Impact of nutrition on the pathophysiology of bovine trypanosomiasis

P. H. HOLMES*, E. KATUNGUKA-RWAKISHAYA[1], J. J. BENNISON[2], G. J. WASSINK[3] *and* J. J. PARKINS

University of Glasgow Veterinary School, Bearsden Road, Glasgow G61 1QH, UK

SUMMARY

Trypanosomiasis is a major veterinary problem over much of sub-Saharan Africa and is frequently associated with under-nutrition. There is growing evidence that nutrition can have a profound effect on the pathophysiological features of animal trypanosomiasis. These features include anaemia, pyrexia, body weight changes, reduced feed intake and diminished productivity including reduced draught work output, milk yield and reproductive capacity. Anaemia is a principal characteristic of trypanosomiasis and the rate at which it develops is influenced by both protein and energy intakes. Pyrexia is associated with increased energy demands for maintenance which is ultimately manifested by reductions in voluntary activity levels and productivity. Weight changes in trypanosomiasis are markedly influenced by the levels of protein intake. High intakes allow infected animals to grow at the same rate as uninfected controls providing energy intake is adequate whilst low energy levels can exacerbate the adverse effects of trypanosomiasis on body weight. Reductions in feed intake are less apparent in animals which are provided with high protein diets and where intake is limited by the disease animals will often exhibit preferential selection of higher quality browse. Further studies are required to evaluate the minimum levels of protein and energy supplementation required to ameliorate the adverse effect of trypanosomiasis, the nature and quality of protein supplement to achieve these benefits and the influence these have on digestive physiology.

Key words: Bovine trypanosomiasis, nutrition/parasite interactions, pathophysiology of trypanosomiasis.

INTRODUCTION

Trypanosomiasis, a protozoan disease transmitted by tsetse flies, is probably the most serious veterinary and animal production problem in sub-Saharan Africa and prevents, or seriously curtails, the keeping of ruminants and equines over millions of square kilometres of potentially productive land. The most pathogenic trypanosome species affecting cattle in Africa are the vascular trypanosomes *Trypanosoma congolense* and *T. vivax*. Through the absence of cattle to provide draught power, milk, manure and meat, rural development is severely impaired. Much research and extension application effort has been devoted to the control of this disease through the use of drugs and control of the fly vector yet only 5 % of the affected area has been cleared of tsetse flies over the past century.

BACKGROUND

Attempts to control the disease are compromised by the problem of geographical scale and the often unusual biology of the trypanosome itself. Drug resistance is apparently increasing and there seems to be little hope of producing a conventional vaccine within the foreseeable future. The difficulty here is that trypanosomes possess multiple mechanisms for immune evasion since, in the infective state, they are in constant contact with the immune systems of the host which they are able to evade by antigenic variation by switching their major variant surface glycoproteins (VSG). Added to this problem, each trypanosome species comprises a number of different strains or serodemes, all capable of eliciting a different repertoire of VSG variation (Murray & Black, 1985; Barry & Turner, 1991).

These factors and the sheer enormity of the tsetse fly control problem largely restricts the keeping of cattle and other ruminants to areas where tsetse flies are less abundant, such as the edges of semi-arid areas where the supplies of water and feed are frequently compromised. The only exception to this general rule is in West Africa where a number of breeds of cattle, e.g. N'Dama and West African Shorthorn and some breeds of small ruminants have developed over many centuries a degree of innate resistance or trypanotolerance to the pathogenic effects of trypanosomes, and as a result those breeds can at least survive in areas with high tsetse populations. However, even in these areas, feed supplies through the prolonged dry season can be severely restricted and trypanotolerance can be compromised. Therefore across Africa, cattle infected with trypanosomes frequently also suffer from undernutrition.

* To whom all correspondence should be addressed.
Present address: [1] Dean, Faculty of Veterinary Medicine, Makere University, PO Box 7062, Kampala, Uganda. [2] Agrimin Limited, Elsham Wold Estate, Brigg, Lincolnshire DN20 0SP, UK. [3] Groom's Cottage, Knowlegate, Ludlow, Shropshire S78 5JA, UK.

THE DISEASE

Bovine trypanosomiasis is characterized by the development of a moderate to severe anaemia, loss of condition and pyrexia, which is especially prominent during the early phases of the disease when the waves of parasitaemia are particularly high. Although the disease can occur in an acute form, it is normally associated with infections which last weeks or months and a slow and insidious loss of condition resulting in eventual death.

ROLE OF NUTRITION

There can be no doubt, in the face of increasing drug resistance, concerns about pesticide residues entering the food chain and the difficulties with the ongoing search for effective practical selection markers for resistance for use in cattle breeding programmes, that host nutrition remains an important factor influencing the host–parasite relationship and the ability to withstand the impact of parasitic infection. The purpose of this paper is to review the impact that nutrition has on the pathophysiology of bovine trypanosomiasis.

TRYPANOSOMIASIS AND PARASITIC GASTROINTESTINAL INFECTIONS

Until relatively recently studies on the interactions between nutrition and the pathophysiology of parasitic infections have largely been confined to helminth infections and there is now a large body of literature on this subject which has been reviewed periodically in more recent years (e.g. Parkins & Holmes, 1989; Coop & Holmes, 1996; Van Houtert & Sykes, 1996). These have clearly demonstrated that poor nutrition, especially low protein intake, can have a profound effect on the pathophysiology of gastrointestinal nematode infections. The earlier experimental studies evolved from original field observations in Australia which were later confirmed in controlled laboratory studies in Scotland and elsewhere. Similarly designed studies have only recently been conducted in ruminants infected with pathogenic trypanosomes and they are still far from complete.

As with the helminth infections, the studies with trypanosomes have been stimulated by field observations which showed that in areas of endemic trypanosomiasis the disease is exacerbated when there is obvious undernutrition of the livestock. One series of important observations was made in Ethiopia in the 1970s in oxen at a settlement scheme in a lowland valley heavily infested with tsetse flies (Bourn & Scott, 1978). These oxen could only be maintained by frequent and judicious use of trypanocidal drugs and good husbandry. As a result the cattle acquired a degree of 'resistance' and could continue working despite the presence of trypanosomes in their blood. However, during a period of drought when the level of feed provision fell, the oxen developed severe signs of trypanosomiasis (despite a decrease in the numbers of tsetse flies) and many succumbed to the disease despite the drug treatments. Similarly, in West Africa it has been reported that the clinical signs of trypanosomiasis in trypanotolerant cattle are more severe at times of the year when they are under nutritional stress, for example during the dry season (Agyemang *et al.* 1992). Interestingly, in the same area of West Africa when peasant farmers were asked whether they thought trypanosomiasis in their cattle was associated with tsetse flies or poor nutrition, nearly half of them replied that it was associated with poor nutrition rather than tsetse flies (Snow, personal communication).

However, in contrast to these observations, other studies have suggested that the onset of parasitaemia in cattle infected with trypanosomes may be earlier in animals which are receiving a supplemented diet (Little *et al.* 1990), possibly because of the raised levels of trypanosome nutrients in the blood. It is known from laboratory studies that the onset and degree of parasitaemia can influence the blood levels of various constituents used metabolically by trypanosomes such as lipoproteins and cholesterol (Black & Vanderweed, 1989). However, it is also recognized that host nutrition can have profound effects on many aspects of the immune response e.g. antibody production which in turn is known to influence the level of parasitaemia.

Feed and water intakes

A complication of most parasitic infections is varying degrees of anorexia. Another important feature of parasite infection is the change in preference or rather the specific selection of particular dietary components by infected animals. A number of observations have noted that decreased intake of the 'poorer' components (i.e. those with most fibre and least protein contents) occurs with a counterbalancing preference for the most 'nutritious' feeds in an otherwise overall depressed dry matter intake in both helminth (Kyriazakis *et al.* 1994) and trypanosome infections (Romney *et al.* 1997). There can also be marked changes in water intake and loss, and, as a result, changes in water retention and body composition. Again, observations on these changes have been largely restricted to studies conducted with helminth infections (Holmes & Bremner, 1971; Parkins, Bairden & Armour, 1982) and it is clearly important to see if similar effects are observed with trypanosomiasis.

Although gastrointestinal helminth and trypanosome infections are fundamentally quite different in their respective pathophysiological characteristics, it is worth briefly reconsidering here the known effects of gastrointestinal parasitism on host metabolism and digestive function so that these observations may act as a comparison template for the evaluation of nutrition-host effects with trypanosomiasis.

Reduction of voluntary feed intake is a major feature of the pathogenesis of both infections but for apparently different reasons. It is now accepted that there is no convincing evidence, in either infection type, of a single mechanism controlling anorexia. In gastrointestinal infection, local pain, changes in abomasal pH, alterations in gut motility, digesta flow rates and elevation of the hormone gastrin, have all been implicated in reduced feed intakes in such parasitized animals. Water intake and retention are also commonly increased and may confuse observations on body weight as an indicator of tissue loss in gastrointestinal infections (Parkins & Holmes, 1989). However, in trypanosomiasis, anorexia is a noted feature of the acute phase of infection and is possibly partly due to a release of interleukin-1 (McCarthy, Kluger & Vander, 1985; van Miert, van Duin & Koot, 1990) acting on the hypothalamus. Gut motility may also be affected by interleukins.

Effects on feed utilization

It is entirely reasonable to predict that gastrointestinal parasitisms of cattle and sheep produce serious disruption, or at least some evident impairment of digestive and physiological efficiency when abomasal and small intestinal function is clearly challenged by these disease processes and leads to the devastations to the nitrogen economy of the host caused by losses of blood plasma proteins (Parkins & Holmes, 1989). This is not so predictable in the case of trypanosomiasis.

Also, reduced nitrogen retention in gastrointestinal infections of both cattle and sheep has been clearly attributed to increased urinary nitrogen losses and logically gave rise to the interpretation that somehow a reduced efficiency of utilization of absorbed amino acids and possibly also tissue protein catabolism in the host were ultimately the main causes of the decreased production performance of infected livestock.

The limited work performed with trypanosome infections to date has implied that the effects of the disease on digestive function (i.e. apparent digestibility which is strictly defined as that part of the feed intake fraction not excreted in the faeces) are minimal and not allied to the degree of trypanotolerance exhibited by the breed type under investigation in each particular trial. For example, trypanotolerant West African Dwarf goats showed no apparent differences in the digestibility of the organic matter and nitrogen fraction of the feed despite reduced feed intake as a result of infection (Akinbamijo *et al.* 1990, 1992, 1994*a*, *b*; van Dam *et al.* 1996). With Gobra and N'Dama cattle. Akinbamijo *et al.* (1997) concluded that infection had no effect on the digestive physiology due to similar *in vivo* dry matter digestibilities and rate of passage.

RECENT STUDIES WITH RYPANOTOLERANT N'DAMA CATTLE

Most work on the interactions between nutrition and trypanosomiasis in cattle have been conducted at the International Trypanotolerance Centre in The Gambia using local N'Dama cattle where early studies (Agyemang *et al.* 1990*a*, *b*; Little *et al.* 1990) provided clear evidence of an interaction between nutrition and trypanosomiasis. However, the feeding regimes were not always representative of traditional livestock systems so, in later studies emphasis was placed on assessing the minimum quantities of supplementary feed required, the timing of supplementation in relation to stage of infection and the effects of trypanosomiasis on animals subjected to diets that were close to or below those required for maintenance purposes (Bennison *et al.* 1998*a*, 1999).

Anaemia and parasitaemia

A significant decline in PCV in response to trypanosomiasis is a characteristic feature of the disease and a primary criterion for assessing its severity (Murray & Dexter, 1988). Trypanotolerance is characterized by an ability to regulate the parasite population and a capacity to control anaemia (Dwinger *et al.* 1992). This was clearly demonstrated in the study by Akinbamijo *et al.* (1997) where Gobra (susceptible *Bos indicus*) and N'Dama bulls were infected with *T. congolense*. The degree of anaemia was more severe in the Gobra animals and parasitaemias were correspondingly higher.

However, Bennison (1997) demonstrated in N'Dama cows that the degree of anaemia responds to short-term changes in the plane of nutrition after the onset of infection. Cows, infected with *T. congolense* in the late dry season were offered either 0, 1 or 2 kg/d fresh groundnut hay as a supplement to native pastures. Results are shown in Fig. 1. The supplementation with groundnut hay did not commence until peak parasitaemia, 2 weeks post-infection. During the 8 week period of infection cows were withdrawn from the trial if their PCV fell below 15% (a critical level) but only 1/8 animals was withdrawn from the group given 2 kg/d supplement compared to 5/8 and 4/8 in the nil and 1 kg/d groups, respectively. The necessity of withdrawing 41% of the infected animals was unexpected. All were mature, multiparous females originally from areas of low to medium tsetse fly challenge. Of the previous experiments with grazing N'Damas in The Gambia, none had reported the loss or the need to treat animals

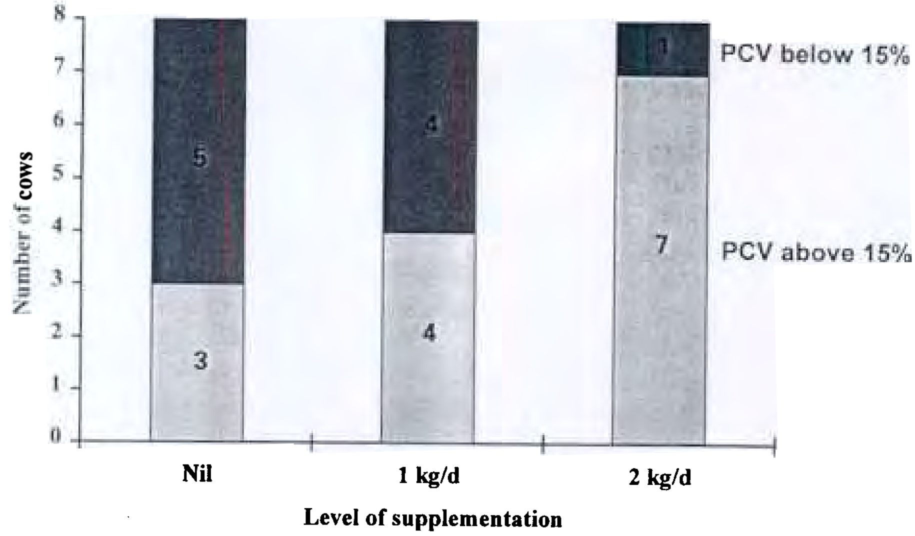

Fig. 1. Effect of feed supplementation of N'Dama cows infected with *T. congolense* on anaemia status.

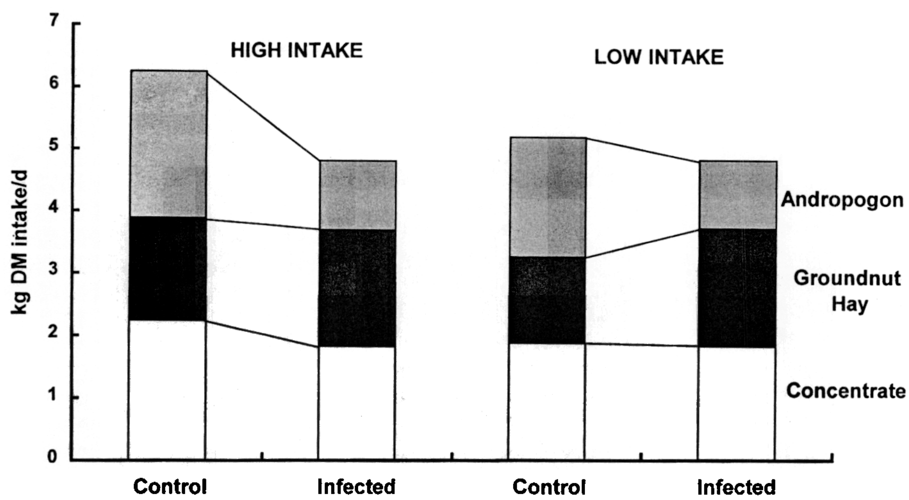

Fig. 2. Effect of infection with *T. congolense* and level of feeding on the voluntary intake of different dietary components in N'Dama cows.

Fever

Fluctuating fever, particularly in susceptible animals, is a typical sign of trypanosomiasis and is the result of an increase in metabolic rate. There is some evidence that trypanotolerance is linked to the suppression of fever during infection. Murray *et al.* (1981) found that the N'Dama, in contrast to the Zebu, did not become febrile even during waves of parasitaemia. The study by Akinbamijo *et al.* (1997) adopted a pair-feeding approach in which the food intake of uninfected control animals was matched to the intake of their infected partners. This ensured that the effects of anorexia on liveweight gain could be isolated from those associated with a change in the host's metabolism during infection. The trial showed that the decrease in liveweight gain in trypanotolerant animals is principally due to changes in intake, whereas in susceptible animals, the cause was a combination of decreased intake and increased energy requirements for maintenance which confirmed a previous finding by van Dam (1996).

Voluntary feed intake

Romney *et al.* (1997) observed that N'Dama heifers maintained intakes of groundnut hay and concentrate while apparently selectively decreasing intake of the poorest quality component, *Andropogon* hay, which was high in fibre and low in crude protein. It was suggested that animals may have selected against the feed expected to have the greatest effect on gut fill. Evidence that the intake response to infection might be influenced by factors other than those affecting gut fill is provided by Bennison *et al.* (1999) who offered lactating N'Dama cows two different total amounts of a diet consisting of fixed ratios of groundnut hay, concentrate and *Andropogon* hay. Results are shown in Fig. 2. When animals were

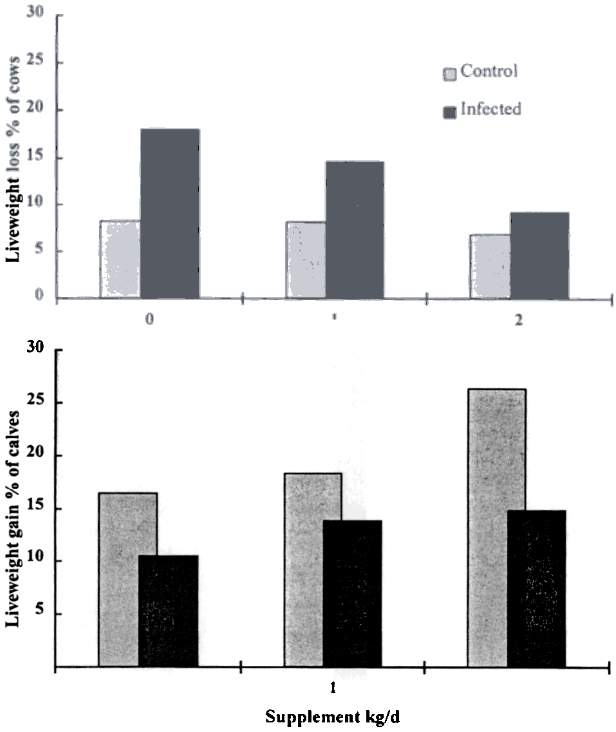

Fig. 3. Liveweight changes (%) of N'Dama cows and their calves following infection with *T. congolense* and supplemented with 0, 1 or 2 kg/d groundnut hay.

offered the high ration allowance, infection reduced intakes of all three dietary components by at least 20%, however, when the low ration was offered only the intake of *Andropogon* hay was reduced (by 26%). Furthermore, a draught animal study demonstrated significant interactions between infection, diet and work on food intake. Animals responding to the demands of work, appeared to be able to maintain feed intake during infection if the diet included a higher quality component equivalent to groundnut hay (Bennison *et al.* 1998*a*).

However, it now appears unlikely that trypanotolerance is linked to a superior ability to maintain intake during infection. In a comparison of pair-fed infected and control Zebu and N'Dama bulls, the relative decline in food intake was similar for both breeds (Akinbamijo *et al.* 1997).

Liveweight

The majority of trials provide evidence of the negative effect of trypanosomiasis on liveweight. Nevertheless, there are marked variations in the liveweight response to infection in N'Dama cattle. Paling *et al.* (1991) reported an impressive (for the N'Dama) average daily gain of 360 g/d over 694 days using 1 year old bulls and heifers with an initial weight of 150 kg. During this period the bulls were subjected to a sequential challenge of four different serodemes of *T. congolense*. The rate of liveweight gain was not significantly different from the uninfected controls. Unfortunately, no details of the diet were provided but it is thought that the animals received *ad libitum* hay and 1 kg/d of concentrate with a crude protein (CP) content of 140–160 g/kg DM (Leak, S.G.A, personal communication). One of the *T. congolense* clones used in the study by Paling *et al.* (1991) was IL1180. Romney *et al.* (1997) used the same strain in a study of the effects of infection and diet on 12 month old N'Dama heifers weighing 89–146 kg. The rations were devised such that animals in the basal group received less than the estimated ME requirements with a CP content of approximately 80 g/kg DM. The second group received in addition to groundnut hay and *Andropogon* hay, supplementary groundnut cake at 3·9 g/kg liveweight. This ensured ME intake was above the requirements for maintenance and the CP content was between 140–150 g/kg DM. Infected animals on the basal diet lost significantly more weight than their non-infected counterparts (71 g compared with 14 g/d). While infected animals supplemented with groundnut cake still gained weight, it was at a lower rate than the controls (52 g compared with 168 g/d). Both Romney *et al.* (1997) and Akinbamijo *et al.* (1997) suggested that in N'Dama cattle, weight change was primarily a function of a change in feed intake as a consequence of infection and not a change in metabolism. This might suggest that the quality of diet in the study by Paling *et al.* (1991) was sufficient for the infected animals to maintain liveweight gain despite a change in intake.

Feeding behaviour

It might be expected that these effects of infection would also be associated with detectable differences in animal behaviour, activity levels and diet selection by free-grazing animals. In The Gambia, Wacher (1993) provided circumstantial evidence that infected animals appeared to move more slowly, rest more and select higher proportions of browse and fruit. Although, these effects were often not statistically significant, the trends were most noticeable in the harshest conditions i.e. the dry season. These responses however, were not observed in a later study by Bennison *et al.* (1998*b*). Trypanosomiasis infection had no significant effect on diet selection, although there was a significant interaction between supplementation and infection on behaviour. Infected cows supplemented with groundnut hay spent a greater proportion of the day resting. In experimental conditions, infected goats masked part of the energy costs of infection by reducing their standing time (van Dam, 1996). This suggests that animals adopt energy-saving behaviour patterns in response to infection.

Interactions between infection, nutrition and milk yield

Although trypanosomiasis has a direct effect on milk yield of N'Dama cows, the relative decline in milk output and liveweight response to infection may be related to the plane of nutrition at the time of infection. Work from The Gambia reported by Bennison (1997), shown in Fig. 3, depicts the liveweight changes in N'Dama cows and their calves (expressed as a % of the original liveweights) following infection of the dam with *T. congolense* and dietary treatments which consisted of supplementation with 0, 1 or 2 kg groundnut hay per day. The figure shows the *relatively* reduced loss of liveweight in infected cows with increasing levels of groundnut hay supplementation and the mirrored *relative* increases of liveweight gain in their suckling calves. Thus the liveweight gain increases observed in the calves reflect the milk output benefits of supplementation in both control and infected cows. However, Bennison (1997) also showed that supplementation with groundnut hay of diets well below maintenance alleviated liveweight loss of infected cows but had little effect on milk yield as represented by calf liveweight change but when breeding cows were given rations close to, or marginally above requirements, there were significant interactions between diet and infection on the productivity response. Animals given a higher plane of nutrition attempted to maintain milk yield at the expense of liveweight, whereas the infected cows on the basal diet had insufficient liveweight reserves with detriment to the milk yield and consequently the calf (Bennison *et al.* 1999). *Pre-partum* nutrition had no effect on the relative change in milk yield and liveweight during a trypanosomiasis infection *post-partum*. However, despite differential *pre-partum* feeding, the cows were still in a lean but moderate condition at parturition and so the trial was unable to determine whether a marked improvement in body condition would influence the response. The study by Akinbamijo *et al.* (1994*b*) on West African Dwarf sheep suggests that this is possible.

STUDIES IN SMAL RUMINANTS

Trypanosome infection does sometimes appear, from the available literature, to exhibit remarkably similar nutritional outcomes to gastrointestinal infections in that negative nitrogen balance and retention in the host are adversely affected by both parasitic diseases (Parkins & Holmes, 1989; Akinbamijo *et al.* 1990; Verstegen *et al.* 1991; van Dam, 1996) but interpretation of some of these results is difficult where reduced feed intakes are not matched by a system of pair-feeding the control animals.

Most work investigating the influence of nutrition on the pathogenesis of trypanosome infections has used two contrasting planes of nutrition. Under such conditions, the respective roles of protein and energy are difficult to evaluate. A novel series of experiments were conducted in Glasgow, (Katunguka-Rwakishaya *et al.* 1993, 1995) which investigated the direct influences of protein and energy on various aspects of the pathophysiology of trypanosome infection using Scottish Blackface sheep, a breed which displays a significant degree of trypanotolerance. The sheep, aged 4–6 months, were given a diet which consisted of a mixture of sugar beet pulp, barley siftings (these are the 'awns' of the thrashed seed only equal in nutritive value to the barley straw itself), soyabean meal and vitamin/minerals in differing proportions. For the protein studies, animals on a 'high protein' diet received 1 kg of fresh matter per day which provided 116 g digestible crude protein (DCP) and 9·8 MJ of Metabolisable Energy (MJME) per day. Animals on 'low' protein intake received 51·5 g DCP and 10·0 MJME/d.

For the energy studies, the diets and feeding regimes were formulated so that animals on a 'high' energy ration received 9·9 MJME with 109 g crude protein (CP) per day and the animals given the 'low' energy ration received 6·1 MJME and 109 g CP/d. The animals in each study were divided into two groups and placed on either high or low protein or energy intake. Each group was further subdivided into an infected and control group. Trypanosome infection was achieved by inoculation with about 10^5 *T. congolense 1180* parasites, a cloned derivative of an isolate made in the Serengeti, Tanzania (Nantulya *et al.* 1984). Parasitaemia, haematological and blood biochemical changes were observed for periods ranging from 10 to 14 weeks.

Parasitaemia

It was observed that neither dietary energy intake had a significant effect on the prepatent periods or the intensity of parasitaemia that followed trypanosome establishment. These observations are in agreement with those of Little *et al.* (1990) and Reynolds & Ekwuruke (1988).

Bodyweight

Where energy intakes offered were adequate for normal growth (*c.* 10 MJME/d), the low protein intake significantly reduced growth in infected animals but not in their pair-fed controls, whereas the high protein intake apparently overcame any effect of infection and animals grew at the same rate as their controls. However, when a reduced intake of energy was offered, growth rates were greatly reduced in both infected and their control partners despite an adequate protein intake. The findings in the protein study are in agreement with those of

Hecker *et al.* (1991) and Agyemang *et al.* (1990*b*) in sheep and cattle infected with trypanosomes respectively. The poorer growth in the low protein infected group could not be attributed to any observed decrease in feed intake which contrasts with the reports of reduced feed intakes in goats infected with *T. brucei* (van Miert *et al.* 1990) or *T. vivax* (Zwart *et al.* 1991). Experiments in goats infected with *T. vivax* (Verstegen *et al.* 1991) have demonstrated that development of fever during a course of trypanosome infection is associated with increased heat production and increased metabolizable energy for maintenance. The consequence of this is that a proportion of nutrients that could be used for growth is reduced, as it is metabolized to provide the extra energy required. This effect appears to be greater for animals receiving a low protein diet, which was just above maintenance, compared to those on a high protein intake. It is interesting that this effect was not observed in the dietary energy study where infected animals on low energy intake grew at the same rate as their controls, which was significantly lower than the rates of infected and control animals on high energy intake. It can therefore be concluded that increased dietary protein ameliorates the effect of trypanosome infection on growth rate where energy intakes are not limiting.

Packed red cell volume (PCV)

In all dietary treatments infection caused a significant drop in PCV, the decline beginning with the appearance of trypanosomes in the circulation. The mean PCV values of the infected animals given either the low or the high protein diets were not significantly different but infected animals given the low energy diet had a significantly lower PCV than the high energy diet. The observation of similar degrees of anaemia in the high and low protein groups is consistent with the observations of Agyemang *et al.* (1990*a*). It would suggest that trypanosome establishment and the rate of development of anaemia are not influenced by dietary protein. In contrast, the observation from the energy study supports those of Little *et al.* (1990) in N'Dama cattle experimentally infected with *T. congolense* and given either a low or a high plane of nutrition and those of Fagbemi *et al.* (1990), and Makinde, Otesile & Fagbemi (1991) in pigs infected with *T. brucei* and given either a low or a high energy intake. In addition, Makinde *et al.* (1991) observed that pigs given a low protein diet developed significant increases in plasma volume, while the increase in plasma volume was not significant in pigs that were given a high energy ration. An increase in plasma volume as a factor in the development of anaemia in trypanosome infected animals has been documented (Katunguka-Rwakishaya, Murray & Holmes, 1992). It would appear that high energy intake may not prevent the rapid establishment of trypanosomes following inoculation but the ensuing anaemia is less severe than that observed in animals receiving a low energy ration.

Recovery from anaemia and erythropoietic responses

Studies conducted in cattle suggested that the rate of recovery from anaemia was faster in animals supplemented with groundnut cake (Agyemang *et al.* 1990*a*). In sheep infected with *T. congolense* and given either a low or a high protein diet, it was observed that following treatment with a trypanocidal drug (isometamidium chloride) at 70 days after infection, animals given a high protein diet recovered from anaemia much faster than those receiving a low protein diet (Katunguka-Rwakishaya *et al.* 1993). This was associated with macrocytosis in the high protein infected group as opposed to normocytosis in the low protein infected group. A further experiment was conducted using similar protein levels to the previous studies, to investigate the influence of dietary protein on erythropoietic responses in sheep by using the Evans Blue dilution technique to measure plasma volume and [^{59}Fe] ferric citrate to assess red cell synthesis based on plasma iron turnover rates (PITR) and red cell iron utilization (Katunguka-Rwakishaya, 1992). Infected animals given the low protein diet had a significantly lower circulating red cell volume than uninfected control animals but there were no differences between the high protein groups. Similarly, infection and dietary protein apparently had no influence on total blood volumes.

Ferrokinetic measurements indicated that the plasma [^{59}Fe] half lives and estimated red cell lifespan were lower while the PITR, [^{59}Fe] utilization and RBC [^{59}Fe] incorporation rates were higher in infected animals than in control animals. This is consistent with enhanced erythropoietic activity in infected animals particularly during the early stages of infection. Another interesting observation was that PITR and RBC [^{59}Fe] incorporation rates were higher in the infected animals given high protein than those given low protein although the differences were not statistically significant. This supports previous observations and suggests that improved protein nutrition enhances erythropoietic activity even as early as 4 weeks after infection. This could explain the faster rate of recovery from anaemia recorded in previous studies (Katunguka-Rwakishaya *et al.* 1993). It can therefore be concluded that dietary protein has a major influence on erythropoietic activity which accounts for improvement in PCV values especially in long-standing infections.

Serum lipid fractions

It was observed that *T. congolense* infection in sheep is associated with profound biochemical changes some of which are modulated by dietary protein or energy. Infected animals showed significant decreases in plasma cholesterol and serum phospholipid concentrations with resultant decrease in serum total lipids. The decline in these fractions was greater in animals receiving low protein or low energy rations compared to those receiving high protein or energy rations.

A reduction in serum cholesterol and phospholipids confirms previous observations in sheep infected with *T. congolense* by Roberts *et al.* (1977) and Traore-Leroux, Fumoux & Pinder (1987). There is evidence that lipids constitute 15–20% of trypanosomal dry weight (Venkatessan & Ormerod, 1976) and that trypanosomes obtain cholesterol from the host by uptake and degradation of low density (Coppens *et al.* 1987; Gillet & Owen, 1987) or high density lipoproteins (Traore-Leroux *et al.* 1987). It has also been demonstrated that trypanosomes require cholesterol for growth and multiplication (Black & Vanderweed, 1989) and that it is the main sterol in trypanosomes (Carrol & McCroire, 1986). In addition, the trypanosomes take up free fatty acids which may be circulating freely (Tizard *et al.* 1978) or bound to albumin (Vickerman & Tetley, 1979). The decline in serum cholesterol commences with the appearance of trypanosomes in the circulation. While uptake by trypanosomes might seem to largely account for the decline in serum cholesterol, it may not be entirely so because even at the peak of parasitaemia, total cholesterol content of the trypanosomes may not be sufficiently high to have any impact on the serum concentration.

It was observed that control animals receiving a high protein diet had higher concentrations of serum total lipids, cholesterol and non-esterified fatty acids than those given the low protein diet. It has also been observed that high cholesterol concentrations are associated with higher parasite numbers (Traore-Leroux *et al.* 1987). It therefore appears that provision of higher protein has a sparing effect on lipid metabolism of the host, making these nutrients available for trypanosome growth while at the same time, the host is able to continue gaining weight.

Albumin metabolism

Infected animals on all dietary treatments developed hypoalbuminaemia and hypoproteinaemia which were more severe in animals receiving low protein and energy ration compared to those on high protein and energy rations. In addition, control animals on high protein diet had higher concentrations of plasma albumin and serum urea. These observations support those of Otesile, Fagbemi & Adeyemo (1991) in boars placed on different energy levels and infected with *T. brucei*. It has been suggested that the degree of hypoalbuminaemia may be related to the severity of trypanosome infection (Holmes, 1976) and it is possible that trypanosomal uptake of albumin-bound fatty acids and lipoproteins for their metabolism and growth (Vickerman & Tetley, 1979) and haemodilution (Holmes, 1976) may account for the decrease in plasma albumin. The observation that the decrease in plasma albumin concentration was mild in animals receiving high protein and energy rations indicates that adequate protein and energy nutrition enhances the ability of trypanosome infected animals to withstand the adverse effects of infection.

INFLUENCE OF TRYPANOSOME INFECTIONS ON THE DIGESTIVE PHYSIOLOGY OF SHEEP

Similarly controlled studies were conducted in Scottish Blackface wethers given diets of different compositions in order to investigate the effects of trypanosome infections on feed intake, digestive function and gross nitrogen balance. Studies were performed on the apparent digestibility of the diets and also the rate of passage of the roughage component of the diet using chromium as a marker (Uden *et al.* 1980, 1982).

In order to distinguish the effects of a reduced feed intake from the other effects of infection, the animals were matched in pairs and the control animal was offered the average amount of food consumed by its infected counterpart during the previous 2 days (pair-feeding regime). In one such study (A), eight sheep arranged in four pairs, were each offered a diet of chopped barley straw *ad libitum* plus a pelleted barley/soyabean mixture supplying 70 g metabolizable protein (MP) and 8·3 MJME/d. Another eight sheep similarly arranged in four pairs, were offered a diet of chopped lucerne hay *ad libitum* plus the same quantity (366 g DM) of a pelleted barley concentrate supplying 140 g MP and 13·3 MJME/d (Wassink *et al.* 1997). One animal of each pair was infected with *T. congolense* 1180 (Nantulya *et al.* 1984).

In a second study (B) also reported by Wassink *et al.* (1997), each sheep of four pairs received 200 g DM grass hay and 425 g DM crushed barley grain (plus minerals) in the morning and barley straw *ad libitum* in the afternoon (low roughage/high concentrate diet; 60 g MP, 10 MJME/d). The sheep in the other four pairs were offered 400 g DM grass hay and 315 g DM crushed barley grain (plus minerals) in the morning and barley straw *ad libitum* in the afternoon (high roughage/low concentrate diet; 60 g MP, 10 MJME/d). Again, one animal of each pair was infected with *T. congolense 1180* (Nantulya *et al.* 1984).

The third study (C) described by Wassink (1997) was designed so that each sheep of four pairs received 150 g DM grass hay and 319 g DM crushed barley grain (plus mineral mix) in the morning and barley straw *ad libitum* in the afternoon (low roughage/high concentrate diet; 45 g MP, 7 MJME/d). The sheep in the other four pairs were offered 300 g DM grass hay and 236 g DM crushed barley grain (plus mineral mix) in the morning and barley straw *ad libitum* in the afternoon (high roughage/low concentrate diet; 45 g MP, 7 MJME/d). One animal of each pair was infected with *T. vivax* strain Y486, isolated by Leeflang, Ige & Olatunde (1976). In contrast to study A, in which there was a large difference in the level of nutrition between the two dietary groups, in studies (B) and (C) the levels of energy and protein were similar between the two dietary groups. However, as the source of protein was different between the dietary groups, the proportion of effective rumen degradable protein (ERDP) and digestible undegraded protein (DUP) intake was different.

The general course of the *T. congolense* infections in the first two experiments followed the same pattern as previously found by Katunguka-Rwakishaya (1992) using the same strain of trypanosomes in Scottish Blackface sheep. Parasitaemia levels were relatively low and packed cell volume (PCV) decreased only moderately, but significantly ($P < 0{\cdot}01$), between day 10 and 20 post infection. PCV levels stabilized after 20 days post infection.

The effects of the disease were much more pronounced in the sheep infected with *T. vivax* (C). These sheep had higher levels of parasitaemia throughout the study period compared to those infected with *T. congolense*. PCV levels declined significantly ($P < 0{\cdot}01$) post infection and, unlike the *T. congolense* infection, continued to decline towards the end of the experiment. The effects of the disease on plasma cholesterol and albumin levels were also greater in the *T. vivax*-infected sheep compared to those infected with *T. congolense*. However, no significant interaction effects were found between the diets and the blood parameters measured.

Feed intake

In all three studies, the depression in feed intake due to the trypanosome infection was relatively small compared to previous experiments in both sheep (Akinbamijo *et al.* 1994*a*, *b*) and goats (Verstegen *et al.* 1991; Akinbamijo *et al.* 1992). The apparently more virulent *T. vivax* infection (C) did not result in a greater decrease in feed intake compared with the milder *T. congolense* infection (A and B). The slight depression in total feed intake that was observed in the last two experiments was mostly due to the reduction in the intake of the *ad libitum* barley straw.

Body weight gain

The relatively small effect of the infection on the feed intake may be partly reflected in the data on body weight gain, which in A was no different between infected animals and their pair-fed controls. Although significant differences were found in body weight gains in (B) ($P < 0{\cdot}05$) and (C) ($P < 0{\cdot}01$) between infected sheep and their pair-fed controls, those differences were small and not thought to be of biological significance.

Apparent digestibility

Organic matter digestibility, OMD, was significantly lower in the *T. congolense*-infected sheep in study (A) ($P < 0{\cdot}01$) and (B) ($P < 0{\cdot}05$) with no interaction effect being observed between the diet and infection. In study (A), OMD of the pair-fed control animals increased and this may have been due to the below potential feed intake in these animals as a result of being paired to the infected ones. The OMD of the *T. vivax*-infected sheep was not found to be significantly different to the OMD of their pair-fed controls. These results indicate no apparent effect of infection on the OMD of the diets. Van Dam (1996) also found no evidence of a decrease in organic matter digestibility in *T. vivax* infected West African Dwarf goats fed lucerne pellets or grass straw.

Crude protein digestibility was also significantly lower in the infected sheep of study (A). However, unlike OMD, the digestibility of crude protein of the pair-fed control animals did not change during the experiment, but the crude protein digestibility values of the infected animals decreased. Again, no interaction between the diet and infection was observed. In studies (B) and (C), the nitrogen digestibility values also appeared to be lower in the infected sheep compared to their pair-fed controls, but most differences were not statistically significant. As the infected animals and their controls were pair-fed, the difference could be attributed mostly to a slightly higher faecal nitrogen excretion in the infected animals of studies (B) and (C) compared to their pair-fed controls. Whether the source of this extra faecal nitrogen excretion was dietary or endogenous was not clear and requires further investigation.

Mean retention time of the roughage in the digestive tract

The mean retention time of the roughage in the digestive tract was found to be significantly longer in the *T. congolense*-infected sheep in study (A). No significant interaction effect was found between the type of diet and infection. A similar effect was observed in study (B) with the *T. congolense*-infected animals having a significantly longer mean retention time than their pair-fed controls and this effect was more pronounced in the animals on the low roughage/high concentrate diet.

The mean retention time of the *T. vivax*-infected sheep (study C) was only slightly longer than that of their pair-fed controls ($P < 0.05$) and in this experiment no interaction between diet and infection was observed. These results indicate an effect of trypanosome infections on the mean retention time of roughage in the digestive tract irrespective of the pathogenicity of the disease. There was some evidence to suggest that the type of diet influenced the effects of a *T. congolense* infection on the mean retention time. No such evidence was found in the *T. vivax*-infected animals of study (C).

Nitrogen retention

Although there was a tendency towards a lower nitrogen retention in *T. congolense*-infected sheep compared to their respective pair-fed controls none of the differences were statistically significant. In the *T. vivax* study (C), however, differences were found to be statistically significant, with the infected sheep having a lower nitrogen retention than their pair-fed controls. Significantly higher urinary nitrogen excretion and to a lesser extent a higher faecal nitrogen excretion appear to be responsible for this lower nitrogen retention in the infected animals. Van Dam (1996) and Akinbamijo *et al.* (1994*a*, *b*) also reported a lower nitrogen retention in *T. vivax*-infected West African Dwarf goats and West African Dwarf sheep, respectively, compared with the controls. However, no change in the efficiency of nitrogen utilization was found and the difference could be attributed to a decreased feed intake in the infected animals.

A significant interaction effect was also found between infection and diet with the infected animals on the low roughage/high concentrate diet being affected more by the infection in terms of nitrogen retention than the ones on the high roughage/low concentrate diet. Close examination of the data revealed that the lower nitrogen retention in the *T. vivax*-infected sheep offered the low roughage/high concentrate diet was mainly caused by a higher urinary nitrogen excretion in these animals. There was, however, a high variation in response between animals. It was calculated that the digestible undegraded protein (DUP) intake of the sheep was lower in the sheep fed low roughage/high concentrate diet compared to those on the high roughage/low concentrate diet (1 and 1·5 g/kg $M^{0.75}$/day respectively). Whether this factor was involved in the difference between the dietary groups needs further investigation.

It was concluded in these carefully controlled nutrient intake studies that trypanosome infections did indeed affect the digestive function of sheep, an outcome that previously had been partly demonstrated but with some doubts being raised over the experimental designs employed. Here clearly nitrogen digestibility was lower in the infected animals, but organic matter digestibility was unaffected. The mean retention time of the roughage in the digestive tract was significantly longer in infected animals, which was found to be affected by the type of diet in the case of *T. congolense*. However, the pathogenicity of the trypanosome infection did not appear to affect the level of changes in digestive function.

The nitrogen retention of the *T. congolense*-infected sheep was similar to that of their pair-fed controls. In contrast, the nitrogen retention of the *T. vivax*-infected sheep was significantly lower than that of their pair-fed controls. This lower nitrogen retention was mainly due to a higher urinary nitrogen excretion and was significantly affected by the type of diet.

CONCLUSIONS

Across Africa, cattle infected with trypanosomes frequently also suffer from undernutrition and the impact that nutrition has on the pathophysiology of bovine trypanosomiasis is recognized as a key area for investigation where the outcomes may afford practical husbandry strategies to counter the effects of the disease. This review has attempted to collate relevant recent work specifically designed to determine the direct and compounded effects of nutrition on the disease, digestive efficiency and productivity.

It is apparent from the studies described in this review that nutrition can have a profound effect on the pathophysiology of bovine trypanosomiasis. The severe problems of impaired growth or weight loss, reduced productivity and increased mortality associated with trypanosomiasis are considerably exacerbated by undernutrition. It is noteworthy that almost all the studies carried out to date have been conducted in breeds of ruminants which show significant resistance or trypanotolerance to trypanosomiasis such as N'Dama cattle, West African sheep and goats and Scottish Blackface sheep. Nevertheless, even in such breeds the impact of undernutrition is clearly evident. It can be anticipated that the effects in trypanosusceptible breeds are likely to be even more pronounced.

Although there are differences between the various studies described in terms of the species, age and history of the hosts, species and strains of trypanosomes, and the level and composition of the feed, the results of the numerous studies do allow a number of tentative conclusions to be drawn. These are best described by examining the effects that nutrition has on the principal clinical and pathophysiological features of trypanosomiasis. These features include anaemia, pyrexia, body weight changes, reduced feed intake and diminished productivity.

Anaemia is characteristic of trypanosomiasis and begins with the first waves of parasitaemia and is progressive. It is clear that nutrition does not influence the onset of the anaemia but the rate of its subsequent development and severity may be affected by the levels of energy intake with lower levels being frequently associated with more rapid development of the anaemia. Following treatment higher protein levels are associated with a more rapid recovery of the packed cell volume.

Pyrexia also follows the onset of parasitaemia and is cyclical with the waves of parasitaemia. It is also associated with increased energy demands for maintenance which is ultimately manifested by reduction in voluntary activity levels and reduced productivity.

Body weight changes (rate of gain or loss) in trypanosomiasis are particularly influenced by the levels of protein intake. High intakes allow infected animals to grow at the same rate as uninfected control animals and indicate that as in the case of gastrointestinal helminthiasis high protein intakes can largely ameliorate the adverse affects of parasitism providing energy intake is adequate whilst low energy levels can exacerbate the adverse effects of trypanosomiasis on body weight.

Reductions in feed intake are common in parasitized animals and this is a feature of trypanosomiasis. However, this is less apparent in animals which are provided with high protein diets, since there are benefits from both the quality and quantity of the dietary intake resulting in significantly reduced impairment from the disease. Where intake is limited by the disease, animals will often exhibit preferential selection of higher quality browse. Although feed selection effects are still difficult to evaluate and the apparent preference for the 'better food' does not always occur.

A common feature of trypanosomiasis is impaired productivity including reduced draught work output, lower milk yields and reduced reproductive capacity. All of these adverse features can be ameliorated to a considerable extent by the provision of higher quality feeds which helps maintain appetite, reduces the severity of the anaemia and supplies energy for work in addition to that required for increased maintenance.

It can therefore be concluded that increased dietary protein and energy intakes ameliorate the effect of trypanosome infection on growth rate and that it would appear that high energy intake may not prevent the rapid establishment of trypanosomes following inoculation but the ensuing anaemia is less severe than that observed in animals receiving a low energy ration. The work reviewed here reinforces the finding that dietary protein has a major influence on erythropoietic activity which accounts for improvement in PCV values especially in long-standing infections.

Despite these tentative conclusions many aspects of the interactions between nutrition and trypanosomiasis remain unclear. The areas which require further study include detailed evaluations of the minimum levels of protein and energy supplementation required to ameliorate the adverse effects of trypanosomiasis, the nature and quality of the protein supplement to achieve these benefits and the influence these have on digestive physiology. Through such knowledge, practical husbandry strategies may be developed, which will allow animals to survive and be productive in many of the large areas of Africa infested with tsetse flies.

REFERENCES

AGYEMANG, K., DWINGER, R. H., JEANNIN, P., LAPERRE, P., GRIEVE, A. S., BAH, M. L. & LITTLE, D. A. (1990*a*). Biological and economic impact of trypanosomes infections on milk production in N'Dama cattle managed under village conditions in The Gambia. *Animal Production* **50**, 383–389.

AGYEMANG, K., DWINGER, R. H., LITTLE, D. A., LAPERRE, P. & GRIEVE, A. S. (1992). Interactions between physiological status in N'Dama cows and trypanosome infections and its effect on health and productivity of cattle in The Gambia. *Acta Tropica* **50**, 91–99.

AGYEMANG, K., DWINGER, R. H., TOURAY, B. N., JEANNIN, P., FOFANA, D. & GRIEVE, A. S. (1990*b*). Effects of nutrition on the degree of anaemia and liveweight changes in N'Dama cattle infected with trypanosomes. *Livestock Production Science* **26**, 39–51.

AKINBAMIJO, O. O., ADEMOSUM, A. A., ZWART, D., TOLKAMP, B. J. & BROUWER, B. O. (1990). Effect of *Trypanosoma brucei* infection on liveweight, organic matter intake, digestibility and N-balance in West African Dwarf goats. *Tropical Veterinarian* **8**, 140–148.

AKINBAMIJO, O. O., BENNISON, J. J., ROMNEY, D. L., WASSINK, G. J., JAITNER, J., CLIFFORD, D. J. & DEMPFLE, L. (1997). An evaluation of food intake, digestive physiology and live-weight changes of N'Dama and Gobra zebu bulls following experimental *Trypanosoma congolense* infection. *Animal Science* **65**, 151–158.

AKINBAMIJO, O. O., HAMMINGA, B. J., WENSING, T., BROUWER, B. O., TOLKAMP, B. J. & ZWART, D. (1992). The effect of *Trypanosoma vivax* infection in West African Dwarf goats on energy and nitrogen metabolism. *The Veterinary Quarterly* **14**, 95–100.

AKINBAMIJO, O. O., REYNOLDS, L. & GORT, G. (1994*a*). Effects of *Trypanosoma vivax* infection during pregnancy on food intake, nitrogen retention and liveweight changes in West African Dwarf ewes. *Journal of Agricultural Science* **123**, 379–385.

AKINBAMIJO, O. O., REYNOLDS, L., SHERINGTON, J. & NSAHLAI, I. V. (1994*b*). Effects of post-partum *Trypanosoma vivax* infection on feed intake, liveweight changes, milk yield and composition in West African Dwarf ewes and associated lamb growth rates. *Journal of Agricultural Science* **123**, 387–392.

BARRY, J. D. & TURNER, C. M. R. (1991). The dynamics of antigenic variation and growth of African trypanosomes. *Parasitology Today* **7**, 207–211.

BENNISON, J. J. (1997). *The Effects of Nutrition and Trypanosomiasis on the Productivity of Trypanotolerant N'Dama Cattle*. Ph.D. thesis, Wye College, University of London.

BENNISON, J. J., AKINBAMIJO, O. O., JAITNER, J., DEMPFLE, L., HENDY, C. R. & LEAVER, J. D. (1999). Effects of nutrition *pre-partum* and *post-partum* on subsequent productivity and health of N'Dama cows infected with *Trypanosoma congolense*. *Animal Science* **68**, 819–829.

BENNISON, J. J., CLEMENCE, R. G., ARCHIBALD, R. F., HENDY, C. R. C. & DEMPFLE, L. (1998*a*). The effects of work and two planes of nutrition on trypanotolerant draught cattle infected with *Trypanosoma congolense*. *Animal Science* **66**, 595–605.

BENNISON, J. J., SHERINGTON, J., WACHER, T. J., DEMPFLE, L. & LEAVER, J. D. (1998*b*). Effects of *Trypanosoma congolense* infection and groundnut (*Arachis hypogaea*) hay supplementation on ranging, activity and diet selection of N'Dama cows. *Applied Animal Behaviour Science* **58**, 1–12.

BLACK, S. & VANDERWEERD, V. (1989). Serum lipoproteins are required for multiplication of *Trypanosoma brucei brucei* under axenic culture conditions. *Molecular and Biochemical Parasitology* **37**, 65–72.

BOURN, D. & SCOTT, M. (1978). The successful use of work oxen in agricultural development of tsetse infected land in Ethiopia. *Tropical Animal Health and Production* **10**, 191–203.

CARROL, M. & McCROIRE, P. (1986). Lipid composition of blood stream forms of *Trypanosoma brucei brucei*. *Comparative Biochemical Physiology* **83B**, 647–651.

COOP, R. L. & HOLMES, P. H. (1996). Nutrition and parasite interaction. *International Journal for Parasitology* **26**, 951–962.

COPPENS, I., OPPERDOES, F. R., COURTOY, P. J. & BAUDHUIN, P. (1987). Receptor-mediated endocytosis in the bloodstream form of *Trypanosoma brucei*. *Journal of Protozoology* **34**, 465–473.

DWINGER, R. H., CLIFFORD, D. J., AGYEMANG, K., GETTINBY, G., GRIEVE, A. S., KORA, S. & BOJANG, M. A. (1992). Comparative studies on N'Dama and Zebu cattle following repeated infections with *Trypanosoma congolense*. *Research in Veterinary Science* **52**, 292–298.

FAGBEMI, B. O., OTESILE, E. B., MAKINDE, M. O. & AKINBOADE, O. A. (1990). The relationship between dietary energy levels and the severity of *Trypanosoma brucei* infection in growing pigs. *Veterinary Parasitology* **35**, 29–42.

GILLETT, M. P. T. & OWEN, J. S. (1987). *Trypanosoma brucei brucei* obtains cholesterol from plasma lipoproteins. *Biochemical Society Transactions* **15**, 258–259.

HECKER, P. A., COULIBALY, L., ROWLANDS, G. S., NAGDA, S. M. & d'ITEREN, G. D. M. (1991). Effect of plane of nutrition on trypanosome prevalence and mortality of Djallonke sheep exposed to high tsetse challenge. In *Proceedings of the 21st Meeting of the International Scientific Council for Trypanosomiasis Research and Control*, Cote d'Ivore, ISCTRC, Yamoussoukro, 21–25th October, 1991.

HOLMES, P. H. (1976). The use of radioisotopic tracer techniques in the study of the pathogenesis of trypanosomiasis. In *Nuclear Techniques in Animal Production and Health*. Vienna, International Atomic Energy Agency, pp. 663–674.

HOLMES, P. H. & BREMNER, K. C. (1971). The pathophysiology of ovine ostertagiaisis. Water balance and turnover studies. *Research in Veterinary Science* **12**, 381–383.

KATUNGUKA-RWAKISHAYA, E. (1992). *The Pathophysiology of Ovine Trypanosomiasis Caused by* Trypanosoma congolense. Ph.D. thesis, University of Glasgow, Scotland: UK.

KATUNGUKA-RWAKISHAYA, E., MURRAY, M. & HOLMES, P. H. (1992). Pathophysiology of ovine trypanosomiasis: Ferrokinetic and erythrocyte survival studies. *Research in Veterinary Science* **53**, 80–86.

KATUNGUKA-RWAKISHAYA, E., PARKINS, J. J., FISHWICK, G., MURRAY, M. & HOLMES, P. H. (1993). The pathophysiology of *Trypanosoma congolense* infection in Scottish Blackface sheep: Influence of dietary protein. *Veterinary Parasitology* **47**, 189–204.

KATUNGUKA-RWAKISHAYA, E., PARKINS, J. J., FISHWICK, G., MURRAY, M. & HOLMES, P. H. (1995). The influence of energy intake on the pathophysiology of *Trypanosoma congolense* infection in Scottish Blackface sheep. *Veterinary Parasitology* **59**, 207–218.

KYRIAZAKIS, I., OLDHAM, J. D., COOP, R. L. & JACKSON, F. (1994). The effect of subclinical intestinal nematode infection on the diet selection of growing sheep. *British Journal of Nutrition* **72**, 665–677.

LEEFLANG, P., IGE, K. & OLATUNDE, D. S. (1976). Studies on *Trypanosoma vivax*: The infectivity of cyclically and mechanically transmitted ruminant infections for mice and rats. *International Journal for Parasitology* **6**, 453–456.

LITTLE, D. A., DWINGER, R. H., CLIFFORD, D. J., GRIEVE, A. S., KORA, S. & BOJANG, M. (1990). Effect of nutritional level and body condition on susceptibility of N'Dama cattle to *Trypanosoma congolense* infection in The Gambia. *Proceedings of the Nutrition Society* **49**, 209A.

McCARTHY, D. O., KLUGER, M. J. & VANDER, A. J. (1985). Suppression of food intake during infection: is Interleukin-1 involved? *American Journal of Clinical Nutrition* **42**, 1179–1182.

MAKINDE, M. O., OTESILE, E. B. & FAGBEMI, B. O. (1991). Studies on the relationship between dietary energy levels and the severity of *Trypanosoma brucei* infection: The effects of diet and infection on blood and plasma volumes and the erythrocyte osmotic fragility of growing pigs. *Bulletin of Animal Health and Production in Africa* **39**, 161–166.

MURRAY, M. & BLACK, S. J. (1985). African trypanosomiasis in cattle: working with nature's solution. *Veterinary Parasitology* **18**, 167–182.

MURRAY, M., CLIFFORD, D. J., GETTINBY, G., SNOW, W. F. & McINTYRE, W. I. M. (1981). Susceptibility to African trypanosomiasis of N'Dama and Zebu cattle in an area of *Glossina morsitans submorsitans* challenge. *The Veterinary Record* **109**, 503–510.

MURRAY, M. & DEXTER, T. M. (1988). Anaemia in African trypanosomiasis. A review. *Acta Tropica* **45**, 389–432.

NANTULYA, V. M., MUSOKE, A. J., RURANGIRWA, F. R. & MOLOO, S. K. (1984). Resistance of cattle to tsetse-transmitted challenge with *Trypanosoma brucei* and *Trypanosoma congolense* after spontaneous recovery from syringe passaged infections. *Infection and Immunity* **43**, 735–738.

OTESILE, E. B., FAGBEMI, B. O. & ADEYEMO, O. (1991). The effect of infection on some serum biochemical parameters in boars on different planes of dietary energy. *Veterinary Parasitology* **40**, 207–216.

PALING, R. W., MOLOO, S. K., SCOTT, J. R., GETTINBY, G., MCODIMBA, F. A. & MURRAY, M. (1991). Susceptibility of N'Dama and Boran cattle to sequential challenges with tsetse transmitted clones of *Trypanosoma congolense*. *Parasite Immunology* **13**, 427–445.

PARKINS, J. J., BAIRDEN, K. & ARMOUR, J. (1982). *Ostertagia ostertagi* in calves: a growth, nitrogen balance and digestibility study conducted during winter feeding following thiabendazole anthelmintic therapy. *Journal Comparative Pathology* **92**, 219–227.

PARKINS, J. J. & HOLMES, P. H. (1989). Effects of gastrointestinal parasites on ruminant nutrition. *Nutrition Research Reviews* **2**, 227–246.

REYNOLDS, L. & EKWURUKE, J. O. (1988). Effect of *Trypanosoma vivax* infection on West African Dwarf sheep at two planes of nutrition. *Small Ruminant Research* **1**, 175–188.

ROBERTS, C. J., TIZARD, I. R., MELLORS, A. & CLARKSON, M. J. (1977). Lysophospholipases, lipid metabolism and the pathogenesis of African trypanosomiasis. *The Lancet* December 3, 1187–1188.

ROMNEY, D. L., N'JIE, A., CLIFFORD, D. J., HOLMES, P. H., RICHARD, D. & GILL, M. (1997). The influence of nutrition on the effects of infection with *Trypanosoma congolense* in trypanotolerant cattle. *Journal of Agricultural Science* **129**, 83–89.

TIZARD, I., NIELSEN, K. H., SEED, J. R. & HALL, J. E. (1978). Biological active products from African trypanosomes. *Microbiological Reviews* **42**, 661–681.

TRAORE-LEROUX, T., FUMOUX, F. & PINDER, M. (1987). High density lipoprotein levels in the serum of trypanosensitive and trypanoresistant cattle. Changes during *Trypanosoma congolense* infection. *Acta Tropica* **44**, 315–323.

UDEN, P., COLUCCI, P. E. & VAN SOEST, P. J. (1980). Investigation of Chromium, Cerium and Cobalt as Markers of Digesta. Rate of Passage Studies. *Journal of the Science of Food and Agriculture* **31**, 625–632.

UDEN, P., ROUNSAVILLE, T. R., WIGGANS, G. R. & VAN SOEST, P. J. (1982). The measurement of liquid and solid digesta retention in ruminants, equines, and rabbits given timothy (*Phleum pratense*) hay. *British Journal of Nutrition* **48**, 329–339.

VAN DAM, J. T. P. (1996). *The Interaction Between Nutrition and Metabolism in West African Dwarf Goats, Infected with Trypanosomes*. Ph.D. thesis, Wageningen Agricultural University: The Netherlands.

VAN DAM, J. T. P., VAN DER HEIDE, D., VAN DER HEL, W., VAN DEN INGH, T. S. G. A. M., VERSTEGEN, M. W. A., WENSING, T. & ZWART, D. (1996). The effects of *Trypanosoma vivax* infection on energy and protein metabolisms and serum metabolites and hormones in West African Dwarf goats on different food intake levels. *Animal Science* **63**, 111–121.

VAN HOUTERT, M. F. J. & SYKES, A. R. (1996). Implications of nutrition for the ability of ruminants to withstand gastrointestinal nematode infections. *International Journal for Parasitology* **26**, 1151–1168.

VAN MIERT, A. S. J. P. A. M., VAN DUIN, C. T. M. & KOOT, M. (1990). Effects of *E. coli* endotoxin, some interferon inducers, recombinant interferon-a_{2a} and *Trypanosoma brucei* infection on feed intake in dwarf goats. *Journal of Veterinary Pharmacology and Therapeutics* **13**, 327–331.

VENKATESAN, S. & ORMEROD, W. E. (1976). Lipid content of the slender and stumpy forms of *Trypanosoma brucei rhodesiense*: a comparative study. *Comparative Biochemistry and Physiology* **53B**, 481–487.

VERSTEGEN, M. W. A., ZWART, D., VAN DER HEL, W., BROUWER, B. O. & WENSING, T. (1991). Effect of *Trypanosoma vivax* infection on energy and nitrogen metabolism of West African dwarf goats. *Journal of Animal Science* **69**, 1667–1677.

VICKERMAN, K. & TETLEY, L. (1979). Biology and ultrastructure of trypanosomes in relation to pathogenesis. In *Pathogenicity of Trypanosomes* (ed. Losos, G. & Chouinard, Amy.), pp. 23–31. Ottawa: Canada, IDRC 132e.

WACHER, T. J. (1993). Interactions of season and trypanosome infection with ranging, activity and diet selection parameters in individual village managed N'Dama cattle in The Gambia. *Consultancy Report for the Natural Resources Institute (NRI)*, January 1993.

WASSINK, G. J. (1997). *The Effect of Nutrition on the Pathophysiology of Trypanosomiasis in Scottish Blackface Sheep*. Ph.D. thesis, University of Glasgow: UK.

WASSINK, G. J., FISHWICK, G., PARKINS, J. J., GILL, M., ROMNEY, D. L., RICHARD, D. & HOLMES, P. H. (1997). The patho-physiology of *Trypanosoma congolense* in Scottish Blackface sheep: influence of diet on digestive function. *Animal Science* **64**, 127–137.

ZWART, D., BROUWER, B. O., VAN DER HEL, W., VAN DEN AKKER, H. N. & VERSTEGEN, M. W. A. (1991). Effect of *Trypanosoma vivax* on body temperature, feed intake and metabolic rate of West African dwarf goats. *Journal of Animal Science* **69**, 3780–3788.