THE RELATIONSHIPS OF NUTRITION AND PARASITISM

A Thesis

by

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INTRODUCTION

It has long been recognised that there is a close relationship between parasitism and nutrition although its true nature has not always been clear.

Some of the relationships have been discussed in the reviews published by Lapage, (1937), von Brand, (1952), Hunter, (1953) and Smith, (1955).

In the association as seen and described by field workers, there is likely to be some confusion of cause and effect because, in the human field, areas where parasitism is most serious - in the backward tropical and sub-tropical countries where overcrowding, lack of sanitation, poverty and illiteracy are the rule, both under-nutrition and malnutrition are almost universal.

Under such circumstances, parasitic infections are frequently multiple, infection rates are high and at the same time, individuals may suffer the effects of one or more dietary deficiencies.

The reason for much of the difficulty is obvious when it is realised that many of the main symptoms of parasitic infection such as retarded growth, loss of vigour, loss of appetite, anaemia and dropsy, are also seen in malnutrition when the intake of certain dietary factors is sub-optimal or in undernutrition when the diet is low in energy.

In the field of animal husbandry also confusion is cormon because parasitism and malnutrition are likely to occur together on properties where management practices are faulty.

In recent years many attempts have been made to determine what essential dietary factors affect the resistance of hosts to particular parasites. It has been shown quite clearly that inadequate diets, especially those lacking vitamins, minerals and high quality protein, tend to render the host more susceptible to parasitism. In the undernourished host, the parasites may establish themselves with greater certainty, grow more rapidly to maturity, reach a greater size, live for a longer time and produce more progeny. They tend to produce more severe symptoms in the host and after their removal recovery is delayed.

Several different aspects of the association come into play independently or in combination. Parasites require certain food materials from the gut contents of the host for their own nutrition but are quite independent of host dietary sources for other materials. The diet of the host may have a direct effect on the nutrition of the parasite in respect of some of its requirements at least. In addition, the nutritional state of the host may have other effects on the parasite which reflect its resistance to invasion which stems from the vigorous health enjoyed by a well nourished body or by other indirect effects the nature of which are at present not

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thoroughly understood.

In addition, diet may have effects due solely to physico-chemical conditions in the gut which create an environment which is unsatisfactory for the parasite and which may result in its failure to establish or its elimination from the host. This effect may be seen quite apart from the presence in the food of recognised anthelmintic substances whether administered mixed with the diet or occurring naturally in the foodstuffs.

Where an immune reaction plays a part in the hostparasite relationship, the role of the diet may be an indirect one through its effect on the efficiency of antibody production which is known to be modified by the quality of the diet particularly in regard to protein and certain vitamins.

Several reports have been published indicating that heavy parasitism results in less efficient food utilization and several hypotheses have been advanced to explain it. It has been suggested that the diarrhoea that is such a common symptom in many parasitic infections, results in reduced absorption of nutrients from the food ingested, simply because of the rapid rate at which it passes through the regions of the alimentary tract where absorption takes place. It has been suggested also that stimulation and irritation of the mucosae by the parasites results in greater gut activity and consequently greater energy consumption leading to lowered efficiency of food

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utilization. Another suggestion has been that as the parasites must protect themselves from digestion by the host's enzymes, they themselves secrete antienzymes, which may be produced in sufficient quantity to have a depressing effect on digestibility especially of protein.

Some evidence has been produced indicating that mineral balance is disturbed in heavy parasitic infections, although much of it is conflicting.

The effect of parasitism on appetite is probably one of the most important factors causing the liveweight changes seen in severe helminthiasis.

Many papers have been published attempting to outline the role of deficiencies of various essential nutrients in increasing the host's susceptibility to parasitic infection and disease resulting therefrom. The following section attempts to review the more important papers published on these aspects of the subject.

HISTORICAL REVIEW

Total Nutrients

Chandler (1932) in studies with <u>Nippostrongylus</u> <u>muris</u> concluded that diet had little or no effect on the acquisition or retention of infection as long as the rats were not harmed by it. No differences were seen in rats on balanced diets, or those rich in carbohydrate or protein. On a diet of coarse fruit and vegetables on which the rats lost weight, the infections were heavier and lasted for a longer time.

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Shorb (1933) studied the host-parasite relationship of <u>Hymenolepis fraterna</u> in rats and mice. He found that a diet of white bread and water reduced the resistance of rats to the mouse strain and it also reduced age resistance. In mice, the worms did not reach maturity on this diet even when it was started one day after the infection was given.

Foster and Cort (1931, 1932, 1935) carried out trials with dogs that were repeatedly infected with the hookworm Ancylostoma caninum while on a normal diet. They were then transferred to a generally deficient vegetarian diet which rendered them more susceptible to the parasite. Fatal cases showed a terminal breakdown of resistance with a sharp rise in daily egg production over the last two weeks. The resistance of young dogs was broken down more rapidly than was the case with older animals. The diet which caused a marked body weight loss in all animals resulted in a lower resistance to invasion and establishment of the worms, an increased rate of development and increased egg production per female. In addition the deficient host showed a decreased ability to compensate for the injurious effects of the worm infestation. When the deficient dogs were transferred to the high plane diet, the egg counts dropped, the worms were eliminated and the dogs were then resistant to reinfection.

They showed that a deficient diet also increased

the susceptibility of both dogs and cats to nonspecific strains of hookworms but even on a deficient diet they could not infect dogs with the human hookworm, Necator americanus.

Ross and Gordon (1933) showed that dietary deficiencies similar to those that may occur frequently in the field in Australia under unfavorable seasonal conditions, in which protein and minerals were low, resulted in the breakdown of the resistance of sheep of all ages to <u>Haemonchus</u>. They suggested that such a result might only occur when the deficiency was severe enough to be reflected in marked loss of weight.

Taylor (1934) carried out experiments with three pairs of lambs fed on different planes of nutrition. One group was fed hay and concentrate, one hay alone and one straw alone. All lambs were given 40 doses of 500 mixed larvae and were killed 10 weeks later. The group fed straw harboured $3\frac{1}{2}$ times as many worms and passed 5 times as many eggs as the high plane group, and the hay group was intermediate in position. In a second trial, a hay group and a hay and concentrate group were fed over a seven month period. Even in the absence of reinfection, the low plane group showed a fivefold increase in egg count while that of the high plane group fell to one ninth of its original figure.

Kauzal (1934) studied the effect of a nutritional déficiency on the resistance of lambs to infection with Dictyocaulus filaria. He fed two 7 month old lambs, previously infected and subsequently shown to be resistant, on a diet made up of equal parts of wheaten straw and oaten chaff, for a period of 3 months. This diet was low in both protein and phosphorus and the animals lost weight over the experimental period. When they were exposed to an artificial infection, there was little or no effect on the resistance as the resultant infection was light and short-lived. A repeated attempt to infect the same animals after a further 2 months on the deficient diet showed that they were still highly resistant.

Frazer, Thomson, Robertson and George (1938) kept two groups of 20 sheep on high and low planes of nutrition. They infected each group with equal numbers of larvae and examined the abomasa at slaughter. The results showed marked differences in the establishment of the worms in the two groups, the high plane group harbouring an average of 2 <u>Haemonchus</u> and 11 <u>Ostertagia</u>, and the low plane group an average of 81 Haemonchus and 229 Ostertagia.

Lucker and Neumayer (1947) carried out tests with 8 lambs, four of which were infested with the hookworm <u>Bunostomum trigonocephalum</u>. From the age of two months, half of each group was kept on a high plane ration of alfalfa hay and concentrates, while the other group was fed timothy hay, a ration deficient in protein, calcium, iron, vitamins and energy. The two low plane infested animals died of anaemia whereas on the high plane the infested animals recovered after becoming moderately anaemic. The low plane control animals did not become anaemic. The infections depressed appetite, but only after the anaemia and unthriftiness had become marked. The poor diet affected both the resistance and the ability to compensate for blood loss.

Gordon (1948) has reported on the effect of two different diets on the development and maintenance of resistance to Haemonchus contortus infection in sheep. Nine 18 months old sheep were placed on a high plane ration of lucerne chaff 60, oats 20, and linseed meal The daily intake per head was 1.82 lb of feed con-20. taining 0.93 lb starch equivalent and 0.31 lb digestible protein. Ten similar animals were placed on a low plane ration of wheaten chaff 50, oats 30, maize 10, and bran 5. The daily intake on this ration amounted to 1.56 lb of feed containing 0.74 lb starch equivalent and 0.16 lb digestible protein per head. After a period of three months each sheep was dosed with 10,000 infective larvae of Haemonchus. On the high plane diet the infections were all light or negligible, and were thrown off an average of 19 days after the eggs appeared in the faeces, while on the low plane diet, four infections were light, six were moderate to very heavy. Three animals died of acute Haemonchosis, and, in the survivors, the infection was not thrown off until an average of 57 days after the

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eggs appeared in the faeces.

Observations on infections with <u>Trichostrongylus</u> spp. and <u>Oesophagostomum columbianum</u> in sheep on the same diets showed that in the case of the former the infection was lost more rapidly on the high plane diet while in the case of the latter parasite the results were less striking but the egg counts increased more on the low plane diet. (Gordon 1949).

Whitlock (1949) discussed the relationship of nutrition to the development of the Trichostrongylidoses in sheep. He drew a distinction between primary and secondary disease. The primary disease was one caused by a parasite infesting a host maintained on a diet capable of supporting maximal growth. He considered that Haemonchosis alone fell into this category. All other infections were considered to be secondary since they only produced symptoms of disease in the presence of dietary or other stresses.

Laurence et al (1951) in South Africa carried out an experiment to study the relation of nutrition to parasitism in merino sheep. Two groups of twenty lambs, 7-8 months old, were fed a high plane ration of veld hay, lucerne hay and 200 g maize, and a low plane ration of veld hay, lucerne hay and 50 g maize. Half of each group was infested with 8,000 larvae of <u>Haemonchus</u> contortus and 1,600 larvae of Oesophagostomum columbianum. All the infested animals died from acute verminosis. A similar infestation in 10 - 11 months old animals caused only chronic verminosis. Later the maize portion of both rations was doubled. The high plane groups and the uninfected groups were superior in liveweight, appetite for hay, and fleece weight. In the case of the haemoglobin the trend was the same except that the high plane infected group had a lower figure than the low plane uninfected group, a situation that might be expected from the vigorous blood sucking activities of the adult <u>Haemonchus</u>. The above differences were fairly obvious ones in spite of the fact that there were only four animals in each of the subgroups.

White and Cushnie (1952) carried out a field trial on the effect of a dietary supplement on the parasite infection of hill sheep. Two groups of ewes were used and they were grazed on a hill pasture during the day, returned to a small field during the night where one group was fed a supplement of one part of oats and two parts of linseed cake, allowing 1 lb of the mixture per sheep per day. Parasite burdens were followed by means of weekly egg counts over a two year period. Observations were also made on their lambs until they were 18 months old. The results showed that the supplement gave an increase in fleece weight but no significant differences in body weight nor in the faecal egg counts of the ewes. The lambs of this flock on the other hand showed reduced faecal worm egg counts, smaller seasonal rises in egg production, and greater body weights and fleece weights. Throughout the trial the faecal egg counts were low, the geometric mean never rising above 700 e.p.g. and at no time were there any clinical signs of helminthiasis. Total worm counts were done on the lambs after slaughter and on a small sample of the ewes but differences between the groups did not reach significant levels owing to the wide variation within the groups.

It has been noted by the authors that the cost of the supplement was about twice the value of the increased wool and mutton produced, but economies in the amount of supplement used, either by feeding it only over the winter months, or by using it more for the growing rather than the adult sheep might alter the picture. It is difficult to see how a significant difference could arise in the fleece weights without a difference appearing in the body weight of the ewes. The true picture might have been revealed if the clean scoured as well as the greasy fleece weight had been determined, since the difference might be seen in the yield due to greater quantities of wool grease in the supplemented animals. A further complicating factor in this type of trial is that the supplemented animals probably graze less than those not supplemented, as the former could satisfy their appetite with a smaller grazing effort. This situation at once introduces a difference in exposure to infection, even though the same area is grazed by each group during the day.

Mayhew (1945) made six attempts to break down an established immunity to <u>Haemonchus contortus</u> in calves by placing the host animals on a very deficient diet of cotton seed hulls. The criterion he used was a rise in egg count after reinoculation. A slight rise occurred only in three out of the six animals in spite of the fact that most of the calves became very weak and in some cases a supplement was necessary to prevent death from starvation.

Starvation

P.P. Levine (1938) confirmed Wetzel's (1932) observation that in the case of <u>Davainea proglottina</u>, there was a daily rhythm of segment discharge in the faeces and he showed that this was related to the feeding habits of the chickens. It occurred between 5-9 p.m. in the summer, but between 2-4 p.m. in the winter months, and it could be altered at will by changing the feeding times of the birds. Under optimal conditions worms produced one segment every 24 hours but by feeding a ration of low nutritive value, segment production was reduced or suspended altogether. Starvation for a period of 24 hours resulted in reduced segment production over the next week but later, normal output was resumed.

Hager (1941) found that in the case of <u>H. diminuta</u> in rats, a reduction of food intake to one third of appetite did not result in any reduction in the number of eggs produced.

The effect of starvation on the poultry cestode <u>Raillietina cesticillus</u> has been studied by Reid and Ackert (1941) and Reid (1942). They found that short periods of starvation of 24-48 hours caused destrobilization of the cestode but that the scolex was unaffected even by starvation periods as long as 20 days. Sudden changes in diet could also cause it. The glycogen content of the worms was reduced to about 10 % of the normal figure by starvation for 20 hours and they thought that destrobilization followed from loss of activity on the part of the parasite.

Later, Reid (1945) studied the effect of starvation on the roundworm, <u>Ascaridia galli</u>. After starving for 24 hours some of the worms were expelled, and after 96 hours, 70 % of the birds were free of parasites. Between 75 and 90 % of the glycogen stored in the parasites was used in a period of 48 hours.

Larsh (1947) showed that mice that were debilitated by the administration of alcohol were more susceptible

to infection with Hymenolepis fraterna. Non-alcoholic mice whose food intake was reduced to the same level as that of the alcoholic mice, were equally susceptible. It was concluded that the effect of the alcohol was through a secondary avitaminosis brought about by the lowered food intake. Larsh and Fletcher (1948) found that alcohol could break down the immunity of mice to T. spiralis under certain circumstances. There was no effect on muscle invasion nor was there any effect on the immunity that followed a single infection or more than 5 infections. An effect was seen however when 2 - 5 infections were given. This might be due to the fact that the additional doses might give a more solid immunity that could not be broken down. Larsh and Kent (1949) found that alcohol had no effect on the natural immunity of mice to T. spiralis even in cases showing extreme debilitation. Similar results were obtained with a single stimulating dose, but by using repeated stimulating doses alcohol had a marked effect on immunity. Alcoholic mice showed greater numbers of both adults and larvae than the controls, and the burdens were even higher if the alcohol was given before as well as after the stimulating infections.

Larsh (1950) found that thiouracil in young mice failed to affect their resistance to <u>H. fraterna</u> but thyroid extract so reduced the resistance of old mice that they harboured more worms than young mice that were dosed to test the viability of the eggs used in the experiment. It is possible that this may occur through a vitamin depletion at the higher metabolic rate.

Read (1951) has examined the effect of crowding in cestode infections in rats and has calculated the 2/3relative surface area per worm, (weight /weight) and plotted it against the number of worms present. In infections of over 8 worms the index was proportional to the logarithm of the number of worms present. He considered that a similar situation appeared to hold for the data published by Reid for <u>R. cesticillus</u>, Chandler for <u>H. diminuta</u>, and Shorb for <u>H. nana</u>. Stunting could be explained by a need for oxygen which may be obtained through close contact with the mucosa and this contact would be limited in heavy infestations.

Vitamin A

Among the factors affecting resistance to infection Vitamin A has received most attention and appears to be the nutrient that has given positive results most frequently and in the widest variety of host-parasite systems. This fact might have been expected because of the role of vitamin A in the maintenance of healthy epithelial tissues and of its proven function in resistance to bacterial infections, particularly those of the respiratory system. Hiraishi (1927) found that he was able to infect only one out of thirty pigs with <u>Ascaris lumbricoides</u>, but when placed on a diet deficient in vitamin A, he was able to infect 89 % and 100 % in two different series. The infections occurred before any signs of avitaminosis were apparent. In depleted pigs the clinical signs of infection appeared earlier, were more severe, and lasted for a longer time.

In 1928 Hiraishi reported on a series of experiments carried out by Koino and added the results of some observations of his own. In Koino's experiments 57 pigs were used and were infected with human and pig Ascaris while the pigs were fed diets adequate or deficient in vitamin A. The results are given in the following table.

Normal diet	10 pigs	Human strain	Negative
Normal diet	20 pigs	Pig strain	19 Negative, l(?)
Vitamin A de- ficient	9 pigs	Pig strain	l Negative, 8 positive
Vitamin A de- ficient	6 pigs	Human strain	6 Positive
Vitamin A de-) ficient) Cod liver oil)	6 pig s	Pig strain	6 Negative
Vitamin A de- ficient	6 pigs	Nil	5 Negative, 1 positive

In Hiraishi's own experiment 9 pigs were used. On a diet with adequate vitamin A the worms did not develop beyond the liver and the lungs but on a diet low in vitamin A the pigs could be infected before any signs of avitaminosis were evident. On the deficient diet the effect of the worm infection appeared earlier and was more prolonged.

De Boer (1935) on the other hand, found that by repeated infection he was able to infect pigs with both the human and the porcine strain of <u>Ascaris lumbricoides</u>, suggesting that the two strains are physiologically identical. He found that a diet low in vitamin A did not facilitate infection.

Ackert, McIlwaine and Crawford (1931) showed that a diet lacking vitamin A lowered the resistance of chickens to infection with the nematode <u>Ascaridia galli</u>. The criteria that they used were the numbers of worms developing from an artificial infection and the length attained by them in the three weeks between infection and slaughter of the experimental birds. They considered that the effect might be due, in part, to reduced peristalsis and the greater bacterial populations in the bowel which would provide a greater food supply for the developing worms. In these trials three diets were used, a natural diet that was adequate in vitamin A, and a semi-synthetic diet, with and without added vitamin. The deficiency of the latter was such that symptoms of deficiency were produced in five weeks. The natural diet gave rise to a resistance that was greater than that seen with the semi-synthetic diet with added vitamin A.

Spindler (1933) found that vitamin A deficiency reduced the resistance of rats to <u>Nippostrongylus muris</u>. This finding has also been confirmed by Watt et al (1943) and by Riley (1943).

Clapham (1933) found that the amount of vitamin A in the diet of rats affected the fate of the larvae of <u>Parascaris equorum</u>. In deficient rats more larvae migrated in the lungs and their development was more rapid than on the normal diet. The same author was unable to show any effect of avitaminosis A in infections with <u>Heterakis gallinae</u> in chickens. She recorded differences in the number of worms present but they failed to reach significant levels.

Nagoya (1931) found that a diet low in vitamin A increased the susceptibility of dogs to the hookworm Ankylostoma caninum.

Wright (1935) observed that vitamin A deficiency increased the susceptibility of dogs artificially infected with <u>Toxocara canis</u> and <u>Toxascaris leonina</u>. Eleven deficient dogs harboured 2,674 worms (average 243), while nine control dogs on a normal diet harboured only 448 worms (average 50).

McCoy (1934) studied the effect of a vitamin A deficient diet on the resistance of rats to <u>Trichinella</u> <u>spiralis</u>. He placed the rats on the deficient diet at 25-27 days of age, and they showed a markedly lowered resistance as early as 2-3 weeks. There was a greater persistence of the adult worms in the intestine and greater numbers of larvae in the muscles. The deficient rats failed to develop immunity to a second infection whereas the control rats were completely immune. Rats that were placed on the deficient diet when they were half grown usually showed only a slight breakdown of resistance after 1-3 months even though the liver reserves of vitamin had been completely exhausted.

Larsh and Gilchrist (1950) found that a diet deficient in vitamin A did not affect either the natural or the acquired resistance of mice to <u>T. spiralis</u>. This finding is in sharp contrast to McCoy's results with rats, and the authors conclude that if mice need vitamin A for resistance to <u>T. spiralis</u> they do so at a much lower level than do rats.

Lawler (1941) studied the relationship of vitamin A deficiency to the immunity to <u>Strongyloides ratti</u> in white rats. He found that deficient rats were less resistant to primary infection but the effect failed to show up if the reserves were only partially depleted. The rate of loss of worms was not significantly lower in deficient rats. The deficiency resulted in a weakening of the immune response as shown by the number of worms established and the presence of embryos in the worms. His results are difficult to appraise as there were only five rats in each group and no statistical treatment of the data was attempted.

Shaw (1937) fed six wethers for 18 months on wheat straw, oats and beet pulp, a diet deficient in vitamin A. The treatment did not increase the susceptibility of the animals to experimental infection with the lungworm, <u>Dictyocaulus filaria</u>, although all animals showed symptoms of avitaminosis A, such as muscular incoordination, night blindness and urinary calculi.

Chandler (1942) in his studies on tapeworm nutrition concluded that <u>Hymenolepis diminuta</u> was independent of supplies of vitamin A in the diet of the host as the growth of the worm was normal in rats on a diet free of vitamin A.

Little work has been published on the effect of dietary deficiencies on trematode infections but Krackower, Hoffman and Axtmeyer (1940) working in Puerto Rico with artificial infections of <u>Schistosoma mansoni</u> in rats showed that, on a normal diet, the destruction of the parasites in the liver reached a maximum after 5-7 weeks, and that a few flukes still remained at the end of the experimental period of 32-115 days. On a vitamin A deficient diet the destruction of parasites was maximal up to 6 weeks, more dead worms were found after that time but still less than in the rats on the adequate diet. Vitamin A deficiency permitted the proliferation of fibroblasts and histiocytes but the resolution of a lesion made up largely of these elements was retarded, although the dead parasites were quickly digested. The regeneration of more specialised cells such as liver parenchyma was retarded on the deficient diet.

Vitamin A deficiency is of particular interest in relation to the lesions produced in the squamous stomach of rats, especially when infested with <u>Gongylonema neoplasticum</u>. The work of Fibiger (1913) on the experimental production of carcinoma of the stomach of rats by infection with <u>G. neoplasticum</u> is well known. A long controversy has raged over the years, as few, if any, other workers have been able to confirm Fibiger's findings. The position has been discussed recently by Hitchcock and Bell (1952) who have concluded, from the results of experimental work and a re-examination of some of Fibiger's original slides, that the lesions described by him were not neoplastic but were hyperplasia and hyperkeratosis of the squamous epithelium

of the forestomach. It occurred in both infested and non-infested rats on a vitamin A deficient diet. In the infested animals on the low vitamin A diet there was a marked epithelial disorganisation with extensive epithelial downgrowths, whereas in non-infested rats on the low vitamin A diet there was ulceration and an oedema and inflammation of the submucosa. Infested rats on the complete diet showed only minimal changes. Fibiger's "lung metastases" were probably metaplastic areas of respiratory tract epithelium, and his "lymph node metastasis" an epithelial inclusion or arrest. The authors conclude that all critical experiments using G. neoplasticum have failed to produce acceptable neoplastic growths; that the lesions described by Fibiger are seen in avitaminosis A; and that G. neoplasticum heightens the effect while, at the same time, vitamin A deficiency accentuates the effect of the parasite. It was pointed out that the diets of dried bread used by Fibiger were almost certainly low in vitamin A. Gongylonema acts as a chronic irritant and produces only minimal lesions in rats on a normal diet.

Vitamin B

Yeast

Ackert and Nolf (1931) showed that baker's yeast contained a factor that stimulated the growth of <u>Ascaridia</u> galli in chickens. The worms were longer in the chickens on the yeast supplemented diet than on a deficient diet or one that derived its B vitamins from other sources such as wheat, eats, corn, and meat. More worms were present on a diet of polished rice and casein than on the same diet with added yeast, or the diet of mixed grains and meat. The length of the worms was greatest on the yeast supplemented diet but there were no differences between the other two groups. The authors considered that vitamin B deficient diets give the worms a better chance of survival due to reduced peristalsis in the host's intestine.

Clapham (1933) states that the results obtained by Ackert and Nolf fail to reach statistically significant levels, but her reasons for saying so are not clear.

Baker (1941) has used brewer's yeast in calves, and Whitlock (1949, 1952) has used it in sheep in an advanced state of parasitic debility with a view to stimulating appetite.

Thiamin

Chandler (1943) who studied the nutritional requirements of the tapeworm <u>Hymenolepis diminuta</u> concluded that the worms were able to absorb nutrients directly from the host's tissues with which they were in close contact. The parasite was independent of the protein in the host's diet but was dependent on the carbohydrate. It was independent of vitamins A, D and E and Thiamin, but was affected by lack of vitamin G in female but not in male rats.

Watt (1944) found that a diet deficient in thiamin reduced the resistance of rats to a single dose of <u>Nippostrongylus muris</u>, and to a more striking degree, to superinfection when the test dose was given 12 days after the initial infection, or when the rats immunised by repeated infection were subsequently placed on the test diet and challenged after a period of 69 days. There was also reduced protection from the serum of rats on the thiamin deficient diet.

Chandler, Read and Nicholas (1950) studied the thiamin requirements of <u>H. diminuta</u>. Complete lack of thiamin in the diet of the host had no effect on the worms. The thiamin content of the worms was fairly constant irrespective of whether the host diet was deficient or whether the host received additional amounts of the vitamin parenterally. By the use of injected radio-thiamin it was shown that the worms derived their thiamin from the host. They pointed out also that oxygen may be the limiting factor responsible for the stunting of cestodes that is seen in heavy infections.

Riboflavine

Watt (1944) found that a diet deficient in

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riboflavine markedly reduced the resistance of rats to an initial infection of <u>Nippostrongylus muris</u>, or to superinfection when the test dose was given 12 days after the initial dose of larvae. The protection afforded by hyperimmune serum was reduced when the donor rats were on a diet low in riboflavine.

Vitamin G

Hager (1941) studied the effect of dietary modifications on the tapeworm H. diminuta in rats. She found that a diet low in vitamin G resulted in a lowered egg output. She considered that the effect was probably a direct one since other diets that were detrimental to the host, as shown in changes in body weight and appetite, did not affect the egg production of the She found that there was no direct relationship worm. between the number of worms present and the number of eggs produced, but there was an inverse one between the number of worms and the number of eggs produced per worm. This effect was probably due to crowding. In heavy infestations the worms are shorter and may fail to produce eggs.

Chandler (1942) showed that <u>H. diminuta</u> was affected by a deficiency of vitamin G in female but not in male rats, and assumed that there was a relationship of some kind between vitamins and hormones. However Beck (1951) found that a diet deficient in vitamin B complex resulted in a reduction in the number of tapeworm eggs passed in the faeces of female rats after 30 days, but in the case of male rats a similar drop did not occur until they had been on the diet for 90 days.

Addis and Chandler (1944) studied the vitamin requirements of H. diminuta in rats. In three experiments the rats were subjected to different periods of depletion, namely, 0, 4, and 17 days. Six female rats in each group were infected with 10 cysticercoids each. The numbers of worms were reduced on diets deficient in fat soluble vitamins or vitamin G. There was no effect on a diet deficient in thiamin. The lower establishment on diets low in fat soluble vitamins might have been due to a disturbance of bile secretion through a failure of stimulation by bile salts for the evagination of the scolex. The authors concluded that the presence of some factor associated with vitamin G brought about a stunting of the worms, whereas a lack of the mixed fat soluble vitamins A, D, and E, or thiamin, resulted in an increase in size. This may have been due to decreased peristalsis resulting in the worms establishing in a more anterior and therefore more favorable position in the intestine.

In a later paper Addis and Chandler (1946) examined

the problem further and concluded from trials with experimental diets containing synthetic vitamins that the factor associated with vitamin G that stimulated the growth of <u>H. diminuta</u> in female rats was none of the eight known factors, nicotinic acid, pyridoxine, pantothenic acid, riboflavine, biotin, p-amino-benzoic acid, inositol, choline or folic acid. The component was heat stable. Liver extract reduced the stunting by one half.

Folic acid and vitamin B12

Keith et al (1949) and Sadun et al (1949) reported on experiments comparing the infections of <u>Ascaridia</u> in chickens that were fed purified diets with and without folic acid and a commercial diet. They showed that chicks on the deficient diet harboured more and larger worms, indicating that the deficiency of folic acid lowered the resistance. They found that the worms on the purified diets were stunted when compared with those on the commercial diet. The stunting was not overcome by dried liver, but liver extract gave a partial response. The commercial liver extract therefore contains a factor that is required by the worms in relatively large amounts, and this factor may be vitamin B_{12} or the "animal protein factor."

Todd et al (1950) compared the resistance of the offspring of hens that were fed six different rations,

three based on soybean meal and three on corn gluten, supplemented with whitefish meal or vitamin B_{12} . Differences in the weight gains in the chickens after parasitisation were taken as the criterion of resistance. Soybean and whitefish gave the best growth and corn gluten and whitefish the poorest growth. Vitamin B_{12} supplements were without effect. The differences noted were significant at the 5 % level.

Hansen, Norris and Ackert (1953) studied the resistance of chicks fed an all plant diet supplemented with "Aurofac" and vitamin B12. The birds were artificially infested with Ascaridia galli. The supplemented groups had a lower mortality, parasitism increased the mortality on both diets, and fewer chicks on the supplemented diet became infested. There were no differences in the weight gains of the infested and control birds on the basal diet, but on the supplemented diet it approached significance at the 1 % level. The authors suggest that this might be due to the fact that at peak efficiency a few worms might have an effect greater than that produced by larger numbers of worms in birds growing more slowly on the basal diet. The supplemented chicks had fewer worms but the sizes of both male and female worms were the same on both diets.

In 1954 Hansen, Petrie and Ackert compared supplements of Aureomycin and vitamin B12 used separately and together in an all plant basal ration. Half of all groups were parasitised, with <u>A. galli</u>. The highest infestion rate and mortality occurred on the basal diet. The supplements together stimulated both the infested and control birds. There were very small differences in feed efficiency with some decrease due to parasitism. Aureomycin alone and with vitamin B_{12} reduced the number of worms. Aureomycin restricted and vitamin B_{12} stimulated worm growth, and when used together the effect on worm size was cancelled.

Maldonado and Asenjo (1953) studied the effect of supplementing a deficient diet with vitamin B₁₂ and folic acid, separately and together, on <u>Nippostrongylus</u> <u>muris</u> in rats. Neither substance affected the course of parasitization or the number or the size of the worms.

In the case of infestation with the fish tapeworm, <u>Diphyllobothrium latum</u>, in man, only a few of the cases infested develop megaloblastic anaemia. Von Bonsdorff (1948) has shown that the anaemia develops in those cases in which the worm is situated high up in the small intestine. The symptoms can be relieved either by removing the worm, or by administering vitamin B_{12} parenterally without anthelmintic treatment. Many attempts have been made to isolate a toxin from the worm, but without success. It now appears that the dried worm is curative, being a rich source of the vitamin. Presumably the worm living high in the intestine absorbs the vitamin in competition with the host.

Vitamin D.

Ackert and Spindler (1939) in four experiments with chickens found that a deficiency in vitamin D, severe enough to cause marked leg weakness, failed to have any effect on the resistance to A. galli. In one trial cod liver oil was used, and an observed effect on the number of worms and the number of infested birds may have been due to the vitamin A present. In one trial, irradiation from an ultra-violet lamp failed to produce any effect on resistance. They stated that vitamin D deficiency had an effect in rendering the birds less resistant to the effects of parasitism, but this conclusion is not justified from their data, as uninfested control birds on both diets were not included for comparison and the difference in growth rate after infection might have been solely a diet effect.

Chandler (1942) found that the tapeworm <u>H. diminuta</u> was not affected by the amount of vitamin D in the diet of the host rat.

Wantland (1934, 1938) showed that the administration of irradiated ergosterol after an artificial infection of <u>Trichinella spiralis</u> resulted in a more rapid calcification of the cysts, the acceleration being greater in the rabbit than the rat.

Vitamin E.

Zaiman (1940) used a synthetic diet deficient in vitamin E. After six months on the diet, experimental guinea pigs were infected with <u>T. spiralis</u>. The deficient animals harboured more than twice the number of muscle trichinae as those fed a standard ration. Vitamin E was chosen for this study because of the known effects on voluntary muscle, the tissue invaded by the parasite in question.

Carbohydrate.

Chandler (1942) in his studies with <u>Hymenolepis</u> <u>diminuta</u> in rats, found that the tapeworm was dependent on carbohydrates in the host's diet and was sensitive even to partial restriction. In a later study, Chandler, Read and Nicholas (1950) found that starch was a better source of carbohydrate than dextrose, and that dextrose was better than sucrose. Read (1950) discussed the gastro-intestinal tract as a habitat for parasites and reviewed the work of previous workers on the effect of milk in the diet of the host, and assumed that lactose was the constituent responsible for the effects reported. There appears, as yet, to be no sound basis for this opinion.

Protein.

In their classical study of the effects of dietary

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factors on the health of two African tribes, Orr and Gilkes (1931) brought forward evidence of large differences in the parasitic infections between the Masai, who were predominantly eaters of milk, meat, and blood, and the Kikuyu, who were mainly vegetarian. The incidence of helminthiasis as a percentage of total cases among outpatients was 0.1 % for the Masai, and 7.1 % for the Kikuyu, and among the inpatients of five native hospitals was 0.8 % for the Masai, and 10.3 % for the Kikuyu. The diets of the two tribes differed in many respects but the difference in protein was the most striking one.

Ackert and Beach (1933) compared the effects of protein supplements of meat and skim milk, meat alone, and peanut meal and skim milk, added to a cereal ration that was adequate in minerals and vitamins, on the resistance of chickens to <u>Ascaridia galli</u>. The skim milk and meat supplement was superior to the other two groups as measured by the number of worms developing after artificial infection and by the size of the worms when the birds were killed three weeks after infection. It was considered that the difference was due to the greater variety of amino acids provided by the supplement.

Ackert (1939) found that as a supplement to an otherwise adequate cereal ration peanut meal was inferior to soybean meal. The latter had a greater variety of amino acids and a greater digestibility, that of the soybean being about 76 % as compared with 30 % for peanut meal. In a later paper, Ackert, Brandson and Ameel (1943) found that soybean meal was as effective as meat scrap, or meat scrap and skim milk, in producing resistance to Ascaridia in chickens.

In 1950 Reidel and Ackert compared supplements of soybean meal and skim milk, and soybean meal and meat The soybean meal and skim milk gave the greatest scrap. resistance to Ascaridia based on the number of worms developing after artificial infection, although the lengths of the worms in the two groups were not significantly different. They considered that the difference was due to the greater range of amino acids especially of lysine and tryptophane. The same authors (1951) compared three diets of different protein content, one containing 11 % protein of plant sources, one containing 18 % from both plant and animal sources, and one containing 24 % mainly from animal sources. The first diet was low in all amino acids except tryptophane, whereas the third diet was low in argenine, glycine, lysine and leucine. The second diet gave the greatest resistance as measured by the number of worms developing, although the differences in length of the worms were not significant.

Reidel (1950) found that diets containing 0.7 % and

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Todd et al (1950) found small differences in resistance, as measured by the growth rate of artificially infested chickens which were the offspring of birds kept on diets based on soybean meal and corn gluten supplemented with whitefish meal and vitamin B12.

Hansen et al (1953) compared the resistance of birds on an all plant basal diet with and without supplements of Aureomycin and vitamin B_{12} . The supplemented chicks had a lower mortality rate, parasites increased the mortality on both diets. There was no difference between the weight gains of infested and control chicks on the basal diet, but on the supplemented diet differences approaching significance at the 1 % level were recorded. It was suggested that at peak efficiency a few worms had a greater effect than a larger number of worms on the basal diet. Supplemented chicks had fewer worms but there were no differences in the sizes of either male or female worms.

Luttermoser and Allan (1942) compared the growth rates of chickens with and without infections with <u>Raillietina cesticillus</u>, on diets containing 13 % and 26 % protein. No difference in growth rate was recorded on the high protein diet, but they claim that there was a retardation in the growth of the infested birds on the low protein diet. The authors presented no statistical treatment of their results, but an inspection of the growth curves would suggest that the differences observed might not be significant. The differences noted amount to less than 10 % of the weight and the curves ran almost parallel for the greater part. Such a result could occur with ordinary errors of weighing on the first weighing day. Further, with a real difference one might expect a progressive divergence between the two curves.

Donaldson and Otto (1946) compared infections with <u>Nippostrongylus muris</u> in rats on a 9 % all plant protein diet and a 22 % protein diet, half of the protein being of animal origin. Rats on the low protein diet were unable to develop a highly protective immunity. Immune rats lost their immunity after 36 days on the low protein diet, although they still retained some immunity when compared with previously uninfected rats. Rats on the high protein diet were also more resistant to primary infection.

Venkatachelam and Patwordhan (1953) found in children a lower digestibility of protein associated with an infection with <u>Ascaris lumbricoides</u>. On low protein intakes they suggested that this factor might be important in the causation of nutritional oedema.

Corkill (1950) has reviewed the relation of dietary

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protein to disturbed host-parasite balance. He points out that there is a continuous interaction between serum protein, body protein and diet. Serum gamma globulin has a half life of about two weeks. He considers that a poorer antibody response may occur when there are two or more antigens competing for the available gamma globulin especially when the diet is low in lysine. Lysine is quantitatively the most important amino acid in gamma globulin and is also lacking in the vegetable diets of tropical countries where multiple infections are common. A poor antibody response in subnutrition is well known and in many infections there is a reduction in serum albumen. Since there is more lysine in albumen than in globulin, this is thought to be the mechanism by which limited lysine supplies are spared for antibody production.

Minerals

Iron

Rhoads et al (1934) carried out a study in Puerto Rico, based on a clinical study of 83 patients with hypochromic anaemia associated with hookworms. In the same area a similar hypochromic anaemia occurs not associated with hookworms. It was thought to be due to the low level of iron and other haemopoietic substances in the poor diet, to gastro-intestinal changes as well as to blood loss from the hookworms

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present. The removal of hookworms produced little change in the haemoglobin level but the erythrocyte count did improve over several weeks. The addition of meat and milk to the diet likewise had little effect on the haemoglobin. On the other hand, the administration of 0.6 g ferric citrate by mouth, with or without the removal of the parasites, had a striking effect on the anaemia and on the clinical condition of the patients. Only a moderate effect was obtained with liver extract. They pointed out that it is important to direct therapy first against the anaemia and secondly against the parasites.

Foster and Landsberg (1934) showed that the anaemia of hookworm disease was a microcytic hypochromic type and that it could be simulated haematologically by repeated bleeding. It responded well to iron therapy but there was no evidence of any toxic action on the haemopoietic tissues.

Otto and Landsberg (1940) found that dogs fed a generally deficient diet, with or without added iron, failed to survive the anaemia produced by an infection of 900 Ancylostoma larvae. Iron stimulated a transient erythropoiesis but had no effect on the development or persistence of the hookworms. They considered that a balanced diet was of more importance than iron and they doubt the validity of the conclusions reached by Rhoads et al. McFadzean and Wong (1952) found that there was a quicker response when iron was given intravenously than when given orally. The amount given was calculated to make up the haemoglobin, plus 50 % for an unknown deficiency of non-haemoglobin iron. It was given daily as saccharated iron oxide in doses of 50, 100 and then 200 mg per day. The oral treatment was 1.5 g ferrous sulphate with 150 mg ascorbic acid daily. The patients were discharged when their haemoglobins reached 60 %. There were 25 patients and controls. The quicker recovery that followed treatment allowed hospital beds to be vacated earlier. The response was about twice as rapid as with oral dosing. The worms were removed with an anthelmintic when the patients were discharged.

Spindler (1936) artificially infested anaemic pigs, on a diet low in iron and copper, with <u>Oesophagostomum</u> <u>dentatum</u>. Higher infections were present in the animals that were supplemented with both iron and copper. He suggested that the supplement may have favoured the parasites or that the anaemia in the unsupplemented pigs was detrimental to the worms. His conclusions are of doubtful significance because of the small number of animals used in the trials.

Phosphorus and Calcium

Phosphorus and calcium have been inadequate in many of the generally deficient diets previously discussed.

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Only a small number of trials have dealt with deficiencies of these elements alone.

Ackert and Gaafar (1949) found that a deficiency of phosphorus was the limiting factor in Ascaridia infection in poultry. A high calcium-low phosphorus diet produced smaller worms than one in which the calcium:phosphorus ratio was optimal. An excess of calcium injected into birds fed on a normal diet had no effect on the parasites. In a later paper Gaafar and Ackert (1953) found that diets low in calcium of phosphorus gave fewer and smaller worms, but this was thought to be due to the mineral requirements of the worms themselves. The resistance of growing chicks did not appear to be affected by low calcium or phosphorus in the ration.

Shearer and Stewart (1933) concluded that heavy nematode infections in lambs interfered with the metabolism of calcium and phosphorus and this would result in retarded skeletal growth. Andrews (1938) failed to find any interference with calcium or phosphorus storage in lambs artificially infested with <u>Cooperia curticei</u>, as did Andrews et al (1944) in the case of Trichostrongylus. Franklin et al (1946) on the other hand didfind some evidence of impaired calcium and phosphorus utilization.

Manganese

Harwood and Luttermoser (1938) considered that on a diet low in manganese the growth of chickens appeared

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Copper

In the peat areas of New Zealand there has been a long experience of gradual reduction of production of dairy cows. On one occasion a field worker demonstrated worms in the stomach of a cow that had died of the disease, and recommended a copper sulphate anthelmintic drench. A definite response was obtained but subsequent work by Cunningham (1946) has shown that the soils and pastures are deficient in copper and that the effect of copper sulphate drenching was to supply the missing mineral rather than to kill the parasites.

There is some field evidence suggesting that parasitism is a somewhat greater problem in young cattle on soils low in copper. The infestations appear to be more severe and more persistent, but definite proof of any relationship has yet to be furnished.

Cobalt

It has often been suggested that there is an increased susceptibility to parasites in lambs grazed on cobalt deficient pastures although definite proof of this fact has still to be produced. Illustrative of some of the difficulties that were encountered in the

early days in New Zealand is the unpublished report of some observations made by W.M. Webster in the Morton Mains district of New Zealand. Farmers in the area reported a wasting disease of lambs, and investigation associated high worm counts with the unthriftiness. The presence of numerous swampy areas on one farm suggested that parasitic infestation was taking place in the moist pasture surrounding the swamps. It was recommended that the sheep could be protected by fencing off the offending areas. This was done, and the following season little or no mortality occurred. It was subsequently shown by Askew and Dixon (1936) that the primary cause of the disease was a cobalt deficiency and, since cobalt topdressing resulted in thrifty lambs, the interest in parasites as a contributing factor waned. The cobalt status of the soils at Morton Mains, however, is marginal so that there is considerable variation in the severity of the symptoms from year to year. To this fact may be attributed the apparent response of the lambs following attempts to limit the intake of larval parasites.

A recent experiment in the same area, reported by Anderson and Andrews (1955), provides evidence suggesting that there is a greater response to the anthelmintic use of phenothiazine in lambs on cobalt deficient pastures than there was when a cobalt supplement was supplied. The differences both in weight gain and in mortality

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between phonothiazine treated and control lambs were not significant so the part played by the parasites in contributing to the syndrome of cobalt deficiency remains uncertain. The lack of response to phenothiazine in the cobalt dosed groups indicates that parasites were not an important factor in limiting the growth of the cobalt dosed lambs.

Digestibility

Stewart (1933) at Cambridge carried out observations on lambs that were heavily infested with gastro-intestinal nematodes. He concluded that the parasites prevented the host from digesting protein to the full extent. Ether extract and N-free extractives were not affected, while crude fibre digestion was reduced in very heavy infections. Shearer and Stewart (1933) found that the same animals showed an interference with calcium and phosphorus metabolism, but that sodium and potassium metabolism was normal. Stewart (1933, b), in an attempt to explain the above observations, showed that, in vitro, extracts of the parasites contained a substance that inhibited proteolytic action. It was considered that the action of the inhibitor in vivo was to protect the parasite from the proteolytic enzymes of the host's digestive fluids, and that in heavy infections they were produced in sufficient quantity to depress protein digestion in the gut of the host.

One criticism of the work is that the animals that were used were taken from the field where their heavy natural infection with parasites was assumed to be the cause of their unthriftiness. Such an assumption can not be made without risk since the unthriftiness may have preceded the parasitism, which might, therefore, be only a secondary factor. Rumen function, for example, might be impaired and might improve while the animals are under experiment and, at the same time, although independently, the parasite load may decrease either through the development of resistance or as a result of reduced re-infection rate. A drop in egg count and an improvement in digestibility may well be correlated although the association may be entirely fortuitous.

Andrews (1938) carried out paired feeding experiments with 8 sheep that were artificially infested with <u>Cooperia curticei</u>. There were no differences in the digestibility of protein or fibre. The infections produced were clinically non-pathogenic but the infested lambs showed a decreased efficiency of food utilization apparently due to increased energy metabolism. This may have been accounted for by irritation of the gut or by a possible although not demonstrated accumulation of guanidine in the blood.

In a later paper Andrews, Kauffman and Davies (1944) studied the effect of T. colubriformis on the digestibility of nutrients and again failed to find depressed protein or fibre digestibility or decreased absorption of calcium or phosphorus. In one animal the apparent digestibility coefficients of all nutrients, except ether and crude fibre, were depressed when the vitality of the animal was so reduced by starvation and diarrhoea that the normal digestive processes could no longer take place. In another animal the bones were demineralised to carry on metabolic processes. Energy metabolism in all infected sheep was increased and the efficiency of food utilization reduced. This may have been brought about by inflammation and irritation of the gut. In those showing diarrhoea it may have been due to increased intestinal activity and the retention of guanidine as a result of the severe dehydration.

Franklin et al (1946) carried out digestibility trials on weaned lambs artificially infested with <u>T. colubriformis</u>. The infested lambs showed a lower liveweight gain and a depression of protein digestibility. The liveweight differences were not significant, although they did approach it at the 5 % level. The differences in protein digestibility were significant in one period at the 5 % level, and in another at the 1 % level, but the difference was not significant in the period that corresponded with the period of greatest egg production. Venkatachelan and Potwordlan (1953) examined

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9 children harbouring Ascaris. They were kept in hospital on a diet allowing a constant amount of nitrogen per day. Faecal nitrogen before and after anthelmintic treatment was determined. In all cases faecal nitrogen was lower after treatment. The average values were 1.35 g per day and 0.755 g per day respectively - a difference that was significant at the 1 % level. The nitrogen content of Ascaris eggs was determined and was 19.2 - 26.1 mg in a total faecal nitrogen of 0.875 - 2.47 g. Two worm-free patients were given the anthelmintic treatment and catharsis but showed no fall in faecal nitrogen. The authors concluded that, on a low protein diet, impaired protein digestion may be an important factor in the cause of nutritional oedema, a syndrome which is seen primarily in young children as a subacute protein deficiency.

Rogers (1941) has studied the metabolism of trichinosed rats during the early phase of the disease. Anorexia and diarrhoea occurred 8 - 12 days after infection. Protein digestion decreased to a low figure 8 - 12 days after infection. He suggested that this could be due to the presence of antiproteases and to mechanical damage to the mucosa of the gut by the adult worms. There was no difference in fibre digestion. In the intermediate phase of the disease there was a minor disturbance about 30 days after infection which may have been due to the adults leaving the gut. Inorganic phosphorus excretion fell to a very low figure 4 - 8 days after infection, 24 days later it rose to 2 - 3 times the normal figure. Calcium assimilation fell to a low level during the second 4 day period, and again at the 9th. Weight loss was greatest in the 8 - 11 day period. (Rogers 1942).

Bray (1953) on the other hand made a study of the nitrogen metabolism in West African children. She was able to find no consistent change in the nitrogen balance after the elimination of Ascaris or Ancylostoma. There were however data from only 3 treated individuals with infections with each parasite.

Anthelmintic Factors in the Diet.

Hall, Shillinger and Cram (1925) have reviewed some of the early literature on the presence of anthelmintic substances in certain foodstuffs, but there is little or no scientific evidence in support of the claims made for them. The factors mentioned include pumpkin seeds, blueberries, pepper, tobacco, raw sugar, an antidiabetic diet and onions. The authors carried out tests with raw onions in the diet of dogs over a sixty day period and found little or no anthelmintic activity. Some whipworms, hookworms and ascarids were passed but, as they point out, their elimination may have been spontaneous and not related to the diet. Milk

Porter (1935) carried out a trial with 3 - 6 rats in each group with diets of either dried or reconstituted milk with and without added iron and copper. The rats on the milk diet showed a greater susceptibility to <u>Nippostrongylus muris</u> than those on a balanced grain ration. The greater susceptibility was seen both in primary and repeated infections. Added iron and copper appeared to increase the resistance but this group was intermediate between the other two. The author presented no statistical treatment of the results of these experiments.

Reidel (1949) fed a normal diet and water or milk to mice which were then infected with 100 Trichinella larvae. There were no differences in the number of adults present when the mice were killed at 6 days, but when they were killed at 28 days marked differences were found in the larvae that had developed. The variation was larger in the group fed milk, presumably due to variations in intake.

Ackert and Reidel (1946) infested with <u>Ascaridia</u> <u>galli</u> chicks that were fed milk on alternate days. The milk-fed chicks harboured only 40 % as many worms as those given water.

Spindler, Zimmerman and Hill (1944) fed 14 pigs on skim milk for 3 - 5 days and found that they eliminated 60 - 100 % of the Trichuris, 0 - 90 % of the Ascaris, and 90 - 100 % of the Oesophagostomum present. The elimination was associated with a profuse diarrhoea. In a later paper Spindler and Zimmerman (1944) carried out two trials under conditions of reinfection, with groups of pigs fed milk plus grain each day or milk for one day every 2 - 3 weeks, and a control group fed a grain ration. Purgation occurred with the feeding of the milk and with it was associated the elimination of parasites. Autopsy showed low infestations with Ascaris, Trichuris and Oesophagostomum, Spirurids and lungworms. The effect of a milk diet on lungworms is difficult to explain unless it is an indirect one.

Shorb and Spindler (1947) repeated the same feeding regime with two virtually worm-free pigs per group. The liveweight gains were 1.31, 1.13 and 1.15 lb per day, whereas under conditions of reinfection Spindler and Zimmerman obtained gains of 1.17, 0.85 and 0.32 lb per day. The differences noted in the weight gains in the earlier trial were then considered to be due to the parasites primarily and to the diet secondarily.

Whitlock (1951) carried out an experiment under conditions of pasture grazing with concentrate feeding in creeps, in which lambs that were weaned at 2 months of age were found to be more susceptible to parasites than those that were not weaned until 5 - 6 months of age. It is doubtful if the egg count differences between the groups were significant. It is doubtful also whether the effect observed should be ascribed solely to the absence of milk from the diet since the early-weaned lambs would be forced to do more grazing than the unweaned lambs and consequently would be exposed to a greater risk of infection.

A further examination of early weaning under more practical conditions has been attempted in New Zealand. Clarke (1954) has shown that similar worm infections occurred in lambs that were weaned in early December as in those weaned 5 weeks later. However early weaning has a very considerable advantage from an economic and flock management point of view. The main features are the more economical utilization of feed through grazing by the lambs, reducing as it does, ewe overhead and feed conversion losses, by the reduced competition with the ewes for the available pasture, and the ease with which the ewes can be handled after weaning in order to reduce their condition for sale or for flushing prior to mating. Early weaning facilitates pasture utilization, weed control and shearing procedures.

Mackerras (1953) found that a milk diet suppressed <u>Plasmodium bergei</u> infections in rats. The infection disappeared in 23 - 27 days in rats on the milk diet while two animals on the control diet succumbed. No effect was seen in infections with Trypanosoma lewisi.

Sen Dutta and Ray (1955) reported that a milk diet had no effect on <u>T. evansi</u> infection but it did affect mammalian plasmodia but not trypanosomes, <u>Babesia canis</u>, or, avian plasmodia. They suggested that it might be due to a deficiency of para-aminobenzoic acid, folic acid, or cortizone.

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Anthelmintic Factors in Pasture

There is a high concentration of hydrogen cyanide in a number of pasture plants and Melville et al (1940) have shown that some strains of white clover contain up to 0.033 %. Tetley (1953) has considered the possibility that this may be toxic to internal parasites in sheep grazing such pastures. He showed that, in vitro, potassium cyanide, in similar concentrations, killed some of the infective larvae of the Trichostrongyles from sheep. An aqueous extract of clover leaves was also toxic to some of the larvae immersed in it. He grazed lambs on worm-free pastures of (1) New Zealand certified pedigree white clover which was high in the cyanogenetic glucoside, lotaustralin (0.023 % hydrogen cyanide), (2) red clover with a doubtful trace of cyanide, and (3) cocksfoot with no trace of glucoside. After four days on the pasture all animals were dosed with 2,000 infective larvae of Haemonchus contortus. Fourteen days later the lambs were killed and the number of worms

present determined. Mean counts of 509.6 on the white clover, 613.9 on the red clover, and 698.6 on the cocksfoot were obtained. The cocksfoot group was significantly higher than the other two groups, but there were no significant differences between the two clover groups. It is doubtful therefore whether the cyanide content was the only factor involved.

Southcott (1955) has reported from Australia the results of some observations on the effect of grazing oats on the adults of <u>Oesophagostomum columbianum</u> in sheep. He found that sheep grazing oats continuously eliminated many adult parasites. Pastures of <u>Phalaris</u> <u>tuberosa</u>-subterranean clover and red clover resulted in some elimination, but the effect was much less than with oats. On oats, not all of the parasites were expelled and there was an associated softening of the faeces and a fall in faecal pH. It was seen to occur with and without large increases in body weight. There also appeared to be some inhibition of emergence of larvae from the nodules during the oat-grazing period.

Self-aure.

The term "self-cure" was first used by Stoll (1929) to describe a sudden spontaneous elimination of parasites from an infested host. Stoll's observations were made with two sheep infested with <u>Haemonchus contortus</u>, and similar observations have since been made with other host-parasite systems by Sarles (1932) with <u>Tricho-</u> <u>strongylus calcaratus</u> in rabbits, Gordon (1948) and Stewart (1950, 1953, 1955) with <u>Haemonchus contortus</u> in sheep and Michel (1952) with <u>Trichostrongylus</u> retortaeformis in rabbits.

Some early observations on the phenomenon were made by the writer at the Regional Pastoral Laboratory, Armidale, New South Wales, in 1940. At the time, routine collections of faeces were being made from several flocks of sheep, and it was observed repeatedly that there was a dramatic fall in faecal egg count, not only in individual sheep, but sometimes in virtually all members of a flock of 30 - 40 animals. The change from high to low counts occurred between two weekly sampling days. On some occasions large numbers of adult Haemonchus were found in the small "grab" samples of faeces removed from the rectum. More detailed later work by Gordon (1948) showed that the phenomenon could occur simultaneously in sheep of different ages, in sheep with infections of different ages, in sheep with heavy or light infections, and in sheep in different flocks situated several miles apart and grazing pastures of very different botanical composition. It could occur in the field on pasture, but not in members of the same flock recently removed to pens and fed a constant diet. It was not a manifestation

of an immune reaction as sheep could show self-cure and almost immediately pick up a new infection which might prove to be fatal. Self-cure was often seen after rain and freshening of the pasture but before there was any marked improvement in the nutritive value of the pasture as reflected in liveweight changes. He postulated the presence of an anti-helminthic factor in the fresh pasture.

Subsequent work by Gordon (1949) and Stewart (1950, 1953, 1955) showed that the phenomenon was an allergic sensitization which was precipitated by a large intake of infective larvae. It was associated with a rise in circulating complement-fixing antibodies, eosinophiles and blood histamine, and with skin sensitivity. The behaviour of the abomasum observed under anaesthesia was one of increased motility with a striking oedema of the abomasal wall.

Stewart also showed that the larvae of Haemonchus could elicit self-cure against the adults of Haemonchus in the abomasum and also those of Trichostrongylus in the small intestine while the larvae of Trichostrongylus could elicit self-cure against adult Trichostrongylus but not against those of Haemonchus. The assumption was that the reaction occurring in the abomasum affected adults in the stomach and lower down in the small intestine, but in the case of Trichostrongylus the reaction did

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not occur until the larvae had reached the intestine and therefore could have no effect on adults living in the abomasum.

PARASITES OF CATTLE IN NEW ZEALAND

Unthriftiness, and mortality in their first year of life, is a serious problem of calf raising on many New Zealand dairy farms. The relative importance of the many factors involved has not been determined, but it is certain that coccidiosis, parasitic gastro-enteritis, and parasitic pneumonia all play a part, as does also general malnutrition and specific dietary deficiencies such as occur in areas low in available copper or cobalt.

Until recently all laboratory examinations for the diagnosis of parasitism in cattle have been carried out at the Wallaceville Animal Research Station, and data collected over several years have been examined in an attempt to improve the interpretation of the results of faecal egg counts, which at the present time, is beset with many difficulties. A study of these observations will also provide a background for the better appreciation of the experimental results presented in the following section on the effects of rotational grazing of cattle.

The specimens that were forwarded to the laboratory included a relatively small number of viscera for total worm counts and a larger number of faecal specimens for worm egg counts. The viscera were sent either unpreserved or formalinised. On arrival they were opened, washed and screened through a fine sieve. The material retained in the sieve was diluted to 2 litres, and four 5 ml aliquots were taken after thorough mixing. After the smaller worms were counted the remainder of the material was examined for smaller numbers of the larger parasites, such as Haemonchus and Bunostomum, before it was discarded. The contents of the large intestine was examined after screening through a coarser sieve (20 meshes per inch), and the worms present counted individually. The lungs were examined by opening the bronchi and estimating the numbers of lungworms present.

In the case of the worm count data, the numbers available have been too small to consider in detail but the following table (Table 1) presents some of the heavier counts from which tentative conclusions as to the relative abundance of the species of parasites present can be drawn. In many cases the lungs or the large intestines were not submitted so the data for these organs are incomplete.

It is obvious that the data presented in the table do not represent a random sample because of the selection that has been made from the records and the fact that more cases suspected of being heavily parasitised would be

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submitted by veterinarians in the field. It is true also that many clear cut cases of parasitism would not be submitted but there would tend to be more where the actiology was obscure or where there were complicating factors. It is clear also that there was not a Dominion-wide cover and that there was a preponderance of specimens from certain areas. Several factors contributed to this, such as proximity to the laboratory, the availability of good express services, the interest of the field worker concerned, and also the density of the cattle population. Another factor that limited the amount of material submitted was the bulky nature of bovine viscera, and unless suitable containers were available at the time of the autopsy either no samples or only inadequate samples were collected.

Specific identification of the parasites collected have shown that the species most commonly found in large numbers were <u>Ostertagia ostertagi</u>, <u>Trichostrongylus axei</u> and <u>Cooperia onchophora</u>. Associated with these but occurring in smaller numbers are <u>O. trifurcata</u>, <u>C. McMasteri</u> and <u>C. punctata</u>. Heavy infection with Haemonchus was uncommon and Bunostomum and Bosicola were relatively rare. In isolated cases, however, heavy infestation with the larval stages of Bosicola have been seen. <u>Dictyocaulus</u> viviparus was commonly associated with outbreaks of

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parasitic pneumonia in both calves and yearlings. In the case of the latter the field diagnosis, both clinically and on post-mortem examination, was so simple that specimens were seldom sent to the laboratory.

TA	B	E	1

Date	Locality	Haem.	0st. ('000)	Trich. ('000)	<u>Coop</u> . (1000)	Bun.	Trich- uris	Dicty.
April	Palmerston North	0	42	4 8	30	0	0	0
tt :	Pahiatua	0	47	0	0	0	0	0
11	Taumaranui	0	76	33	37	0	0	-
11	Ruakura	0	10	0	0	0	0	0
11	Taumaranui	0	68	52	22	0	0	-
May	Featherston	0	36	28	20	0	-	
Ŧŧ	Paeroa	134	16	8	14	0		-
June	Ruakura	0	11	2	7	0	0	0
tt :	Wanganui	0	54	63	3	0	1	30
11	Taumaranui	0	24	24	36	1	-	0
11	Trentham	0	116	93	84	0	0	0
July	Upper Hutt	0	125	16	0	0	20	0
Aug.	Whakatane	0	26	16	36	0	150	0
tt .	Whakatane	0	80	34	30	0	20	0
tt	Whakatane	0	22	32	5	0	40	0
17 .	Whakatane	0	15	0	68	0	10	0
117	Hamilton	0	20	14	18	0	0	-
11	Galatea	184	3	29	10	0	0	30
TT	Ruakura	0	48	8	4	0	-	0
Ħ	Hamilton	0	18	29	3	1	-	-
tt :	Taumaranui	0	36	56	0	37	0	
Sept.	Whangarei	0	24	14	16	17	0	
11	Whakatane	20	27	2	0	0	7	-
Nov.	Hamilton	0	67	10	0	0	-	***
11	Timaru	0	15	37	14	0	0	

The results presented provide evidence of a marked . seasonal occurrence of heavy worm infections in the autumn and winter months. It should be pointed out, however, that in New Zealand virtually all calves kept for herd replacements are born in July or August. Roberts (1941) has shown that in Queensland the pattern of worm infection was similar in calves born at different times of the year. It is probable therefore that in New Zealand the same is true, and that the apparent seasonal occurrence of heavy infection was a reflection of the fact that there was a population of young animals that all reached their most vulnerable age in the autumn months, rather than that the autumn months provided weather conditions most favourable for the development of the parasites on the pasture.

The faecal specimens were sent unpreserved and were examined using the McMaster technique. In most instances examinations were done within 48 hours of collection. Provided the sample bottles were filled completely, thus excluding air, the development of the eggs was arrested and specimens were suitable for examination under these circumstances for several days.

The analysis of the egg count data has been carried out by means of punched cards on which were entered the relevant information for each animal. The cards were then sorted to give a breakdown according to the year, month, locality, count, and the consistency of the faecal specimen. The vast majority of the specimens were from cattle under one year of age although there were some from older animals, but the records were so often incomplete in this respect that all specimens have been considered together. The intensity of the infection has been expressed as the mean egg count and also as the percentage of specimens with counts of above two arbitrary levels of 500 and 1,000 eggs per gram - representing moderate and heavy infections respectively. The latter figures may give a more reliable measure, since the mean egg count is influenced so much by a few very high values.

The following tables show the number of specimens examined over recent years, the distribution of the counts recorded, the seasonal and regional comparisons, and the relationship of count to the consistency of the faeces. Because of the small numbers of specimens examined in the early years the figures for 1941-1949 have been combined.

TABLE 22 Yearly Comparisons 1941-1954

Year	Samples Examined	Mean E.G.P.	% over 500	% over 1,000
194 1	7)		D.d.I.	Dec at
1942	6)			
1943	108)			
1944	51)			
1945	192)	290	20	12
1946	20 j			
1947	30)			
1948	144)			
1949	370)			
1950	714	170	8.5	3
1951	430	400	17	10
1952	312	400	20	11
1953	332	550	21	12
1954	582	450	20	12
Total	3,305	330	15	8

TABLE 3

	DISCIPIDUCION	Or COUNTRS	1941 - 1904
Eggs per	Gram	Frequency	Percentage of Total Samples
Negativ	7e	1,533	46
1 -	500	1,351	41
500 -	1,000	161	5
1,000 -	2,000	135	4
2,000 -	3,000	56	2
3,000 -	5,000	28	0.8
5,000 -1	L0,000	24	0.7
10,000 ar	ndover	17	0.5
Total		3,305	100

Distribution of Counts 1941 - 1954

TABLE 4

	Month	ly Compari	sons 19	41 - 1954	
Month	Samples Examined	Percent. of Total	<u>Mean Egg</u> Count	Percent. of Counts over 500	Percent. of Counts over 1,000
Jan. Feb. Mar. April May June July Aug. Sept. Oct. Nov. Dec.	182 199 231 216 305 355 346 465 253 287 285 181	5.5 6 7 6.5 9.5 11 10.5 14 7.5 8.5 8.5 5.5	240 320 400 570 350 650 290 310 190 120 120	12 14.5 17 22.5 24.5 16.5 24.5 11 8 7 5 8	$ \begin{array}{r} 6 \\ 6 \\ 9 \\ 12 \\ 14 \\ 7 \\ 5 \\ 14 \\ 5 \\ 4 \\ 2 \\ 5 \\ 1 \end{array} $
Total	3,305	100	350	15	8

TABLE 5

Seasonal Comparisons			1941 - 1954			
Season	Samples Examined	Percent. of Total	Mean Egg Count	Percent. of Counts Over 500	Percent. of Counts Over 1,000	
Spring (SeptNov.)	748	24.5	310	7.5	4.5	
Summer (DecFeb.)	534	17	330	12.0	5.0	
Autumn (MarMay)	721	23	500	21.0	12.0	
Winter (JunAug.)	1,060	33.5	430	17.0	8.0	
Total	3,063	100	350	15.0	8.0	

TABLE 6

North Island - South Island Comparisons 1941 - 1954

Number of Specimens	<u>Mean Egg</u> Counts	Percent.of Counts over 500	Percent.of. Counts over 1,000	2
North Island 2,871	350	15	7.8	ł
South Island 399	410	13.5	8.5	
Total				
3,270	350	15	8.0	



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TABLE 7

Regional Comparisons 1941 - 1954

Region	Number of Specimens	<u>Mean Egg</u> Count	Percent. of Counts over 500	Percent. of Counts over 1,000
Northland Auckland -	405	330	15	8
Waikato	1,085	420	16	9
Bay of Plenty	141	440	20	10
Taranaki	476	180	9	3
Wellington -				
Manawatu	753	350	15	7.5
East Coast	11	200	18	9
Westland	141	290	10	5
Nelson -				
Marlborough	103	350	15	8
Canterbury	76	780	20	16
Otago	34	180	9	6
Southland	45	450	14	10
Unknown	35	360	23	11.5
Total	3,305	350	15	8

TABLE 8

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Climatological Data

Region	Annual Rainfall (Inches)	Mean Summer Max. Temp.	Mean Winter Min. Temp.
Northland	50	75	46
Waikato	50	75	43
Bay of Plenty	50	75	40
Taranaki	65	70	43
Wellington -			
Manawatu	45	70	40
East Coast	35	75	40
Westland	100	65	36
Nelson -			
Marlborough	35	72	36
Canterbury	25	70	34
Otago	30	65	36
Southland	45	65	34

Since diarrhoea is a prominent symptom in many parasitic infections this point was examined in the present series. The consistency of the faeces was recorded by classifying the specimens into one of five categories according to the moisture content. The assessment was a visual one. By a further grouping into three categories, samples showing evidence of constipation, those which might be considered normal for calves grazing on pasture, and those where there was evidence of scouring, no marked difference was seen with the exception of the first group, but this was a small sample and no definite opinion can be reached.

TABLE 9

Consistency	Number of Samples	Percent. of Total	<u>Mean Egg</u> Count	Percent. of Counts over 500	Percent. of Counts over 1,000
Soft-formed Soft Very soft Diarrhoeic	31 1,578 560 539	1.0 43.0 17 16	700 330 430 380	32) 13 18.5 17	20 7 9 9
Very Diarrhoeic Not recorded	289 308	9 9	320 275	15 10.5	10.5 7
Total	3,305	100	350	15	8

Faecal Consistency Comparisons 1941 - 1954

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TABLE 10

Consistency Comparisons 1941 - 1954

<u>Consistency</u>	Number of Samples	Percent. of Total	<u>Mean Egg</u> Count	Percent. of Counts over 500	Percent. of Counts over 1,000
Constipated faeces (soft	t-				
formed)	31	l	700	32	20
Normal faeces	3				
(soft, very	soft)				
	2,138	71	350	15	7.5
Abnormal faeces (diarrhoeid) ,				
very diari	hoeic)				0.0
	828	28	360	15	9.2
Total	2,997	100	350	15	8.0

Two of the main difficulties met with in the interpretation of egg counts from diagnostic material are the facts that an ailing animal showing inappetence of several days standing will show an automatic increase in the number of eggs per gram due to the lowered faecal output, and, on the other hand, when diarrhoea is present the increased water content of the faeces effects a dilution of the eggs present, provided the dry matter content of the faeces is maintained. It is possible, however, for an animal with diarrhoea to show a falsely high egg count if the total faecal output is reduced through a lowered food intake. Smaller changes in the same direction can be introduced where the feed has a high digestibility, as in the case of young pasture or of root crops. The results showed a remarkable consistency from year to year, although there was one year, 1950, when the infestation was considerably lower than normal. Reference to meteorological data for that year shows that the rainfall over the early part of the year was very much below average, the winter was mild and the spring was a very favourable one. The low counts, therefore, would appear to be a result of the summer and early autumn drought preventing a build up of infection in the late autumn and winter months.

The seasonal comparisons showed a well-marked autumn peak and a spring trough. As previously mentioned it was not certain whether these effects were truly seasonal ones, or due to the age of the host.

The regional comparisons showed a complete lack of a north to south gradient in the severity of infection although 800 miles separated the most northerly districts from the most southerly ones. The combined figures for the two islands, which overlap to a very small extent, and vary considerably in climate and farming practice, failed to show a significant difference. On the other hand there appeared to be a well-marked west to east gradient occurring in both islands, with the higher figures to be found in the eastern and drier areas, although rainfall may not be the determinant. The results were consistent whether the mean count or the proportion of moderate or high counts was considered. The use of the chi square test showed that Taranaki and Westland had fewer high counts, while the Bay of Plenty and Canterbury had more high counts than the remaining areas, which showed no differences between themselves. Because of the small numbers involved no conclusions could be reached concerning Otago, Southland or the East Coast.

The position in other countries where conditions are not similar might be quite different. It is possible that the warm temperate to sub-tropical parasites, such as Haemonchus, Bunostomum and Bosicola, might play a more important part elsewhere, and the lack of a difference in New Zealand between the North and the South Islands might be due to the fact that the species concerned are mainly those of the cool temperate regions, and a much greater range of temperatures might be necessary for a difference to appear.

The real difference between the eastern and the western districts was an unexpected one if one assumed that high rainfall favoured the development of the free living stages. A possible explanation may be a difference in the plane of nutrition, possibly, although not necessarily, associated with the rainfall. Taranaki, for instance, is an area of high natural fertility, whereas the Bay of Plenty and Canterbury have a lower

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soil fertility, and also possess certain areas where deficiencies of minerals such as copper, cobalt and iodine are recognised, and such factors may affect the levels of infection. An alternative explanation might be that the higher rainfall resulted in a level of infection so high that, under the good nutritional conditions existing, a solid immunity was established with greater certainty than elsewhere. On the other hand, although the rainfall on the eastern side is lower than on the west, it may still be high enough to allow ample development of the larval stages, and the difference observed might be quite unrelated to rainfall. Another effect of heavy rainfall might be to wash infective larvae from the pasture into the soil so that they become available to the grazing animal in smaller numbers.

THE EFFECT OF A SYSTEM OF ROTATIONAL GRAZING ON THE NEMATODE PARASITES OF DAIRY CALVES.

For a number of years at the Ruakura Animal Research Station comparisons have been made between groups of calves raised under different systems of grazing management.

In each year one group has been raised under conditions of set-grazing which are very similar to those seen on many typical New Zealand dairy farms. The calves graze from the age of a few days, and are confined
to one small paddock near the milking shed, where they remain until weaning when the pasture conditions usually make a change of paddock essential. Pasture deterioration occurs under the highly selective grazing behaviour of the calves and when grass growth is vigorous the pasture becomes rank and the calves then refuse to eat the rough herbage. Grass contaminated with dung is unattractive so that the area grazed becomes smaller and smaller and is cropped very close to the ground. It has been estimated that under such conditions only 20 - 30 % of the pasture area would be grazed. In January the calves are run with the yearlings and dry stock, and the calf paddock is then left vacant.

On the other hand, the system of rotational grazing that has been developed has been shown to result in more satisfactory growth and a marked reduction in mortality. The system has been described briefly by Ballinger (1942), Gerring (1947), and McMeekan (1947, 1948, 1954). The system consists essentially of rotational grazing of the calves ahead of the milking herd. The cows graze the paddocks only after the calves have selected their ration and been moved on to the next area in the rotation. After the cows have eaten much of what remains, the dry stock - the yearlings and the non-lactating cows - are used to clean up the rough unpalatable grass left by the milking cows.

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It is obvious that adequate subdivision and watering facilities are necessary for the adoption of such a system of rotational grazing. It is obvious also that such subdivision will allow better utilization of pasture, better opportunities for the conservation of hay, ensilage and autumn-saved pasture, as well as resulting in an increase in total pasture production which normally follows an on-and-off type of grazing management. The maintenance of a uniform high plane of nutrition for the growing and producing animals is consequently greatly facilitated.

The effect of the system on productivity has been discussed elsewhere (McMeekan, 1954) but the following figures can be quoted to illustrate the magnitude of the differences between the set-stocked and rotationallygrazed groups.

		<u>T.</u>	ABLE 11	Body	Weights	on 31st	March
Season		Rotated	Set-sto	cked	Differ	ence	
1940-1941 1941-1942 1942-1943 1943-1944 1944-1945 1945-1946 1946-1947 1946-1947 1946-1947 1947-1948 1947-1948 1948-1949 1948-1949	Twins Twins Twins	416 1b 423 439 385 405 327 424 414 410 410 410 410 414 408 357	354 360 374 334 279 263 326 341 325 338 288 325 325 297	lb	62 63 65 51 126 64 98 73 85 72 126 83 60	lb	
1949-1950 1950-1951 1951-1952 1952-1953 1953-1954 1954-1955	TWINS	408 439 446 405 363 385	525 295 301 328 275 288		83 144 145 77 88 97		

At the average age of 20 months the body weight difference increased to about 140 lb.

In the early years of the trial, 1940/41 to 1944/45, rotational grazing was begun when the calves were weaned at 4 months of age and the pastures were grazed by the cows as soon as the calves were taken out of the paddock. The cows were then followed immediately by the dry stock, after which the paddocks were harrowed if necessary to spread the faeces and the area was rested until it was grazed again by the calves on their next round. Although grazing times and resting times varied throughout the season, depending on the amount of feed available and the number of paddocks that were shut up for hay and ensilage, in general the paddocks were grazed for 2 - 3 days and there was a period of about 10 - 14 days between successive grazing periods. While these figures are typical there were very wide variations depending on the rate of growth of the pasture, and the time taken for a paddock to recover after grazing.

In the later years, since 1945/46, the system has been modified slightly so that the calves begin rotational grazing at the age of 4 weeks and graze each paddock for 1 - 3 days, but the grazing by the calves falls about midway between successive grazings by the milking cows. Under this modified system there is some recovery

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of the pasture before the cows graze it. The periods between grazings are then usually about 7 - 10 days.

It is important that the cows and the calves should graze the same paddocks since the pasture that is unpalatable for the calves is more readily eaten by the cows and later the regrowth is again acceptable to the calves. This appears to be quite important since there are instances in the field where farmers have practised rotation through a series of calf paddocks without the use of adult stock, with a result not very different from that seen under the set-grazing system.

The results are less satisfactory when the calves follow too closely behind the cows, or when a weekly rotation is used. During dry periods ensilage and hay are fed but the rotation around the paddocks is continued. The calves soon become accustomed to the hay and they are kept on an ample hay ration after the autumn rains fall, so that the quickly growing grass is not consumed to any great extent until it has had time to mature. Material and Methods

In several seasons parasitological observations were made. Faecal samples were collected directly from the rectum of all the calves in the trial and were sent to Wallaceville where worm egg counts were done. The McMaster technique was used, taking a 4 g sample and using half saturated brine as the flotation fluid.

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Three areas of 1 sq. cm. were scanned so that each egg counted represented 33 eggs per g. of faeces. Most of the samples were of normal consistency and rarely did more than 20 per cent. of the samples show evidence of severe diarrhoea. It was considered reasonable, therefore, to ignore differences in dry matter content of the faeces and the dilution effect that might be expected to accompany scouring. Further, no attempt has been made to apply a correction for the increasing quantity of faeces passed as the calves grow older.

Statistical analyses have been carried out on square root transformations of the original number of eggs counted per slide, on each day that samples were collected. The results are presented in graphical form, each graph showing on a logarithmic scale the mean number of eggs per g. of faeces for each group. The level of significance of the differences between the group means is indicated on the graphs.

There were a number of variations in the experimental procedure in different years but, for the sake of clarity, only data from the set-stocked and the rotationally-grazed groups are considered. Details of the treatments in the seasons when samples were taken are given below.

1940/41: In the first season there were two

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groups of 14 and 15 calves. One was set-stocked and the other was rotationally grazed from weaning. The first examination for parasites was carried out on April 17, 1941, and at this stage the condition of most of the animals in the set-stocked group, and even in some of those in the rotationally-grazed group, was far from satisfactory. At this time both groups were run together and a small scale drenching trial was carried out, grouping all calves at random into 3 groups. One group was drenched with copper and nicotine sulphates, one with tetrachlorethylene following sodium bicarbonate, and the third group left as an undrenched control. Three drenches were given during June and July. The results failed to indicate any beneficial effect of treatment, as measured by reduction in worm egg count in the treated animals. 0n August 14, all animals were drenched with phenothiazine. In this season seven calves died during May, June and July, five of the deaths were in the set-stocked group, and two in the rotationally-grazed group. For simplicity, the graph of the egg counts of the calves in their original groups is given, while the drenching treatment of the calves and the fact that they were run together after mid-April has been ignored.

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<u>1941/42</u>: In the second season there were 15 calves in the rotationally-grazed group and 18 in the set-stocked group. This was the first year that phenothiazine was available in New Zealand in more than small experimental quantities and it was used for the regular treatment of half the animals in the set-stocked group. Drenching was carried out at three-weekly intervals from January until June. The dose rate was 20 g. in January, February and March, and 30 g. in April, May and June. On a body weight basis the above doses ranged from 0.1 g. per 1b. body weight in the lightest to 0.05 g. per 1b. body weight in the heaviest calves in the group. Drenched and undrenched set-stocked animals were run together.

Rotational grazing was begun after weaning. In this season there were two deaths in May and June, both of which were in the set-stocked undrenched group.

<u>1942/43</u>: In the third season there were four groups. One group was grazed ahead of the cows after weaning, a second group grazed with the cows, the third group was the usual set-stocked group, and the fourth group was set-stocked but the excess pasture growth was controlled by the use of additional stock when this was necessary. None of the groups was drenched. The graph presents only the data for the standard set-stocked and rotationally-grazed groups of 11 and 10 calves.

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<u>1943/44</u>: In this season, in addition to the standard groups of 13 calves each, a third group was set-stocked from weaning until January and then transferred to the rotational system. The same treatments were replicated in a further three groups of animals. This trial was terminated rather earlier than usual owing to unfavourable dry weather and feed conditions. No drenching was carried out and there were no deaths.

At this stage the scope of the project was changed from a study of methods of calf rearing to one of lifetime productivity of dairy cattle under good and poor conditions of management, with provision for a switch over of half of each group from one level to the other after the first calving. The new project involved the setting up of two similar farms, each selfcontained and differing only in the management practices used.

The same amount of topdressing was used on each farm, i.e., 2 cwt. superphosphate per acre and one ton of lime on the peat areas.

On the so-called low plane farm there were five paddocks of 10 - 15 acres each, where the calves were reared under a set-stocking system and the cows were managed on the day paddock/night paddock method of grazing. No ensilage was saved but hay was conserved at the rate of approximately $\frac{1}{2}$ ton per milker and $\frac{1}{4}$ ton per head of dry stock. The pastures were not topped with the mower and harrows were used only to a limited extent.

On the high plane farm there were 20 paddocks of 3 - 4 acres each. Hay and ensilage were conserved at the rate of approximately $1\frac{1}{2}$ tons per milker and $\frac{3}{4}$ ton per head of dry stock, although the amount of fodder conserved and fed varied rather widely from season to season. Autumn-saved pasture was also available to the cows 2 - 3 weeks before and after calving.

At this point, regular examination of faecal samples was discontinued but after some trouble was experienced in the autumn of 1947 examinations were resumed for the two seasons 1947/48 and 1948/49. In these two seasons drenching was restricted to the low plane group, where the primary object was to ensure the survival of the maximal number of experimental animals in this group, rather than as a part of an experiment to study the effect of drenching <u>per se</u>. For the same reason treatment was not given to the high plane group as it was clear that it was not necessary. For these two seasons therefore the comparison is between animals rotationally-grazed without anthelmintic treatment, and animals set-stocked and given repeated treatment with phenothiazine. As there were no set-stocked animals that were not treated, the effect of anthelmintic treatment alone cannot be assessed.

The dose rates of phenothiazine and dates of drenching the low plane animals were as follows: -

			Average per 1b. body weight
1947/48	Jan. 31	28 g.	0.12 g.
	Feb. 25	17	0.11
	Mar. 31	Ħ.	0.10
1948/49	Feb. 2	tt.	0.12
	Feb. 16	tt	0.12
	Mar. 2	11	0.11
	Mar. 23	35 g.	0.13

The above dose rates are considered by some workers to be rather low for calves (Cauthen, 1945), the reason that they were not exceeded being that on two occasions, at Ruakura, drenching with phenothiazine has been followed by keratitis and so the smaller doses were used to reduce the risk of a recurrence of this condition which, in itself, can give a severe check to growing animals. (Whitten, Clare and Filmer, 1947).



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Results

In all seasons there were significantly more eggs passed by the set-stocked calves at one time or another during their first year. The level of infestation varied widely from year to year and the period over which significant differences were noted in the egg counts also varied widely. The earliest that it appeared was in December, and in one year it persisted until the end of July. In most seasons it was restricted to the period from January or February until April or May. It is of interest to record that the high plane group did not show significantly higher egg counts than the low plane group on any of the 61 sampling days. This was true even in those years when the low plane group was drenched and the high plane group was not. On the other hand, the low plane group passed significantly more eggs than the high plane group on 27 occasions.

It is true that there would be a greater output of faeces by the heavier high plane animals and that this might be expected to have some effect in reducing the egg count per gramme of faeces in this group. It is certain, however, that the difference due to this factor would be but a fraction of the real differences recorded.

There were no significant differences between the

groups drenched in 1941 with bluestone and nicotine sulphate, with tetrachlorethylene, and those that were undrenched. In the following year there were no differences in the egg counts of the animals drenched repeatedly with phenothiazine and those that were undrenched and grazing the same area as the drenched animals.

The low infestations recorded in 1943/44 were apparently due to the fact that this was a very dry season when the pasture growth was so poor that the calves were largely maintained on conserved fodder. It is of interest also that in this season the egg counts were the lowest recorded, and also the body weight differences were smaller than those noted in any other year. Discussion

The poor results obtained in 1941/42 with phenothiazine were unexpected at the time, but, as has been pointed out earlier, the parasites that are most frequently found in large numbers in New Zealand are <u>Ostertagia, Trichostrongylus axei</u> and <u>Cooperia</u> spp., whereas <u>Haemonchus</u> and <u>Bosicola</u> are relatively rare. Most workers are generally agreed that <u>Haemonchus</u> and <u>Bosicola</u> are very susceptible to phenothiazine, and in most other countries where anthelmintic tests have been carried out, <u>Haemonchus</u> and <u>Bosicola</u> are much more common than they are in New Zealand. There are a

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number of published papers giving data on the efficiency of phenothiazine against the species of cattle parasites which are common in New Zealand. Swanson, Porter and Connelly (1940) found that doses ranging from 0.44 to 1.1 g. per 1b. body weight were highly effective against Ostertagia and T. axei. Taylor and Sanderson (1940) used doses of 25 - 40 g. and reported that they were highly effective against Ostertagia. Porter (1941) showed that at 0.1 g. per 1b. body weight the drug was highly effective against T. axei but not Ostertagia, and that to be effective against the latter parasite the dose had to be increased to 0.2 g. per lb. body weight. Porter, Simms and Cauthen (1941) stated that at 0.2 g. per 1b. body weight it was not effective against Cooperia spp. Roberts (1941) found that at a dose rate of 0.16 - 0.8 g. per lb. body weight it was effective against Ostertagia and Trichostrongylus but not against Cooperia. Recently Riek (1951) in Queensland has reported observations on the efficiency of the drug in doses of 0.1, 0.2 and 0.3 g. per lb. body weight, and his data suggest that against Trichostrongylus it was highly efficient only at the two higher dose rates, but against Ostertagia and Cooperia the efficiency was poor even at 0.3 g. per 1b. hody weight.

The poor results with the drug at Ruakura are therefore not at variance with most of the published data when the dose rate on a body weight basis is taken into account and the fact that the three most common species at Ruakura are among those that are less readily controlled by anthelmintics.

Rotational grazing has long been considered as one means of reducing the risk of parasitic disease outbreaks. The free-living stages of the worms are influenced by such factors as heat, cold and dryness, and although different species vary widely in their resistance to adverse environmental factors, death of the larvae will follow the mere passage of time if they are not ingested by a suitable host.

Information on the survival of infective material of cattle parasites on pasture is scanty. Most workers, who have reported observations on longevity of larval stages, have made their measurements by grazing wormfree animals on areas kept vacant for known periods and later slaughtering them to recover the worms acquired from the pasture. Few data have been published on the rate of loss of infection and this is due, no doubt, to the very real technical difficulties that are encountered in this type of work. It is true, however, that while a few larvae may survive for a long period, the fact that the vast majority may disappear after a much shorter period may have an important influence in reducing the uptake of infection by grazing animals.

Baker (1939) at Cornell University states that Ostertagia and Cooperia larvae survived on pasture for 9 months. Threlkeld and Johnson (1948) noted that Ostertagia survived 4 불 - 6 불 months in Virginia. Swanson, working in Florida, found that Ostertagia, Trichostrongylus and Cooperia were able to survive for 5 months on pasture that was not grazed. Roberts, in Queensland, on the other hand, states the larvae, mainly Haemonchus, can survive for 2 months in large numbers but that the numbers are reduced after 3 months. He also points out that even in the absence of rain, eggs in cattle faeces can develop to the infective stage but that the larvae remain trapped beneath the hard surface crust of the faecal pad. The effect of rainfall then, is considered to favour the uptake of larvae by the host not so much be allowing development of eggs to proceed, but mainly by effecting dispersal of the larvae when the pad disintegrates. He points out also that larvae from cattle faeces are capable of lateral migration through several feet. It is clear then that, under grazing conditions, larvae could migrate from faeces where the grass is unpalatable to grass sufficiently far away to be eaten readily.

Recommendations for the use of rotational grazing to control parasitism usually provide for the periods

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between successive grazings to be as long as possible. From the figures quoted above it is clear that the relatively rapid rotation practised at Ruakura would be of limited value in this respect. This is particularly true of a locality such as Ruakura with a temperate climate lacking extremes of temperature, and with a very well distributed rainfall. The annual rainfall at Ruakura is 43 inches and very rarely does less than one inch of rain fall in any month. The maximum summer temperatures seldom exceed 80°F, and minimum winter temperatures seldom fall below 24°F.

Under optimal conditions of temperature $(80^{\circ}F)$ and moisture, larvae may reach the infective stage in 4 - 6 days, so that the calves would return to the paddocks in many instances when the larvae developing from faeces deposited on the previous round, would be at their most vigorous stage.

At Ruakura it is only under special circumstances that the pastures are left unstocked for periods of longer than those mentioned above. When calves are grazed on paddocks that have been cut for silage the period of rest varies from 4 - 6 weeks, and in the case of hay aftermath, for periods of 8 - 10 weeks. During the late autumn and winter months the animals have access to autumn-saved pasture. In this case paddocks are shut up in mid-April and fed off in small breaks by the use of the electric fence. The earliest breaks fed will have been rested for 8 - 10 weeks and the later breaks for 18 - 20 weeks. It is considered that the effect of the above periods will have little influence on the general uptake of larvae. For instance, in the case of aftermath, less than 50 % of the grazing will come from these paddocks and on the second and subsequent grazing periods there will be available infective larvae which have developed from eggs deposited at the time of the first grazing. The same is true also in the case of the autumn-saved pasture but, at the time of the year when this feed is used, differences between the groups have usually disappeared and the egg counts have fallen to a low level following the development of resistance by the host.

Factors that would appear to be of greater importance than the loss of infective material due to the resting of the paddock are those connected with the amount of contamination seeded on to the pasture and the numbers of larvae that subsequently become available to the calves. It is probably true that the infections in the calves are initiated when the young animals begin to graze and pick up larvae, most of which have been deposited on the pasture by the previous season's calves. The infections are then built up, slowly at first, but then more rapidly by auto-

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infection from material seeded on to the paddocks by the calves themselves. It is unlikely that the cows would make any appreciable contribution in this direction, as the infection in adult cattle is always very much lower than that seen in the younger stock. It is obvious, then, that under a set-grazing system, where the calves alone are confined to a small area of the farm, the bulk of the infective material which is deposited would have the opportunity of being picked up by the animals that are most susceptible to infection. On the other hand, under rotational-grazing, the stocking rate of calves per acre would be much lower, the eggs would be spread over a much greater area, and only a fraction of the larvae after development would be ingested by the calves. Since the same area is being grazed by the cows, most of the larvae on the pasture would be eaten by the cows, and in these older and more resistant animals they will fail to develop and be destroyed.

It has been pointed out by Taylor (1934) that while large numbers of larvae are able to reach the upper leaves of clover plants, this is not the case on the grasses where they tend to be deflected and come to rest on the outer and lower leaves, and only relatively few reach the upper portions of the younger and taller inner leaves of the plant. This fact would

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suggest that where the calves need only graze a paddock lightly, the parts of the grass that are less heavily contaminated would be available to them, and the more heavily infected lower leaves would be eaten by the cows or the dry stock. Conversely, where animals are forced to graze close to the ground, as in the set-grazing system, the number of larvae ingested would be greatly increased.

It is likely that, in the animals on the low plane of nutrition, the percentage development of ingested larvae would be greater than in those on the high plane. It is true also that the effect of a given infestation would be less when the host is on a high plane of nutrition: in other words, a greater number of worms could be harboured without the appearance of clinical signs of disease. Further, there is evidence in several nematode infections that the rate of development, fecundity and longevity, are all reduced when the host is maintained on a high plane of nutrition.

It is at present impossible to assess the relative influence on body weight of parasitological and nutritional factors but, in the writer's opinion, it is fairly certain that the nutritional factors are by far the most important. This view is borne out by the fact that body weight differences between the two

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groups were fairly constant from year to year whereas differences in the worm burden, although constantly in favour of the rotationally-grazed group, varied very widely from year to year, both with regard to the magnitude of the difference and in the period over which the differences were detected.

Ascaris lumbricoides in New Zealand Pigs.

It has often been considered that <u>Ascaris</u> <u>lumbricoides</u> is one of the most common parasites of the pig and also one of the most pathogenic. There is no doubt that this true in many parts of the world but until recently there was little information available on its importance in New Zealand. Many workers, notably Ransom and Foster (1920) and Roberts (1934), have shown that young pigs are more susceptible and show a higher incidence than older animals. In slaughtered pigs the incidence is usually higher in porkers than in baconers or older breeding animals.

With the help of the meat inspection staff of the Division of Animal Industry of the Department of Agriculture a survey of the incidence of roundworms in a series of porkers was undertaken in several abattoirs in New Zealand. The animals were selected at random without reference to the condition of the carcase or the presence of lesions in the liver. The small intestine was run out into a bucket and the contents examined for the presence of the parasites. It is obvious that with such a procedure it would be unlikely for an inspector to overlook a parasite as large as a mature Ascaris. Infestations with only migrating stages would not be detected and small immature stages in the small intestine might be overlooked. The results of the survey are presented in Table 12.

TABLE 12

Incruence of Asc	saris lumpricoldes	In Figs.
North Island	Number Examined	Number Infested
North Auckland Auckland Hawkes Bay Taranaki Wairarapa	13,761 45,868 43 475 50	0 55 0 0 2
Wellington-Manawatu	369	
	60,566	57

Incidence of Ascaris lumbricoides in Pigs.

Percentage of Total Infested - 0.1 %

South Island	Number	Examined	Number	Infested
Nelson Christchurch Dunedin		467 663 302:	:	10 130 45
Invercargill		400	-	0
	1,	832	:	185
			-	
Perce	ntage of To	otal Infest	ed - 10.	1 %

The results showed that the incidence in the North Island was very low but it was much higher in the South Island. At Invercargill, in Southland, the incidence was very similar to that seen in the North Island.

In view of the findings of Spindler et al (1944), that the feeding of skim milk was associated with an elimination of Ascaris and other nematodes, it is pertinent to consider the environment in which pigs are raised in New Zealand.

Approximately 90 % of the pigs are found in the North Island, where they are fed mainly on skim milk, whey, or buttermilk supplemented with barley or other grains, meatmeal and pasture. There is some feeding of garbage near the cities. In the South Island, on the other hand, grain feeding is more common, although in some areas milk by-products are also fed.

There are a number of points that should be made regarding the survey figures. The Auckland sample comprised some pigs from the Auckland area, but most of them came from the Waikato, Bay of Plenty, and North Auckland dairying districts, and were fed mainly on milk. The sample from Taranaki included 200 from Patea, a location chosen specifically for the reason that virtually all pigs there were fed whey. The Wellington sample included 225 pigs that were known to be from garbage-feeding establishments. The pigs from Invercargill were all milk-fed.

If the feeding of skim milk were responsible for

the low incidence, a similar incidence in garbage-fed pigs might be explained by the fact that the pigs bought by the garbage feeders would come from a population in which the incidence was low, and it would be unlikely that infected pigs would be bought. Even under suitable conditions for spread the infection might never make its appearance.

An additional very interesting point has been brought out by Ineson (personal communication) who examined feral pigs from both Islands. Nine out of a sample of 21 pigs, or 43 %, were infested. He also found Ascaris ova in 14 out of 40 faecal samples from feral pigs. He examined 1,024 domestic pigs, slaughtered in Wellington, in which the incidence was 1.3 %. His results lend support to the theory that a lack of milk favoured infection, but would fail to support an alternative one that the important place of grazing in pig-feeding practice in New Zealand was the operative factor.

Feeding Skim Milk to Parasitised Pigs

To investigate further the effect of feeding skim milk on nematode infections in pigs a number of animals were purchased after faecal examination showed them to be infected with Ascaris and/or Oesophagostomum. The pigs were placed in metabolism crates which allowed collection of the faeces passed each day. The collections were not quantitative but 70 - 90 % of the faeces passed was collected, screened and examined for parasites. As a rule in these experiments when there was a large amount of faeces only a fraction of the total, usually 1/2 to 1/8 was examined. If the parasites were very numerous several aliquots of 1/100 of the total were taken after thorough mixing. Observations made in the study of six pigs are given below.

Pig No. 1

A faecal examination showed the presence of both Ascaris and Oesophagostomum eggs. It was fed a sole ration of fresh skim milk, and consumed 10 gallons over a period of three days. It began to scour three hours after the first meal, and on that day it passed 50 Oesophagostomum; on the second day it passed 41 Oesophagostomum and one male Ascaris; on the third day it passed 68 Oesophagostomum, and on the fourth day 2 Oesophagostomum. When the animal was killed it still harboured 1,332 Oesophagostomum, 5 mature and 2 immature Ascaris. There were 10 lungworms in the bronchi.

Because of the bulky nature of the contents of the large intestine of the pig and the difficulty of detecting and counting small worms like Oesophagostomum in the very fibrous material, the opportunity was taken during the autopsy on this animal to determine the distribution of the parasite in the bowel. Four regions were examined separately: (1) the caecum from the blind tip of the viscus to the ileo-caecal valve, (2) from the ileo-caecal valve to a point half way to the centre of the coiled portion of the colon, (3) from this point to the centre of the coiled portion of the colon, (4) from the

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centre of the coiled portion to a point half way out of the coiled portion. In this animal this was the point where faecal pellets were beginning to form. The number of Oesophagostomum in each of the four regions was as follows: - Region No. 1 28 worms No. 2 1,300 worms •• No. 3 4 worms No. 40 worms • • • •

With this information the counting procedure could be simplified by discarding much of the bulky bowel contents from regions outside the normal habitat of the parasite.

Pig No. 2

The pig was placed in the crate and fed a grain ration for two days, then for three successive days it was fed 3 lb of dried skim milk in about 2 gallons of water. On the second day of the milk feeding the animal scoured and passed 6 Oesophagostomum. On the following day it scoured more severely and passed a further 15 Oesophagostomum and one Trichuris. The pig was killed two days later and the large and small intestines and the lungs examined for worms. It harboured 5 Ascaris, 28 Trichuris, 6 Oesophagostomum, and one lungworm.

Pig No. 3

An examination of the faeces of pig No. 3 showed the presence of both Ascaris and Oesophagostomum eggs. It was fed dried skim milk in water at the rate of 3 lb per day for three days. Within three hours of the first feeding it scoured, and during that day 296 Oesophagostomum and one Trichuris were passed. During the next day 238 Oesophagostomum, 3 Trichuris and one immature Ascaris were passed. When the animal was slaughtered, two days later, 850 Oesophagostomum were collected from the colon, and three mature Ascaris from the small intestine.

Pig No. 4/5

This pig was used for two experimental feedings, and so the results from this animal provide a double record in the table. Faecal examination showed the presence of a very small number of Oesophagostomum eggs but a moderate number of Ascaris eggs. The pig was fed one 1b of lactose in one gallon of water on each of three successive days. During the period of the experiment there was no diarrhoea but some mucus was passed in the faeces. Only four Oesophagostomum were passed, and, since it was known that a number of Ascaris were present the animal was retained for a further experiment. It was fed a grain ration for the period of 5 days between the two trials. The second treatment consisted of feeding a quantity of dried skim milk with a minimum of water added. A total of 9 lb was fed over a period of 4 days. No worms were passed and the animal was slaughtered. On post-mortem examination 16 mature Ascaris were collected, there were no Oesophagostomum, and there were 4 lungworms in the bronchi.

Pig No. 6

Pig No. 6 was fed 1 lb of lactose in water on each of 4 successive days. There were 23 Oesophagostomum passed in the faeces. When the animal was killed two days later 23 Oesophagostomum remained in the colon. There were 4 Ascaris in the small intestine, and 25 lungworms in the bronchi.

Pig No. 7

Pig No. 7 was placed in the crate and fed one lb of casein mixed with water. The mixture was eaten slowly on the first day and was obviously unpalatable. The second pound was not eaten completely even after a further two days without other food. The pig passed 14 Oesophagostomum between the first and the third day of the trial. On post-mortem 2,040 Oesophagostomum

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remained and there were approximately 200 lungworms in the bronchi.

TABLE 13

	Ratio of Worms Passed to Total Worms Present.							
Pig	No.	Treatment	<u>Oesophago-</u> stomum	<u>Ascaris</u>	<u>Tri</u> - churis	Lungworms remaining		
1		Fresh skim milk	161:1493 (11 %)	1:8 (12 %)	-	10		
2		Dried skim milk	21:27 (78 %)	0::5	1129 (3 %)	l		
3	:	Dried skim milk	536:1386 (38 %)	1:4 (25 %)	3:3 (100 %)			
4	:	Dried skim milk	0:0	0:16	***	4		
5		Lactose	4:4 (100 %)	0:16	-	4		
6		Lactose	23:46 (50 %)	0:4		25		
7		Casein	14:2055 (7%)	0::0		200		

The results of these trials are difficult to appraise because of the small number of animals available, and the varied infections that they carried. The percentages of the worms removed are not impressive when viewed according to the standards required of anthelmintics, but the fact that some worms were eliminated following the feeding of skim milk, either as the fresh liquid material or when reconstituted from the dried powder, is important. Larger quantities or a more prolonged feeding regime may result in the removal of greater numbers of worms. A limited attempt to determine the factor in the milk that is responsible gave inconclusive results, although pure lactose gave results which indicated that this constituent might be the important one.

The proof that dried skim milk will cause elimination of worms provided a suitable starting point for further investigation which would be simplified if a convenient host-parasite system could be found in a small laboratory animal.

Ascaris lumbricoides in Rats

Infection with <u>Ascaris lumbricoides</u> in rats was chosen for study since it provides a convenient hostparasite system, and, although it is an abnormal one, its pathology is well known. The lesions are severe and are caused principally by the mechanical damage resulting from the migration of larvae through the liver and the lungs.

Preliminary Trials

The first trial carried out was designed to determine whether a diet of skim milk fed during the stage of infection would have any effect on the subsequent development of the disease. Such an effect might occur if the milk diet produced conditions in the gut that were unfavourable to the hatching of the eggs or the subsequent penetration of the larvae. In addition the trial served to show which criteria were most satisfactory for assessing the damage caused by the larvae. It also provided data on the dose of infective eggs necessary to produce a severe but not fatal disease.

Eighteen young male rats weighing 150 g were weighed and divided into two even groups. Group 1 was fed a standard ration of commercial feed pellets. Group 2 was fed nothing but liquid milk for 48 hours before and 24 hours after infection. They were then fed the standard pelleted ration. Both groups were divided into three subgroups - a control untreated group, a group given 12,000 eggs, and a group given 24,000 eggs by stomach tube. All rats were killed 8 days after infection, liver and lung weights were recorded, and the number of larvae in the lungs estimated after peptic digestion.

The weight changes recorded suggested that the liveweight gain noted 8 days after infection might prove to be the most suitable quantitative measure of the effect of the infection. This figure would include, in addition to true body weight changes, loss of gastrointestinal contents resulting from reduced food intake and possibly also loss from dehydration. These effects, however, were part of the syndrome under study and there was therefore no reason why crude body weight

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changes should not be used to measure the severity of the disease. The weight gains, expressed as a percentage of the initial weight, were as follows: -

	Control	12,000	24,000
Norma <u>l</u> ration	20 %	11 %	0 %
Milk	25 %	3 %	-6 %

An analysis of variance revealed that there were significant differences between treatments (dose rates), but not between the diets. Actual lung weights showed a difference between diets, but not between treatments, whereas lung weights relative to body weight showed a difference between treatments, but not between diets. The apparent reversal of effect seen here was a reflection of the liveweight changes.

In the second trial the same treatments were repeated, except that Gaines' dog food was used as the standard ration, since it was easier to handle in the measurement of residues for calculating food consumption of the groups. The rats used in this trial were immature males with an average body weight at the beginning of the trial of 112 g. The same doses of eggs were used as in the first trial, but the effect on liveweight was greater since the animals were lighter initially. Food intakes of the groups between the 4th and 8th day after infection were measured. Liveweight changes were taken at the 8th day, and at autopsy lung and liver weights were recorded and the number of larvae in the lungs estimated. The mean percentage liveweight gains were as follows: -

	Control	12,000	24,000
Normal Ration	10 %	-10 %	-16 %
Milk	13 %	-5 %	-7 %

Analysis of the results showed that there were significant differences in weight gain both between treatments and between diets. Actual lung weights showed no difference, but the treatment differences in relative lung weights were significant at the 5 % level.

The food consumption for the groups between the 4th and the 8th days after infection were as follows : -

	Control	12,000	24,000
Normal ration	17 g/rat/day	8.0	4.8
Milk	17 g/rat/day	10.6	10.4

There were wide variations in the numbers of larvae recovered from the lungs in both trials, but no obvious differences were seen between the two diets in this respect.

Effect of Ascaris Infection on Growth and Feed Consumption

Twelve male and twelve female rats with an initial mean weight of 115 g were used. They were caged individ-

ually and fed Gaines' commercial dog food, and individual food consumption records were kept. They were divided on a body weight basis into 6 groups of 4 rats each. Groups of each sex were infected with 6,000 eggs and 12,000 eggs, and two groups were kept as controls.

The mean liveweight gains between the 5th and the 10th days after infection were as follows: -

	Controls	6,000	12,000
Males	15 %	2 %	3 %
Females	11 %	5 %	3 %

An analysis of variance showed significant differences between control and infected rats, but not between levels of infection nor between sexes. Daily food consumptions between the 5th and the 10th days showed a marked reduction following infection :

	Controls	6,000	12,000
Males	18 g	12 g	ll g
Females	16 g	1 3 g	12 g

The treatment differences were significant but the sex differences were not. However, when food consumptions between infection and 20 days were considered, the differences were no longer apparent. Mean daily food consumptions between infection and 20 days :

	Controls	6,000	12,000
Males	18 g	17 g	17 g
Females	16 g	16 g	16 g

Twenty days after infection when the liveweight differences had disappeared the animals were re-grouped and half of each original group infected with 24,000 eggs. On this occasion the percentage weight gains between the 5th and the 10th day after infection showed significant differences between the infected and the controls among the rats that were previously infected, and although their numbers were small, previously uninfected rats showed differences of a much greater magnitude.

	Previously	Infected	Previously	Uninfected
	Male	Female	Male	Female
Infected	1 %	3 %	-12 %	-7 %
Controls	8 %	4 %	13 %	6 %

Effect of Plane of Nutrition

Two experiments were designed to study the effect of Ascaris infections on rats that were maintained on high and low planes of nutrition. In the first trial 16 male and 22 female rats were used. The low plane diet used was whole wheat flour fed at somewhat below appetite to give a slow rate of gain. The high plane diet was whole wheat 69, dried skim milk 23, maize oil 4.5 (containing 40 units per rat per day of vitamin A), calcium lactate 2.5, calcium phosphate (monobasic) 0.8, and salt 0.2. It was fed <u>ad libitum</u> and resulted in good growth rates. Half of the rats on each diet were infected with 12,000 eggs three days after the experimental feeding began. At this point the male rats weighed 130 g. and the female rats 110 g.

The percentage weight gain during the 7 days after infection showed significant differences between treatments, diets, and sexes.

	Infected		Cont	rol
	Male	Female	Male	Female
High plane	0 %	5 %	28 %	13 %
Low plane	-6 %	-2 %	8 %	9 %

Six weeks after the first infection, when full recovery had taken place in all animals, the same rats were reinfected but, since there were wide differences in body weight, three levels of infection, 9,000, 12,000, and 18,000 eggs, were used, depending on the size of the animal. The percentage weight gain during the 7 days after infection showed a difference between diets but not between treatments, indicating that the second infection produced no effect at all.

	Infected		Control	
	Male	Female	Male	Female
High plane	0%	-2 %	4 %	-2 %
Low plane	-7 %	-4 %	-8 %	-1.5 %

The second trial in this series was similar to the first one except that the infections were not given until some body weight difference had been established between the diet groups. Twenty rats of mixed sexes were used and at the time of the first infection, three weeks after the differential feeding began, the average weight of the low plane rats was 100 g., and that of the high plane rats 150 g. Seven days after infection the percentage weight gains were :

	Infected	Control
High plane	-3 %	17 %
Low plane	-10 %	2 %

The second infection, which was carried out one month after the first, consisted of 6,000 eggs for the rats on the low plane diet, and 12,000 eggs for those on the high plane. The mean percentage weight gain 7 days after infection was :

	Infected	Control
High plane	4 %	1 %
Low plane	-5 %	-3 %

The differences between diets were significant but those between treatments were not.

Vitamin A Deficiency

A single trial was carried out on the effect of a diet deficient in vitamin A on the response to an artificial infection with Ascaris. Soon after the birth of their litters, breeding does were placed on a diet deficient in vitamin A and the young rats were maintained on the same diet after weaning. The diet consisted of sucrose 68, easein 15, corn oil 5, yeast 8, and U.S.P. salt mixture #2, 4. The groups were infected with 6,000 eggs at the point when about half the deficient group were showing moderate signs of xerophthalmia. After dosing there were a number of deaths in the vitamin deficient groups. Following are the records of the percentage weight gains in the 10 days after infection and the number of deaths occurring in the same period. There were no deaths in the groups with adequate vitamin A.

	Infected	Control
Adequate vitamin A	4 %	20 %
Deficient vitamin A	-21 %	-11 %

	Dead	Alive	Total
Infected	7	4	11
Control	l	10	11
Total	8	14	22

 $x^2 - 4.9$

It is clear from these results that the vitamin A deficiency in the rats resulted in an increase in the susceptibility and also in a poorer survival rate under the stress of infection. It is true that the vitamin supplemented groups weighed somewhat more than the deficient groups and all were given the same dose of eggs. In the supplemented groups the males averaged 89 g. and the females 86 g., while in the deficient groups the males weighed 76 g. and the females 67 g. There is a reduction in the apparent effect because of the heavy mortality in the infected groups since the casualties included most of the lighter animals and consequently the mean weight of the survivors was greater than if there had been no mortality.

Discussion

The experiments with <u>Ascaris lumbricoides</u> infection in rats have shown that there is virtually no effect from the feeding of skim milk during infection. The hopes that the infection might be a useful one in the study of milk consumption in relation to parasitic infection were not realised. A low plane diet resulted in more severe disease and delayed recovery. A second infection had a much smaller effect than the primary one due, undoubtedly, to the development of an acquired immunity. The results with vitamin A deficiency showed that in severely depleted rats the mortality rate was greatly increased under the stress of infection.

Trichinella Infection in Rats

Starvation

An experiment was carried out to observe the effect of complete starvation at the time of infection on the establishment of <u>Trichinella spiralis</u>. Seventeen adult male rats were used. One group was starved for 72 hours

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before and 48 hours after infection, while the control group was fed <u>ad libitum</u>. Half of each group was killed after a week and adult parasites in the intestine were counted. The remainder was killed after 4 weeks and counts made of the larvae in the musculature. Each carcase was skinned, beheaded, and eviscerated, then it was minced and digested with pepsin for 18 - 24 hours at 39° C. The number of larvae present was counted using a dilution method. The counting method consisted of making the digest fluid up to 1,200 ml., mixing thoroughly, and taking 12 samples of 0.1 ml., placing them in a Kline concavity slide, and then counting the larvae under the dissecting microscope. The total in the samples constituted 1/1,000 of the larvae present.

The results were as follows :

Starved		Control		
Adult Worms	Larvae	Adult Worms	Larvae	
7 6 3 5 5	74,000 2,900 4,700 1,700	5 4 2 5	2,100 1,300 2,300 3,600	

The rate of establishment of adult worms was low and may have been due in part to the age of the rats, and in part to the prolonged period that the larvae were kept in the peptic fluid which may have impaired their viability. However there is no evidence that there was any interference with the development due to the starvation over the time of infection.

Effect of Plane of Nutrition

After the completion of the experiment with <u>Ascaris</u> <u>lumbricoides</u> the remaining control animals were infected with <u>T. spiralis</u>. At the time of infection the low plane animals averaged 150 g. and the high plane animals 250 g. All rats were infected with 100 larvae liberated from infected meat by peptic digestion. The rats were maintained on the two planes of nutrition for 12 days after infection and then both groups were fed the same diet of commercial feed pellets. Four to six weeks after infection the rats were killed and the number of larvae which had developed was counted. The results were as follows :

Low Plans		High Pla	ine
Male rats	Female rats	Male rats	Female rats
74,400 79,500 97,100	35,000 13,200 10,000 33,600 106,000	5,000 6,400 10,000	7,700 6,200 4,500

The difference between planes of nutrition was highly significant. Since the low plane diet was deficient in energy, protein, minerals, and vitamin A, a trial was set up to study the effects of various factors, separately and in combination, as supplements to a basal ration of whole wheat flour. Twenty-eight female rats were used with 3 or 4 in each group. The basal group was fed whole wheat flour <u>ad libitum</u> and the other groups were fed the basal diet with or without 25 % dried skim milk, with or without vitamin A and with or without 5 % of a mineral supplement consisting of calcium lactate 70, dibasic calcium phosphate 22, and sodium chloride 8. After the diets had been fed for 7 weeks the rats were infected with 100 larvae. A month later they were killed and the larvae present in the muscles were counted.

The results are given in the following table :

	No Milk		Milk		
	No Minerals	Minerals	No Minerals	Minerals	
<u>No Vitamin A</u>	1,175 950 1,975	1,075 155 360 300	950 450 250 1,250	750 1,100 700 800	
Vitamin A	10 20 76 3	1,400 1,500 10,400	1,500 1,750 4,450 5,750	2,750 2,100 3,100 5,700	

The results of the trial are difficult to interpret . because of the small number of rats in the groups and the wide variability within groups. The very low counts in one group are difficult to explain, and a further trial will be necessary before any conclusion can be drawn. The figures suggest that more larvae have developed in the groups supplemented with milk and minerals but not with vitamin A. The body weight differences between the basal and the supplemented groups were smaller than in the previous trial because of the fact that the diet was fed <u>ad libitum</u> whereas in the earlier trial intake was restricted in the low plane group. A repetition of this trial is desirable but has not yet been attempted.

The Effect of Trichinella Infection on Appetite

There is a considerable loss in body weight in rats infected with <u>T. spiralis</u> and with it is associated a loss of appetite that is very marked in heavy infections. In one non-fatal case there was a weight loss of 30 % following infection with 8,000 larvae. In order to obtain more figures of both body weight loss and reduction in food intake 11 young male rats were used with infections at two different levels for comparison with uninfected controls. Food consumption was measured over two 5-day periods before the two groups of 4 rats were infected with 1,000 and 4,000 larvae each. The larvae used were freshly isolated from infected meat by 6 hours' digestion in artificial gastric juice. The results are set out in the following table :

Average Daily	Food	Consumption	in Trie	chinella	Infection
			(Grams	per rat	per day)
Period	1	2 3	3	4 5	6
Control Group Infected 1000 Infected 4000	25 21 22	25 24 22 21 22 13	4 21 L 21 3 14	5 23 L 22 4 17	25 23 23

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The food intake of the most severely affected animal in the trial fell to 3 g. per day over one 5-day period. This was only 14 % of its normal consumption.

The Effect of Vitamin A Deficiency on Trichinella Infection

Young rats of mixed sexes were used in the trial. They had been raised by dams that were placed on the vitamin deficient diet at parturition and the young rats were maintained on the same diet after weaning. A supplement of vitamin A was given to one group while the other was kept as a control. They were infected with 600 larvae when their average weight was 200 g. After 6 weeks all the rats were killed and the number of larvae present in the carcase determined after digestion. The results of the larval counts are given in the table.

	Male Rats	Female Rats
Low Vitamin A	437,500 52,000 83,000	4,000 53,000
<u>High Vitamin A</u>	11,750 6,750 6,750	8,500 8,500 2,000

During the infection the first female rat on the low vitamin diet regurgitated some of the suspension of larvae so that the figure of 4,000 for this rat was not comparable with the remainder of the group. In spite of the small numbers of rats in the groups the difference

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between diets was a striking one and it approached significance at the 1 % level. It was of interest to record also that, at slaughter, all the low vitamin rats showed a very heavy infestation with lice, while there were very few lice on the rats that received the supplement.

In a second experiment the effect of a shorter period of depletion was studied. On this occasion the rats were fed commercial feed pellets and when their average weight was 150 g. they were placed on the vitamin deficient diet with one group receiving a supplement of 40 units per rat per day. After 4 weeks on the diet all rats were infected with 2,300 larvae by stomach tube. They were killed 3 weeks later and counts made of the larvae present in the muscles. The results are given in the table :

<u>High Vitamin A</u>	Low Vitamin A
74,000	13,500
35,000	2,500
65,000	30,000
50,000	25,000
14,000	

The difference between the groups is not significant but it does approach significance at the 5 % level.

A third trial was carried out with 28 rats of mixed sexes that were similar in age and weight to those in the last experiment. They were treated in a similar manner but were kept on the deficient diet for 6 weeks before they were infected. They were infected with 2,000 larvae and were killed 3 - 4 weeks later. The results are given in the table. Differences were not significant.

	Male Rats	Female Rats
Low Vitamin A	5,800	29,000
+	7,500	5,000
	2,500	3,000
	3,300	9,000
	1,700	15,000
	5 ,8 00	34,000
	6,400	16,000
High Vitamin A	4,000	27,000
	9,000	36,000
	4 , 000	12,000
	11,000	23,000
		35,000
		4,000
		18,000
		15,000

The difference between the diets was not significant while the difference between sexes may be due to the fact that the males were killed nearly a week before the females so that more mature larvae would be present in the latter.

Oxygen Consumption of Trichinosed Rats

Using an even line of young male rats an attempt was made to determine the effect of infection with Trichinella on the oxygen consumption, using the method of Farmer and Crampton (1948). The rats were divided into 5 groups of 4 animals. Two groups were infected with 1,000 larvae and two groups with 4,000 larvae. A 3-day interval separated the two groups on the same dose level. The fifth group was an uninfected control group. The reason for staggering the infections was to obtain bi-weekly readings with only a single weekly test day.

The tests were carried out in the mornings after overnight starvation and readings were taken of the times required for the consumption of 200 ml. oxygen. The results fail to show any short term changes but there were significant differences between groups when the combined figures for the first four tests were considered, although differences on any one day were not great enough to reach significant levels. Furthermore, the difference was only seen in the more heavily infected group and it disappeared on the fourth week after infection.

One might expect an increase in the metabolic rate to follow infection but it was considered that the low figures recorded were the result of reduced activity due to the malaise and to muscular pain both of which are prominent symptoms of Trichinosis.

After the tests were completed the rats were killed and the number of larvae present in the muscles estimated. The average number of larvae in the rats dosed with 1,000 larvae was 80,000, and in those dosed with 4,000 larvae, 250,000.

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Discussion

The experiments with Trichinella infections have confirmed that there was greater fecundity in the females when the host was maintained on a low plane of nutrition, and, in the case of a vitamin A deficiency, there was a marked increase in the number of larvae which developed from an artificial infection. When the reserves of vitamin A were incompletely exhausted these differences were not apparent, and there even appeared to be an increased resistance with a partial deficiency of vitamin A, although the results failed to reach the 5 % level of significance. It is difficult to suggest an explanation for this fact, but the same phenomenon has been noted by Riley (1943) in the case of Nippostrongylus infection.

Trichinella is a very pathogenic parasite both during the intestinal phase and when the muscles are being invaded. During the intestinal phase there is a marked loss in weight and a serious reduction in food consumption through loss of appetite. Non-fatal cases in rats have lost 30 % of their body weight in 2 - 3 weeks. The observations on oxygen consumption have shown that there is a reduction in oxygen consumption in the 3 - 4 weeks after infection. There are at least three factors that may influence oxygen consumption. There may be an increase in metabolic rate associated with the infection, and oxygen consumption would be increased in the later stages of the infection when body fat is being consumed as a source of energy, following reduced food intake. The third factor which appears to more than balance the two already mentioned is the reduction in activity that is associated with malaise and muscular pain. The lethargy and disinclination to move on the part of the heavily infected animals was noted during the tests.

THE EFFECT OF A FISH DIET ON THE PARASITES OF DOGS

It has been reported (Wolfgang, unpublished) that there was some evidence suggesting that a change to a fish diet was associated with a reduction in the incidence of intestinal parasites, and that there appeared to be an inflammation of the intestines of dogs on such a diet. In order to study the problem an experiment was Three litters of Beagle puppies 6 - 8 weeks of set up. age were used. They were divided into 4 groups fed as follows: Group 1 - basal diet of commercial calf meal; Group 2 - equal parts of basal meal and head, fins and scales; Group 3 - equal parts of basal meal and fish gut, liver and ovary; Group 4 - equal parts of basal meal and fish fillets. Freshwater fish, mainly carp and pike, were used. The fish was chopped in a food chopper, mixed with the meal, packed into 300 g. cartons and frozen until required for feeding. The trial ran for 40 days, except for Group 2 which was killed after

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33 days when the batch of food was exhausted. Body weights and individual food consumption were recorded. The puppies were treated for Ascarids before the trial began and throughout the trial faecal samples were collected weekly for worm egg counts. After 2 weeks on the diet several dogs lost weight, lost appetite, and developed diarrhoea. The cause was not determined but it was not associated with the diet since there were some cases in 3 out of the 4 groups, including 2 cases in the group fed the basal meal. Seventeen days after the trial began all animals were infected with 3,000 embryonated eggs of Toxocara canis. At the end of the trial all animals were killed and autopsied. The worms present were counted and tissue from 3 different sites in the small intestine was taken for histological examination.

There was no evidence either macroscopically or histologically of any inflammatory changes in the intestinal mucosa.

The numbers of specimens of Toxocara, both adults representing a residual infection and larval stages which were probably the outcome of the artificial infection, are given in the table. There was no obvious effect of the diets on either the adult or larval numbers. Faecal egg counts were low throughout the trial and no differences were seen between the groups.

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TADLE 15				
Group	Adults	Larvae		
1	5 0 5 3	0 4 0 0		
2	1 4 2 7	0 1 0 0		
3	2 4 4 5	0 0 3 5		
4	5 5 3 1	2 10 0 4		

A single animal was used to study the effect of a fish diet on an established infection with <u>Uncinaria</u> <u>stenocephela</u> in the dog. The dog was infected artificially from larvae cultured from the faeces of wild carnivora - fox, wolf, and coyote, from the Granby Zoo. After egg production by the worms had begun the infection was followed by means of faecal egg counts over a period. The dog was fed, at different times, a sole ration of fish material as indicated in the table (Table 14), which also gives the detailed egg counts. It is clear from the table that there was no marked effect of any of the diets containing fish material on the course of an established infection with Uncinaria. There was a steady and regular decline in the numbers of eggs passed, but that is fairly typical of artificial infections, and no change recorded can be related to the diets fed to the dog. A sole diet of fish was not tested. Many dogs in the northern districts subsist for considerable periods on nothing but fish, and infections with Uncinaria are very common in such dogs.

TABLE 14

Date		Eggs per gram of Fa	leces
June 14	Fed canned dog food and	7,500	
July 8 10 11		3,700 4,000 3,400	
12	Fed fish fillets and meal	-	
13 18 24 26 30	mixture tt tt tt tt tt tt tt tt tt tt	2,200 2,300 1,800 3,200 700	
Sept. 24	17 17	3,800	
Sept. 25	Fed fish scales and meal mixture		
Oct. 11 24	17 17 17 17	1,400 1,900	
31	Fed fish vizcera and meal		
Nov. 10 12		1,300 700	
30	Fed canned dog food and com	m-	
Dec. 11		900	

The author was absent during August and September but the same feeding regime was continued during the period although no egg counts were carried out.

THE EFFECT OF VARIOUS DIETS ON OXYURID INFECTIONS IN MICE

In order to study the effect of skim milk on an established infection it was desirable to find a suitable host-parasite system in a small laboratory animal so that experiments could be carried out with larger numbers. Oxyurid infections in mice appeared to offer the most promise. A colony of white mice was available in which there was a heavy natural infection with both Syphacia obveolata and Aspiculuris tetraptera. Experiments were carried out by selecting suitable animals usually from the weaner stage to young adults. They were divided at random into groups, and each group fed the selected diet for 4 days. On the fifth day all the mice were killed, the caecae and colons were removed and the worms counted, either individually, or, if more than 100 were present, one tenth of the total number was counted. All counting was done under the dissecting microscope. Both sexes, both species, and adult and immature worms are combined in the tables.

In the first experiment a diet of dried skim milk was compared with one of commercial feed pellets, using 10 mice in each group. All the mice on the skim milk diet developed a severe diarrhoea and the large intestines on post-mortem contained fluid contents which was yellowish in colour and frequently contained a large

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number of gas bubbles.

Analysis of the results was carried out using logarithmic transformations - $\log (x + 1)$, or where dilution counts were made - $\log a(x + 1)$; "x" is the number of worms counted and "a" the dilution factor.

The worms present after 4 days on the experimental diets were as follows :

Commercial Feed Pellets	Dried Skim Milk
69	0
76	0
87	0
89	0
150	0
157	0
212	0
264	1
462	8
484	13

A second experiment was carried out using 4 groups to compare the effect of different proportions of skim milk and whole wheat flour. The results were as follows :

Whole Wheat Flour	33% Skim Milk	67% Skim Milk	Skim Milk
8	0	0	0
120	õ	õ	õ
170	0	0	0
250	l	5	0
280	60	6	0
360	60	20	0
480	140	150	0
500	160	150	8
680	210	180	118
1,200	240	270	360
1,370	520	1,400	1,430

The mice in this trial also scoured, the degree of scouring being proportional to the amount of skim milk in the diet. It is believed that the failure in some animals in the milk groups was due to the fact that the animals were somewhat overcrowded and therefore the less aggressive members of the group would not have consumed the ration to appetite.

A third trial was carried out to compare the effects of various carbohydrates and of whole egg which contains virtually no carbohydrate, with the standard wheat flour and skim milk groups. Six groups of six animals were used with the following results : Whole Wheat Lactose Sucrose Starch Whole Skim Flour Egg Milk 0 0 7 0 0 1 21 12 0 0 0 4 54 29 0 23 0 0 100 44 45 0 2

The mice that were fed lactose consumed very little and consequently the death losses were high. The egg and the milk groups harboured significantly fewer worms than the other groups.

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130

130

150

In view of the striking results with whole egg a further experiment was carried out to compare the effect of egg albumen and egg yolk fed separately with a whole wheat meal group.

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Whole Wheat Flour	Egg Albumen	Egg Yolk
0	0	0
0	0	0
1		0
13	0	0
25	0	0
36	23	0
130	90	0
400		0
		0

The results were as follows :

The mice that were fed albumen appeared to suffer from an intercurrent infection since there were 3 deaths and several of the survivors showed a bloody diarrhoea when they were killed. The results with albumen therefore were inconclusive although the effect of the egg yolk was again very striking. A further attempt therefore was made to determine the effect of egg albumen and on this occasion it was combined with an equal amount of wheat flour on a dry matter basis. The results were as follows :

Whole	Wheat	Flour	and Albumen	Whole	Wheat	Flour
		0			0	
		0			0	
		0			2	
		0			Ţ	
		1			9	
		4			360	
		140			760	

Owing to the low incidence of parasites in the control group the results with albumen were still inconclusive. A further trial was carried out with egg albumen and wheat flour with an additional group fed

Egg Albumen and Wheat Flour 1:1	Sucrose and Wheat Flour 1:1	Wheat Flour
16	2	40
33	5	75
110	25	130
120	94	170
560	730	240
660		
1,030		

equal parts of sucrose and wheat flour. The results were as follows :

There is no significant difference between the groups.

In the next experiment comparisons were made of mixtures of wheat flour with lactose and with casein at a 1:1 ratio. The results were as follows :

Wheat Flour	Wheat Flour and Casein	Wheat Flour and Lactose
		•
380	35	0
420	86	0
490	170	0
740	240	0
790	360	1
810	470	7
880	480	7

The results show a marked effect with lactose but none with casein.

Discussion

Further work on this phenomenon is desirable since an elucidation of the mechanism by which the parasites are eliminated might possibly give a lead for the development of new anthelmintics or to a method of treatment by diet therapy. The observations are important also because Oxyurids in mice are frequently employed in parasitological experiments, particularly in anthelmintic screening tests. Erroneous conclusions might be drawn if dietary factors were overlooked. Although the Oxyurids are usually considered to be virtually non-pathogenic there remains the possibility that in the interpretation of nutritional experiments an effect might be attributed directly to the diet when in fact it might be an indirect one brought about by a reduction in the parasite load.

SOME SPECIAL PROBLEMS IN A GRAZING ENVIRONMENT

The economy of New Zealand is one based primarily on livestock, and its livestock rely almost entirely on pasture as the main source of food. With a climate that is almost ideally suited to pasture growth, and with high natural or induced fertility, very heavy concentrations of stock per acre are possible. Under these conditions the risk of parasitism is ever present. It is certain that the high nutritive quality of rapidly growing pasture plays a very important part in limiting the ravages of parasitism in such an environment.

The results of Clarke, Barton, and Wilson (1953) at Massey College, and those of Walker (1955) at Ruakura have shown that high stocking rates are not incompatible with a high standard of stock health, and are essential if high per acre production is to be achieved. In the former study breeding ewes, grazed entirely on improved pastures at concentrations of about 8 ewe equivalents per acre, were able to raise their lambs to give a high quality carcase of 33 - 36 lb dressed weight in 100 - 110 days. No anthelmintics were used. The mortality in the ewes was 6 % and in the lambs it was 12 %, virtually all of which occurred at, or within, 3 days of birth. In the Ruakura study, ewes grazed at 4 or 6 per acre with cattle, or 8 ewes per acre without cattle, were capable of producing 250 lb of high quality meat per acre on pasture alone. There was a 5 % mortality in the ewes and 13 % in the lambs. Again it was not necessary to give any anthelmintic treatment before weaning. Total worm counts carried out by the writer on representative lambs from the flock showed that the infections were relatively light, although experience under similar conditions would suggest that a fairly rapid rise would take place soon after this time if the lambs were not slaughtered.

Recent work in New Zealand (Clarke and Filmer, 1954) has added another example of some of the difficulties in the interpretation of field experiments. The cause of "hogget unthriftiness" is still unknown, but the disease is characterised by loss of weight, anaemia, and diarrhoea. It occurs to some extent every autumn but it is more in evidence in a wet season than in a dry one.

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It appears after weaning, increases in severity until April, May or June, and slow recovery takes place on the same pastures after the middle of June. Parasite counts have been made and show that the worm burdens mirror very closely the body weight changes. Regular, and even intense, courses of anthelmintic treatment with phenothiazine have failed to eliminate the disease, although on some occasions they have helped to check weight losses and have reduced the mortality. It has been observed that unthrifty lambs, removed from regular sheep pastures and grazed throughout the autumn and winter on dairy pastures, have made much more satisfactory growth and suffered lower mortality than their mates grazed throughout this period on sheep pastures. This fact has been confirmed repeatedly at Ruakura and has provided a basis for field experimentation. The results so far have shown that dairy pastures are invariably more satisfactory than sheep pastures. They are even superior to sheep pastures that have been allowed to grow long to resemble very closely the dairy pastures. Long sheep pastures are, however, better than short sheep pastures. The intake on short pasture is less than on long pastures, and the times spent grazing are longer. Lambs on pastures that are relatively worm-free show a similar growth pattern, which suggests that the cause is a nutritional one rather than parasitic.

Taylor (1934) has made the suggestion that pastures rich in clovers tend to increase the risk of parasitic disease, and he outlines what he considers to be the mechanism by which the effect is brought about. The structural difference between the clover and grass plants results in a greater availability of infective larvae, since a greater number of the migrating stages are able to climb to the top of the clover leaf whereas there is a tendency for them to be deflected on to the lower outer leaves of the grass plant. These larvae then only become available to the grazing animal when close grazing is practised. Taylor has also suggested that the prevalence of parasitism in certain districts, notably in Northumberland, is referable to the increased use of basic slag as a fertilizer and the consequent increase in the proportion of clover in the sward.

In sharp contrast to the findings of Taylor are some experiences in lamb growth studies in the Poverty Bay district of New Zealand, where the growth of lambs on white clover pastures is definitely superior to that on pastures dominant in perennial ryegrass. The magnitude of the differences observed can be seen from the growth curves in Figure 10, drawn from data collected by Sinclair (unpublished) who carried out controlled grazing trials on the two pastures. The superiority of the clover pastures is obvious and the lack of any significant



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difference between the worm burdens of the two groups when they were slaughtered suggests that this factor was not involved in the differential growth rates. The true explanation of the difference has yet to be found.

New Zealand presents good opportunities for the study of the epidemiology of parasitism in a grazing environment because although the islands are small, and the climate is an oceanic one, the presence of a north to south chain of high mountains results in a very wide difference in rainfall, ranging from 200 inches on the west coast of the South Island to as low as 10 inches not many miles away on the eastern slopes of the same mountains. There is no very great range in either diurnal or seasonal temperatures but there are widely differing soil types within a fairly small area. Few New Zealand soils have a high natural fertility, but the poorer soils have been brought into a high state of production by the use of artificial fertilizers, modern high producing strains of pasture plants, and special methods of grazing management that have been developed to cope with the rapid growth of the herbage.

The results of the calf raising experiment at Ruakura are of value in that they provide a practical means of reducing calf wastage under intensive dairy farming conditions, whereas, under the poor system of grazing management, many difficulties arise. The better thrift of the young animals follows automatically the enlightened system of pasture utilization and animal management, and it has been achieved without recourse to drug therapy.

Another factor of considerable importance is that a reduced survival rate in the young animals of a herd retards genetic improvement. Under some circumstances the wastage may be so high that all survivors are required for recruitment as herd replacements. The fruits of progress in selective breeding are concentrated in the young stock, and poor survival reduces the scope for selection, and slower growth to maturity lengthens the generation interval and so reduces the rate of genetic improvement.

It is obvious that an improvement in the level of nutrition is the natural solution to problems of this nature but in New Zealand it is difficult to implement in a simple manner that would be economic. Concentrates are always in short supply; they are highly priced and therefore of limited value in this respect. Forage crops such as rape, kale, lupins, and turnips, are useful, but the greatest potential lies in pasture and adaptations of methods of management to eliminate the seasonal fluctuations in available food supply. Pasture conservation in the form of hay, silage and autumnsaved pasture is extremely valuable, but dried grass is out of the question because of the high costs of fuel and labour. The improvement of pasture production by the sowing of improved strains of herbage plants, by better use of fertilizers, by rotational grazing, and by the use of special purpose pastures, designed to provide extra feed during periods of shortage in the regular pastures, offers the best means of raising the plane of nutrition of grazing animals and thereby promoting a state of resistance to parasitic infection.

GENERAL DISCUSSION

The relationships between parasitism and nutrition are complex. Some of the aspects of the problem can be discussed under the following headings: <u>The Effect of the Host Diet on the Nutrition of the</u> <u>Parasite</u>

Although the effect of the host's diet on the parasite is the most direct of those under discussion there is still very little known about it. The work of Chandler and his school gives perhaps the best synthesis. As far as the tapeworm Hymenolepis is concerned, carbohydrate appears to be the most important nutrient in this respect, while the parasite's protein requirements are provided directly from the host tissues and the worm is thus quite independent of dietary sources of protein. Another example is that of Ascaridia in chickens; the growth of the worm is restricted when the host if fed a diet low in either calcium or phosphorus. The Effect of the Host Diet on the Resistance to Parasitic Invasion

Much more work has been done on this aspect of the problem but there are still many gaps in our knowledge. Vitamin A is undoubtedly one of the most important factors in this respect and a sound basis for this fact is provided both by theoretical considerations and by the results of experimental work. Another important factor is the protein content of the diet which must provide a full range of essential amino acids.

The Effect of Host Nutrition on Parasitic Disease

It is natural to expect that in parasitic infections a high level of nutrition will raise the threshhold at which symptoms become evident, and will limit the severity of the disease both in the acute and chronic stages. A diet that is rich in body-building elements is essential for the repair of damaged tissue, or to replace body reserves that have been depleted during the acute phase of the infection. Where an immune response is involved a low protein intake may impair antibody production which, in turn, may disturb the host-parasite balance.

The Effect of Host Immunity on the Nutrition of the Parasite

In some infections there is a strong immune response that is manifested by the production of pre-
cipitates in and about the openings of the excretory and digestive organs of the parasite. It has been assumed that such precipitates might bring about their harmful effect on the parasite by mechanical interference with its normal digestive processes.

The Effect of Parasite Activity on the Nutrition of the Host

In many parasitic infections it is not known how the pathogenic effect is brought about. There are a number of examples where the activity of the parasite is responsible for a serious effect on the host. The best known ones are the worms that produce anaemia in the host simply through the haemorrhage caused by their blood-sucking activities. The hookworms of man and dogs and Haemonchus of ruminants fall into this category. The strongyles of horses, although more strictly they are tissue feeders rather than blood suckers, produce a similar result. While the blood that is lost contains plasma proteins as well as erythrocytes, the haemoglobin, and more particularly its iron, is probably the most important factor.

In the case of Trichinella infection the damage produced by the adults in the gut is thought to be sufficient to limit digestive processes during the acute phase of the disease. A more chronic effect is seen with parasites such as Oesophagostomum when many calcified nodules are present in the wall of the intestine. In heavy infections of long standing bowel motility is so reduced that it interferes with digestive processes. The presence of a large number of nodules in the gut may result in intussusception which also arises from the impaired motility of the intestinal wall.

A third example is seen in the case of Diphyllobothrium in man, where there is competition between the parasite and the host for the available vitamin B_{12} in the food. It is only when the tapeworm is established well forward in the intestine that it is able to compete successfully to the disadvantage of the host. In the majority of cases the worm is situated further back and there is adequate absorption of the vitamin by the host before the food reaches the point where the worm is residing.

An aspect that is less well documented is that of interference with the digestion of several nutrients, particularly of protein. There are many contradictions in the literature and much disagreement among authors. The Cambridge workers brought forward evidence that there was a lowered digestibility of protein associated with heavy mixed infections, but American workers were unable to confirm it with pure infections of either Cooperia or Trichostrongylus. In Australia, on the other hand there was some confirmation of it in the case of Trichostrongylus, but in these trials the period when differences were most marked did not correspond with the period of greatest egg production by the parasites. SUMMARY AND CONCLUSIONS

The results of the present study can be considered in four separate parts.

(1)Survey techniques have been used in two investigations. One utilised the accumulated data from a diagnostic service in New Zealand to elucidate certain aspects of the epidemiology of internal parasitism of dairy cattle. The main pathogenic species present were Ostertagia, Trichostrongylus axei, Cooperia and Dictyocaulus. Other species occurred but because of their small numbers they were of little consequence from the point of view of parasitic disease. There was a well marked peak of worm infection in the autumn and early winter months, but it was not certain whether this was due to seasonal conditions or due to the fact that calves all reached their most susceptible age at about the same time. The level of infection was higher in two eastern districts and lower in two western districts. The rainfall in the western districts is considerably higher than that in the east. There was no difference between the North Island and the South Island and no evidence of any north to south trend in level of infection. There was no clear relationship between the level of infection and the presence of scouring in the cattle.

The second survey was carried out in an attempt to collect incidence data for <u>Ascaris lumbricoides.</u> It was shown quite clearly that there was a much lower incidence of Ascaris in domestic pigs in the North Island than in the South Island. There appeared to be a relationship between the feeding of dairy by-products and the low incidence, since in those areas in the South Island where skim milk was fed the incidence was very similar to that seen in the North Island. In New Zealand grains are used to supplement a basal diet of skim milk rather than the reverse, and this may be responsible for the fact that the incidence of Ascaris in pigs is lower than in any other area for which figures are available.

(2) A study of the parasite population in two experimental herds of cattle that were managed under conditions similar to those found on well-managed and poorly-managed commercial herds, has shown that a system of rotational grazing resulted in better growth of young animals and in a lower level of parasitic infection. This difference occurred even when the set-stocked animals were drenched with phenothiazine and the rotationally grazed animals were not drenched. The factors that were thought to be involved were the higher level of nutrition, lower intake of infective larvae, and a greater resistance to infection of the rotationally grazed calves. From this study has arisen a well tested and thoroughly satisfactory method of parasite control which is compatible with modern enlightened techniques of management of grassland and the grazing animal.

(3) Small laboratory animal experimentation, using a variety of host-parasite systems, has been used where larger numbers and standardised infections were more feasible than similar experiments with large animals. Several trials were made to seek a suitable host-parasite system for studying the effect of skim milk on the infection, and pinworm infections in mice proved to be the best. With infections of Ascaris in rats, the severity of the disease, as measured by the percentage weight gain in the 7 - 10 day period after infection, was increased when a generally deficient diet was used, and also when the ration was low in vitamin A. With a severe deficiency of vitamin A infected rats suffered a heavier mortality than uninfected controls, whereas in rats that were adequately

supplied with vitamin A the infection produced moderate symptoms of pneumonia but no mortality.

Infections with <u>Trichinella spiralis</u> in rats showed that there was an increased number of larvae developed in the muscles of rats that were maintained on a low plane ration and in rats that were fed a diet low in vitamin A. The effect occurred only in rats that were severely depleted, since it was not seen in rats that were placed on a deficient diet at weaning and maintained on it for 4 - 7 weeks.

A series of experiments with mice infected with pinworms showed that there was a striking elimination of the parasites when the mice were fed for four days on dried skim milk, lactose, or egg yolk. The effect was also seen when varying proportions of these materials were mixed with cereal. The result was not seen with sucrose, starch, casein or egg albumen. The effect of skim milk was probably due to its lactose content, but the factor present in egg yolk has not yet been determined.

(4) The fourth part of the investigation required returning to the large animal for experiment on a limited scale. In the present series, the pig was used to determine the effect of dietary modifications on its parasites. The available pigs were infested with Oesophagostomum and, in some cases, Ascaris as well. It was shown that some parasites were eliminated with fresh skim milk, with reconstituted powdered skim milk, and with lactose. Casein resulted in the elimination of very few parasites. Further work along these lines is necessary before definite conclusions can be reached.

A trial was also carried out to test the effect of a diet of fish on the parasites of dogs. A basal meal was mixed with equal quantities of fish fillets, fish viscera, or fish scales and heads. There was no effect either on the adult Toxocara present in the intestine, or on the establishment of a new infection from embryonated eggs administered during the trial. The same three rations fed in turn to a dog harbouring <u>Uncinaria stenocephela</u> had no apparent effect on the course of the infection.

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