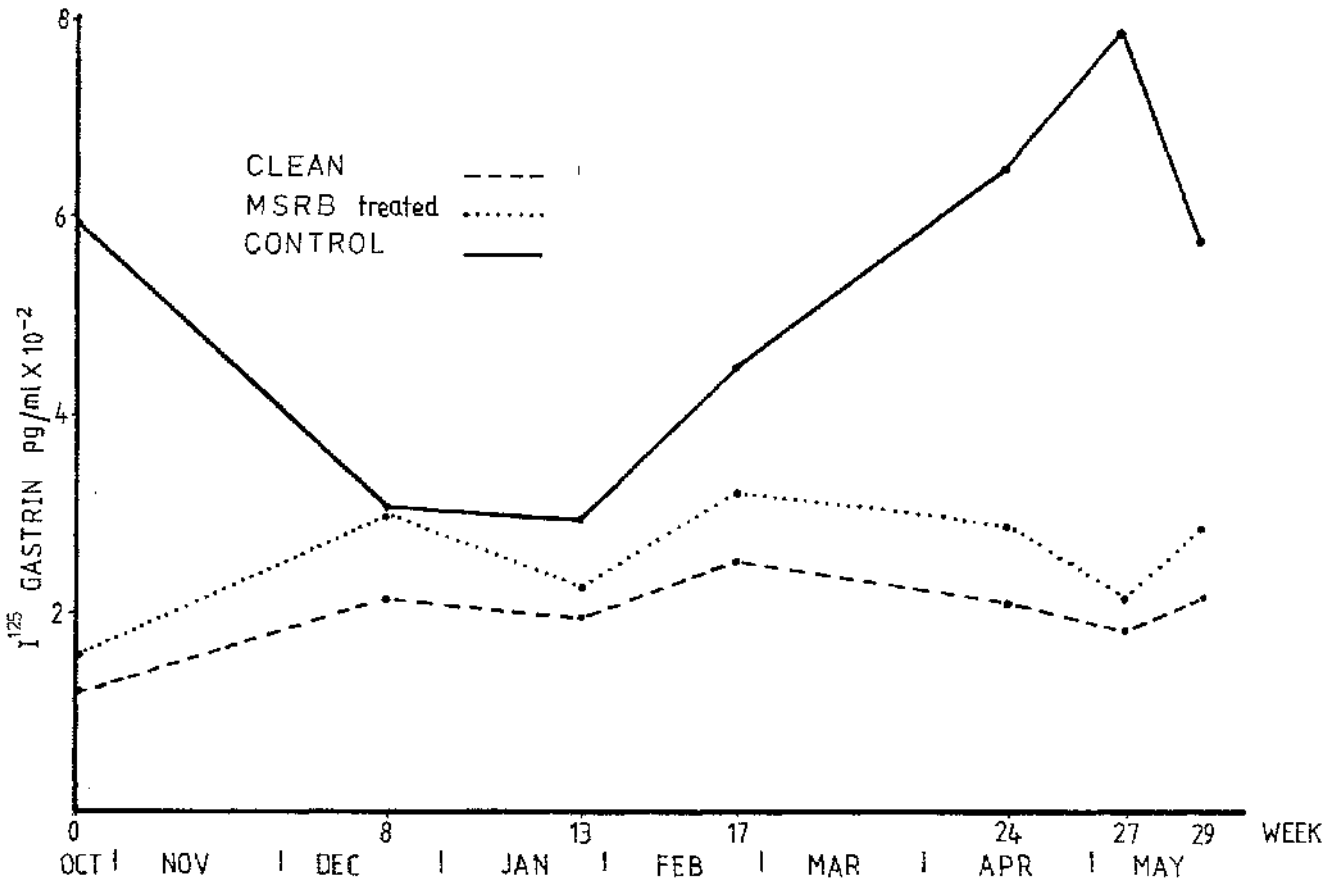


Fig. B5

FAECAL NITROGEN OUTPUT (% OF INPUT)

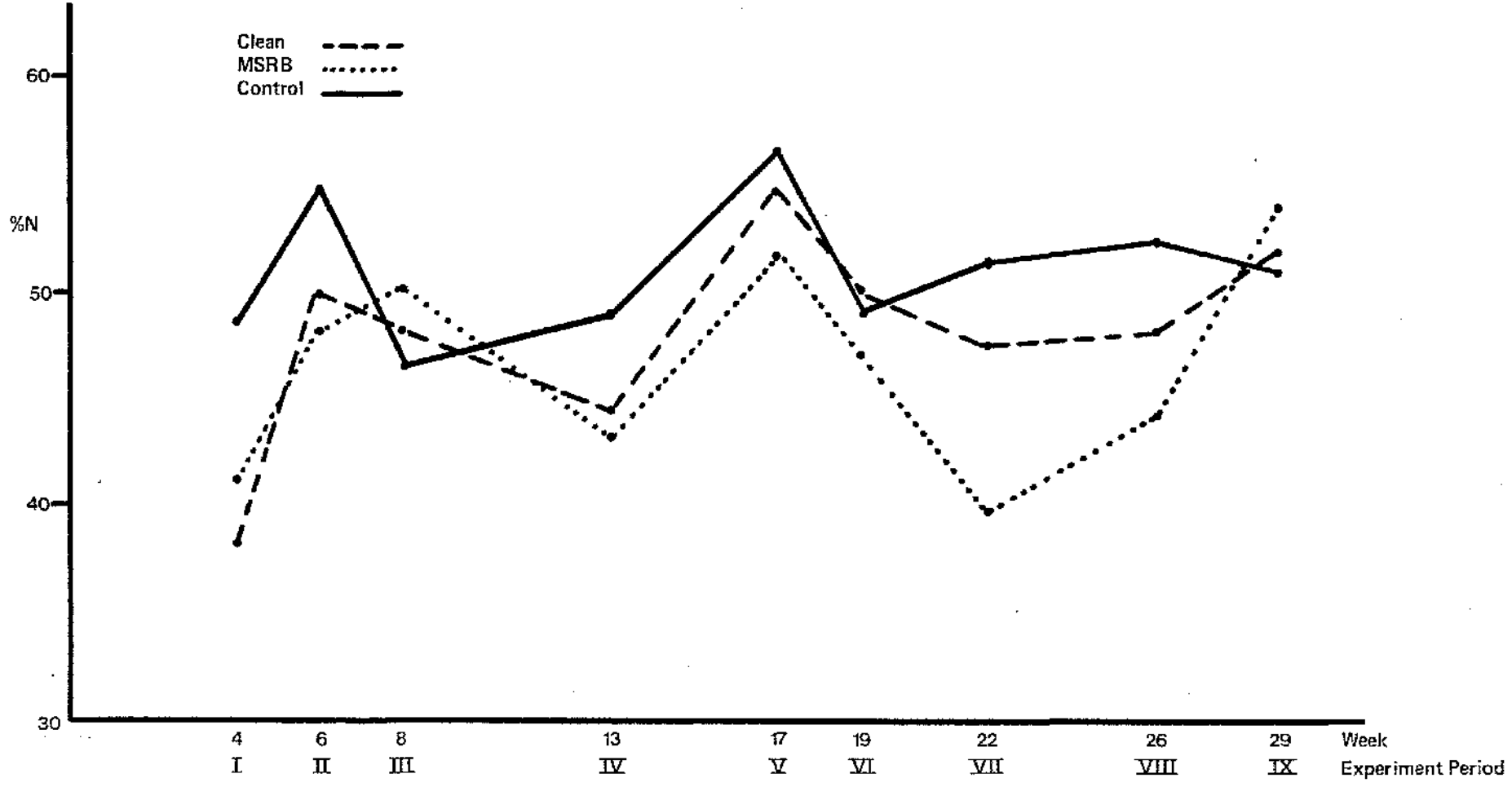


Fig. B6

URINE NITROGEN OUTPUT (% OF INPUT)

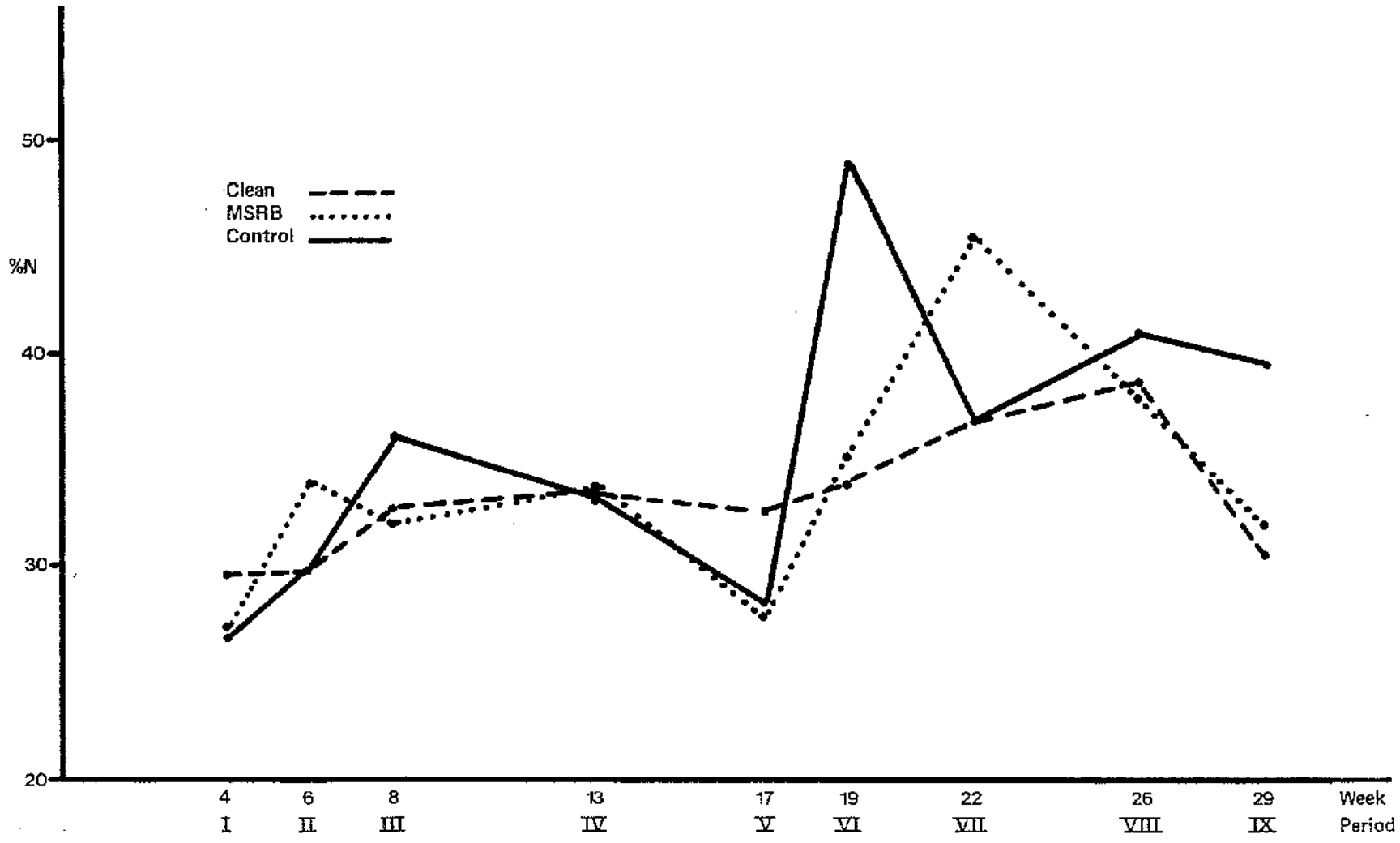


Fig. B7

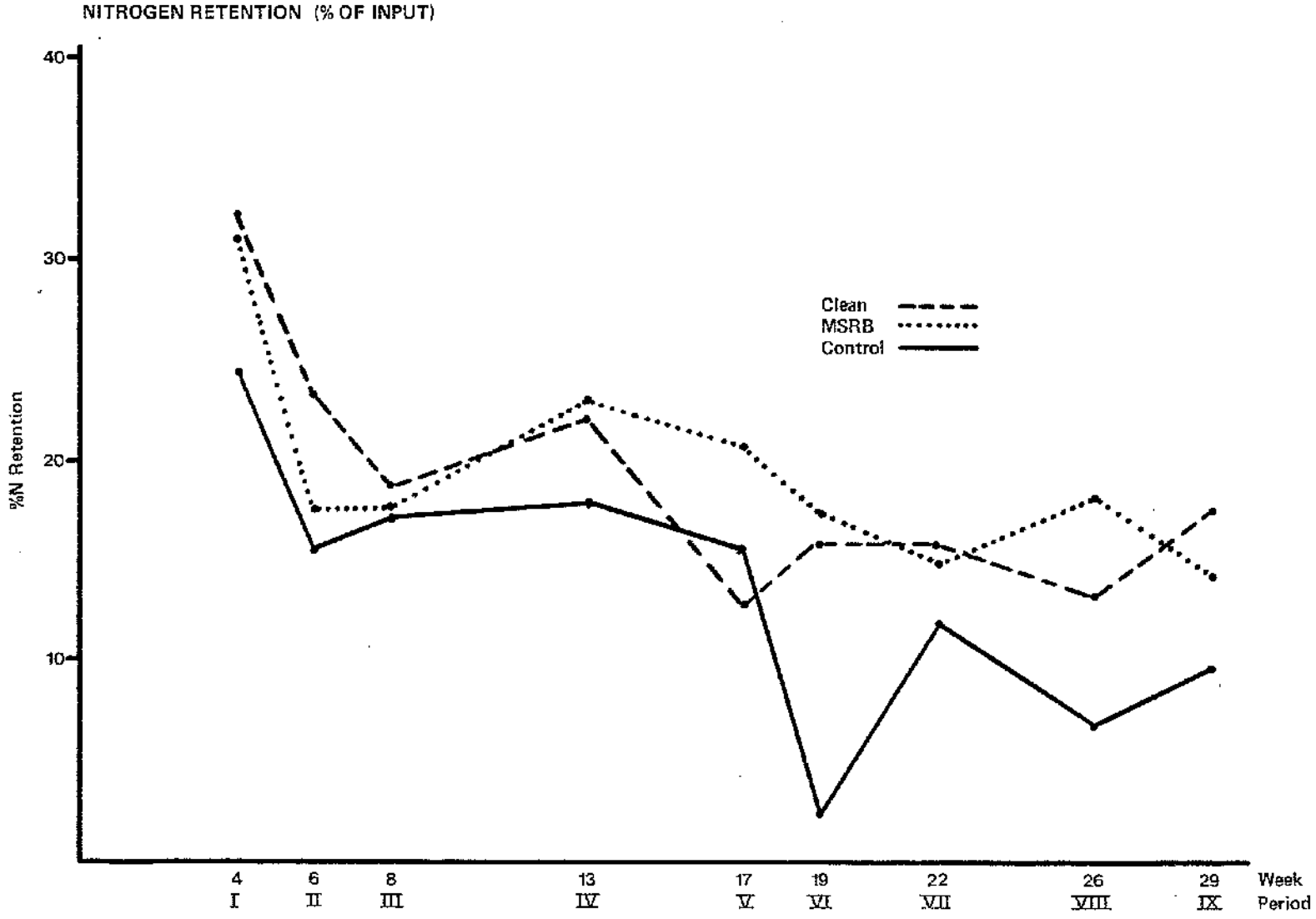

NITROGEN RETENTION (% OF INPUT)
40
30
20
10
%N Retention
Clean
MSRB
Control
4
I
6
II
8
III
13
IV
17
V
19
VI
22
VII
26
VIII
29
IX
Week
Period

Fig. B8

NITROGEN BALANCE DATA

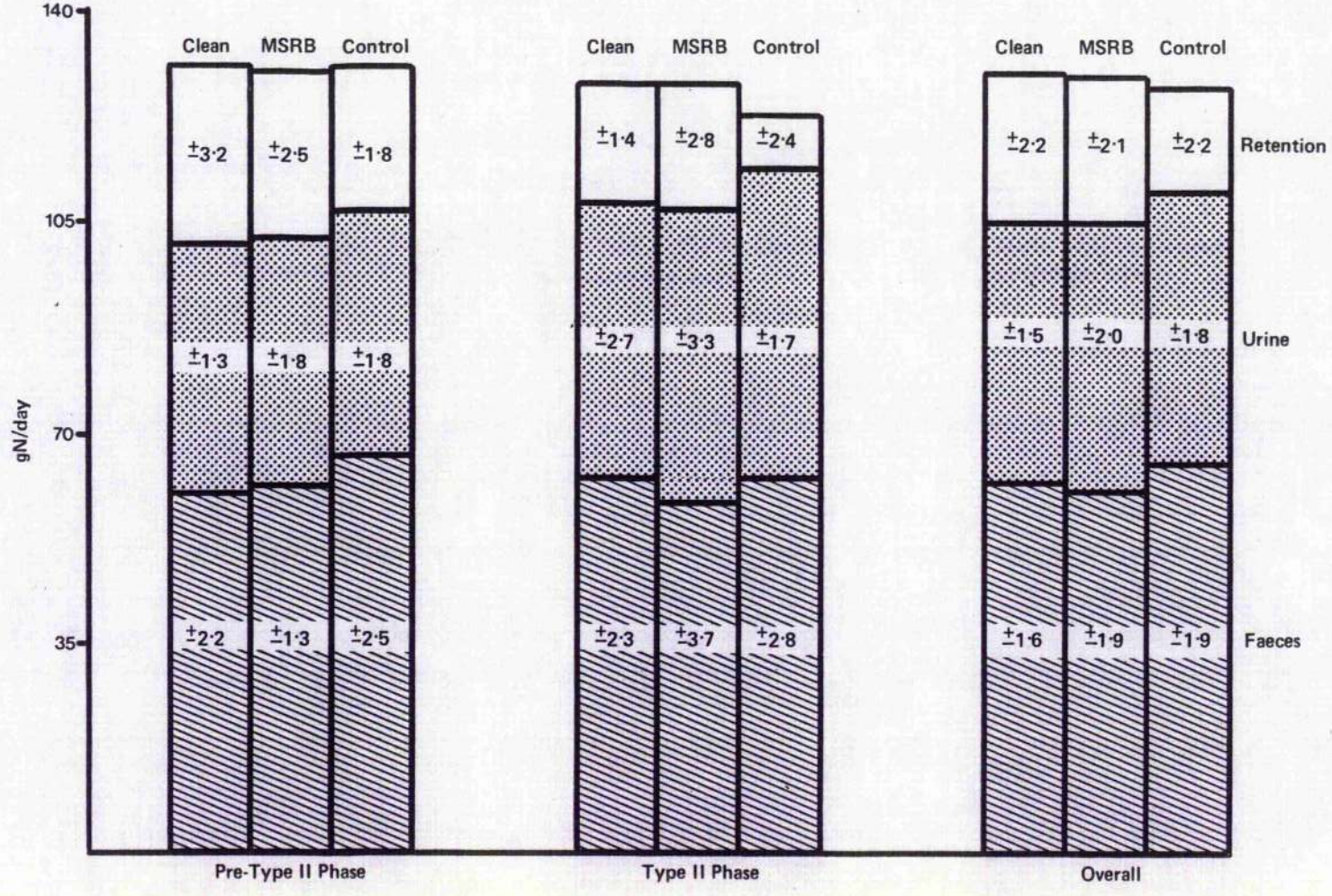


Table B2. Mean liveweight kg (±SE) of Clean, MSRB treated and Control steers during the winter period following the first grazing season

Week	Clean	MSRB	Control
0	264 ± 9.7	251 ± 6.1	230 ± 4.8
2	273 ± 9.8	255 ± 5.6	240 ± 5.4
4	267 ± 9.9	256 ± 6.3	234 ± 4.5
6	283 ± 9.3	269 ± 5.2	247 ± 5.4
8	296 ± 10.3	277 ± 6.4	254 ± 5.3
10	313 ± 9.9	296 ± 6.3	275 ± 5.6
13	313 ± 9.8	300 ± 6.5	276 ± 6.9
15	326 ± 10.1	310 ± 6.9	288 ± 6.5
17	332 ± 9.7	320 ± 6.8	297 ± 7.2
19	343 ± 10.3	321 ± 6.7	307 ± 8.8
20	344 ± 9.8	330 ± 8.9	310 ± 7.9
22	352 ± 8.8	343 ± 7.2	318 ± 8.9
24	357 ± 9.3	346 ± 8.1	319 ± 7.8
26	352 ± 8.1	347 ± 7.0	317 ± 8.6
28	359 ± 7.7	345 ± 7.8	321 ± 10.0
29	360 ± 7.9	345 ± 8.7	318 ± 10.2
Total Gain	96 kg	94 kg	88 kg

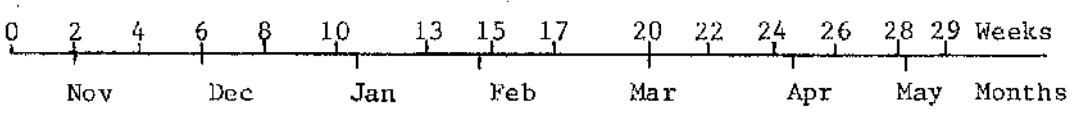


Table B3. Mean trichostrongyle egg count in faeces of Clean, MSRB treated and Control steers during the winter period following the first grazing season

Week	Clean	MSRB	Control
0	0	70	426
2	0	25	240
4	0	25	205
6	0	35	60
8	0	10	55
11	0	15	50
13	0	0	50
15	0	35	45
17	7	50	90
19	0	35	55
20	7	60	65
22	0	45	50
24	0	50	95
26	0	83	140
27	7	55	85
29	0	50	70

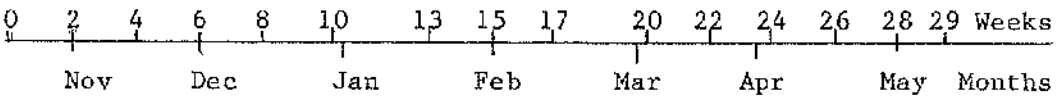


Table B5. Mean plasma pepsinogen levels (I.u. tyrosine) of Clean, MSRB treated and Control steers during the winter period following the first grazing season

Week	GROUP		
	Clean (7)	MSRB (10)	Control (10)
0	1.1 ± 0.04	1.6 ± 0.16	6.3 ± 0.52
2	0.9 ± 0.05	1.3 ± 0.11	5.1 ± 0.56
4	0.9 ± 0.04	1.2 ± 0.11	3.8 ± 0.55
6	0.8 ± 0.07	1.2 ± 0.17	2.6 ± 0.37
8	0.9 ± 0.04	1.2 ± 0.21	2.2 ± 0.26
10	0.9 ± 0.04	1.1 ± 0.11	2.6 ± 0.27
13	0.8 ± 0.04	1.2 ± 0.08	2.5 ± 0.25
15	1.0 ± 0.05	1.3 ± 0.11	2.9 ± 0.30
17	0.9 ± 0.06	1.4 ± 0.13	3.6 ± 0.26
19	0.9 ± 0.04	1.4 ± 0.12	4.2 ± 0.43
21	0.9 ± 0.04	1.4 ± 0.14	4.2 ± 0.40
22	0.9 ± 0.04	1.6 ± 0.23	4.9 ± 0.42
24	0.9 ± 0.03	1.6 ± 0.16	5.8 ± 0.49
26	0.9 ± 0.04	1.5 ± 0.15	6.1 ± 0.59
27	0.8 ± 0.05	1.5 ± 0.13	6.3 ± 0.60
29	0.9 ± 0.04	1.6 ± 0.17	7.0 ± 0.60

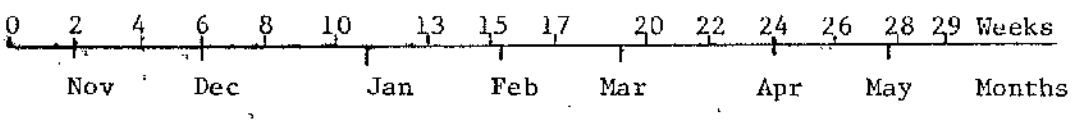


Table B6. Mean plasma gastrin (pg/ml) concentrations (±SE) determined in Clean, MSRB treated and Control cattle during the winter housing period

Week	Date	Clean	MSRB Treated	Control
0	27.10.82	121 ±13.3^{b}	123 ±25.3 ^{b}	590 ±112.4 ^{A}
8	22.12.82	215 ± 7.7	295 ±28.1	304 ± 68.1
13	26. 1.83	193 ± 8.7	224 ±17.9	292 ± 78.3
17	24. 2.83	252 ±16.9^{b}	320 ±48.1	446 ± 83.4 ^{a}
24	13. 4.83	211 ±21.3 ^{b}	286 ±40.3 ^{a,b}	464 ±108.5 ^{a}
27	4. 5.83	180 ± 5.8	213 ±24.2	784 ±363.8
29	18. 5.83	214 ±21.8^{b}	284 ±27.8	573 ± 98.1 ^{A}

Statistics: a,b $P<0.05$ A,b $P<0.01$

Table B7. Blood biochemical analyses for Clean, MSRB treated and Control cattle during the winter period following the first grazing season

Week	-2	4	15	17	24	29
Urea mmol/l						
Clean	2.4±0.4^{b}	5.1±0.2	2.6±0.2	3.2±0.1^{b}	4.0±0.1^{b}	3.2±0.4
MSRB	5.8±0.2A	5.4±0.3	3.0±0.1	4.4±0.2A	4.9±0.2a	3.6±0.3
Control	5.9±0.3 A	5.3±0.2	2.8±0.2	3.4±0.1^{b}	3.9±0.2^{b}	2.6±0.4
Inorganic P mmol/l						
Clean	2.6±0.2	2.8±0.1	2.6±0.1	2.7±0.1	2.6±0.1	2.3±0.1
MSRB	2.4±0.1	2.7±0.1	2.7±0.1	2.9±0.1	2.8±0.1	2.4±0.1
Control	2.3±0.1	2.9±0.1	2.8±0.1	2.9±0.1	2.8±0.1	2.6±0.1
Total Protein g/l						
Clean	66.8±1.8^{b}	69.4±1.1	70.8±2.5	67.6±1.5	68.6±2.0	70.0±1.0^{a}
MSRB	73.6±1.6^{a}	66.0±1.2	67.2±0.9	65.2±1.4	64.7±1.2	66.6±1.0^{b}
Control	71.3±1.9^{a,b}	65.2±2.1	66.6±2.6	66.6±1.4	66.2±1.6	68.2±1.9^{a}
Albumin g/l						
Clean	30.8±1.3^{b}	34.7±1.1^{A}	39.0±1.2^{A}	37.0±0.7	37.1±0.8^{A}	36.3±0.6 A
MSRB	35.6±0.7A	34.0±0.1A	36.6±0.6A	35.1±0.7	35.4±0.6^{a,b}	34.3±0.6 A
Control	28.7±1.1^{b}	27.4±1.2^{b}	32.7±0.8^{b}	32.2±0.9	33.4±0.8^{b}	29.6±1.0^{b}

a,b, P<0.05

A,b, P<0.001

Table B8. Digestibility coefficient of the whole diet given to individual animals in the Clean, MSRB treated and Control groups during the winter housing period following the first season at grass

Clean Group

Pd.	Week	No.	DM	CP	CF	EE	Ash	OM	GE
I	6	G30	0.59	0.64	0.54	0.75	0.35	0.62	0.59
	7	66	0.59	0.61	0.55	0.81	0.38	0.61	0.60
II	8	88	0.56	0.49	0.48	0.63	0.19	0.44	0.58
		73	0.55	0.51	0.44	-	0.19	0.43	0.57
III	10	74	0.56	0.52	0.48	0.84	0.25	0.60	0.58
		9	0.57	0.52	0.49	0.78	0.29	0.61	0.58
IV	15	G30	0.56	0.55	0.56	0.65	0.11	0.61	0.59
		66	0.56	0.56	0.51	0.68	0.28	0.59	0.58
V	19	88	0.59	0.45	0.50	0.63	0.34	0.62	0.59
Pre-type II									
Mean			0.57	0.54	0.51	0.72	0.24	0.57	0.58
±SE			0.005	0.019	0.012	0.076	0.028	0.024	0.003
VI	21	74	0.56	0.52	0.52	0.77	0.13	0.61	0.58
		9	0.56	0.49	0.50	0.71	0.13	0.60	0.58
VII	24	G30	0.60	0.50	0.49	0.71	0.40	0.62	0.58
		66	0.58	0.53	0.48	0.74	0.38	0.60	0.57
VIII	28	88	0.60	0.52	0.49	0.58	0.33	0.63	0.60
		73	0.53	0.49	0.39	0.67	0.18	0.57	0.55
IX	31	74	0.60	0.44	0.53	0.68	0.32	0.62	0.59
		9	0.67	0.53	0.58	0.66	0.43	0.69	0.67
Type II									
Mean			0.59	0.50	0.50	0.69	0.29	0.62	0.59
±SE			0.014	0.010	0.018	0.019	0.041	0.012	0.011

MSRB Treated Group

Pd.	Week	No.	DM	CP	CF	EE	Ash	OM	GE
I	6	76	0.58	0.59	0.53	0.75	0.37	0.60	0.59
II	8	75	0.58	0.53	0.51	0.69	0.27	0.46	0.60
		384	0.56	0.50	0.48	0.74	0.16	0.45	0.58
III	10	67	0.57	0.49	0.48	0.79	0.30	0.60	0.59
		91	0.58	0.51	0.50	0.72	0.26	0.61	0.59
IV	15	76	0.58	0.58	0.54	0.69	0.30	0.60	0.59
		86	0.58	0.56	0.55	0.63	0.23	0.62	0.60
V	19	75	0.59	0.50	0.51	0.70	0.29	0.62	0.59
		384	0.56	0.47	0.45	0.67	0.22	0.60	0.57
Pre-type II									
Mean			0.58	0.53	0.51	0.70	0.27	0.58	0.59
±SE			0.003	0.014	0.010	0.014	0.019	0.022	0.003
VI	21	67	0.60	0.56	0.55	0.72	0.24	0.64	0.62
		91	0.58	0.49	0.54	0.72	0.18	0.62	0.59
VII	24	76	0.60	0.53	0.50	0.71	0.37	0.63	0.60
		86	0.66	0.68	0.57	0.78	0.48	0.68	0.65
VIII	28	75	0.64	0.60	0.52	0.73	0.37	0.68	0.64
		384	0.61	0.51	0.52	0.58	0.31	0.64	0.60
IX	31	67	0.63	0.49	0.55	0.63	0.38	0.66	0.64
		91	0.61	0.43	0.54	0.56	0.33	0.64	0.62
Type II									
Mean			0.62	0.54	0.54	0.68	0.33	0.65	0.62
±SE			0.009	0.026	0.007	0.026	0.031	0.007	0.007

Control Group

Pd.	Week	No.	DM	CP	CF	EE	Ash	OM	GE
I	6	8	0.50	0.45	0.48	0.88	0.29	0.52	0.52
		90	0.58	0.58	0.51	0.78	0.43	0.59	0.59
II	8	65	0.53	0.45	0.45	0.62	0.16	0.40	0.55
		7	0.51	0.46	0.40	0.64	0.18	0.36	0.52
III	10	79	0.56	0.51	0.47	0.75	0.28	0.59	0.57
		92	0.61	0.56	0.55	0.79	0.27	0.65	0.63
IV	15	8	0.55	0.53	0.52	0.60	0.26	0.58	0.56
		90	0.53	0.50	0.52	0.33	0.22	0.57	0.55
V	19	65	0.55	0.47	0.46	0.62	0.22	0.59	0.55
		7	0.50	0.41	0.36	0.63	0.17	0.54	0.51
Pre-type II Mean			0.54	0.49	0.47	0.66	0.25	0.54	0.56
±SE			0.011	0.017	0.018	0.045	0.023	0.027	0.010
VI	21	79	0.56	0.50	0.51	0.74	0.11	0.61	0.58
		92	0.58	0.52	0.50	0.70	0.19	0.63	0.61
VII	24	8	0.56	0.48	0.44	0.77	0.31	0.59	0.57
		90	0.60	0.56	0.52	0.69	0.34	0.63	0.60
VIII	28	65	0.59	0.49	0.54	0.56	0.19	0.63	0.59
		7	0.53	0.43	0.40	0.48	0.21	0.57	0.53
IX	31	79	0.62	0.47	0.56	0.61	0.35	0.65	0.61
		92	0.64	0.51	0.56	0.54	0.40	0.67	0.64
Type II Mean			0.59	0.50	0.50	0.64	0.26	0.62	0.59
±SE			0.012	0.012	0.019	0.034	0.055	0.011	0.011

Table B9. Mean apparent digestibility of the whole diet given to Clean, MSRB treated and Control cattle during the winter (Pre-type II and type II) following the first grazing season

Feed Fraction	Clean	MSRB	Control
Pre-type II Weeks 0-17 Oct-Feb	(n=9)	(n=9)	(n=10)
DM	0.57 ± 0.005[a]	0.57 ± 0.003[a]	0.54 ± 0.011[b]
CP	0.54 ± 0.019	0.52 ± 0.014	0.49 ± 0.017
CF	0.51 ± 0.012	0.51 ± 0.010	0.47 ± 0.018
EE	0.72 ± 0.076	0.71 ± 0.014	0.66 ± 0.045
Ash	0.26 ± 0.028	0.27 ± 0.019	0.25 ± 0.023
OM	0.57 ± 0.024	0.57 ± 0.022	0.54 ± 0.027
Energy	0.58 ± 0.003[a]	0.59 ± 0.003[a]	0.55 ± 0.010[b]
Type II Weeks 17-29 Feb-May	(n=8)	(n=8)	(n=8)
DM	0.58 ± 0.014	0.62 ± 0.009	0.59 ± 0.012
CP	0.50 ± 0.010	0.54 ± 0.026	0.50 ± 0.012
CF	0.50 ± 0.018	0.54 ± 0.007	0.50 ± 0.019
EE	0.69 ± 0.019	0.68 ± 0.026	0.64 ± 0.034
Ash	0.29 ± 0.041	0.33 ± 0.031	0.26 ± 0.033
OM	0.62 ± 0.012[b]	0.65 ± 0.007[a]	0.62 ± 0.011[ab]
Energy	0.59 ± 0.011[b]	0.62 ± 0.007[a]	0.59 ± 0.011[b]
Total	(n=17)	(n=17)	(n=18)
DM	0.58 ± 0.007[ab]	0.59 ± 0.006[a]	0.56 ± 0.010[b]
CP	0.52 ± 0.012[ab]	0.53 ± 0.014[a]	0.49 ± 0.011[b]
CF	0.50 ± 0.011	0.52 ± 0.017	0.48 ± 0.014
EE	0.71 ± 0.017	0.69 ± 0.015	0.65 ± 0.029
Ash	0.27 ± 0.024	0.30 ± 0.019	0.25 ± 0.019
OM	0.59 ± 0.015	0.61 ± 0.015	0.58 ± 0.019
Energy	0.59 ± 0.005[a]	0.61 ± 0.008[A]	0.57 ± 0.009[b]

Table B10 Individual Animal Nitrogen Balance Data (gN/d) together with Mean Values (±SE) for Experimental Periods I to IX

Period	Group	No.	Intake	g N/d Faeces	Urine	Retained
I (Week 4)	Clean	G30	135.3	48.2	43.9	43.2
		96	135.3	53.3	36.5	45.5
		Mean	135.3	50.7	40.2	44.3
		±SE	0	1.8	2.6	0.8
	MSRB	76	135.3	55.5	36.5	43.3
	Control	8	135.3	74.6	28.8	31.9
		90	135.3	56.3	43.5	35.5
		Mean	135.3	65.4	36.2	33.7
		±SE	0	6.5	5.2	1.3
II (Week 6)	Clean	88	129.3	66.4	42.7	20.2
		73	129.3	62.5	36.2	30.5
		Mean	129.3	64.6	39.5	25.4
		±SE	0	1.3	2.3	3.7
	MSRB	75	129.3	60.3	48.8	20.2
		34	129.3	64.5	39.6	25.2
		Mean	129.3	62.4	44.2	22.7
		±SE	0	1.5	3.2	1.8
	Control	7	129.3	70.2	36.7	22.4
		99	129.3	71.2	40.2	17.9
		Mean	129.3	70.7	38.4	20.1
		±SE	0	0.4	1.2	1.6
III (Week 8)	Clean	9	118.6	57.4	39.3	21.9
		74	118.6	57.2	38.7	22.7
		Mean	118.6	57.3	39.0	22.3
		±SE	0	0.1	0.2	0.3
	MSRB	91	118.6	58.2	39.0	21.4
		67	118.6	60.9	37.2	20.5
		Mean	118.6	59.6	38.1	20.9
		±SE	0	1.0	0.6	0.3
	Control	92	118.6	51.8	46.1	20.7
		79	118.6	58.6	39.8	20.2
		Mean	118.6	55.2	42.9	20.4
		±SE	0	2.4	2.2	0.2

Period	Group	No.	Intake	Faeces	g N/d Urine	Retained
IV (Week 13)	Clean	G30	141.2	63.6	46.6	31.0
		96	141.2	61.6	48.1	31.5
		Mean	141.2	62.6	47.3	31.2
		±SE	0	0.7	0.5	0.2
	MSRB	76	141.2	59.6	44.7	36.9
		86	141.2	62.1	51.0	28.1
		Mean	141.2	60.8	47.8	32.5
		±SE	0	0.9	2.2	3.1
	Control	90	141.2	70.6	48.6	22.0
		8	141.2	67.1	45.4	28.7
		Mean	141.2	68.8	47.0	25.3
		±SE	0	1.2	1.1	2.4
V (Week 17)	Clean	88	129.5	71.1	42.5	15.9
	MSRB	384	129.5	69.3	35.1	25.1
		75	129.5	64.9	36.0	28.6
		Mean	129.5	67.1	35.6	26.8
		±SE	0	1.6	0.3	1.2
	Control	7	129.5	76.7	34.0	18.9
		99	129.5	69.3	38.8	21.5
		Mean	129.5	73.0	36.4	20.2
		±SE	0	2.6	1.7	0.9
VI (Week 19)	Clean	74	138.3	67.0	54.9	16.4
		9	138.3	70.8	39.4	28.1
		Mean	138.3	68.9	47.1	22.2
		±SE	0	1.3	5.5	4.1
	MSRB	91	138.3	69.8	57.1	11.4
		67	138.3	60.4	41.1	36.8
		Mean	138.3	65.1	49.1	24.1
		±SE	0	3.3	5.7	9.0
	Control	92	97.3	46.8	55.1	-4.5
		79	138.3	68.8	60.1	9.4
		Mean	117.8	57.8	57.6	2.5
		±SE	14.5	7.8	1.8	4.9

Period	Group	No.	Intake	Faeces	g N/d Urine	Retained
VII (Week 22)	Clean	96	131.5	61.7	46.6	23.2
		G30	131.5	62.9	50.1	18.5
		Mean	131.5	62.2	48.3	20.8
		±SE	0	0.4	1.2	1.7
	MSRB	76	131.5	62.3	57.1	12.1
		86	131.5	41.8	62.7	27.0
		Mean	131.5	52.0	59.9	19.6
		±SE	0	7.3	2.0	5.3
	Control	8	131.5	68.2	49.8	13.5
		90	131.5	66.7	46.9	17.9
		Mean	131.5	67.4	48.3	15.7
		±SE	0	0.5	1.0	1.6
VIII (Week 26)	Clean	88	134.3	63.9	56.1	14.3
		73	118.5	57.5	41.9	19.1
		Mean	126.4	60.7	49.0	16.7
		±SE	5.6	2.3	5.0	1.7
	MSRB	384	134.3	66.3	45.8	22.2
		75	118.5	44.9	50.1	23.5
		Mean	126.4	55.6	47.9	22.8
		±SE	5.6	7.6	1.5	0.5
	Control	99	134.3	67.9	53.3	13.1
		7	118.5	63.9	50.4	4.2
		Mean	126.4	65.9	51.8	8.6
		±SE	5.6	1.4	1.0	3.2
IX (Week 29)	Clean	74	125.5	70.1	34.9	20.5
		9	105.5	49.6	35.8	20.1
		Mean	115.5	59.8	35.3	20.3
		±SE	7.1	7.3	0.3	0.1
	MSRB	91	125.5	71.7	36.8	17.0
		67	105.5	53.3	36.6	15.6
		Mean	115.5	62.5	36.7	16.3
		±SE	7.1	6.5	0.1	0.5
	Control	79	125.5	65.9	44.3	15.3
		92	105.5	51.5	47.3	6.7
		Mean	115.5	58.7	45.8	11.0
		±SE	7.1	5.1	1.1	3.0

Table B11. Mean Nitrogen Output and Retention expressed as a percentage of N input in Clean, MSRB treated and Control calves during the winter housing period

Period	Group	% of Input N		
		Faecal N	Urine N	Retained N
I	Clean	37.5	29.7	32.8
(Week 4)	MSRB	41.0	27.0	32.0
	Control	48.4	26.7	24.9
II	Clean	49.9	30.5	23.5
(Week 6)	MSRB	48.2	34.2	17.6
	Control	54.7	29.7	15.6
III	Clean	48.3	32.9	18.8
(Week 8)	MSRB	50.2	32.1	17.7
	Control	46.5	36.2	17.2
IV	Clean	44.3	33.6	22.1
(Week 13)	MSRB	43.1	33.9	23.0
	Control	48.8	33.3	17.9
V	Clean	54.9	32.8	12.7
(Week 17)	MSRB	51.8	27.5	20.7
	Control	56.4	28.1	15.5
VI	Clean	49.8	34.1	16.1
(Week 19)	MSRB	47.1	35.5	17.4
	Control	49.0	48.9	2.1
VII	Clean	47.4	36.8	15.8
(Week 22)	MSRB	39.6	45.6	14.8
	Control	51.3	36.8	11.9
VIII	Clean	48.0	38.8	13.2
(Week 26)	MSRB	44.0	37.9	18.1
	Control	52.2	41.0	6.8
IX	Clean	51.8	30.6	17.6
(Week 29)	MSRB	54.1	31.8	14.1
	Control	50.8	39.7	9.5

Table B12. Mean daily nitrogen balance data (±SE) for Clean, MSRB treated and Control cattle during the winter housing following the first grazing season in the pre-type II, type II and overall experimental periods.

	Group	No.	Input	Faeces		Urine		Retained	
				g	%	g	%	g	%
Pre-type II Periods I-V inclusive	Clean	9	130.9 ± 2.6	60.2 ± 2.2	46.1 ± 1.9	41.5 ± 1.3	31.8 ± 0.8	29.2 ± 3.2	22.1 ± 2.2
	MSRB	9	130.3 ± 2.6	61.7 ± 1.3	47.5 ± 1.3	40.5 ± 1.8	31.2 ± 1.1	28.1 ± 2.5	21.1 ± 1.6
	Control	10	130.8 ± 2.4	66.6 ± 2.5	50.9 ± 1.7	40.2 ± 1.8	30.9 ± 1.6	24.2 ± 1.8	18.2 ± 1.3
Type II Periods VI-IX inclusive	Clean	8	127.9 ± 3.7	62.9 ± 2.3	49.2 ± 1.0	44.9 ± 2.7	35.1 ± 1.6[a]	20.1 ± 1.4	15.7 ± 1.1[a]
	MSRB	8	127.9 ± 3.7	58.8 ± 3.7	46.0 ± 2.6	48.4 ± 3.3	37.8 ± 2.2[a,b]	20.7 ± 2.8	16.1 ± 2.0[a]
	Control	8	122.8 ± 4.8	62.5 ± 2.8	50.8 ± 0.6	50.9 ± 1.7	42.0 ± 2.3[b]	9.4 ± 2.4	7.2 ± 1.9[b]
Total All Periods I-IX inclusive	Clean	17	129.5 ± 2.3	61.5 ± 1.6	47.5 ± 1.2[a]	43.1 ± 1.5	33.3 ± 1.0	24.9 ± 2.2	19.1 ± 1.5[a]
	MSRB	17	129.2 ± 2.3	60.3 ± 1.9	46.8 ± 1.4[a]	44.4 ± 2.0	34.3 ± 1.5	24.5 ± 2.1	18.8 ± 1.4[a]
	Control	18	127.2 ± 2.7	64.8 ± 1.9	50.9 ± 1.0[b]	44.9 ± 1.8	35.8 ± 1.8	17.5 ± 2.2	13.3 ± 1.7[b]

SECTION C: PERFORMANCE AND CARCASS STUDIES ON GROUPS OF CATTLE DURING THE SECOND GRAZING SEASON AND SUBSEQUENT FINAL HOUSED FATTENING PERIOD.

INTRODUCTION

The significant reduction in the accumulation of trichostrongyle L_3 on the pasture during the second half of the summer due to the previous administration of a morantel sustained release bolus to first season calves at spring turnout and, as a consequence, the better performance of these animals compared to untreated control calves has already been documented (Armour et al, 1981; Jacobs et al, 1981; Jones, 1981; Tharaldsen and Helle, 1981; Tornquist et al, 1981; Tornquist and Tolling, 1983; Borgsteede, 1983; Raynaud et al, 1983).

In the first grazing season of the current experiment (see Section A) a significant increase in liveweight of 20 kg in MSRB treated animals over the controls was recorded, despite the fact that parasitic bronchitis occurred in both groups necessitating treatment with diethylcarbamazine, and that an unscheduled treatment of levamisole was given to the control group to combat severe symptoms of type I ostertagiasis during the second part of the summer.

Following the post-mortem of 11 calves in October some carcass measurements were taken in order to further compare the groups. It was shown that the killing-out percentage was better in the MSRB treated animals (49.5%) than in the control group (46.9%). The mean weight of the indicator 7-10th forerib of the control group was 1990 g compared with the mean of the MSRB treated group of 2575 g, (but this difference was not significant owing to a high standard error), the lean muscle of the forerib in the MSRB treated animals showed a significantly higher weight (1621 g) than the controls (1190 g), with no significant differences being found in the weight of bone and fat. The dry matter content of the eye muscle of the control group was significantly less than that of the MSRB treated group (274 compared with 301 gDM/kg respectively).

During the second part of the winter period (Section B of this thesis) severe type II ostertagiasis occurred in some of the control calves. Digestibility and nitrogen balance studies were performed on these calves and also on a clean group which had been kept free of parasites with fortnightly treatments of fenbendazole during the grazing season. The MSRB treated group showed significantly better digestibilities of dry matter (DM), crude protein (CP) and energy, and the nitrogen retention was also greater than that of the control group.

However, most late winter or spring-born beef or dairy/beef cattle require a second grazing season before they are sold or sent to slaughter at an appropriate weight. All replacement heifers need a second grazing season before entering the milking herd, but little information is available on the epidemiology, incidence and economic importance of parasite infection during the second grazing season.

Epidemiological observations on ostertagiasis in cattle over two grazing seasons in Scotland were conducted by Armour et al (1979). They found that first year animals showed clinical ostertagiasis towards the end of the grazing season (September), due to the high levels of infective larvae which had accumulated on the pasture. This outbreak resulted in the classical features of high trichostrongyle faecal egg counts, high O. ostertagi worm burdens and elevated plasma pepsinogens. By the following May, at the beginning of the second grazing season, very low numbers of larvae were recovered from the paddocks because of an almost complete disappearance of the overwintered L_3 larvae from the pasture followed by the the appearance of moderately high numbers of new populations of L_3 (up to 9000 L_3 per kg) by September. This increase was reflected by negligible faecal egg counts, low worm burdens and only a moderate elevation of plasma pepsinogen in the second year animals. In the opinion of the authors young cattle acquire a good immunity to O. ostertagi after one season at grass, but small infections which established in the early part of the second season are capable of contaminating the pasture to levels which could be dangerous to susceptible young stock. They also proposed that the moderately elevated plasma pepsinogen levels recorded in the second year animals could be explained by an allergic reaction in the gastric mucosa.

Some conflicting results have been reported on the benefits of anthelmintic treatments for animals in the second grazing system. In Austria, Prosl, Supperer, Jones, Lockwood and Bliss (1983) reported on the efficacy of the MSRB in controlling gastrointestinal parasitism during two consecutive grazing seasons. In the first grazing period the treated animals were 34.8 kg ($P < .001$) heavier than the control but no further beneficial effect in weight gain was observed during the second season following a second bolus treatment. However, Conder, Jones and Bliss (1983) also reporting on the effect of the MSRB in second-season grazing cattle from seven farms in south east England, found that although there was hardly any measurable reduction in faecal

egg counts and herbage larval counts between treated and control animals, there was a significant overall 15.1 kg liveweight gain over the controls, and the latter showed no compensatory growth in the subsequent housing period. However, in Germany, Guldenhaupt and Burger (1983) found a non-significant production advantage of the use of MSRB in second season cattle. However, in all these studies individual parameters of, at the most, three animals were assessed and there has been no attempt to integrate fully the parasitological, biochemical, pathological, production and clinical findings with a carcass evaluation at marketing. This section reports on these parameters together with a carcass evaluation at slaughter.

MATERIALS AND METHODS

Experimental Design

The animals were grazed for 22 weeks (19th May to 17th October, 1983) and were then housed for a final fattening period prior to marketing and slaughter. In order to monitor any effect of climatic variation between years on the epidemiology of trichostrongylosis and to study the possible acquisition of immunity to Ostertagia and Cooperia, a group of four first season helminth-naive calves, aged four to six months, were grazed from 23rd May on a nearby permanent cattle pasture comparable to that used by the second season cattle. This group is designated 1983 naive-controls.

Grazing Management

The experimental cattle were turned out into the grazing plots on 19th May 1983. In order that the nine steers now remaining in each of the control and MSRB treated groups would receive an equivalent larval challenge from overwintered L_3, the plot assignment was altered from that of the previous (1982) season so that control animals grazed plots 3 and 4, (see diagram given on page 42 in Section 1 of this thesis), and the MSRB treated animals grazed plots 2 and 5. Thus, each group alternatively grazed the same paddock as in 1982 and also a paddock grazed by the other group in that year. The six steers remaining in the clean group were again set-stocked in plot number 1 for the whole of the summer as that group had been during the first grazing season. The stocking rate, in terms of head of cattle/ha, was reduced from the previous year to about 4.5/ha in all three groups.

The grazing received an 'early-bite' fertilizer application of 50 kg N/ha and an equivalent dressing following each of the two grazings. The cattle in the MSRB and control groups were each allowed to graze one of their allocated plots until one group had eaten down the pasture. Both groups were then moved into the alternate plot on the same day. Grazing moves were made in each of the months of June, July, August and September. Supplementation with a trough-fed concentrate was necessary from mid-August. The cattle were finally removed from grass on October 18th, 1983.

Final fattening in the second winter housing period

The animals were rehoused, in their own groups, in the previously described accommodation comprising individual self-locking headgates for concentrate feeding and hayracks for group feeding hay. They were bedded on straw. Following housing on October 18th 1983 the cattle were introduced to a ration of 4 kg commercial concentrate together with ad lib access to hay. This was achieved within one week since the cattle had been accustomed to concentrate feeding from the necessary supplementation at grass. After two weeks the commercial concentrate supply was depleted and a careful changeover to mineralised crushed barley was effected. The cattle remained on this finishing diet of hay and barley until slaughter. The crushed barley allowance which was individually given in two equal feeds per day was increased to a total of 5 kg FM/day in late November and to 6 kg in mid-December and remained at that allowance for the remainder of the experiment. The group ad-lib intake of the medium quality hay on offer was measured on a weekly basis.

Selection of animals for marketing, grading and carcass measurements

The original experimental design proposed that cattle would be slaughtered at a constant finish i.e. at the time as and when each individual animal had gained a satisfactory body fat-cover as subjectively assessed by eye. This design would thus permit an assessment of the additional feed and time cost necessary to take the late finishing cattle to market compared with the earlier finishing cattle. However, it became apparent that the extra time that would be needed in order to do this would be excessive and so the design was altered such that the cattle, except for three early-finishing animals which were marketed directly from grass in October, were killed in

December 1983 and January 1984. It was possible to kill only three animals on any one occasion because the time involved in properly undertaking the pathological, parasitological and carcass measurement procedures was considerable. Thus the experimental design associated with animal marketing was altered to that of a modified 'constant-time' slaughter. Animals were selected for slaughter on the basis of a subjective body score assessment and eye appraisal. Guidelines for the body score method are given by an information leaflet published by the Meat and Livestock Commission (1982) 'Selecting Cattle for Slaughter', and essentially comprises the assessor feeling the fat cover development in the animal over the pin bones and around the tailhead and thigh area, and over the transverse processes of the loin, the spring of the ribs and also the chine and the shoulder blade ridge. This was performed when the steers were tethered in the self-locking gates at feeding time. Animals in the clean and MSRB groups all reached an assessed score suitable for marketing in December and January. The control animals were generally in a poorly-finished condition at this time. The experiment was terminated in January when the last control cattle were slaughtered.

Marketing, grading, carcass measurements and necropsy procedures

The 24 animals remaining in the clean, MSRB treated and control groups were slaughtered at a commercial abattoir in Paisley. The cattle were weighed before sending them to the abattoir where they spent overnight in the lairage before slaughter early during the following morning. The necropsy procedures adopted are fully described in the General Materials and Methods Section of this thesis. The hot carcasses were weighed and, at the same time, officially graded by an MLC grader according to the prescribed regulations of the EEC Beef Premium Scheme. Carcass measurements of the side, leg and total lengths, leg circumference and 7th rib width were taken directly using a tape measure.

Following at least five days hanging in the cold room, a commercial 7-10th rib joint was removed from the left side of each of the carcasses and taken for physical dissection measurement and laboratory analysis.

Only three animals could be killed on any one day owing to the restrictions of time and space. The animals were killed in December and January, except for three early finishing steers which were

slaughtered in September and October (numbers 9, 13, 19)

Observations

As in the first grazing season the animals were clinically observed daily and carefully examined at weighing. Faecal, blood and herbage samples were collected at two weekly intervals when liveweight was also recorded.

Parasitological procedures i.e. faecal egg counts, pasture larval counts, post-mortem worm burdens, blood analyses, plasma pepsinogen and gastrin levels were performed using the techniques already described in the General Materials and Methods section. At slaughter the lungs and gastro-intestinal tract were removed and examined as before for pathological changes and abomasal mucosal weight.

RESULTS

GRAZING PERIOD OF THE SECOND YEAR ANIMALS

Clinical observations

During the second grazing season the animals were observed daily at grass and more carefully at the routine two-weekly sampling and weighing time. During the first week of the second grazing season three animals in the control group showed some lack of appetite, in fact one of the animals (number 25) had to be treated with the anthelmintic levamisole because of its very poor condition, diarrhoea and lack of appetite. Interestingly, the same animals had shown severe symptoms of type II ostertagiasis during the last part of the winter. From there onwards, the animals of the different groups were clinically normal.

The climatic data shown generally in Fig. A1 is repeated in Fig. C7 and shows that the weather pattern was similar to that recorded in other years except that in August, the dry conditions led to a shortage of grass and the cattle had to be supplemented with commercial concentrates; thereafter the autumn rains were particularly heavy. In mid August the daily supplement was 3 kg/hd of a commercial beef concentrate, this was increased to 4 kg/hd in mid-September.

Liveweight changes at grass

The mean liveweight of the three groups are shown graphically in Fig. C1 and numerically in Table C1.

At the beginning of the second grazing season the mean liveweights of the clean, MSRB treated and control groups were 353, 350, and 324 kg respectively; the differences already being significantly higher in both clean and MSRB treated animals.

The first animal was slaughtered, in a fat-finished condition, on September 6th 1983 and was a member of the MSRB treated group. At that point the MSRB group was reduced to eight in number. Comparisons of performance with continuous numbers of animals in all groups thus ends after 112 days at grass when the mean liveweight gains were 88, 112, and 92 kg for the clean, MSRB treated and control groups respectively. However grazing, albeit with considerable (4 kg/d) concentrate supplementation, continued until October 17th. The mean liveweight gains for the groups (with the MSRB group now eight in number) after 140 days at grass were 97, 129 and 104 kg for the clean, MSRB treated and control groups respectively. This equates to mean daily liveweight gains of 0.7, 0.92 and 0.74 respectively for the same groups.

Two further animals were selected for marketing on October 6th 1983 (i.e. number 19 from the control group and number 13, again from the MSRB treated group) thus reducing the numbers left at grass after that date to six in the clean group (unchanged), seven in the MSRB group and eight in the control group. In hindsight, this selection for slaughter was premature as both the cattle, although they had an assessed reasonable fat cover, were a little underweight and failed to grade; both being lacking in conformation.

However, taking the mean values of the remaining animals, the liveweight gain over the total 152 days at grass was 105, 131 and 109 kg for the clean, MSRB treated and control groups, corresponding to overall mean daily liveweight gains of 0.7, 0.9 and 0.7 kg respectively. This improved LWG performance of the MSRB treated group of cattle is significantly greater than that of both the clean and control groups at all stages of the grazing season. Most particularly so in the first month at grass when an apparent gain of 34 kg/hd was noted in the MSRB group compared with 30 kg/hd in the clean group, but only 19 kg in the control group. The growth performance of the clean cattle declined markedly as the season progressed, even though there did not appear to be any shortage of grass until later on in the

season, when all three groups had to be supplemented with concentrate because of an acute grass shortage in all plots.

Calculation showed that, ignoring the requirement for concentrate supplementation from mid-August onwards, which amounted to about 200 kg FM per head, (at a current (1983) cash value of about £30 Sterling) the liveweight gain produced from the grazing was 450 kg LW/ha in the clean group, 480 kg/ha in the MSRB treated group and only 403 kg/ha for the control cattle.

Plasma pepsinogen levels

The levels of plasma pepsinogen recorded in all groups during the second season are shown in Table C2 and are graphically displayed in Fig. C2. During the entire season the plasma pepsinogen levels (I.u. tyrosine) of clean animals remained below 1.0 with one exception at week 14 (August) when the value was 1.3. The MSRB treated group showed a mean level of 1.6 at the beginning of the grazing season, thereafter a gradual decrease occurred. There was little variation in those levels from week 8 onwards with slightly higher values than those of the clean group being seen. Highly significantly greater pepsinogen values were recorded from the control group in May with 6.7 being seen. A progressive decrease then occurred throughout the remainder of the experiment to a final value of 2.6 was observed.

Plasma gastrin levels

The mean plasma gastrin levels of the clean, MSRB treated and control groups are shown in Fig. C3 and are also given in Table C3. Plasma samples were evaluated for weeks 0, 2, 6, 12, 16 and 22. From the second week of grazing, at the end of May, the mean values of the MSRB treated and control groups increased while that of the clean animals remained virtually unchanged. At week six the MSRB treated group had a mean value of 473 pg/ml. This increase coincided with an increase of Ostertagia spp. L_3 on the pasture (see Fig. C5). As the larval count on the pasture decreased so also the gastrin levels returned to the previous levels. In week 16, (beginning of September), an increase in gastrin levels was observed in the clean and MSRB treated group but not in the control group; this coincided with the introduction of concentrates due to a shortage of grass towards the end of the grazing season.

Blood biochemical analyses

Results of the blood biochemical analyses from the clean, MSRB treated and control groups performed on five different occasions are presented in Table C4. The concentrations of urea (mmol/l) showed a marked increase in all the groups at the second sampling, probably as a reflection of the high protein concentration of the grass at this time, being significantly higher in the clean (7) than in both the MSRB treated (5.2) and control groups (5.3). Similar results were recorded in week 7. There were no differences in the phosphate levels between the groups which all showed normal values throughout the grazing season. The levels of total protein showed some variation among groups at the different sampling times, but the mean albumin levels in the control animals were consistently lower than those of the MSRB treated and clean groups. While frequently these differences were significant, they always remained within accepted normal values.

Faecal egg counts

Mean trichostrongyle egg counts in the faeces of the three groups are detailed in Table C5 and illustrated in Fig. C4. Throughout the whole grazing season only one positive result was recorded (in week 14) from the clean animals with a mean value of 7 derived from the minimal count of 50 epg in one animal. At the beginning of the season the MSRB treated group had an average of 56 epg followed by a sharp decrease to zero by week 16. Thereafter an increase from a mean of 7 to 29 epg in week 22 was observed. The control group showed the highest egg count at the beginning of the season with a mean of 72 epg being present, this then decreased gradually over the following 8 weeks. Apart from the 6 epg found in week 16 no other positive egg count was recorded in this group over the remainder of the grazing season.

Pasture larval counts

The recoveries of Ostertagia spp. L_3/kg DM from the paddocks grazed by clean, MSRB treated and control animals are illustrated in Fig C5. and detailed in Table C6. No larvae were recovered from the clean group paddock throughout the grazing season. Very low numbers of larvae were found on two occasions, (weeks 4 and 8) during the first part of the second grazing season of the MSRB treated group paddocks. However, from week 12 (August) onwards a gradual increase occurred, reaching a maximum of 353 L_3/kg DM in week 20. In the last sampling recovery was again zero. Similar results were recorded from the

paddocks grazed by control animals during the first part of the second grazing season, with only 236 Ostertagia spp. L_3/kg DM in week 12 with a distinct rise to a maximum of 8396 L_3/kg DM observed in week 18. Thereafter larval recoveries fell to 1698 L_3/kg DM at the end of the season. Cooperia spp. larvae were recovered on only one occasion, namely in the control plot in September.

FINAL FATTENING PERIOD IN SECOND WINTER PERIOD

Liveweight changes

The cattle were receiving an allowance of ad-lib hay and 5 kg of bruised barley (0.81 DM) by November 8th. In mid-December, the barley allowance was further increased to 6 kg/d. The mean liveweight of the three groups of fattening steers are shown in Table C7. As individual cattle attained a satisfactory body score with respect to fat cover they were marketed. This occurred for the clean and MSRB treated cattle through December and January. Most of the control steers were eventually slaughtered in January irrespective of the degree of fat-finish attained in order to follow the reviewed design whereby the cattle were, as reasonably practical, slaughtered at a constant-time.

Table C7 Mean Liveweight of clean, MSRB treated and control steers during the final winter fattening period

Week	Date	Clean (N=6)	MSRB (N=7)	Control (N=8)
0	17.10.83	458	474	430
2	31.10.83	477	487	439
4	14.11.83	491	500	449
6	29.11.83	501	508	465
8	13.12.83	518	519	479
		(N=4)	(N=5)	(N=8)
12	9.01.84	528	528	491

This table has some difficulty associated with its interpretation since clean and MSRB treated animals were removed for slaughter from early December but the control cattle were only taken between January

13th to 26th. However, six weeks, after housing, at the end of November, the mean liveweight gain for the six steers in the clean group was 43 kg compared with 34 kg for the seven remaining MSRB cattle and 35 kg for the eight control animals. The mean liveweights were 501, 508 and 465 kg for each of these groups respectively. At the last date of group mean presentation (January 9th) the mean gains for the four surviving clean steers was 70 kg, with 54 kg gain apparent for the surviving five in the MSRB group compared with 61 kg for the same eight animals of the control group. The results would strongly suggest that the clean cattle, who had shown very poor performance in the second half of the grazing season, made some genuine compensatory growth during the whole of this fattening period. Growth rates for the MSRB treated and control bullocks were comparable although a mean liveweight difference of some 43 kg was apparent at the beginning of December. Overall the control steers had a daily liveweight gain of about 0.8 kg for the period up until January 9th which was regarded as being satisfactory for the estimated total dietary input (MAFF et al. 1975).

Faecal egg counts

Mean trichostrongyle egg counts in the faeces of the three groups are subject to the same decreases in group numbers of animals as has been previously described in the results for liveweight changes. A table of results is not warranted since there were no eggs detected in the clean steers at any time and on only two occasions a mean count of 6 epg was noted for the control bullocks. The MSRB treated cattle however recorded 29 epg on arrival from grazing and had counts of 0, 14, 21, 10 and 30 for each of the weeks 2, 4, 6, 8 and 12.

Plasma pepsinogen levels

Table C8 records the mean group values for plasma pepsinogen activity. These results also should be viewed in the knowledge of the decreasing animal numbers in the clean and MSRB treated groups.

Table C8 Mean plasma pepsinogen (I.u. tyrosine) for clean, MSRB treated and control cattle in final winter fattening period

Week	Clean	MSRB	Control
0	0.7	1.2	2.6
2	0.7	0.8	1.6
4	0.8	0.7	1.6
6	0.8	0.8	1.6
8	0.8	0.8	1.3
12	0.7	0.8	1.1 (N=8)

The interesting decline in plasma pepsinogen values for the control group is a valid data set since none of the animals were removed in that period. Clearly the value (I.u. tyrosine) fell from 2.6 at housing to 1.1 by January. These results will later be discussed in relation to the highly significantly increase in pH, abomasa and abomasal mucosa weights at necropsy in this group of animals. (Table C13) Individual pepsinogen values at slaughter for all the steers are shown in Table C9.

Plasma gastrin levels

The plasma gastrin levels at slaughter are shown in Table C9 and indicate that the values of individual calves in group had returned to pre-infection levels.

RESULTS - 1983 NAIVE CONTROLS

From late September, clinical signs of type I ostertagiasis occurred in three of the four calves, characterised by weight loss, bright green diarrhoea and loss of appetite.

The mean faecal egg counts and plasma pepsinogen levels of the naive 1983 control calves are given in Fig. C6 and the herbage larval counts of trichostrongyle infective larvae are also shown in Fig. C6. The means and, where appropriate, standard errors of the above parameters are shown in Table C10.

The pattern of infection in these 1983 controls was broadly similar to that of the naive-controls in 1982. Thus the mean faecal egg counts increased gradually from June 13th when they were first positive at a mean 12 epg to 137 epg in early September; thereafter a

sharp increase was observed to a mean maximum of 733 epg by the end of October; mean plasma pepsinogen levels which were initially less than 1.0 I.u. increased markedly in late September to reach 7.3 I.u. by early October.

Pasture larval recoveries were low until the end of July and then increased steadily to reach a maximum of 17,931 L_3/kg DM of Ostertagia and 10,345 L_3/kg DM of Cooperia spp. on October 17th. At post-mortem, heavy burdens of both O. ostertagi and C. oncophora were present, the numbers ranging from 41,700 to 225,400 and from 1,400 to 63,800 respectively. Interestingly, the proportion of EL_4 was less than that recorded from the second year animals. Individual worm burdens are shown in Table C11.

POST-MORTEM EXAMINATION

Bolus Recovery

At the slaughter of the nine MSRB treated animals all 18 of the administered slow release devices (i.e. two per steer) were satisfactorily recovered.

Parasitological and pathological findings

Details of the abomasal and small intestinal worm burdens from the individual clean, MSRB treated and control animals are shown in Table C12.

At slaughter, no parasites were found in the gastrointestinal tract of the clean group. Very low O. ostertagi worm recoveries were obtained from both MSRB treated and control groups with only 100 Cooperia spp. being found in one animal of the MSRB treated group.

The mean values shown in Table C12 include data from three animals that were taken for slaughter directly from grass in September and October, some considerable time before the slaughter of the remaining animals. If the results (which show very low worm burdens) from these animals are removed from the groups, the mean total numbers of O. ostertagi in the MSRB treated and control animals becomes 2656 and 2561 respectively, in which 85% were assessed to be inhibited L_4 larvae.

The abomasa and abomasal mucosa weights together with the calculated 'mucosal fraction', pH and liveweight of the steers at slaughter are individually detailed in Table C13. Mean values for each

of the three groups, are also presented. The mean liveweight of the control group at slaughter was significantly lower than the means of both the clean and the MSRB treated groups. The mean fresh weights of the empty abomasum, the abomasal mucosa (and the 'mucosa fraction') together with the pH of the abomasal contents were all significantly greater in the control group cattle compared with the means of both the MSRB treated and clean animals.

Table C14 Mean of linear measurements, hot carcass weight and MLC grade from Clean, MSRB treated and Control animals at slaughter

			Length cm					
Group	Wt.kg	KO%	Leg	Carc	Total	Circum	7th Rib	Grade
Clean	283.2^{a}	53.0	87.3	133.7^{A}	215.8^{a}	114.7^{a}	69.2^{a}	5/6^{A}
MSRB	287.0^{a}	54.5	87.9	133.4^{a}	217.3^{a}	115.4^{A}	70.8^{A}	7/9^{A}
Control	264.0^{b}	53.5	84.6	127.7^{b}	207.4^{b}	108.8^{b}	66.6^{b}	2/9^{b}

Values with different superscripts a,b $P<0.05$ A,b $P<0.01$

Carcass grading

The carcasses were graded on the basis of conformation and fat cover by Meat and Livestock Commission (MLC) officials according to regulations set down by the EEC Common Agriculture Policy (CAP). This is more fully described in the General Materials and Methods Section of this thesis. Individual grading results are given along with the carcass measurements in Table C15. All the failures to grade well were described as being due to 'lack of conformation' (LC). Only two carcasses out of a total of nine presented graded, both at 03, in the control group. In contrast, in the clean group only one out of six carcasses presented was ungraded with the other five all attaining fat scores of 4 and two out of the five receiving conformation grades of 'R'. In the MSRB group of cattle only two out of the nine carcasses presented failed to grade (LC), but five out of the seven which did grade successfully achieved fat scores of 4, three of which also received an 'R' score for conformation. Chi-squared analyses (Minitab, Penn State University 1983 version) showed that both clean and MSRB treated carcass gradings were highly significantly better than the control group results but different one to another.

Carcass measurements

Measurements were taken from the left side of the hot bissected carcasses using a tape measure. Table C15 (in three parts) details the individual animal numbers, date of slaughter, liveweight of animal before slaughter, hot carcass weight, calculated killing-out percentage, and also the carcass, leg and total lengths, the width of the side at the 7th rib, leg circumference and MLC grade result. Mean results and errors for each group of carcasses are also presented. For ease of reference these mean results are also presented in the preceding table, Table C14.

The mean hot carcass weights of the clean and MSRB treated groups (283 and 287 kg) were about 20 kg heavier than the control group (265 kg). These differences were significant. The killing-out percentage (or 'dressing percentage') was highest in the MSRB group (54.5) compared with both the control cattle (53.5) and the clean group (53.0). (This measurement does not account for water loss on chilling which would normally account for a further 5-10 kg of deadweight loss in these carcasses).

As regards the linear measurements of the carcasses, the mean control cattle results for side, leg, total lengths and width at the 7th rib were all significantly lower than both the clean and MSRB treated groups thus reflecting an overall smaller skeletal size in these animals. Additionally, the circumference measurement of the leg was significantly less in the control cattle reflecting a reduced musculature compared with the larger clean and bolus treated cattle.

Rib joint dissection and eye muscle analysis

The 7-10th ribs, inclusively, were removed from the set left side of each carcass following a minimum period in the chill room of five days. The rib bones were trimmed to the normal length that is present in the commercial 'forerib' presentation of this joint as described in the General Materials and Methods Section of this thesis. Mean (±SE) and individual results of the physical dissection and separation of the joint into lean, bone and fat tissue on a weight basis and as a percentage of the total rib weight are presented in Table C16.

The mean weight of the total rib joint was significantly greater in both the clean (5893 g) and the MSRB treated group (5978 g) than in the control group (5196 g). On a weight basis, the total lean tissue and fat contents, but not bone, were significantly greater in both the clean and the MSRB treated groups than in the control animals. When

the results for the lean, bone and fat contents are expressed as a percentage of the total rib weight and the mean values are compared it was seen that statistically, the bone content in the joint of the control group, at 23.8%, was significantly greater than the mean values of 20.9% for the clean group and 21.5% for the MSRB treated steers. However, the lean tissue contents were all very similar at about 61.5% but the increases in the mean % of fat content of 3.5 and 2.1% for the clean and MSRB rib joints respectively just failed to be significant. These results, however, would indicate that there were considerable differences in the total muscle and fat masses between steers which, as a group, had displayed clinical ostertagiasis and the other two groups which had not. The compositional (%) differences of the rib joints indicate the presence of more bone and less fat in the control cattle.

The measurement of the area of the eye muscle of the rib joint was performed by weighing a cut-out section of the muscle from a life-size monochrome photograph and weighing it as described in the General Materials and Methods Section of this thesis. (Other analyses which were performed upon samples of the eye muscle tissue are also described). Table C17 details the individual animal and group mean results for the measurement of eye muscle weight and area, dry matter content, water holding capacity %, protein concentration and total eye muscle protein content. The actual size of the eye muscle, as shown by the area measurements was much smaller in the ribs from the control steers (49.7 sq cm) compared with the other groups (both about 60 sq cm). These differences were highly significant. Also, the mean weight of the control group dissected eye muscle (1076 g) was significantly lower than the mean values for the other two groups (both about 1300 g). The mean dry matter contents of the eye muscles were all similar and were not significantly different. However, the control group mean value was the lowest at 258 g DM/kg compared with 263 and 265 g DM/kg for the clean and MSRB treated groups respectively. The calculated weights of dry muscle had the same order of highly significant difference as the fresh weight of this tissue. The water holding capacity measurements revealed little difference between groups. Control cattle had the lowest mean value of 20.3% (i.e. 203 g 'free' water per kg fresh muscle) compared with identical values of 21.9% for each of the other two groups. Thus, the 'free' water content of the tissue apparently represented about 27% of the total water content of the muscle samples.

The crude protein concentration in the dry matter of the eye muscle tissue (total N x 6.25; g CP/kg DM) was highest in the control group (847) and lowest in the clean group (831) although the differences were not significant. The difference was speculated to be due to the higher content of intramuscular fat in the clean animals. However, total mean CP yield from the eye muscle samples of the control cattle was only 235g which was highly significantly lower than the other groups, both of which had a mean yield of 288 g CP.

Plate 12 shows indicator joints at the 10th rib face from each of a control, MSRBtreated and clean animal.

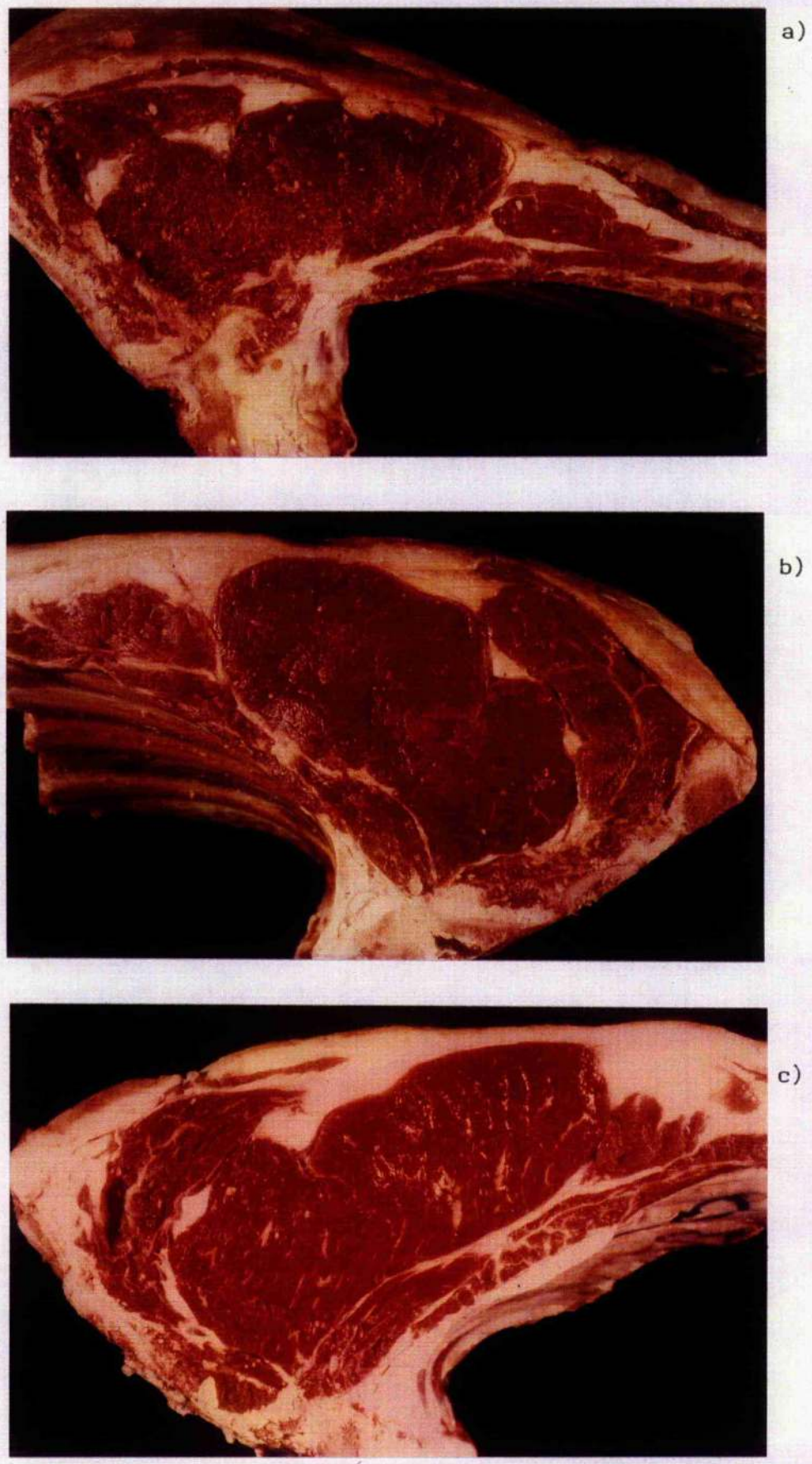


Plate 12 Indicator joint (10th rib) from steers at marketing: a) control, b) MSRB, c) clean.

DISCUSSION

SECOND GRAZING SEASON

The weather pattern for 1983 was reasonably typical for the West of Scotland with rain occurring in most months and being particularly heavy in the Autumn (see Fig. C7). Consequently, the epidemiology of ostertagiasis in the group of naive-control calves grazed for the first time in 1983, was typical, with the disease surfacing in early September and just two weeks later than the naive-control calves which grazed in 1982.

However, when the faecal egg counts from the different groups of second year cattle and the larval counts from the pastures grazed by these cattle, are compared to the same parameters in the naive controls, there are clear differences. Thus the epg's of the second year animals never exceeded 72 epg and were frequently negative, whereas the naive controls achieved a mean maximum of 773 epg; the highest numbers of trichostrongyle L_3 recovered from the pastures of the second year animals were 8396 Ostertagia and 312 Cooperia L_3 in a control plot in September and 353 Ostertagia from the MSRB plot in October compared to 17,930 O. ostertagi and 10,345 C. oncophora on those grazed by the naive controls. Furthermore, clinical type I ostertagiasis occurred in the 1983 naive controls whereas the 1982-born cattle were clinically normal.

The differences in plasma pepsinogen and gastrin levels were not quite so clear cut and this is discussed later.

At post-mortem the mean Ostertagia burden of the 1983 calves was 118,750 and the mean Cooperia burden was 26,025; low burdens of T. axei were also recorded although no larvae of this species were recovered from the pasture. In contrast, the Ostertagia burden in the 1982 born cattle of the MSRB treated and control groups were all below 11,000 and the Cooperia burdens were negative except for one animal.

Clearly, a very strong immunity to Ostertagia spp. had developed in the second year animals and an almost absolute immunity to Cooperia spp., the latter agreeing with the results of Coop et al(1979) and Smith and Archibald (1968). This immunity was strongly developed in cattle from both the controls and the MSRB treated group

and the introduction of the bolus had no detrimental effect on the development of immunity. The relatively low numbers of Ostertagia L_3 and negligible Cooperia L_3 which were present in the second year pastures were evidently sufficient to stimulate the strong immunity to re-infection. These results are very similar to the results of Armour et al (1979) and confirms that under natural grazing conditions a good immunity to the common gastro-intestinal nematodes of cattle in Britain, develops during the second grazing season, assuming that the cattle are exposed to infection.

An interesting finding was the presence of higher numbers of O. ostertagi L_3 in the control grazed plots from early September until October, compared to those in the MSRB plots. This occurred despite the epg's of the cattle in both groups being very similar and therefore contamination of the pastures with eggs during the grazing season of 1983 also being similar. The source of the extra larvae found in the control plots is not definitely known but may reflect a reduced hatchability of the eggs produced by the MSRB treated calves indicating a larvicidal activity of the morantel excreted in the faeces as suggested by Prosl et al (1983); alternatively, the larvae may have originated from eggs or larval reservoirs in the faeces or soil as demonstrated by Al Saqur, Bairden and Armour (1982). However, since the plots grazed in 1982 were partly interchanged between the groups for the 1983 season one would have expected a larval reservoir to be present in one of the areas grazed by the different groups in 1983; there was no evidence of this and it therefore seems more likely that a reduced hatchability of the trichostrongyle eggs produced by the MSRB treated calves was responsible for the reduced larval counts recorded from the MSRB grazed plots.

The fact that the mean plasma pepsinogen values in the clean group remained at or below 1.0 I.u., except on one occasion, when it was 1.3 I.u., indicates that these animals remained uninfected by Ostertagia during the second grazing season. This was confirmed by the parasitology data. These figures are very important since they are the first pepsinogen values available from Ostertagia-free cattle in their second grazing season. The mean values were also consistent with those of Ostertagia-naive calves less than six months old and indicate that age per se does not alter the normal levels of plasma pepsinogen to any extent.

The mean pepsinogen values from the MSRB treated calves in the

spring were slightly higher than is generally considered 'normal' (ranging from0.9 tol.6) but probably reflect the true normal value in animals which have been moderately exposed to Ostertagia infection in their first grazing season and to a light infection in the second season. They steadily declined towards the end of the season, presumably as their immunity developed.

The values in the control group are particularly interesting. Following recovery from the type II phase there was a steady fall in pepsinogen values from a mean of 6.6 I.u. in May to 2.8 I.u. in August. The fact that the decline in values ceased from then until housing, and coincided with an increase in L_3 populations on the pastures, suggests that despite the development of immunity, the reaction in the mucosa to the moderate larval challenge was sufficient to sustain the pepsinogen values around a mean of 2.8 I.u.. The reason for this is not known, but probably reflects the increased permeability of the mucosa at the time of the immune reaction to the ingestion and rejection of larvae, coupled with the establishment of low populations of adult Ostertagia. It has been suggested that the immune reaction to larval challenge at the mucosal level in gastro-intestinal parasites is based on a hypersensitivity response (Barth, Jarrett and Urquhart, 1966; Anderson, 1977; and Armour et al, 1980) with an increased permeability to macromolecules such as pepsinogen. The current results are consistent with such a hypothesis and it is interesting that once the larval challenge was removed at the housing of the cattle, the mean pepsinogen levels fell to 1.2 I.u. by the time of slaughter in December and January. What is important from a diagnostic point of view is that clinically normal cattle aged 18 months to two years had pepsinogen values around 3.0 I.u., a level generally considered to be diagnostic for animals suffering from clinical ostertagiasis (Selman et al, 1976).

Of course, it should be remembered that the abomasa of the MSRB treated cattle may not have been functioning optimally and that this was reflected in the moderately elevated plasma pepsinogens, but clinically masked by compensatory changes further down the intestinal tract.

The serum gastrin levels provide some clues to the abomasal functioning but also create some doubts as to what was happening. Thus, as stated previously, a limited number of technique kits were available for gastrin estimation and in one individual with a high

value, sufficient dilutions could not be made in order to get the actual value. In such cases, an estimated value was used and therefore the average values were slightly lower than the exact level. Another difficulty is in the interpretation of gastrin levels and what can be considered 'normal'. The situation is also complicated by the use of a human gastrin assay kit and the possibility that some cholescystokinin, which is structurally similar, is also being assayed by the technique.

However, despite the above strictures on the use of serum gastrin as an indicator of abomasal changes in ostertagiasis, the results presented in Fig. C3 are helpful in formulating an explanation of what is happening in the abomasum. Serum gastrin levels are known to increase in response to a reduction in gastric acidity such as occurs in clinical ostertagiasis, but also responds to physiological changes in the abomasum, such as created by dietary change or larval challenge in the immune animal. If the mean gastrin levels of the animals in each group are examined it appears that various stimuli are creating changes in the gastrin levels. Thus in the late spring (May and early June) there were increases in the levels of serum gastrin in the controls and the MSRB treated group which were under a low larval challenge from the pastures, but no change in the clean group which received no challenge at this time. The serum gastrin of the controls and the MSRB treated groups then decreased rapidly and coincidentally with the drop in larval challenge. The larval challenge in the autumn in the control pastures seemed to halt the drop in mean gastrin levels in the control calves and these, rather like the plasma pepsinogen levels, then plateau until near the end of the grazing season. Following removal of this larval challenge by housing the serum gastrin levels of the controls had fallen to pre-infection levels by the time of slaughter (Table C9). There was a rise in the the serum gastrin levels of the MSRB treated and clean control calves in the autumn, coincident with the introduction of concentrates.

What then is the value of serum gastrin levels in estimating the degree of abomasitis in ostertagiasis? It appears from Fig. C3 that the levels of gastrin closely follow the trend for pepsinogen in cattle infected with O. ostertagi and that elevated gastrin levels are also a good indication of an actively developing lesion of ostertagiasis. Until more information is available on what these levels are in different grades of infection and different ages of cattle, the

ultimate value of the test in diagnosis will not be known. However, it is possible that consideration of gastrin and pepsinogen levels in the serum of cattle will prove helpful in instances where pepsinogen levels in older cattle are moderately elevated but there are no clinical signs. In such cases a high serum gastrin, say more than 600 pg/ml could indicate the changes in acidity associated with developing Ostertagia infections and treatment would be indicated; a low gastrin level, say less than 400, would indicate the reverse. However, more information is required on what are significant levels of serum gastrin before the test could be recommended for routine use.

Over the whole of the second grazing season (0-22 weeks) the mean liveweight gains of the clean, MSRB treated and the control groups were 105, 131 and 109 kg respectively. Particularly interesting was the marked increase in weight observed for the MSRB treated animals during the first two weeks of grazing which was 10 kg greater than that of the other two groups. This result could not be explained by the larval challenge because it was either negative or very low. A probable explanation was a compensatory gut-fill effect due to the lack of available grass which occurred in the MSRB plots (Plate 9) at the end of the previous grazing season and which was not fully compensated for during the housing period because of the restriction of the pair-feeding system employed.

Differences in performance between the MSRB treated and the control animals during the whole season may also be explained by a lengthy physiological recovery from the clinical type II ostertagiasis suffered by the control animals, due to the persistence of developing larvae and adults whose presence was supported by higher plasma pepsinogen and gastrin values, faecal egg count, lower albumin values, and later on in the season, by an increase in the larval challenge from the pasture.

Parkins et al (1982 b) concluded that the detrimental effects of nematode infections on production in cattle during the winter might persist for two or three months even following an effective anthelmintic treatment at housing. However, it is difficult to explain the difference of 10 kg which accumulated in the first 12 weeks of grazing in favour of the MSRB treated animals over the clean group, since there was no apparent difference in grass availability. Some speculation might be offered. Firstly, the repeated treatment (11 per season) with fenbendazole could have produced some alteration in the normal digestive physiology of the clean controls which lowered production since substituted benzimidazoles have been shown to decrease acid production in the abomasum (Fellenius, Berglindh et al, 1981). However an increase in plasma pepsinogen and/or gastrin might also have been expected. The second, and indeed a more controversial explanation, is that a low parasitic burden could produce an overall beneficial effect due to an increase in digestion and absorption of the small intestine. After all, animals with parasites are the 'normal'

under nearly all global grazing situations.

A factor produced by a parasite having physiological effects in the host was described by Steelman, Glitzer, Ostlind and Mueller (1971) in rats infected with Spirometra mansanoides. A larval (sparganum) form of this parasite has a growth hormone-like effect and was associated with increased weight gain over the controls, apparently acting similarly to growth hormones on bone, protein synthesis and in increasing liver glycogen. Another possibility is that morantel has an anabolic effect since the differences occurred during the expected release period of the drug, calculated as 90 days, although this seems unlikely since, in many of the trials reported by Jones (1981), there were no significant differences between the MSRB treated cattle and those with light infections. However, an interesting feature at post-mortem was the much higher mucosal fraction in the control group compared to the other two groups (2.2 compared with 1.1 in each of the control and the MSRB treated animals).

The reason for these persistently high mucosal fractions, despite the very low worm burdens and the relatively normal plasma pepsinogen and serum gastrin levels, is not definitely known. It cannot be attributed solely to oedema and hyperaemia since these were not particularly obvious on examination of the gut. Presumably, hyperplasia of the mucosa must make a major contribution to the increased weight of the mucosa. However, the reasons for the persisting hyperplasia are not known. One possible stimulus might be the continued ingestion of L_3 larvae or perhaps the presence of low numbers of adult parasites.

Data obtained from the carcass measurements and analyses should properly be considered in relation to the cumulative effects of type I and type II ostertagiasis. The sometimes highly significantly improved results obtained from the clean and MSRB treated groups over those of the control animals with respect to the hot carcass weight, carcass, leg and total length, leg circumference and width at the 7th rib, are clearly expressive of the detrimental effects of prolonged infection with O. ostertagi on growth and protein deposition.

There were few significant differences in the carcass linear measurements taken between the control and the MSRB treated groups at the end of the first grazing season in which type I ostertagiasis occurred in the control animals. However, type I ostertagiasis occurs

over a relatively short period and was, in this experiment halted by deliberate anthelmintic treatment, so it is unlikely to have had any effect on skeletal development. However, the more persistent action of O. ostertagi throughout the winter, mainly during the clinical type II ostertagiasis, and probably during the second grazing season, clearly contributed to the significant differences recorded at slaughter at the end of the experiment.

The control cattle at slaughter were smaller in weight and skeletal size and had a lower fat cover. Weight itself is a major determinant of composition, as has been shown in many studies (e.g. Burton and Reid, 1969). It has also been recorded that for any given species, (and the breed type or cross thereof), the weight of the body alone provides the best single indicator of its composition (Lister, Perry and Wood, 1983). However, in the current work, liveweight at slaughter differed by a mean of 35 kg (and the dressed carcass weight by about 20 kg) between the smaller control cattle and both the clean and MSRB treated groups of steers and, clearly alone, does not account for the differences noted in the leg circumference and the compositions observed in the indicator rib joint analyses. Protein deposition was clearly affected by the course of both the type I and the type II occurrence of ostertagiasis in the control animals.

The reduced leg muscle mass as shown from the lowered circumference measurements at the symphysis pubis and the lowered lean tissue weights from the 7-10th forerib dissection together with the eye muscle analyses for area, weight and crude protein concentration confirms the occurrence of a lowered protein deposition in the control animals. The compositional analyses of the ribs shows the controls to have a significantly greater bone content (as a percentage of total joint weight), a decreased fat content and apparently the same% lean tissue content. However, on a fresh weight basis the actual weights of lean tissue are considerably lower in these control cattle than in the other two groups.

In contrast to the lowered killing out percentages reported in the control calves after the type I occurrence in Section A, the killing out percentages of the hot carcasses here, after two grazing periods, were similar in the three groups. Also the water holding capacity and the total dry matter contents of the eye muscle were similar, contrasting with the lowered dry matter (and probably increased WHC as presumed from the watery appearance and 'drip' of the eye muscle

surface) of the control cattle indicator ribs at slaughter after one grazing.

Fig. C1

Mean liveweights of clean, MSRB treated and control cattle recorded during the second grazing season

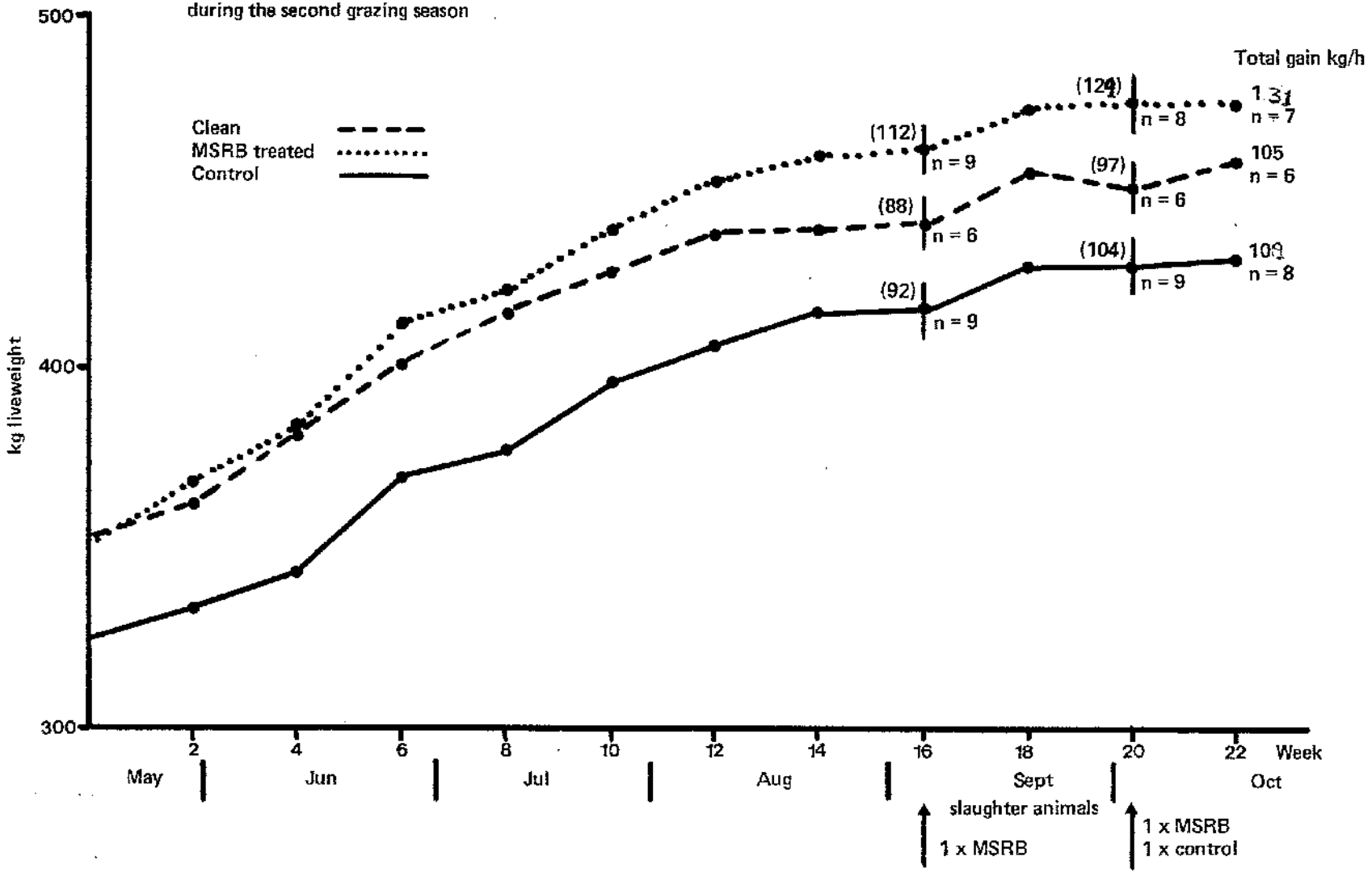


Fig. C2

Mean Plasma Pepsinogen Values (IU Tryosine) of clean, MSRB treated and control cattle during the second grazing season.

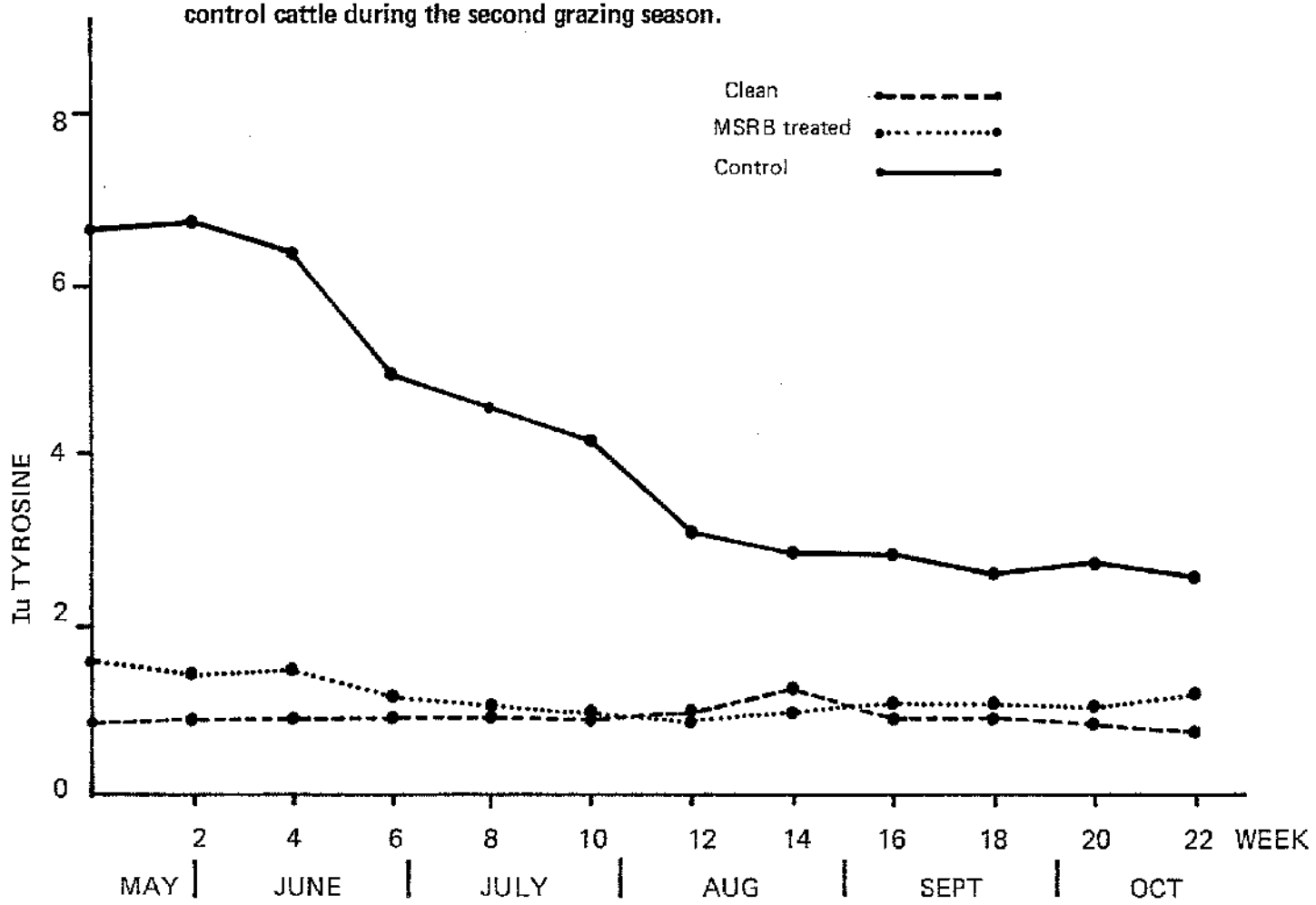


Fig. 03

MEAN PLASMA GASTRIN LEVELS OF CLEAN MSRB TREATED AND CONTROL CATTLE DURING SECOND GRAZING SEASON

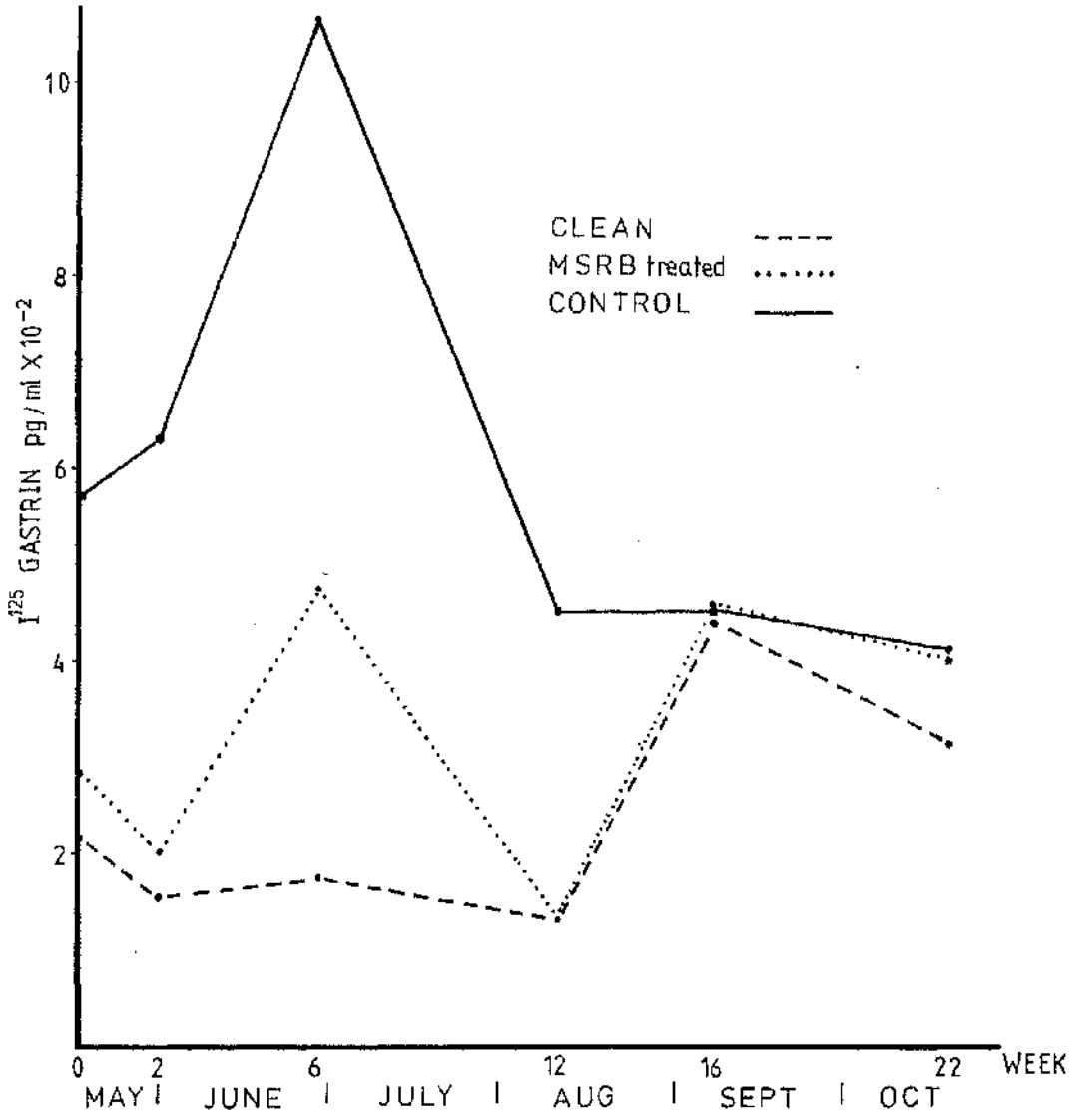


Fig. 04

Mean trichostrongyle egg count in the faeces of clean, MSRB treated and control cattle during the second grazing period

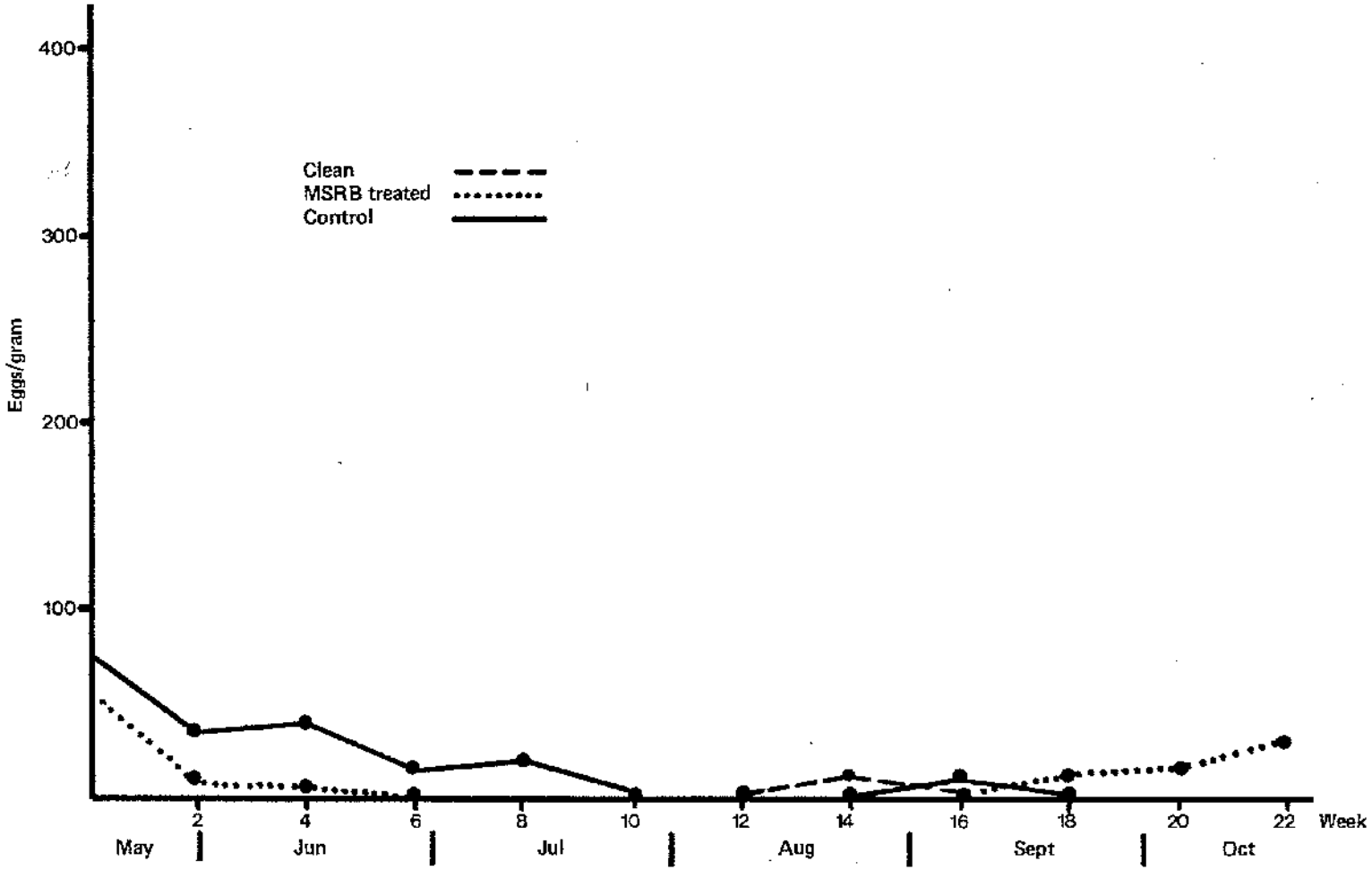


Fig. C5

Numbers of O.ostertagi L_3 larvae recovered from paddocks of clean, MSRB treated and control cattle during the second grazing period

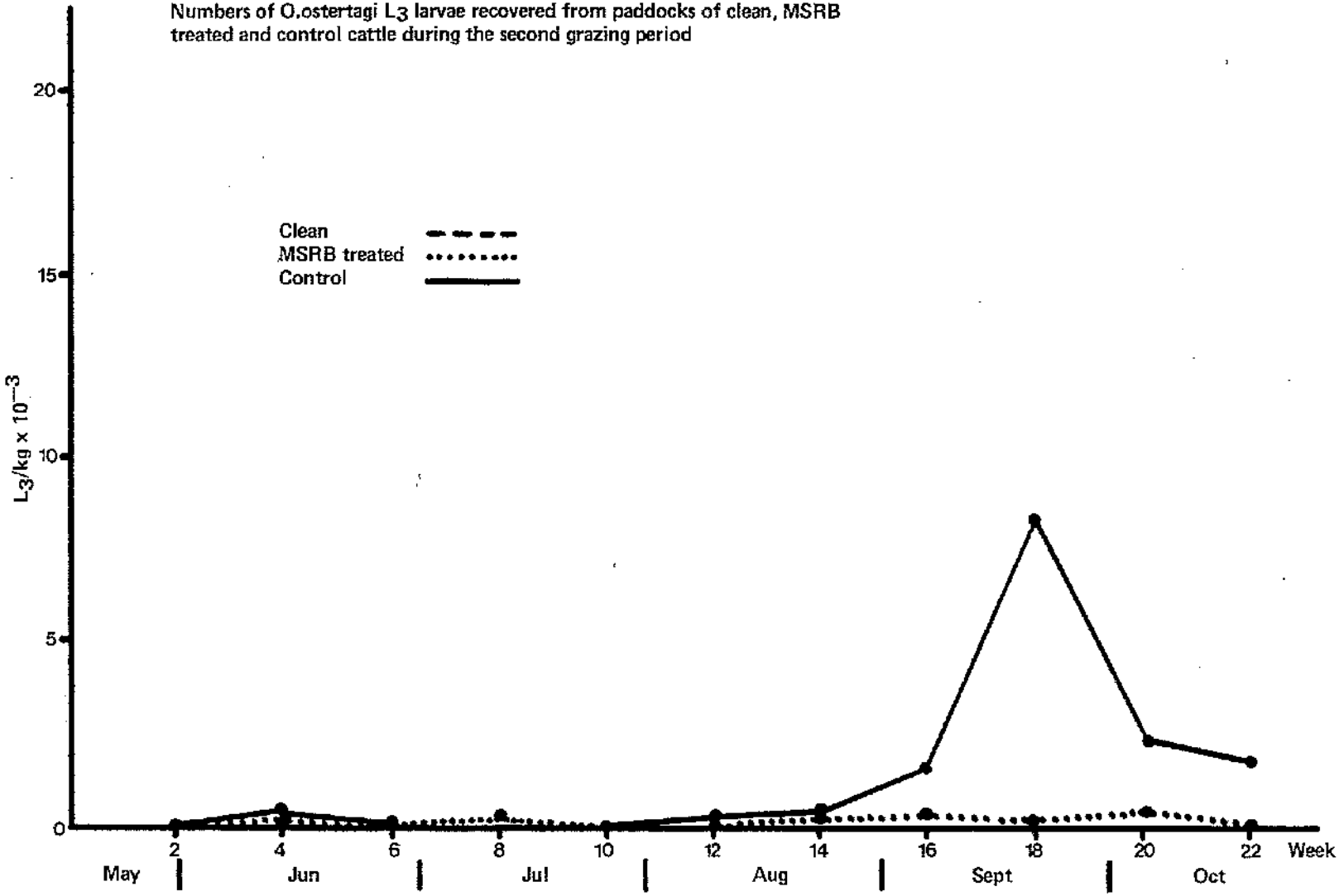


Fig. C6 Mean herbage larvae, egg count and plasma pepsinogen of helminth-naive calves during the second grazing season.

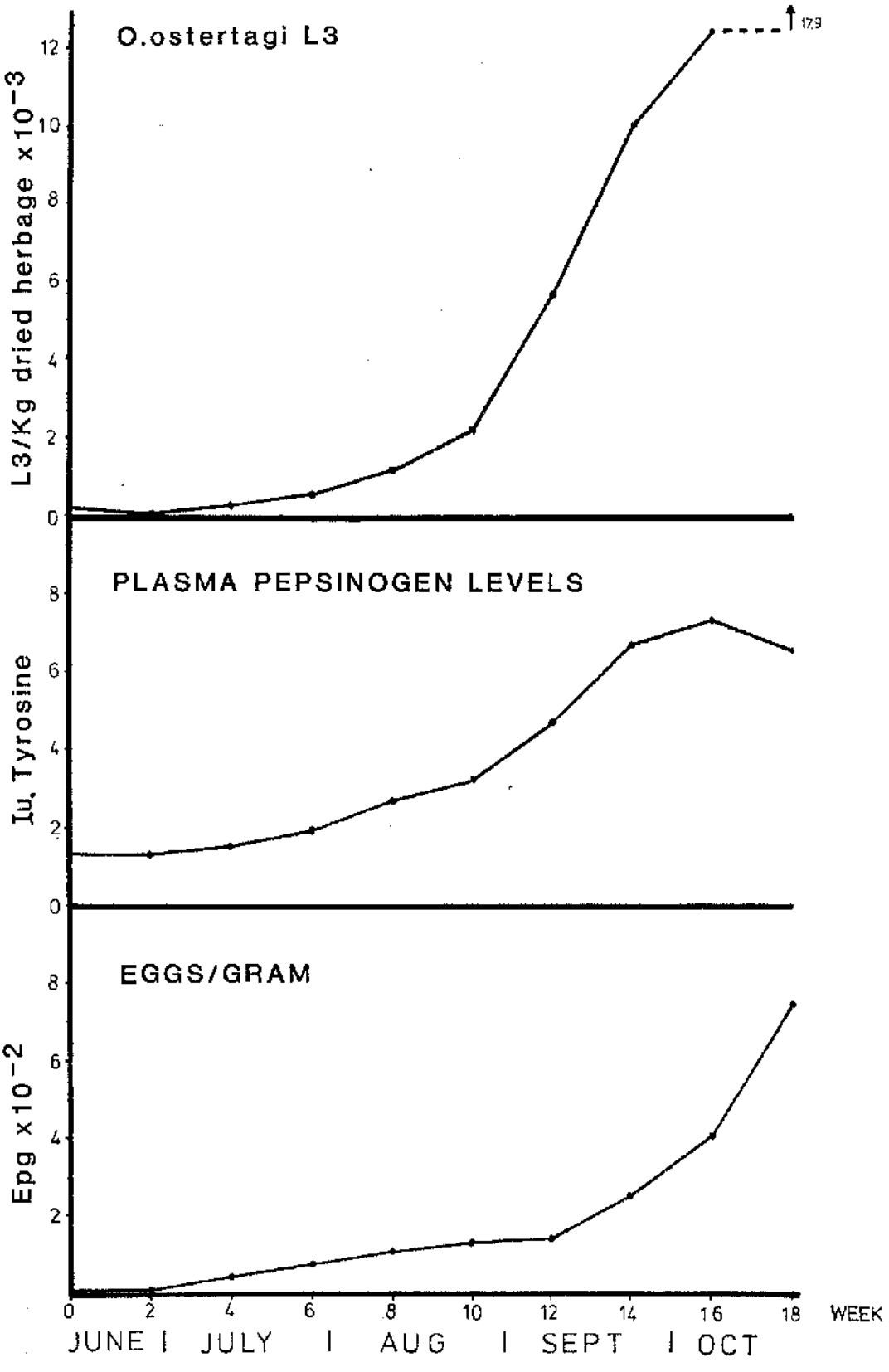

O.ostertagi L3
17.9
L3/Kg dried herbage x10⁻³
12
10
8
6
4
2
0
PLASMA PEPSINOGEN LEVELS
Iu. Tyrosine
8
6
4
2
0
EGGS/GRAM
Epg x10⁻²
8
6
4
2
0
2
4
6
8
10
12
14
16
18
WEEK
JUNE | JULY | AUG | SEPT | OCT

Fig. C7 Temperature and Rainfall from April to November in the experimental years 1982-1983 compared with those of 1975-1979

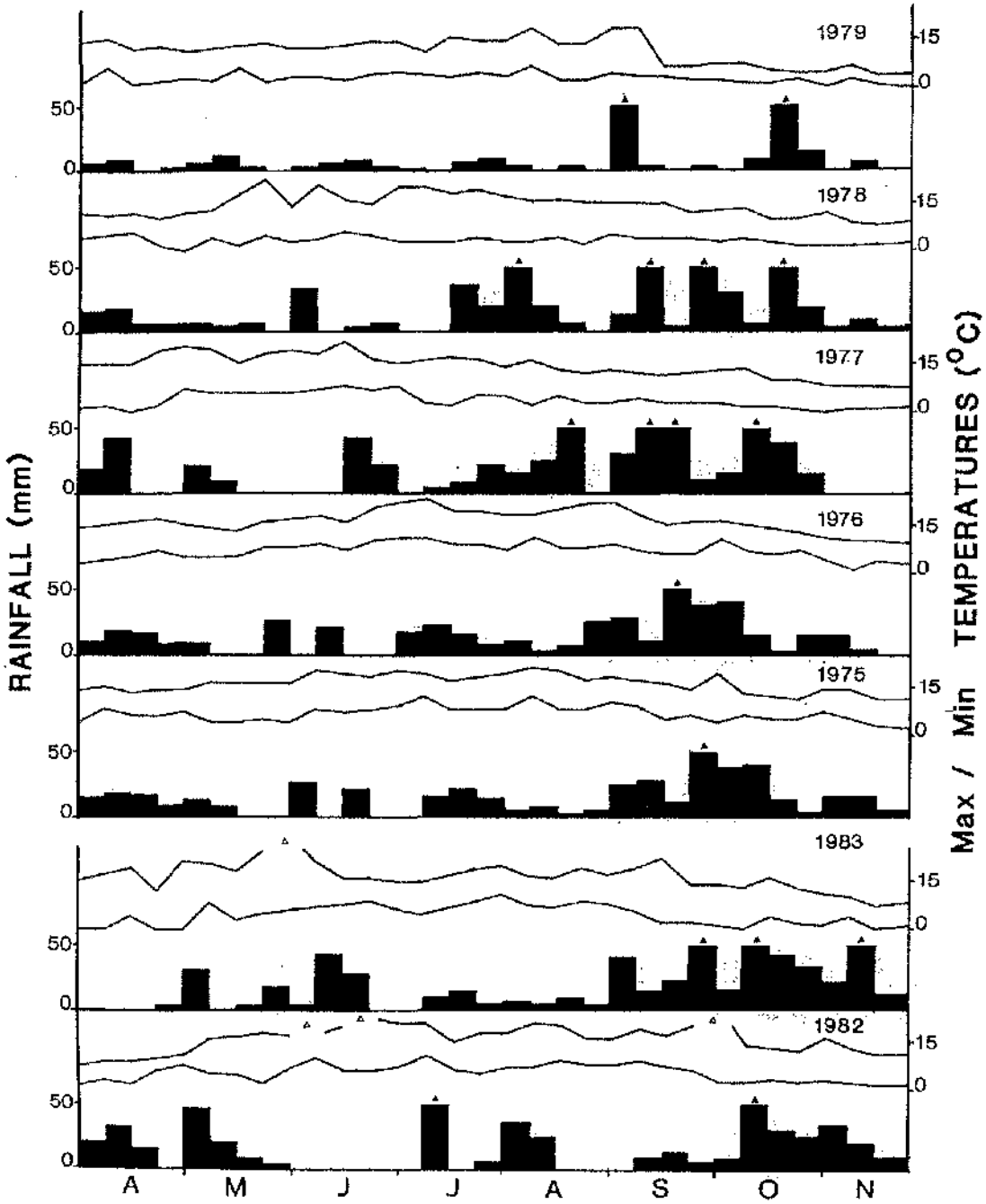

1979
1978
1977
1976
1975
1983
1982
50
0
15
0
RAINFALL (mm)
Max / Min TEMPERATURES (°C)
A
M
J
J
A
S
O
N

Fig. C8

PLASMA PEPSINOGEN AND GASTRIN LEVELS OF 5 CONTROL CALVES THROUGHOUT THE EXPERIMENTAL PERIOD

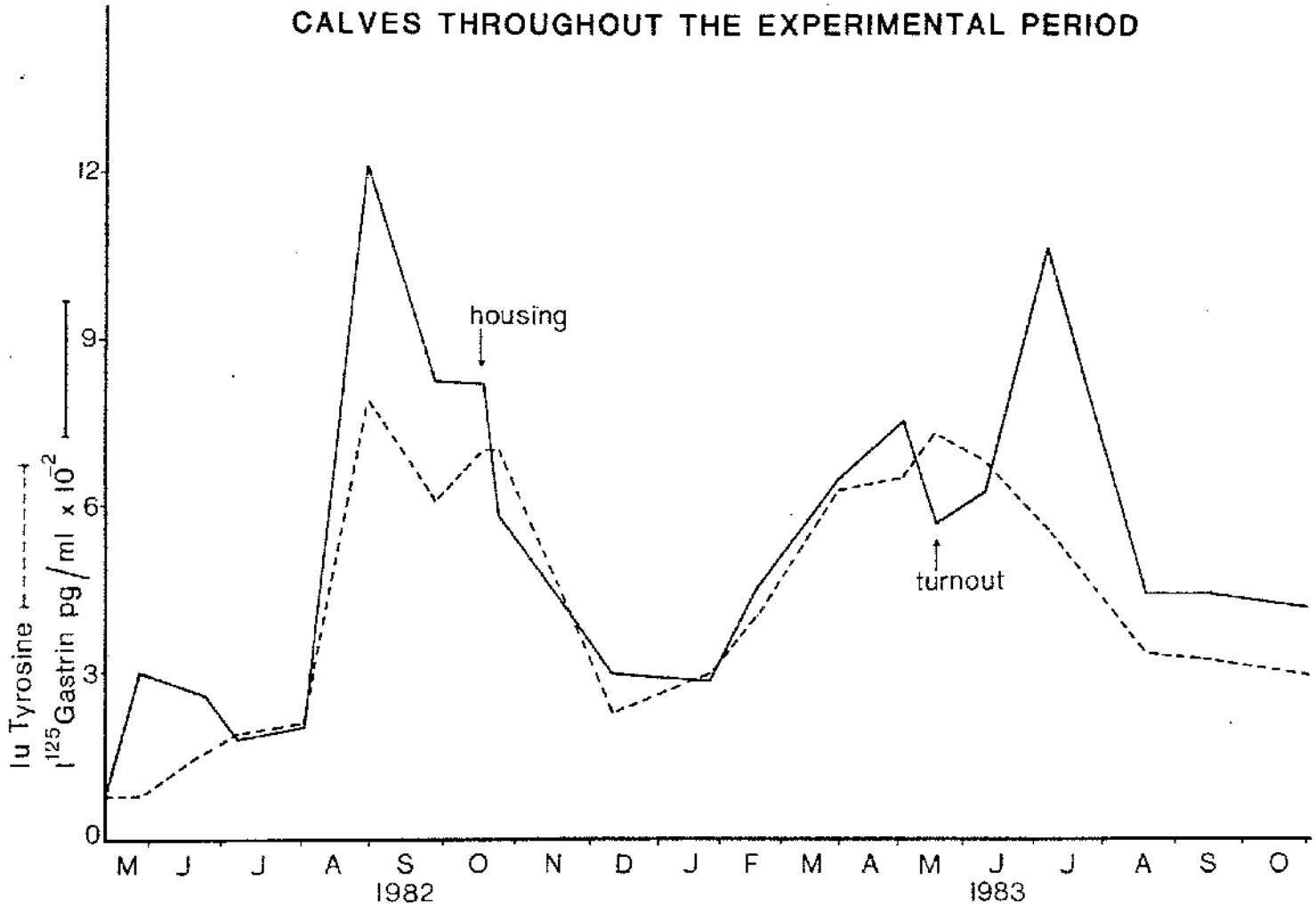


Table C1. Mean liveweight (kg±SE) of Clean, MSRB treated and Control cattle during the second grazing season together with mean liveweight gain data

Week	Date	Clean (n=6)	MSRB (n=9)	Control (n=9)
0	18.05.83	353 ± 5.1	350 ± 7.7	324 ± 9.1
2	31.05.83	362 ± 4.5	369 ± 6.9	332 ± 8.7
4	13.06.83	383 ± 5.6	384 ± 9.1	343 ± 9.4
6	27.06.83	402 ± 5.2	413 ± 9.3	370 ± 9.9
8	11.07.83	416 ± 6.1	422 ± 9.9	377 ± 8.8
10	26.07.83	427 ± 8.4	439 ± 9.9	396 ± 7.3
12	9.08.83	438 ± 7.5	453 ± 9.1	406 ± 7.3
14	23.08.83	439 ± 7.8	459 ± 8.7	415 ± 6.6
16(1)	5.09.83	441 ± 6.0	462 ± 7.2	416 ± 6.6
18	20.09.83	455 ± 4.8	473+ ± 8.6	428 ± 6.4
20(2)	3.10.83	450 ± 7.6	474 ± 7.4	428 ± 5.0
22(3)	17.10.83	458 ± 7.2	474● ± 7.4	430+ ± 6.4
Mean liveweight gain over 112 days kg (1)		88	112	92
LW gain kg/d		0.78	1.00	0.82
Mean LW gain over 140 days (2)		97	129+	104
LW gain kg/d		0.70	0.92	0.74
Mean LW gain over 152 days (3)		105	131°	109+
LW gain kg/d		0.69	0.86	0.70

\+ n = 8 animals
● n = 7 animals

Table C2. Mean plasma pepsinogen values (I.u. tyrosine) recorded in the Clean, MSRB treated and Control cattle during the second grazing season

Week	Clean[b]	MSRB[b]	Control[A]
0	0.8 ± 0.04	1.6 ± 0.17	6.6 ± 0.60
2	0.9 ± 0.04	1.4 ± 0.15	6.7 ± 0.48
4	0.9 ± 0.04	1.5 ± 0.15	6.3 ± 0.57
6	0.9 ± 0.04	1.2 ± 0.08	4.9 ± 0.57
8	0.9 ± 0.06	1.1 ± 0.06	4.5 ± 0.56
10	0.9 ± 0.06	1.0 ± 0.04	4.1 ± 0.52
12	0.9 ± 0.05	0.9 ± 0.04	3.0 ± 0.33
14	1.3 ± 0.05	1.0 ± 0.06	2.8 ± 0.27
16	0.9 ± 0.05	1.1 ± 0.10	2.8 ± 0.27
18	0.9 ± 0.04	1.1 ± 0.09	2.6 ± 0.20
20	0.8 ± 0.04	1.0 ± 0.10	2.7 ± 0.26
22	0.8 ± 0.04	1.2 ± 0.09	2.6 ± 0.25

0 2 4 6 8 10 12 14 16 18 20 22 Weeks

May June July Aug Sept Oct Months

A,b $P<0.001$

Table C3. Mean plasma gastrin (pg/ml) concentrations (+SE) determined in Clean, MSRB treated and Control cattle during the second grazing season

Week	Date	Clean	MSRB	Control
0	10/ 5/83	214 ±21.8^{b}	284 ±27.8^{b}	573 ± 98.1^{a}
2	31/ 5/83	154 ± 8.6 ^{b}	200 ±16.1^{b}	626 ±161.3 ^{a}
6	27/ 6/83	174 ±17.3 ^{c}	473 ±51.6 ^{B}	1064 ±171.8 ^{A}
12	9/ 8/83	131 ± 5.6 ^{b}	131 ± 3.8 ^{b}	450 ± 72.7 ^{a}
16	5/ 9/83	438 ±17.5	456 ±25.1	448 ± 43.3
22	18/10/83	312 ±11.0	400 ±34.0	410 ± 54.0

AB; a,b,c, $P<0.005$

A b,c $p<0.001$

Table C4. Blood biochemical analyses on five occasions from Clean, MSRB treated and Control calves (±SE) during the second grazing season

On any one occasion values with different lettered superscripts are significantly different viz.,

a, b, c $P<0.05$

A, b $P<0.001$

Week / Group	0 18.05.83	2 31.05.83	7 26.07.83	18 30.09.83	22 18.10.83
Urea mmol/l					
Clean	3.2 ±0.48	7.0 ±0.17^{A}	6.5 ±0.25^{A}	5.5 ±0.20^{b}	5.4 ±0.22
MSRB	3.2 ±0.34	5.2 ±0.08^{b}	4.9 ±0.16^{b}	6.6 ±0.21^{a}	5.3 ±0.18
Control	2.5 ±0.41	5.3 ±0.22^{b}	5.0 ±0.26^{b}	6.5 ±0.31^{a}	5.7 ±0.32
Inorganic Phosphate mmol/l					
Clean	2.3 ±0.07	2.3 ±0.05	2.3 ±0.08	2.6 ±0.08	2.1 ±0.01
MSRB	2.4 ±0.08	2.4 ±0.06	2.4 ±0.10	2.4 ±0.09	2.4 ±0.09
Control	2.5 ±0.09	2.3 ±0.10	2.4 ±0.11	2.2 ±0.07	2.2 ±0.05
Total Protein g/l					
Clean	69.5 ±1.1	78.8 ±2.0	69.8 ±1.0	64.0 ±2.2	69.8 ±1.8
MSRB	67.3 ±0.8	79.1 ±1.0	72.0 ±1.0	62.5 ±1.7	69.8 ±0.9
Control	69.2 ±1.1	77.7 ±0.9	73.6 ±1.0	66.6 ±1.4	65.0 ±2.2
Albumin g/l					
Clean	36.0 ±0.6^{A}	39.2 ±0.7^{A}	38.0 ±0.7^{A}	36.8 ±0.8^{A}	39.2 ±0.4^{a}
MSRB	34.4 ±0.7^{A}	36.3 ±0.9^{b}	35.7 ±0.2^{b}	35.8 ±1.0^{a}	39.1 ±0.6$^{a\ b}$
Control	30.3 ±0.8^{b}	32.6 ±1.1^{c}	33.6 ±0.6^{c}	32.1 ±0.1^{b}	37.4 ±0.6^{b}

Table C5. Mean trichostrongyle egg count (eggs per gram faeces) in Clean, MSRB treated and Control cattle during the second grazing season

Week	Clean	MSRB	Control
0	0	56	72
2	0	6	33
4	0	6	38
6	0	0	11
8	0	0	17
10	0	0	0
12	0	0	0
14	7	0	0
16	0	0	6
18	0	7	0
20	0	12.5	0
22	0	29	0

Table C6. Recovery of L_3 Ostertagia larvae (per kg dry grass) in the paddocks grazed by the Clean, MSRB treated and Control cattle in the second season at grass

Week	Clean	MSRB	Control
0	0	0	0
2	0	0	0
4	0	133	236
6	0	0	0
8	0	68	0
10	0	10	0
12	0	50	192
14	0	169	233
16	0	278	1447
18	0	126	8396
20	0	353	2500
22	0	0	1698

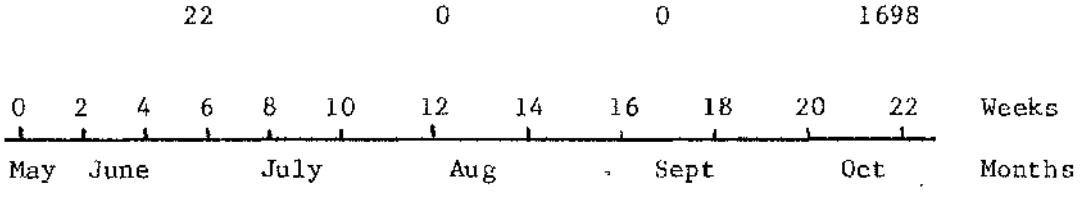


Table C9. Mean plasma gastrin and plasma pepsinogen values determined in Clean, MSRB treated and Control cattle at slaughter after two periods at grass

Clean			MSRB			Control		
No.	Gastrin	PP	No.	Gastrin	PP	No.	Gastrin	PP
1	145	0.6	9	140	1.1	18	140	0.8
2	125	0.8	10	160	0.8	19	200	1.6
3	150	0.8	11	210	0.7	20	175	1.0
4	140	0.8	12	185	0.7	21	210	1.7
5	190	0.7	13	145	1.7	22	145	1.0
8	190	0.7	14	140	0.7	23	170	1.1
			15	180	1.0	24	210	1.0
			16	150	0.7	25	200	1.3
			17	210	0.8	26	120	1.1
Mean	156.6	0.73		168.9	0.91		174.4	1.2
±SE	10.1	0.03		8.9	0.10		10.5	0.009

Gastrin pg/ml

Plasma Pepsinogen I.u. tyrosine

Table C10. Parasitological and biochemical parameters from four parasite naive calves grazed on Trichostrongyle contaminated pasture in 1983

Date	Week	Eggs per gm faeces	Plasma Pepsinogen (I.u.)	Trichostrongyle L_3 on Pasture		
				Ostertagia	Cooperia	Nematodirus
13.06	0	12	1.3 ±0.1	128	--	1154
27.06	2	12	1.3 ±0.1	--	--	2444
12.07	4	37	1.5 ±0.2	275	--	92
27.07	6	--	1.9 ±0.2	615	--	--
08.08	8	100	2.7 ±0.5	1133	--	--
25.08	10	125	3.2 ±0.6	2135	--	--
06.09	12	137	4.7 ±1.0	5644	--	--
21.09	14	250	6.7 ±1.0	9126	1942	--
04.10	16	400	7.3 ±1.8	12381	1905	--
17.10	18	733	6.5 ±1.6	17931	10345	--

Table C11 Post-mortem worm counts, plasma pepsinogen (I.u.) and gastrin (pg/l) of calves grazed from May to September, 1983 on pastures contaminated with Trichostrongyle L_3.

				O.ostertagi								
An.	pH	P.P.	Gastrin	L_4	L_5	Ad	Total	Trich. spp	L_4	L_5	Ad	Total
38	5.4	9.1	1200	36,800	7,400	93,600	137,800	2,200	15,700	900	9,900	26,500
39	4.9	7.8	740	28,100	—	42,000	71,000	1,600	—	—	1,400	1,400
50	4.6	2.7	320	37,400	200	4,100	41,700	—	3,400	—	9,000	12,400
32	6.1	5.6	1900	22.200	60,400	142,800	225,400	200	49,200	5500	9,100	63,800
$\bar{x}$		6.3	1040	31,125	17,001	70,625	118,750	1,001	17,076	1601	7,350	26,025
$\bar{x}$ + SE		1.2	16.12	3,166	12,616	26,199	35,391	463	9,722	1140	1,726	11,777

Table C12, Individual and mean worm burdens of Clean, MSRB treated and Control groups at slaughter following two grazing seasons

O. ostertagi

Group	An. No.	L_4	L_5	Adults	Total	$\%L_4$
MSRB	9	--	--	200	200	0
	13	400	--	--	400	100
	14	400	--	200	600	67
	17	1000	--	--	1000	100
	16	200	--	400	600	34
	12	800	--	200	1000	80
	10	8200	400	1200	9800	84
	11	1800	--	1000	2800	64
	15	7400	--	100	7500	99
	Mean	2245	48	376	2656	70 (84)
	SE	±1005	±41	±137	±1109	±11.3
Control	19	--	--	--	--	
	22	10800	--	400	11200	96
	26	--	--	150	150	0
	18	800	--	900	1700	47
	23	1400	--	300	1700	82
	25	--	--	200	200	0
	24	1400	--	700	2100	67
	20	800	--	--	800	100
	21	4800	--	400	5200	92
	Mean	2223	--	340	2561	60 (87)
	SE	±1113	--	±95	±1135	±14.5

Clean:— No worms found at all in 6 animals.

( ) = % L_4 of total worms

Table C13. Liveweight (LW kg), Abomasum weight (AW g), Abomasal mucosa weight (MW g), pH and the Mucosal Fraction (MF g/kg, MW/LW) of Clean, MSRB treated and Control cattle slaughtered after two grazing seasons

Group	No	LW	AW	MW	MF	pH
Clean	2	534	1436	654	1.2	3.4
	1	510	1210	356	0.7	2.5
	3	543	1427	548	1.0	3.2
	5	553	1425	677	1.2	3.5
	4	530	1369	623	1.2	2.2
	8	502	1296	601	1.2	3.2
	Mean	529^{a}	1361^{b}	577^{b}	1.1^{b}	3.2^{b}
	SE±	7.2	33.8	43.6	0.06	0.2
MSRB	9	505	1303	750	1.5	3.4
	13	480	1764	640	1.3	2.8
	14	549	1683	710	1.3	3.1
	17	523	1455	528	1.0	3.5
	12	565	1398	526	0.9	2.9
	16	556	1440	542	1.0	3.0
	10	507	1072	501	1.0	3.4
	11	541	1370	607	1.1	2.5
	15	510	1515	680	1.3	3.6
	Mean	526^{a}	1444^{b}	609^{b}	1.1^{b}	3.1^{b}
	SE ±	8.9	63.9	28.5	0.06	0.1
Control	19	445	1680	540	1.2	3.6
	22	530	2177	954	1.8	4.9
	26	540	2137	1306	2.4	3.2
	18	510	1820	856	1.7	4.0
	23	491	1600	806	1.6	3.5
	25	490	3041	1200	2.4	4.5
	24	494	2638	1194	2.4	3.6
	20	460	1988	902	2.0	4.3
	21	493	1896	1028	2.2	4.0
	Mean	495^{b}	2109^{A}	956^{A}	2.0^{A}	4.0^{A}
	SE ±	9.5	146.6	74.4	0.13	0.2

a,b $P<0.05$ A,b $P<0.001$

Table C15 · Carcass Measurements of Clean group of Cattle slaughtered after two grazing seasons

No	Date	IW	Carcass wt	KO%	Leg Length	Car. Length	Total Length	Leg Circum.	Width 7th Rib	MLC Grade
2	7.12.83	534	296	52.9	92.0	134.5	221.0	114.5	70.0	R4L
1	9.12.83	510	286	51.6	88.5	132.0	213.5	112.0	68.0	04L
3	11.01.84	543	284	52.3	85.0	136.0	218.5	113.0	69.0	R4L
5	11.01.84	553	292	52.8	88.0	135.0	216.0	119.5	70.0	LC
8	16.01.84	502	270	53.8	84.0	130.0	208.5	112.0	67.5	04L
4	16.01.84	530	289	54.5	86.0	135.0	217.0	117.0	71.0	04L
Mean		528.7[a]	283.2[a]	53.0	87.3	133.7[A]	215.8[a]	114.7[a]	69.2[a]	5/6[A]
SE ±		7.2	4.35	0.39	1.08	0.8	1.61	1.13	0.55	

Table C15 Carcass Measurements of the MSRB treated group of Cattle slaughtered after two grazing seasons

No	Date	LW kg	Carcass wt kg	KO%	Leg Length cm	Car. Length cm	Total Length cm	Leg Circum. cm	Width 7th Rib cm	MLC Grade
9	6.09.83	505	267	52.9	84.0	132.0	213.0	113.0	68.5	03
13	6.10.83	480	268	55.8	82.0	127.0	211.5	111.0	67.5	LC
14	7.12.83	549	290	52.3	91.0	131.0	219.5	117.0	74.0	R4L
17	9.12.83	523	278	53.2	88.5	133.5	218.5	114.5	70.0	04L
16	12.01.84	556	310	55.8	89.0	130.0	213.5	119.5	72.0	R4L
12	12.01.84	565	312	55.2	93.0	135.0	221.0	117.0	74.0	R4L
10	19.01.84	507	284	56.0	89.0	137.0	221.0	114.0	71.0	03
11	19.01.84	541	293	54.1	86.5	142.0	224.0	117.0	71.0	04L
15	27.01.84	510	280	54.9	88.0	133.0	214.0	116.0	69.5	LC
Mean		526.2[a]	287[a]	54.5	87.9	133.4[a]	217.3[a]	115.4[A]	70.8[A]	7/9[A]
SE ±		8.86	5.1	0.43	1.05	1.36	1.98	0.81	0.7	

Table C15 Carcass Measurements of the Control group of Cattle slaughtered after two grazing seasons

No	Date	LW kg	Carcass wt kg	KO%	Leg Length cm	Car. Length cm	Total Length cm	Leg Circum. cm	Width 7th Rib cm	MLC Grade
19	6.10.83	445	248	55.7	78.0	122.0	186.0	105.0	63.0	LC
22	13.01.84	530	282	53.2	90.2	128.0	215.0	107.0	69.0	LC
26	13.01.84	540	281.5	52.1	88.0	133.0	216.0	112.0	69.0	LC
18	18.01.84	510	265	52.0	88.0	128.0	213.5	112.0	69.0	03
23	18.01.84	491	256	52.1	85.0	128.0	210.0	113.0	67.3	03
25	25.01.84	490	260.5	53.2	80.0	126.0	204.0	104.0	65.0	LC
24	25.01.84	494	272.0	55.1	85.0	130.0	210.0	110.0	66.0	LC
20	26.01.84	460	246.5	53.6	83.0	122.0	202.0	106.0	65.0	LC
21	26.01.84	493	270.0	54.8	84.0	132.0	210.0	110.0	66.0	LC
Mean		495 [b]	264.6 [b]	53.5	84.6	127.7 [b]	207.4 [b]	108.8 [b]	66.6 [b]	2/9 [b]
SE ±		9.5	4.1	0.44	1.2	1.2	2.9	1.05	0.67	

Table 016 Lean, Bone and Fat measurements of the 7-10th rib joint dissections in the Clean, MSRB treated and Control calves slaughtered after two grazing seasons

		Lean		Bone		Fat		Total Wt
Group	No	g	%	g	%	g	%	g
Clean	2	3563	61.5	1270	21.9	963	16.6	5795
	1	3725	64.2	1296	22.3	785	13.5	5805
	3	3580	59.6	1180	19.6	1250	20.8	6010
	5	3690	59.3	1250	20.1	1280	20.6	6220
	8	3455	60.8	1230	21.6	1001	17.6	5686
	4	3520	60.3	1150	19.7	1170	20.0	5840
	Mean	3589[a]	61.0	1229	20.9[b]	1075[a]	18.1	5893[A]
	SE ±	38.1	0.7	20.6	0.5	71.5	1.2	71.4
MSRB	9	3378	63.3	1190	22.3	768	14.4	5336
	13	3450	63.1	1210	22.1	810	14.8	5470
	14	3854	61.2	1281	20.3	1160	18.3	6295
	17	3675	62.3	1190	20.2	1030	17.5	5895
	16	3820	61.0	1370	21.9	1070	17.1	6260
	12	4105	62.9	1405	21.5	1020	15.6	6530
	10	3685	63.4	1310	22.5	820	14.1	5815
	11	3570	57.9	1220	19.8	1380	22.3	6170
	15	3709	61.1	1380	22.7	980	16.1	6069
	Mean	3694[A]	61.8	1284	21.5[b]	1004[a]	16.7	5978[A]
	SE±	69.0	0.6	27.0	0.4	60.7	0.8	123.3
Control	19	3185	67.6	1051	22.3	475	10.0	4711
	22	3760	67.4	1165	20.9	650	11.7	5575
	26	3506	62.4	1240	22.1	875	15.5	5621
	18	3164	60.3	1320	25.2	760	14.5	5244
	23	3126	61.1	1289	25.2	702	13.7	5117
	25	2737	53.3	1195	23.4	1190	21.3	5210
	24	2796	53.7	1305	25.0	1109	21.3	5210
	21	3292	64.0	1320	25.7	530	10.3	5142
	20	3458	63.0	1212	24.2	645	12.8	5015
	Mean	3192[b]	61.4	1233	23.8[a]	771[b]	14.6	5196[b]
	SE±	99.5	1.7	27.9	0.6	77.1	1.4	86.6

Table C17. Indicator joint (7-10th rib) eye muscle (EM) analyses of weight (g), area A (cm^2), dry matter content DM (g/kg), total DM weight EM DM (g), crude protein concentration CP g/kg DM, total g CP contained in the EM and Water Holding Capacity WHC % for Clean, MSRB treated and Control steers after two seasons at grass

Control

No	EM g	Acm^2	DM	EM DMg	gCP/kgDM	gCP EM	WHC%
18	980	48.9	255	249.8	868.6	217.0	18.2
19	1083	48.9	263	284.8	834.6	237.7	19.1
20	1048	41.1	253	264.8	874.7	231.6	25.2
21	1060	52.6	247	261.4	856.7	223.9	16.3
22	1250	52.6	247	308.3	886.2	273.2	16.9
23	946	46.6	264	249.4	836.0	208.5	23.6
24	1102	45.5	273	300.8	806.2	242.5	22.3
25	1005	55.6	263	263.9	807.6	213.1	19.2
26	1210	55.3	261	315.3	857.1	270.2	22.1
Mean	1076^b	49.7^b	258	277.6^b	847.5	235.3^b	20.3
±SE	31.6	1.5	27.0	8.0	9.5	7.8	1.0

Table (Continued) (- C 17)

Clean Group

No	EM g	Acm^2	DM	EM DMg	gCP/kgDM	gCP EM	WHC %
1	1320	72.6	254	334.9	851.2	285.1	24.0
2	1380	63.9	251	346.9	862.1	299.1	21.9
3	1370	72.9	253	346.1	900.4	311.6	21.8
4	1180	45.5	278	329.9	764.7	252.3	20.6
5	1410	61.6	269	379.3	801.9	304.2	23.9
8	1250	53.8	275	342.2	808.4	277.4	19.3
Mean	1318^A	61.7^A	263	346.7^A	831.5	288.3^A	21.9
±SE	32.8	4.0	4.5	6.4	20.0	8.8	0.7

MSRB

No	EM g	Acm^2	DM	EM DMg	gCP/kgDM	gCP EM	WHC %
9	1153	48.5	252	290.6	938.1	272.6	25.1
10	1210	45.9	281	340.1	751.6	255.6	17.9
11	1230	46.6	273	335.7	835.5	280.5	17.5
12	1430	72.2	253	361.2	877.9	317.1	24.9
13	1255	63.9	270	339.1	836.3	283.6	21.8
14	1365	65.4	270	368.4	851.9	313.8	21.5
15	1219	63.9	255	310.5	836.9	259.9	22.1
16	1470	69.5	279	410.3	798.2	327.5	23.6
17	1275	59.0	252	321.3	871.4	280.0	22.1
Mean	1290^A	59.4^A	265	341.9^A	844.2	287.8^A	21.9
±SE	33.9	3.1	3.7	11.0	17.3	8.6	0.9

GENERAL DISCUSSION

This thesis reports the first ever study on the sequential development of type I and type II ostertagiasis in young cattle over a two year fattening period. During this period the effects of the disease on performance was assessed and compared with calves in which the disease was controlled or absent. At slaughter, a comparative carcass evaluation study was also undertaken.

During the two grazing seasons the climatic pattern was fairly typical for the area, with the exception of a rather prolonged dry period in the summer of 1982. However, the dry period did not affect the expected normal course of the disease in the control animals. Thus, type I ostertagiasis, complicated by moderate infections with C. oncophora, occurred in late August and necessitated their treatment with the anthelmintic levamisole. Concommitant infection with lungworm was also treated. Following these treatments the majority of the larvae ingested became arrested in development at the EL_4 stage, as usually occurs during autumn, and at housing in October the sacrificed calves had high burdens of arrested larvae.

In contrast, a group of cattle given the MSRB bolus at turnout showed no clinical signs of ostertagiasis and the animals sacrificed at housing had only low worm burdens thus confirming the prophylactic value of the MSRB (Jones, 1983). However parasitic bronchitis also occurred in this group and required specific anthelmintic treatment confirming the lack of control by the MSRB over this parasite (Armour et al, 1981). A third group of cattle which were given anthelmintic treatment at 14 day intervals had zero worm burdens as indicated by consistantly negative faecal egg counts and normal plasma pepsinogen values. These differences in levels of infection were reflected in the liveweight gain performance of the different groups such that the control calves gained 0.55 kg/d compared with 0.70 kg/d overall for both the MSRB treated and the clean group of calves. During winter housing the animals of all the groups were clinically normal for the first 17 weeks when similar weight gains of about 0.6 kg were noted when the calves were given a normal winter store diet of hay and concentrate. In the 17th week some of the control animals showed marked softening of the faeces and three developed severe diarrhoea, typical of type II ostertagiasis, necessitating the slaughter,for humane

reasons, of one of the control animals. The sudden onset of type II ostertagiasis after four months of housing was again typical of what has been seen in the field by Anderson et al (1965) and produced experimentally by Armour and Bruce (1974) and completely different from the apparent field and experimental situation reported by Michel, Lancaster and Hong (1976 a,b) in which they found asynchronous development of arrested larvae. In the current study it appeared that the development of a large proportion of L_4 larvae was highly synchronous, clinical, biochemical and parasitological data pertaining to calf number 83, thus the onset of the disease was rapid and at post-mortem the worm burdens were predominantly adult indicating that the EL_4 had matured over a short period. The reasons for the differences between the Weybridge and the Glasgow data are not definitely known, however there can be no doubt that the explosive nature of the type II disease development also agrees with descriptions given by Anderson, Armour et al, (1965,6), and Bruce (1974) in Scotland; Williams et al (1980) in the United States; Raynaud and Bouchet (1976) in France; Wedderburn (1970) in New Zealand and Anderson (1971) and Hotson (1967) in Australia; Smith and Perreault (1972) in Canada.

It should also be pointed out that the Weybridge work was based solely on post-mortem worm counts with no sequential clinical examination of the animals. The current experiment is, in fact, the first description of the sequential development of type II ostertagiasis.

As might be expected from the low or negative worm burdens at the end of the grazing season there was no clinical disturbance to the animals given the MSRB bolus or to the clean animals and weight gains during the 17-29 week period of the type II phase were 28, 25 and 21 kg/hd for clean, MSRB treated and control animals respectively. A full discussion on the reasons for the rather low LWG during this time is given in Section B of the thesis.

During the second grazing season there were no clinical signs of ostertagiasis in any of the animals despite the presence of larvae in numbers up to 8,900/kg DM on the pasture paddocks grazed by the MSRB and control animals indicating that these animals had acquired a good immunity which was not interfered with by the two treatments with the MSRB device at the start of each grazing. The high degree of acquired immunity to Ostertagia was confirmed by the presence of very low burdens at post-mortem at the end of the second grazing season,

compared to the the very high burdens in first year animals grazing an adjacent paddock. The almost total absence of Cooperia L_3 on the pastures and worm burdens in the host indicated that a very high degree of immunity to this parasite had been acquired after the first grazing season. Of course the immune status of the clean animals could not be ascertained as they received no larval challenge from the pasture. Liveweight gain performance of the three groups was also similar during this period.

The high degree of immunity acquired confirms the only other study conducted on this topic by Armour et al (1979). Although it is unlikely that the clean cattle would have an immunity since there are literature reports of ostertagiasis in helminth-naive adult cattle, e.g. Selman et al (1975); Bailey (1978); Petrie et al (1983). Undoubtedly, the most important aspect of this is the fact that the MSRB bolus, whilst controlling clinical ostertagiasis in both years, permitted sufficient larval numbers to develop on the pasture to stimulate good immunity.

The parasitological and biochemical parameters used to monitor the helminth status of the cattle fully reflected the described course of events. Thus, in the first grazing season, the faecal egg counts of the control calves were positive after three weeks grazing and remained so throughout that season reaching a peak coincident with the development of type I ostertagiasis. The eggs deposited in the first half of the season being responsible for the marked increase in pasture larval counts of Ostertagia and Cooperia which occurred from August onwards. In contrast the egg count of the MSRB treated cattle remained negative until week 5 and so the accumulation of pasture L_3 larvae was delayed and at a greatly reduced concentration. Only very few larvae were recovered from the pasture grazed by the clean animals; the source of these being unknown. Even during the period of clinical type II in the winter housing, there were few eggs recovered from either the MSRB treated or the clean steers.

In the second grazing season, egg counts were again low although larval counts of up to 8,900/kg DM were recorded in the control plots, as is reported in part three of this thesis. The reason for this is not entirely clear. While it might reflect the control exerted by the MSRB bolus in the second year animals, it may also be a reflection of the ubiquitous nature of the Ostertagia parasite and the obvious difficulty in predicting when pastures would be free from infection.

In this context it is interesting that the author has noted burdens of ostertagia in excess of 5,000 in naive calves grazed for one week on pastures vacated by cattle some 18 months previously and not grazed subsequently (Bairden. Personal communication).

Biochemical Parameters

Plasma pepsinogen levels of the control calves during the first grazing season paralleled the course of infection with Ostertagia larvae and rose to a level of 6.0 I.u. during the clinical type I phase and remained elevated until housing in October. A very high correlation existed between the numbers of active Ostertagia (L_5+ adults) and the plasma pepsinogen values of the individual animals at necropsy indicating the usefulness of this measurement in the diagnosis of type I ostertagiasis and directly relating to the potential degree of abomasal dysfunction and damage. After housing and the removal of pasture larval challenge the pepsinogen values steadily reduced to a value between 2 and 3 I.u. by the month of December and then remained steady until the beginning of February at which time some increase was first noted. Coincident with the onset of the type II disease there was a sharp upturn in the activity of the pepsinogen, peaking at a value of 7.0 I.u. by the end of the somewhat extended housing period in May.

In the second grazing the pepsinogen values of the control cattle fell continuously from turnout to a mean plateau level of 2-3 I.u. in the middle of the summer. Subsequently in the housing period prior to slaughter, the plasma pepsinogen decreased to a level only a little higher (1.2 I.u) than that of naive calves. In the MSRB group the trend of plasma pepsinogen levels was similar albeit at a very much lower level than in the controls. Values in the clean animals were normal. Clearly from these results the plasma pepsinogen levels provide a much better indication of clinical ostertagiasis, be it type I or type II, compared with faecal egg counts. Furthermore, the correlation between plasma pepsinogen and the numbers of active parasites but not L_4 was very high in the animals slaughtered in the type I disease. Unfortunately it was not possible to make the same correlation at the end of housing in the type II phase since only one animal from each group could be slaughtered. However, data was available from a separate field outbreak of type II in a group of nine Friesian steers on which it was possible to perform regression studies

when these animals were killed. Again the correlation between plasma pepsinogen and active worms was high (r^2 62%) but the correlation with L_4 was again low (r^2 17%). Also most importantly, the correlation of gastrin and active worms at slaughter was very high (r^2 80%), whereas the relationship with L_4 was virtually nil (r^2 2%).

In view of these findings it is difficult to understand why the plasma pepsinogen test has not been more universally accepted for the diagnosis of ostertagiasis. There are, however, two possible reasons for this attitude; one is the dogma expressed by state authorities that the 'normal' value of plasma pepsinogen is below 1.0 I.u.. In the present study, which is the first time pepsinogen levels have been measured throughout episodes of type I, pre-type II and type II ostertagiasis in the same animals, it is quite clear that once exposure to Ostertagia has occurred and the larval challenge continues, albeit at a relatively low level, the 'normal' plasma pepsinogen value is in excess of 1.0 I.u and indeed in animals in their second grazing season it may exceed 2.0 I.u.. Clearly, there are uncertainties in the use of this test in animals in this category, but if the grazing history and the level of larval challenge are taken into account together with the absence of clinical signs, the test is still of value.

Obviously, if the plasma pepsinogen value could be supplemented with another indicator of abomasal dysfunction due to the presence of developing adults and related to the reduction in gastric acidity, it would be very useful and for this reason the levels of plasma gastrin were evaluated concurrently with the pepsinogen assays. This parameter was chosen as it is known that gastrin secretion by the G cells increases in response to increased abomasal pH such as that induced by an Ostertagia worm burden in sheep (McLeay et al, 1973). The correlation coefficient between gastrin and plasma pepsinogen, the multiple correlation of adult worm burdens at necropsy to gastrin and plasma pepsinogen function were very high, giving cause for extreme optimism for its future use as a diagnostic tool, particularly as the graphic trend of the the two assays were generally similar. However, before plasma gastrin, either alone or in conjunction with plasma pepsinogen, could be recommended as a routine clinical biochemical assay, further studies on other factors which might influence gastrin levels, such as larval challenge and change in diet will have to be

investigated in depth.

The other parasitological/pathological finding which is worthy of general comment is the mucosal index. This index which is based on the weight of the abomasal mucosa over the liveweight of the animal was significantly increased in cattle with severe ostertagiasis. The fact that the differences between infested controls and the MSRB treated cattle at the end of the first grazing season were maintained in the animals slaughtered at the end of the second season, despite the low worm burdens and insignificant sesions present in all groups of cattle, suggests that the mucosal index is a good indicator of both current and previous Ostertagia infection.

Production, metabolism and carcass studies

Clearly significant superiority was reported in the liveweight gain performance at grass in each of the two summer periods of the bolus treated cattle over the control animals and also sometimes the clean steers which had been routinely dosed with fenbendazole. During the grazing periods there was no prolonged enforcement of feed restriction in that the experimental design permitted the movement of both the control and the bolus treated cattle groups to fresh grass in the alternate plot as soon as the grass supply dwindled in any one group. Nevertheless the MSRB treated cattle, particularly later in the first season, often were somewhat short of available grazing and their potential for growth expression may have been greater than that recorded. The clean group of cattle only became short of grass at the very end of the first grazing and had to be supplemented with hay for two weeks before housing.

However, during the first winter period the feed supply to the three groups was strictly limited to a fixed store ration and then this was further limited to the intake of the control group of cattle when the appetite fell during the clinical type II phase in February. Again full potential for growth via an ad-lib access to feed in the unaffected cattle was deliberately not permitted by the nature of the experiment design since one of the pricipal objectives was to examine differences in feed digestibility and nitrogen retention between these groups of cattle at the same level of feed intake. Thus the pair-feeding control practice for the metabolism studies had been extended to the group feeding of the animals in this period. The poor performance of all cattle in the last few weeks of winter housing after

the type II occurrence may partly and jointly be attributable to; the enforced partial feed restriction (mainly a hay reduction), the increase maintenance cost of the heavier animals and the nature of the commercial concentrate containing the large grit-size particles of limestone of unknown dietary availability. Complaint to the feed company resulted in an acceptance of responsibility for the contribution to the early demise of the steer with the ulcer and the acknowledged poor performance of the other cattle. Compensation was paid for the loss of the steers and a refund on the 5% inclusion value of the concentrate.

No compensatory growth was seen in the control group of cattle which suffered both type I and type II ostertagiasis in any of the post-disease periods i.e. during the first winter in the so called pre-type II phase, or at grass and final fattening following the type II occurrence. However this phenomenon was observed in both winter periods in the clean cattle which received fenbendazole every 14 days, at grass, but not during housing. Clearly caution must be used in attempting to produce a simulated parasitologically clean and otherwise 'normal' group of grazing cattle in this way since growth was apparently depressed (particularly in the middle to late stages of grazing) even though an apparently good supply of grass was available. The results of Jorgensen et al (1978) showed previously infected cattle to exhibit compensatory growth at winter housing when offered a high energy diet ad-libitum. However, the magnitude of the burdens of arrested larvae present at housing were not known. In the current work the appetite of the control animals was often depressed, as in the type II phase of the disease, and also in the first winter the cattle were restricted to a store diet.

With respect to the production results from the two consecutive grazings it might appear that the second grazing did not attain the same output as the first season since calculation of the liveweight gain produced per hectare of grazing (LWG kg/ha) in the first season amounted to 515, 706 and 530 for clean, MSRB treated and control animals respectively, compared with 450, 480 and 403 for the same three groups in the second season. However, it should also be borne in mind that stocking rate differences are misleading if only the numbers of head/ha are considered. For example, during the first season 16 calves each grazed a 2.4 ha allocation in each of the MSRB and control groups, whereas this was decreased to only nine steers on the same plots in the

second season. The carrying capacity in terms of kg animal/ha, however, was greater in the second year, i.e. taking the mean liveweight of the steers at the start of each grazing;

1982 16 head x 140 kg = 2.24 t and

1983 9 head x 350 kg = 3.15 t.

Also the maintenance cost of the second year animals was considerably greater than that of the first year calves again further reducing the potential for liveweight gain production. Considerable concentrate supplementation (0.2 t/hd) was needed in the late second season, nevertheless, within any one year, clear superiority of production was demonstrated for the MSRB cattle. It also cannot be argued that pasture differences may have had an effect with the control and the MSRB groups since half the grazing was interchanged for the second season.

The digestibility and nitrogen (N) retention studies demonstrated a lowered digestibility of the whole diet and clear disturbances in the N retention of the housed control cattle during the winter period between the two grazings. This is the first known report of such studies being conducted with cattle throughout a pre-type and type II occurrence of ostertagiasis since the earlier work of Parkins et al (1982 a,b) recorded digestive efficiency and balance results from cattle following a type I occurrence at grass for only the first 18 weeks of housing (i.e. the pre-type II phase) and no further. Also as a first report, is the novel use of a dual pair-feeding system, operated throughout the 52 individual digestibility studies, where the exact intake of both the hay and concentrate components of the store diet consumed by the control animals were given to the paired steers in the clean and MSRB groups on the following day. Such a system has, of course, been used many times in nutritional studies conducted with sheep infected with parasites (e.g. Abbott, 1983).

In the pre-type II phase increased faecal N output reduced the apparent protein digestibility in control cattle, but in the clinical type II phase, where reduced intakes were noted, an increased urinary N output severely reduced the overall N retention. The results of comparative work in which pair-fed animals were used is summarised in a review by Topps (1983). Sykes and Coop (1976, 1977) confirmed the lower N retention balance observed in their infected animals by showing a lower content of protein in the carcass. The increased faecal N output is due either to an extra leakage of endogenous nitrogen into

the gut or a lower true digestibility of nitrogen (which unfortunately the simple input-output technique cannot measure). The increase in urinary loss would indicate a lower efficiency of utilization of absorbed nitrogen. Also an increase in urea synthesis, irreversibly lost in the urine, can also be derived from the ammonia produced by gut tissue amino acid deamination. However, no marked increases in plasma urea were noted in the current experiment during the type II phase.

The carcass evaluation studies conducted here would confirm the digestibility and N retention evidence in the demonstration of the decreased muscle masses of the indicator rib joints and even in the physical measurement of the leg circumference of the carcass. The acute type I visually produced 'wetter' muscles in the indicator joints and the combined effects of type I and type II ostertagiasis ultimately resulted in a smaller carcass, which did not grade, mainly on a 'lack of conformation' basis and these findings are in agreement with those of Sykes and Coop (1976, 1977) who, with sheep, found that the percentage of water in infected animals was higher than normal and that the deposition of fat, protein and skeletal minerals was considerably lower than in worm-free controls. This thesis clearly supports the contention of Topps (1983) that "carcass quality can be affected adversely by parasites".

The carcass evaluations at both stages were a useful exercise in attempting to quantitatively measure the effects of bovine ostertagiasis on production. Ultimately it is the yield of lean meat, irrespective of other interesting anatomical differences, which is economically important to the livestock industry. At the present time the economic penalty incurred by the control cattle can be simply calculated. There are of course many complex costing systems which might generate somewhat different figures. The liveweight difference overall of about 35 kg below that of the best performance group, the MSRB treated animals, is valued at £38.50 alone in the live animal. Further, the loss of grading meant that no EEC Beef Premium Scheme subsidy was payable which currently is valued at about £50/hd for animals of this weight. And further still, the poor finish of the carcass, with a known poorer yield of lean meat, would always mean a retail at a market price below that payable for quality carcasses. This loss of income is extremely variable.

However, a per head deficit of £85-plus is the known monetary loss for the mean control animal which failed to grade compared with

the mean animal of the MSRB group. Further accounting should include the cost of necessary medication which the control animals received as a result of the disease and also the virtual write-off value of the one animal which had to be destroyed *in extremis* at grass, and perhaps also a considerable part of the value of the steer destroyed after the type II occurrence, with the gastric ulceration. Proper account has also to be taken of the commercial cost of the MSRB bolus.

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