AB

TRYPANOTOLERANCE IN DJALLONKE SHEEP AND WEST AFRICAN DWARF GOATS IN THE GAMBIA.

Importance of trypanosomosis, nutrition, helminth infections and management factors.

Sabine Osaer

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TRYPANOTOLERANTIE IN DJALLONKE SCHAPEN EN WEST AFRIKAANSE DWERGGEITEN IN GAMBIA

Belang van trypanosomosis, voeding, wormbesmettingen en management factoren

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CHAPTER 1

GENERAL INTRODUCTION

1. INTRODUCTION

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1. GENERAL INTRODUCTION

1.1. Trypanosomosis in sub-Saharan Africa

Approximately 10 million km^2 of sub-Saharan Africa is infested by the tsetse fly, the vector of trypanosomosis. Due to this disease, livestock production cannot be fully exploited in these areas (FAO, 1992). These areas, however, hold the greatest potential for expansion of agricultural output (Winrock, 1992). In mixed farming systems, trypanosomosis may constrain the number of draught animals kept and therefore reduce the average area planted. In addition, trypanosomosis causes direct losses in different breeds and species of livestock through reduced calving rates (1-20 %), increased calf mortality by (0-20 %), reduced milk off take (10-26 %) and reduced lambing and kidding rates (4-38%) (Swallow, 1997). All mammals including man are potential hosts for tsetse flies, the vectors which are able to transmit pathogenic trypanosome species. Certain wild life species are not suffering from clinical signs of trypanosomosis when infected but are important as potential reservoir for domestic livestock (Claxton et al., 1992; Weitz, 1963).

There is no easy solution to solve the problem of trypanosomosis. Chemoprophylactic or curative trypanocidal drugs are expensive and not always available for poorer sections of the population or in remote areas of the countries. Since animal trypanosomosis occurs in poor countries, development of new drugs is commercially unattractive and the development of new trypanocidals is unlikely in the near future. Moreover, resistance against trypanocidal drugs is becoming a serious problem and has already been reported in 13 countries in sub-Saharan Africa (FAO, 1992; Peregrine, 1994; Geerts & Holmes, 1998). Reduction or eradication of the vector, the tsetse fly can be achieved using various physical or chemical agents but is expensive and might cause environmental pollution. The use of insecticide impregnated targets to control the fly is feasible in certain regions but needs continued investments to avoid re-infestation of the area. Furthermore, it is unlikely that in the near future a vaccine will be developed providing full protection against all different trypanosome species and all their variable antigen types (Murray et al., 1982) due to the existence of variable surface glycoproteins (VSG), although recently some promising reports were published with reference to the use of flagellar pocket antigens (Mkunza et al., 1995).

The above control methods are very often not sustainable, especially when used over longer periods with the aim of increasing animal production. Over the last decade increasing efforts were put into investigating the possible role of breeds of livestock with a natural resistance against trypanosomosis. These breeds, most of them indigenous to West and Central Africa, are called trypanotolerant because they survive and remain productive in tsetse infested areas where other breeds do not survive without treatment (Murray et al. 1982). Trypanotolerant cattle breeds include West African Longhorn (N'Dama) and West African Shorthorn (Baoule, Lagune, Muturu) as the most important ones. Trypanotolerant small ruminants mainly include Djallonke sheep and West African Dwarf (WAD) goats. These trypanotolerant breeds represent only small percentages of the total livestock population in Africa (6 % of the cattle, 6 % of the sheep and 11 % of the goats). However, their numbers are slowly increasing (Hoste et al., 1988). The ability of any animal species to succeed depends on its ability to adapt to local nutritional, environmental and health related circumstances (French, 1970). Based on these criteria, N'Dama cattle, Djallonke sheep and West African Dwarf goats are very well adapted. Trypanotolerant breeds of small ruminants are of a relatively small size, but have proven to be productive under difficult conditions (Wilson et al., 1989). This indicates their potential role for a more sustainable solution to the growing demand for animal protein in much of tsetse infested Africa.

1.2. The Gambia - general information and statistics.

1.2.a. The country and its population.

The Gambia is a small country in West Africa of some 400 km long and 30 km wide on both sides of the river Gambia. It stretches between 13°54' and 13°20' North and between 17°05' and 14°05' West. The Gambia covers an area of 11,300 km^2 with a total land area of 10,689 km^2 and is completely surrounded by Senegal except for a coastal stretch of 30 km.

Its population is 1.2 million (1996) with a natural annual increase of 2.7 % which implies a doubling of the current population in 26 years (PRB, 1996). Population density is one of the highest in Africa with on average 96 inhabitants per km^2. Fourty percent of the population is living in urban centres with an annual urban growth rate of 6% (FAO, 1997). This high population density and urban growth is a serious constraint to development. The Gambia, having an annual per caput income (1996/1997) of 350 US $, (UNDP, 1998) is among the poorest and least developed countries.

About 79 % of the economic active population is working in the agricultural sector, however, with a decreasing trend. The contribution of the agricultural and natural resources' sector to the GDP was 40 % in 1980 but declined to 20 % in 1997 (UNDP, 1998). The annual value of agricultural imports is three times higher than the export. The national calorie intake is clearly insufficient and The Gambia relies for half of its total staple food requirements on importation and donor aid. The Gambia is facing serious problems concerning the development of infrastructure and agriculture development in order to be able to feed its growing population.

1.2.b. Climate and vegetation.

The Gambia is situated in the Sudano-Sahelian agro-ecological zone. This zone varies from semi-arid (600-1000 mm per annum) in-land to subhumid (more than 1000 mm per annum) at the coast. The humid and rainy season lasts from July to October with peak rainfall in August. The dry season lasts from November to June. Temperature varies between 14-40°Celsius. The vegetation is mainly savannah woodland with swamp areas.

The Gambia faces a problem of environmental degradation and shows early indicators of desertification (Schwartz, 1999). Constant overgrazing of natural pastures and slash-and-burn techniques in order to prepare land for cultivation are major causes. Additional deforestation is caused by collecting wood for fencing materials and cooking. It is clear that mainly people are responsible for environmental degradation. A given ecosystem will be balanced when natural resources can cope with the pressure opposed on them by people and livestock. Overstocking and high population pressure will result in a too slow regeneration or complete depletion of natural resources. The main factor for land clearance and deforestation in Africa is crop production for human consumption. Contrary to what often is stated, livestock is negligible as factor in deforestation (de Haan et al, 1996).

1.2.c. Agriculture-livestock production system.

The traditional farming system in the Gambia is either agropastoralism or small-scale mixed farming associated with rain fed cultivation of cash and food crops. Crop production accounts for 60 % to the total agricultural production. Food crops are maize, millet, sorghum and rice. The main cash crops are groundnuts and to a lesser extent cotton seed and sesame. Due to the short rainy season there is one major cropping season and cropping rotation is practised.

The income derived from livestock and their products in such mixed farming systems can be responsible for 10-50 % of the household income (Wilson, 1991). For The Gambia, livestock contributes approximately 24 % of the agricultural GDP in 1997 with an annual growth rate of 3.3% (FAO, 1997). This growth follows an increased livestock integration into agriculture. Draught animals are linking both sub-sectors since animal traction is used for land preparation and transportation. Faeces of cattle and small ruminants are used for manuring cereal fields and fruit trees. Agro-pastoralists will invest surplus of their income from sales of cash crops, to buy animals as protection against crop failure. This is a main reason for the shift of pure agriculturalist to a mixed agro-pastoral rural society. Livestock production is traditional and extensive with the number of animals being more important than their individual productivity. Supplementary feeding of livestock is limited and reserved for draught oxen, horses and donkeys. This extensive low-input system results in an overall low productivity.

The land tenure system is not well organised and areas demarcated as rangeland are increasingly used for crop cultivation which creates conflicts between stock owners and crop farmers (FAO, 1997). This conflict between feed-for-livestock and food-cultivation for human consumption will intensify equally with growing population and urbanisation. As for the Gambian situation, swamp lands, used as grazing areas during the dry season have now been put under cultivation for human food consumption.

Farmers associations are generally weak, badly organised or non-existing. An increase of institutional and technical support combined with other support services such as credit, training, extension and marketing techniques for the rural farmer would lead to an increased agricultural output.

1.2.d. Role of small ruminants in mixed farming systems.

Sheep and goats are an important component of the mixed farming system. Small ruminants are often considered as the small change, while cattle constitute the capital amount. Cattle take traditionally the most important position in the livestock sector. This is linked to social status and cultural, historical and religious reasons. In terms of exchange for cash or kind, goats are considered the cheapest form of trade followed by sheep. This hierarchy of trade is followed by donkeys, cattle and horses (Itty et al., 1997). The number of small ruminants in The Gambia has been increasing steadily till present with goats outnumbering sheep (Table 1).

Table 1 : Numbers of livestock in consecutive censuses in The Gambia.

	sheep	goats	cattle*	reference
1958	50.000 est	75.000	143.000	Aalfs 1959
1978	146.000	158.000	296.000	Janneh 1979
1986/87	150.000	193.000	213.000	Sumberg
1987/88	175.000	208.000	282.000	Sumberg
1993	155.000	213.000	278.000	ITC/DLS census
1994	157.000 est	216.000 est	308.000 est	FAO 1996
1996	159.000 est	224.000 est	323.000 est	FAO 1996

* Cattle including draft oxen and cows.
'est' : estimates

Factors in favour of rearing small ruminants at village level are their precocity and fast breeding cycle. Small ruminants are also popular in extensive systems since they are thought to be less demanding than cattle on feeding and watering requirements. Sheep and in particular goats use a broader range of available grasses and shrubs than cattle do. Increase in livestock numbers has in no way kept the same rate as human population growth because the number of people has more than doubled over the past two decades.

Sheep and goat flocks are usually small, ranging between 2 and 40 heads. They are owned by the poorer section of the rural population. A survey carried out in four localities of the Gambia between 1986 and 1988 showed that 80-95% of the compounds owned small ruminants (Rawlings et al., 1992). The main objectives for raising them are income, savings and ceremonial (Itty et al., 1997). Manure, domestic meat and milk consumption were other reasons in decreasing order of importance (Bennison et al., 1997). Small ruminants are sold for cash for various family expenses such as the purchase of grains for the coming planting season. They are often exchanged for cattle to improve the farmers social status and when flock size becomes too big. Small ruminants also contribute considerably to equity issues because many of them are owned and managed by women and children. A sample survey carried out in 55 villages over the five divisions of The Gambia indicated that 68% of the small ruminants were owned by women (WID, 1993). When mutton is rather used for religious or family festivities, goat meat is more frequently commercialised and very popular as 'afra' (grilled meat). Milking of goats is practised in 18 % of the compounds, the milk being consumed within the compound (WID, 1993). The potential of goat milk production is promising with an average of 23 litres over 100 days lactation period at village level (Jaitner, personal communication, 1999). The milk provides additional protein for children or elderly people. However, low social value of goats is the main reason which makes farmers reluctant to this.

Management in the small ruminant production system is traditionally extensive. During the dry season, animals are left to roam around and feed on crop residues of harvested fields or on natural pasture. During the cropping season, sheep and goats are tethered or herded to avoid crop damage. In general they are not supplemented apart from some kitchen waste and salt. Medical treatment is often scarce and housing facilities are basic.

Semi-intensified production systems exist on a small scale, either in de peri-urban sites or in the rural areas. It is mainly practised for ram fattening for the Tobaski feast. Rams are confined and put on a zero-grazing fattening regime a few months before the feast.

The main breeds of sheep and goats in The Gambia and the subregion are the Djallonke sheep and West African Dwarf (WAD) goats. These are considered trypanotolerant (FAO, 1980; Mawuena, 1987). The West African Dwarf goat is an achondroplastic dwarf goat with a height ranging between 30-50 cm and weight between 18-25 kg. The Djallonke sheep is also a dwarf sheep with a height of 40-60 cm and an average weight between 20-35 kg depending on the sex. Both breeds originate from the Fouta Djallon and are found in many countries of the subhumid zone of West Africa (Wilson, 1991). These breeds are very fertile and productive with varying age at first parturition according to management system (range 12 -18 months) and goats being more precocious than sheep (Wilson, 1991). Both the West African Dwarf goats and the Djallonke sheep are able to survive and produce under stressful conditions based on heat tolerance, capacity to utilize low quality forage and their resistance to certain diseases e.g. trypanosomosis (Mawuena, 1986; Murray, 1982).

In the most recent livestock survey (ITC-DLS, 1993) very small percentages of 'non-Djallonke were found. Gambian farmers are increasingly interested in importing larger breeds from Senegal. These are fattened for a short period in preparation of the Muslim feast of Tobaski. These breeds of sheep and goats - Sahelian Long legged, belong to the Savanna group, which includes many Saharan types as Touabire and Peul-Peul sheep and Sahel goats. They have a height of between 70-85 cm for the goats and 65-90 for the sheep (Wilson, 1991). These breeds are considered as trypanosusceptible. Because they are not adapted to the local conditions and endemic diseases such as trypanosomosis, these Sahelians are usually fattened for a short period and slaughtered.

1.2.e. Tsetse challenge in The Gambia.

Two tsetse species occur in the Gambia, both of which are near the northern limit of their distribution. *Glossina palpalis gambiensis* is a typically riverine species and *G. morsitans submorsitans* occurs in savannah woodland. The pre-dominant tsetse species, *G. m. submorsitans* (Rawlings et al., 1993; Wacher et al., 1993) has been identified as the main vector of trypanosomosis in The Gambia. Five major foci of *G.m. submorsitans* were identified in the Gambia and very few livestock can be considered as being exposed to zero challenge (Rawlings et al., 1993). However, 90% of the tsetse flies feed on wild animals, mainly warthogs and only ten percent feed on domestic animals (Snow 1979). *G. morsitans* feeds mainly on suidae and bovidae. Although in some areas all hosts are available, some tsetse flies prefer to feed on one species only, suggesting that the feeding habit of the flies might be genetically determined (Weitz, 1963). Influencing the number of warthogs in a region may also influence the feeding behaviour of tsetse flies feeding almost exclusively on this species. Demographic, climatic and environmental factors affect tsetse populations, but it is expected that these foci of *G. m. submorsitans* infestation in the Gambia will persist for at least the next 5-10 years. Nevertheless, it has been suggested that the presence of increasing numbers of horses and donkeys in most parts of the country is an indication of reduced trypanosomosis risk compared to former times (Rawlings et al., 1993). Their owners seem to accept high mortality rates and make use of trypanocidal drugs, especially for horses (Snow et al., 1996).

1.3. The need for research on small ruminant production and development

The overall productivity from livestock in The Gambia is far below the potential. This reflects in a low per caput consumption of meat and milk. The human population in The Gambia is increasing at a rate of around 2.7 % per annum (PRB, 1996) expecting a total population of 1.6 million by 2010. An estimated additional 6000 ton/year of meat is needed to maintain the current rate of domestic meat consumption per caput. The increase in livestock numbers has not followed the same rapid trend as for the human population. This is partly due to the low productivity of the low-input traditional system and the limited availability of natural pastures. Enhancing livestock productivity through selection has been thought to be too slow for today's economic requirements. In addition, the importation of high yielding exotic breeds for pure or cross-breeding purposes is only successful if the environment is suitable or if it is economically justified to alter it (French, 1970).

For Gambian farmers, the survival of the animal is more important than milk or meat production and therefore owners try to maximize numbers of animals. Farmers' perceptions of primary importance for productivity parameters of their cattle were savings or insurance. This clearly differs from parameters set by government or potential donors which would like to increase productivity (Bennison et al., 1997). Small ruminants are valuable to all farmers in their day to day live, yet they receive little attention. The main objectives in keeping small ruminants are income savings and ceremonial. Manure, domestic meat and milk consumption are of secondary importance (Bennison et al. 1997). In the prospect of increasing overall productivity from the livestock sector, small ruminants are well positioned. Therefore, policy makers in the region have focused on the potential of short cycle animals in meeting the growing demand for animal protein (ITC, regional workshop, 1997). In the traditional Gambian system, sheep production, specially ram-fattening has been identified as a very profitable enterprise (Itty et al., 1997; Loum, 1998).

Increase in domestic production will be of benefit to all classes of population, both rural and urban. Potential high benefits from sheep and goats in both the traditional farming systems and in more intensified production systems are linked to a minimum of organisation and appropriate investment. Both systems have their own constraints which need to be evaluated for their importance in attaining or hindering increased productivity. Constraints which hamper improved productivity include socio-economic, traditional and institutional factors such as land-use and marketing problems besides bio-technical factors such as disease pressure, reproductive wastage, nutrition and management.

Researchable key factors to improve productivity are disease constraints, nutrition, genetic potential and disease tolerance, and management. Establishment of marketing systems, processing facilities and land tenure systems are key factors in hands of the government. Intensification and increase of individual animal production will concurrently demand for intensification of fodder production and novel conservation methods to limit further environmental pressure. Understanding all influencing factors and their possible interactions would lead to appropriate intervention measures with farmer community participation. This will in turn assure sustainability. Impact of the interventions and evaluation of their cost/benefits for the target population is subject of continuous socio-economic follow-up.

1.4. Diseases in sheep and goats influencing productivity.

Apart from trypanosomosis there are other important diseases which are not controlled by routine vaccination. These include some viral and bacterial diseases which play a role in the respiratory syndrome, tick borne diseases and gastro-intestinal parasites. Veterinary care is not widely practised in the rural areas where traditional farmers can not always pay for treatment of small ruminants. In addition, animal health care services in rural areas are facing problems with respect to drug distribution and lack of guidance in proper drug use. The more intensified production systems, situated mainly in the peri-urban areas, are faced with diseases due to intensification. These need different approaches, but the higher cost for disease control will be easier accepted.

1.4.a. Trypanosomosis.

Trypanosomosis is a disease caused by protozoans of the genus *Trypanosoma*. *Trypanosoma vivax*, *T. congolense* and *T. brucei* are the 3 causal species of Nagana or African trypanosomosis. *T. evansi* can also be found in small ruminants, although infections are mainly seen in camels causing the disease Surra. Nagana is transmitted cyclically by flies of the genus *Glossina* (tsetse fly) in sub-Saharan Africa, but mechanical transmission by *Tabanidae* or other biting flies is also possible. The pathogenicity depends on the trypanosome species, the strain and the susceptibility of the host. Anaemia is a predominant symptom and a reliable indicator for the severity of the infection. In the acute phase of infection, a severe anaemia appears with progressing parasitaemia. In the chronic phase, the animal remains anaemic but often with low or non detectable parasitaemia levels. This phase is followed by a slow recovery to normal haematological parameters, pending on the presence of parasites and provided there is an active haematopoietic response.

Apart from anaemia, other clinical symptoms during infection include fever, weakness, lameness, anorexia, weight loss, swollen lymph nodes and oedema of different regions of the body. Mortality can occur at all stages of the disease. Without exception the parasitaemia in the different stages of infection and with different trypanosomes was very high at the time of death (Losos et al 1972). The pathological changes caused by the different trypanosomes depend further on whether the parasite is located intra- or extra vascularly. *T. congolense* and to a lesser extent *T. vivax* are considered as strictly bloodstream forms causing no direct tissue lesions. However, splenomegaly, subcutaneous or intramuscular haemorrhages *(T. vivax)* and serous fluid in pericardial, pleural and peritoneal cavities *(T. congolense)* have been observed (Losos 1972). *T. brucei* is found extra vascularly and invades different tissues and organs. In addition to the above mentioned pathology, a *T. brucei* infection will cause severe lesions as splenomegaly, hepatomegaly, interstitial keratitis, epididymitis and encephalitis which eventually can lead to death. In *T. brucei* infections the parasitaemia is often mild and trypanosomes are more likely to be detected in the lymph nodes and the cerebrospinal fluid.

Haematological changes have been described in detail (Anosa & Isoun, 1976, Anosa 1988, Griffin 1978). They include erythrocyte, leucocyte, trombocyte and plasma changes together with changes in the blood chemical parameters such as red blood cell enzymes. In trypanosome infections, factors causing anaemia are numerous but haemolysis is by far the most important one. Haemolysis is related directly or indirectly to the trypanosome antigen subsequently coated with antibodies and to haemolysins produced by live trypanosomes or released through trypanolysis. Other causes of haemolysis are the direct adhesion of the parasite to the red blood cell and to a lesser extent through fever. All the different causes lead to extravascular erythrophagocytosis and haemosiderosis in spleen and liver (MacKenzie and Cruickshank, 1973, Anosa 1988). During the acute phase of infection, the rate of red cell destruction is higher than the erythropoiesis, but in the chronic phase both mechanisms are more balanced. In the recovery phase the erythropoiesis becomes more important.

The immunosuppressive effect of trypanosomosis exceeds the actual course of the disease. The host becomes more susceptible to secondary infections, whether from viral, bacterial or parasitological origin (Urquhart et al., 1973; Mackenzie, 1975; Sacks and Askonas, 1980; Nantulya et al., 1982; Rurangirwa et al., 1983).

Trypanosomosis is associated with serious reproductive disorders in both female and male small ruminants (Ikede et al., 1988; Sekoni, 1994). In females, infection causes pathological changes in the ovaries with as a consequence temporary or permanent anoestrus and abortions, stillbirths or neonatal deaths in pregnant animals (Ikede and Losos, 1972; Llewelyn et al., 1987; Edeghere et al., 1992). The devastating effect of *T. brucei* infection is due to its invasive character through which it can cause severe lesions in genital organs (Ikede and Losos, 1975), and abortion in all stages of pregnancy (Edeghere et al., 1992). Atrophy of reproductive organs in the non-invasive trypanosome infections is more likely to be caused by indirect mechanisms as thrombosis in the gonadal blood vessels, severe anaemia, thermal effects of pyrexia and possible stress-induced perturbation of the hypothalamus-pituitary-gonadal axis (HPG) resulting in a reproductive hormonal imbalance (Mutayoba et al., 1995 a, 1995b). However, *T. vivax* and *T. congolense*, although both considered as intra vascular species, have been associated with intrauterine infection (Elhassan et al., 1989). They were observed in amniotic fluid and ovarian cysts (Isoun and Anosa, 1974) and in the blood of a newborn lamb (Ikede and Losos, 1972). Moreover, they were able to cross the placental barrier and to cause abortion in East African goats (Ogaa et al., 1991). In males, severe inflammation of testis and epididymis and testicular degeneration are described (Losos and Ikede, 1972; Isoun and Anosa, 1974; Anosa and Isoun, 1980). The inflammatory changes in the genital organs, due to the invasive character of the parasites in *T. brucei* infections, are not found in infections with *T. vivax* and *T. congolense*. In the latter infections testicular degeneration is mainly caused by thrombosis in the spermatic blood vessels and prolonged period of fever. These testicular and epididymal lesions lead to drastic and progressive deterioration in semen quality revealed by cessation of sperm production, a decrease in sperm motility, a higher percentage of dead and abnormal spermatozoa in rams (Agu et al., 1986; Akpavie et al., 1987; Sekoni, 1992).

Under natural tsetse challenge in The Gambia sheep and goats get infected with different species of trypanosomes. Infections with *T. vivax* and/or *T. congolense* are more common than with *T. brucei* (Greenwood et al. 1988). However, sheep and goats are not the preferred hosts for tsetse flies as only 0.8 % of blood meals in tsetse flies were identified as from sheep or goats (Snow et al., 1996) .

1.4.b. Nematodosis.

Nematodosis in ruminants is causing high losses through mortality or production losses not only in Africa but worldwide (Beck et al., 1985; Over et al., 1992). Farmers in rural areas do not always have access to anthelmintics or do not buy them for small ruminants. Good knowledge and understanding of the epidemiology of helminthosis is the basis for any intervention in order to reduce or control clinical outbreaks. Among the gastrointestinal nematodes, *Haemonchus contortus* (Allonby and Urquhart, 1975; Schillhorn Van Veen, 1978) and *Trichostrongylus colubriformis* (Eysker and Ogunsusi, 1980) are generally considered as the most important ones in sheep and goats. Epidemiology of helminthosis is influenced by climatological, environmental, animal and breed related factors. The epidemiology of helminth infections in sheep and goats in West-Africa is well documented (Eysker and Ogunsusi, 1980; Fritsche et al., 1993; Ndao et al., 1995; Ankers et al., 1994, 1997, Goossens et al., 1998). During a one year sampling period Fritsche et al., (1993) found that over 95% of sheep and goats of all age classes carried nematodes. *T. colubriformis* and *Oesophagostomum columbianum* were most frequently seen while *H. contortus* was found in 67% of the animals with the adult population peaking during the rainy season (July till October) (Fritsche et al., 1993). In a later abattoir survey in The Gambia, *Trichostrongylidae* were found in 85 % of the goats and 73 % of the sheep during the rainy season. For both species, younger animals had significantly higher egg output then older animals, indicating acquisition of immunity in older animals (Goossens et al., 1998). Pregnant does tended to have a higher egg excretion than empty ones. This phenomenon is well known from temperate regions as 'peri-parturient' rise and related to a stress induced immunodepression, but has also been observed in the tropics (Schillhorn van Veen & Ogunsusi; 1978; Zajac et al., 1988; Agyei et al., 1991). Ankers et al. (1994) observed that re-infestation with helminths was negligible during the dry season. Nematodes survive the long dry season either as hypometabolic adults or as hypobiotic larvae (Ankers et al., 1994; Kaufmann & Pfister, 1990; Zinsstag et al., 1994). Ndao et al. (1995) found similar infestation levels for small ruminants in Senegal although sheep carried more helminths than goats. *H.contortus* was found as most important nematode in sheep and *T. colubriformis* in goats. Ankers et al. (1997) in Guinea found nematodes in 99 % of the autopsied small ruminants of which 96 % with more than one species. The most important nematodes found in sheep were *T. colubriformis, Cooperia* spp and *H. contortus*. In goats the most important nematodes were *T. colubriformis, H. contortus* and *Oesophagostomum* sp. Age in sheep and age and sex in goats had no influence on the nematode populations (Ankers et al., 1997). In general, goats carry less gastrointestinal nematodes than sheep (Ndao et al., 1995; Fritsche et al, 1993). Differences in worm burden between species might be related to the browsing habits of goats which may result in a reduced uptake of infective larvae (Aumont, 1999).

Trematoda belonging to the family *Paramphistomatidae* (rumen flukes) are prevalent in small ruminants (Fritsche et al., 1993; Ankers et al., 1994) but only the immature worms are of some pathological importance. Liver flukes have never been reported in small ruminants but were found in very low prevalence in slaughtered cattle (*Fasciola gigantica)* in some villages in Eastern Gambia, of which the occurrence is probably very restricted because of the required presence of the snail intermediate hosts (Swiss Helminthosis project, 1993). In Senegal, *F. gigantica* was found in sheep and goats with a prevalence of 3% and 1%, respectively (Ndao et al., 1995). *Cestoda* such as *Monieza spp.* are frequently encountered in the gastrointestinal tract of small ruminants. They generally do not cause clinical disease and are therefore of lesser economic importance (Ankers et al., 1997; Ndao et al., 1995; Fritsche et al., 1993). These gastrointestinal parasites occur throughout the year without seasonal fluctuation.

Resistance to anthelminthics in small ruminants is increasing throughout the world both in numbers of species and range of drugs involved (Sangster, 1999). Very few reports from West Africa have been published up to now (Geerts and Dorny, 1996). Ba and Geerts (1998) reported on benzimidazole resistance in some small ruminant flocks in The Gambia. The authors observed more resistance in sheep and goats under improved conditions than in animals kept at village level. They attributed this to more frequent drug use in research centres. In general, a higher prevalence of anthelmintic resistance was found in goats than in sheep (Geerts

& Dorny, 1996). This might be due to the fact that goats are more susceptible to nematode infections (Le Jambre & Royal, 1976; Huntley et al., 1995; McKenna; 1984) and therefore need more frequently anthelmintic treatment. In addition, using the same dosage of benzimidazoles and levamisole for both species whilst differences in pharmacokinetics may result in underdosage of goats creating quicker resistance (Hall et al., 1981; Hennessy, 1994). A more rapid elimination of closantel was also demonstrated in goats compared to sheep, resulting in a shortened remanent effect in goats (Hennessy et al., 1993; Dorny et al., 1994). Reasons for increasing anthelmintic resistance include incorrect dosage, extended single use of the same drugs as well as uncontrolled or wrongly applied strategic and therapeutic control strategies (Smith et al., 1999). To slow down the growing problem of anthelmintic resistance, combinations of existing and alternative control regimes are tested. Grazing management techniques, such as rotational grazing and alternation of host species on pastures, can support worm control programmes and therefore reduce the reliance on anthelmintic treatments (Barger, 1999; Waller, 1997; Rew, 1999). A novel alternative is biological worm control with nematophagous fungi, which reduce free-living stages of parasitic nematode larvae on pasture (Larsen et al.,1998, 1999; Faedo et al., 1998). Another approach is the selective treatment of haemonchosis in sheep, with use of colour cards to inspect the paleness of the mucosae (Van Wyk et al.,1997). There is also a continuous search for new antiparasitic agents (Witty, 1999) or development of vaccines (Smith, 1999). However, sofar only *Dictyocaulus viviparus* and *D. filaria* vaccines are commercially available. Given the high costs involved for these control methods with the continuous threat of drug resistance, considerable effort is given to study natural resistance against helminthosis in small ruminants.

1.4.c. Other diseases influencing productivity.

Another prevalent disease, which is a threat to small ruminants in this region and under these climatic conditions, is the viral disease Peste des Petits Ruminants (PPR). Morbidity and mortality may be high in particular when secondary bacterial infections occur. In a recent outbreak of PPR in Senegal mortality among goats was 63 %. However, a secondary infection with *Escherichia coli* might have boosted the virus effect and the mortality rate (Akakpo et al., 1996). The disease is observed as a complex of respiratory and enteric symptoms. Another important disease is Pasteurellosis. It has a respiratory tropism, but can become septicemic. Both diseases can be controlled by vaccination. Outbreaks of PPR continue to occur in The Gambia, because vaccination campaigns are not effective enough neither in frequency nor in covered areas. During a country-wide questionnaire 70 % of the compounds reported vaccination against PPR for their small ruminants (WID-ITC, Sheep and goats survey, 1993). An abattoir survey during 1993 and 1994 showed antibodies against PPR in 39 % of the goats and 49.5 % of the sheep (Goossens et al., 1998). This implies a level of immunity which is not sufficient to prevent outbreaks (Rossiter and James, 1989).

Peaks of tick infestations occur during the wet and humid season, whilst tick-borne diseases such as cowdriosis are endemic with clinical cases occurring throughout the year. Under Gambian conditions, ticks in small ruminants do not appear as an important constraint. An on-station study revealed that only 6 % of the ticks were found on sheep and 2% on goats compared to 92 % of the ticks counted on cattle. During the latter study, the predominant species in small ruminants was *Amblyomma variegatum* (72%) (Goossens and Osaer, unpublished results). In cattle, *A. variegatum* was the most abundant tick besides *Hyalomma truncatum*, *Rhipicephalus senegalensis* and *Boophilus geigi* (Claxton and Leperre, 1991). Small ruminants carry fewer ticks as compared to cattle during the rainy season and this is in line with observations made by Gueye et al. (1989) in Senegal. Depending on the species, ticks will attach to different regions of the body but they are most frequently encountered on the ears, in the perianal region and between the hoofs of sheep and goats. The latter facilitates secondary bacterial infections such as interdigital dermatitis and footrot. The most important tick-borne disease is cowdriosis caused by the rickettsia *Cowdria ruminantium* and transmitted by *A. variegatum*. Since it is an endemic disease in the country and the subregion southwards, the indigenous breeds seem to be resistant to a certain extent. Clinical cases mainly occur when stress factors or other diseases interact or when susceptible and/or exotic breeds are imported. In that respect, the introduction of Sahelian sheep for crossbreeding with the local Djallonke breed resulted in high numbers of deaths. Out of ten Sahelian sheep that died 7 were confirmed positive for *cowdriosis* (Goossens and Osaer, unpublished results). This has also been reported earlier by Uilenberg (1983; 1989).

1.4.d. Interactions of diseases.

Under field conditions animals will rarely be infected with a single pathogen. When deaths occur it is mostly due to a combination of additional pathogens which aggravates the initial infection. Some pathogens cause immunodepression and render the host more susceptible or less resistant in combatting simultaneous infections.

The interaction of *T. congolense* and *H. contortus* has been studied experimentally in young N'Dama cattle (Kaufmann et al., 1992). It was observed that dual infections were more severe than single infections. The most harmful combination was a *H. contortus* infection one week after a *T. congolense* infection. In dual infected groups they found a reduced prepatent period for the *H. contortus* infection. Evidence of this interaction was also found in village N'Dama cattle (Dwinger et al., 1994). Griffin et al (1981) studied dual infections with *T. congolense* and *H. contortus* in two different breeds of goats. They used East African goats which are trypanotolerant but susceptible to haemonchosis and Saanen and Galla crossbred which are resistant against haemonchosis but susceptible to trypanosomosis. They concluded that *T. congolense* infection suppressed the innate resistance of the goats against haemonchosis. Specht (1982) conducted a field survey in Mozambique on the effects of trypanosomes and gastrointestinal nematodes on the productivity of sheep and goats. Animals receiving treatment against both the pathogens gained more weight than groups where only one of the two pathogens was controlled. Gretillat (1981) made the observation that a treatment against nematodes resulted in a proliferation of coccidiosis. The latter author explained that this was due to a disequilibrium of the established interaction between nematodes, cestodes and coccidia in the gastro-intestinal tract of the host. Yvoré et al. (1980) made similar observations in sheep which received anthelmintic treatment following experimental infections with *Eimeria ovinoidalis* and *T. colubriformis*. Rahman (1994) studied the interaction between *Eimeria spp.* and *H. contortus* infection in goats in Malaysia. Goats challenged with an artificial infection of *Eimeria* and consecutively with *H. contortus* shed more eggs of *H. contortus* and gained weight more slowly compared to the animals initially treated against *Eimeria*. Cox (1987) reviewed the interactions between trypanosomosis, babesiosis and theileriosis. Several indications were given for the immunosuppressive action of trypanosomosis with secondary pathogens becoming more activated and causing more severe disease as a consequence. Moreover, it may hinder proper immunisation by means of vaccination to other infections.

1.5. Non-disease factors influencing productivity.

1.5.a. Animal nutrition.

The influence of nutrition on productivity is self evident. In addition, deficiencies in mineral or trace elements may have impact on the reproductive performance. Research on improvement of animal nutrition in developing areas implies study of available feed resources or introduction of new feedstuffs. Furthermore, existing grazing management in the rural areas differs between seasons and influences greatly the nutritional condition of the animals. Studying grazing behaviour and applied management is essential to formulate possible improvements.

The nutrition factor is often underestimated as a limiting factor for productivity. Research often concentrates on the disease constraint without taking into consideration the nutritional status of the animal and its possible interaction with the pathogen. The course of a disease may be aggravated by poor nutrition because it undermines the resistance of the host against infection. Vice versa, sick animals will loose appetence due to certain physiological changes and voluntary food intake will reduce. In severely *H. contortus* infected animals, the pH of the abomasum increases, factor which may have an effect on appetence (Holmes 1987). The same author suggested that similar as in gastrointestinal infections, it is most likely that in haemoprotozoan infections voluntary food intake is reduced. This was corroborated by Romney et al. (1997) who observed reduced intake of *Andropogon* hay in trypanosome infected cattle. Similarly, other studies have reported trypanosome induced depression of feed intake in the order of 15% in small ruminants and have linked this to decreased live weight gain (Verstegen et al., 1991; Akinbamijo et al., 1994; Wassink et al.,1997; Reynolds & Ekwuruke,1988).

Diseases imply a metabolic cost which negates gain in productivity. Trypanosome infections lead to increased maintenance requirements in the order of 15 % in small ruminants (Verstegen et al., 1991; Akinbamijo et al., 1994) resulting in decreased nitrogen and energy retention. In the course of disease, higher nutrient supply is required for recovery and immune responses. Therefore, Agyemang et al. (1990) concluded that the plane of nutrition plays an important role in the rate of recovery from anaemia following trypanosome infection of N'Dama cattle. In trypanotolerant cattle, malnutrition has been reported as a factor which can reduce trypanotolerance (Murray et al, 1982; Agyemang et al., 1990; Little et al., 1990; Bennison, PhD Thesis, 1997). A better erythropoietic response was observed in trypanosome infected Scottish Blackface sheep kept on a high protein diet compared to those on a low protein diet (Katunguka-Rwakishaya et al., 1993).

1.5.b. Livestock management.

The importance of the husbandry system and management including housing, herding and reproductive management should be emphasized since it might influence indirectly or directly the health and productivity of small ruminants.

Reproductive performance of a flock is measured by its yearly produced offspring. Management of reproduction, apart from the foregoing factors, may influence overall performance. If mating is uncontrolled, some animals might become pregnant at an earlier age with abortion or premature birth as a result. Together with the risk of inbreeding the reproductive outcome is negatively influenced. If mating is controlled, minimum targets for breeding are often not met and age at first parturition is higher then in uncontrolled systems (Wilson et al., 1989). Poor husbandry and housing influence disease risk and productivity.

Kaufmann et al. (1993) observed a lower infection with helminths in cattle in herds frequently moved to new night holding places than in herds which were not moved. Raised and slatted platforms for sheep and goats reduce the risk for helminth infections and footrot during the rains. Tethering small ruminants for the daytime during the cropping season without moving them frequently results in inadequate nutritional supply and increases the risk of rapid helminth reinfection. In the rural areas where both cattle and small ruminants depend on natural grazing, overstocking may occur in some areas, particularly in the dry season. This results in depletion of natural pastures and in environmental degradation. This again will affect the productivity.

1.6. Innate disease resistance in Djallonke sheep and West African Dwarf goats.

Native breeds of sheep and goats are adapted to the climatic conditions of the ecological system they live in and to the diseases occurring in the area through long term and natural selection. Djallonke sheep and the West African Dwarf goats are considered trypanotolerant (Touré et al. 1981; Mawuena 1986, 1987; Bengaly et al., 1993). Murray et al (1982) defined trypanotolerance as the trait that certain breeds possess which gives them the possibility to survive and reproduce in tsetse-infested areas without the aid of chemotherapy where other breeds cannot. The same author described the mechanism of trypanotolerance in cattle as an ability to limit reductions in haematocrit and to control the level and duration of parasitaemia following infection. There are suggestions, which will be confirmed further in this thesis, that the mechanism of this trait in small ruminants differs from that in cattle. Trypanotolerant cattle breeds are the N'Dama breed and West African Shorthorn. These breeds are dwarf breeds and productivity is generally believed to be low. Genetic selection for meat (and milk) production in the pure breeds is one solution to enhance individual productivity. Cross-breeding with larger, trypanosusceptible breeds is another option but disease resistance may be reduced, especially the trypanotolerance. Therefore, trypanotolerant breeds can play an important role in the improvement of livestock productivity in Sub-Sahara Africa. Furthermore, the use of trypanotolerant breeds in tsetse infested areas is an economically viable and environmentally safe alternative to overcome the constraint of trypanosomosis on animal production. Apart from trypanotolerance, there are strong indications that these breeds possess resistance to other diseases such as helminthosis (Assoku, 1981; Claxton & Leperre, 1991; Baker, 1995), tick infections and tick-borne diseases (Mattioli et al., 1993). Distinction, however, should be made between resistance to infection and resilience to the effects of infection (Clunies-Ross, 1932).

In Togo, Mawuena (1986, 1987) described trypanotolerance in indigenous Djallonke sheep as a quasi absence of clinical signs following infection with trypanosomes whilst they had similar weight gains as the non-infected sheep. Moreover, fertility in the infected animals seemed not affected since they lambed normally. In Burkina Faso, Bengaly et al. (1993) compared Djallonke and Sahelian sheep under artificial trypanosome challenge and concluded that the former breed had a superior resistance in terms of clinical outcome and better control of the parasitaemia. In Senegal, Touré et al., (1981) observed that 60 percent of the Sahelian and none of the Djallonke sheep died following *T. congolense* infection, but there was no difference between breeds following *T. vivax* infection. This difference could be rather explained by a different pathogenicity of the strains used. In Nigeria, West African Dwarf (WAD) goats were more resistant following *T. congolense* infection compared to the Red Sokoto (RS) goats (Adah et al., 1993). The infected RS goats had initially a more severe anaemia leading to a 100% mortality while the infected WAD had positive weight gain and only one death occurred.

Trypanotolerance has also been observed to a certain extent in native East African sheep and goats. Griffin and Allonby (1979a) compared two breeds of sheep and three breeds of goats under high tsetse challenge. Sheep included the indigenous Blackhead Persian (B.P) and Karakul and for goats they compared Saanen x Galla crosses, Galla and East African breed. Weight gain in B.P. was higher than in the Karakul, whereas the latter breed had more severe anaemia and higher mortality rates. Similar observations were also seen in the goats with the indigenous East African and Galla goats performing best. They concluded that under high tsetse challenge, the indigenous breed was less susceptible to trypanosome infection than the imported breed. It was also observed that the indigenous breeds were less infected than the exotic breeds under same level of tsetse challenge indicating that there is a variation in severity according to the breed at risk. The same authors evaluated three different breeds of sheep and four breeds of gaots following an experimental *T. congolense* infection (Merino, B.P. and Red Masai for the sheep and Saanen, Saanen x Galla crosses, Galla and East African). Based on PCV % and weight changes they concluded that the indigenous breeds of sheep (Red Masai and B.P.) and goats (East African and some what lesser Galla) resisted infection better than the exotic Merino sheep or Saanen goats (Griffin and Allonby 1979b). Kanyari et al. (1986) and Mutayoba et al. (1989) evaluated indigenous goat breeds of East Africa and also recognised differences in susceptibility to trypanosomosis.

Since long it has been suggested that the trypanotolerant trait has a genetic base. In comparative studies with N'Dama, Zebu and their F1 crosses, the crossbred expressed an intermediate resistance to trypanosome infection compared to the two pure breeds (Chandler 1952). Touré et al. (1978) in Senegal and Murray et al. (1981) in The Gambia came to the same conclusion. In trypanotolerant sheep the basis for trypanotolerance also seems genetically driven, although not much evidence is available. In Kenya, Griffin and Allonby (1979b) observed that only five out of 10 Red Masai sheep became parasitaemic after experimental *T. congolense* infection and concluded that the nature of this resistance was innate since the animals originated from a tsetse free area. Crossbred Saanen X Galla goats reacted in an intermediate way to an experimental trypanosome infection compared to the pure breeds (Griffin and Allonby, 1979b).

Apart from innate resistance, there is also acquired resistance to trypanosomosis. Previous exposure to trypanosomes, the age of the host as well as the species, strain and number of infecting parasites are additional factors determining the host's resilience to infection. Finally, intercurrent disease factors and the nutritional status of the host interfere to a certain extent with the expression of trypanotolerance but this needs to be further studied in small ruminants. This thesis will further provide evidence on the trypanotolerant quality of Djallonke sheep and West African Dwarf goats.

Several authors reported that native breeds reveal a superior resistance to helminth infections as compared to improved or imported breeds and explained this as a combination of innate and acquired resistance (Preston and Allonby 1979; Bradley et al., 1973). In a village study in Bansang, Zebu cattle were found to carry significantly higher number of endo- and ecto parasites compared to N'Dama cattle (Claxton and Leperre 1991). They concluded that the lower parasite burden in the N'Dama must be partially due to an innate resistance to strongyle infection. Research on resistance against helminths in small ruminants mainly concentrates on sheep, given their higher economic importance as compared to goats in the production of protein and wool in industrialised countries. Research in sub-Saharan Africa revealed the existence of resistance to helminths in some breeds of sheep and goats. These breeds include the Red Masai, Blackhead Persian and Merino (Preston and Allonby, 1979; Al-Khshali and Altaif, 1979;

Wanyangu et al., 1997). Further evidence is given for resistance in St.Croix against *H. contortus* (Gamble and Zajac, 1992). Similarly also for St. Croix, Barbados Blackbelly, Sumatra, Florida Native and Louisiana Native, the latter two also grouped as Gulf Coast Native (Zajac et al., 1990; Romjali et al., 1997, Miller et al., 1998; Yaswinski et al., 1979,1980). St. Croix and Barbados Blackbelly sheep originate from West Africa and are probably related to the Djallonke breed. There are suggestions that Djallonke sheep are resistant to helminth infections, however, not much experimental evidence is available (Assoku, 1981; Baker, 1997). A higher resistance to *Haemonchus contortus* was found in East African (EA) goats than in Toggenburger x EA-crosses or in the Galla goat, but there were no significant differences in EPG nor PCV% (Shavulimo et al., 1988). There is no evidence of resistance to helminth infection in the West African Dwarf goat.

The nature of resistance to infection with nematodes is not completely clear. More than in trypanosomosis, resistance against trichostrongylosis is based on history of exposure, age of the host and on genetics (Gamble and Zajac, 1992). Evidence of genetic variation comes from three sources: variation among breeds, variation within breeds and the identification of genes contributing to the variation (Stear & Murray, 1994). To study the genetic variation in resistance to helminths the following traits are used: number of adult worms of slaughtered ewes, faecal egg output (EPG) and anaemia (Gray, 1987; Albers et al., 1987). Until present, it is generally supported that the EPG in sheep and goats is the single most important and currently available criterium in selecting for resistance (Baker, 1999). Furthermore, assessment of resistance to gastrointestinal nematode parasites should be based on experimental infections with a known larval dosage, since there is evidence of a strong genetic correlation with natural pasture challenge and the possibility of cross-protection amongst nematode species (Baker, 1999). Zajac et al. (1990) compared three breeds of sheep challenged with an experimental *H. contortus* infection. Whereas no breed differences were found in total worm burden, the Dorset/Rambouillet sheep showed higher EPG and lower PCV % levels compared to the Florida Native and St.Croix breeds. Based on these traits they concluded that the Dorset/Rambouillet are less resistant to haemonchosis than the Florida Native and St. Croix. More recent research has investigated the underlying mechanism of resistance to *Ostertagia circumcincta* in Scottisch Blackface lambs, which is an important nematode parasite in temperate zones (Stear et al., 1997a, 1997b; 1999). They found that these lambs showed considerable genetic variation in EPG following infection, which was a consequence of genetic variation in worm length and worm fecundity, and not due to variation in worm burdens. Worm length is a marker of worm fecundity and both traits are strongly phenotypically correlated. The same authors also revealed that genetic resistance operates through control of an acquired response, of which the immune response is the most prominent one (Stear et al.,1999). However, these findings can not be extrapolated to other nematode genera or other resistant breeds until further proven (Baker, 1999). The main immunological mechanism involved in resistance to *O. circumcincta* is the production of more parasite-specific immunoglobulin A (Ig A) than in susceptible lambs. Local IgA responses regulate worm length and hence fecundity and develops before the more general effective hypersensitivity responses which regulate worm burdens (Stear et al., 1997a; 1999). This IgA is therefore a good indicator to screen for resistant or susceptible lambs to *O. circumcincta* infection (Stear et al., 1997b). Resistance to most nematode infections is believed to be immunologically mediated (Stear and Wakelin, 1998).

2. OBJECTIVES OF THIS THESIS

2.1. Overall objective

The studies described hereafter generally aimed at improving survival and develop sustainable methods of enhancing productivity of trypanotolerant small ruminants in trypanosome-endemic areas of West Africa. All studies were carried out at the International Trypanotolerance Centre and/or in rural areas of The Gambia.

2.2. Justification

The use of trypanotolerant breeds of small ruminants in tsetse infested areas is an alternative approach to control trypanosomosis. The Djallonke sheep and West African Dwarf (WAD) goats, the main breeds in the Gambia, are considered as trypanotolerant. Expression of trypanotolerance can be negatively influenced by intercurrent diseases, nutrition, husbandry factors and their interactions with trypanosomosis. In addition, these indigenous breeds are well adapted to local conditions and possess a certain disease resistance to other endemic diseases. Results obtained in The Gambia are to a certain extent applicable to the sub-region.

2.3. Expected outputs

The results of this thesis will hopefully contribute to a better understanding of the role of stress factors in the mechanism of trypanotolerance in the WAD goats and Djallonke sheep and of their disease resistance in general. Generated results should support income generation and increased food supply from small ruminants for rural small-holding farmers, particularly women. It is hoped that they will contribute to the high demand of protein by the rapidly growing urban population. Altered feeding strategies and husbandry practises for small ruminants will contribute to environmental protection.

2.4. Detailed objectives

- To study the impact of trypanosomosis as sole factor or in combination with other stress factors on survival and productivity of indigenous trypanotolerant West African small ruminants both under artificial and natural challenge.
- To study the influence of nutritional status on the pathogenic effects of trypanosomosis on health and reproductive performance in Djallonke sheep.
- To study the interaction of a helminth infection and trypanosome infection on the trypanotolerance in Djallonke sheep.
- To evaluate the influence of genetics on disease resistance and production potential of Djallonke sheep and their F1- crossbred with the trypanosusceptible Sahelian breed.
- To survey the traditional rural sheep and goat production systems in The Gambia and to identify existing constraints, importance of diseases, nutrition and husbandry practices related to small ruminant production
- To evaluate interventions concerning helminth control for on-farm improvement of small ruminant health and productivity in the rural areas.
- To identify constraints on feeding strategies and fodder production for small ruminants in rural areas and peri-urban production sites.

- To test certain feeding strategies to allow seasonal strategic feeding of selected livestock and at the same time contribute to sustainable environmental conservation.

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CHAPTER 2

TRYPANOTOLERANCE
IN DJALLONKE SHEEP AND WEST AFRICAN DWARF GOATS

2.1

A comparison of the susceptibility of Djallonke sheep and West African Dwarf goats to experimental infection with two different strains of *Trypanosoma congolense.*
Osaer, S., Goossens, B., Clifford, D.J., Kora, S. and Kasama, M. 1994.
Veterinary Parasitology, 51: 191-204

2.2

Haematological changes in trypanotolerant sheep and goats following experimental *Trypanosoma congolense* infection.
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2.3

Health and productivity of traditionally managed Djallonke sheep and West African Dwarf goats under high and moderate trypanosomosis risk.
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2.1
A comparison of the susceptibility of Djallonke sheep and West African Dwarf goats to experimental infection with two different strains of *Trypanosoma congolense*.

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A comparison of the susceptibility of Djallonké sheep and West African Dwarf goats to experimental infection with two different strains of *Trypanosoma congolense*

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Abstract

Two cloned strains of *Trypanosoma congolense*, of West and East African origin, were used to infect by intradermal inoculation two groups of young adult female Djallonké sheep and West African Dwarf goats. For a 3 month period post-infection, packed red cell volume (PCV), parasitaemia, body weight and clinical parameters were followed to evaluate their trypanotolerant nature and to control the pathogenicity of the two strains of *T. congolense*. Although the West African strain of *T. congolense* was more pathogenic than the East African strain, it seemed that the Djallonké sheep and the West African Dwarf goats, despite high levels of parasitaemia and a concomitant drop in PCV, showed a high degree of trypanotolerance, as reflected by zero mortality and an increase in body weight during 12 weeks of observation.

Key words: *Trypanosoma congolense*; Sheep-Protozoa; Goat; Pathogenicity-Protozoa

Introduction

In sub-Saharan Africa, livestock production is severely limited in tsetse-infested areas. However, certain breeds of cattle such as N'Dama and West African Shorthorn are able to remain productive under trypanosomosis challenge and are considered as trypanotolerant (Murray et al., 1982). Moreover, certain breeds of sheep and goats also seemed to survive in tsetse-infested areas where trypanosomosis is endemic (Griffin and Allonby, 1979a; Kanyari et al., 1986; Mawuena, 1986, 1987; Mutayoba et al., 1989). There are suggestions that the mechanism of tolerance in small ruminants may differ from that of cattle. Thus, while trypanotolerant cattle have an ability to control the trypanosome growth, develop a less severe anaemia and withstand the effects of the disease (Murray et al., 1982), it is not evident that trypanotolerant sheep and goats resist in the same way. In West Africa, it is generally accepted that the indigenous Djallonké sheep and West African Dwarf goats

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are trypanotolerant, although the underlying mechanism of this tolerance has not been studied in detail. Cameroon Dwarf goats showed a considerable resistance to *Trypanosoma vivax* and *Trypanosoma congolense* (Büngener and Mehlitz, 1976) compared with the fatal infection of *Trypanosoma brucei*. Djallonké sheep were found to be more resistant than Fulani sheep to syringe-passaged *T. congolense* (Touré et al., 1981). The Djallonké sheep and West African Dwarf goats in Togo showed a high level of trypanotolerance under natural challenge (Mawuena, 1986). To evaluate the trypanotolerance of the local Djallonké sheep and West African Dwarf goats in The Gambia, West Africa, two on-station experiments were carried out in which groups were artificially inoculated with an East African or a West African strain of *T. congolense*. Both these strains are known to be highly pathogenic and lethal in Zebu cattle (Dwinger et al., 1992). Parasitaemia, packed cell volume (PCV) and weight changes were monitored.

Materials and methods

The two experiments took place on-station at the coastal site of the International Trypanotolerance Centre (ITC), The Gambia, West Africa. The climate is sudano-sahelian with a mean annual rainfall of approximately 1000 mm. Since susceptible Zebu cattle around the station had remained free of trypanosomosis for more than 5 years, it was concluded that tsetse challenge and hence trypanosomosis risk was effectively zero (W.F. Snow and D.J. Clifford, personal communication, 1992). For the first experiment, 13 Djallonké sheep and 13 West African Dwarf goats were purchased from different village flocks near the coast where trypanosomosis prevalence is low (Agyemang et al., 1990). All animals were negative for trypanosomes following examination of the buffy coat by dark ground (DG) microscopy (Murray et al., 1977), while no trypanosomal antibodies were detected using the immunofluorescence antibody test (IFAT) (Katende et al., 1987). All animals were females aged between 6 and 18 months. Before the start of the experiment, they were vaccinated against Peste des Petits Ruminants (PPR-Tissue Culture Rinderpest vaccine, Hann Laboratory, Dakar), dewormed (Panacur®, 10%, Hoechst, Germany, 7.5 mg kg^{-1} body weight) and sprayed with acaricide (Gammatox®, Coopers, UK, 8 g per 10 l water). Deworming and spraying was repeated every 3 weeks during the wet and humid season. The animals were herded around the station during the day for grazing and housed at night. One male sheep and one male goat were added to the respective groups for reproduction purposes.

Prior to infection, all animals were weighed and bled for assessment of PCV percentages. Ten female animals of each species were artificially infected by syringe inoculation with 10^4 bloodstream forms of *T. congolense* ITC 50 (hereafter referred to as *Ea-Tc*); IL 1180, a cloned parasite originating from

East Africa (Dwinger et al., 1992) was used as reference stabilate to produce the bulk stabilate, ITC 50, by passaging it once through mice. The animals were inoculated intradermally at four sites on the left flank. Three sheep and three goats were kept as uninfected controls. All animals were bled daily for the first 16 days following infection and, thereafter, three times per week. Jugular vein blood samples (3–5 ml) were collected in ethylenediamine tetraacetic acid (EDTA) coated vacutainer tubes. PCV percentages were measured as an estimation of anaemia using the capillary microhaematocrit centrifugation method. Trypanosomes were detected and an estimation of parasitaemia level was made by examining the buffy coat by the dark ground (DG) method (Murray et al., 1977). The number of trypanosomes was scored as described by Paris et al. (1982). Clinical signs and mortality rates were recorded and body weights were measured weekly using a Salter spring balance. Additional parameters, including oestrus dates, service dates and abortions were also recorded. The experiment started in the late rainy season (August 1989).

The second experiment took place under the same conditions with 24 female Djallonké sheep and 20 female West African Dwarf goats. They were all aged between 6 and 18 months. The animals were related to the sheep and goats of the first experiment (second or third generation). For both the sheep and the goats, the animals were divided equally into two groups, infected and control, based on age and weight (goats 10/10, sheep 12/12). This experiment started in the dry season (February 1992). The animals were constantly confined during the first 4 weeks post-infection and fed mainly with groundnut hay and small amounts of supplement (sesame cake and spent brewers' grain). Owing to maladaption of the goats to confinement, the animals were released during daytime for grazing in the fifth week. The protocol and methods used before and during this experiment were similar to those used for the first experiment, except for the strain of *T. congolense*: a West African strain, ITC 84, (hereafter referred to as *Wa-Tc*), a cloned stabilate derived from a goat at Kunting on the north bank of the River Gambia (Upper River Division, The Gambia). The stabilate was expanded in mice and the animals were inoculated intradermally at four sites on the left flank with 1 ml of the infected mouse blood containing at least 10^4 bloodstream forms of *Wa-Tc*.

In addition, before the start of the second experiment, haemoglobin typing was performed using electrophoresis on cellulose acetate paper in Tris–glycine buffer (pH 9.0) running at 200 V for 120 min (Huisman, 1963). All sheep and goats belonged to the AA group. The nematode egg excretion (eggs per gram of faeces, EPG) was examined monthly using the McMaster technique (Thienpont et al., 1979).

A comparison was made between the groups of infected sheep and infected goats in both experiments, for the first 3 months post-infection. The control animals are considered as a reference for the changes in PCV values and body weights.

Data were analysed with the Minitab Statistical computer package (Ryan et al., 1985). One- and two-way analyses of variance were used on the group data to compare control and infected groups (sheep and goats), and the two strains of *T. congolense*. The 95% confidence level was regarded as significant. The results are presented as arithmetic mean ± 1 SD.

Results

Clinical observations

None of the animals infected either with *Ea-Tc* or the *Wa-Tc* strain died. No obvious clinical changes were observed that could be attributed to trypanosome infection, although sporadic respiratory problems and diarrhoea occurred in both infected and control animals. Overall, the sheep and goats remained in good health. As the observation period was only 12 weeks, few results could be drawn concerning reproduction. However, it was observed that in both infected and control groups the animals came into oestrus at regular intervals and were mounted by the male. One goat in the *Wa-Tc* infected group aborted 14 days post-infection. The foetus was mature but hairless.

Parasitaemia

The daily (Days 0–16) and weekly mean parasitaemic scores for sheep are presented respectively in Figs 1 (a) and 1 (b); those for goats are presented in Figs. 2(a) and 2(b).

Sheep

In the first experiment following *Ea-Tc* infection, all sheep became parasitaemic between 5 and 9 days post-infection (PI). The first parasitaemia peak was reached in the second week with a mean score of 4.1 at Day 13. The parasitaemia dropped from Week 8 to remain steady with an overall average score of 1.1 until Week 12. Following *Wa-Tc* inoculation, a prepatent period between 4 and 7 days was observed. The sheep attained their first peak at Day 12 with a similar mean score of 4.1 as in the *Ea-Tc* experiment. After the first peak, the parasitaemic waves continued and there was slight tendency to decrease (average score of 2.8 between Weeks 5 and 12). Considering the entire observation period, the mean parasitaemia level of the *Wa-Tc* infection was significantly higher than during the *Ea-Tc* experiment.

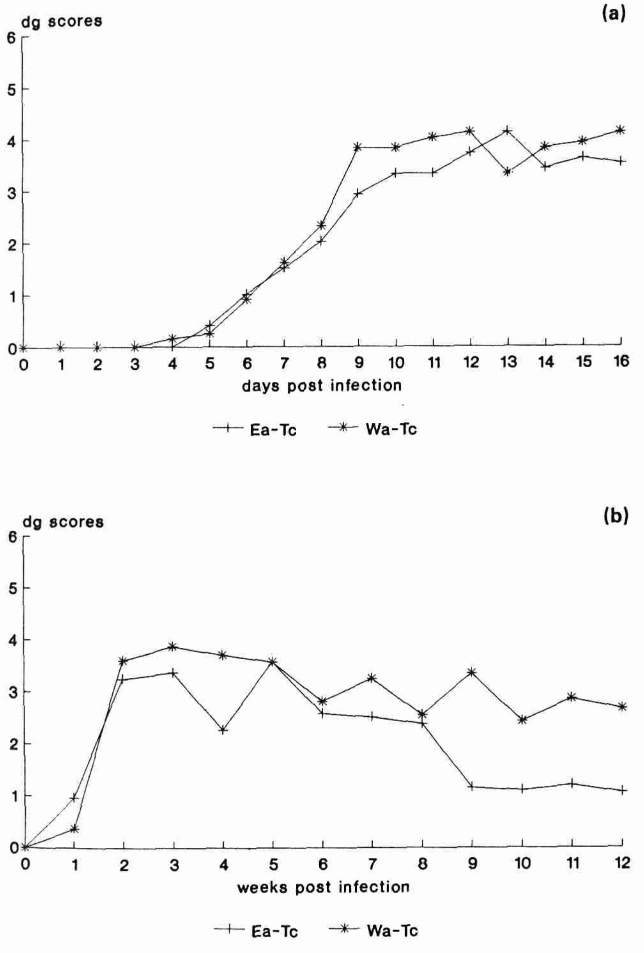

Fig. 1. Mean DG scores of sheep: (a) daily; (b) weekly.

Goats

The prepatent periods were the same as those recorded for the sheep. Following *Ea-Tc* infection, the first parasitaemia peak was reached on Day 10 with a mean score of 3.4, whereas for the *Wa-Tc* infection, the peak was attained on Day 8 with a mean score of 3.8. Following *Ea-Tc* infection, parasitaemia in the goats dropped markedly from the second week, and an overall mean score of 1.7 was maintained up to Week 12. Parasitaemia in the group infected with the *Wa-Tc* strain remained at a high level after the first peak until Week 4 with subsequent parasitaemic waves, still at high levels with an average score of 2.7. They followed the same pattern as for the sheep with *Wa-Tc* infection.

In general, during both infections, there was no significant difference in

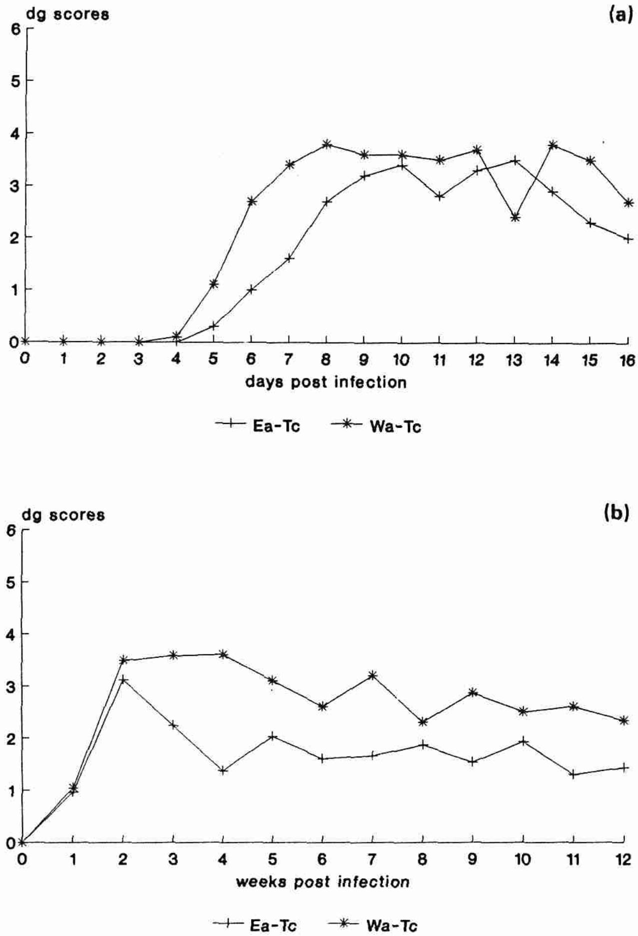

Fig. 2. Mean DG scores of goats: (a) daily; (b) weekly.

mean parasitaemic score between goats and sheep. However, as for the sheep, the parasitaemia was significantly higher following *Wa-Tc* infection.

Packed cell volume

The mean weekly PCV percentages are presented in Figs. 3 and 4. The onset of anaemia was directly related to the first wave of parasitaemia and from Week 2 a decrease of PCV values occurred in both groups of infected sheep and goats.

Sheep

In both experiments, the mean PCV values initially showed a progressive decrease, although the *Wa-Tc* strain caused a more severe drop from $32 \pm 2.4\%$

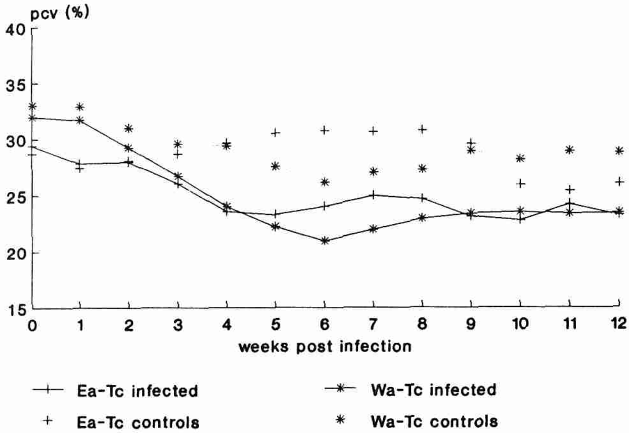

Fig. 3. Mean weekly PCV percentages in sheep.

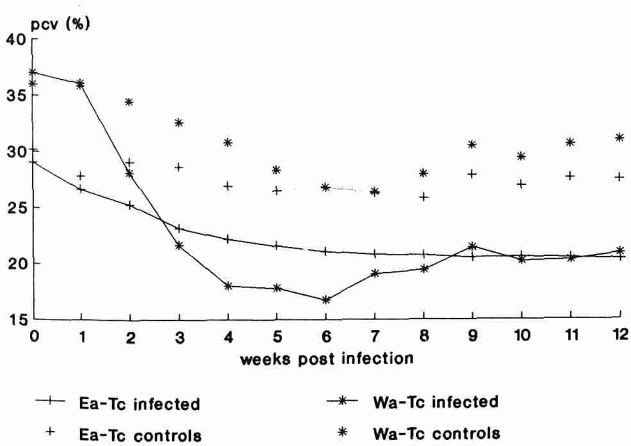

Fig. 4. Mean weekly PCV percentages in goats.

(mean ± 1 SD) at the start to 20.9 ± 2.3% at Week 6, a fall of 11.1 percentage points, compared with the *Ea-Tc* strain which induced a drop from 29.4 ± 2.1 to 23.3 ± 3.1% at Week 5, a fall of 6.1 percentage points. After this fall both groups tended to increase slowly, although at Week 12 they still remained at levels lower than pre-infection values. However, comparing the two strains for the entire observation period, the mean PCV values of the infected groups were not significantly different. Considering the control groups in the *Wa-Tc* experiment, the uninfected sheep showed a drop in mean PCV value equivalent to −7, possibly as a result of frequent bleeding. The PCV values of the three *Ea-Tc* control sheep did not follow this pattern, although they were bled as frequently as the other animals, and showed a decline only after the eighth week. Overall, the average PCV levels of the infected groups were significantly lower than those of the controls.

Goats

The drop in PCV in the infected goats was more marked than in the sheep, especially after the *Wa-Tc* infection. They showed a decline from an initial 37 ± 3.5% to 16.8 ± 2.3% at Week 6, a fall of 20.2 percentage points. The other group (*Ea-Tc*) decreased slowly until Week 12 from 29.1 ± 3.0% to 20.4 ± 3.2%, a drop of 8.7 percentage points. The PCV levels of the infected goats of both infections were significantly lower than those of the infected sheep, while there was no difference in mean PCV comparing the species in general. This might suggest that goats develop a more severe anaemia and suffer more from the infection than sheep. As a result of the frequent bleeding procedure, the control goats of both experiments also showed a drop in mean PCV values. The *Wa-Tc* control goats, which were a more reliable control group ($n=10$) had a more important decline of up to 10 percentage points compared with −4.5 percentage points of the three *Ea-Tc* controls. However, the mean PCV values of the infected and control groups of both experiments were significantly different over the 12 weeks following infection.

Body weights

The average weekly body weights are presented in Tables 1 and 2. The weights are given as arithmetic means ± 1 SD. The weight changes of the dif-

Table 1
Mean (± 1 SD) weekly body weights (kg) of sheep and goats following *Ea-Tc* infection

Week	Goats		Sheep	
	Control	Infected	Control	Infected
0	13.3 ± 3.3	13.5 ± 3.5	15.7 ± 1.9	15.5 ± 2.5
1	15.0 ± 2.6	14.1 ± 3.7	16.3 ± 1.6	15.6 ± 2.4
2	15.7 ± 3.2	15.0 ± 3.9	18.6 ± 2.1	18.3 ± 3.4
3	16.3 ± 2.1	16.0 ± 4.2	19.6 ± 1.5	18.6 ± 3.1
4	16.2 ± 2.8	14.9 ± 3.3	19.5 ± 1.5	18.3 ± 2.8
5	16.3 ± 3.8	14.5 ± 3.6	18.3 ± 1.9	17.8 ± 3.7
6	16.3 ± 2.8	14.8 ± 3.3	19.6 ± 2.3	18.5 ± 2.5
7	15.5 ± 3.5	14.5 ± 2.6	20.6 ± 1.6	19.1 ± 3.3
8	16.0 ± 3.9	13.8 ± 2.9	20.6 ± 1.9	19.3 ± 2.5
9	16.2 ± 4.0	14.5 ± 2.4	20.5 ± 2.3	19.0 ± 2.9
10	16.9 ± 4.0	14.9 ± 2.2	21.1 ± 2.0	19.2 ± 3.0
11	17.7 ± 4.2	15.4 ± 2.3	22.2 ± 1.9	18.6 ± 4.6
12	18.1 ± 3.1	14.7 ± 2.4	23.0 ± 2.6	19.3 ± 3.4

Table 2
Mean (±1 SD) weekly body weights (kg) of sheep and goats following *Wa-Tc* infection

Week	Goats		Sheep	
	Control	Infected	Conotrol	Infected
0	13.6±3.1	13.3±4.0	17.0±4.8	18.0±5.9
1	13.9±3.0	13.2±4.0	17.2±5.0	17.8±6.0
2	13.7±3.2	13.1±4.0	17.2±4.8	17.7±5.8
3	13.9±3.2	12.9±3.5	17.7±5.1	18.2±5.9
4	13.8±3.2	12.6±3.2	17.0±4.9	18.0±5.7
5	14.0±3.2	13.0±3.3	17.3±5.0	18.1±5.5
6	13.7±3.1	12.9±3.3	16.4±4.8	17.1±5.3
7	13.6±3.0	12.8±3.3	16.5±4.6	17.1±5.4
8	14.0±3.3	13.0±3.3	17.0±4.6	17.5±5.4
9	14.6±3.5	13.2±3.4	17.8±4.7	18.2±5.3
10	15.1±3.8	13.5±3.4	18.8±4.9	18.7±5.3
11	15.2±3.7	13.3±3.4	18.9±5.1	18.8±5.3
12	15.4±3.7	13.5±3.3	19.1±5.4	18.9±5.4

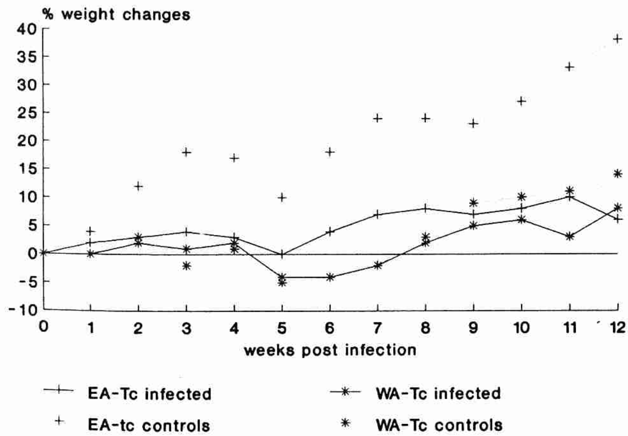

Fig. 5. Percentage weight changes in sheep.

ferent groups are shown in Figs. 5 and 6, calculated as a percentage of their mean initial body weight.

Sheep

The mean weights of both groups had increased by the end of the 12 weeks of observation. However, there was a decrease between Weeks 4 and 6 for the *Ea-Tc* infected group and between Weeks 4 and 8 even below their pre-infection weights for the *Wa-Tc* infected group. The final weight gain was not more than +6% for *Ea-Tc* infected sheep and +8% for *Wa-Tc* infected sheep. The

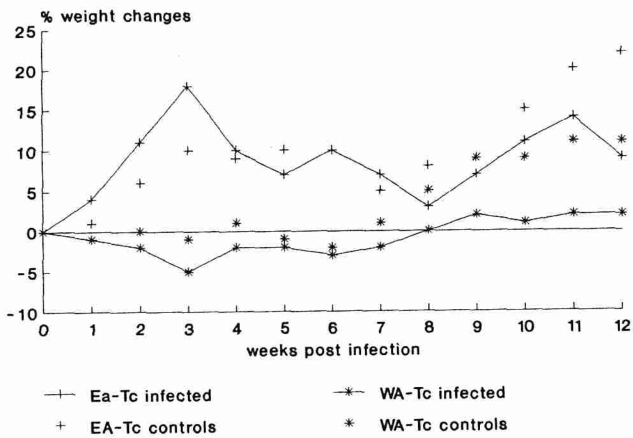

Fig. 6. Percentage weight changes in goats.

mean weight gain for the control group in the *Ea-Tc* experiment was much higher (+38%), although it should be emphasised that there were only three animals in the control group. In contrast, the control group in the *Wa-Tc* experiment, which had the same number of animals as the infected group, did not exhibit significantly greater in weight changes (+14%) than the infected group during the observation period. Overall, there were no significant differences in mean body weights between control and infected groups during the 12 week observation period.

Goats

By 12 weeks after infection, both groups had gained weight. For the *Wa-Tc* infected group, however, the gain was very small (2%): during the first 8 weeks the animals lost weight, but never more than 5% of the starting weight. The other group (*Ea-Tc*) performed better. After an initial increase of 18%, followed by a decline between Weeks 3 and 8 to the lowest value of +3%, they increased again to a final gain of +9% compared with their pre-infection mean weight. In the *Wa-Tc* experiment, the control goats followed a similar pattern as for the infected group up to Week 9. The control goats in the *Ea-Tc* experiment did not differ significantly from the infected animals until Week 11. Once again, there were no significant differences in mean body weights between control and infected goats for both experiments. A comparison between the two strains for the mean weights of infected sheep and goats did not show any significant differences.

Faecal egg output

During the *Wa-Tc* experiment, rectal faeces samples were taken on three occasions to check for helminth infection. Out of the 132 samples examined in total, 12 animals were positive for strongyles, of which ten had an EPG of 200 and two had an EPG of 400. Strongyloides were not found. Considering the low prevalence of positive samples with low EPG, the results were found to have no significance.

Discussion

The trypanotolerant nature of cattle breeds such as N'Dama appears to be related to an ability to control the level and duration of parasitaemia and to develop a less severe anaemia (Murray et al., 1982).

The Djallonké sheep and West African Dwarf goats used in this experiment, resisted both infections of *T. congolense*. Despite high parasitaemia and severe anaemia, no deaths were recorded following infection. The general condition and appearance of the animals during the observation period remained good. Comparing the different parameters measured (PCV, parasitaemia, body weight, mortality), only a significant difference in mean parasitaemia level indicated the difference in virulence between the two strains used. ITC 84, the West African strain was more pathogenic than ITC 50, an East African strain, as shown by shorter prepatent periods and significantly higher levels of parasitaemia. These two strains of *T. congolense* have also been used in an on-station experiment where N'Dama and Zebu cattle were artificially infected twice, first with ITC 50 (*Ea-Tc*) followed by ITC 84 (*Wa-Tc*). The *Wa-Tc* strain was found to be more virulent than the *Ea-Tc* strain, since 60% of the trypanosusceptible Zebu needed treatment after infection with the former strain compared with 30% following infection with the latter (Dwinger et al., 1992).

The clinical and pathogenic effects of a *T. congolense* infection in trypanosusceptible breeds of small ruminants are well described (Losos and Ikede, 1972; MacKenzie and Cruickshank, 1973; Kaaya et al., 1977; Mutayoba et al., 1989; Dwinger et al., 1990; Mwangi et al., 1990). Infections are usually accompanied by severe weight loss, lethargy, debilitation and mortality.

In both experiments, the Djallonké sheep and West African Dwarf goats showed a significant degree of resistance to two strains of *T. congolense* of known pathogenicity, in terms of weight changes and clinical outcome (i.e. final weight gains were not different from control animals, no clinical signs such as lethargy, no mortality). However, under the conditions of these experiments, there is evidence that the nature of trypanotolerance in Djallonké sheep and West African Dwarf goats may be different from that of trypanotolerant cattle breeds. Thus, following infection by both strains of *T. congolense*, sheep and in particular goats developed significant degrees of anaemia and had high persistent levels of parasitaemia. In the *Wa-Tc* experiment, while the mean PCV levels in both infected sheep and goats were significantly lower than in the controls, the positive weight changes and zero mortality were taken as the real index of resistance. Touré et al. (1981), who compared Djallonké sheep with Fulani sheep following *T. congolense* and *T. vivax* inoculation, reported a remarkable resistance of the Djallonké to *T. congolense*, but not to *T. vivax*. Mawuena (1987), who studied the Djallonké sheep under natural challenge in the south of Togo, showed that the small ruminants were even more resistant to trypanosome challenge than trypanotolerant cattle; despite high parasitaemia levels and severe anaemia among these animals, their reproduction capacity was not affected. Because this author studied animals that had been naturally exposed to trypanosomosis all their lives, both acquired and innate resistance must be considered in evaluating the trypanotolerance of the Djallonké sheep used in his experiment. In the present study, the experimental sheep and goats had no known previous contact with trypanosomosis as reflected by a lack of detectable antibodies prior to infection. Thus it was concluded that the degree of resistance that was demonstrated could be considered as the extent of innate resistance.

In East Africa, Griffin and Allonby (1979a,b) compared indigenous breeds of sheep and goats in Kenya with exotic breeds and found that local breeds were more resistant to trypanosomosis following both natural and experimental infection. In contrast, Whitelaw et al. (1985) detected no significant difference among indigenous breeds of goats and crossbreeds following experimental infection by tsetse-transmitted *T. congolense*. Kanyari et al. (1986) and Mutayoba et al. (1989) evaluated indigenous goat breeds of East Africa and also recognised a difference in their susceptibility to the disease. These contradictory results might be attributed to differences in various 'stress factors', e.g. helminth infections, poor nutrition and management which can be regarded as affecting the trypanotolerance of small ruminants (Greenwood and Mullineaux, 1989). In the present study, for the *Wa-Tc* experiment, the animals were confined during the first 4 weeks. This confinement might explain the weight losses observed in both infected and control goats before they were released in the fifth week. This management regime might also have had a negative impact on the trypanotolerant status of the animals. With respect to the different parameters measured, it was observed in this study that goats seem to suffer more from experimental trypanosome infection than sheep, especially in terms of anaemia. Despite this conclusion, in epidemiological studies the natural prevalence of trypanosomosis in goats is usually very low. It has often been observed that goats are able to live in tsetse-infested zones without showing obvious signs of disease (Murray et al., 1982). The low incidence of infection in goats exposed to natural challenge may be explained by the fact that goats are not often fed on by tsetse (Weitz, 1963). Blood-meal analyses of tsetse flies indicated that small ruminants are rarely selected as a source of feeding (Snow and Boreham, 1979; W.F. Snow, personal communication, 1992). In this context, the different grazing patterns of small ruminants should also be considered as this determines the contact with tsetse. In The Gambia, during the wet season, infection prevalence is low as a result of restricted grazing and therefore less contact with tsetse. In the dry season, when sheep and goats are grazing free and go further in the bush, a higher incidence of infection is found (Greenwood and Mullineaux, 1989). Studies on the grazing behaviour of small ruminants compared with cattle showed that small ruminants stay closer to the village for grazing and therefore have less chance of being fed on by tsetse (T. Wacher, unpublished data, 1992).

It was concluded from the present study that following experimental infection with *T. congolense*, the local Djallonké sheep and West African Dwarf goats in The Gambia, West Africa show a significant level of trypanotolerance which is largely due to innate resistance.

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2.2
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Haematological changes and antibody response in trypanotolerant sheep and goats following experimental *Trypanosoma congolense* infection

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Abstract

Ten West African Dwarf (WAD) female goats and twelve Djallonké ewes were artificially infected with a West African strain of *Trypanosoma congolense* and monitored during 36 weeks over an acute phase (weeks 0–12) and chronic phase (weeks 13–36) to evaluate their haematological and immunological response. Parasitaemia, packed cell volume, red blood cells, haemoglobin, white blood cells and trypanosomal antibodies were assessed. Mean corpuscular volume and mean corpuscular haemoglobin concentration were calculated. The infected animals showed a persistent parasitaemia together with a chronic anaemia and significantly lower packed cell volume, red blood cell count and haemoglobin. The infected sheep developed a macrocytic, hypochromic anaemia during the acute phase changing to normocytic, hypochromic during the chronic phase, whereas, the infected goats developed a normocytic, normochromic anaemia during the acute phase and normocytic, hypochromic during the chronic phase. A significant increase in WBC counts was observed only in the infected sheep during the chronic phase. Trypanosomal antibody titres were significantly higher in the infected sheep than in the infected goats. Both species are regarded as trypanotolerant but Djallonké sheep mount a better haematopoietic and immunological response to infection with *T. congolense* than WAD goats.

Keywords: Sheep-protozoa; Goat; Haematology; *Trypanosoma congolense*; Trypanotolerance

1. Introduction

In African animals, acute, chronic and recovery phases of trypanosomosis are recognised and haematological changes, weight loss, anorexia, weakness and elevated

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body temperature are observed. In *T. congolense* infection the acute phase is generally characterized by high levels of parasitaemia and a concomitant rapidly developing anaemia which will remain stable but low in the chronic phase during which parasites may be present or not. Anaemia is a predominant symptom and a reliable indicator for the severity of a trypanosome infection and measured by decrease in PCV, RBC and Hb levels as indicators of erythrocytic response (Losos and Ikede, 1972; Kaaya et al., 1977; Ogunsanmi et al., 1994). Red cell indices as mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC) may indicate the type of anaemia which developed following infection. Leucopenia is observed in trypanosomosis (Naylor, 1971; Anosa, 1988) specially during the acute phase and the onset of anaemia (Valli et al., 1979); however, leucocytosis also has been reported specially during the chronic phase or in breeds known to be more tolerant (Adah et al., 1993; Ogunsanmi et al., 1994). In addition, as in many other parasitic diseases, trypanosomosis also has a clear immunodepressive effect and renders animals more sensitive to secondary infection (Griffin et al., 1981a; Van Dam et al., 1981; Nantulya et al., 1982).

Indigenous breeds of sheep and goats in West and East Africa possess a certain degree of tolerance against trypanosome infections (Griffin and Allonby, 1979; Kanyari et al., 1986; Mawuena, 1987; Goossens et al., 1997a). In West Africa, natural infection with trypansomosis in sheep and goats is reported as low with *Trypanosoma vivax* more prevalent than *Trypanosoma congolense* (Kramer, 1966; Mawuena, 1987; Greenwood and Mullineaux, 1989; Snow et al., 1996). In East Africa, Griffin et al. (1981b) showed that the indigenous goats reacted to severe anaemia, whether caused by infection or frequent bleeding, with a marked erythropoietic response which was not the case in the exotic breeds. This marked response might be the trait to judge trypanotolerance in small ruminants. In trypanosusceptible cattle, the detection of neutralizing antibodies against *T. congolense* was delayed and the titre was lower as compared to trypanotolerant cattle (Pinder et al., 1988). It was also observed in trypanosusceptible Zebu cattle infected with *T. brucei* or *T. congolense*, that falling concentrations of antibodies are related to self-cure (Wilson and Cunningham, 1971). While, trypanotolerance in N'Dama cattle is related to the ability to control the level and duration of parasitaemia (Dargie et al., 1979) this seems not to be the case in Djallonke sheep and WAD goats (Osaer et al., 1994; Goossens et al., 1997a). Few reports are available on haematological changes in Djallonke sheep and WAD goats following experimental infection with *T. congolense* (Edwards et al., 1956; Adah et al., 1993; Bengaly et al., 1993). This experiment contributes to the knowledge about trypanotolerance in Djallonke sheep and WAD goats based on haematological changes and antibody response following artificial infection with *T. congolense*.

2. Materials and method

The experiment took place between February 1992 and October 1992 at the coastal station of the International Trypanotolerance Centre (ITC), Gambia, West Africa. The climate is sudano-sahelian with a mean annual rainfall of approximately 1000 mm.

Nineteen female WAD goats and 23 female Djallonke sheep, aged between 6 and 18 months at the beginning of the experiment, were selected from the ITC breeding flock and randomly allocated within species into control and infected groups based on age and weight groups. All experimental animals were born and reared at the coastal site, an area known to be free of tsetse flies (Rawlings et al., 1993) and could not have been infected with *T. congolense* before. Before the experiment, all the animals were negative for trypanosomes following examination of the buffy coat by dark ground (DG) microscopy (Murray et al., 1977) and negative for antibodies against *T. congolense* following indirect fluorescent antibody test (IFAT) (Katende et al., 1987). All animals were vaccinated against Peste des Petits Ruminants at 6-month intervals (PPR-Tissue Culture Rinderpest vaccine, ISRA/LNRV, Dakar). They were dewormed (Panacur®, 10%, Hoechst, Germany, 7.5 mg kg bodyweight or Ivomec®, 1.0% Merck Sharp and Dohme, USA, 1 ml 50 kg^{-1} bodyweight) and sprayed with acaricide (Gammatox®, Coopers 8 g 10 l^{-1} water or Ovipor®, 2.5% C-Vet, UK), and deworming and spraying were repeated every 4 weeks during the wet season. During the experiment, medical treatment was provided where needed and a post mortem was performed whenever death occurred. In the first month p.i. the animals were kept confined, afterwards they went out for grazing. In the duration of the experiment, they received a supplementary diet of groundnut hay, rice bran and sesame cake. During the night, they were kept confined.

In week zero, 12 out of 23 sheeps and 10 out of 19 goats were inoculated with ITC 84, a West African strain of *T. congolense*. This West African strain was compared with ITC 50, an East African strain of *T. congolense* (ITC 50) in Djallonké sheep and WAD goats and the West African strain was found more pathogenic than the East African strain (Osaer et al., 1994). The same observation was made in trypanosusceptible Zebu cattle of which 30% needed treatment following infection with ITC, 50 and 70% following infection with ITC 84 (Dwinger et al., 1992). The stabilate was expanded in mice and the animals were inoculated intradermally at five sites on the left body flank with 1 ml infected mouse blood, containing at least 10^4 trypanosomes. Infection took place on day 0 of the experiment and all times referred to in the text relate to time p.i. Following infection all animals were bled daily for the first 16 days, thereafter three times per week from week 3 until week 12, then once weekly until week 36. Jugular vein blood samples (3 ml) were collected in ethylenediamine tetra-acetic acid (EDTA)-coated vacutainer tubes. Packed cell volume (PCV) levels were measured as an estimation of anaemia using the capillary microhaematocrit centrifugation method. The buffy coat zone of the capillary was examined for trypanosomes by the DG technique (Murray et al., 1977) and parasitaemia was quantified according to Paris et al. (1982). WBC counts, RBC counts and haemoglobin (Hb) were assessed by means of an ABX MINOS ST Haematology Analyzer (ABX-International, Levallois, France). MCV and MCHC were calculated according to Riedler and Zingg (1983). The titre of trypanosomal IFAT antibodies for both infected sheep and goats was determined according to Magnus (1988) using Flourescein-conjugated Rabbit IgG fraction to sheep or goat.

Statistical analysis was carried out using SAS® statistical package version 6.11 SAS (1989)–(1996). The various traits were examined by unbalanced analyses of variance. PCV percent was subjected to a model, which included the effect of infection (treatment), animal (nested in treatment), sampling week and the interaction treatment* weeks. All traits were analysed for two different periods. The period over week 0–12 was regarded as the acute period and period from week 13–36 was defined as chronic. This separation between acute and chronic periods is based on the observation in the infected animals that, 12 weeks p.i. the PCV levels stabilised with a tendency to increase. In addition, the parasitaemia level had reached its peak and was clearly decreasing (Osaer et al., 1994; Goossens et al., 1997a). Results were regarded as statistically significant when Type I error probability was smaller than 5%. Means of the different groups are presented as least square means ± standard error (i.e. mean±SE).

3. Results

Pre-infection values of all animals and values of control animals over 36 weeks for haematology traits were obtained and least square means were calculated. Table 1 shows these values for PCV, RBC, Hb, MCV, MCHC and WBC of clinical healthy sheep and goats. During the 36-week observation period, one infected goat died (week 16) with a PCV of 22 and parasitaemia score of 4+. Post mortem showed inflamed lymph nodes, enlarged spleen, hydropericard and hydrothorax symptomatic for trypanosomosis. No infected sheep and none of the control animals died. Animals of all groups gained weight during the whole observation period of 36 weeks. Prepatent period for occurrence of parasites in the blood of infected sheep and goats was between 4 and 7 days. The first peak was reached at day 12 for the sheep and day 8 for the goats. Parasitaemia of infected sheep and goats followed the same pattern with decreasing peaks, but some animals remained parasitaemic up to the end of the observation period. There were no significant differences between species in level of parasitaemia (Fig. 1).

Haematological traits are presented in Table 2. Initial decline in PCV, RBC count and Hb level was more marked in the infected goats. Infected sheep and goats had significantly lower PCV levels (Fig. 2), lower mean RBC counts (Fig. 3) and lower mean Hb levels (Fig. 4) than their respective control groups over the whole observation period. For the infected goats, mean levels of PCV, RBC counts and Hb did not reach pre-infection values within 36 weeks, whereas, in the infected sheep, RBC counts reached pre-infection values in week 23. MCHC and MCV levels are plotted, respectively, in Figs. 5 and 6. Comparing infected goats with non-infected controls, analysis indicated no significant differences for MCV but a significantly lower MCHC ($p<0.05$) in the infected animals only during the chronic phase resulting in a normocytic, normochromic anaemia for the acute phase and a normocytic, hypochromic anaemia in the chronic phase. The

Table 1
Values of haematological traits of non-infected control goats ($n=351$) and sheep ($n=429$) determined over the whole observation period expressed as least square means±SE

	PCV	RBC	Hb	MCV	MCHC	WBC
Goats	31.6±0.1	15.2±0.7	10.2±0.5	20.2±0.4	33.5±0.6	18.9±3.7
Sheep	29.6±0.1	10.6±0.8	10.7±0.5	28.3±0.5	36.4±1.5	12.6±2.7

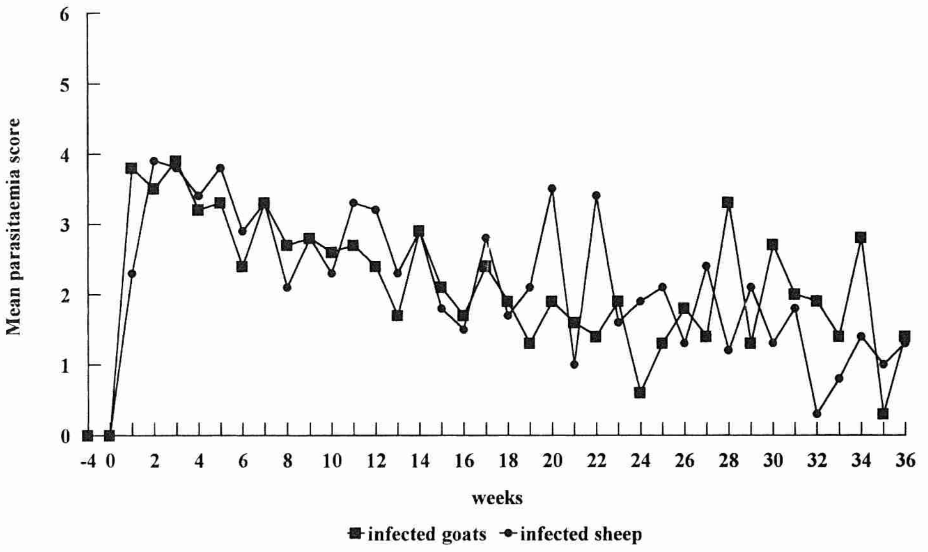

Fig. 1. Mean weekly parasitaemia scores in Djallonke sheep and WAD goats following an experimental *Trypanosoma congolense* infection.

infected sheep showed significantly higher MCV levels compared to the controls during the acute phase (macrocytic) but not during the chronic phase (normocytic). MCHC in the infected sheep was significantly lower (hypochromic) during both the acute ($p<0.05$) and chronic phase ($p<0.01$).

Table 2
Values of haematological traits expressed as least square means±SE for control and infected sheep and goats during the acute and chronic periods. Comparison between treatments within species and period with levels of significance per species

Parameter and period		Control goats	Infected goats	Level of significance	Control sheep	Infected sheep	Level of significance
PCV	Acute	30.6±0.1	22.2±0.1	$p<0.001$	28.9±0.1	24.8±0.1	$p<0.001$
	Chronic	30.1±0.1	24.3±0.2	$p<0.001$	28.9±0.1	25.5±0.1	$p<0.001$
Hb	Acute	9.2±0.05	6.8±0.05	$p<0.001$	9.7±0.05	8.1±0.05	$p<0.001$
	Chronic	10.5±0.04	7.4±0.05	$p<0.001$	11.2±0.03	9.2±0.03	$p<0.001$
RBC	Acute	14.1±0.07	9.8±0.07	$p<0.001$	9.8±0.05	8.0±0.05	$p<0.001$
	Chronic	15.8±0.08	11.6±0.1	$p<0.001$	11.0±0.05	9.2±0.05	$p<0.001$
MCV	Acute	21.6±0.1	21.8±0.1	NS	29.5±0.2	30.9±0.2	$p<0.01$
	Chronic	20.7±0.1	20.4±0.2	NS	28.5±0.1	28.8±0.2	NS
MCHC	Acute	30.6±0.1	30.2±0.1	NS	34.0±0.01	33.4±0.1	$p<0.05$
	Chronic	33.0±0.1	32.4±0.02	$p<0.05$	37.0±0.01	36.1±0.1	$p<0.01$
WBC	Acute	18.0±0.3	19.9±0.2	NS	13.1±0.2	13.4±0.2	NS
	Chronic	19.1±0.2	18.1±0.3	NS	12.1±0.2	16.9±0.2	$p<0.001$

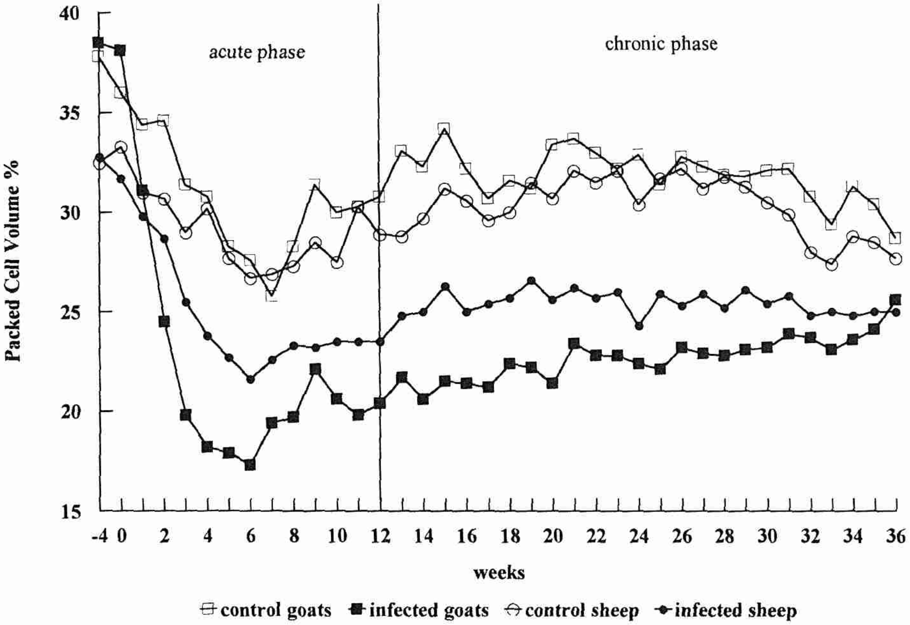

Fig. 2. Mean weekly packed cell volume % in *Trypanosoma congolense* infected and control Djallonke sheep and WAD goats.

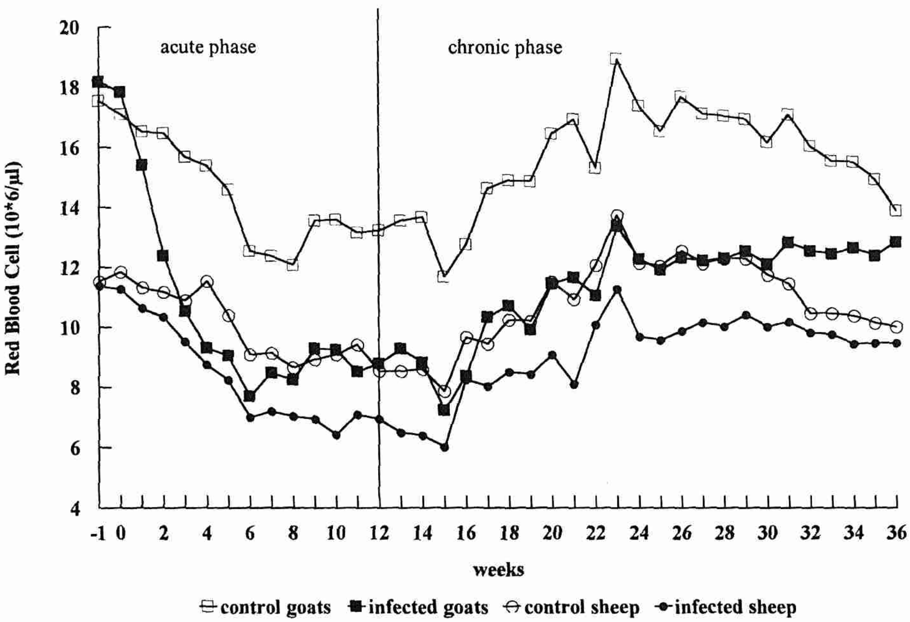

Fig. 3. Mean weekly red blood cell counts in *Trypanosoma congolense* infected and control Djallonke sheep and WAD goats.

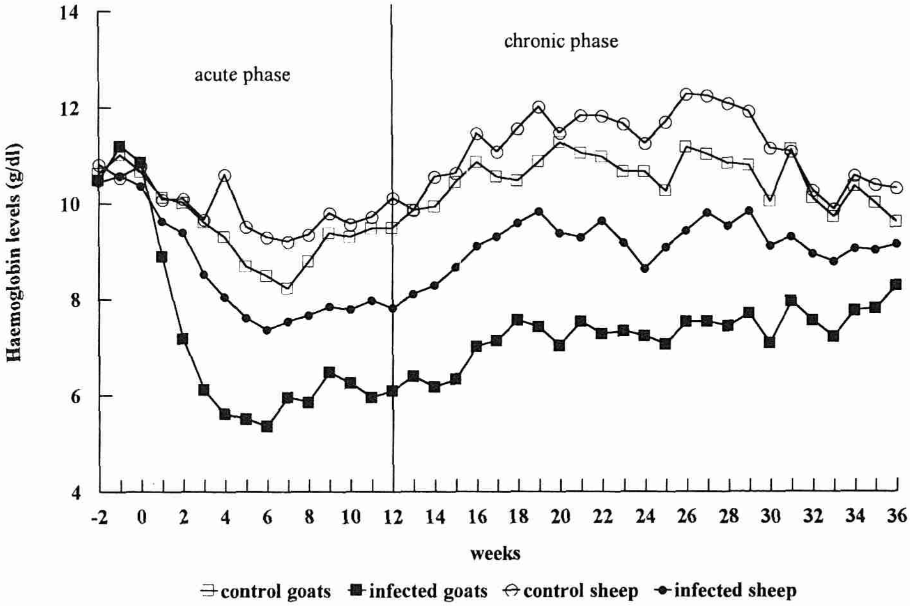

Fig. 4. Mean weekly haemoglobin level in *Trypanosoma congolense* infected and control Djallonke sheep and WAD goats.

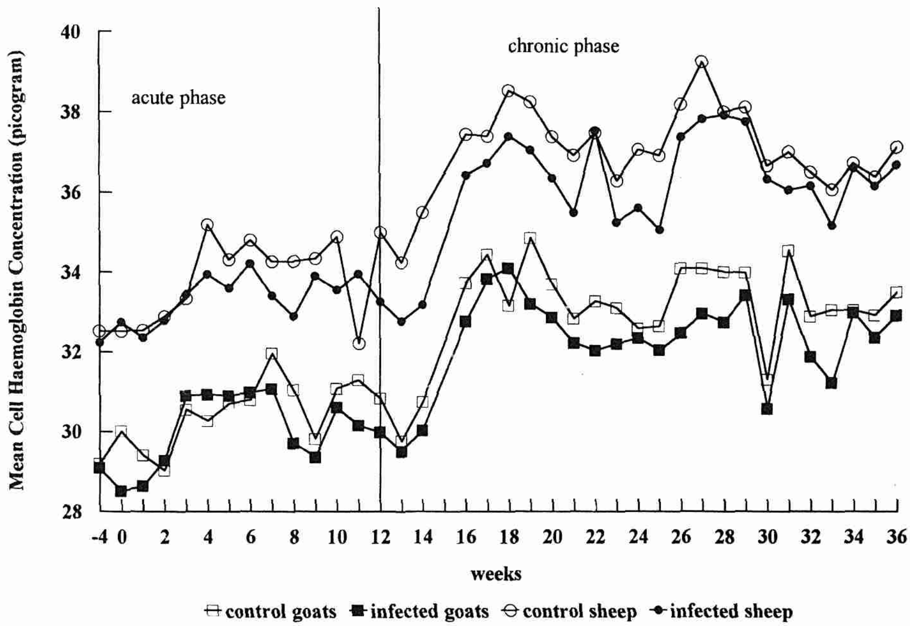

Fig. 5. Mean weekly mean cell haemoglobin concentration (MCHC) level in *Trypanosoma congolense* infected and control Djallonke sheep and WAD goats.

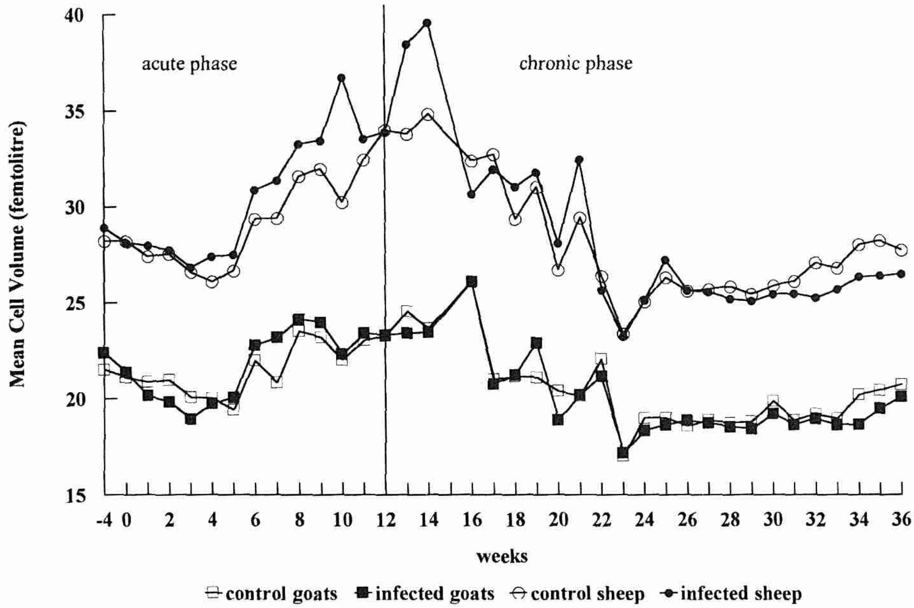

Fig. 6. Mean weekly mean cell volume (MCV) level in *Trypanosoma congolense* infected and control Djallonke sheep and WAD goats.

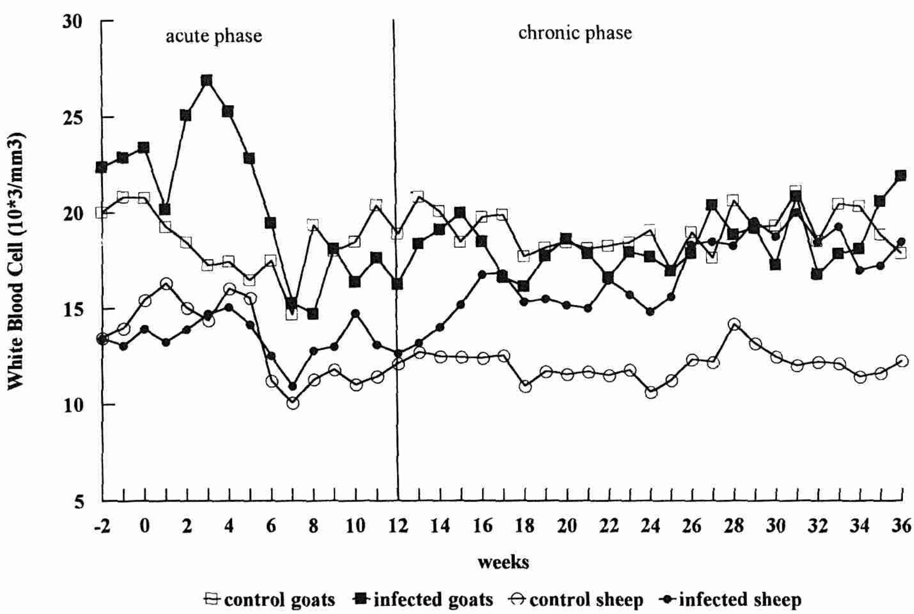

Fig. 7. Mean weekly white blood cell counts in *Trypanosoma congolense* infected and control Djallonke sheep and WAD goats.

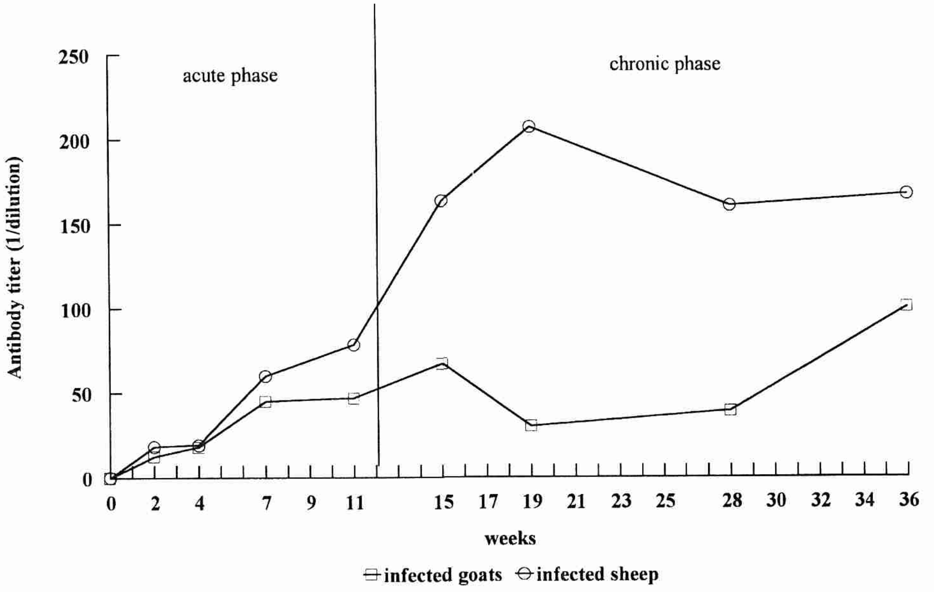

Fig. 8. Mean antibody titres against *Trypanosoma congolense* in infected Djallonke sheep and WAD goats.

The total leucocyte counts are presented in Fig. 7 for goats and sheeps. The infected goats showed a sharp increase but short in duration in the acute phase, and no differences were found between treatment groups during the two periods. In the sheep, a slower increase in WBC counts was observed with significantly higher ($p<0.001$) levels of WBC during the chronic period. The titres of trypanosomal IFAT antibodies (1/dilution) for both infected sheep and goats are presented in Fig. 8. The development of antibodies followed a similar pattern during the acute phase after which the infected sheep built up significantly higher levels of antibodies compared to the infected goats.

4. Discussion

The values for haematological traits of clinically healthy sheep and goats were similar to those found during previous studies (Oduye, 1976; Adah et al., 1993). The initial decrease in PCV, RBC and HB also observed in the control groups is related to the frequent bleeding regime in the beginning of the experiment, a feature which was also observed by Griffin et al. (1981b).

One infected goat died due to trypanosomosis, whereas, no infected sheep died and the decline in PCV, RBC and Hb, concomitant with the onset of parasitaemia, was more significant in infected goats compared to infected sheep. Macrocytic, hypochromic anaemia as seen in infected sheep during the acute phase is attributed to a regenerative anaemia, whereas, a normocytic, normochromic anaemia observed in the infected goats during the same phase is a non-regenerative anaemia which is often caused by failure of erythropoiesis (Morris and Dunn, 1992). During the chronic phase the anaemia changed to normocytic, hypochromic in both the infected sheep and goats.

Following artificial *T. congolense* infection, the Djallonke sheep responded with a marked erythropoiesis resulting in the release of young red blood cells in the circulation. This is accompanied by a leucocytosis which, although not defined, is assumed to be a neutrophilia which often accompanies a regenerative anaemia indicating increased marrow activity (Morris and Dunn, 1992) and this leucocytosis became apparent during the chronic phase of the infection. The erythropoietic response in the goats following infection was weak and transient and much different than in infected sheep. The anaemia following infection is normocytic and normochromic, indication of a non-regenerative anaemia and failure of erythropoiesis. Although there was a slight increase in MCV and decrease in MCHC during the acute phase of the infection, these differences were non-significant. The increase in WBC counts during the acute phase, although sharp, was not significantly higher when compared to the control group.

Both trypanosusceptible and trypanotolerant breeds of cattle, sheep and goats have been infected artificially with different trypanosomes and followed for their haematological response whose results could be used in relating to degrees of tolerance. Griffin (1978) in Kenya, using a *T. congolense* infection, reported a marked erythropoietic response in indigenous sheep and goat breeds known as trypanotolerant and only a slight increase in the more exotic non-trypanotolerant breeds. A normocytic, normochromic anaemia was observed in East African goats infected with *T. congolense* (Kaaya et al., 1977) or macrocytic, normochromic anaemia in different breeds of goats (Whitelaw et al., 1985; Adah et al., 1993). Djallonke sheep infected with *T. brucei* developed a normocytic, normochromic anaemia during the acute phase, changing to macrocytic during the chronic phase (Ogunsanmi et al., 1994). Trypanosusceptible Zebu cattle and Holstein Friesian calves infected with *T. congolense* developed a normocytic, normochromic anaemia (Losos et al., 1973; Valli et al., 1978) and Naylor (1971) reported dyshaemopoiesis in trypanosusceptible Zebu shorthorns as one of the reasons of anaemia. When infected with *T. congolense*, the observed peak parasitaemia in trypanotolerant N'Dama cattle attained later, was lower and of shorter duration as compared to trypanosusceptible Zebu cattle. In both breeds, the anaemia was due to intravascular red cell destruction which was more pronounced in the Zebu; however, they showed a higher degree of red cell synthesis. Ellis et al. (1987) compared N'Dama with Boran cattle during *T. congolense* infection and observed that N'Dama had lower levels of parasitaemia and maintained fairly high PCV, whereas, Boran cattle developed severe anaemia and needed drug treatment. In N'Dama the higher tolerance is related to the control of parasitaemia and not primarily to a more efficient erythropoietic response (Dargie et al., 1979). There appears to be a different mechanism in trypanotolerant Djallonke sheep and WAD goats. Although, there was no difference in the parasitaemia levels of the infected sheep and goats in this experiment and the infected animals remained parasitic over extended periods, their respective haematological responses were different. There seems to be no correlation between parasitaemia and severity of anaemia or other parameters, indicating that both are regulated by different mechanisms (Authié et al., 1993). An increased phagocytosis of leucocytes and erythrocytes in *T. congolense* infection (MacKenzie et al., 1978) suggests a generalized stimulation on the mononuclear phagocytic system. Phagocytosis of these cells is thought to lead to anaemia and leucopenia which is positively correlated with high parasitaemia (Anosa et al., 1992). In infected sheep, the erythropoietic response on the increased phagocytosis was earlier and faster compared to infected goats resulting in a less severe drop of erythrocyte values (PCV, RBC and Hb).

Due to the immunosuppressive effect of a trypanosome infection, secondary infections may become more pathogenic in combination with trypanosomosis (Griffin et al., 1981a; Van Dam et al., 1981; Goossens et al., 1997b). In most cases of animal trypanosomosis, leucopenia is observed (Anosa, 1988). Leucocytosis was observed in the more tolerant WAD goats compared to the Red Sokoto goats infected with *T. congolense* (Adah et al., 1993) and in WAD sheep infected with *T. brucei*, but this is only during the chronic phase of the disease (Ogunsanmi et al., 1994). Trypanosusceptible Boran cattle infected with *T. congolense*, developed leucopenia and trypanotolerant N'Dama cattle leucocytosis (Paling et al., 1991) and also Zebu cattle and Holstein calves infected with *T. congolense* developed leucopenia (Naylor, 1971; Valli et al., 1979). The development of leucocytosis in trypanotolerant breeds following infection with trypanosomes is confirmed during the current experiment, where a significantly higher WBC count was observed in the sheep during the chronic phase of the infection. The initial sharp rise in WBC counts in the goats was of very short duration and returned quickly to normal value, again indicating less tolerance as compared to sheep.

Apart from innate resistance, acquired resistance is an important component when describing trypanotolerance. Animals infected with a certain strain of trypanosomes and reinfected with the same strain may not become parasitaemic due to the development of specific immunity (Murray et al., 1982). All animals used in this experiment were negative for trypanosomal antibodies before infection, were born and reared in a tsetse-free region and kept there during the experiment and all control animals remained negative for trypanosomes throughout the experimental period. Although, there was a difference in age, it is concluded that before artificial infection, the experimental animals could not have had contact with the strain of *T. congolense* used and therefore, development of specific immunity is excluded.

The role of the immune response in resistance to African trypanosomosis was demonstrated by using mice models indicating differences in susceptibility and related immunological responses (Roelants, 1986). Pinder et al. (1988) infected Baoule and Zebu cattle classified as lower and higher susceptible with *T. congolense* and detected neutralizing antibodies earlier and of a significantly higher titre in the less susceptible animals. In primary infection, N'Dama but not Boran cattle mount a prominent IgG response to congopain (Authié et al., 1993) and it was noted that the response was detected at a time during infection when the levels of parasitaemia appear to be similar in both types of cattle. A positive correlation was observed between the mean level of antibody and the degree of resistance of the breed (Authié, 1994). It appears that the

Djallonke sheep in the current experiment responded in the same way with high levels of IgG to congopain. It has been demonstrated in cattle that declining levels of antibodies are associated with self-cure (Wilson and Cunningham, 1971) and that the resistance of N'Dama compared to Zebu against trypanosomosis is in their ability to limit-level and duration of parasitaemia (Dargie et al., 1979). This is contrary in Djallonke sheep and WAD goats since they showed no tendency for self-cure, remained parasitaemic up to 36 weeks p.i. and the antibody titre increased while the parasitaemia decreased. In the infected sheep, antibodies built up slowly but steadily and were significantly higher than in the infected goats. Apart from the haematological response reported here, other evidence was found earlier for the lower tolerance in WAD goats as compared to Djallonke sheep. A higher mortality and a more affected reproductive capacity were observed in WAD goats but nevertheless, both groups of infected Djallonke sheep and WAD goats showed weight gain not significantly different from the control groups (Goossens et al., 1997a). Griffin et al. (1981b) suggested that, due to shorter generation interval in small ruminants and the higher breeding potential, natural selection over decades in these species for trypanotolerance was quicker than compared to cattle in the same regions of tsetse-infested sub-Sahara Africa. It has also been reported that small ruminants are not the preferred host of Glossina confirmed by blood meal analysis of engorged tsetse flies in which only 0.8% of the blood meals came from these species (Weitz, 1963). Particularly, goats seem to be less preferred by tsetse because of their anti-feeding behaviour such as skin rippling (Snow et al., 1996). The same authors studied the grazing patterns of sheep and goats and it was observed that small ruminants stay closer to the villages, mostly in fallow fields and therefore do not intrude as far into tsetse-infested habitat as cattle do. If goats get less bitten and thus less infected, this may support the hypothesis that acquired resistance developed slower in goats as compared to sheep.

Based on the haematological and antibody response following trypanosome infection, we may conclude that, innate resistance was demonstrated in Djallonke sheep but to a much lesser extent in WAD goats. This better erythropoietic response may be used as an indicator of tolerance in Djallonke sheep and WAD goats against an artificial *T. congolense* infection. Combining this with the observations made on productivity parameters of the same animals over a longer period (Goossens et al., 1997a), provides clear indicators of trypanotolerance for both species; however, Djallonke sheep respond overall better than WAD goats.

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2.3
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Health and productivity of traditionally managed Djallonke sheep and West African dwarf goats under high and moderate trypanosomosis risk

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Abstract

Trypanosome infections, packed red cell volume levels (PCV), body weight and nematode faecal egg counts of village-based small ruminants were monitored in two areas in The Gambia with either moderate or high trypanosomosis risk for 24 and 30 months respectively. Outflows from the flock and new-born animals were recorded and data on housing and management were compiled. Reported mortality rates were higher in goats than in sheep, but for both species highest in the moderate risk area. The peak of trypanosome infections lagged the peak of tsetse densities by 1–3 months in both areas. *Trypanosoma vivax* was the predominant species found in the infected animals, followed by *T. congolense*. Trypanosome prevalence was, in general, higher in sheep than in goats but only significantly higher during Year 1 in the moderate risk area. Trypanosome infection reduced the PCV level significantly and seasonal effects indicated significantly lower PCV levels during the rains. Trypanosome infection significantly depressed weight gain in both species at periods where infection rates were highest. In both species considerably lower weight gains were observed during the rainy season. Abortion rates were higher in goats than in sheep in both study sites, and highest in the high-risk site. Trypanosome infection in ewes in the high risk area increased lamb mortality significantly but had no effect on birth weights, nor on growth rates up to 4 months. Offspring mortality up to 4 months was generally high at both sites. Trypanosome infection in the dam between 3.5 to 7 months post parturition significantly increased parturition interval in both species. Peak faecal egg output occurred at the end of the rainy season and was highest for both species in the moderate risk site. Poor grazing management was found responsible for a seasonal nutritional constraint. Based on these results, these breeds of sheep and goats can be considered as trypanotolerant since they are able to remain productive under high and moderate levels of trypanosome challenge. Nevertheless, trypanosomosis affected their health and production level as shown by reduced PCV levels, depressed weight gains, longer parturition intervals and higher lamb mortality. In addition, during the rains, helminth infections and poor management

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leading to nutritional constraints had also a negative impact on health and production and therefore influenced the innate resilience to trypanosomosis in those indigenous breeds. Adaptations in management may have an equal impact as certain disease control measures to improve biological and economical returns from small ruminants in tsetse infested rural areas.

Keywords: *Trypanosoma* spp.; Sheep protozoa; Goat; Rural development; Nutrition; Helminths

1. Introduction

Trypanosomosis is one of the major constraints for livestock productivity in sub-Saharan Africa. Only trypanotolerant breeds survive, reproduce and remain productive without treatment in tsetse-infested areas (Murray et al., 1982). In the West African traditional farming systems, small ruminants play a crucial role in providing protein (meat, milk) and non-food commodities (manure, hides). They also serve as a cash reserve and a form of saving for the rural population and as protection against agricultural crop failure. Therefore, the number of animals is more important than their individual productivity. Traditional sheep and goat production in different parts of Africa is a very profitable enterprise but returns are strongly determined by biological performance (Sumberg and Mack, 1985; Upton, 1985; Panin and Mahabile, 1997; Itty et al., 1997). For The Gambia, sheep production is even more favourable than cattle production from a financial, gender and equity perspective (Itty et al., 1997). Livestock surveys in The Gambia demonstrated that the cattle population has plateaued while the goat and sheep population are still increasing (DLS/ITC, 1993). The Djallonké sheep and West African Dwarf (WAD) goats are the primary breeds in The Gambia and their trypanotolerant nature has been described as an ability to maintain production under infection (Mawuena, 1987; Osaer et al., 1994; Goossens et al., 1997a). Murray et al. (1982) defined the criteria of trypanotolerance in cattle as an ability to limit reductions in packed red cell volume and to control the level and duration of parasitaemia and showed that these traits were positively genetically correlated with productivity (Murray et al., 1991). However, stress factors such as malnutrition, poor management and intercurrent diseases which are often encountered by animals kept under traditional management, may interact with this natural resistance to trypanosome infection (Agyemang et al., 1990, 1992; Little et al., 1990; Goossens et al., 1997b). Intercurrent disease factors imply peste des petits ruminants (PPR), pasteurellosis, helminthosis and cowdriosis. Under experimental conditions, concurrent infections of *Trypanosoma congolense* and *Haemonchus contortus* resulted in more severe disease in Djallonke sheep (Goossens et al., 1997b) and in N'Dama calves (Kaufmann et al., 1992) than the single infections. Year-round inadequate fodder supply is a major constraint for village-based small ruminants and although they are important in the economy of rural households, farmers will not easily provide supplementary feeding (Russo, 1991). Productivity of trypanosome-infected Djallonke ewes on a low nutrition level was more affected than productivity of those on an adequate level of nutrition under experimental conditions (Reynolds and Ekwuruke, 1988; Osaer et al., 1998a, b, 1999). Nutritional supplementation of young Djallonké sheep kept under high natural tsetse challenge resulted in reduced mortality rates, delayed onset of trypanosomosis and reduced body weight losses (Hecker et al., 1991).

Few reports are available on the importance of trypanosomosis in traditional small ruminant production systems. As a follow-up to controlled experiments, the present village surveys were conducted to study the health and productivity of Djallonke sheep and West African Dwarf goats under various levels of tsetse challenge. Moreover, the impact of natural trypanosome infections was assessed in combination with nutritional and other management constraints which are interfering with the trypanotolerance of these species.

2. Materials and methods

2.1. Study areas and animals

The surveys were carried out in Central River Division South, The Gambia, West Africa. The first study site is situated in the Niamina East area (13°40′N, 14°58′W) and the second study area in the Bansang south area (13°27′N, 14° 41′W), at 200 km and 230 km from the Atlantic coast respectively. Both areas are mainly infested with the tsetse fly *Glossina morsitans submorsitans*, which is considered as the major vector of animal trypanosomosis in The Gambia (Rawlings et al., 1991) and mainly found in dry, canopied, savanna woodland. The prevalence of the riverine species *G. palpalis gambiensis* in both study sites is very low (between 1 and 2.5% of the catches). Population densities of *G. morsitans submorsitans* appears to correlate with the abundance of warthogs (*Phacochoerus africanus*), the preferred host as confirmed by blood-meal identification (Rawlings et al., 1993; Snow and Boreham, 1979). The Niamina East region was ranked as an area of high tsetse challenge and Bansang south region as an area of low to seasonally moderate tsetse challenge which were expected to remain the same for at least 5–10 years, despite influence of demographic, climatic and environmental factors on tsetse populations (Rawlings et al., 1993).

In The Gambia, the wet and humid season lasts from June to the end of October with varying precipitation from 600 to 1600 mm, showing a latitudinal gradient with declining rainfall from west to east of the country. The onset of rains may be earlier in the east (Jabang, 1990; Rawlings et al., 1991). Highest rainfall is concentrated around July and August. The data recorded on the total rainfall for the years 1995, 1996 and 1997 were in the Niamina East area 828, 701, 638 mm, and in the Bansang area 561, 608, 808 mm respectively (Goossens, unpublished data). The dry season lasts for 7 months, from November till May. The Niamina East study site comprises seven adjacent villages situated 1.5 km apart. Each village is surrounded by fields and fallow and situated in an area of open savanna woodland, with riparian woodland and freshwater swamps extending 2.5 km to the north and south. In the Bansang South study site, five villages less than 2 km apart situated in an area of degraded savannah woodlands were included. In both study areas, flocks of sheep and goats are managed in a traditional extensive way. During the dry season, most of the animals roam freely but sheep staying closer to the villages than goats. During the cropping season, starting with planting in July and lasting until after harvest and threshing in November, grazing of small ruminants is restricted in time and area. Other livestock mainly constitute cattle and horses and donkeys as draught animals. An influx of migratory herds and flocks into Niamina East during the dry season in search for natural grazing increases the cattle number from 300 sedentary head to 5000 and coincides with the time of highest tsetse abundance (Wacher et al., 1993). In Bansang, seasonal changes are less extreme.

Apart from these seasonal fluctuations, the overall number of livestock in both study areas remained fairly constant during the survey. In total for both areas, 650 West African Dwarf (WAD) goats and 500 Djallonké sheep were selected at random from the different flocks within each village and identified by ear tags. Sex and age were determined, and for the latter parameter the dentition code as described by Goossens et al. (1998) was used. The newborn animals were consequently ear tagged and included in the monitoring programme. The two study sites Niamina East and Bansang will hereafter be referred to as respectively high and moderate risk site.

3. Sampling methods

The monthly monitoring of animals began in 1995 and was carried out for 24 (moderate risk) and 36 months (high risk), respectively. Animals older than 5 months were bled from the jugular vein (ethylene diaminetetraacetic acid (EDTA)-coated vacutainer tubes (2–3 ml)) for assessing packed red cell volume levels (PCV) and trypanosome infections. The latter were diagnosed using the dark ground technique (DG) and the number of trypanosomes was scored by the method of Paris et al. (1982). The trypanosome species were identified by their movement, size and morphology, if necessary confirmed with a stained blood smear as described by Hoare (1972). In Year 1 and 3 in the high risk site, and during the entire period in the moderate risk site, monthly prevalences were measured according to Thrusfield (1995). Because of the existence of persistent trypanosome infections and quasi absence of self cure in small ruminants following artificial infection (Goossens et al., 1997a) and of the risk of continuous reinfection, monthly trypanosome incidence rate was measured during Year 2 in the high risk study site. Monthly incidence differs from prevalence in that it measures the new infections over the previous month (Thrusfield, 1995). Therefore, from March 1996 till April 1997, all animals which were found infected with trypanosomes during the monthly blood sampling, were treated with a trypanocidal drug (Berenil^R, at a dose of 3.5 mg/kg or 7 mg/kg body weight in case *T. brucei* was diagnosed).

From 20% randomly selected animals, faecal samples were collected monthly during the rainy season and every 2 months during the dry season. The number of strongyle eggs per gram of faeces (EPG) was determined using a McMaster technique with a sensitivity of 100 EPG (Thienpont et al., 1979). Monthly weighing was done with Salter spring balances (50 kg/200 g accuracy). New entries, birth weights and exits were recorded continuously.

Tsetse fly monitoring was carried out using blue F3 box traps (Flint, 1985; Green and Flint, 1986). Arrays of traps were sited, as far as possible, in habitats throughout the area grazed by the village livestock and were operated for 3 days per month. From the data on trap catches, a monthly mean number of tsetse per trap per day (CTD) was calculated (Rawlings et al., 1991). In Year 2, additional traps were included within each village in the high risk area. A seasonal challenge index was calculated as a measure of trypanosomosis risk for the season-year where the peak of trypanosome infections appeared in both study sites, as the product of the mean CTD for that season and percentage mature infection rate (Rawlings et al., 1991).

3.1. Statistical analyses

Continuous variables (PCV, live weight gain, parturition interval, birth weight) were analysed by linear model techniques (GLM procedure; SAS, 1997). The following main effects were included: trypanosome infection status (infection/no infection), sex, age class (6–12/12–24/ > 24 months), village, animal (sex × village) and the interaction trypanosome infection status × age. The animal effect was regarded as random. Additional factors inherent to the observed trait were included. Individual daily weight gains (linear regression, $Y = b_0 + b_1$ days) were calculated and analysed per period, which were defined as a losing and a gaining period. The losing period coincide with the rainy season (June to October), whereas the gaining period corresponded to the dry season (November to May). The model for the estimated regression coefficient (b_1) fitted, apart from the above mentioned effects, also for the effect 'lambed/kidded status'. For the analysis of the parturition interval, censored data were taken into account following the methodology suggested by Tanner (1993). These data were derived from dams which lambed/kidded only once during the first half of the survey but not a second time, despite their continuous presence in the flock. The analysis was carried out with programs specifically written for these data sets (H. Simianer, 1998, personal communication). The linear model included the effects trypansome infection, abortion, season-year, study site, village (within study site) and litter mortality. Two periods were considered to study the effect of trypansomosis on parturition intervals, e.g. trypanosome infection in the first 3.5 months post partum (Period 1) and trypanosome infection between 3.5 and 7 months post partum (Period 2). All hypothesis were tested by the *F*-test (Snedecor and Cochran, 1980). Means of the different groups are presented as least square means ± standard error (mean ± s.e.). Trypanosome prevalence/incidence rates and offspring mortality rates were compared using χ^2-tests.

4. Results

4.1. Flock dynamics

In the original flocks, the proportion of male animals of both species was low in the high risk area (16% rams, 20% bucks) and somewhat higher in the moderate risk area (35% rams, 38% bucks). In addition, males were young, with 80% under 2 years. During the study the male/female ratio remained stable in the moderate risk area, whereas there was a shift to 35% in the high risk area, but males remained predominantly very young. The outflow rate from the flocks ranged between 21% and 26% for both species and both

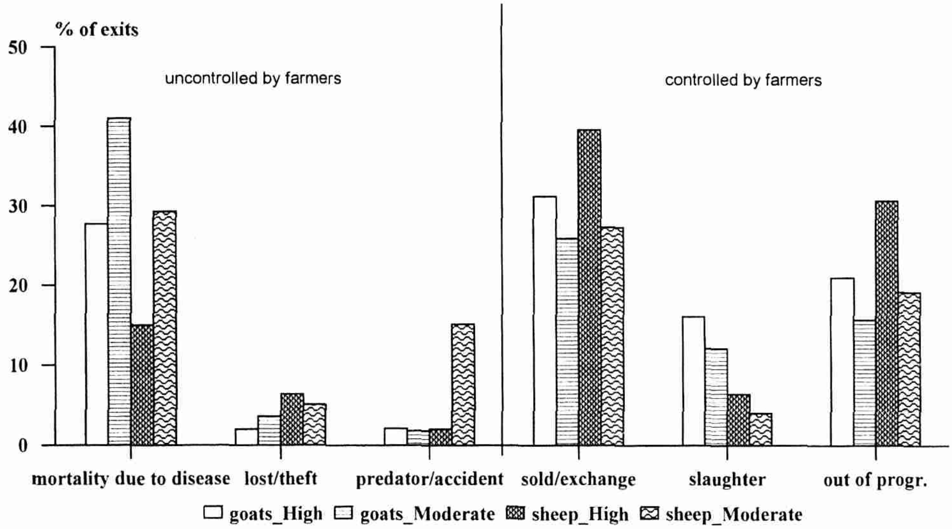

Fig. 1. Exit reasons in flocks of sheep and goats in an area with high or moderate trypanosomosis risk (relative percentages).

areas at the end of the study period. Mortality rates due to disease were higher in goats than in sheep and higher in the moderate than in the high risk site (Fig. 1). A large proportion (70%) of these mortalities occurred during the rainy season. In both study areas, the farmer-controlled offtake was high, with more sheep being sold or exchanged and more goats being slaughtered. Remaining outflow reasons were theft, accident, predator or lost in the bush.

4.2. *Trypanosome infections*

Fig. 2 shows the monthly trypanosome prevalences for Year 1 and 3 (i.e. the number of trypanosome infected animals per monthly number of animals sampled), the monthly trypanosome incidence for Year 2 (i.e. the proportion of newly acquired trypanosome infections since previous sampling) and the mean monthly tsetse catches per trap per day (CTD) for the high risk area. Fig. 3 shows the monthly trypanosome prevalences and the CTD for the moderate risk area.

Peak trypanosome prevalences and incidences for both species in the high risk site (Fig. 2) occurred in the early dry season (November–December). These peaks lagged maximal tsetse densities by 1 month. Peak incidences in Year 2 were reduced by 50% as compared to peak prevalence rates in Year 1 and 3. These rates in Year 1 and 3 indicate the occurrence of persistent infections combined with newly acquired infections in sheep and goats. The predominant tsetse species found was *G. morsitans submorsitans*. *G. palpalis gambiensis*, a riverine species, constituted only 1.1% of the catches. The challenge indices during the early dry season (November till February) in Year 1, 2 and 3 were estimated at 128, 81.1 and 48.3 respectively. The reduction in tsetse densities

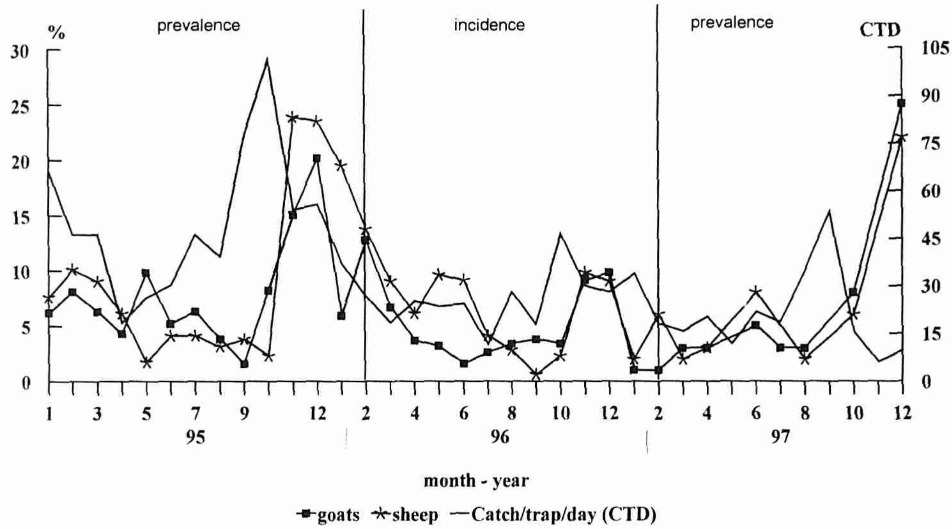

Fig. 2. Prevalence/incidence of trypanosome infections in sheep and goats and mean density of tsetse (mean catch per trap per day) in high trypanosomosis risk area.

(Fig. 2) and challenge indices as observed from Year 2 were due to inclusion of village traps. No statistically significant differences in trypanosome prevalence or incidence were found between sheep and goats in the high risk area (see Table 1; $\chi^2 = \text{ns}$). In the moderate risk site (Fig. 3), peak trypanosome prevalences were seen at the end of the dry

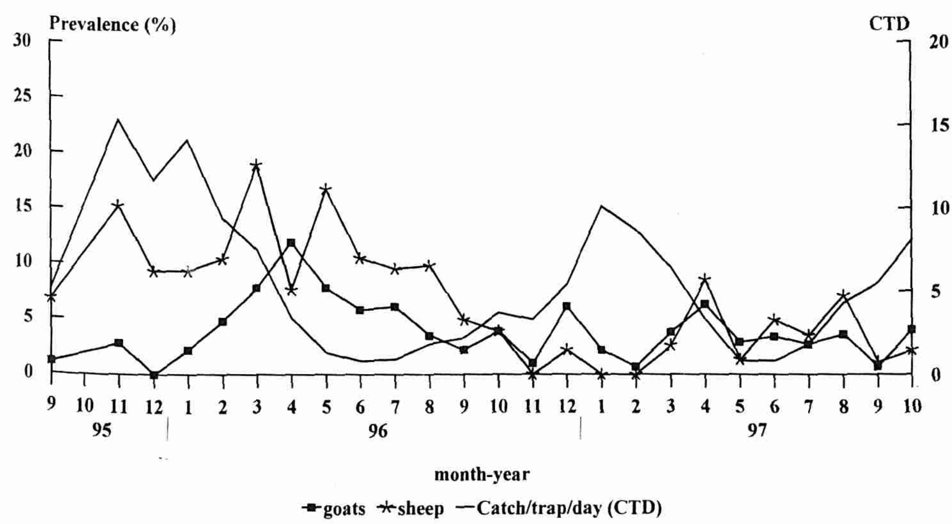

Fig. 3. Prevalence of trypanosome infections in sheep and goats and mean density of tsetse (mean catch per trap per day) in moderate trypanosomosis risk area.

Table 1
Mean trypanosome prevalence/incidence (in %) over different years in sheep and goats in two areas with different trypansosomsosis risk (high or moderate) and levels of significance

Species	High risk area[a]			Moderate risk area[b]	
	Year 1	Year 2	Year 3	Year 1	Year 2
Goats	8.1%	4.0%	7.1%	4.5%	3.2%
Sheep	9.4%	5.6%	7.1%	10.6%	2.8%
χ^2	ns	ns	ns	$p < 0.001$	ns

[a] High risk area: prevalence (Year 1 and 3) and incidence (Year 2).
[b] Moderate risk area: prevalence (Year 1 and 2).

season (March–June), with higher prevalence rates in the first than in the second year. These peak prevalences were preceded by maximal CTD 2–3 months earlier. As in the high risk site, the predominant tsetse species found was *G. m. submorsitans*, constituting 97.5% of the catches. The challenge indices during the late dry season in Year 1 and 2 were 10.5 and 10.2 respectively. Table 1 shows that overall trypanosome prevalence was higher in sheep than in goats in Year 1 ($\chi^2 = p < 0.001$). The age of the animal had a significant effect on trypanosome prevalence in both species ($p < 0.001$) and both areas, with highest infection rates for age groups above 24 months and significantly fewer infections in the age group 6–12 months. In both areas and for both species, significant differences in trypanosome prevalence were seen between villages ($p < 0.01$). The predominant trypanosome species found in both species was *T. vivax* (73% in high risk and >50% in moderate risk), followed by *T. congolense* and very few *T. brucei* infections.

4.3. Packed red cell volume

Trypanosome infection significantly reduced the PCV levels in both species in both areas, even in Year 2 in the high risk study site, despite the treatment of parasitological positive cases with monthly intervals (Table 2). The lower PCV due to trypanosome infection was more pronounced in goats than sheep. In addition, there was a seasonal

Table 2
Effect of trypanosome infection on packed cell volume (PCV) levels in sheep and goats in two areas with different trypanosomsis risk (high or moderate) and over different years (ls means ± s.e.)

	Goats			Sheep		
	High		Moderate	High		Moderate
	Year 1	Year 2[a]	2 years	Year 1	Year 2[a]	2 years
Negative	24.4 ± 0.2%	24.8 ± 0.2%	23.6 ± 0.1%	23.2 ± 0.2%	23.0 ± 0.3%	21.7 ± 0.2%
Positive	20.1 ± 0.4%	21.1 ± 0.4%	20.2 ± 0.3%	21.2 ± 0.4%	21.0 ± 0.4%	20.4 ± 0.3%
Significance	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$

[a] Year 2 in high risk area with monthly treatments of trypanosome infected animals and in Year 1 without treatments. Other effects included age: $p < 0.001$; village: $p < 0.001$; sex (goat: $p < 0.001$; sheep: ns); sampling month: $p < 0.001$.

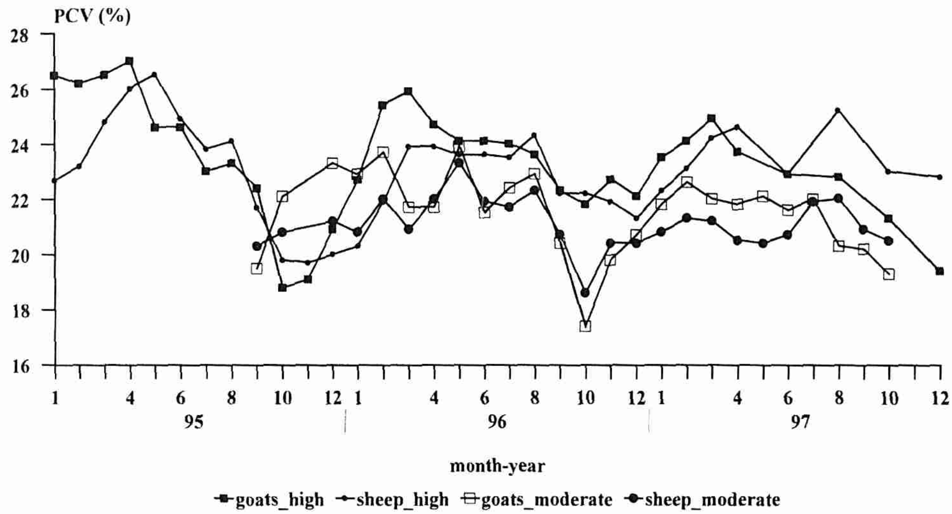

Fig. 4. Mean packed cell volume levels in sheep and goats in high or moderate trypanosomosis risk area.

effect ($p < 0.001$) with lowest PCV levels observed at the end of the rainy and the beginning of the dry season (September–November) (Fig. 4). It was also observed that PCV levels of sheep and goats were lower in the moderate risk area.

4.4. Weight changes

Live weight patterns of adult sheep and goats showed seasonal fluctuations with decrease during the rainy season and increase during the dry season. Tables 3–4 show the effect of trypanosome infection on weight gain in both study sites. For goats in the high risk area (Table 3), a negative effect of trypanosomosis on weight gain was observed in all periods except during the '1996/1997 gaining' period. For the sheep, trypanosome infection depressed weight gain only during '1995/1996 gaining' period or dry season. The overall observed differences between periods confirmed weight losses or lower weight gains in both species during the rains. In the moderate risk area, trypanosome-infected animals of both species showed lower weight gain compared to the negative ones but were only significantly reduced in infected sheep during the losing periods (Table 4). Similar to the high risk site, the difference in weight gain between periods (seasons) was obvious. In general for both study sites, the effect of age on weight gain was significant and confirmed differences between the age groups. In addition, if parturition occurred within a period, daily weight gain was influenced significantly ($p < 0.001$).

5. Reproduction

The monthly distribution of parturitions showed no clear seasonality, although the number of parturitions was slightly higher between August and November as compared to

Table 3
Effect of trypanosome infection on daily weight gain (g/day) in sheep and goats in an area with high trypanosomosis risk: daily weight gain for trypanosome-infected and negative animals in gaining (dry season) and losing (rainy season) periods (levels of significance and ls means ± s.e.)

Infection	Sheep				Goats			
	1995 losing ($n = 174$)	1995/1996 gaining ($n = 179$)	1996 losing ($n = 181$)	1996/1997 gaining ($n = 159$)	1995 losing ($n = 222$)	1995/1996 gaining ($n = 208$)	1996 losing ($n = 219$)	1996/1997 gaining ($n = 201$)
	ns	$p < 0.01$	ns	ns	$p < 0.05$	$p < 0.01$	$p < 0.05$	ns
Negative	13.8 ± 9.0	43.2 ± 4.4	13.9 ± 5.2	58.1 ± 4.4	1.7 ± 6.5	61.6 ± 4.0	25.6 ± 3.7	56.6 ± 3.1
Positive	21.4 ± 15.4	30.9 ± 4.9	10.7 ± 10.5	58.4 ± 6.2	−23.0 ± 12.4	41.8 ± 6.0	7.9 ± 9.4	59.2 ± 5.8

Other effects included age: $p < 0.01$; trypanosome infection × age: ns; village (goat: ns, sheep: $p < 0.05$); sex (goat: $P < 0.05$, sheep: $p < 0.05$); parturition (sex): $p < 0.001$.

Table 4
Effect of trypanosome infection on daily weight gain (g/day) in sheep and goats in an area with moderate trypanosomosis risk: daily weight gain for trypanosome-infected and negative animals in gaining (dry season) and losing (rainy season) periods (levels of significance and ls means ± s.e.)

Infection	Sheep				Goats			
	1995/1996 gaining ($n = 137$)	1996 losing ($n = 120$)	1996/1997 gaining ($n = 140$)	1997 losing ($n = 128$)	1995/1996 gaining ($n = 137$)	1996 losing ($n = 126$)	1996/1997 gaining ($n = 216$)	1997 losing ($n = 180$)
	ns	$p < 0.05$	ns	$p < 0.05$	ns	ns	ns	ns
Negative	34.6 ± 8.3	24.2 ± 4.8	34.9 ± 3.4	29.7 ± 3.4	31.3 ± 24	6.8 ± 5.6	48.7 ± 4.8	14.8 ± 4.3
Positive	43.9 ± 9.7	2.6 ± 9.7	29.9 ± 10.6	13.9 ± 7.0	83.1 ± 40	0.0 ± 11.4	33.7 ± 10.5	10.7 ± 7.7

Other effects included age: $p < 0.01$; trypanosome infection × age: ns; village: $p < 0.05$; sex: $p < 0.05$; parturition (sex): $p < 0.05$.

Table 5
Effect of trypanosome infection and offspring mortality on parturition interval in sheep and goats in an area with high or moderate trypanosomosis risk (levels of significance and ls means ± s.e.)

	Goats ($n = 237$)		Sheep ($n = 175$)	
	p	ls means ± s.e.	p	ls means ± s.e
Overall average		273.7 ± 4.9		287.3 ± 6.2
Unaffected reference	–	266.6 ± 6.3	–	271.9 ± 7.6
Trypanosome infection Period 1[a]	ns	262.6 ± 14.5	ns	297.6 ± 19.6
Trypanosome infection Period 2[b]	$p < 0.001$	357.3 ± 18.0	$p < 0.001$	347.1 ± 21.3
Litter mortality < 30 days	ns	243.5 ± 10.6	–	–
Litter mortality 30–91 days	$p < 0.01$	215.2 ± 17.0	ns	241.6 ± 15.7
Study site	ns		ns	
High		281.8 ± 7.0		294.0 ± 8.5
Moderate		262.0 ± 8.7		273.7 ± 12.4

[a] If infection occurs during the 3.5 months post partum.
[b] If infection occurs between 3.5 and 7 months post partum.
Other effects included abortion (goats: $p < 0.05$); village (area) (goats: $p < 0.001$, sheep: ns); season-year (goats: ns, sheep: $p < 0.05$).

other months (between 25% and 30% of the total parturitions). Average litter size and offspring's birth weights for both species were not different between study sites and arithmetic means were respectively for goats and sheep for littersize 1.6 and 1.2 and for birth weight 2.1 kg and 2.4 kg. Abortion rates were higher in goats than in sheep in both areas, and highest in the high risk site (goats high: 5.3%, goats moderate: 2.4%, sheep high: 1.1%, sheep moderate: 1.4%). The occurrence of premature births was below 2% in both study sites and in both species. Age at first lambing/kidding was calculated for animals born during the survey and were, despite high individual variations, higher in the high risk site, with averages for high and moderate sites in goats 383 days ($\sigma = 100$) and 344 days ($\sigma = 93$), respectively, and in sheep 437 days ($\sigma = 85$) and 401 days ($\sigma = 115$) respectively. The effect of trypanosome infection on parturition intervals was analysed and results are presented in Table 5. The unaffected reference is an estimated value based on dams without trypanosome infection and with surviving offspring. A trypanosome infection in Period 2 significantly increased the parturition interval in both species. Parturition intervals for both species were shortened when litter died before they reached 90 days. For goats this shortening was statistically significant ($p < 0.05$). There was no effect of trypanosome infection on offspring's birth weight during pregnancy in the dam. Similarly, infection during late pregnancy and lactation did not influence offspring growth up to 4 months, although lambs from infected ewes had lower weight gains than those from non-infected ewes (Table 6). Trypanosome infection in the ewe, during late pregnancy and/or lactation period, increased ($p < 0.01$) lamb mortality in the high risk site. Estimates of mortality in offspring up to 4 months are presented in Table 6.

5.1. Nematode egg excretion

The mean monthly EPG for sheep and goats are given in Fig. 5 for both areas and both species. Peak EPG values were observed at the end of the rainy season (September–

Table 6
Effect of trypanosome infection during late pregnancy and lactation period of the dam on offspring daily weight gain (g/day; ls means ± s.e.) and offspring mortality up to 4 months (%) in sheep and goats in an area with high or moderate trypansosomosis risk (levels of significance)

	Goats				Sheep			
	High		Moderate		High		Moderate	
	Daily gain (g/day)	Mortality (%)	Daily gain (g/day)	Mortality (%)	Daily gain (g/day)	Mortality (%)	Daily gain (g/day)	Mortality (%)
Not infected	63.6 ± 4.8	36.1	61.4 ± 2.8	31.3	89.5 ± 7.4	31.7	71.6 ± 8.4	34.0
Infected	61.9 ± 6.1	33.8	66.5 ± 5.0	36.4	82.4 ± 7.3	46.4	60.3 ± 12.8	46.9
Significance	ns	ns	ns	ns	ns	$p < 0.01$	ns	ns

October) with no clear differences between sheep and goats. EPG values were significantly higher in the moderate risk area than in the high risk area for both species (see Fig. 5).

5.2. Housing-management for contact host-vector

Overall 65% of the households ($n = 56$) provided a roof for the goats against 6.5% for sheep. More than 50% of the night shelter for goats had a wall against only 21% for sheep. Sheep were tethered at night in 40% and goats in 25%. In the moderate risk site some households provided a slatted floor for sheep and goats. In the high risk area and

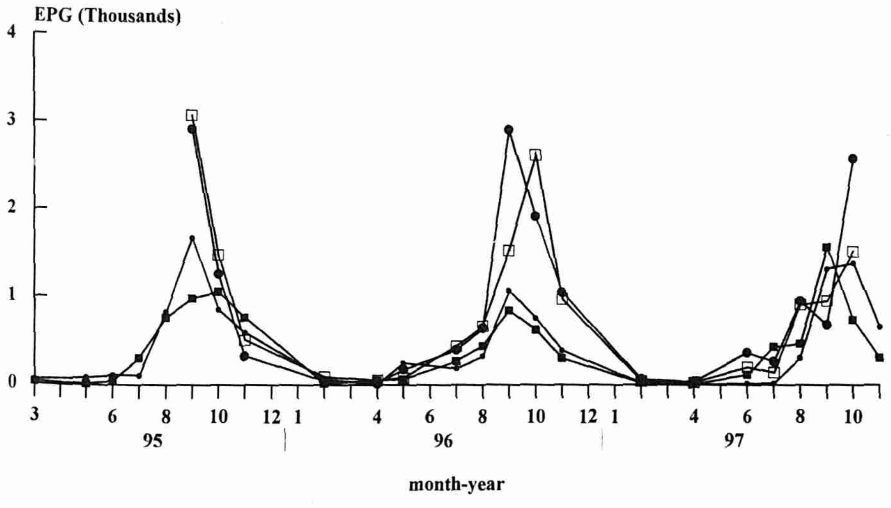

Fig. 5. Mean Trichostrongylidae egg excretion (EPG) in sheep and goats in high or moderate trypanosomosis risk area.

during the rainy season, all sheep and goats were herded either in bush area (60% for both species), swamp area (24% for sheep and 32% for goats) and close to the villages (10% for sheep and 13% for goats). In the moderate risk area, 37% of households were tethering their animals, 50% were herding and the remainder were left free during the rains. Tsetse traps sited within the village in the high risk area caught a non-negligible number of tsetse flies, which followed a similar seasonal density pattern as described above (Figs. 2 and 3). Therefore, it was clear that throughout the year, small ruminants were entering tsetse infested habitats, even by staying close to the villages in the high risk area. During the dry season grazing management was non existing, animals were roaming free, either close to the villages or further down to the river sides to graze on rice fields.

6. Discussion

These surveys were carried out to broaden the knowledge on the health and productivity in the rural small ruminant production systems with special attention for the impact of trypanosomosis. Based on the results, both sheep and goats could be considered as trypanotolerant since they remained productive under different levels of trypanosome challenge. Nevertheless, whilst production was affected by trypanosomosis, other intercurrent factors such as helminthosis and seasonally poor nutrition were found equally important. Improvements are suggested with the aim of increasing the biological and economical benefits from small ruminants in rural settings.

The low proportion of male animals and their young age before and during the study demonstrated that males are sold and/or slaughtered at a young age. This could result in a poor breeding potential for the flocks at village level. This is in agreement with previous studies in The Gambia by Greenwood and Mullineaux (1989) and in Nigeria by Sumberg and Mack (1985). Overall mortality rates were higher in goats than in sheep which corresponds with earlier studies in The Gambia (Greenwood and Mullineaux, 1989). The majority of mortality occurred in the rainy season, which coincided with the peak helminth infestation, poorer nutritional condition and lowest haematocrit levels. Farmer-controlled offtake was revealed to be important. This confirms that small ruminants serve as an insurance against crop failure and emergency cash reserve as recorded in The Gambia (Bennison et al., 1997) and in Nigeria (Sumberg and Mack, 1985).

In the high risk area, a slightly higher rate of trypanosome infections were found in sheep than in goats. In the moderate risk area, the prevalence was significantly higher in sheep than in goats in year one. Compared to cattle herds resident in the high challenge area, where peak prevalences went up to 50% around the same time (Clifford, 1997, personal communication), peak trypanosome prevalences were lower in small ruminants. Factors contributing to this include the low feeding success of tsetse on sheep and goats, illustrated previously by blood-meal analysis in The Gambia (Snow et al., 1996). This may be related to their smaller body size and anti-feeding behaviour such as leg kicks and skin rippling (Vale, 1977). Another factor may be a limitation of contact between tsetse and small ruminants due to grazing patterns. When tsetse densities are highest, sheep and goats in the high risk site are still grazing on harvested fields in the vicinity of villages. Here challenge to tsetse is low because these open fields are not a suitable habitat.

However, a continuous trypanosomosis risk within the villages was confirmed in the high-risk site. In that respect, horses and donkeys, which are kept within the village, showed monthly incidence rates up to 35% during the peak period in this site (D. Faye, 1998, personal communication). When the dry season progresses, small ruminants graze further into tsetse-infested areas whilst migrating cattle herds have joined the same pastures, resulting in a reduction of the number of tsetse per head (Wacher et al., 1993). Nevertheless, peak trypanosome infections in small ruminants followed peak tsetse density as predicted by a model of the epidemiology of trypanosomosis (Milligan and Baker, 1988). Since the rainfall patterns at the two sites were not very different, it is likely that the observed differences in the apparent seasonality of *G. m. submorsitans* are related to differing host densities and/or availability and factors, particularly density dependent mortality, regulating the tsetse populations (W.F. Snow, 1998, personal communication). The monthly incidence rate in Year 2 in the high risk area reflected infections newly acquired during one month whilst the higher infection rates observed in Year 1 and 3 measured in addition the persistent infections acquired during previous months. These observed differences confirmed the existence of persistent trypanosome infections which has been reported earlier in sheep and goats following a single artificial infection (Goossens et al., 1997a). The cattle prevalence rates in the high risk site remained the same (12%) during the 3 years with the predominant trypanosome species being also *T. vivax* (D.J. Clifford, 1997, personal communication). The lower infection rates measured in younger animals have also been observed by Greenwood and Mullineaux (1989). As young offspring graze with their dam, thus lower infection rates could only be due to their smaller size.

Packed red cell volumes, measured as an indicator of health, are affected significantly by trypanosomosis and are a reflection of anaemia in both species. This corresponds to what is found in experimental infections with a clone of *T. congolense* causing chronic anaemia (Osaer et al., 1994; Goossens et al., 1997a). Apart from the effect of trypanosome infection on PCV, the seasonal effect significantly lowered levels during the rains. A major factor contributing to this reduction in PCV is the peak of helminth infestations, demonstrated by the peak egg excretions during the rains. Fritsche et al. (1993) found an overall prevalence of 67% of *Haemonchus contortus* in small ruminants in The Gambia whilst the adult populations peak during the rains. The latter species is pathogenic for ruminants because the larval stages (L4 and L5) and adult worms are blood-sucking parasites of the abomasum and cause anaemia. Heavy acute infections may lead to death. Dual infections of *T. congolense* and *H. contortus* in controlled experiments have proven the severeness of the mixed infections as opposed to both single infections on haemotological parameters in sheep (Goossens et al., 1997b) and N'Dama calves (Kaufmann et al., 1992). Another factor reducing PCV levels during the rains may be inadequate nutrition due to poor grazing management, whereby small ruminants are either tethered or herded during a restricted time. During the wet season, crops have to protected and all available labour is involved in crop cultivation. This management causes a nutritional stress for small ruminants which reflects immediately in depressed growth or weight losses but may reduce also PCV. Previous work has shown that low levels of nutrition decrease PCV levels in sheep (Wassink et al., 1997) and aggravate the course of trypanosomosis in sheep (Katunguka-Rwakishaya et al., 1993, 1995; Osaer et al., 1998b).

During the rainy season small ruminants had low weight gains or lost weight because of helminth infestations and nutritional constraints as mentioned earlier. Trypanosomosis depressed weight gain in both species in high and/or moderate risk site, but more obviously in the periods and study site with higher infection rates. In comparison, following artificial trypanosome infections, weight gains were reduced but not significantly in female WAD goats and Djallonke sheep post infection (Osaer et al., 1994; Goossens et al., 1997a). However, in another study, Djallonke rams showed significantly lower weight gains compared to their controls in the chronic stage of infection (Osaer et al., 1997).

Trypanosome infection increased significantly parturition intervals of both sheep and goats. Reproductive disorders such as periods of anoestrus, or embryonic or early foetal death in early stage of pregnancy might occur as a result of trypanosomosis (Goossens et al., 1997a; Osaer et al., 1998a) and considerably increase the parturition intervals. The higher abortion rate primarily found in goats under high tsetse challenge, accords with earlier observations on experimental trypanosome infections (Goossens et al., 1997a). Trypanosome infection of the ewe increased mortality in the lambs. Negative effects on lamb survival and/or growth following *T. congolense* infection in Djallonké ewes have been shown earlier (Goossens et al., 1997a; Osaer et al., 1998b, 1999), giving evidence for reduced milk production in the latter. In contrast, kids from infected does had similar growth rates as those from negative ones in this study, which was also observed by Goossens et al. (1997a). Offspring mortality rates in the present study were found higher than those reported for village based sheep and goats in Nigeria (Sumberg and Mack, 1985; Otesile and Oduye, 1991). Apart from trypanosomosis in the dam, other determining factors such as litter size, sex and season of birth, genuine poor milk production or an affected udder could also contribute to this high offspring mortality (Nash et al., 1996). Especially in Djallonke ewes milk production seems to be low and of a short duration. Highly pregnant and early lactating dams could be fed supplements to boost the milk production. In addition, harsh environmental conditions with the presence of predators are a constant threat to young animals.

The seasonal pattern of strongyle egg output found during this survey was comparable to other epidemiological studies done in The Gambia (Greenwood and Mullineaux, 1989; Fritsche et al., 1993; Goossens et al., 1998). Sheep and goats in the moderate site showed a higher nematode egg excretion compared to the high risk site which might be due to more frequent tethering in the former site, leading to higher infection risks. Higher helminth infestation reflected also in the lower PCV levels and higher mortality rates observed in that site. A reduced re-infection was demonstrated in N'Dama cattle when they were moved more frequently to new night holding places during the rainy season (Kaufmann et al., 1993). In that respect, the most appropriate housing for small ruminants which prevents reinfection are slatted floors.

Out of these surveys, it was concluded that both species are trypanotolerant since they remain productive under various tsetse challenges. Despite trypanosomosis being an important constraint to production, other factors such as helminth infections and nutritional constraints during the rainy season equally affect the health and productivity of village-based sheep and goats. In addition, they may interact with their resilience to the effects of trypanosomosis. Offspring mortality was found an important constraint limiting productivity. It was suggested that on-farm improvements on management, housing and nutritional aspects could considerably contribute to a reduction of mortality and increased productivity. These measures, apart from disease control, could considerably increase biological and economical returns from these species.

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CHAPTER 3

REPRODUCTIVE PERFORMANCE OF TRYPANOTOLERANT SMALL RUMINANTS FOLLOWING TRYPANOSOME INFECTION

3.1

Long term effects of an experimental infection with *Trypanosoma congolense* on reproductive performance of trypanotolerant Djallonke ewes and West African Dwarf does.

Goossens, B., Osaer, S., and Kora, S. 1997.
Research in Veterinary Science 63: 169-173.

3.2

Evaluation of semen quality and reproductive performance of West African Dwarf rams experimentally infected with *Trypanosoma congolense*.

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Long-term effects of an experimental infection with *Trypanosoma congolense* on reproductive performance of trypanotolerant Djallonké ewes and West African Dwarf does

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SUMMARY

Ten West African Dwarf (WAD) does and 12 Djallonké ewes were artificially infected with a West African strain of *Trypanosoma congolense* and observed over two years. The infected animals showed a chronic anaemia together with a persistent parasitaemia but very low mortality and increase in body weights was not significantly different from the control. In the infected sheep significant differences were found in offspring production at three and five months due to a higher mortality among the lambs. The infected goats had more abortions and stillbirths and period to first kidding, total number of parturitions, production at birth, one, three and five months were significantly different from the controls. A productivity index was calculated and revealed that a chronic *T congolense* infection significantly decreases the productivity of WAD goats during two years whereas in Djallonké sheep, the loss in productivity is recovered after one year. Although both species are regarded as trypanotolerant, the Djallonké sheep show a better tolerance to a chronic *T congolense* infection that the WAD goats.

BREEDS of small ruminants, such as WAD goats and Djallonké sheep which are indigenous to West Africa are highly prolific (Sumberg and Mack 1985, Wilson 1989) and are considered as trypanotolerant (Mawuena 1987).

In trypanosomosis an acute, chronic and recovery phase is recognised. Apart from anaemia and other haematological changes, weight loss, anorexia, weakness and elevated body temperature are also observed following trypanosome infection (Anosa 1988, Griffin 1978). Pathological changes in reproductive organs as a result of trypanosomosis with as a consequence temporary or permanent anoestrus, abortions, stillbirths or neonatal deaths have been observed and described (Ikede and Losos 1972, Ikede et al 1988, Sekoni 1994, Llewelyn et al 1987, Edgehere et al 1992). The mechanism through which the reproductive system is damaged depends on the trypanosome species. Although *T congolense* is considered as strictly intravascular it has also been reported to cross the placental barrier and cause abortion in East African goats (Ogaa et al 1991). Atrophy of reproductive organs in non-invasive trypanosome infections is more likely to be caused by indirect mechanisms such as thrombosis in the gonadal blood vessels, severe anaemia and thermal effects of pyrexia. It was also suggested that fever, stress due to fever and anorexia activate the hypothalamo-pituitary-adrenal (HPA) axis resulting in production of corticosteroids which will stimulate the production of prostaglandins and oestrogen from the placenta causing intra-uterine contractions and abortion (Elhassan et al 1989). In addition, activation of the HPA axis might lead to changes in gonadotrophin level and cause infertility (Mutayoba et al 1995a, b).

Indigenous breeds of small ruminants in Kenya showed more resistance to a trypanosome infection than the exotic breeds based on weight changes and mortality rates (Griffin and Allonby 1979). Trypanotolerant breeds of sheep and goats also seem to possess the capacity of returning quickly to normal oestrus cycle after infection with trypanosomes. The greater the tolerance, the higher the retention of fertility resulting in the term higher residual fertility (Mutayoba et al 1988a). In trypanotolerant breeds of goats following *T congolense* infection, the cyclic activity declined more slowly compared to trypanosusceptible breeds and the decline in the hormonal values was less (Mutayoba et al 1988b). They concluded that clinical tolerance is correlated with residual fertility which is based on the quicker return of a normal oestrus cycle after infection with trypanosomes. In WAD goats reduced kidding percentages following *T congolense* have been reported (Kanyari et al 1983). Djallonké sheep under natural trypanosome challenge showed high parasitaemia levels and severe anaemia but their reproduction capacity was not affected (Mawuena 1986, 1987).

The present experiment was designed to study the effect of a chronic experimental trypanosome infection on the clinical performance of WAD does and Djallonké ewes, to assess the long-term effect on fertility and to evaluate the survival and growth rates of the offspring.

MATERIALS AND METHODS

The experiment took place between February 1992 and August 1994 on station at the coastal site of the International Trypanotolerance Centre (ITC), The Gambia, West Africa. The climate is sudano-sahelian with a mean annual rainfall of approximately 1000 mm.

Nineteen female goats and 23 female sheep were selected from the ITC breeding flocks. All animals were born and reared at the coastal site, an area known to be free of tsetse with virtually no risk of trypanosomosis (Rawlings et al 1993). Before the start of the experiment all animals were found negative for trypanosomes following examination of the buffy coat by dark ground (DG) microscopy (Murray et al 1977). All animals were aged between six and 18 months at the beginning of the experiment. Five out of 23 sheep

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and 11 out of 19 goats had one parity and were equally distributed over both groups. The animals were allocated according to age and parity into control and infected groups. All animals were vaccinated against Peste des Petits Ruminants (PPR-Tissue Culture Rinderpest vaccine, Hann lab., Dakar) and this was repeated every six months. They were wormed (Panacur®, 10 per cent, Hoechst, Germany, 7·5 mg kg^{-1} bodyweight or Ivomec®, 1·0 per cent, Merck Sharp and Dohme, USA, 1ml per 50 kg bodyweight) and sprayed with acaricide (Gammatox®, Coopers 8 g 10^{-1} litre water or Ovipor®, 2·5 per cent C-Vet, UK). Worming and spraying was repeated every four weeks during the wet season. During the experiment, medical treatment was provided where needed and a postmortem was performed whenever death occurred. The animals were confined and separated from males during a period of three months prior to infection. Five days after infection a ram and a buck were introduced into the flocks. All animals were confined for another month in order to optimise heat detection and servings.

While being confined they were given a full base diet of groundnut hay, sesame cake and rice bran, calculated on daily weight gain of 20 g. After one month they went out for grazing and received a supplement of rice bran and sesame cake. During the night they were kept confined. The ram and the buck were replaced every eight months and male offspring were castrated at the age of six months.

On February 19, 1992, 12 out of 23 sheep and 10 out of 19 goats were infected with a West African strain of *T congolense*, ITC 84. The strain used was a cloned stabilate derived from a goat found positive on an earlier occasion during a village survey (Upper River Division, The Gambia). The pathogenicity of the strain has been assessed in trypanosusceptible Zebu cattle (Dwinger et al 1992). The stabilate was expanded in mice and the animals were inoculated intradermally at five sites on the left body flank with 1 ml infected mouse blood, containing at least 10^4 bloodstream forms. Infection took place on day 0 of the experiment and all times referred to in the text relate to time post-infection. Following infection all animals were bled daily for the first 16 days, thereafter three times a week from week 3 until week 12, then once weekly until week 37 and thereafter once monthly until August 1994. Jugular vein blood samples (3 ml) were collected in ethylenediamine tetraacetic acid (EDTA)-coated vacutainer tubes. Packed red cell volume (PCV) levels were measured as an estimation of anaemia using the capillary microhaematocrit centrifugation method. Trypanosomes were detected and an estimation of parasitaemia level was made by the DG-method. The number of trypanosomes was scored as described by Paris et al (1982). The animals were weighed weekly until week 40, thereafter monthly using a Salter Spring balance (50 kg, accuracy 200 g).

Reproduction parameters included oestrus/serving detection, abortions, stillbirths, premature or normal parturition, twin or single births. Offspring were weighed within 24 hours of birth and at one, three and five months. In addition, a productivity index was calculated per female and per year based on the annual kidding/lambing percentage, offspring mortality up to five months and the weight of offspring at five months (FAO 1980).

Statistical methods

The various traits were analysed by unbalanced analysis of variance. PCV percentage and body weight were subjected to a model which included the effect of treatment, animal (nested in treatment), sequence of sampling, and the interaction treatment* sequence. These parameters were analysed for an acute phase (0–12 weeks) and for a chronic phase (week 13–96). To compare the reproductive performance of infected and control animals, the reproductive traits (total number of offspring, average production at birth, average production at month one, three and five and the time to first lambing/kidding) were calculated per dam for the period of one year (19.02.1992–19.02.1993) and for the period of two years (19.02.1992–19.02.1994) post infection. All these reproductive traits were subjected to analysis of variance with as main effects treatment and age of the dam, regarded as a quadratic regression. The offspring performance data were analysed only with respect to the dam. In addition, a chi-square test was performed to compare the number of parturitions between treatments. All hypothesis were tested by the F-test, where in some tests the approximation of Sattherthwaite (Snedecor and Cochran 1980) had to be used. Error probability of less than 5 per cent was regarded as significant.

RESULTS

Clinical observations

During the two year period few deaths occurred. Out of the infected goats two died, one in week 16 and one in week 69. On postmortem, the infected goats had pathological signs of trypanosomosis including pale mucosae, inflamed lymph nodes, enlarged spleen, hydrothorax and hydropericardium. One control goat died (week 100) of a severe metritis two days after delivering a triplet. In the control sheep there were two deaths, one died in week 49 and one in week 68. One sheep died after severe pneumonia and the other one of unknown reasons. None of the infected sheep died. During the first month post infection it was observed that most of the sheep and some goats came in oestrus and were seen to be served by the males.

Packed cell volume, parasitaemia score and body weights

During both acute and chronic phase, the infected sheep and goats had significantly lower PCV levels than the control groups (Tables 1 and 2). In all infected animals, parasites occurred in the blood between four and seven days post infection. The first peak was reached at day 12 for sheep and day eight for goats. All infected sheep and goats

TABLE 1: Least square means ± SE for PCV percentage and body weights during acute and chronic phase for control and infected goats

	Acute phase (0-12 weeks)			Chronic phase (13-96 weeks)		
	Control	Infected	Sign	Control	Infected	Sign
PCV %	30·6±0·1	22·2±0·1	***	30·1±0·1	24·3±0·2	***
Body weight (kg)	14·3±0·05	13·1±0·05	ns	20·4±0·3	18·0±0·1	ns

TABLE 2: Least square means ± SE for PCV percentage and body weights during acute and chronic phase for control and infected sheep

	Acute phase (0-12 weeks)			Chronic phase (13-96 weeks)		
	Control	Infected	Sign	Control	Infected	Sign
PCV %	28·9±0·1	24·8±0·1	***	28·9±0·1	25·5±0·1	***
Body weight (kg)	17·7±0·05	18·1±0·05	ns	24·3±0·3	23·4±0·1	ns

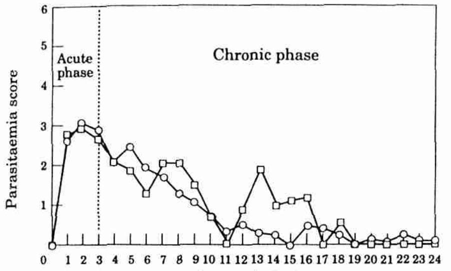

FIG 1: Average parasitaemia scores of *T congolense* infected WAD goats and Djallonké sheep. Key: □ goats; ○ sheep

remained parasitic for 11 months with decreasing peaks. After month 11, the sheep had low mean levels of parasitaemia but some stayed parasitaemic until month 24, whereas the goats had a second increase at month 12 but became all negative at 18 months (Fig 1). There were no significant differences between sheep and goats in parasitaemia level. There were no significant differences between infected and control animals in mean body weights throughout the experiment (acute and chronic phase) indicating that all animals gained equally in weight. (Tables 1 and 2, Fig 2).

Reproduction

Table 3 summarises some reproduction parameters of all four groups for the complete two year period. Abortions were only seen in the infected groups whereas all groups had stillbirths but more in the infected groups. Although the control goats had a lower number of parturitions they produced more kids than the control sheep due to higher numbers of twin births. It was also observed that two of the infected sheep and six of the infected goats did not produce any offspring during year 1. A productivity index for the dams/does of the different treatment groups was calculated for year 1 and year 2 separately to observe possible differences in the potential of infected dams and does to return to normal productivity (Table 4).

The results of the analysis of variance given as least-square means ± SE and significance of treatment effects are presented in Tables 5 and 6 for goats and sheep respective-

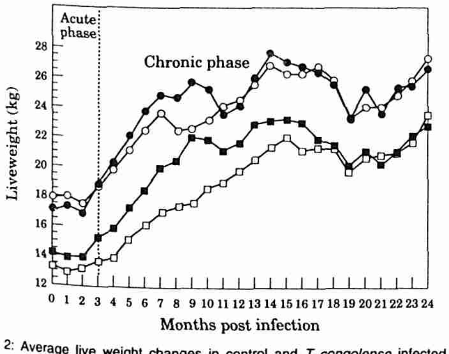

FIG 2: Average live weight changes in control and *T congolense* infected WAD goats and Djallonké sheep. Key: ■ control goats; □ infected goats; ● control sheep; ○ infected sheep

TABLE 3: Parameters of reproduction (absolute numbers) of sheep and goats for the two year period

Parameter	Sheep cont	Sheep inf	Goats cont	Goats inf
Number of females	11	12	9	10
Total number of parturitions	25	31	21	13
– single	24	27	7	7
– twin	1	4	13	5
– triplet	–	–	1	–
Total number lambs/kids	26	35	36	17
Number of stillbirths	1	4	4	5
Total number surviving five months	23	22	27	11
Abortions	–	1	–	3
Prolificacy rate	1·04	1·12	1·71	1·30

ly. For all reproductive traits, the age of the dam/doe was not significant. In the infected goats all traits were significantly lower than those of the controls regarding both the first year and the complete two-year period. In the infected sheep the average production at birth calculated per dam was not significantly different compared to the controls. In the first year the average production at month one, three and five in the infected ewes was significantly lower compared to those of the control ewes in the first year. Regarding the two-year period, the infected ewes still had lower average production of three- and five-month-old offspring. The number of parturitions was significantly lower for the infected goats compared to the control group. For the sheep this parameter was not significantly different. Subsequent kidding/lambing intervals were found shorter in goats than in sheep, but there was no significant difference between control and infected groups.

DISCUSSION

Following *T congolense* infection moderate clinical signs were noticed among the infected animals. Mortality possibly due to trypanosomosis only occurred in two goats. The infected sheep and goats developed anaemia concomitant with the onset of parasitaemia and both species remained anaemic for an extended period while having persistent parasitaemia. The infected sheep and goats only self-cured after a period of 18–24 months. PCV decline was more important in infected goats compared to the sheep, indicating that goats suffered more from the *T congolense* infection in terms of clinical appearance. Similar observations were made in a previous on-station experiment where the trypanotolerant nature of the Djallonké sheep and WAD goats was evaluated using two *T congolense* strains with different pathogenicities (Osaer et al 1994). Following artificial infection with *T congolense*, comparing trypanotolerant N'Dama with trypanosusceptible Zebu cattle, the latter showed higher levels of parasitaemia and lower levels of PCV percentage (Dwinger et al 1992). Trypanotolerant sheep and goats seem to react differently than trypanotolerant cattle in terms of controlling the parasitaemia and the anaemia. Although trypanosome infected Djallonké rams

TABLE 4: Productivity index of sheep and goats under an experimental infection with *T congolense*

	Year 1		Year 2	
	Control	Infected	Control	Infected
Sheep	13·3 kg	4·9 kg	15·5 kg	16·5 kg
Goats	11·1 kg	1·2 kg	15·6 kg	7·5 kg

Weight of five-month-old offspring produced per female maintained per year (kg). Results for year 1 and year 2

TABLE 5: Reproductive parameters for West African Dwarf goats: least-square means ± SE over one year and two years for control and infected groups with significance

Trait	1 year Control (n = 9)	1 year Infected (n = 10)	Significance	2 years Control (n = 8)	2 years Infected (n = 9)	Significance
Average number of offspring	1·0±0·2	0·1±0·2	**	3·7±0·3	1·3±0·3	***
Average production at birth (kg)*	1·7±0·3	0·2±0·3	**	6·0±0·5	2·3±0·5	***
Average production at 1M (kg)*	4·3±0·8	0·6±0·7	**	12·8±1·1	4·8±1·1	***
Average production at 3M (kg)*	8·0±1·4	1·1±1·4	**	19·4±1·7	6·4±1·6	***
Average production at 5M (kg)*	10·7±2·0	1·5±1·9	**	24·4±2·6	8·1±2·5	***
Time to first kidding (days)†	280·4±17·1	369·6±17·1	**			
Average kidding interval (days)				231·2±18·2	229·4±24·4	ns

** P<0·01
*** P<0·001
* all observations per doe
† one single observation per doe

seemed to be able to control the parasitaemia and anaemia (Osaer et al 1997) this was not the case in Djallonké ewes, indicating the immunodepressive effect of pregnancy and lactation in female trypanotolerant sheep.

Regarding their reproductive performance following infection, goats were clearly more affected than sheep. In the infected goats, time between infection and first kidding was significantly higher and kidding rates significantly lower compared to the control goats. This difference indicates a period of cessation of ovarian activity with lower conception rates or early embryonal death and resorption. A shorter inter-kidding interval was also observed which might be due to the higher number of abortions and stillbirths resulting in a quicker return to oestrus. However, kids born out of infected goats seemed to have an equal probability of survival and there was no evidence of affected milk production in the infected goats. Therefore, the significant lower average production of offspring per infected doe was mainly the result of fewer offspring. Similar clinical observations were made in goats following *T congolense* infection (Llewelyn et al 1987, Mutayoba et al 1988b). Bealby et al (1993) reported lower kidding rates in naturally infected goats and Ogaa et al (1991) found *T congolense* to be a cause of abortion in goats.

The sheep performed much better than the goats following infection in terms of reproduction. There were no significant differences between control and infected ewes in lambing rate and total offspring born. The average production at birth per dam was not different for the control and infected sheep. However, the lamb performance seemed to be worse in the infected group revealed by significant lower average production at one, three and five months in the first year. Regarding the two-year period, there was still lower average production at three and five months among the infected group. Since there were no differences in lambing rates between control and infected ewes, this could only be explained by higher lamb mortality and/or poorer lamb growth. Lamb mortality was indeed much higher in the infected group and this together with poor lamb growth could be attributed to lowered milk production in the infected ewes.

Body weight, which is a good indicator for trypanotolerance, increased throughout the experiment for all animals with no significant differences between treatments. Mawuena (1986, 1987) observed trypanotolerance in Djallonké sheep and described a positive weight gain in trypanosome infected sheep despite low PCV. Fertility of the animals was apparently not affected since they lambed normally and no abortion was observed. Djallonké ewes in this experiment seem to be resistant to *T congolense* infection since, except for the poorer lamb performance, no severe reproductive disorders were found compared to the control group. FAO (1980) compiled data of 18 countries in West and Central Africa for a productivity index for Djallonké sheep and WAD goats, raised under different conditions. This index, based on the total weight of five-month-old offspring produced per ewe or doe maintained per year ranged for Djallonké sheep between 9·4 kg and 18·7 kg and for WAD goats between 11·0 kg and 15·6 kg. Comparable values for this productivity index were found for the control groups of sheep and goats in the present experiment. This index was used to measure differences between the infected and control groups. The infected goats in year one were 89 per cent less productive than the controls. In the second year they were not able to recover completely and remained 52 per cent less productive. The infected sheep appeared to be 63 per cent less productive than the control sheep in one year. However, in year two

TABLE 6: Reproductive parameters for Djallonké sheep: least-square means ± SE over one year and two years for control and infected groups with significance

Trait	1 year Control (n = 11)	1 year Infected (n = 12)	Significance	2 years Control (n = 10)	2 years Infected (n = 12)	Significance
Average number of offspring	1·0±0·1	0·7±0·1	ns	2·7±0·2	2·7±0·2	ns
Average production at birth (kg)*	2·0±0·3	1·2±0·3	ns	6·1±0·5	5·2±0·5	ns
Average production at 1M (kg)*	5·9±0·6	2·8±0·6	**	14·6±1·4	11·6±1·2	ns
Average production at 3M (kg)*	9·5±1·2	4·3±1·1	**	23·6±2·2	16·9±1·9	**
Average production at 5M (kg)*	13·2±1·6	4·8±1·6	**	31·5±3·0	20·8±2·6	**
Time to first lambing (days)†	251·5±29·0	279·9±25·1	ns			
Average lambing interval (days)				253·6±21·0	231·2±18·1	ns

** P<0·01
*** P<0·001
* all observations per dam
† one single observation per dam

the effect of infection disappears since the infected group becomes more productive than the control group. Based on this productivity index, there is a clear reduction in productivity due to a *T congolense* infection.

CONCLUSION

Although trypanotolerance can be shown by positive weight gain and low mortality, an infection with trypanosomosis has a clear negative influence on reproduction in both Djallonké sheep and WAD goats. The Djallonké sheep seem to recover within one year from an artificial *T congolense* infection in terms of reproductive performance. In WAD goats the return to normal reproduction parameters takes at least two years.

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Evaluation of the semen quality and reproductive performance of trypanotolerant Djallonké rams following an artificial infection with *Trypanosoma congolense*

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Abstract

Following an experimental infection with *Trypanosoma congolense*, the semen quality and reproductive performance of 17 mature Djallonké rams was studied during a period of 26 weeks. The rams were randomly divided into three groups, namely one control group (G1) and two infected groups (G2 and G3). During the 5 weeks pre-infection the normal semen parameters were assessed. Following infection, moderate clinical symptoms associated with trypanosomosis were seen among the infected rams. Thirteen weeks post-infection, diminazene aceturate was administered to group 3. Both the treated group and the control group had significantly better weights than the infected group in the period following the trypanocidal treatment. The impact of trypanosomosis on the reproductive performance of the rams was seen in reduced libido with a higher rate of refusals and on semen quality with some temporary effects on mass motility, percentage live sperm cells and minor sperm cell abnormalities. However, these temporary changes in semen parameters were not significant when analysed as impact of infection. From the present study it was concluded that reproductive performance was not significantly impaired following artificial *T. congolense* infection. In addition, the Djallonké rams also showed an important clinical tolerance. However, significant differences between rams indicated a large variation in clinical response and reproductive performance following trypanosome infection. © 1997 Elsevier Science B.V.

Keywords: Djallonké rams; Semen quality; Trypanosomosis; Trypanotolerance; The Gambia

1. Introduction

Trypanosomosis has long been associated with reproductive disorders in man and animals (Apted, 1970, Ikede et al., 1988, Boersma et al., 1989, Kimato et al., 1994). Some published work exists on the pathology of infection by different trypanosomes (*T. brucei*, *T. congolense* and *T. vivax*) on the male reproductive system of small ruminants. Severe inflammation of testis and epididymis and testicular degeneration are described (Losos and Ikede, 1972, Isoun and Anosa, 1974, Ikede, 1979, Anosa and Isoun, 1980). The inflammatory changes in the genital organs due to the invasive character of the parasites in *T. brucei* infections are not found in infections with *T. vivax* and *T. congolense* where testicu-

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lar degeneration is more marked, mainly as the result of thrombosis in the spermatic blood vessels and prolonged fever. These testicular and epididymal lesions lead to drastic and progressive deterioration in semen quality revealed by cessation of sperm production, a decrease in motile sperm, a sharp increase in the percentage of dead sperm and a higher percentage of abnormal spermatozoa in rams (Isoun and Anosa, 1974, Anosa and Isoun, 1980, Agu et al., 1986, Akpavie et al., 1987, Sekoni, 1992).

Djallonké (West African Dwarf) sheep are the main breed found in The Gambia and are widely distributed in other West African countries. They are generally considered to be trypanotolerant because they survive and remain productive in tsetse-infested areas (Toure et al., 1981, Mawuena, 1987). However, there are suggestions that the mechanism of tolerance in small ruminants may differ from that in cattle. Trypanotolerance in cattle is related to an ability to control the level and duration of parasitaemia and to develop a less severe form of anaemia (Murray et al., 1982). There is no evidence that trypanotolerant sheep and goats resist in the same way. However, they do show an important degree of tolerance following experimental infection, which is reflected in very low mortality and an increase in body weights (Osaer et al., 1994). The response of Djallonké rams to heavy trypanosome infections in terms of their reproductive performance is not known. Some studies on West African Dwarf rams in Nigeria following *T. brucei* infections described severe lesions in the genital organs (Ikede, 1979) and following mixed infection with *T. brucei and T. congolense* a marked decrease of plasma testosterone levels (Adeyemo et al., 1989) was found. More research has been done in trypanotolerant cattle breeds. Despite their ability to control a trypanosome infection in terms of clinical outcome, the reproductive system and libido also seems to be affected (Djabakou et al., 1984, Grundler et al., 1988, Sekoni et al., 1988, Cloé et al., 1989, Bataille, 1990, Boly et al., 1991).

The present experiment was designed to examine the effects of an artificial *T. congolense* infection on the libido and semen characteristics in trypanotolerant Djallonké rams.

2. Materials and methods

2.1. Animals and husbandry

Seventeen Djallonké rams, all aged between 1 and 1.5 years, were purchased at the local market. They were all negative for trypanosomes on darkground (DG) buffy coat technique (Murray et al., 1977) and no trypanosomal antibodies were found using the immunofluorescence antibody test (IFAT; Katende et al., 1987). The 17 animals were randomly divided into three different treatment groups before the start of the experiment. Group 1 consisted of five rams and there were six rams in groups 2 and 3 respectively.

The three groups of rams were kept together on station at the coastal site of the International Trypanotolerance Centre (ITC). The climate is sub-humid with a mean annual rainfall of approximately 1000 mm. Since susceptible zebu cattle at ITC remained free of trypanosomosis for more than 5 years, it was concluded that tsetse challenge was effectively zero (Snow, pers. comm., 1992). No contact was allowed with female sheep except during days of semen collection. The animals received a supplement corresponding to 6.6 MJ and 9% Crude Protein to ensure a daily gain of 20 g. Before the start of the experiment they were vaccinated against Peste des Petits Ruminants (PPR-Tissue Culture Rinderpest vaccine, Hann lab., Dakar), dewormed (Panacur, Hoechst, Germany) and sprayed with acaricide (Gammatox, Coopers, UK).

2.2. Experimental design

2.2.1. Period 1: Assessment of semen parameters

In order to establish the normal semen parameters of Djallonké rams, semen was collected during a period of 5 weeks. Semen was collected every week with the artificial vagina using a teaser ewe in oestrus. Weights were recorded weekly and their Packed Cell Volume percentages (PCV%) were determined.

The ejaculate volume was read in graduated collection tubes (Chemineau and Cagnie, 1991). Mass motility (expressed in percentage) was assessed by microscopic examination, 40 × magnification, of an undiluted drop of ejaculate on a warmed stage (37–38°C) soon after semen collection. The percentage of live spermatozoa was determined in thin smears stained with eosine–nigrosine by counting 200 sperm cells. The concentration of spermatozoa was measured by a haemocytometer following a 1/400 dilution of the semen sample in formol–physiological saline (Chemineau and Cagnie, 1991). The abnormalities were classified into major and minor defects (Ott et al., 1987). Major defects included underdeveloped spermatozoa, pyriform and tapered heads, nuclear vacuoles, proximal cytoplasmic droplets, deformed midpiece and coiled tails. The minor defects were recorded as detached heads, abnormal and detached acrosomes, micro and macrocephalic forms, distal cytoplasmic droplets and abaxial implantation of midpiece. The scrotal circumference was measured using a flexible canvas tape. The libido was recorded as the number of seconds before the first mount.

2.2.2. *Period 2: Experimental infection*

In the second phase, two of the groups (groups 2 and 3, each with six rams) were infected with a *T. congolense* stabilate ITC84, while group 1 (five rams) served as an uninfected control. ITC84 is a cloned stabilate derived from a goat at Kunting on the north bank of the river Gambia (Central River Division, The Gambia). The pathogenicity of this stabilate has been assessed in trypanosusceptible Zebu cattle (Dwinger et al., 1992) and in trypanotolerant N'Dama cattle (Bennison, pers. comm., 1993). The 12 rams of groups 2 and 3 were inoculated intradermally at five sites on the left body flank with at least 10^4 bloodstream forms. Following infection, blood samples were taken daily for the first 2 weeks and thereafter weekly for 11 weeks. To assess the degree of anaemia PCV% was measured. The parasitaemia level was assessed by the dark ground (DG) method and the number of trypanosomes was scored by the method of Paris et al. (1982). The weights were recorded weekly. Rectal temperatures were taken daily in the early morning for 2 weeks following infection.

The weekly semen collection continued in the same way as in period 1 and the same parameters as described above were determined. In addition, clinical examination of the genital organs was performed to detect gross lesions like oedema, induration, alopecia of scrotal skin or atrophy of the testes.

2.2.3. *Period 3: Trypanocidal treatment*

Thirteen weeks post infection, one of the infected groups (group 3) was treated with a trypanocidal drug, diminazene aceturate (Berenil, Hoechst, Germany) at 7 mg/kg bodyweight by deep intramuscular injection. Bleeding and weighing procedures as well as determination of semen characteristics continued as in period 2. This period ended 26 weeks post infection.

2.3. *Statistical methods*

The various traits were analysed by unbalanced Analysis of Variance using a mixed model. The following main effects were included: treatment (control, infected, infected plus treated groups), period (pre infection, post infection and post treatment), ram (nested within treatment), week (nested within period) and the interaction effects treatment * period, ram * period and week * treatment. The effects ram, week, ram * period and week * treatment were regarded as random factors. All hypotheses were tested by the *F*-test, where the approximation of Satterthwaite (Snedecor and Cochran, 1980) had to be used.

The interaction between treatment and period was of greater interest than then the main effect of treatment alone. Considering the design of the experiment in the first period there should be no difference between the groups, in the second period there should be no difference between group 2 and 3. With the interaction, four contrasts were defined. Contrast 1 expresses the difference between control rams and infected rams for the changes in period 2 compared to period 1 (this measures the impact of infection). Contrast 2 expresses the difference in infected rams between treated and untreated, with respect to the difference between the last period and the average of the first two periods (this measures the impact of trypanocidal treatment). Contrasts 3 and 4 measure the differences between control rams and infected rams and treated rams respectively with respect to the difference between periods 3 and 2.

3. Results

3.1. Reference parameters

The normal semen parameters as assessed in period 1 are 0.74 ± 0.02 ml (least square mean $\pm$ standard error) for volume, $3.8 \pm 0.09 \times 10^9$/ml for concentration, $81.4 \pm 1.2\%$ for mass motility and $73.6 \pm 1.3\%$ for live sperm cells. Major and minor abnormalities were 3.1 ± 0.3 and $2.1 \pm 0.8\%$ respectively.

3.2. Clinical parameters

Following the *T. congolense* infection (period 2), there were no obvious clinical signs due to trypanosomosis among the infected rams. One ram (No. 4) died suddenly at 7 weeks post-infection (p.i.) of unknown causes. Palpation of the genital organs did not reveal any abnormalities within the different groups.

The mean rectal temperatures are presented in Fig. 1. Although seven out of 12 rams had temperatures above 40°C on one or more occasions during the 2 weeks p.i., the mean rectal temperature of the infected groups never rose above 40°C. The highest mean rectal temperature was reached at day 8 p.i. for both infected groups with averages of 39.39°C and 39.81°C for groups 2 and 3 respectively. There were no significant differences between the groups during the 2 weeks p.i. All the rams from groups 2 and 3 became parasitaemic between 4 and 7 days p.i. The first peak for both groups was reached at day 12 p.i. with mean scores of 5.0 and 4.8 for groups 2 and 3 respectively (see Fig. 2). As expected the overall mean score was not different for the groups 2 and 3 during time of infection.

The PCV values for the groups in different periods are presented in Table 1 as least square means. Group 2 and 3 had a drop following infection from 23.7 to 21.9 and from 26.3 to 23.5 respectively and recovered both during period 3 with group 3 almost reaching the pre-infection value. Despite these drops in PCV levels, no significant differences were found for the interaction treatment ~ period.

The weight changes are presented in Table 1. During period 3 there was a marked decline from 28.8 ± 2.1 kg to 26.9 ± 2.1 kg for the infected rams (group 2). The interaction effect treatment ~ period was significant, as well as contrast 2 and 3 ($P <$

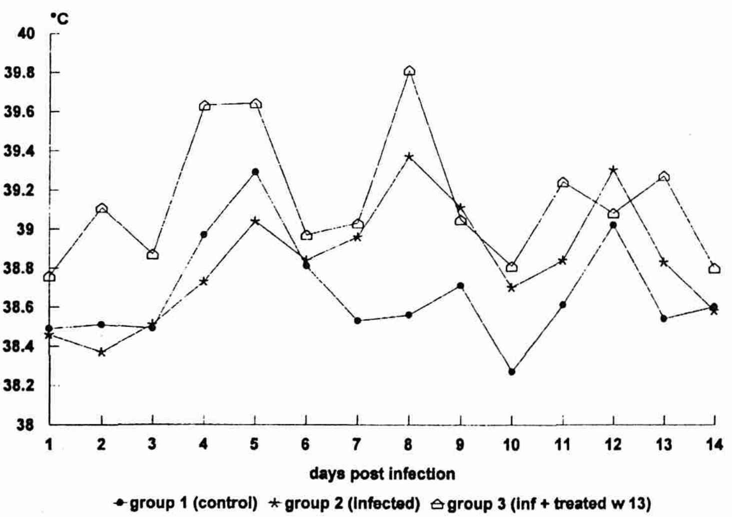

Fig. 1. Mean rectal temperature (°C) during 14 days p.i.

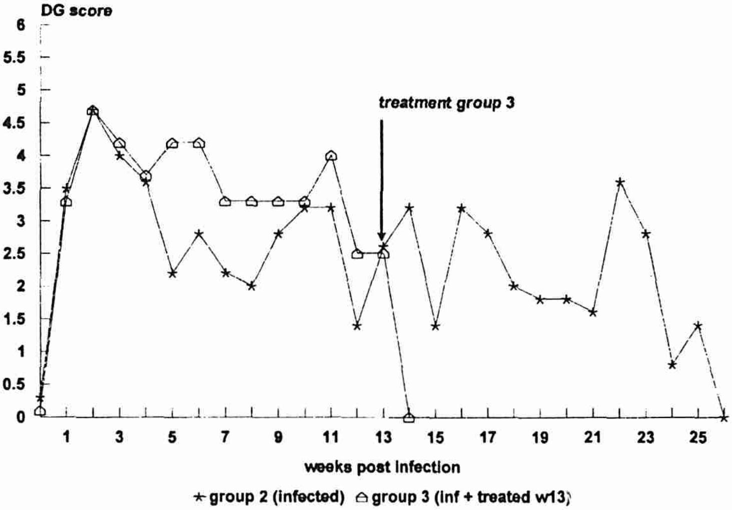

Fig. 2. Mean weekly parasitaemia-score (darkground).

0.05). These significant contrasts indicate that the infected/treated rams and the control rams had better body weights compared to the infected/not treated ones.

Differences ($P < 0.001$) between rams were found in rectal temperature, parasitaemia score, PCV% and body weight.

3.3. Semen characteristics

The total sperm output, determined by the volume and concentration of the ejaculate, is presented in Table 1. The total sperm output did not seem to be affected following infection. Contrast 2 revealed differences in total sperm output ($P < 0.05$) indicating a higher sperm output for the treated group compared to the infected group in period 3. Nevertheless, contrast 1 which measures the impact of the infection did not show any difference between control and infected rams.

The weekly mean values for mass motility are plotted in Fig. 3. Fluctuations were seen for all three groups, but marked drops for group 2 appeared at week 1, between week 9 and 15 and at week 24 p.i.,

Table 1
Averages of Packed Cell Volume % – Bodyweight – Sperm output: least square means (± SEM)

Treatment group	Packed cell volume (%)			Bodyweight (kg)			Sperm output [1]		
	Period I	Period II	Period III	Period I	Period II	Period III	Period I	Period II	Period III
1	25.8 (±1.2)	25.0 (±1.1)	25.7 (±1.1)	25.7 (±2.2)	29.7 (±2.1)	29.6 (±2.1)	1.5 (±0.2)	1.4 (±0.1)	1.6 (±0.1)
2	23.7 (±1.2)	21.9 (±1.1)	22.4 (±1.1)	25.6 (±2.2)	28.8 (±2.1)	26.9 (±2.1)	1.8 (±0.1)	1.7 (±0.1)	1.7 (±0.1)
3	26.3 (±1.1)	23.5 (±1.0)	25.6 (±1.0)	26.5 (±2.0)	30.9 (±1.9)	30.0 (±1.9)	1.6 (±0.1)	1.7 (±0.1)	1.9 (±0.1)

[1] Figures are based on square root transformed observations; to get the real sperm output, the figures have to be squared and multiplied by 10^9.

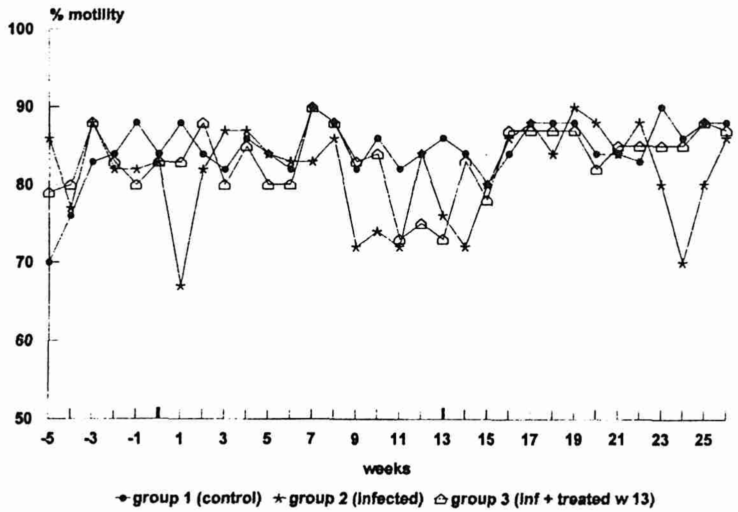

Fig. 3. Mean weekly mass sperm motility (in percentages).

whereas group 3 had a decline between week 10 and 14. Despite these temporary drops in motility following infection, there were no differences between treatment ~ period combinations. An overall impact of infection and/or treatment was not present.

There was a profound decline in percentage live sperm cells for both infected groups at week 13, from which both groups recovered quickly (see Fig. 4). Group 2 had a second decline at week 24 to an average of 63.2% live spermatozoa. Despite these

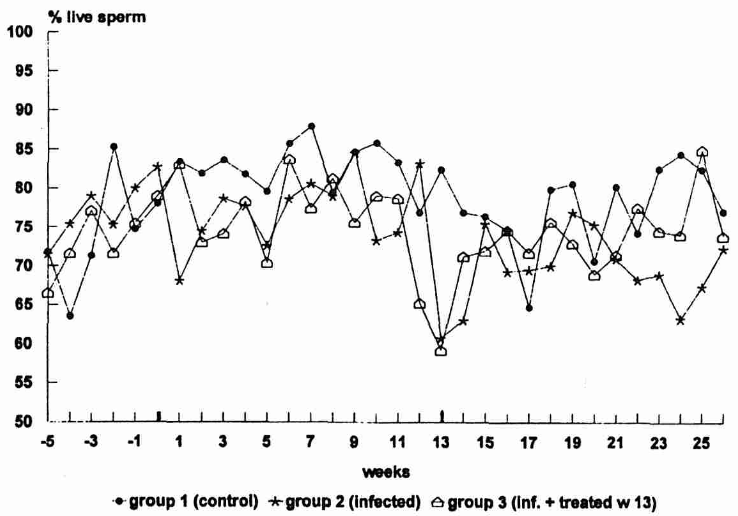

Fig. 4. Mean weekly live sperm cells (in percentages).

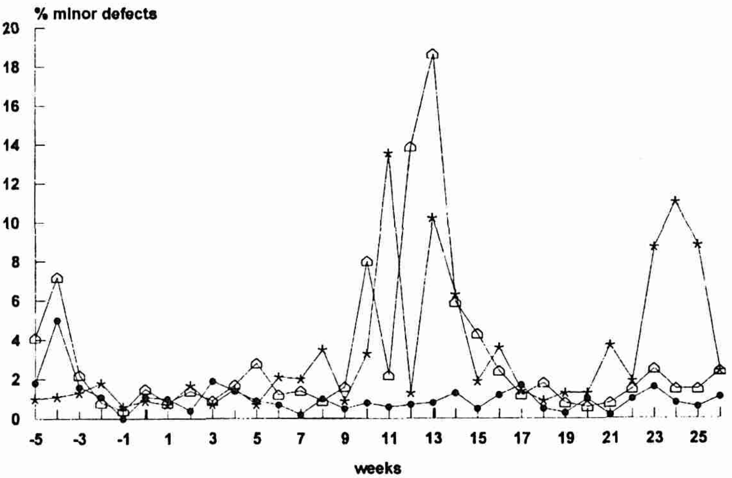

Fig. 5. Mean weekly minor sperm cell defects (in percentages).

drops, the interaction effect treatment ~ period was not significant. However, contrast 1 revealed a higher % live sperm cells for the control rams in period 2 compared to the infected rams ($P < 0.05$).

Fig. 5 shows the weekly mean percentages for minor sperm cell defects in each group. Between week 10 and 14 p.i., there was a general increase in minor defects for both infected groups, followed by a second peak at week 24 p.i. for group 2. These peaks were transient and due to a few individual high values, with a maximum value of 93.5% in group 3 and 38.0% in group 2. The major sperm cell defects showed fluctuations for all groups throughout with a magnitude not more than 5.1% on average. For both traits the interaction effect period ~ treatment was not significant.

The mean scrotal circumference did not show noticeable changes following infection. In general, all the semen characteristics revealed important ram differences ($P < 0.05$).

3.4. Libido

For all groups, there was an overall increase in reaction time from week 6 p.i. towards the end of the experiment, although the weekly values for all groups had important fluctuations during period 2 and 3. The interaction treatment ~ period was not significant for reaction time but there were differences between rams ($P < 0.001$). However, the rate of mounting failures was highest in the infected rams (5 ×), compared to infected/treated rams (2 ×) and control rams (1 ×).

4. Discussion

Following infection, the Djallonké rams showed moderate clinical signs of trypanosomosis in terms of anaemia, pyrexia and total weight changes. Mortality due to trypanosome infection was virtually zero. Pyrexia did not occur in all the infected animals during the 2 weeks p.i. and there was no significant difference between the groups. Despite the drop in PCV levels following infection for groups 2 and 3 no significant differences to the control group were found. Both control group and treated group had significantly better body weights than the infected group in period 3.

The significant differences between rams for weight changes, PCV levels, parasitaemia score and rectal temperature may indicate an individual variation in clinical response to trypanosomosis. The ability to control the disease in terms of minimising the level of trypanosomes as it is occurring in trypanotolerant N'Dama cattle was not seen as such (Dwinger et al., 1992). Group 2 remained parasitologically positive until the end of the experiment (26 weeks p.i.). During a previous experiment on female Djallonké sheep following infection with the same stabilate, animals remained positive for trypanosomes on DG until 24 months p.i., without showing severe clinical signs and having positive weight gains. (Osaer and Goossens, pers. comms.).

The impact of trypanosomosis on the reproductive performance of the rams was not evident in all the semen characteristics. The total sperm output was not clearly affected by infection. The mass motility showed some temporary drops following infection although no significant differences were found. The negative effects on percentage live sperm cells were of short duration and there were no differences between the groups except for contrast 1. However, the latter was not very likely an effect of trypanosomosis since there was no decline in the infected groups. The increases in minor abnormalities following infection were of transient nature and return to normal values was remarkably quick, taken into account the duration of a spermatogenic cycle which is about 7 weeks. It is generally assumed that total abnormal sperm morphology exceeding 20% is associated with lowered fertility. The rams which had very high values for sperm cell defects following infection could have been regarded as temporarily unsuitable for breeding. Significant ram differences for all these reproduction parameters indicated that some rams suffered more from infection than others.

The libido, measured as first mount reaction time, was not clearly affected by the infection. However, when considering the rate of mounting failures, the impact of infection on libido was more obvious with the highest rate found in the infected group. This discrepancy could be due to the fact that reaction time is also influenced by other factors.

In *T. congolense* infections in small ruminants testicular degeneration (Kaaya and Oduor-Okelo, 1980, Anosa and Isoun, 1980) and reduction in plasma testosterone levels (Adeyemo et al., 1989, Mutayoba et al., 1993) have been described. According to the severity of testicular degeneration, one can find increased numbers of pathological heads and proximal droplets associated with poor motility in mild cases, whereas in more severe cases poorly concentrated semen, low motility and condensed proximal droplets are found (Wenkoff, 1988). The main defects observed in this study were detached heads, proximal and distal cytoplasmic droplets and coiled tails, which occur during maturation of the spermatozoa (Barth and Oko, 1989). There was a very low incidence of deformed midpieces, nuclear vacuoles and detached acrosomes. Because of the rapid disappearance of the defects, it is very unlikely that the seminiferous epithelium was severely affected, although no further histological examination could be performed. Plasma testosterone was not measured but it can be assumed that the level declined and therefore partly caused a depression of libido in the infected rams, together with clinical factors such as the onset of anaemia, parasitaemia and pyrexia.

5. Conclusions

The impact of the *T. congolense* infection on the clinical and reproductive performance of the Djallonké rams was modest. It was only evident in negative weight changes, a higher rate of mounting refusals and on semen quality with some transient, but no significant effects on mass motility, percentage live sperm cells and minor abnormalities. The positive effect of treatment was seen in better weight changes.

Although these are not the criteria of the trypanotolerant trait, the large individual differences in clinical and reproductive response found in this study could be used to rank for the trypanotolerant quality of Djallonké rams, based on both clinical tolerance and reproductive performance following trypanosome infection.

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CHAPTER 4

INTERACTION OF NUTRITION AND TRYPANOSOMOSIS ON PRODUCTIVITY OF TRYPANOTOLERANT DJALLONKE SHEEP.

4.1

Effects of *Trypanosoma congolense* infection and nutritional supplementation on establishment and outcome of pregnancy in trypanotolerant Djallonke Ewes.

Osaer, S., Goossens, B., Jeffcoate, I., Jaitner J., Kora S. and Holmes, P. 1998.

Animal Reproduction Science, 51: 97-109

4.2

Effects of *Trypanosoma congolense* and nutritional supplements in Djallonke ewes on live weight during pregnancy, post partum weight, haematology parameters and lamb performance.

Osaer, S., Goossens, B., Jeffcoate, I., and Holmes, P. 1998.

Research in Veterinary Science, 65: 65-69.

4.3

Effects of *Trypanosoma congolense* infection and diet on puberty, age at first lambing and haematology changes in Djallonke ewe lambs.

Osaer S., Goossens, B., Kora S. and Jeffcoate, I. 1999.

Veterinary Parasitology, 80: 215-230.

4.4

Biochemical changes following experimental *Trypanosoma congolense* in young Djallonke ewes kept on two planes of nutrition.

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Effects of *Trypanosoma congolense* and nutritional supplements on establishment and outcome of pregnancy in trypanotolerant Djallonké ewes

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Abstract

Interactions between *Trypanosoma congolense* and nutritional status were studied in 42 ewes, bred at the peak of parasitaemia after synchronisation of oestrus. As experimental design a randomised block design was used with four treatment combinations (2 × 2 factors), of which two were on a restricted diet (L), the remainder on an unrestricted diet (H) and half of each nutritional group infected with *T. congolense* (LI, HI), the remainder serving as controls (LC, HC). Severity of parasitaemia was not influenced by supplementation and mortality rates were higher in the HI and LC groups, but these differences were found not significant. Progesterone levels during the synchronised cycle were significantly lower in the infected groups. Levels of pregnant specific protein B (PSPB) in pregnant sheep at days 21 and 26 were not significantly affected by nutrition or infection, despite the tendency of a decrease in infected groups. *T. congolense* clearly affected establishment of pregnancy, as shown by lower rates of pregnancy and extended intervals between breeding and confirmation of pregnancy, nor was there any benefit of nutritional supplementation. Mean progesterone concentration during pregnancy, in those ewes which lambed, was not different between groups. The effect of the *T. congolense* infection on the outcome of pregnancy was not clear with the LI and HC performing well and poor pregnancy outcomes in groups HI and LC, although differences in litter size might explain these anomalies. It is concluded that the most pronounced effect of *T. congolense* was a negative influence on establishment of pregnancy, with nutritional supplementation unable to overcome this effect but having a beneficial influence on maintenance and successful outcome of pregnancy. However, individual exceptions indicate that

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some ewes cope better with the negative effects of infection and poor nutrition.

Keywords: Sheep—pregnancy; *Trypanosoma congolense*; Feeding and nutrition

1. Introduction

The development and exploitation of trypanotolerance is an important consideration before improvements in livestock productivity can be achieved in tsetse infested areas. Djallonke sheep, indigenous to West Africa and the main sheep breed in the Gambia, are considered trypanotolerant (Mawuena, 1986, 1987; Osaer et al., 1994) but even so, common situations such as pregnancy and lactation, concurrent disease and malnutrition tend to reduce trypanotolerance (Murray et al., 1982; Agyemang et al., 1990, 1992; Little et al., 1990). West African Dwarf goats seem to suffer reduced kidding percentages following *Trypanosoma congolense* infection (Kanyari et al., 1986). Workers who have studied the effects of trypanosomosis as a single factor in pregnant small ruminants have observed such reproductive disorders as abortions, and stillbirths or neonatal deaths (Ikede and Losos, 1972; Ikede et al., 1988; Sekoni, 1994; Llewelyn et al., 1987; Elhassan et al., 1989; Edeghere et al., 1992). Although *T. congolense* is considered as strictly intra vascular, it has been reported to be able to cross the placental barrier and cause abortion in East African goats (Ogaa et al., 1991). In the nonpregnant animal, *T. vivax* infection resulted in anoestrus in Djallonke ewes, varying from 40 to 96 days in duration and characterised by prolonged low levels of progesterone (Elhassan et al., 1994). In goats, *T.congolense* infection results in anoestrus due to persistent corpora lutea (Llewelyn et al., 1987). Present evidence therefore seems to suggest that trypanosome infection can reduce ewe productivity by acting directly on the pregnant uterus and also by disrupting cyclicity and preventing a return to oestrus. Dietary conditions are important in these effects, so both energy-restricted and protein-restricted diets can aggravate the detrimental effects of *T. congolense* infection on growth and productivity in sheep (Katunguka et al., 1993, 1995), while dietary supplements are beneficial in young Djallonke sheep by reducing mortality rates, delaying onset of trypanosomosis and reducing live-weight losses (Hecker et al., 1994). Nutritional supplements also have a beneficial effect in pregnancy in trypanosome-infected dams including reduced ewe and perinatal lamb mortality (Reynolds and Ekwuruke, 1988).

The present experiments were therefore designed to study the effects of *T. congolense* and nutritional status on the endocrinology of the conception cycle, conception rates and outcome of pregnancy in Djallonke ewes. To this end, infected animals were maintained on one of two planes of nutrition to see whether supplementation can alleviate the effects of trypanosomosis on reproductive performance.

2. Materials and methods

2.1. Animals and husbandry

Forty-two nonpregnant ewes were selected from the ITC breeding flock. All were routinely treated with anthelmintic (Panacur, Hoechst, Germany) and vaccinated against peste des petits ruminants (Tissue Culture Rinderpest vaccine, Hann Laboratory, Dakar, Senegal), and clostridial infections (Covexin, (Pittman Moore), Mallinckdrodt, Uxbridge, UK). All ewes were kept confined at the coastal site of the ITC. The climate is subhumid with a mean annual rainfall of approximately 1000 mm. Since susceptible zebu cattle at ITC have remained free of trypanosomosis for more than five years, it has been concluded that there is no risk of tsetse challenge at this site (W. Snow, personal communication). For the experiment a randomised complete block design was essentially used. Ewes were allocated to blocks based on their live weight at the start of the experiment. Within blocks, ewes were randomly allocated into a 2 × 2 factorial design resulting in four treatment combinations: high level of nutrition and infected (HI), high level of nutrition and not infected (HC), low level of nutrition and infected (LI) and low level of nutrition and not infected (LC). High level of nutrition comprised per ewe 120 g crude protein (CP) and 9 MJ/day whereas the low diet offered 90 g CP and 7 MJ per animal per day. All ewes were placed on the appropriate diet, consisting of ground nut hay and concentrate, eight weeks from the start of the experiment having been previously fed a basic supplement. Prior to trypanosome infection, all animals were confirmed free of trypanosomosis using the dark ground (DG) buffy coat technique (Murray et al., 1977). Some of the animals had been used in a previous on-station experiment, where the same *T. congolense* strain was used but all had been treated with diminazene aceturate (Berenil, 7 mg/kg live weight, Hoechst, Germany). However, those animals had to be withdrawn (see results) from the experiment since boostered immune responses in the ewes prevented the development of parasitaemia following the trypanosome infection in the present study.

2.2. *Breeding and trypanosome infection*

Oestrus was synchronised by 12-day treatment with an intravaginal progestagen-impregnated sponge, placed at day −12 (Veramix, Upjohn, Crawley, UK), followed by an injection of serum gonadotrophin upon sponge withdrawal (on day 0) (PMSG-Intervet, 500 IU/ewe, Intervet, Cambridge, UK). The trypanosome infection took place during progestagen treatment, at day −10, in such a way that the peak of parasitaemia would coincide with oestrus, which was expected within 48 to 72 h following sponge withdrawal (day 0). This time prediction to reach peak parasitaemia was known from previous experiments using the same strain (Osaer et al., 1994, 1997; Goossens et al., 1997). Following this method it is anticipated that the trypanosome infection interferes with establishment of pregnancy. During expected oestrus, one ram was introduced into each of the four groups and swapped around twice daily to ensure optimum mating activity. Rams were fitted with a colour marking harness to help confirm mating times. The rams remained with the ewes for a period of 3 months.

The ewes allocated to the groups LI and HI were inoculated with a stabilate of the *T. congolense* strain ITC84, which is a cloned stabilate of *T. congolense* derived from a goat at Kunting on the north bank of the River Gambia (Central River Division, The Gambia). The pathogenicity of this stabilate has been assessed in trypanosusceptible Zebu cattle (Dwinger et al., 1992), in trypanotolerant N'Dama cattle (J. Bennison, personal communication) and in West African Dwarf goats and Djallonke sheep (Osaer et al., 1994, 1997; Goossens et al., 1997). It was expanded in mice and the ewes were inoculated intradermally with at least 10^4 bloodstream forms at five sites on the left flank.

2.3. Blood sampling and analysis

Blood samples were taken on the eleventh day post infection, i.e. the day of expected oestrus, and thereafter at weekly intervals to assess the degree of anaemia (PCV %) and the parasitaemia level by the dark ground (DG) method. The number of trypanosomes was scored by the method of Paris et al. (1982). In addition, plasma was collected on day 12, day 17 and day 19 post oestrus and stored from the weekly blood sampling during the first 3 oestrus cycles, thereafter at three weekly intervals, for measurement of progesterone concentration. Additional plasma samples were collected on days 21 and 26 after synchronised breeding for measurement of pregnancy specific protein B (PSPB). Plasma PSPB concentration was measured by a heterologous double antibody radioimmunoassay using procedures described for sheep by Willard et al. (1987). This assay employed a rabbit anti PSPB antiserum (RGS 41-5) and bovine PSPB (R37) for both iodination and standard as described by Humblot et al. (1990) at a similar stage of pregnancy. Assay sensitivity was 0.2 ng/ml and inter- and intraassay coefficients of variation were 15% and 10% respectively. Plasma progesterone concentration was measured using a double antibody enzyme immunoassay as described by Meyer and Güven (1986) and Prakash et al. (1987) but with modifications in the form of rat anti-progesterone monoclonal antibody (Sigma Clone 2H4), anti-rat coating antibody (Sigma, R5130) and peroxidase labelled progesterone label (Sigma P3659). The assay standard curve was prepared in plasma from a castrated ram and ranged from 10–0.31 ng/ml. The sensitivity of this test was determined to be 0.59 ng/ml ($\sigma = 0.19$, $n = 21$). Four control values with known values of 0.5, 1.0, 3.5 and 5.0 ng/ml were included in each assay. The interassay coefficients of variation for the four control values were 16.1%, 18.3%, 16.4% and 9.2% respectively. The intraassay coefficients of variation for the 4 controls were 7.1%, 5.2%, 6.9% and 8.1% respectively. Conception rates were assessed based on a progesterone level > 1.5 ng/ml at days 17 and 21 post oestrus and detectable PSPB on days 21 and/or 26, the latter only measured following the first induced oestrus.

2.4. Statistical analyses

Statistical analysis was carried out using SAS® statistical package version 6.11 (1996) (Statistical Analysis Systems Institute SAS, 1989–1996).

Effects of treatments on mortality rate, conception rate, pregnancy outcome and litter size were tested using the Chi-square test. Continuous variables were analysed by a linear mixed model. The following main effects were included wherever appropriate: infection (control vs. infected), diet (low vs. high), block, date of sampling, animal (nested within infection, diet and block), lambing month and the interaction between infection and diet. The animal effect was regarded as random. All hypothesis of continuous variables were tested by the F-test (Snedecor and Cochran, 1980). Results were regarded as statistically significant when the type I error probability was smaller than 5%. Means of the different treatments are presented as least square means ± standard error (i.e. mean ± S.E.).

3. Results.

3.1. Animal breeding and parasitaemia.

Following oestrus synchronisation, all ewes were mated within 3 days except for one ewe in group LI which was not served until 17 days later. Several ewes returned to oestrus repeatedly during subsequent cycles but accurate rebreeding records are not available as much of the colour marking by the rams was deemed to be due to excessive zeal and the close confines of the pens. Mortality rates during the observation period were not significantly different between groups. Three ewes from group HI died, two as a result of trypanosomosis in the third and fourth week post infection and one died from cowdriosis in week 22 and was found pregnant in an early stage. The ewe which died in the third week was excluded from further analyses since no sufficient data were available. One ewe died in each of groups LI and HC, the former as a result of trypanosomosis in week 12 and the latter from tetanus shortly after parturition. Four ewes in the LC group died before their predicted delivery respectively in week 11, 13, 15, and 18 of pregnancy, the latter three probably died from undernutrition compounded by the presence of multiple pregnancies.

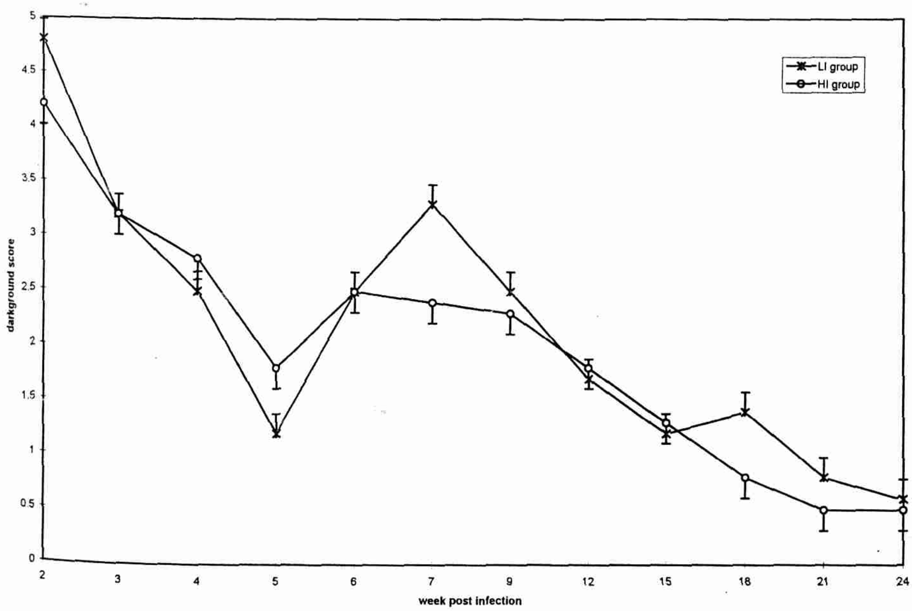

Fig. 1. Mean parasitaemia level (dark ground score) during 24 weeks post infection in both infection groups LI and HI.

At no point in the experiment were trypanosomes detected in the control ewes. Trypanosomes were never detected in five ewes from group LI and four from group HI, animals which had been infected with the same strain in the previous experiment and consequently these animals were omitted from further study. Trypanosomes were detectable by the eleventh day post infection in the remaining ewes, with parasitaemia scores being maximal by the second week post infection and remaining detectable until the end of the experiment, 24 weeks post infection as shown in Fig. 1. Dietary level had no effect on parasitaemia score ($P > 0.05$).

3.2. PSPB and progesterone during first cycle

Mean plasma progesterone increased in all groups after synchronised oestrus (Fig. 2), but was lower in both infected groups. Between days 1–12 after oestrus, a significant negative effect of infection on plasma progesterone concentration was found, with the estimated increase in plasma progesterone between days 1 and 7 being on average 4.3 ± 1.7 ng/ml higher in control vs. infected ewes (infection × cycle-stage; $P < 0.01$). Neither diet nor the interaction diet × infection had a significant influence on luteal progesterone production during the first cycle.

Of those ewes which conceived successfully after the synchronised oestrus, plasma concentrations of PSPB at day 21 and day 26 post breeding revealed no significant differences between the four treatments (see Fig. 2), but did increase significantly in all

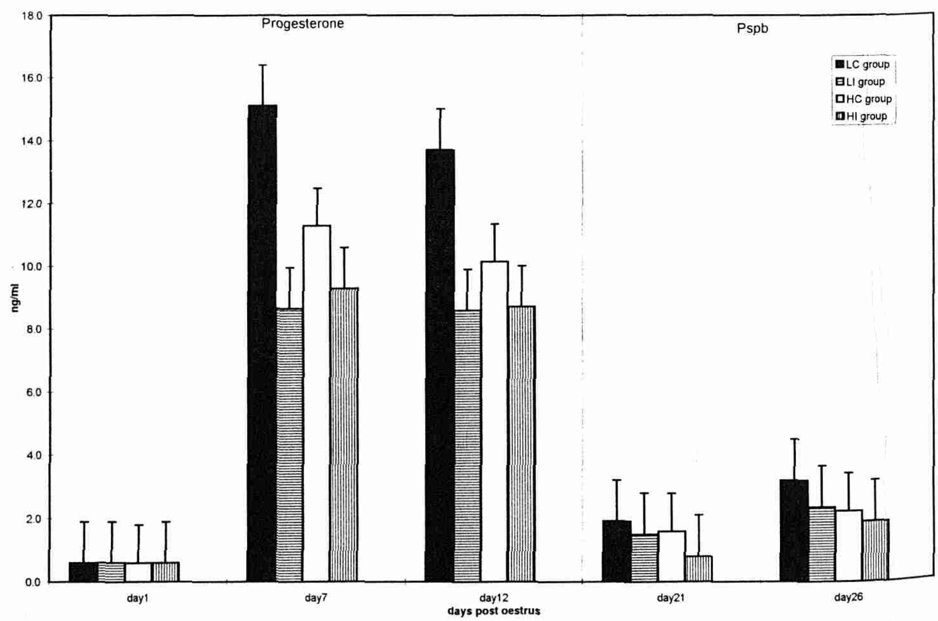

Fig. 2. Plasma progesterone concentration on days 1, 7 and 12 of first synchronised cycle and PSPB concentration at day 21 and day 26 post breeding in groups Low Control (LC), Low Infected (LI), High Control (HC) and High Infected (HI).

ewes between day 21 (1.4 ± 0.4 ng/ml) and day 26 (2.4 ± 0.3 ng/ml). There was also a tendency for plasma PSPB to be lower in trypanosome infected ewes than controls (1.5 ± 0.4 and 2.3 ± 0.2 ng/ml respectively).

3.3. Conception rates and time to conceive.

Conception rates after the first synchronised oestrus were based on detectable PSPB on days 21 and/or 26, and plasma progesterone concentration > 1.5 ng/ml on days 17 and 21. Conception rates after two or more oestrus cycles (repeat breeders) were similarly based on progesterone levels. Overall conception rates per group are presented in Table 1 as the proportion of the number of ewes that were exposed to rams. Trypanosome infection influenced negatively conception rates ($P < 0.05$). In addition, a difference between the four groups ($P < 0.01$) indicated that the HI group was performing worse than both control groups, but the LI was not different to the control groups neither to the HI group. Between level of diets there was no difference in conception rates. PSPB was not detected in three ewes which turned out to be repeat breeders, one in each of the LI, HI and HC groups, whereas plasma progesterone concentration had been elevated on day 17. Amongst the repeat breeders in the control groups (LC, HC), all had conceived by the second cycle; whereas in the infected groups (LI, HI) all conceived much later (two conceived after three cycles, one after six cycles and one after seven cycles). In each of the trypanosome-infected groups, one ewe entered a long period of anoestrus after the synchronised cycle as shown by persistently basal plasma progesterone. The time needed to conceive was reflecting the presence of these repeat breeders within the different groups, with mean intervals between induced oestrus and conception for respectively LC, LI, HC and HI groups of 29.8 ± 17.2 days, 58.2 ± 19.1 days, 0.6 ± 11.5 days and 98.6 ± 48.9 days. Trypanosome infection significantly increased the time to conceive ($P < 0.05$), the overall means for control and infected sheep being 15.2 ± 10.0 and 78.4 ± 25.0 days. Both dietary level and interaction

Table 1
Number of ewes per group at start, number of excluded ewes, group conception rates and percentage, lambing outcome and percentage of the four treatment groups

Treatment	No. of ewes at start	No. of ewes excluded[1]	Conception rates		Lambing outcome	
			No. of ewes conceived after one or multiple cycles/ no. of ewes exposed to rams	(%)	No. of ewes lambing/ no. of ewes alive at predicted lambing date	(%)
LC	11	0	11/11	100[a]	5/7	71.4[a]
LI	11	5*	5/6	83.3[ab]	5/5	100[a]
HC	10	0	10/10	100[a]	10/10	100[a]
HI	10	4* + 1**	3/5	60[b]	0/2	0[b]

Different superscripts within a column indicate a significant difference ($P < 0.01$).
[1]Ewes per group that were excluded due to their non parasitaemic status* or early death**.
LC, Low Control; LI, Low Infected; HC, High Control; and HI, High Infected.

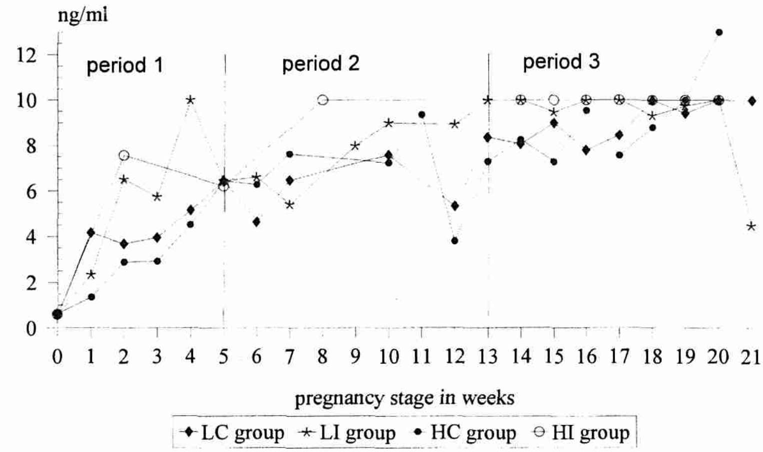

Fig. 3. Plasma progesterone concentrations during pregnancy in groups Low Control (LC), Low Infected (LI), High Control (HC) and High Infected (HI).

diet * infection had no significant effect on intervals from synchronised oestrus to conception.

3.4. *Progesterone during pregnancy*

Plasma progesterone concentrations during pregnancy in each of the four groups are shown in Fig. 3. The results were split into three periods for analysis; firstly, the period of linear increase in progesterone ($Y = b_o + b \times$ sampling day + error) up to week 5, secondly, weeks 6–13 and thirdly, weeks 13 to 21, analysed as repeated measurements of progesterone. Apart from individual variations ($P < 0.05$), neither infection nor diet nor their interactions influenced the level of progesterone during different stages of pregnancy. General means (σ) were 0.13 (0.19), 7.1 ng/ml (2.0) and 9.2 ng/ml (1.2) respectively for the daily increase in progesterone during the first five weeks of pregnancy and the averages during weeks 6 to 13 and weeks 13 to 21.

3.5. *Pregnancy outcome.*

Lambing rate, defined as the ratio of ewes which delivered at the predicted date to those that conceived and stayed alive are shown in Table 1. Of those ewes which died before they could deliver several were found pregnant (four in LC, one in HI). Of the ewes which failed to lamb at predicted date, none aborted but, some ewes became empty (one in LC, one in HI) but died at the end of the experiment, and others lambed but much later than expected (one in LC, one in HI), thus all four ewes experienced early foetal death. There was a significant difference in pregnancy outcome between groups

($P < 0.001$) with lowest rate in group HI, having significantly lower lambing outcome than the other groups.

4. Discussion.

In the present study it was shown that *T. congolense* had a detrimental effect on the establishment of pregnancy in Djallonke ewes, as shown by extended intervals from ram introduction to confirmation of pregnancy. Thus, the noninfected repeat breeders returned to oestrus once whereas infected ewes required the equivalent of from three to seven oestrous cycles before conceiving. Moreover, there was no improvement in conception rates with increased energy and protein intake.

Comparison of the progesterone profiles obtained from infected and control sheep during the first cycle showed a lower progesterone level in the infected groups. Progesterone appears to be one of the major factors in establishment of pregnancy and one of its functions is to synchronise development of the maternal endometrium with arrival of the embryo in the uterus (Wilmut and Sales, 1981; Lawson and Cahill, 1983). Wilmut et al. (1985) have demonstrated an association between progesterone profiles and embryo survival in ewes. Growth of the sheep embryo during the first few weeks of pregnancy is influenced by the concentration of progesterone in the maternal plasma (Lawson, 1977), so a certain minimum level of progesterone is essential for maintenance of pregnancy (Trounson and Moore, 1974; Parr et al., 1982). It therefore seems likely that low luteal-phase progesterone levels contributed to the poor pregnancy rates in the infected ewes. Indication of early foetal death was seen in ewes which either died empty or lambed later than predicted from their P4 and PSPB levels. The four ewes which experienced early foetal death may also have had a decrease in plasma progesterone levels leading to termination of pregnancy and three of them, two from the HI group, were showing transient decreases in plasma progesterone profiles around day 30 of gestation. However, since foetal loss mostly occurred after day 50 of gestation, when blood samples were only being collected once weekly, it was not possible to confirm a cause and effect relationship between circulating progesterone and loss of pregnancy. In a previous study, Mani et al. (1995) showed that undernutrition did not affect circulating progesterone level in goats and in the present experiment, conception rates were not altered by level of nutrition, so lower progesterone levels and poor pregnancy rates seem unlikely to be simply related to level of nutrition but more to do with trypanosome infection.

There was also evidence of early embryonic death in some trypanosome-infected ewes (one from group LI and one from HI) and also in one control ewe, since plasma PSPB concentration was undetectable on days 21 and 26 but high plasma progesterone had been measured on the seventeenth day after breeding.

The effect of the *T. congolense* infection on the outcome of pregnancy was not clear. Thus, despite the poor performance in the HI group, the LI group did very well compared to group HI. A positive effect of nutrition on lambing outcome was more apparent in the control groups. In the LC group, two ewes experienced early fetal death and four died carrying multiple pregnancies. These ewes appeared to be anorexic despite the extra ME allowance for pregnancy and while feed refusals were never measured, the combination of anorexia and multiple pregnancy resulted in poor pregnancy maintenance and fetal survival until term.

The effects of infection and supplementation seemed to be equally important in the maintenance of pregnancy in the present study. Experiments to ascertain the effects of feed restriction during gestation in ewes have shown an inverse relationship between plane of nutrition and plasma progesterone concentration (Parr et al., 1982; Wilmut et al., 1985). In the present experiment, high nutrition did not cause lower progesterone levels in those ewes who completed pregnancy, since there was no difference between groups, however, it might have lowered the level of progesterone before termination of pregnancy in the HI group. This was certainly not the case in the control groups. Since progesterone seems to be a key factor in establishment and maintenance of pregnancy, it is clear that any impairment of luteal and/or placental progesterone production would have serious implications for pregnancy. Previous studies to evaluate the effects of another haematic type trypanosome, *T. vivax*, on the oestrous cycle and fertility of Djallonke ewes revealed extended periods of anoestrus and extended luteal function with reduced plasma progesterone levels. However, those ewes which survived all became pregnant and had normal gestations (Elhassan et al., 1994). In East African Dwarf goats, reduced kidding rates after naturally acquired trypanosome infections have been interpreted as a result of trypanosome effects on oestrus and conception as opposed to pregnancy (Bealby et al., 1993).

Parasitaemia score was not influenced by nutrition in the present study. In contrast, previous studies have reported on effect of supplementation, especially of protein, on the intensity of parasitaemia (Katunguka et al., 1993). The mechanism by which haematic forms of trypanosomes can cause a reduction in both luteal and placental progesterone secretion might include gonadal atrophy due to thrombosis in the gonadal blood vessels, severe anaemia and thermal effects of pyrexia (Mutayoba et al., 1988). Alternatively, trypanosomes have been shown to reduce secretion of, and gonadal sensitivity to, gonadotrophins (Mutayoba et al., 1995b,c, 1996) possibly as a result of hypothalami–pituitary–adrenal activation (Mutayoba et al., 1995a). In addition, haematic forms of trypanosomes have been shown to cross the placental barrier (Ogaa et al., 1991; Elhassan et al., 1994) possibly leading to disturbance of placental progesterone production and fetal death.

The effects of nutritional supplements on the impact of trypanosomiasis during pregnancy in sheep have been described by Reynolds and Ekwuruke (1988) and have mainly revealed a reduction in both perinatal mortality and ewe mortality in the better-nourished ewes. Mortality rates, which are the ultimate measure of trypanotolerance, are affected by plane of nutrition (Reynolds and Ekwuruke, 1988). In the present study, highest mortality rates were seen in the HI and LC groups, probably as a result of infection and low nutrition combined with multiple pregnancy respectively. In the LI group mortality rates were lower but the fact that there were no multiple pregnancies could have been an advantage and maybe as a consequence these ewes were more trypanotolerant. The latter became also apparent in better pregnancy outcome rates and overall pregnancy rates.

5. Conclusions.

Since the mean circulating progesterone levels of those who completed gestation showed no significant differences between groups but only between individuals, we would conclude that the rates of conception and maintenance of pregnancy is a more justified way of measuring effects of infection and level of nutrition on reproductive performance. *T. congolense* infection impaired the establishment of pregnancy. Maintenance of pregnancy was influenced by both infection and nutrition. However, the importance of individual variation indicates that some ewes cope better with the effects of trypanosome infection and low nutrition on their reproduction performance.

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4.2

Effects of *Trypanosoma congolense* and nutritional supplements in Djallonke ewes on live weight during pregnancy, post partum weight, haematology parameters and lamb performance.

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Effects of *Trypanosoma congolense* and nutritional supplements in Djallonké ewes on live weight during pregnancy, post partum weight, haematology parameters and lamb performance

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SUMMARY

The effects of *Trypanosoma congolense* infection and nutritional supplements on live weight changes during pregnancy, haematology traits and offspring performance were studied in 42 Djallonké ewes. A randomised block design was used to allocate ewes to four treatment combinations, of which two were on a restricted diet (L) and the remainder on an unrestricted diet (H). Half of each nutritional group were infected with *T congolense* (LI, HI), the remainder serving as controls (LC, HC). The degree of anaemia following infection was similar in both infection groups ($P<0{\cdot}001$), but the erythropoietic activity, as judged by the increase in mean corpuscular volume, was significantly greater in the HI group ($P<0{\cdot}01$). Live weight gains during pregnancy attributable to higher supplements were significantly depressed by infection ($P<0{\cdot}01$). Post partum weight was lower in the LI group as compared with the LC control. Diet interacted significantly ($P<0{\cdot}01$) with infection and resulted in the lowest lamb growth rates in the LI group. It was concluded that dietary supplementation of trypanosome-infected Djallonké ewes during pregnancy and lactation improves productivity in terms of ewe live weight and improved lamb growth rates to weaning.

TRYPANOSOMOSIS is a major constraint to the numbers and productivity of livestock in sub-Saharan Africa where virtually all the costs of the disease are attributed to production losses (ILRAD 1993). Djallonké sheep, which are indigenous to West Africa and the main sheep breed in the Gambia, are considered as trypanotolerant (Mawuena 1986, 1987, Osaer et al 1994). Maintenance of adequate reproductive performance is essential for achieving successful animal production and the nature of trypanotolerance in sheep and goats has been described as an ability to maintain production under infection (Mawuena 1986, Osaer et al 1994, 1997, Goossens et al 1997). This definition is at variance with that of Murray et al (1982), who described trypanotolerance in cattle as an ability to limit reductions in haematocrit and to control the level and duration of parasitaemia. In trypanotolerant breeds, physiological factors such as pregnancy and lactation, intercurrent diseases and malnutrition may interact with resistance to the effects of trypanosome infection (Agyemang et al 1990, 1992, Little et al 1990). Under experimental conditions, both energy restricted and protein restricted diets clearly aggravate the negative effects of *Trypanosoma congolense* infection in sheep (Katunguka et al 1993, 1995). Under natural high tsetse challenge, the effect of supplementation in young Djallonké sheep was mainly seen in reduced mortality rates, delayed onset of trypanosomosis and reduced bodyweight losses (Hecker et al 1991). Akinbamijo et al (1994) studied the effect of *T vivax* during mid and late pregnancy in Djallonké ewes and revealed reduced feed intake and nitrogen retention leading to a decreased live weight gain, lower lamb birth weights, and lower growth and survival rates in the offspring of infected dams. The positive effect of nutritional supplements of trypanosome-infected dams during pregnancy was shown by Reynolds and Ekwuruke (1988), where ewe mortality and perinatal deaths were lower in the groups fed to maintenance than in groups on a submaintenance diet. The present experiment was designed to study the effects of *T congolense* and nutritional supplements on the health and productivity of Djallonké ewes during pregnancy and during lactation, as measured by the performance of their lambs.

MATERIAL AND METHODS

Animals and husbandry

Forty-two non-pregnant ewes were randomly selected from the breeding flock at the International Trypanotolerance Centre (ITC). All were routinely treated with anthelmintic (Panacur, Hoechst, Germany) and vaccinated against peste des petits ruminants (Tissue Culture Rinderpest vaccine, Hann Laboratory, Dakar, Senegal), and clostridial infections (Covexin, Pittman Moore, Cheshire, UK). All ewes were kept confined at the coastal site of the ITC, where the climate is sub-humid with a mean annual rainfall of approximately 1000 mm. Since susceptible zebu cattle at ITC have remained free of trypanosomosis for more than five years, it has been concluded that there is no risk of tsetse challenge at this site (W. Snow, personal communication). For the experiment a randomised complete block design was essentially used. Ewes were allocated to blocks based on their live weight at the start of the experiment. Within blocks, ewes were randomly allocated to one of four treatment combinations: high level of nutrition and infected (HI; 10 ewes), high level of nutrition and not infected (HC; 10 ewes), low level of nutrition and infected (LI; 11 ewes) and low level of nutrition and not infected (LC; 11 ewes). High level of nutrition comprised 120 g Crude Protein (CP) and 9 MJ per ewe per day, whereas the low diet offered 90 g CP and 7 MJ per animal per day. All ewes were placed on the appropriate diet, consisting of ground nut hay and concentrate, eight weeks from the start of the experiment. After parturition, the diets were adjusted for lactation by feeding the ewes with 155 g CP and 11·4 MJ day^{-1} for the high level and 118 g CP and 8·7 MJ day^{-1} for the low level. Prior to trypanosome infection, all animals were confirmed free

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of trypanosomosis using the dark ground (DG) buffy coat technique (Murray et al 1997). About 15 animals, which were distributed among the four groups, had been used in a previous experiment, where the same *T congolense* strain was used but all had been treated with diminazene aceturate (Berenil, 7 mg kg^{-1} live weight, Hoechst, Germany). Ewe live weights were recorded weekly using a Salter spring balance. Lambs were weighed at birth and thereafter weekly until the age of three months.

Breeding and trypanosome infection

Oestrus was synchronised by a 12-day treatment with an intra-vaginal progestagen sponge (Veramix, Upjohn, Crawley, UK) followed by an injection of serum gonadotrophin upon sponge withdrawal (PMSG-Intervet, 500 IU/ewe, Intervet, Cambridge, UK). The trypanosome infection took place during progestagen treatment, at day −10, in such a way that the peak of parasitaemia would coincide with oestrus, which was expected within 48 to 72 hours following sponge withdrawal (day 0). This time prediction to reach peak parasitaemia was known from previous experiments using the same strain (Osaer et al 1994, 1997, Goossens et al 1997). On the day of expected oestrus, one ram was introduced into each of the four groups and swapped around twice daily to ensure optimum mating activity. Rams were fitted with a colour marking harness to help confirm mating times.

Ewes allocated to the groups HI and LI were inoculated with a stabilate of the *T congolense* strain ITC 184. This is a cloned stabilate of *T congolense* derived from a goat at Kunting on the north bank of the River Gambia (Central River Division, The Gambia). The pathogenicity of this stabilate has been assessed in trypanosusceptible Zebu cattle (Dwinger et al 1992), in trypanotolerant N'Dama cattle (J. Bennison, personal communication) and in West African Dwarf goats and Djallonké sheep (Osaer et al 1994, 1997). The trypanosomes were obtained from mice during their first rising parasitaemia. Each ewe was inoculated intradermally with 1 ml infected mouse blood containing at least 10^4 blood stream forms, divided between five sites on the left flank.

Blood sampling and analysis

Blood samples were taken on the 11th day post infection (p.i.) ie, the day of expected oestrus, and thereafter at weekly intervals for 24 weeks to assess the degree of anaemia (PCV) and the parasitaemia level by the dark ground (DG) method. The number of trypanosomes was scored following the method of Paris et al (1982). Mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC) were determined until week 9 p.i., using an automated blood cell counter (ABX MINOS ST Haematology Analyser, Levallois, France).

Statistical analyses

Statistical analysis was carried out using SAS® statistical package version 6.11 (1996). Effects of treatments on lamb survival rates were compared using the Chi-square test. Ewe live weight during pregnancy and haematology traits were analysed by general linear model (GLM procedure) as repeated measurements. The following main effects were included: infection (control vs infected), diet (low vs high), block, period (pre-infection: week −5 to −1; acute: week 0 to 6; chronic: week 7 to 24), week (nested within period), ewe (nested within treatment, diet and block), pregnancy status (empty, early, late) and the interactions: infection*diet; infection*period; diet*period, and infection*diet*period. In view of the aims of the experiment, joint effect of infection and nutrition was of high interest and this was tested in the model by the interactions infection*diet and infection*diet*period. The ewe effect was regarded as random. Variables such as post partum weight, lamb birth weight and growth rate up to 90 days as calculated from linear regression analysis, were tested in a similar model, which also included the effects of litter size, sex and start weight of dam (as a covariable for lamb performance data only). All hypotheses were tested by the F-test (Snedecor and Cochran 1980). Means of the different groups are presented as least square means ± standard error (ie mean ± SEM). Differences in defined interactions are described in order to estimate the effects of trypanosome infection and/or nutrition for the changes between periods. Results were regarded as statistically significant when the Type I error probability was smaller than 5 per cent.

RESULTS

Parasitaemia and haematology changes

Trypanosomes were never detected by the DG method in five ewes from group LI and four from group HI. These animals had been infected with the same strain in the previous

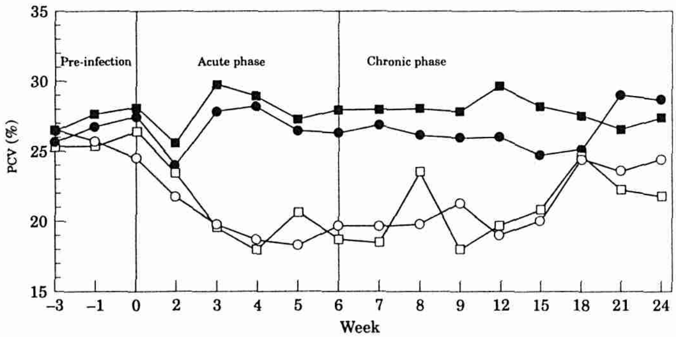

FIG 1: Mean packed cell volume for groups high control (HC, ■), low control (LC,●), high infected (HI, □) and low infected (LI, ○) during pre-infection, acute and chronic phase of infection

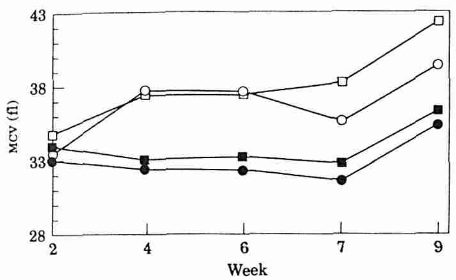

FIG 2: Mean cell volume during nine weeks post infection for groups high control (HC, ■), low control (LC,●), high infected (HI, □) and low infected (LI, ○)

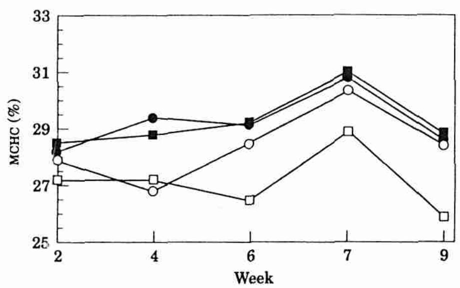

FIG 3: Mean cell haemoglobin concentration during nine weeks post infection for groups high control (HC, ■), low control (LC,●), high infected (HI, □) and low infected (LI, ○)

experiment and consequently were omitted from further analyses. Trypanosomes were detectable by the 11th day p.i. in the remaining ewes, with parasitaemia scores being maximal by the second week p.i. and remaining detectable until the end of the experiment, 24 weeks p.i. At no point in the experiment were trypanosomes detected in the control ewes.

PCV levels dropped following infection (see Fig 1) to reach a minimum at week 4, followed by slight recovery as from week 15 onwards. The reduction in PCV due to trypanosome infection was estimated as 5·2±0·8 percentage points (P<0·001) in the acute period. PCV levels in the trypanosome-infected groups slowly recovered towards the end of the trial, still remaining significantly lower than the controls. Early pregnant females had on average higher PCV levels as compared with empty females with a difference of 1·1±0·6 percentage points (P<0·05). As pregnancy further progressed the average PCV levels declined again, thus the difference between empty and highly pregnant ewes was not significant any longer. PCV levels were not affected by the level of diet.

The mean MCV level was higher in trypanosome-infected sheep than in controls during the first nine weeks p.i. (37·7±0·3 and 33·4±0·2 fl respectively, P<0·001) (see Fig 2). However, the interaction of diet*infection*period revealed a significantly higher MCV for the HI group between week six and nine, with a difference of 2·75±1·1 fl (P<0·01) compared with the LI group. High level of nutrition resulted in higher MCV levels (36·3±0·2 and 34·8±0·2 fl for high and low level, respectively; P<0·05). MCHC levels decreased in both infection groups (27·9±0·2 per cent) vs controls (29·3±0·1 per cent) (P<0·001), but the interaction of infection*diet*period revealed that MCHC was even lower for the HI group as compared to the LI group between week six and nine, with respective means of 27·7 ± 0·4 per cent and 29·4±0·3 per cent (P<0·05) (see Fig 3).

Live weight changes

Group mean live weights for the duration of the experiment are presented in Fig 4. Diet had an overall significant effect on live weight (P<0·05), as well as pregnancy status (P<0·001). The difference in weight due to higher level of supplementation remained until week 18, thereafter a sharp increase was seen for the control groups mainly, until parturitions started. The interaction of infection*diet*period revealed a difference (P<0·01) in live weight of 1·1±0·4 kg between high and low control groups vs the difference between high and low infected groups for the changes between acute and chronic period, being evidence of a better effect of supplementation in the control groups than in infected groups. In all treatment groups, some of the animals failed to become pregnant following the first synchronised cycle and became pregnant following the second or later cycles (repeat breeders). Therefore, the status of pregnancy was taken into consideration when analysing live weights. Highly pregnant animals weighed on average 1·9±0·3 kg more (P<0·001) than non-pregnant ewes.

Post partum weights were recorded in 22 ewes. An overall diet effect (P<0·05) remained, with mean post partum weights for the high and low diets being respectively

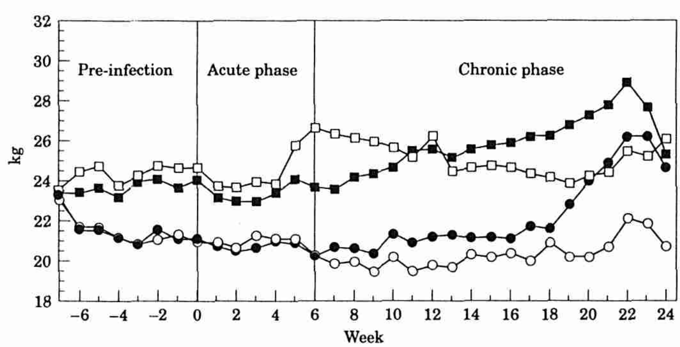

FIG 4: Mean live weight for groups high control (HC, ■), low control (LC,●), high infected (HI, □) and low infected (LI, ○) during pre-infection, acute and chronic phase of infection

TABLE 1: Post partum weight of ewes; birth weight and growth rates of lambs up to three months old

Treatment*diet	Post partum weight (kg)	Birth weight (kg)	Daily gain (g)
HC	24·4 (0·8)	1·6 (0·2)	54·3^{a} (5·9)
LC	22·6 (1·4)	1·8 (0·2)	45·2^{ac} (10·7)
HI	30·7 (3·6)	1·9 (0·5)	121·0^{b} (19·7)
LI	20·4 (1·7)	0·9 (0·3)	20·9^{c} (8·1)
Significance	ns (P<0·07)	ns (P<0·08)	P<0·01)

Least square means (SEM) for treatment groups high control (HC), low control (LC), high infected (HI) and low infected (LI).
Different superscripts within a column indicate a significant difference (P<0·01)

27·6±1·9 and 21·5±1·3 kg. The interaction of diet*infection tended (P<0·07) to show differences between the four groups. Table 1 shows the least square means for the four treatment groups. Litter size did not affect post partum weight.

Offspring performance

Single lambs had higher birth weights and growth rates up to three months than multiple lambs (2·0±0·2 kg and 79·0±8·3 g day^{-1} vs 1·1±0·2 kg and 41·7±9·1 g day^{-1} respectively; P<0·01). Growth rates of lambs whose dams were on the high supplemented diet had faster daily gains than the low supplemented groups (87·7±10 g day^{-1} vs 33·1±8 g day^{-1} respectively, P<0·01). Trypanosome infection had no overall significant effect on either birth weight or growth rate. The interaction diet*infection was significant for growth rates (P<0·01), but not for birth weight. Mean lamb birth weights and growth rates up to three months are presented in Table 1 for the four groups HC, LC, HI and LI. Lamb mortality rates are presented in Table 2. Neither infection nor diet had significant effect on lamb mortality rates.

DISCUSSION

In the present study it was clear that *T congolense* infection and a low plane of nutrition interacted to affect the productivity of female Djallonké sheep. Ewe live weights increased by an average of almost 2 kg during the pregnancy period and while infection per se did not affect live weight significantly, there was an interaction between dietary plane and trypanosome infection so that weight gains due to higher supplementary feeding were significantly depressed by infection. A beneficial effect of nutrition on the effects of trypanosomosis during pregnancy and lactation, namely better ewe live weight gains, lower ewe- and lamb mortality and higher lamb growth rates has been described by Reynolds et al (1988). In the presence of adequate nutrition, no significant live weight differences were seen in weights between control and *T congolense* infected Djallonké breeding ewes, despite evident clinical effects (Osaer et al 1994); although trypanosome infection lowered weight gain in Djallonké rams despite showing less severe clinical signs than breeding females (Osaer et al 1997).

Acute anaemia is a typical pathological finding in trypanosome-infected animals and infected Djallonké sheep and goats manifest low PCV levels until the chronic stage of the disease with persistent parasitaemia (Osaer et al 1994, Goossens et al 1997). In the present experiment, although PCV recovered a little from week 15 onwards when the mean parasitaemia score was below score 1, the effect of infection remained. However, there could have been a confounding effect of pregnancy since PCV increased significantly during early pregnancy. In the present study, a higher level of dietary supplementation did not ameliorate the degree of anaemia following trypanosome infection. This is in contrast to observations in infected sheep and cattle which were fed on high energy diets (Katunguka et al 1995, Little et al 1990). Katunguka et al (1993) showed no effect of higher protein intake on the degree of anaemia following *T congolense* infection, but a better erythropoietic response and a quicker recovery following treatment was observed. Similarly, a better control of the anaemia caused by natural trypanosome infection was observed in N'Dama cattle kept on a high plane of nutrition (Aygemang et al 1990). In the present experiment, there was also evidence of a better erythropoietic response (observed as macrocytic anaemia in the chronic stage of infection) in the infected group kept on a high diet. In both infection groups there was a hypochromic anaemia, but MCHC levels remained lower in the high supplement infection group up to week 9 post-infection whilst those in the low infection group increased. Haemolysis is, by far, the most important mechanism causing anaemia in trypanosome infections (Anosa 1988). In the acute phase during the early stages of infection, the rate of destruction of red blood cells is not sufficiently compensated by erythropoiesis, but when infection persists, destruction and replacement of RBCs becomes more balanced. During the recovery phase, the rate of erythropoiesis exceeds the destruction of RBCs. An increased MCV is indicative of younger erythrocytes released in the blood stream, which can be hypochromic because of their immaturity. A macrocytic, hypochromic anaemia was also found in *T congolense*-infected sheep by Wassink et al (1997). In the latter work, however, there were no differences between diets whereas in this study, the macrocytic and hypochromic responses following infection were more pronounced in ewes on the high plane diet. It is apparent from the present results that infected ewes on the low dietary plane, showed a smaller increase in erythropoiesis than ewes on the high plane. Dietary plane did not however affect PCV levels, which is in contrast to the observations of Abdullahi et al (1986) who observed lower PCV levels in protein deprived sheep and Wassink et al (1997) who noted reduced PCV levels in sheep fed a roughage with lower energy and protein.

Despite finding that post-partum weights were not generally affected by infection in the present study, an interaction between diet and infection revealed a tendency for lower (ns, P>0·05) live weights in the low infected group as compared to the low controls, but this was not the case in the high supplemented groups. In the present study, birth weight and lamb growth rate were not significantly affected by trypanosome infection. Nevertheless, the effects of diet and infection were not just additive and resulted in the low-

TABLE 2: Perinatal mortality and total mortality rate of lambs up to three months (absolute number/total number born) in treatment groups high control (HC), low control (LC), high infected (HI) and low infected (LI)

Treatment*diet	≤24 hours	0 to 3 months
HC	7/19	9/19
LC	0/7	2/7
HI	1/2	1/2
LI	1/5	1/5
Chi-square	ns	ns

est lamb growth rates occurring in the low infected group, but there was no difference between control groups. Goossens et al (1997) studied the productivity of Djallonké ewes on an adequate plane of nutrition following *T congolense* infection. Their findings were that in the first year post infection, there was no significant reduction in productivity per ewe at lambing, but when lamb growth and mortality rates were taken into account, infection caused reduced production per ewe at months 1, 3 and 5 post partum. Akinbamijo et al (1994) showed that *T vivax* resulted in lower lamb birth weight and survival rates when infection took place during late pregnancy as compared with mid pregnancy, although in the present study, neither trypanosome infection nor diet had an influence on lamb mortality rates. The high mortality rate in the high supplement, non-infected group may have been confounded by the fact that more multiple births occurred in this group, resulting in smaller, weaker lambs. In the present study the trypanosome infection was acute at the time of establishment of pregnancy so conception rates were most affected, although infection and diet did also interfere with the maintenance of pregnancy (Osaer et al 1998). However, the trypanosome infection persisted throughout the experiment and thus the infection became chronic around parturition and beyond, with a less severe effect on lamb performance.

We can conclude that under the conditions of the present experiment dietary supplementation of trypanosome-infected Djallonké ewes during pregnancy and lactation improved productivity in terms of ewe live weight, lamb birth weight and improved lamb growth rates to weaning.

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Effects of *Trypanosoma congolense* infection and diet on puberty, age at first lambing and haematology changes in Djallonké ewe lambs

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Abstract

The interactions between *T. congolense* infection and nutritional supplements on onset of puberty and age at first lambing were observed in 24 young Djallonké ewes. As experimental design, a randomised complete block design was used with four treatment combinations, of which two were kept on a restricted diet (L), the remainder on an unrestricted diet (H) and half of each nutritional group being infected with *T. congolense* (LI and HI), the remainder serving as controls (LC and HC). Infection with *T. congolense* took place at an average age of 6 months and 15 days. Mortality due to trypanosome infection was zero and clinical symptoms were not obvious. Intensity of parasitaemia and packed cell volume (PCV) drop following trypanosome infection were similar in both infected groups (HI and LI). High dietary supplementation resulted temporarily in a better haematopoietic response following trypanosome infection, measured as a macrocytic anaemia. Dry matter intake (DMI) was significantly depressed in the HI group immediately following infection. Trypanosome infection had a negative effect on live weight gain during the chronic phase, with the difference being most obvious in the HI group (interaction diet$\times$infection; $p\leq0.05$). Whereas trypanosome infection had no significant effect, high supplementary feeding significantly reduced the age at first cycling. Age at first lambing was similarly reduced by the diet. Trypanosome infection tended ($p\leq0.09$) to delay age at first lambing with a mean difference of 31.5 ± 22.4 days between infected and controls. Interactions between diet and infection for age at first cycling/lambing were not significant, indicating these effects were just additive. Neither birth weights nor growth rates of offspring born to the experimental animals were significantly affected by previous trypanosome infection, nor by the diet of the dam. In contrast, lamb mortality up to 3 months of age was significantly increased by infection of the dam and most losses arose in group LI. In conclusion, the effects of trypanosome infection on puberty and age at first lambing were indirectly

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mediated through depression of growth rates. Nutritional supplementation enabled a better erythropoietic response to *T. congolense* infection and better offspring survival rates but resulted in more depressed weight gains. The results however clearly indicated the delaying effect of insufficient feeding on onset of puberty and reproductive performance in young Djallonké sheep.

Keywords: Sheep-protozoa; Puberty; *Trypanosoma congolense*; Feeding and nutrition; Trypanotolerance

1. Introduction

Trypanosomosis is one of the major constraints limiting numbers and productivity of livestock in sub-saharan Africa. As a consequence, only trypanotolerant breeds do survive, reproduce and remain productive in tsetse infested areas without the need for trypanocidal treatment (Murray et al., 1982).

In the west African traditional mixed farming systems, small ruminants play a crucial role in providing protein (meat and milk) whilst they also serve as a cash reserve and protection against agricultural crop failure. A recent livestock survey in the Gambia (Department of Livestock Services and International Trypanotolerance Centre census, 1993) counted 155 132 head of sheep, mainly of the Djallonké breed (>98%). There are several advantages of keeping small ruminants as compared to cattle in traditional livestock production systems and, in terms of reproductive performance, the list includes their early puberty and age at first lambing, short lambing intervals and non-seasonal breeding. Djallonké sheep are considered as trypanotolerant (Touré et al., 1981; Mawuena, 1986, 1987; Bengaly et al., 1993; Osaer et al., 1994, 1997; Goossens et al., 1997a, b). Adequate reproductive performance is an essential component in efficient animal production and trypanotolerance in sheep and goats has been described as an ability to maintain production under infection (Mawuena, 1986; Osaer et al., 1997; Goossens et al., 1997b). This definition is at variance with that of Murray et al. (1982), who described trypanotolerance in cattle as an ability to limit reductions in haematocrit and to control the level and duration of parasitaemia. In trypanotolerant cattle, malnutrition has been reported as a factor which can reduce trypanotolerance (Murray et al., 1982; Agyemang et al., 1990, 1992; Little et al., 1990). Some studies in Djallonké sheep describe the effects of nutritional status on their resilience to the effects of infection under natural trypanosome challenge (Hecker et al., 1991), and under experimental infection (Reynolds and Ekwuruke, 1988). Both energy restricted and protein restricted diets clearly aggravated the negative effects of an experimental *T.congolense* infection in Scottish blackface sheep (Katunguka-Rwakishaya et al., 1993, 1995). However, malnutrition may also influence directly the reproductive performance of sheep and dietary level should, therefore, be kept adequate. In view of the known influence of body mass on puberty (Foster and Olster, 1985; Bronson and Rissman, 1986) and the absence of literature on the effects of trypanosome infection on puberty, the present experiments were designed to study the effects of *T. congolense* infection with or without nutritional supplements on health, weight gain and onset of reproductive function in Djallonké ewes.

2. Materials and methods

2.1. Animals and husbandry

24 ewe lambs were selected from the breeding flock after weaning at approximately 4 months of age and average live weight of 10 kg. All of the ewe lambs were treated with fenbendazole (Panacur, Hoechst, Germany) and vaccinated against peste des petits ruminants (Tissue Culture Rinderpest vaccine, Hann Laboratory, Dakar, Senegal), and clostridial infections (Covexin, Pittman Moore, Cheshire, UK).

2.2. Experimental site

All lambs were kept confined at the coastal site of the International Trypanotolerance Centre (ITC). The climate is sub-humid with a mean annual rainfall of approximately 1000 mm. Since susceptible zebu cattle at ITC have remained free of trypanosom infections for more than 5 years, it has been concluded that there is no risk of tsetse challenge at this site (W. Snow, personal communication).

2.3. Experimental design and dietary conditions

For the experiment a randomised complete block design was used with animals allocated into blocks based on their date of birth. Within blocks, lambs were randomly assigned to one of the four treatment combinations: high level of nutrition and infected (HI), high level of nutrition and not infected (HC), low level of nutrition and infected (LI) and low level of nutrition and not infected (LC). The feed was offered individually so that daily intake could be measured. The ration comprised of groundnut hay, groundnut cake and rice bran and had a dietary composition of 9.6 Mj metabolisable energy (ME)/kg dry matter (DM) and 19% crude protein (CP) with a metabolisability of 0.55 (Q). The ration was restricted in the low diet groups to allow a daily live weight gain of about 50 g, (i.e., initially 3.7 Mj ME and 66 g CP), whereas the high-diet groups were offered an unrestricted amount far above the requirements (i.e., initially 6 Mj ME and 115 g CP) (McDonald et al., 1978). The rations were adjusted to average live weight during the course of the experiment, however, the same composition of the diet with two dietary levels was kept. All ewe lambs were placed on the appropriate diet five weeks before trypanosome infection and confirmed free of trypanosomes using the darkground (DG) buffy coat technique (Murray et al., 1977). During the experiment a ram, fitted with a colour marking harness was kept with the lambs at all times for oestrus detection and to help confirm mating times. Data on subsequent lambings, birth weights and lamb performance were recorded till 90 days after lambing.

2.4. Trypanosome infection

12 ewe lambs (assigned to either of the groups HI or LI) were infected in blocks, once they had attained 6 months and 15 days of age. All experimental times are related to this date of infection. The inoculum was a stock of *T. congolense* ITC84, a clone derived

(Dwinger et al., 1992) from a goat at Kunting on the north bank of the River Gambia (Central River Division, Gambia). The pathogenicity of this clone has been assessed in trypanosusceptible Zebu cattle (Dwinger et al., 1992), in trypanotolerant N'Dama cattle (J. Bennison, personal communication) and in west African dwarf goats and Djallonké sheep (Osaer et al., 1994, 1997; Goossens et al., 1997a, b). The trypanosomes were obtained from mice during their first parasitaemia. Each lamb was inoculated intradermally with 1 ml infected mice blood containing at least 10^4 bloodstream forms, divided between five sites on the left flank. Although the observation period for haematology and weight parameters for the ewes did not exceed 40 weeks, trypanocidal treatment was only given when ewes were confirmed pregnant (by plasma progesterone measurements) whilst all ewes were further kept on the two dietary levels for the purpose of recording lamb performance data.

2.5. Measurements and analyses

Individual food intake was recorded daily. Live weights were recorded weekly using a Salter spring balance. DM and Nitrogen (N) contents of the offered ration and bulked refusals were determined weekly according to A.O.A.C. (1975) procedures. Blood samples were taken at weekly intervals to assess the degree of anaemia (PCV %) and the parasitaemia level by the dark ground (DG) method, continuing until week 40 post infection. The number of trypanosomes was scored by the method of Paris et al. (1982). Red blood cell (RBC) counts, mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC) were calculated upto week 12 using an automated blood cell counter (ABX MINOS ST Haematology Analyzer, Levallois, France). Plasma progesterone concentration was measured weekly using a double antibody enzyme immunoassay as described by Meyer and Güven (1986) and Prakash et al. (1987) but with modifications in the form of rat anti-progesterone monoclonal antibody (Sigma Clone 2H4), anti-rat coating antibody (Sigma, R5130) and peroxidase-labelled progesterone label (Sigma P3659). The assay standard curve was prepared in plasma from a castrated ram and ranged from 10–0.31 ng/ml. The sensitivity of this test was determined to be 0.59 ng/ml (σ=0.19, n=21). Four control values with known values of 0.5, 1.0, 3.5 and 5.0 ng/ml were included in each assay. The inter-assay coefficients of variation for the four control values were 16.1%, 18.3%, 16.4% and 9.2%, respectively. The intra-assay coefficients of variation for the four controls were 7.1%, 5.2%, 6.9% and 8.1%, respectively.

2.6. Statistical analyses

Statistical analysis was carried out using SAS® statistical package version 6.11 (SAS, 1989–1996).

Continuous variables (live weights, DMI, haematology and parasitaemia level) were analysed by general linear model (GLM procedure) as repeated measurements. The following main effects were included: infection (control vs. infected), diet (low vs. high), block, period (pre-infection: week from −5 to −1; acute: week 0–5, post-acute: week 6–19, chronic: week 20–40), week (nested within period), ewe (nested within treatment, diet and block), and the interactions infection×diet, infection×period, diet×period and infection×diet×period. Considering the design of the experiment, joint effect of infection and nutrition was of high interest and tested in the model by the interactions infection×diet and infection×diet×period. The animal effect was regarded as random. Age at first cycling, age at first lambing, lamb birth weight and lamb growth were tested in a similar model with littersize and sex of the lamb considered as additional effects for the two latter parameters. All hypothesis were tested by the *F*-test (Snedecor and Cochran, 1980). Means of the different groups are presented as least square means±standard error (i.e., mean±SE). Differences in defined interactions are described in order to estimate the effects of trypanosome infection and/or the diet for the different periods.

3. Results

3.1. Clinical observations, parasitaemia and haematology

No obvious clinical symptoms of trypanosomosis were noticed among the infected lambs in the period immediately following infection. One animal out of the LI group died in week 13 (day 92) post infection, without pathological lesions normally associated with trypanosomosis.

All animals of the LI and HI group were positive for trypanosomes by week 2 post infection, reaching first peak during week 3 for the HI group and week 4 for the LI group (see Fig. 1). Mean parasitaemia level stayed high during both the acute and post-acute

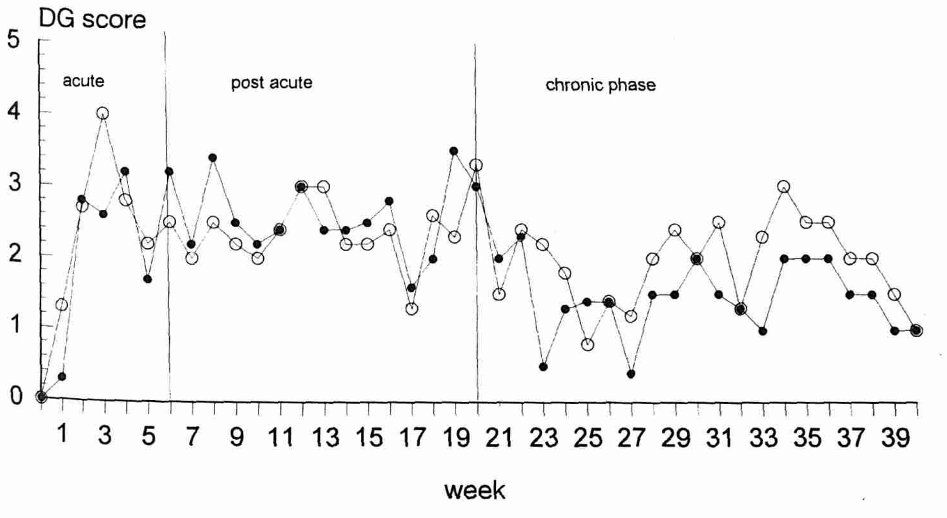

Fig. 1. Mean weekly parasitaemia level (DG-score) HI and LI groups during 40 weeks post infection.

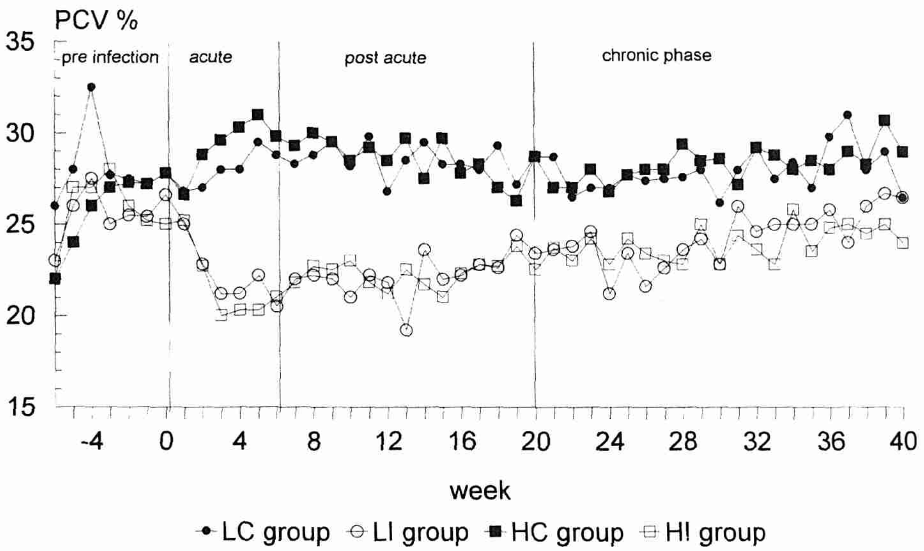

Fig. 2. Mean PCV for groups HC, LC and LI during pre-infection phase, acute, post-acute and chronic phases of infection.

periods, and declined during the chronic phase, giving mean parasitaemia scores 2.0±0.1, 2.5±0.1 and 1.8±0.1, respectively for the three periods. All animals remained parasitaemic until the end of week 40, after which they were treated. Despite the higher parasitaemia score initially in group HI, no overall significant differences were found between groups HI and LI, indicating that diet had no effect on parasitaemia level.

PCV percentages dropped significantly (p<0.001) in both infected groups (see Fig. 2) concomitant with the first wave of parasitaemia, with overall mean PCV levels, respectively, for infected and control groups of 23.6±0.2% and 28.4±0.2%. The low PCV levels in the infected sheep persisted into the acute and post-acute periods of infection declining by 3.6±0.8 and 4.3±0.7% units (p<0.001) respectively compared to their pre-infection values. Overall, diet did not affect the PCV level. Also the finding that PCV levels of the groups HI and LI were not significantly different indicated no positive interaction diet×infection over periods. PCV levels recovered slowly during the chronic phase, with values for groups LI and HI remaining significantly lower than their controls (HC and LC) at the end of the observation period.

RBC counts dropped significantly following infection to reach a minimum at week 4 (see Fig. 3), overall mean RBC counts being $7.8\pm0.1\times10^6/\mu l$ for infected groups versus $9.4\pm0.1\times10^6/\mu l$ for controls (p<0.001). The drop tended to be more severe in the HI group but there were no significant differences between the two dietary levels. By week 12 post infection, both infected groups had almost reached their pre-infection values for RBC.

MCV values in the HI group showed a transient but not significant increase in MCV values between week three and eight as shown in Fig. 4. Diet significantly influenced MCV, with the high supplemented ewes having an overall mean of 31.8±0.4 fl versus

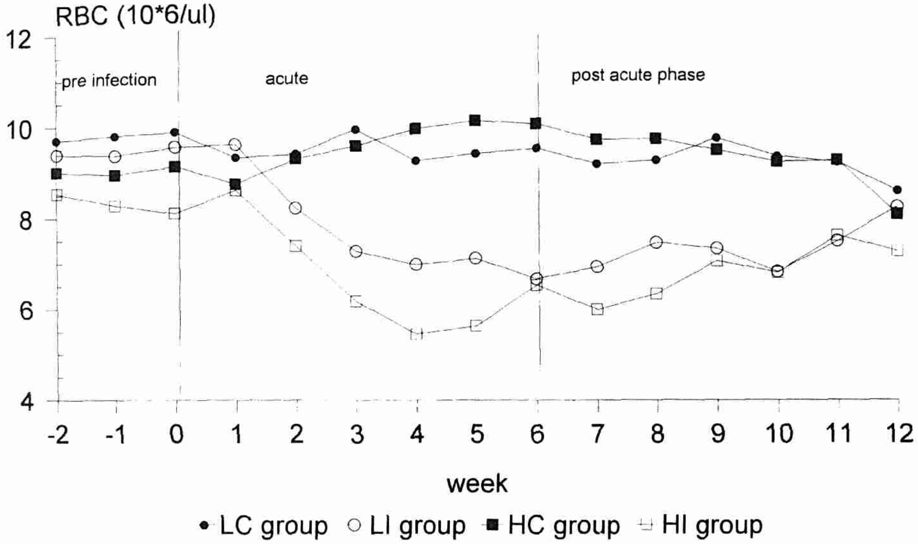

Fig. 3. Mean RBC counts for groups HC, LC, HI and low infected LI till 12 weeks post infection.

29.4±0.4 fl for the low supplemented ones (p<0.01). The interactions diet×infection and diet×infection×period were not significant, so the two factors were merely additive.

Group mean MCHC following infection is plotted in Fig. 5. Despite a transient rise at week 6 and again at weeks 11–12 in both infected groups, neither infection nor diet

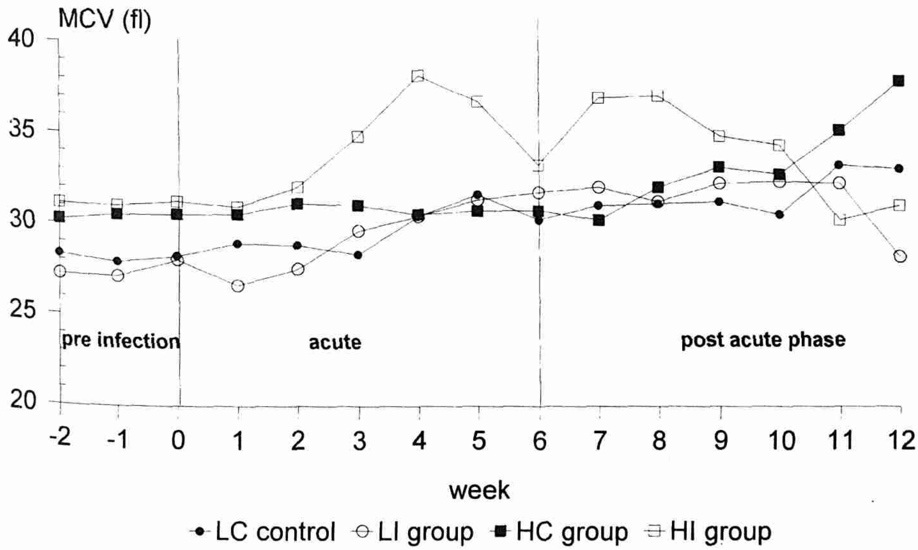

Fig. 4. MCV for groups HC, LC, HI and LI till 12 weeks post infection.

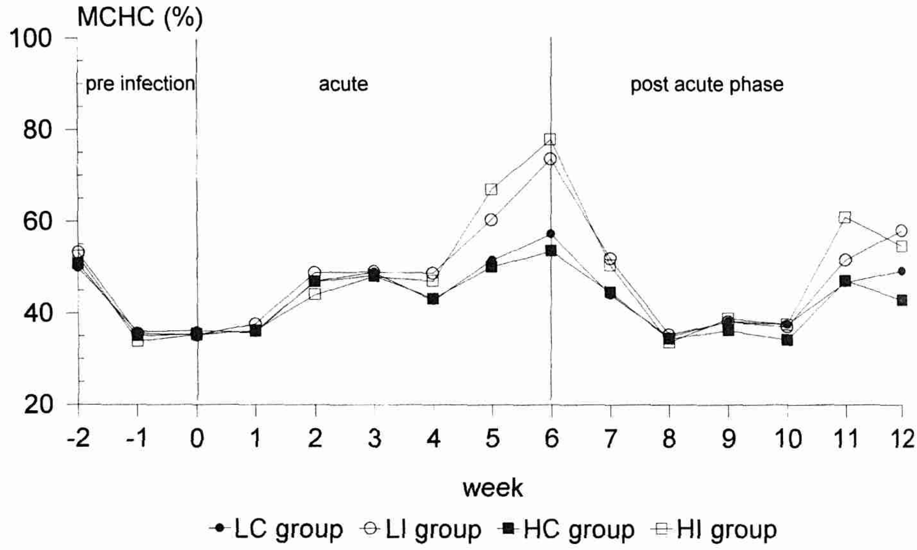

Fig. 5. MCHC for groups HC, LC, HI and LI till 12 weeks post infection.

significantly influenced the MCHC with 47.1±2.5% and 43.6±2.5%, respectively for infected and control groups.

3.2. DMI and live weight responses

Trypanosome infection caused a significant decrease in DMI ($p<0.01$), overall means being 490.0±0.1 g versus 479.1±0.1 g for control and infected groups, respectively (see Fig. 6). The effect of infection did only occur in the acute period with a mean reduction in DMI of 14.7±3.0 g for all infected animals (interaction infection×period; $p<0.001$). However, the drop in DMI was only observed in the HI group, despite its already lower intake level during the pre-infection period. The contrast which described differences in DMI over periods due to infection as compared to low diet groups and pre-infection values, estimated a drop in DMI of 27.7±6.2 g for the HI group in the acute phase of infection (interaction infection×diet×period; $p<0.001$).

Mean weekly live weights are plotted in Fig. 7. Whilst the LI group and their counterpart LC group showed similar increases in live weight during the study, the body weight increase in the HI group was significantly lower as compared to the HC group. Mean live weight of the HI group started to increase in the chronic phase of infection, but remained lower than the HC group. Despite their lower mean weight at the start, the total increase in mean body weight for the HI group over the entire observation period was 7.4 kg compared to 10.3 kg for the HC group, whereas for the low diet groups the increases for LI and LC groups were, respectively, 4.3 and 5 kg. Trypanosome infection significantly reduced live weight gain during the chronic phase of the infection, with control and infected groups having respective mean weights of 16.3±0.05 kg and 14.6±0.06 kg (interaction infection×period, $p<0.001$). The significant diet×infection

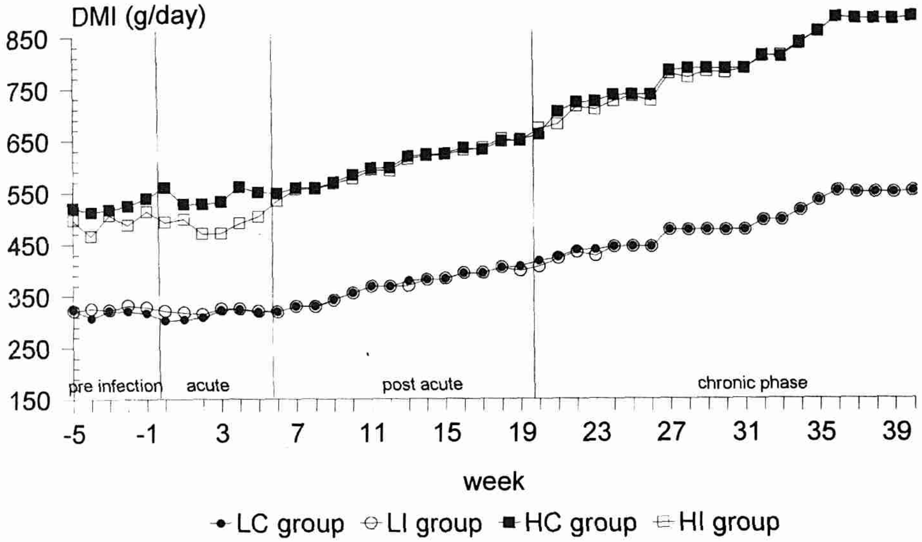

Fig. 6. Mean DMI for groups HC, LC, HI and LI during pre-infection phase, acute, post-acute and chronic phases of infection.

interaction ($p \leq 0.05$) indicated that differences occurred mainly between the high diet groups with overall group live weights of 14.2, 11.9, 11.7 and 12.1 kg (pooled SE=0.06), respectively, for the four treatment groups HC, HI, LC and LI. There was an overall

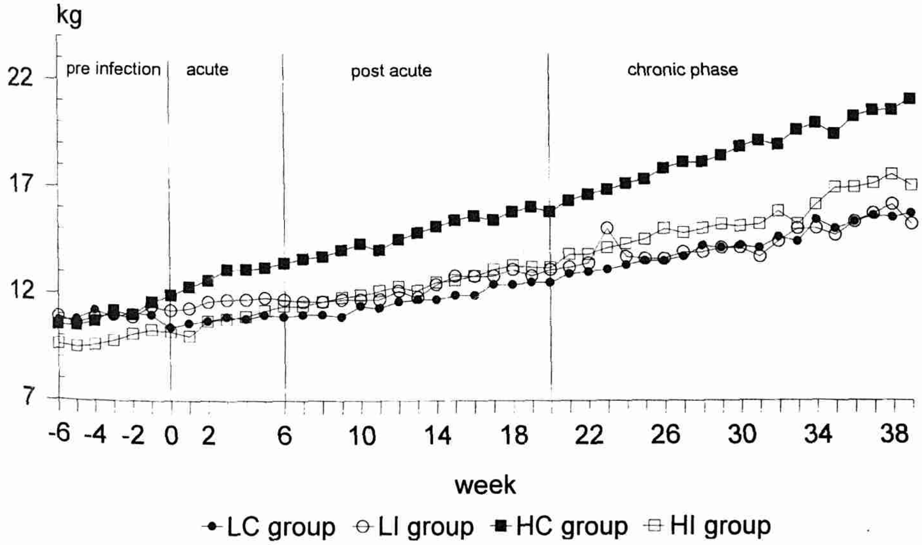

Fig. 7. Mean live weight for groups HC, LC, HI and LI during pre-infection phase, acute, post-acute and chronic phases of infection.

Table 1
Mean age (days±SE) at first cycling (plasma progesterone >1.0 ng/ml) and age (days±SE) at first lambing in groups of ewe lambs on HI, HC, LI and LC

Group	Age at first cycling (days±SE)	Age at first lambing (days±SE)
HC	236.5±47.2	549.8±20.5
HI	273.2±47.2	579.6±23.4
LC	353.3±47.2	599.7±20.5
LI	343.2±47.2	632.9±23.4

positive effect of diet on live weight, with differences between high and low level of diet of 0.9, 2.1 and 3.2 kg (pooled SE=0.2) ($p<0.001$), respectively, during the acute, post-acute and chronic period.

3.3. Onset of puberty and age at first lambing

The age when the first peak in plasma progesterone concentration occurred (i.e., >1.0 ng/ml) varied considerably. It also appeared that some animals had a period of low plasma progesterone (i.e., non- cycling activity) after their first progesterone peak whilst others had subsequently regular cycles. Group means are presented in Table 1. There was a negative correlation between age at first cycling and weight (−0.7; $p<0.001$). Trypanosome infection did not affect significantly the age at first cycling. High supplementary feeding significantly decreased the mean age at first cycling with 254.8±33.4 days versus 348.3±33.4 days respectively for high- and low-diet groups ($p\leq0.05$). The interaction of the diet with infection was not significant, thus both effects were just additive.

Average age at first lambing is presented in Table 1. High supplementary feeding significantly decreased age at first lambing (564.7±15.5 days versus 616.3±15.5 for H and L groups, respectively; $p<0.05$). There was no significant effect of infection on age at lambing (606.3±17.1 days versus 574.8±14.5 days for infected and controls, respectively). The effects of diet and infection were additive, thus, highest age at first lambing was found in the LI group, followed by the LC group.

3.4. Lamb performance

Average birth weights were 2.0±0.2 kg for HC, 1.3±0.2 kg for HI, 1.8±0.2 kg for LC and 1.8±0.3 kg for the LI group (interaction diet×infection, ns: $p\leq0.09$). There was no significant effect of earlier infection of the dam on birth weight of the lambs, nor had supplementary feeding an influence.

Likewise, daily lamb growth till 90 days, calculated as a linear regression, was not significantly influenced either by previous infection or by the diet of the dams (means of 65.1±11.0 g for HC, 52.2±11.0 g for HI, 45.6±8.8 g for LC and 35.2±14.8 g for LI). On the contrary, lamb mortality up to 90 days was significantly elevated due to the earlier infection of the dams ($p<0.01$) with four out of nine lambs which died versus zero mortality in the control groups. Overall, level of diet did not affect significantly lamb mortality, however, three of the four lambs which died originated from the LI group.

4. Discussion

The present results illustrate the complexity of the interactions between trypanosome infection and nutritional status. *T. congolense* infection caused a persistent anaemia which could not be countered by protein and energy supplementation. Growth rates in the infected ewe lambs were depressed, partly as a direct result of reduced DMI intake. Surprisingly *T. congolense* infection did not appear to influence directly age at puberty, a parameter which was lowest in non-infected lambs on the high level feed supplements. Similarly, offspring born to infected ewes had higher mortality rates than those born to controls despite similar birth weights and growth rates.

The ewe lambs resisted the effects of *T. congolense* infection well in clinical terms and mortality rate was zero, in correspondence to previous experimental infections in Djallonké sheep (Osaer et al., 1994, 1997). The level of parasitaemia in the infected animals remained high during the 40 weeks observation period and no difference in intensity of parasitaemia could be ascribed to nutritional status as shown previously in *T. congolense* infected N'Dama cattle fed on different planes of nutrition (Romney et al., 1997) and in *T. vivax* infected Djallonké sheep fed either to maintenance or submaintenance (Reynolds and Ekwuruke, 1988). In contrast, Katunguka-Rwakishaya et al. (1993) have described beneficial effects of protein supplementation on the intensity of parasitaemia in Scottish Blackface lambs infected with *T. congolense*. In the present experiment there were no signs of self cure nor ability to control the level and duration of parasitaemia, also shown with previous experimental *T. congolense* infections of Djallonké sheep (Osaer et al., 1994, 1997; Goossens et al., 1997a, b). The acute drop in PCV in the infected ewe lambs in this experiment was less severe than that observed in older ewes (Osaer et al., 1998), the anaemia, however, persisted into the chronic phase of infection and was not alleviated by high nutritional supplementation. Wassink et al. (1997) similarly found that the effects of *T. congolense* on PCV were not different between the two dietary groups. A temporary macrocytic anaemia was observed in the high supplemented infected group, indicating the presence of a younger RBC population and thus a better erythropoietic response as compared to the LI group. Similar findings have been described in trypanosome-infected Djallonké ewes at the time of breeding kept on a high plane diet (Osaer et al., 1998) and in infected Scottish blackface sheep on a high protein diet (Katunguka-Rwakishaya et al., 1993). The low protein level in this study of around 70 g/day, could have been insufficient in supporting the erythropoietic response. Remarkably, however, that RBC counts in both infected groups almost returned to pre-infection values by week 12, whilst PCV levels in infected ewe lambs were still much lower than the controls at that time. Although no measurements of plasma- and total blood-volume were performed, the latter findings may indicate indirectly evidence of increased plasma volume due to trypanosome infection, which is in correspondence with previous reports in *T. vivax*-infected sheep (Clarkson, 1968; Anosa and Isoun, 1976) and in *T. congolense*- infected sheep (Katunguka-Rwakishaya et al., 1992).

The present dietary levels did not alter haematocrit, in contrast to other findings where lower PCV levels were apparent in Scottish black face sheep fed on low energy and low protein roughage (Wassink et al., 1997). In the present study, the Djallonké ewes on the low plane received even lower levels of energy (about 3.5 ME) and protein (about 70 g) compared to the latter study. This discrepancy may be ascribed to breed differences; however, further work would be required to verify this hypothesis.

DMI was significantly depressed by infection in the acute phase, most noticeably in the high supplemented group. Similarly, Wassink et al. (1997) showed a significant depression of DMI due to *T. congolense* infection in lambs fed on a high plane diet, whilst Reynolds and Ekwuruke (1988) described similar declines in feed intake due to *T. vivax* infection in Djallonké sheep. Because intake varied between individuals ($p<0.001$), pre-infection intake levels were accounted for in the analyses. Other studies have reported trypanosome induced depression of feed intake in the order of 15% in small ruminants and have linked this to decreased live weight gain (Verstegen et al., 1991; Akinbamijo et al., 1994). In addition to the reduced intake, the latter studies also gave evidence of increased maintenance requirements due to infection, which led to a decreased nitrogen and energy retention and therefore reduction of the weight gain. Trypanosome infection also changes energy and nitrogen metabolism in the host because of the induced fever (Verstegen et al., 1991) and catabolism of immunological products (Nielsen et al., 1978).

The retarded growth rates in the HI group, as compared to the HC group, could be partly related to the reduced DMI shortly after infection. Between LC and HC groups there was an overall difference in total weight increase of 4.3 kg, due to a difference in DMI of 230 g (diet effect). It would be difficult to accept that the transitory drop in DMI for the HI group of 28±6 g would cause such depressed growth rates in the HI groups. Although the reduction in feed intake complicates the study of the effects of infection on energy and protein metabolism in the host, the present results suggest an increased metabolic cost due to infection. In contradiction, however, DMI was apparently not depressed in the LI group and total weight increase was not so much lower than in the LC group. Presumably the offered ration for the low-diet groups was so low that intake could not be further reduced by infection. Experimental *T. congolense* infection did also not affect live weight patterns in adult Djallonké ewes when not supplemented (Osaer et al., 1994), nor the growth rates of Scottish blackface lambs, which were similar to their pair-fed controls (Wassink et al., 1997). The latter observations did not show evidence for increased metabolic cost due to infection.

The ewe lambs were generally quite old at puberty with an important variation among individuals. Whereas trypanosome infection did not have any significant effect, high supplementary feeding significantly reduced the age at first cycling. Berger and Ginistry (1980) reported that the age at first oestrus in the Djallonké sheep was around 8 months with a corresponding live weight of 16.3 kg. Only the HC group attained puberty at this target age in the present experiment, compared to 11 months in groups LC and LI. Since age at puberty is highly correlated to body weight, these results are in complete agreement with group live weight pattern.

Age at first lambing was similarly influenced by the diet ($p<0.05$), but there was also a tendency for trypanosome infection to delay age at first lambing. Effects of diet and infection were just additive. Other reports show that age at first lambing in Djallonké ewes can range from 451.4±23 days in the typical Gambian village situation (Osaer et al., 1998, unpublished results) to 456 days in Côte d'Ivoire (Ivory Coast) and to 524 days in Benin (FAO, RAF/88/100, 1993). The mean age for the control groups in the present study (574.8±14.5 days) is higher than the other observations and maybe attributed to the confinement of the animals.

Birth weights and growth rates of lambs born out of the experimental groups were neither significantly affected by trypanosome infection, nor by the diet of the dam. In contrast, lamb mortality up to 3 months was significantly increased by infection and most lamb deaths arose in group LI. There is much similar evidence of the effects of trypanosome infection of pregnant ewes being postponed to affect offspring survival rather than pregnancy per se. Thus, when *T. congolense* infection was arranged at the time of breeding in Djallonké ewes, there were neither effects on lamb birth weight nor on lamb survival, but there was a significant interaction between diet and infection resulting in the poorest lamb growth rates in the low supplemented infected ewes (Osaer et al., 1998). Similarly, birth weights and initial growth rates of lambs derived from ewes with natural trypanosome infections, whether the infection occurred in the first term or second term of pregnancy, or during the early post-partum period, were not affected (Osaer et al., 1998, unpublished observations). Reynolds and Ekwuruke (1988) also found no different growth rates in lambs born to infected Djallonké ewes fed either on maintenance or submaintenance. In contrast, during the first year after experimental *T. congolense* infection, productivity of Djallonké ewes was significantly reduced by infection in terms of ability to nurse lambs (lamb growth rate) and higher lamb mortality (Goossens et al., 1997b). These contrasting results on lamb performance may be partly explained by the interval between infection in the dam and parturition. In the present study, trypanosome infection persisted in all ewes until the chronic phase whilst negative effects due to infection remained. However, whenever ewes were confirmed pregnant they were treated with trypanocidals so by the time parturition occurred all ewes were free of trypanosomes.

5. Conclusions

In conclusion, the results show that whereas trypanosome infection only tended to delay puberty and age at first lambing directly, the effects were more indirectly mediated through depression of weight gain. Interaction between the effects of trypanosome infection and diet were observed in, for example, the high supplemented infected group having depressed weight gain and reduced DMI, but at the same time a better haemopoietic response and better lamb survival rate compared to the low supplemented infected group. Depression of PCV and intensity of parasitaemia following trypanosome infection were similar at both low and high levels of nutrition. Whether an adequate feeding regime would be always beneficial in trypanosome-infected ewes is not clear since large part of the extra feed were used to compensate for infection. Further work should give clarification. The results undoubtedly showed the delaying effect of a low dietary level on onset of reproductive activity and on reproductive performance in young Djallonké sheep.

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4.4
Some biochemical changes following *Trypanosoma congolense* infection in Djallonk ewe lambs and breeding ewes fed on two levels of nutrition.

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SOME BIOCHEMICAL CHANGES FOLLOWING *TRYPANOSOMA CONGOLENSE* INFECTION IN DJALLONKÉ EWE LAMBS AND BREEDING EWES FED ON TWO LEVELS OF NUTRITION.

Osaer S., Akinbamijo O.O. and Goossens, B.

Abstract

The effects of artificial *Trypanosoma congolense* infection and dietary level on biochemical changes were observed in 24 ewe lambs (exp. I) and 42 breeding ewes (exp. II). For both experiments, there were four treatment combinations, of which two were kept on a restricted diet (L), the other two on an at libitum diet (H). Half of each dietary group was infected with *T. congolense* (LI, HI), while the remainder served as uninfected controls (LC, HC). Artificial *T. congolense* infection took place at the age of 200 ± 7 days in experiment I and at the peak of oestrus in experiment II. Irrespective of dietary levels offered, total proteins in lambs and ewes and albumin in lambs declined significantly ($P < 0.001$) post infection. Plasma glucose concentration was reduced by the low dietary level and not by infection. Although plasma urea concentrations were slightly increased in the infected ewe lambs, adult ewes in the HI group demonstrated increased plasma urea concentrations ($P < 0.05$) due to an interaction between infection and diet. Neither infection nor the imposed diet induced significant changes on plasma creatinine concentrations. Transitory peaks in non-esterified fatty acids (NEFA) and beta-hydroxy butyric acid (BHBA) levels in infected ewes on low dietary level indicated temporary changes in the energy metabolism of the host.

It was concluded from this study that, inspite of their trypanotolerance, Djallonke lambs and ewes demonstrated an infection effect on host metabolism pattern due to *Trypanosoma congolense* infection. These changes reflected to some extent trypanosome-induced alteration of the nutrient metabolism, which could not always be negated by diet supplements. Nutrition, as an independent factor, did confer added benefits against the debilitating effects of trypanosomosis under the conditions of the present study.

Introduction

Trypanosomosis is a major constraint for livestock production in sub-Saharan Africa. Consequently, only the trypanotolerant breeds can survive, reproduce and remain productive in tsetse infested areas without the aid of trypanocidal treatment (Murray et al., 1982). The Djallonke breed of sheep is considered trypanotolerant due to an inherent ability to maintain production under trypanosomosis although the mechanism of this trait seems to differ from that in cattle (Mawuena, 1986, 1987; Osaer et al., 1994, 1997; Goossens et al., 1997b). However, intermediary metabolic disorders have been reported in cattle (Akinbamijo et al 1997, Romney et al 1997) and small ruminants (Verstegen et al 1991, Zwart et al 1991, Akinbamijo et al 1992, van Dam et al. 1996, 1998, Wassink et al 1997). Typical biochemical events associated with trypanosomosis include increased maintenance requirements, changes in the concentration of the serum metabolites and associated haemostatic conditions, culminating in a compromised energy and nitrogen retention in the host.

The patho-physiological changes due to trypanosomosis in the host are mediated by the nutritional status of the host. Earlier studies have shown that high protein and/or energy supplements can ameliorate the adverse effects arising from *T. congolense* infection in Scottisch Blackface sheep (Katunguka-Rwakishaya et al., 1993;1995). Similarly, following *T. congolense* infection in pregnant Djallonké ewes, haematopoietic responses and productivity parameters were improved at unrestricted dietary levels as compared to low dietary conditions (Osaer et al., 1998a). As most of the trypanosome-induced metabolic disorders are expressed at the post-absorptive phase (Akinbamijo et al., 1992; Van Dam et al., 1996, 1998), the present authors chose to further investigate the changes that occur in this phase. As the two experiments were carried out consecutively under similar on-station conditions, using the same trypanosome strain and the same breed, data on biochemical changes from both experiments are described in this paper. In addition, differences in age or physiological conditions such as early pregnancy may interact with resistance to trypanosomosis and resulting biochemical/haematological alterations (Osaer et al., 1998a,1998b).

The present study is therefore aimed at evaluating the effects of *T. congolense* infection and level of nutrition on some serum biochemical parameters in both young and adult ewes at different physiological conditions.

Materials and Methods

The study was conducted on-station at the coastal site of the International Trypanotolerance Centre (ITC), The Gambia, West Africa. The climate is sub-humid with a mean annual rainfall of approximately 1000mm. Based on entomological and pathological records, the site is generally considered to be at no risk of tsetse challenge (W. F. Snow, personal communication).

Experiment 1:

Animals and Husbandry

Twenty-four ewe lambs were selected from the ITC breeding flock at their respective weaning age of 4 months and average live weight of 10 kg. A randomised complete block design was used and lambs were allocated into blocks based on their date of birth. Within blocks, lambs were randomly assigned to one of four treatment combinations: infected high level of nutrition (HI), control high level of nutrition (HC), infected low level of nutrition (LI) and control low level of nutrition (LC). The ewe lambs were tethered during feeding time to allow daily feed intake measurements. The diet composed of groundnut hay (GNH), groundnut cake (GNC) and rice bran (RB) in a 5/2/1 ratio, and this ratio was similar for both low and high level. This ration had a dietary composition of 10,69 Mj ME/ kg DM (book value) and 22 % Crude Protein (CP) with a metabolizability of 0.55 (Q). Animals on the low level of nutrition were offered a restricted amount of the diet at the start of the trial, corresponding to 3.7 Mj ME and 66 g CP, sufficient for maintenance and daily growth of 50 g for a 10 kg lamb (McDonald et al., 1978; Agricultural and Food Research Council (AFRC), 1993). On the other hand, the high diet groups were offered from the same ration but higher amounts (=350 g GNH/ 140 g GNC/ 70 g RB) with initially 5.7 Mj ME and 115 g CP, which would allow growth rates above 50 g/d (McDonald et al., 1978; AFRC, 1993). During the experiment, rations were adjusted for changes in live weight, while maintaining the same composition of the diet and the difference between high and low. All ewe lambs were confined in one group and placed on their respective diet five weeks before trypanosome infection. Individual dry matter intake (DMI) was measured daily and live weights were recorded weekly with a Salter spring balance. Prior to inoculation, all animals were confirmed free of trypanosomosis using the darkground (DG) buffy coat technique (Murray et al., 1977). Routine flock health procedures included fenbendazole treatment and vaccination against peste des petits ruminants (Tissue Culture Rinderpest vaccine, ISRA/LNERVE, Dakar, Senegal), and clostridial infections (Covexin, (Pittman Moore), Mallinckdrodt, Uxbridge, UK).

Trypanosome infection

On attainment of 200 ± 7 days of age, 12 animals belonging to the HI and LI group, were inoculated with a pathogenic cloned stock of the *T. congolense* ITC 84, derived from a goat at Kunting on the north bank of the River Gambia (Central River Division, The Gambia). The clone was expanded in mice and the lambs were inoculated intradermally with 1ml containing 10^4 bloodstream forms at five sites on the left flank. The remaining groups (LC, HC) with 12 animals each served as uninfected controls.

Measurements

Following infection, plasma was prepared from the weekly blood samples, collected into Lithium Heparin-coated vacutainer tubes (2-3 ml) during 11 weeks post-infection to assess concentrations of glucose, total protein, urea, creatinine and albumin with 'dry chemistry' kits (Johnson & Johnson Vitros slides).

Experiment 2:

Animals and Husbandry

Forty-two cycling adult open ewes were randomly selected from the ITC breeding flock. The trypanosome-free ewes were subjected to routine flock health checks and procedures described in experiment 1. A randomised complete block design was used in which ewes were divided into blocks based on their live weight at the start of the experiment, followed by allocation to one of the four treatments as described for Expt 1: HI (n =10), HC (n = 10), LI (n=11) and LC (n=11). The diet consisted as in expt. 1 of GNH, GNC and RB but with a different ratio of 5/1/2, which was kept similar for low and high level. The ration had a composition of 10.52 Mj ME/ kg DM (book value) and 14 % Crude Protein (CP) with a metabolizability of 0.55 (Q). Ewes in the high dietary group were offered 580 g GNH, 200 g RB and 120 g GNC, which provided 120 g CP and 9 Mj ME per animal/day, whereas the low level diet offered 450 g GNH, 150 g RB and 90 g GNC, which provided 92 g CP and 7 Mj per animal/ day. The low dietary level was calculated to be sufficient for maintenance and requirements for early pregnancy (=5.5 Mj ME and 60 g CP), calculated for an ewe of 25 kg (AFRC, 1993). All ewes were placed on their respective diet eight weeks prior to commencement of the experiment and feed was offered per treatment group. Live weights were recorded weekly.

Trypanosome infection and breeding

In order to synchronise breeding with peak parasitaemia, all ewes were oestrus synchronised using a 12-day treatment with an intra vaginal progestagen sponge (Veramix, Upjohn, Crawley, UK). This was followed by an injection of serum gonadotrophin after sponges were withdrawn (PMSG-Intervet, 500 IU/ewe, Intervet, Cambridge, UK). The parasite inoculation was administered to HI and LI ewes in a way that the peak of parasitaemia would coincide with oestrus, which was expected within 48 to 72 hours following sponge withdrawal. Peak parasitaemia in the strain is well documented from previous experiments (Osaer et al., 1994; 1997; Goossens et al., 1997a). On the day of expected oestrus, rams were introduced into each of the treatment groups.

Measurements

Plasma was stored serially for nine weeks post-infection from blood samples collected into Lithium Heparin-coated vacutainer tubes (2-3 ml). Biochemical assays on parameters of interest included urea, total protein, non-esterified fatty acids (NEFA) and beta-hydroxy butyric acid (BHBA) using commercial kits (Randox laboratories).

Statistical Analyses

Statistical analyses were carried out using SAS-statistical package version 6.12 (1998). Biochemical parameters were analysed by the general linear model (GLM procedure) as repeated measurements using a mixed model. The following main effects were included: infection (control vs infected), diet (low vs high), block, period (acute (week 0 to 5), post acute (week 6 to 11)), week (nested within period), ewe (nested within treatment, diet and block), and the interactions between infection and diet; infection and period; diet and period; and infection, diet and period. Considering the design of the experiment, joint effect of infection and nutrition was of high interest and tested in the model by the interactions between infection and diet and infection, diet and period. The animal effect was regarded as random. All hypotheses were subjected to the F-test (Snedecor and Cochran, 1980). Differences in defined interactions are described in order to estimate the effects of trypanosome infection and/or the diet for the different periods. Means of the different groups are reported as least square means ± standard error.

Results

General

In experiment 1, no obvious clinical symptoms of trypanosomosis were noticed among the infected lambs and there was no mortality due to trypanosomosis. All animals of the LI and HI group were positive for trypanosomes by week 2 post infection. Despite the initial higher parasitaemia score in group HI, no significant differences in parasitaemia level were found between diets. Mean dry matter intake (DMI) and live weight gains for the four treatment groups during the observation time are presented as arithmetic means (sd=σ) in table 1. The young ewes in the LC group had a mean growth rate of 12 g/d and a mean intake of 3.5 Mj and 73 g CP. The HC group gained an average of 27 g/d with a mean intake of 5.9 Mj and 122 g CP. The effects of trypanosome infection, diet and their interaction on DMI level and live weight gain have been described in more detail earlier (Osaer et al., 1998b). Infected animals on low (LI) and high (HI) dietary regimes grew on average 7 and 25 g/d, respectively.

Table 1: Dry matter intake (DMI) levels (expt. I), mean live weight (LW) and weight changes for the treatment groups LC, LI, HC and HI during an observation period of respectively 11 (expt. I) and 12 weeks (expt. II).

	Expt. I: ewe lambs			Expt. II: breeding ewes	
group	DMI (σ)	LW (σ)	weight gain	LW (σ)	weight gain
LC	325.3 g (18.8)	10.9 kg (0.3)	0.9 kg	20.8 kg (0.3)	-0.5 kg
LI	328.8 g (15.4)	11.6 kg (0.2)	0.5 kg	20.7 kg (0.6)	-1.4 kg
HC	552.5 g (19.7)	13.3 kg (0.7)	2.1 kg	23.7 kg (0.4)	0.4 kg
HI	522.4 g (40.6)	11.2 kg (0.6)	1.9 kg	25.0 kg (1.0)	1.7 kg

In experiment 2, two ewes from group HI and one from group LI died as a result of trypanosomosis during the acute phase of infection. Trypanosomes were never detected in five ewes from group LI and four from group HI. These ewes were consequently omitted from further analyses. Similar to the first trial, dietary level had no influence on parasitaemia level. Live weight gain for the observation period are presented for the four groups in table 1. Although data on individual DMI are not available since there was group feeding, no refusals from either treatment group were observed during the trial. Thus, mean daily DMI levels for the ewes were 650 and 850 g/day for ewes on low and high dietary regimes, respectively. Mean weight changes in LC and LI groups for the observed time were 6 and -17 g/day respectively, while those in the HC and HI groups were 5 and 20 g/day, respectively. Effects of infection, diet and their interactions on live weight changes have been analysed for a longer period (=gestation length) and described in detail (Osaer et al., 1998a).

Plasma Biochemistry

1. Total protein

In experiment 1, infected animals showed a significant decline in total protein level, with mean levels of 61.9 ± 0.8 g/l for control groups and 57.4 ± 0.8 g/l for infected groups ($P<0.01$). Although the trend was reversed in the subacute phase of infection, the concentration of plasma total protein remained significantly lower in infected animals when compared with control groups (see Fig. 1a). In contrast, neither the dietary level nor its interaction with infection had significant influence on plasma protein. Thus, the decrease due to infection was not different for LI and HI groups although a high degree of individual variation in total protein was observed ($P< 0.05$).

Total protein pattern in *experiment 2* was not different from the preceding trial. Infection caused a decline in plasma protein with mean values of 94.2 ± 0.8 g/l and 88.1 ± 1.1 g/l for control and infected groups respectively ($P<0.01$) (Fig. 1b). Similarly, dietary effect on plasma protein level and the difference in plasma protein concentration between LI and HI did not attain statistical significance.

2. Plasma albumin concentration (Expt I)

Trypanosome infection caused a significant decline in the plasma albumin concentration with a difference in overall mean of 2.7 ± 0.4 g/l ($P<0.001$) between control and infected groups (see Fig. 2). Dietary effect on albumin levels was not significant nor was its interaction with infection, so effects of diet and infection were additive. Mean plasma albumin concentrations for groups HC, HI, LC and LI were 29.3 g/l, 25.5 g/l, 27.6 g/l and 26.1 g/l (pooled s.e.: 0.4) respectively, with considerable individual variation in albumin concentration.

3. Plasma creatinine concentration (Expt I)

Group mean concentration of plasma creatinine on a weekly basis are presented in Figure 3. Trypanosome infection did not cause any significant changes in creatinine level during the 11 weeks post infection. In contrast, low diet groups tended to show higher plasma creatinine levels ($P<0.06$) than those on high dietary level. In addition, a significant ($P<0.01$) interaction between diet and infection was observed resulting in higher concentrations for groups LI and HC. Plasma creatinine concentrations for groups HC, HI, LC and LI were 56.58 μmol/l, 48.62 μmol/l, 51.27 μmol/l and 58.34 μmol/l (pooled s.e. 0.01) respectively. Individual variation in creatinine concentration ($P<0.01$) was observed in the trial.

4. Plasma glucose concentration (Expt I)

Trypanosome infection did not influence plasma glucose concentration. In contrast, differences between dietary levels were observed in the post acute phase but did not attain statistical significance ($P< 0.06$), with means for low and high diet groups of 2.63 ± 0.11 mmol/l and 2.84 ± 0.11 mmol/l, respectively. Although the LI group had the lowest value observed especially in the post acute phase (Fig. 4), nevertheless, the interaction of diet, infection and period was not significant. The overall means for the four groups were as follows (HC: 2.84 mmol/l; HI: 2.86 mmol/l; LC: 2.86 mmol/l; LI: 2.78 mmol/l; pooled s.e.:0.09).

5. Plasma urea concentration

The weekly group arithmetic means for plasma urea concentration are as shown in figures 5a and 5b. In *experiment* 1, infection tended to increase the urea concentration with mean levels for infected and control groups of 11.01 ± 0.3 mmol/l vs 10.51 ± 0.3 mmol/l respectively. However, the dietary effect was significant ($P< 0.05$) with mean concentrations of 11.46 ± 0.3 mmol/l for high diet and 10.07 ± 0.3 mmol/l for the low diet groups, reflecting the higher N intake in the high diet groups. The interaction of diet and infection was not significant, thus effects of diet and infection were additive.

In *experiment 2,* trypanosome infection did not seem to affect the level of peripheral urea. The interaction of diet and period indicated significant higher ($P<0.01$) urea levels in the high diet groups during the first five weeks of infection. In addition, there was a significant interaction between diet and infection ($P<0.05$): the HI group indicated significant higher levels of plasma urea compared to other groups (HC:12.5 mmol/l; HI: 14.6 mmol/l; LC: 13.1 mmol/l ; LI: 12.5 mmol/l; pooled s.e.:0.3).

6. Non- esterified fatty acids (NEFA) plasma concentration (Expt II)

Weekly group levels of NEFA are presented as arithmetic means in figure 6. Despite a transitory increase at week 2 for the LI group, neither trypanosome infection nor diet significantly influenced plasma NEFA concentrations during the trial. As from week 7 onwards, there was an increase in the observed values for the LC group.

7. Beta-hydroxy butyric acid (BHBA) plasma concentration (Expt II)

A sharp increase in BHBA levels in week 3 and a subsequent decrease in week 4 was observed for group LI, although weekly averages for the remaining groups were fluctuating considerably throughout the study period (see Figure 7), reflected by period and week (within period) effects ($P<0.001$). Neither infection nor diet influenced BHBA levels significantly in the present study.

Discussion

Metabolic responses under pathologic conditions are influenced by a broad spectrum of factors. Hence, the evidence presented by the experiments only provide useful indications of nitrogen and energy metabolism through the metabolic profile of infected and non-infected animals. In the present study, changes in blood biochemical parameters in young and adult Djallonke ewes following *T. congolense* infection were not drastic. Further, interactive effects between trypanosome infection and diet were absent except for the changes in the concentrations of creatinine in the young ewes and urea in the adult ewes, the other parameters being influenced independently.

Nitrogen metabolism

The major changes induced by trypanosome infection were a decline in total protein level in both young and adult ewes together with a decrease in albumin concentration in the ewe lambs. Decrease in albumin due to parasitic infections has also been reported in ovine haemonchosis (Abott et al, 1986) and in both single and serial infections of *T. congolense* and *Haemonchus contortus* (Goossens et al., 1997a). The present findings agree with those in susceptible breeds of sheep (Katunguka-Rwakishaya et al., 1993, 1995; Wassink et al., 1997) and cattle (Wellde, 1974) following trypanosome infection. In the present experiment, reductions in total protein and albumin due to trypanosome infection were independent of the dietary regime of the host. However, animals on high dietary level tended to increase albumin levels but this difference was mainly observed in uninfected ewe lambs. In contrast, Katunguka-Rwakishaya et al. (1993, 1995) found that protein supplements reduced the rate of decline of albumin post-infection in much the same manner as high energy levels did of plasma total protein. This was also indicated by Wassink et al. (1997) who observed a significantly higher dietary effect compared with infection effects on albumin level while both effects acted in an additive manner. Romney et al. (1997) found reductions in albumin due to both *T. congolense* infection and low dietary levels in trypanotolerant N'Dama cattle. The overall mean protein and albumin concentrations in the infected ewe lambs were close to the minimum level of the reference values in sheep (Total Protein: 56-78 g/l; Albumin: 24-37 g/l) (Johnson & Johnson, 1993; Kaneko J., 1989). This metabolic response might have influenced tissue accretion and general protein metabolism pattern in the young infected animals. Although the protein levels observed in the infected adult ewes were also significantly reduced compared to their control counterparts, the values were higher than those observed in the young ewe lambs. Consequently, the age factor has an important role in the interpretation of serum proteins. In general, we observed an increase in total protein, globulins and decrease in albumin with advancing age, as well as physiological conditions such as pregnancy which may influence these parameters (Kaneko, 1989). The reduction in albumin relative to total protein level suggest in part the commencement of the synthesis of immunoglobulins and the initiation of the immune response (Katunguka-Rwakishaya et al., 1992). However, the protein:albumin index for the young ewes did not alter during trial for any of the treatment groups and this ratio was within normal physiological range of 35-50% in all groups (Kaneko, 1989). Haemodilution has been suggested to be associated with trypanosome infection and could therefore explain partly the declined albumin and total protein concentrations (Anosa & Isoun, 1976; Katunguka-Rwakishaya et al., 1992; Osaer et al., 1998b).

The significant increase in urea levels due to higher supplements (diet effect), are a reflection of the higher N intake in these groups, with a surplus of about 50 g/day CP in the lambs and a difference of 30 g/day CP in the breeding ewes. Previous studies have confirmed the negative nitrogen and energy retention coupled with a high peripheral urea concentration as a result of increased body protein catabolism in response to an increased N demand during infection (Akinbamijo et al., 1994; Van Dam et al., 1996; Verstegen et al., 1991). The results presented in this paper are in agreement with those presented in trypanosome-infected small ruminants that manifest a negative N-balance post-infection . The low plasma total protein and albumin levels induced by infection, combined with a tendency to increase plasma urea levels, suggest a disorder in the nitrogen metabolism pattern as widely reported in trypanosomosis in the literature (Akinbamijo et al., 1994, Verstegen et al., 1991). Based on the plasma creatinine concentration results, the study further indicated that neither the severity of infection nor the imposed diet induced severe muscle degeneration. The interaction between diet and infection showed higher levels for the LI and HC groups. However, these were still near or below the minimum of the reference values in sheep (53-133 μmol/l; Johnson & Johnson, 1993), thus confirming absence of muscle degradation as earlier pointed out in the literature by Akinbamijo et al (1992).

Energy metabolism

In the ewe lambs, low plasma glucose concentration was a reflection of the dietary allowances of the low diet groups and this became emphasised at the onset of trypanosome-induced anorexia in the post acute phase (Osaer et al., 1998b). The mean level for the low diet groups (2.63 $\pm$ 0.11 mmol/l) was also below the normal range in sheep (2.78-4.44 mmol/l). Additive effects of diet and infection, the latter effect not being significant but with a declining tendency, resulted in even lower values for the LI group (2.54 ± 0.14 mmol/l). Based on these low glucose values, and coupled with the metabolic cost of infection, it is not unusual for a shift in the host metabolism process towards endogenous catabolism leading to a state of negative nutrient balance in animals on low dietary regime. Evidence of the latter hypothesis was observed in the second trial. Although the plasma NEFA and BHBA levels in the adult ewes were not significantly affected by infection, the transient sharp increase observed in the LI group in the acute phase of infection supports the hypothesis of lypolysis as an immediate response to post-infection increased energy demands. The initial trauma of the pre-patent period must have induced a condition of temporary ketogenesis in infected ewes. The observations on nutrient metabolism are corroborated by the live weight patterns in both trials. Live weight gains were significantly depressed due to trypanosome infection in the chronic phase (20-40 weeks p.i.), as reported earlier (Osaer et al, 1998b). Similarly, the positive effect of high supplementation on weight changes was depressed by infection, which became obvious in the chronic phase of the trial (Osaer et al., 1998a). Apart from trypanosome- induced reduction in dry matter intake measured in the first trial (Osaer et al., 1998b), an additional metabolic cost of infection was assumed to play a significant role in the nutrient metabolism of infected sheep. This is in agreement with a previous study in Djallonke sheep (Akinbamijo et al.,1994) following *T. vivax* infection. Trypanosome infection changes the nitrogen and energy metabolism in the host because of the induced fever (Verstegen et al., 1991), increased hematopoiesis, production and catabolism of immunological products (Nielsen et al.,1978).

However, since there was a lower rate of weight gain in the infected animals in both trials, it was assumed that the exogenous energy and protein sources were sufficient to cover maintenance requirements while requirements for growth/weight gain were already negated by the increased metabolic cost of trypanosome infection and trypanosome-induced anorexia. Nevertheless, it was observed in both trials that growth rates/weight changes in the non-infected ewe lambs and breeding ewes were much lower than expected from their ME and CP intake levels. It has been observed earlier that Djallonké sheep do not respond with the expected gain to a given ration, which covers growth or production requirements based on AFRC standards (Van Winghem J., pers. comm). This may be due to the genetic composition of the animals (dwarf breed) or lower digestibility of the ration, as values less than 0.55 are not uncommon with the ration used (Jarrige, 1989).

Finally, from the observed changes in biochemical parameters, it was concluded that inspite of their trypanotolerance, a trypanosome-induced alteration of the nutrient metabolism was present in both young and adult Djallonke ewes irrespective of the dietary status. However, since the effect of nutrition was mostly acting as a stronger, independent factor, it conferred added benefits against the debilitating effects of trypanosomosis under the conditions of the present study.

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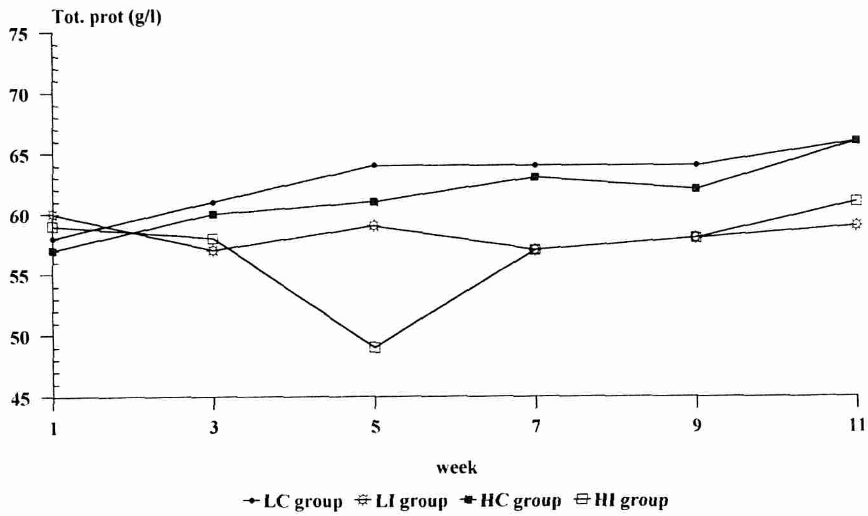

Figure 1a: Mean plasma Total protein level in the young ewes for groups high control (HC), low control (LC), high infected (HI) and low infected (LI) during 11 weeks post infection.

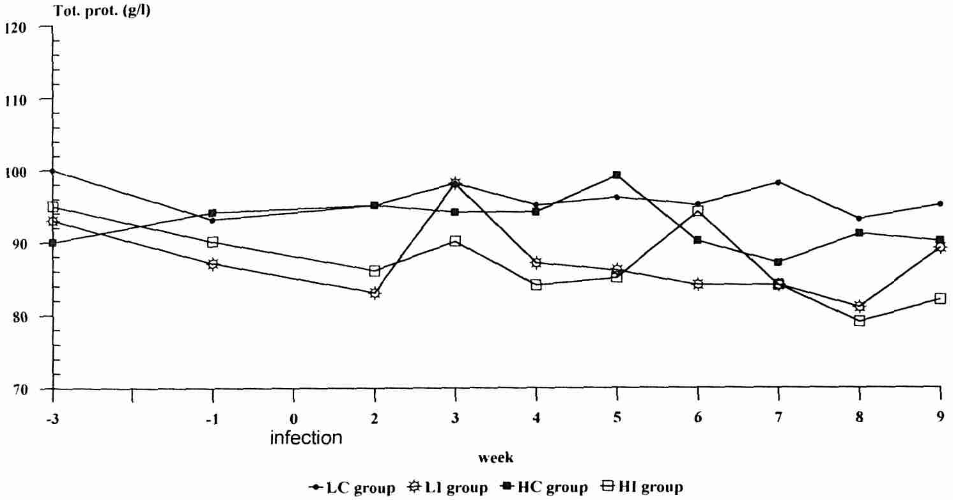

Figure 1b: Mean plasma Total protein level in the adult ewes for groups high control (HC), low control (LC), high infected (HI) and low infected (LI) until 9 weeks post infection.

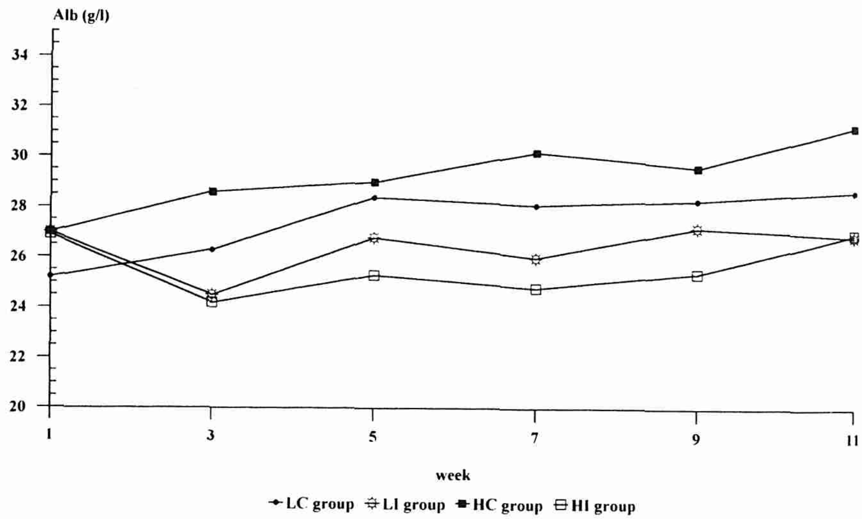

Figure 2: Mean plasma Albumin level in the young ewes for groups high control (HC), low control (LC), high infected (HI) and low infected (LI) during 11 weeks post infection.

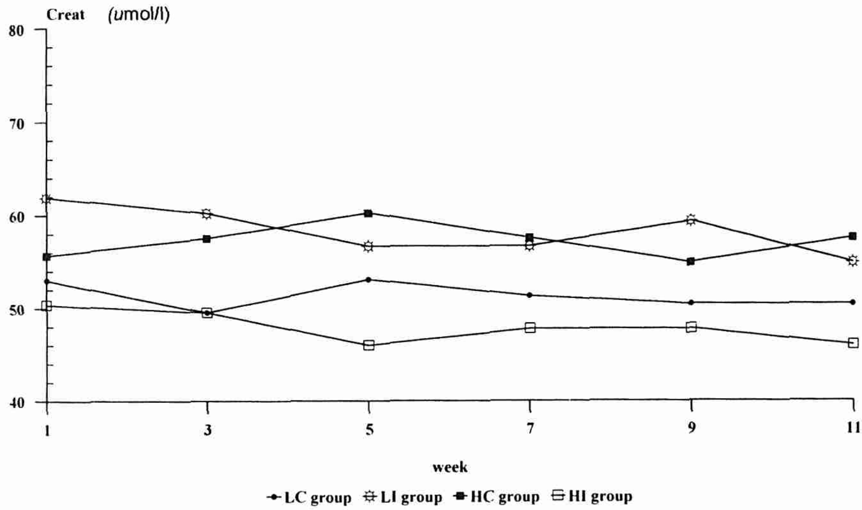

Figure 3: Mean plasma Creatinine concentration in the young ewes for groups high control (HC), low control (LC), high infected (HI) and low infected (LI) during 11 weeks post infection.

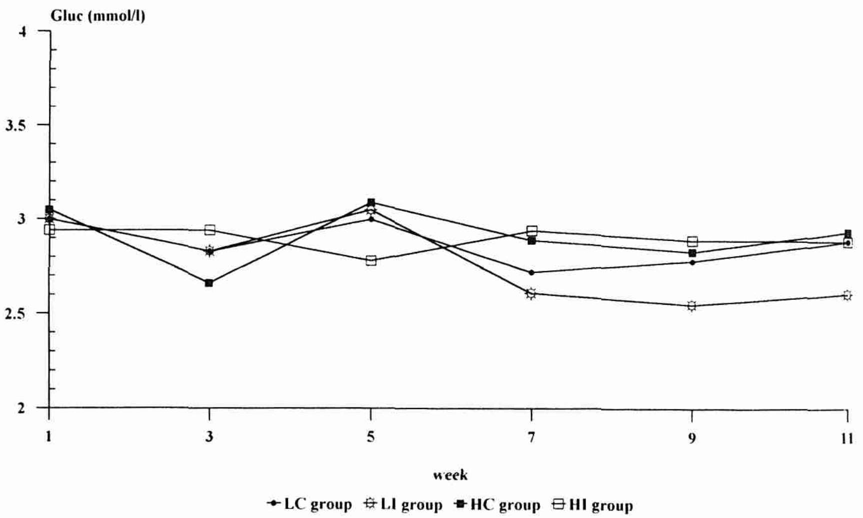

Figure 4: Mean plasma Glucose level in the young ewes for groups high control (HC), low control (LC), high infected (HI) and low infected (LI) during 11weeks post infection.

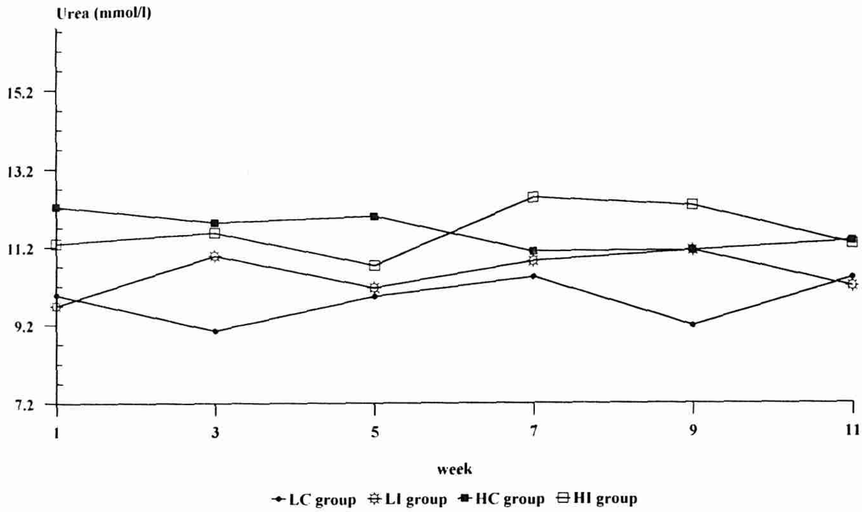

Figure 5a: Mean plasma Urea concentration in the young ewes for groups high control (HC), low control (LC), high infected (HI) and low infected (LI) during 11weeks post infection.

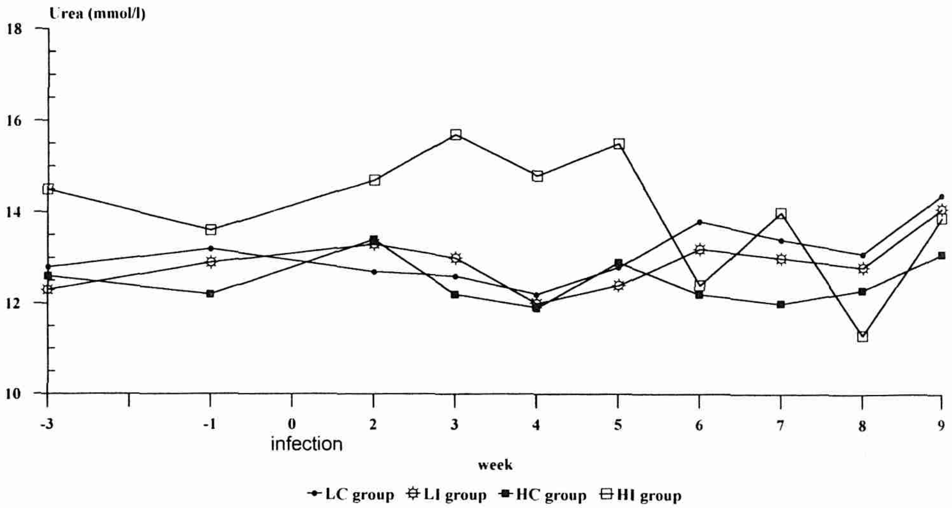

Figure 5b: Mean plasma Urea concentration in the adult ewes for groups high control (HC), low control (LC), high infected (HI) and low infected (LI) until 9 weeks post infection.

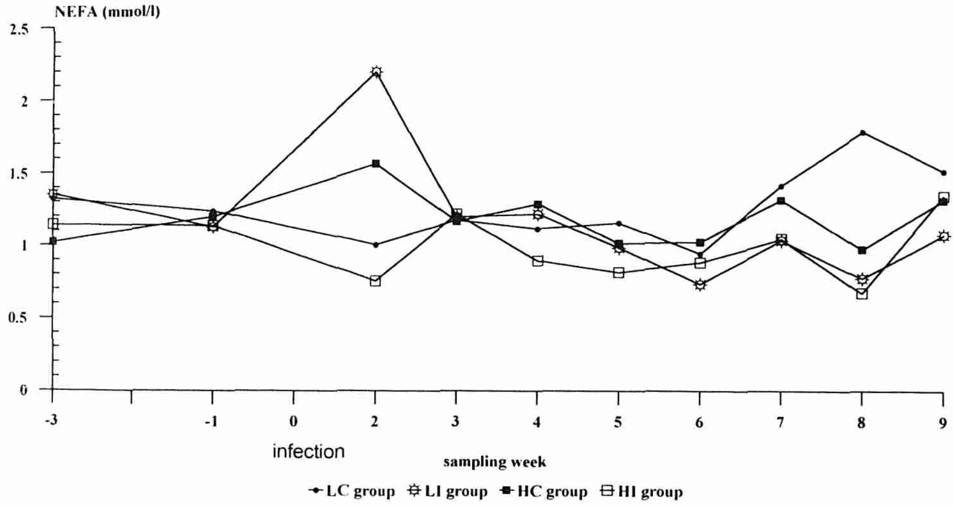

Figure 6: Mean plasma NEFA level in the adult ewes for groups high control (HC), low control (LC), high infected (HI) and low infected (LI) until 9 weeks post infection.

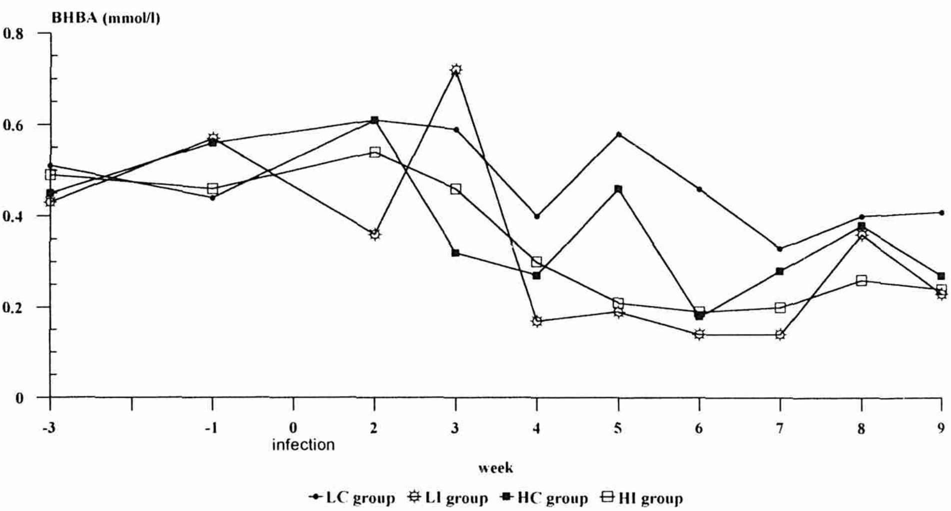

Figure 7: Mean plasma BHBA level in the adult ewes for groups high control (HC), low control (LC), high infected (HI) and low infected (LI) until 9 weeks post infection.

CHAPTER 5

INTERACTION OF HELMINTH INFECTION AND TRYPANOSOME INFECTION IN TRYPANOTOLERANT SMALL RUMINANTS

5.1

The interaction of *Trypanosoma congolense* and *Haemonchus contortus* infections in trypanotolerant Djallonke sheep.

Goossens, B., Osaer, S., Kora, S., Jaitner, J., Ndao, M. and Geerts, S. 1997.
International Journal for Parasitology, 27: 1579-1584.
(Manuscript in the thesis is the version used as basis for the above 'Research Note').

5.2

The effects of prophylactic anthelmintic treatment on the productivity of traditionally managed Djallonke sheep and West African Goats goats kept under high trypanosomosis risk.

Osaer, S., Goossens, B, Eysker, M. and Geerts, S.
Acta Tropica, In Press.

5.1
The interaction of *Trypanosoma congolense* and *Haemonchus contortus* infections in trypanotolerant Djallonke sheep.

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(Manuscript in the thesis is the version used as basis for the above 'Research Note').

THE INTERACTION OF *TRYPANOSOMA CONGOLENSE* AND *HAEMONCHUS CONTORTUS* IN TRYPANOTOLERANT DJALLONKE SHEEP.

B. Goossens, S. Osaer, S. Kora, J. Jaitner, M. Ndao and S. Geerts

SUMMARY

The interaction between *Trypanosoma congolense* and *Haemonchus contortus* was studied in Djallonké sheep using artificial infections. Five groups of 8 Djallonké sheep were used. Two groups received a single infection with either *H. contortus* or *T. congolense*, two groups were infected with both pathogens but in different sequence : *T. congolense* followed by *H. contortus* (TcHc group) and vice versa (HcTc group). One group was kept as uninfected control.

Mortality due to infection was only observed in the dual infection groups. In the TcHc group, the effects were more acute whereas in the HcTc group they were more of a chronic nature as judged by the haematology parameters (packed red cell values PCV, haemoglobin Hb, red blood cells RBC) and the parasitological parameters (eggs per gram faeces EPG and parasitaemia score). No significant differences in weight gain could be demonstrated between infected and control groups.

Djallonké sheep are able to withstand a single infection with either *T. congolense* or *H. contortus* which confirms their trypanotolerant nature and provides some preliminary indication of resistance against helminth infections. However, when exposed to successive infections with both parasites, some of the animals loose the capacity to control the pathogenic effects.

INTRODUCTION

Despite important efforts, trypanosomosis is still a major constraint for livestock production in Sub-Sahara Africa. Anaemia is a predominant symptom and a reliable indicator for the severity of a trypanosome infection (Anosa & Isoun, 1976; Griffin, 1978; Anosa, 1988a & b). In addition, immunosuppression has been associated with trypanosome infected animals which become more sensitive to secondary infections (Urquhart *et al.* 1973; Mackenzie, 1975; Nantulya *et al.* 1982). Also gastrointestinal helminths are regarded as important anaemia-causing pathogens especially *Haemonchus contortus* through its bloodsucking stages in the abomasum. From all parasitic nematodes *H. contortus* is found to be the most common and the most pathogenic for ruminants in Sub-Sahara Africa (Vercruysse, 1983; Preston & Allonby, 1979; Vassiliades, 1981; Fritsche, Kaufmann & Pfister, 1993). Under natural conditions, trypanosomosis and helminthosis often occur in mixed infections.

Djallonké sheep are known to be trypanotolerant (Griffin & Allonby, 1979a, b; Murray, Morrison & Whitelaw, 1982; Mawuena 1987, Osaer *et al.* 1994) but stress factors like poor nutrition or high worm burdens will negatively influence the expression of trypanotolerance. Resistance to trypanosomosis can be of innate or acquired origin and there is evidence that both exist in the Djallonké breed (Bengaly *et al.* 1993; Osaer *et al.* 1994). However, to profit as much as possible from the genetical resistance, intercurrent stress factors should be kept to a minimum.

Few studies have been undertaken to examine the interaction between trypanosomes and helminth infections. The interaction of *T. congolense* and *H. contortus* has been studied in young N'Dama cattle (Kaufmann *et al.* 1992) and in two different breeds of goats (Griffin, Allonby & Preston, 1981a; Griffin *et al.* 1981b). Specht (1982) did some field trials in sheep and goats and used treatments to rule out one or more pathogens. In mice, *T. congolense* has been studied in interaction with a chronic gastro-intestinal nematode infection (Fakae *et al.* 1994).

The interaction between trypanosomosis and helminthosis, both anaemia-causing diseases which often occur together in natural infections, has not been studied in Djallonké sheep under controlled conditions. This experiment has been designed to study the course of the disease in both single and mixed infections and to examine the interaction between *T. congolense* and *H. contortus*.

MATERIALS AND METHOD

The experiment took place during the dry season (December 1993 - July 1994) on Kerr Serigne station of the International Trypanotolerance Centre (ITC), The Gambia, West Africa. Young (6-8 months) female Djallonké sheep (n=40) were purchased on the local market. Vaccination against Peste des Petits Ruminants (PPR-Tissue Culture Rinderpest Vaccine, Laboratoire Hann, Dakar) and Pasteurella was provided (Vaccin Antipasteurelique-mouton, Laboratoire Hann, Dakar). Deworming was done in week -12 and week -5 with ivermectin (Ivomec^R, MSD U.S.A.) and in week -2 with fenbendazole (Panacur^R, 10%, Hoechst, Germany) at the recommended dosages. The animals went out for grazing during the day and were fed a supplementary ration calculated on a daily weight gain of 50 gram. The region is known to be free of tsetse flies (Snow, personal communication). The animals were allocated in 8 blocks (n=5) according to their weight. Five groups (n=8) were randomly selected from the blocks. The experimental set-up is shown in Table 1.

Table 1 : Experimental plan : Sheep were infected with 5000 L3 of *Haemonchus contortus* (HC) and/or with 10^4 bloodstream froms of *Trypanosoma congolense* (TC). The sequence of infection is different according to the groups.

	Week of Infection		
Infection group	Week 0	Week 1	Week 5
Group 1 (Control)			
Group 2 (Tc)	TC		
Group 3 (Hc)		HC	
Group 4 (TcHc)	TC	HC	
Group 5 (HcTc)		HC	TC

Tc : *Trypanosoma congolense* Hc : Haemonchus contortus

Besides a control group, two groups (2 and 3) received a single infection, either *T. congolense* or *H. contortus* and two groups (4 and 5) were successively infected with both parasites: either *T. congolense* followed by *H. contortus* (TcHc) or vice versa (HcTc). The interval between both infections was different (1 week in the TcHc-group and 4 weeks in the HcTc-group), because the second pathogen was given at the moment the effect of the first infection was expected to reach its maximum. All times referred to in the text are related to time following the first *T. congolense* infection (Week 0). The duration of the experiment was 30 weeks.

Treatments

A West African strain of *Trypanosoma congolense* isolated in The Gambia was used. The pathogenicity of this cloned stabilate (ITC 84) has been described previously (Dwinger *et al.* 1992; Osaer *et al.* 1994). The artificial inoculation was done intradermally at five sites of the body flank totalling 1 ml infected mouse blood containing at least 10^4 bloodstream forms.

The infective larvae of *Haemonchus contortus* were provided by The Central Veterinary Laboratory in Lelystad, The Netherlands. The strain was isolated in 1971 and passaged 64 times in sheep. The strain is showing no resistance against anthelmintic products and is also known not to be inhibition-prone (Borgsteede, personal communication). All animals of the three groups receiving the *H. contortus* infection were infected orally using a stomach tube with 5000 infective larvae (L3). The dosage was based on the results of a trial infection.

Parameters

All animals were bled daily during the first week post infection (p.i.) and thereafter once weekly. Trypanosomes were detected by examination of the buffy coat using the darkground (DG) method (Murray, Murray & McIntire, 1977) and the number of trypanosomes was scored as described by Paris, Murray & McOdima (1982). Haematology parameters were determined by means of a Haematology Analyzer (ABX- MINOS ST International, Levallois, France) : packed red cell volumes (PCV), white blood cell counts (WBC), red blood cell counts (RBC), haemoglobine (HB), mean corpuscular volume (MCV) and mean corpuscular haemoglobine concentration (MCHC). Serum albumin was assessed using a colorimetric test (Kodak Ektachem Clinical Chemistry Slide ALB). The animals were weighed weekly for the whole duration of the experiment using a Salter Spring balance (50 kg/200grams accuracy).

Egg output (EPG) was assessed using the Mc Master technique (sensitivity of 100 eggs per gram) as described by Thienpont, Rochette & Vanparijs (1979). From day 12 post *H. contortus* infection (p.H.i.)onwards, the animals were faecal sampled daily until all were positive. Thereafter they were sampled once weekly until the end of the experiment. Rectal body temperatures were taken daily from the start of the experiment until week 7 and thereafter three times weekly until week 29. On any animal that died during the experiment a post mortem examination was performed.

The various traits (group, block, sequence of sampling and animal(nested within group*block)) were analysed by unbalanced analysis of variance using a mixed model. Animal effect was regarded as random factor. Whenever the group effect revealed to be significant for a trait, contrasts were defined to determine statistical differences between the groups. All hypothesis were tested by the F-test, where the approximation of Sattherwaite (Snedecor & Cochran, 1980) had to be used.

Table 2 : Mortality per group on total number of animals, weeks p.i. at time of mortality and symptoms observed before death or reason for death.

Group	Infection	Total mortality (No of deaths /No animals	Mortality due to infection	Time of death (week p.i.)	Reason of death
1	-	0 / 8			
2	Tc	1 / 8	0	17	Respiratoric problems * *Oestrus ovis*
3	Hc	1 / 8	0	6	car-accident
4	TcHc	2 / 8	2 **	3 7	Tc / Hc infection idem
5	HcTc	2 / 8	1**	6 27	Hc / Tc infection bloating *

* : Animals in good body condition; no oedema; no trypanosomes and EPG zero in animal of group 5. In animal of group 2, trypanosomes were present.
** : All animals had low PCV(5-17), clear signs of oedema, hydrothorax/hydropericard in some of them. Large numbers of adult *H. contortus* present in abomasum of two of them.

RESULTS

Although some deaths occurred due to other causes than trypanosomes and helminths, mortality due to the infections was only observed in the dual infected groups (Table 2).

For all parameters analysed the animal and sequence effect was highly significant. Average rectal temperatures were higher in the groups infected with *T. congolense* than in the other two groups. Group 1 and group 3 had significantly lower rectal temperatures than group 2 ($P < 0.01$), 4 and 5 ($P < 0.05$).

All animals of the *H. contortus* infected groups (3-4-5) excreted eggs. The pre-patent period for group 4 was the shortest. However, when subjected to analysis of variance, the group and block effect were both not significant, indicating no differences in prepatent periods between single *H. contortus* infection group and double infection groups. Sheep of group 4 had the sharpest rise in egg output, reaching its peak EPG value at week 4 p.H.i. Also groups 3 and 5 reached a peak at week 4 p.H.i. (Table 3 and Figure 1a and 1b). Analysis of the logarithmic transformation of the EPG data revealed that group 4 had a higher egg output ($P < 0.05$) during the peak period than groups 3 and 5.

Table 3 : Duration of the prepatent period (PPP) and peak EPG figures for the groups infected with *H. contortus*.

Group	PrePatentPeriod (days)		Peak EPG at week 4 p.H.i.	EPG
	mean ± s.e.	range	mean	Range
3 Hc	19.4 ± 1.6	16-30	3,425	0 - 14,000
4 TcHc	17.4 ± 1.6	16-19	10,271	200 - 32,700
5 HcTc	19.4 ± 1.4	14-25	4,029	0 - 22,700

For all *T. congolense* infected groups (2-4-5) the first occurrence of trypanosomes in the blood circulation was between 4 and 7 days following intradermal needle challenge. There was no tendency for 'self-cure' towards the end of the experiment (Figure 2). The levels of parasitaemia were analysed for the 30 weeks observation period. After correction for the different dates of infection they showed a group effect ($P < 0.02$) with group 4 having lower ($P < 0.05$) parasitaemia levels than groups 2 and 5.

All groups except for the control group showed a sharp decline in mean PCV % levels shortly after infection (Figure 3). Group 4 showed a further decline until week 5 before starting to recover and group 5 showed a second decline in PCV % in week 7 at the time of the superimposed *T. congolense* infection and started the recovery only in week 11. Group 4 showed the lowest PCV % levels for 5 weeks and although the initial decline in group 4 was more marked than in group 5, the first group recovered quicker than groups 2 and 5. A significant group effect was found with groups 2, 4 and 5 having lower levels of PCV % than group 1 ($P < 0.04$).

Group 2 had significantly lower RBC values than the control group and group 3.

Groups 4 and 5 were not significantly different from group 2 and from the control group considering the 30 week period.

Mean Hb levels (Figure 4) for groups 2, 4 and 5 were significantly ($P < 0.05$) lower throughout the experiment compared to the controls. Group 3 (Hc) was not different to the control group but had significantly higher Hb values than groups 2 and 4. The differences between groups 2, 4 and 5 were not significant. The MCV values showed an initial increase for all groups, in particular for groups 2 and 4 but analysis revealed no significant differences between groups. Mean MCHC values showed a sharp decrease for all groups including the control group from week 2 onwards. A subsequent increase was present which however never reached the pre infection values. There was a significant difference between the groups ($P < 0.05$) with group 4 showing lower values than group 1 and 3. The contrasts indicated no difference between single and dual *T. congolense* infection groups (2, 4 & 5). Mean WBC counts showed no statistical differences between the groups. Mean serum albumin levels for group 3 and 4 declined below 2.4 g/dl at week 5 of the experiment. Thereafter, the average values for albumin remained within the normal range. The contrasts indicated significant higher values for group 1 compared to groups 4 and 5 and for group 2 which had higher values than group 4. The albumin concentration of groups 2 and 3 were not significantly different and did also not differ from those of the control group.

The mean body weight changes are presented in Figure 5. All groups had increasing body weights till week 26 after which they all showed a small decrease. Group 2 had the lowest weights at the end of the experiment, however, the lowest weight gain was observed in group 5 (23.8 gram/day versus 29.5-38.6 gram/day in other groups). Analysis of variance revealed no significant group effect. Also, statistical analysis of the individual weight gain between week 0 and week 30 of the experiment was not significant different between groups and blocks indicating similar weight gains for all animals under different treatments and in different weight categories.

DISCUSSION

The most striking phenomenon in this experiment is the fact that mortality due to the experimental infections was only observed in the dual infection groups (in the TcHc group it reached 25 %). Only the *T. congolense* infection had a significant influence on the PCV % and was the main cause for anaemia over the entire experimental period. High mortality rates were also observed in dual infected goats, however only the sequence *T. congolense* followed by *H. contortus* was studied in that experiment (Griffin *et al.* 1981a,b). They used two different breeds of goats but mortality was as high in the exotic breed as in the indigenous breed. Kaufmann *et al.* (1992) also carried out a similar experiment using N'Dama calves and similarly to the present experiment the mortality was higher in the mixed (50%in TcHc and 33% in HcTc group) than in the single (11% in Hc and 0 % in Tc group) infected groups. These high mortality rates might also be explained by the fact that the experiment of Kaufmann *et al.* (1992) was carried out under field conditions during the rainy season, so that additional natural infections with *H. contortus* and trypanosomes cannot be ruled out. Furthermore, mortality due to other agents cannot be excluded since the etiology of the mortalities was not verified by post mortem examination. The current experiment on Djallonke sheep took place during the dry season and it is known that no re-infection with helminths occurs during this season (Ankers, Zinnstag & Pfister, 1994). *In addition, only small numbers of helminth eggs were detected in the animals* of the control group and group 2 towards the end of the experimental period (Fig. 1a) which could be due to infections taken up from pasture when humidity was rising. Since the region is also known as having a zero trypanosomosis challenge (Snow, personal communication) natural infection with trypanosomes is therefore excluded.

Analyses of the different parameters measured clearly indicated the severeness of the mixed infections as compared to the single infections. More significant differences were observed between the mixed infection groups and the control group than between the single infected groups and the control. Weight gain was also low in the two mixed infection groups.

Clear differences were noted between the two dual infected groups. In group 4 (TcHc)the effects were more acute, judged by a higher mortality shortly after the infections, a significantly higher peak EPG compared to groups 3 and 5 and a significant lower PCV compared to the control group. Values for Hb, PCV, MCHC and albumin were significantly different from the control group. The shorter PPP noted for group 4 compared to groups 3 and 5 was however not significantly different. Griffin *et al.* (1981a) found a shorter PPP for the goats infected first with *T. congolense* followed by *H. contortus* as compared to the single *H. contortus* infected group. The dual infected goats unexspectedly showed a lower or equal EPG output but a higher worm burden as compared to the single *H. contortus* infected goats.

The effects in group 5 (HcTc) were more chronic. No high or low peak values for the parameters measured were noted, except for a significantly higher parasitaemia compared to group 4. The effect of the infection lasted longer as judged by the values for PCV, RBC, Hb and albumin which did not reach their pre-infection values within the observation period, whereas for group 4 these pre-infection values were reached earlier. A superimposed infection with *T. congolense* on *H. contortus* made the recovery period for the animals much longer. The weight gain in this group was also lowest over the 30 weeks of observation (23.8 gram versus 29.5 - 38.6 gram in the other groups). This factor together with the results of the other parameters indicate a more chronic effect of the sequence of infection in group 5.

The trypanotolerant character of the Djallonké sheep was again confirmed in the present experiment. No mortality due to the infection with *T. congolense* occurred in the single infected group. In previous experimental infections, in which the same *T. congolense* strain was used, it was observed that the Djallonké sheep showed an important level of resistance against trypanosomosis proved by a zero mortality, positive weight gain and a non significant difference in reproductive performance compared to the control group (Goossens & Osaer, unpublished observations) However, once the trypanosome infection is complicated by an intercurrent infection with *H. contortus*, an increased mortality is observed.

It was observed in this experiment that a single *H. contortus* infection did not cause a significant decrease in PCV levels as compared to the control group. The bloodsucking activity of the *H. contortus* worms only lasted for ± three weeks. The EPG output was rather low and the animals recovered afterwards if there was no superinfection with a second pathogen. These observations are in contrast with those in the Netherlands, where the strain gave always a very high EPG and a severe drop in PCV % in lambs up to nine months (Borgsteede, personal communication). Differences in EPG, specially regarding the peak values as observed in this experiment, were also observed in other experiments studying the resistance against gastro-intestinal parasites in different breeds of small ruminants (Gray, 1987). Zajac *et al.* (1990) reported that all animals of three different breeds (Florida Native, St. Croix and Dorset/Rambouillet) experimentally infected with *H. contortus* had a peak EPG in week 4 or 5. The more sensitive breed (Dorset/Rambouillet) had a much higher peak and a slower reduction than the two other breeds (St. Croix and Florida Native) for which tolerance was suggested which was confirmed later by Gamble & Zajac (1992) under artificial and natural challenge.

Acquired and innate resistance was also demonstrated by Preston & Allonby (1979) for the Red Masai breed, based on differences in faecal egg counts and mortality rates.

The results of the present experiment indicate some degree of natural resistance of Djallonké sheep to *H. contortus*, although the infection dose was rather low (5000 L3) and the infectivity of the larvae might have decreased due to the rather long storage time of three months before using (McKenna, 1973).

Although the Djallonké sheep used in this experiment seemed to withstand a single *H. contortus* infection very well, this was much less the case when pre-infected with *T. congolense*. The initial infection with *T. congolense* probably suppresses the immunity of the Djallonké and makes them more susceptible to a *H. contortus* infection. Dwinger *et al.* (1994) studying the effects of trypanosome and helminth infections on production of village N'Dama cattle, found also that animals infected with helminths showed higher egg counts, when they were simultaneously infected with trypanosomes. In addition there were clear indications that animals had an increased susceptibility to trypanosomosis when infected with helminths.

It has also been suggested that the erythropoietic response of indigenous breeds of cattle and sheep may be higher than that in exotic breeds (Anosa, 1977; 1988a, b) and that this factor contributes to a greater tolerance against the disease (Griffin, 1978). For the groups with the mixed infections the increase in MCV and decrease in MCHC were most marked. The worst combination here seemed to be the *T. congolense* infection followed by the *H. contortus* infection (group 4). Hb levels of this group and of groups 2 and 5 remained also significantly lower than the control group throughout the experimental period. The response of the animals to a single *H. contortus* infection was not marked by a severe or long lasting depression in RBC. However, for this parameter the most severe effects were seen in groups 2 and 5. It was remarkable that group 5 performed worse than group 4. Comparable observations on red blood cell destruction were made by Griffin *et al.* (1981b) on goats where a more rapid red cell loss was found in dual infected goats compared to single *H. contortus* goats.

In conclusion, this study has clearly shown that dual infections have a more important impact on Djallonké sheep and their capacity of recovering from an infection than single infections with either *H. contortus* or *T. congolense*. A *H. contortus* infection superimposed on a *T. congolense* infection (TcHc group) caused more acute effects and the mortality was also highest in this group (2 out of 8). The significantly higher peak EPG in these animals increase the risk of infection for other animals grazing together and has thus important epidemiological consequences. In the animals, which received a *T. congolense* infection superimposed on a *H. contortus* infection, the effects were more chronic. It took much longer than in the former group before pre-infection values for several of the examined parameters were reached.

Although, the Djallonke breed showed some resistance against diseases when infected with one pathogen at the time it is clear that under natural conditions mixed infections are more likely to occur. Even disease tolerant animals suffer more from mixed infections than from single infections, influencing significantly their individual productivity. Apart from suggestions (Assoku, 1981; Baker, 1995), up to now for the Djallonké breed no clear evidence for resistance against *H. contortus* in particular is available. In order to further elucidate this aspect, appropriate experiments should be carried out comparing the susceptibility of different sheep breeds towards a well known pathogenic *H. contortus* strain.

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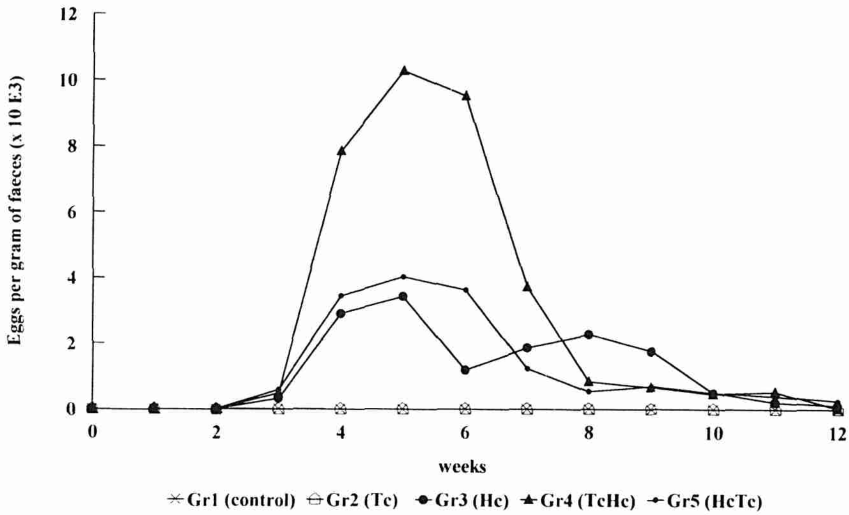

Figure 1a : Faecal worm egg count in eggs per gram of faeces (group averages) over the period week 0 - 12 of sheep infected with *Haemonchus contortus* (Hc group) (○), sheep infected with *Trypanosoma congolense* followed by *Haemonchus contortus* (TcHc group) (▲), sheep infected with *Haemonchus contortus* followed by *Trypanosoma congolense* (HcTc group) (●) and of sheep infected with *Trypanosoma congolense* alone (Tc group) (⌂) or control sheep (∗).

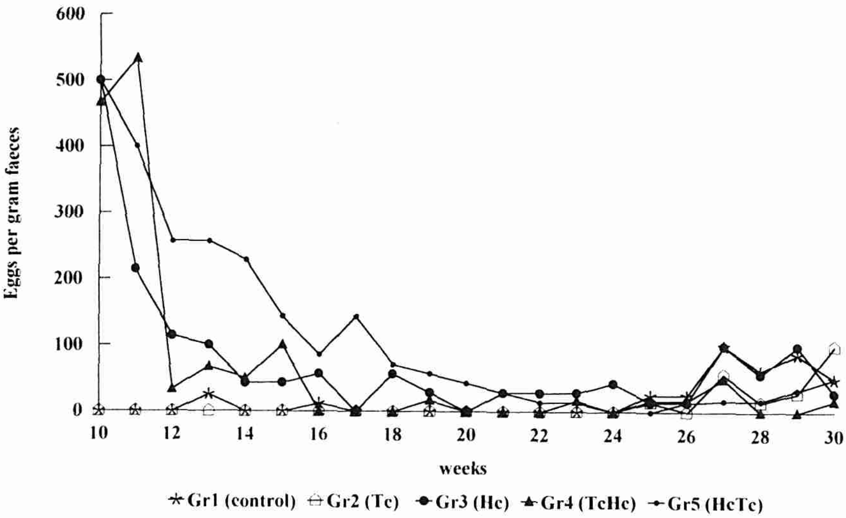

Figure 1b. : Faecal worm egg count in eggs per gram of faeces (group averages) over the period week 10 - 30 of sheep infected with *Haemonchus contortus* (Hc group) (○), sheep infected with *Trypanosoma congolense* followed by *Haemonchus contortus* (TcHc group) (▲), sheep infected with *Haemonchus contortus* followed by *Trypanosoma congolense* (HcTc group) (●) and of sheep infected with *Trypanosoma congolense* alone (Tc group) (⌂) or control sheep (∗).

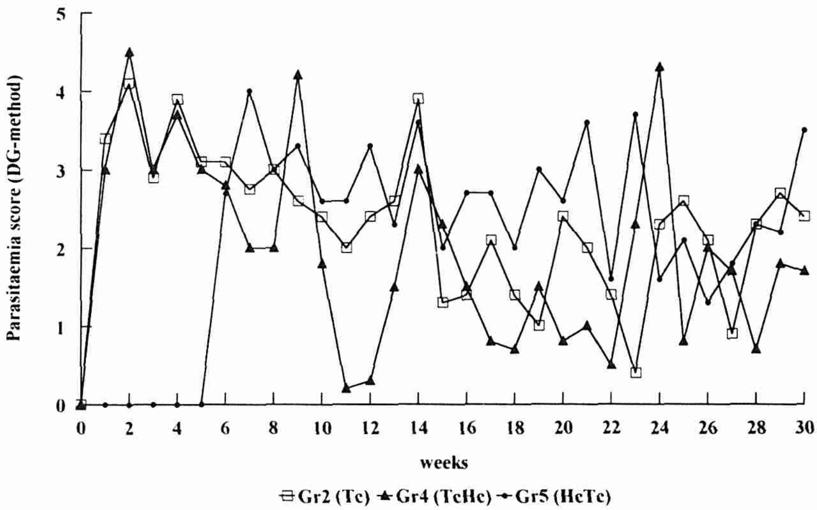

Figure 2 : Parasitaemia level (group averages) as scored by the Darkground Method for sheep infected in week 0 with *Trypanosoma congolense* (Tc group) alone (◻) or followed one week later by infection with *Haemonchus contortus* (TcHc group)(▲) and sheep infected with *Haemonchus contortus* followed by *Trypanosoma congolense* infection in week 5 (HcTc group) (●).

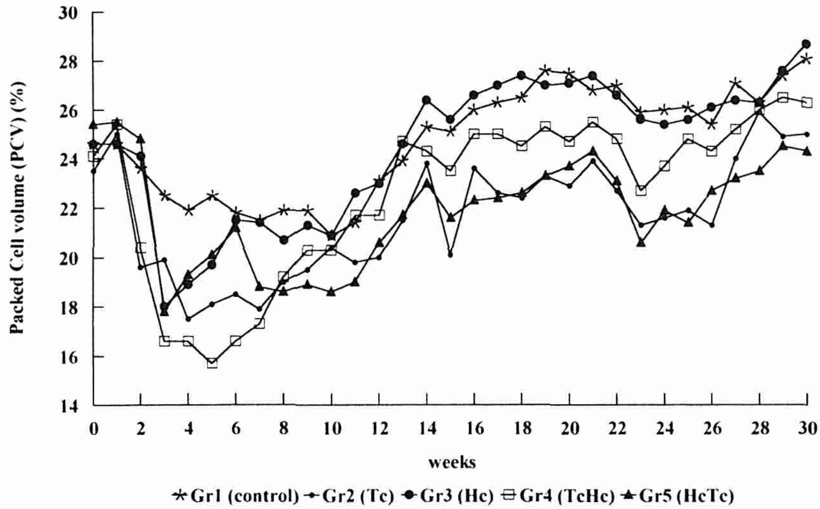

Figure 3 : Packed red cell volumes (PCV) percentages (group averages) over 30 weeks of non infected sheep (∗), sheep infected with *Haemonchus contortus* alone (Hc group) (○) or with *Trypanosoma congolense* alone (Tc group) (⌂), sheep infected with *Trypanosoma congolense* followed by *Haemonchus contortus* (TcHc group) (▲) and sheep infected with *Haemonchus contortus* followed by *Trypanosoma congolense* (HcTc group) (●).

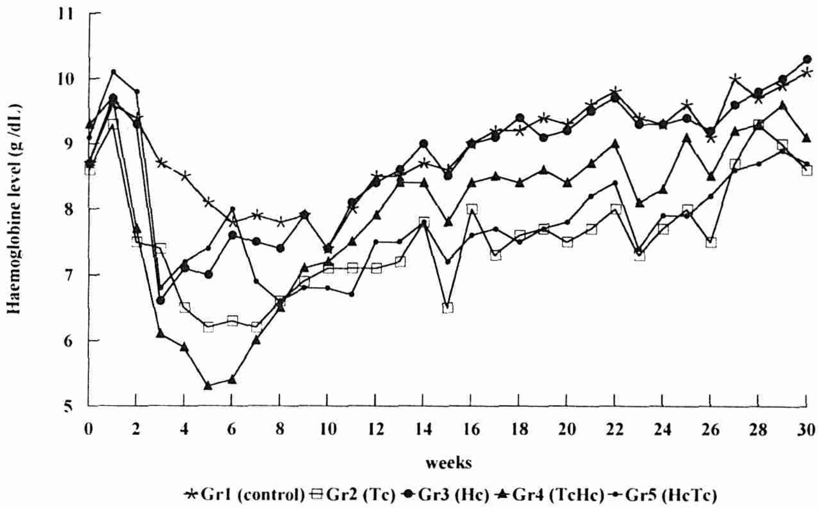

Figure 4 : Haemoglobin levels in g/dL (group averages) over 30 weeks of non infected sheep (*), sheep infected with *Haemonchus contortus* alone (Hc group) (○) or with *Trypanosoma congolense* alone (Tc group) (⌂), sheep infected with *Trypanosoma congolense* followed by *Haemonchus contortus* (TcHc group) (▲) and sheep infected with *Haemonchus contortus* followed by *Trypanosoma congolense* (HcTc group) (●).

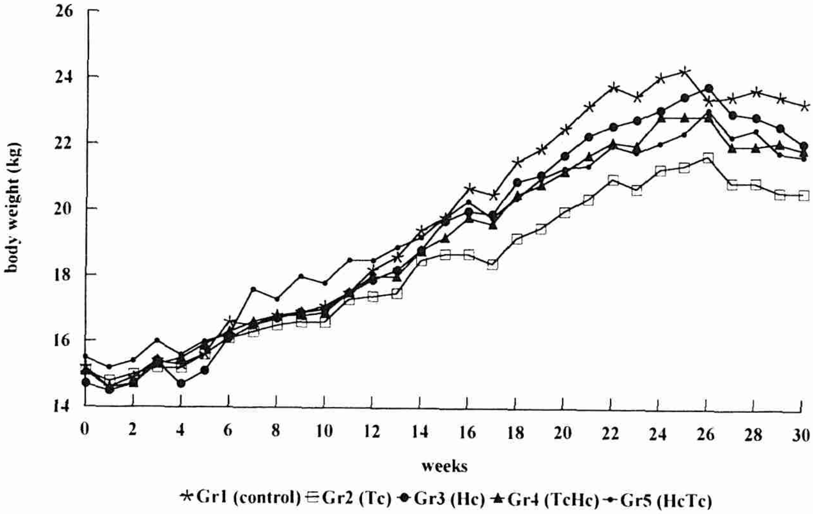

Figure 5 : Body weight changes in kg (group averages) over 30 weeks of non infected sheep (*), sheep infected with *Haemonchus contortus* alone (Hc group) (○) or with *Trypanosoma congolense* alone (Tc group) (⌂), sheep infected with *Trypanosoma congolense* followed by *Haemonchus contortus* (TcHc group) (▲) and sheep infected with *Haemonchus contortus* followed by *Trypanosoma congolense* (HcTc group) (●).

5.2

The effects of prophylactic anthelmintic treatment on the productivity of traditionally managed Djallonke sheep and West African Goats goats kept under high trypanosomosis risk.

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THE EFFECTS OF PROPHYLACTIC ANTHELMINTIC TREATMENT ON THE PRODUCTIVITY OF TRADITIONALLY MANAGED DJALLONKE SHEEP AND WEST AFRICAN DWARF GOATS KEPT UNDER HIGH TRYPANOSOMOSIS RISK.

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Abstract

The effects of a prophylactic anthelmintic intervention on the productivity of village based sheep and goats was studied in an area of high trypanosomosis risk in The Gambia during 2 years and 3 years, respectively. In total 223 sheep and 385 goats from 5 villages were included. Allocation to treatment groups (treated-control) was randomised by village, based on age and sex. Three treatments per rainy season were applied with Fenbendazole (Panacur[R], Hoechst, 2.5%, 5 mg/kg).

Mean nematode egg excretion per gram faeces (EPG) of the treated groups were significantly reduced by prophylactic anthelmintic treatment, indicating the efficiency of the treatment despite the risk of rapid reinfestation. Weight gain benefits of anthelmintic treatment were observed in all age categories (>6 mo.) of sheep but not in goats. Kidding rates were significantly increased whilst the same positive trends were observed for other reproductive parameters (litter size, parturition interval) in both goats and sheep without reaching statistical significance. Birth weights of offspring born out of treated does and ewes were higher ($P< 0.05$) than those from the controls. In contrast, growth rates until 3 months of age were not influenced by the treatment status of the dam. Mortality rates until the age of 3 months of kids from treated does were significantly lower than of those from control does. Mean Packed Red Cell Volume (PCV) levels during the rains were significantly higher in treated goats than in control goats. The same trend was observed in sheep. In general, there were no interactions between trypanosome infections and effect of anthelmintic treatment, thus both factors acted independently. Finally, the live weight productivity index (12 months old-offspring in kg/year/dam) for treated dams was 24% and 47% higher than in control ewes and does, respectively.

It can be concluded that, despite the continuous risk of trypanosome infections which has a negative impact on their productivity, a beneficial effect of anthelmintic treatment was observed in both species but most obviously in goats, measured as an increased production and improved health status. A cost-benefit analysis should be carried out in order to confirm whether prophylactic anthelmintic treatment can be recommended to farmers to increase their income from small ruminant production. Nevertheless, anthelmintic treatment will certainly optimise the trypanotolerance in these breeds.

Key words : sheep ; goats; anthelmintic treatment; trypanotolerance; productivity; rural development

Introduction

Traditional sheep and goat production in different parts of Africa is a very profitable enterprise but returns are strongly determined by biological performance (Panin and Mahabile, 1997; Sumberg and Mack, 1985; Upton, 1985; Itty et al., 1997). For the Gambia, sheep production even is more favourable than cattle production from a financial, gender and equity point of view (Itty et al., 1997). There are more goats (213,017) than sheep (155,132) (DLS/ITC census, 1993). The Djallonke sheep and West African Dwarf (WAD) goats are the main breeds in the Gambia and their trypanotolerant nature has been described as an ability to maintain production under infection (Mawuena, 1987; Osaer et al., 1994,1997; Goossens et al., 1997a). However, trypanotolerance is not absolute and stress or disease factors such as helminth infections, which coincide with trypanosome infections in field situations may interact with their resilience to the effects of trypanosomosis. Under experimental conditions, a *Trypanosoma congolense* infection in Djallonke sheep combined with natural helminth infections, resulted in a more severe anaemia and reductions in weight gain (Goossens et al., 1999). Similarly, concurrent infections of *T.congolense* and *H.contortus* in controlled experiments have proven the severeness of the mixed infections as opposed to both single infections on haematological parameters in sheep (Goossens et al, 1997b).

Gastro-intestinal parasitism in ruminants contribute greatly to production losses and even death (Parkins and Holmes, 1989; Barger et al., 1982; Cobon and O'Sullivan, 1992; Thomas & Ali, 1983; Hoste & Chartier, 1993; Tillard et al. 1992 et 1997). One of the possible interventions to reduce the impact of gastro-intestinal parasitism in developing countries are prophylactic anthelmintic treatments (Bullerdieck P., 1996). However, treatment is often expensive for the small scale farmer and incorrect usage can induce drug resistance. Grazing management techniques can support worm control programmes, and therefore reduce the reliance on anthelmintic treatments (Barger I.A., 1998;1999; Waller, P.J. 1997). A good knowledge and understanding of the epidemiology of helminth infections is a prerequisite for any anthelmintic intervention. Epidemiology of helminth infections in sheep and goats have been well studied in the Gambia (Fritsche et al., 1993; Ankers et al., 1994; Greenwood et al; 1989; Osaer et al., 1999). Over an one year study period, it was found that over 95% of sheep and goats of all age classes harboured nematodes (Fritsche et al., 1993). *Trichostrongylus colubriformis* and *Oesophagostomum columbianum* were most prevalent while *Haemonchus contortus* had an annual prevalence of 67%, but with the adult population peaking during the rainy season (July till October) (Fritsche et al., 1993). Ankers et al. (1994) observed that re-infestation with helminths was negligible during the dry season. Nematodes survive the long dry season either as hypometabolic adults or as hypobiotic larvae (Ankers et al., 1994; Kaufmann & Pfister, 1990; Zinsstag et al., 1994).

Few reports are available on the effects of prophylactic anthelmintic treatments in small ruminants kept under traditional, extensive management in developing countries. In The Gambia, intervention schemes based on 2 anthelmintic treatments during the rains have been tested in Djallonke sheep, and resulted in increased lambing rates and average litter size whilst mortality and weight at 12 months were not affected (Ankers et al., 1998). Despite its large variations, the rate of return was high and therefore this treatment scheme was highly recommendable (Ankers et al., 1998). In Senegal, biannual anthelmintic treatment lead to improved weight gains and lowered mortality in young sheep (Vassiliades G., 1984), whereas Tillard et al. (1992) reported on an improvement of the productivity index in both adult sheep and goats following 3 Fenbendazole treatments.

Because of concurrent infections with trypanosomes and other environmental stress factors, the possible benefits of anthelmintic treatment on productivity and health in sheep and goats might be different. In addition, the expression of trypanotolerance should be preserved as much as possible in an area with high trypanosomosis risk, so that intervention against helminth infections might be one way to reduce stress factors known to reduce resilience in those breeds. The effects of trypanosomosis on health and productivity of sheep and goats kept in the same area and studied during the same period have been described earlier (Osaer et al., 1999) and will not be discussed in this paper.

Whether goats have similar benefits from a prophylactic anthelmintic treatment as sheep has not often been studied under field conditions. The present study aimed at testing a prophylactic anthelmintic treatment scheme on-farm on indigenous goats and sheep kept under high trypanosomosis risk.

Materials and Methods

1. Study site

The study took place in Central River Division South, The Gambia, between July 1996 and December 1998. The study site is situated in the Niamina East area (13°40'N, 14°58'W), at 200 km from the Atlantic coast. This area is highly infested with the tsetse fly *Glossina morsitans submorsitans,* the major vector of animal trypanosomosis in the Gambia (Rawlings et al., 1991). Niamina East region was ranked as an area of high trypanosomosis risk which was expected to remain the same for at least 5 to 10 years, despite influence of demographic, climatic and environmental factors on tsetse populations (Rawlings, et al. 1993). Peak trypanosome infections in small ruminants occurred in the early dry season but annual infection rates for the study years 1996 and 1997 were 4.0 % and 7.1% in goats and 5.6% and 7.1% in sheep, respectively (Osaer et al., 1999). For the year 1996 these figures are incidence rates, because trypanocidal treatments were given to infected animals at monthly intervals. In the subsequent years, no trypanocidals were applied thus prevalence rates were measured (Osaer et al., 1999). In the third year 1998, the mean prevalence rate in goats was 10%. In the Gambia the wet and humid season lasts from June to the end of October, with annual rainfall for the years 1996, 1997 and 1998 in the study site being 701 mm, 638 mm and 593 mm, respectively (Goossens, unpublished results). The dry season lasts for 7 months, from November till May. The study site comprises 5 adjacent villages situated 1.5 km apart. Flocks of sheep and goats are managed in a traditional extensive way. During the dry season, most of the animals roam freely but sheep staying closer to the villages than goats. During the cropping season, starting with planting in July and lasting till after harvest and threshing in November, grazing of small ruminants is restricted in time and area (tethering).

2. Animals and treatments

At the start of the intervention in July 1996, 130 sheep and 180 goats from the 5 villages were selected, followed by a random allocation to treatment groups (T=treated ; C=control) per village and based on age and sex. Lambs/kids born during the trial were ear tagged and allocated to the same treatment group of their dam. Both sheep and goats were treated with anthelmintics during the rainy season of 1996 and 1997 and goats also during the third rainy season in 1998. In total, individual records of 223 sheep and 385 goats were included in the study which fulfilled the conditions for analysis. Three anthelmintic treatments were applied, using Fenbendazole (Panacur^R; Hoechst; 2.5%) at 5 mg/kg: the first two treatments were given during the rains to decrease the pathogenic effects of the nematodes by removing the adult and larval populations under constant infection pressure. The third treatment, given at the start of the dry season, would eliminate all nematodes, including the hypobiotic larvae, with negligible risk of reinfection (Ankers et al, 1994). In 1996, the 3 consecutive treatments were given in mid July, mid September and early November. In 1997 and 1998 the first treatment was postponed till early August, whilst the second and third treatment remained as in 1996. Animals were vaccinated against Peste des petits ruminants (PPR) at 6 monthly intervals. Between control groups and groups treated with anhelminthics of both species, there were no differences found in trypanosome infection rates. No routinely acaricide treatments were applied. Apparently, tick infestation was low and only associated with limping problems.

Sampling methods

The monitoring began in July 1996 and lasted 24 months and 30 months for sheep and goats respectively. Data on new entries, birth weights and exits were obtained at weekly intervals. Animals older than 5 months were bled monthly from the jugular vein into ethylene diamine tetra-acetic acid (EDTA)-coated vacutainer tubes (2-3 ml) for assessing Packed Cell Volume levels (PCV) and trypanosome infections. The latter were diagnosed using the buffy-coat dark ground technique (DG) (Murray et al., 1977) and the number of trypanosomes was scored by the method of Paris et al. (1982). From the total sheep and goats in the trial, for each species 20 % randomly selected faecal samples were collected monthly during the rainy season and every two months during the dry season. The number of strongyle eggs per gram of faeces (EPG) was determined using a McMaster technique with a sensitivity of 100 EPG (Thienpont et al., 1979). Monthly weighing was done with Salter spring balances (50 kg/200 gr accuracy).

Productive performance analyses

Traits on health and productive performance (EPG, PCV, live weight gain, parturition interval, parturition rate, average litter size, birth weight) were analysed by linear model techniques (GLM procedure; SAS, 1998). Parturition rate (number of parturitions/per year/per female) was calculated as the total number of parturitions divided by the number of years of presence per breeding female. Breeding females were considered as from their theoretical age at first parturition of 300 days (doe)- 350 days (ewe). The former ages were based on baseline data from the site and to avoid seasonal effects (Osaer et al., 1999), only dams present for at least 1 year were considered in the analyses. Apart from the main effect anthelmintic treatment (treated=**T**/control=**C**), trypanosome infection status (infected/not infected) and the following effects were included in the model: age-class (6-12 mo./ 12-24 mo./ >24 mo.), sex, village, and the interactions treatment*trypanosome infection and treatment*age-class. Additional factors, inherent to the observed trait, were included. Individual daily weight gains (linear regression, $Y= b_0 + b_1$ days) were calculated and analysed per season, which were also defined as loosing and gaining period, based on earlier observations (Osaer et al., 1999). The model for the estimated regression coefficient (b_1) fitted, apart from the above mentioned effects, also the effect 'lambed/kidded status'. For the analysis of the parturition interval, censored data were taken into account following the methodology suggested by Tanner (1993). These data were derived from dams which lambed/kidded only once during the first half of the survey but not a second time, despite their continuous presence in the flock. The analysis was carried out with programs specifically written for these data sets (H. Simianer, 1998, personal communication). All hypothesis were tested by the F-test (Snedecor and Cochran, 1980). Means of the different groups are presented as least square means ± standard error (mean ± s.e.). Offspring performance data (<3 mo.) were analysed by taking into consideration the treatment status of their dams. Estimates for mortality included also, apart from reported mortalities, animals which disappeared from the monitoring without reporting but which were assumed to have died. Mortality rates (up to 90 days / 90 till 365 days) were compared between treatment groups using Chisquare (Chi^2) tests.

Results

Nematode egg excretion (EPG).

The effect of anthelmintic treatment on the mean nematode egg excretion (EPG) was evaluated by comparing treated groups with controls. The overall means were for goats C: 516.6 ± 73.3 vs T: 319.6 ± 74.3 and for sheep C: 834.2 ± 162.8 vs T: 471.4 ± 166.4, for their respective study period. To approximate the normal distribution, data on EPG were logarithmic (ln) transformed. For both species, anthelmintic treatment reduced EPG significantly ($P< 0.001$), but with highest effect during the rains when peak excretion occurred (interaction month*treatment). In sheep, the reduction in EPG due to treatment was greatest in sheep aged 12 to 24 months (interaction age*anthelmintic treatment; $P< 0.01$). The village effect ($P< 0.05$) indicated important variations in levels of EPG between the villages included in the study. Differences between species were not significant.

Weight changes.

Mean daily weight gains for both species during the different seasons are presented in table 1. In general, a beneficial effect of anthelmintic treatment was present for sheep, especially in the dry season 1996/97 where the increase in weight gain was significant ($P<0.01$). The interaction age* treatment was not significant, indicating that the positive effect of anthelmintic treatment was equal for all involved age categories. In contrast, there was no such positive effect of treatment on weight gain in goats. Generally, weight gains were considerably higher in the dry season than in the rainy season, especially in the rainy season 1998 when weight losses were observed in goats. The interaction of the anthelmintic treatment with trypanosome infection status of the animal was not significant except in sheep during the rainy season '97, where a positive effect of anthelmintic treatment ($P<0.05$) was mainly seen in the trypanosome infected sheep (T-infected: 33.0 ± 12.2 g vs C-infected: -6.8 ± 14.4 g).

Reproductive performance.

The mean parturition rate and litter size per breeding female for both species and for the different treatment groups are presented in table 2. Whilst a positive influence of anthelmintic treatment was observed for both parameters in both species, only kidding rates were significantly ($P<0.01$) increased due to prophylactic anthelmintic treatment. Parturition intervals, including censored animals, were shortened due to treatment but differences were for neither species significant. Interactions of anthelmintic treatment with their trypanosome infection status were not significant, indicating that both factors were acting independently.

Table 1: Effect of prophylactic anthelminthic treatment[1] on daily weight gain (age-classes > 6 months) in the different periods (seasons) : least square means ± s.e. (in g/day).

Species:	Goats					Sheep			
Treatment group	rains 1996 (n=146)	dry 96/97 (n=119)	rains 1997 (n=226)	dry 97/98 (n=122)	rains 1998 (n=130)	rains 1996 (n=105)	dry 96/97 (n=82)	rains 1997 (n=132)	dry 97/98 (n=75)
Control	13.9 ± 7.3	59.9 ± 6.2	11.2 ± 7.3	59.2 ± 5.6	-5.5 ± 8.5	19.2 ± 7.2	38.1 ± 7.2	8.4 ± 8.9	45.7 ± 12.3
Dewormed	13.4 ± 7.8	55.5 ± 7.5	19.1 ± 8.0	57.9 ± 6.2	-4.7 ± 7.9	24.9 ± 7.0	59.5 ± 7.1	26.9 ± 7.7	63.3 ± 9.6
significance	*ns*	*ns*	*ns*	*ns*	*ns*	*ns*	*P<0.01*	*P<0.06*	*ns*

Other effects included are trypanosome infection status, sex, age-class, village, interactions deworm*age-class and deworm*infection status.
(1) : anthelmintic interventions during the rainy seasons of 1996 and 1997 for both species, for goats also in 1998.

Table 2: Effect of prophylactic anthelminthic treatment [1] on reproductive parameters in sheep and goats in an area of high trypanosomosis risk: least square means ± s.e.

Species / Parameter	goats				sheep			
	cases (n)	Control	Dewormed	*Sign*	cases (n)	Control	Dewormed	*Sign*
parturition rate	141	1.00 ± 0.06	1.21 ± 0.06	*P <0.01*	85	1.15 ± 0.09	1.26 ± 0.12	*ns*
average litter size	141	1.57 ± 0.07	1.61 ± 0.08	*ns*	85	1.15 ± 0.06	1.22 ± 0.08	*ns*
parturition interval	143	267.4 ± 10.4	251.1 ± 6.6	*ns*	118	254.3 ± 11.7	236.9 ± 12.1	*ns*

Other effects included are trypanosome infection status, village, interaction deworm*infection status.
[1] : anthelmintic interventions during the rainy seasons of 1996 and 1997 for both species, for goats also in 1998.

Table 3: Effect of prophylactic anthelminthic treatment [1] of sheep and goats on the performance of their offspring in an area of high trypanosomosis risk: least square means ± s.e.

Species / Parameter	Kids				Lambs			
	cases	Control	Dewormed	*Sign*	cases	Control	Dewormed	*Sign*
Birth weight	273	2.4 ± 0.1kg	2.7 ± 0.1kg	*P < 0.05*	111	2.6 ± 0.2 kg	3.1 ± 0.2 kg	*P< 0.05*
Daily gain till 3 months	105	67.9 ± 4.8 g	66.3 ± 7.0 g	*ns*	57	122.4 ± 10.4 g	108.1 ± 13.6 g	*ns*
Mortality till 3 months % (no/no total)	410	31.2 % (68/218)	22.4 % (43/192)	*P < 0.05*	247	28.6 % (36/126)	32.2 % (39/121)	*ns*

other effects included are sex, type (single/multiple birth), dam's trypanosome infection status, village, age at birth weight-recording
[1] : anthelmintic interventions during the rainy seasons of 1996 and 1997 for both species, for goats also in 1998

Offspring performance

Birth weight and daily weight gain of offspring born during the study out of treated or control dams are presented in table 3. In addition, the influence of treating the dam on the offspring's mortality rates till 90 days are presented. In both species, birth weight of offspring of treated dams were significantly higher than those of control dams (P<0.05). There was no significant interaction between treatment status and trypanosome infection status of the dam. Thus, the positive effect of anthelmintic treatment was similar in trypanosome infected or negative dams. Daily gain till 3 months was not influenced by the treatment status of the dam. In contrast, a reduction of the mortality rates in kids up to 3 months was observed (P< 0.05) due to anthelmintic treatment of their dams (see table 3).

Mortality - Packed Cell Volume levels

Mortality rates between 3 and 12 months of age did not differ between treated or control animals with respective means for goats and sheep: C=31.9% vs T=38.9% (G; n=146) and C=33.3% vs T=34.8% (Sh; n=79). Most mortality (>75%) occurred during the rainy season.

Mean packed cell volume levels (PCV) of both control and treated groups of goats and sheep are presented in table 4 for the consecutive rainy seasons during which anthelmintic treatments took place. PCV-values were significantly higher in treated goats than in control goats in the first two grazing seasons; the same trend, though not significant at the 5% level of probability, was seen for goats in the third and sheep in both grazing seasons. The effects of trypanosome infection and anthelmintic treatment on PCV were acting in an additive way (interaction: ns).

Table 4: Effect of prophylactic anthelminthic treatment on Packed Cell Volume (PCV) levels in sheep and goats during the rainy seasons of 1996, 1997 and 1998 (goats only) in an area with high trypanososomsis risk : ls means ± s.e.

Species / Treatment group	goats			sheep	
	rains 1996 (n=138)	rains 1997 (n=189)	rains 1998 (n=144)	rains 1996 (n=91)	rains 1997 (n=138)
Control	20.8 ± 1.2%	19.4 ± 0.9%	21.0 ± 0.7%	20.5 ± 1.1%	20.9 ± 0.8%
Dewormed	23.3 ± 1.3%	21.2 ± 1.0%	21.3 ± 0.8%	23.1 ± 2.2%	22.7 ± 0.8%
significance	*P<0.05*	*P<0.05*	*ns*	*ns*	*ns; P<0.06*

Other effects included: trypansome infection ; age_class; village; sex; age* deworm treatment; deworm treatment*trypanosome infection;

Productivity index

A mean live weight productivity index was calculated for both dewormed and control groups of sheep and goats, based on the following formulae (Ankers et al., 1998): parturition rate x litter size x (1-mortality rate till 3 mo.) x (1- mortality rate 3-12 months) x weight at 365 days. In both species, the productivity index was considerably increased in the groups receiving anthelmintic treatments, but with the greatest increase for the goats. Productivity indices in goats were: T=21.2 kg and C=14.4 kg and in sheep: T=17.6 kg and C=14.2 kg for treated and control groups, respectively.

Flock structure.

The average relative flock structure for both species is presented in figure 1. It shows that male offtake rate is high in both species but highest in sheep with the majority of the outflow occuring before the age of three years. For both species, the number of males older than 4 years is neglible, whilst there are still high number of females in that age category. The ratio of females per male above 2 years was 3 to 1 for both species.

Discussion

The results of the present on-farm trial demonstrated beneficial effects of prophylactic anthelmintic treatment on production and health parameters in both species but most clearly in goats, in spite of a continuous risk of trypanosomosis, contaminated environment (helminths) and the presence of other environmental stress factors.

Nematode egg output

Nematode egg excretion (EPG) was significantly reduced in the treated groups in spite of the immediate risk of reinfection with infective larvae from the communal pastures. During this study animals from both treatment groups were commingled and therefore contributed greatly to rapid reinfestation. Moreover, the usual management practises applied under these conditions have proven to play a role in the re-infection of helminths as observed in cattle (Kaufmann et al., 1993) and small ruminants (Osaer et al., 1999). The concept of strict separation of treatment groups was not withheld because it would not be feasible in the existing traditional production systems. Therefore, the benefits of the anthelmintic treatment may have been underestimated in this study. Nevertheless, based on previous epidemiological studies (Fritsche et al., 1993) it can be assumed that the applied treatment scheme in the present trial had suppressed considerably the worm burden in both sheep and goats.

Weight gain

Weight gain advantages of treated versus control sheep were observed at a similar scale in all age classes, but this trend was not observed for goats. When the same prophylactic treatment scheme was applied under controlled conditions in the Gambia, treated Djallonke sheep had on average 1.1 kg ± 0.1 better weight gain than the controls while grazing on the same pastures (Goossens et al., submitted). In 6-12 month old sheep treated twice a year with an anthelmintic (once in the dry - once in the rainy season), Vassiliades (1984) reported on a 40% improvement of weight gains. Contrary to the present study, there was little risk of reinfection since all animals in the village were treated. In addition, the trial was carried out in the Louga region, which is a more northern, dryer zone in Senegal and where Sahelian sheep are present (Vassiliades, 1984). A biannual treatment scheme was also tested in village-based Djallonke sheep in the Gambia by Ankers et al. (1998), with two Fenbendazole treatments given during the rains, but mean weight at 12 months was not significantly higher in the treated groups. This is in contrast with the present study, where a third treatment at the start of the dry season gave clearly improved weight gains in the sheep of all age categories, also during the following dry season. This last treatment was probably the most efficient with respect to remanence since it was given at a time with low risk of reinfection. Moreover, it supports the recommendations of Ankers et al. (1994) who stressed the importance of an early dry season treatment both in sheep and cattle and of Ndao et al. (1995) who measured a significant weight increase in cattle following a single dry season treatment. The negative effect of gastro-intestinal parasitism on weight gain in sheep has been extensively reported following artificial helminth infections (Albers et al.1984, Kimambo et al. ,1988a; 1988b) and under natural infections in young grazing sheep (Barger, 1982) with a 32% to 79% weight gain reduction in the untreated groups. In this study, an improvement in weight gain was observed not only in the growing sheep but also in the adult stock while this was not apparent in the goats.

Reproductive performance

Kidding rates in treated goats were significantly higher than in control goats while in sheep, the same trend was observed but the difference did not reach significance. Moreover, there was a positive effect of anthelmintic treatment on litter size and parturition intervals in both species. Improved reproductive performance as a result of prophylactic anthelmintic treatment has also been reported in Djallonke ewes by Ankers et al. (1998), with significantly increased lambing rates and litter size. Compared with this trial, the early dry season treatment did not bring in extra benefits for the reproductive performance in sheep. However, a different study area and time, less observations on sheep and uncontrolled environmental effects make it difficult to compare both results. Moreover, the negative effects of trypanosome infection on reproduction in both species (Osaer et al., 1999) were diminishing the beneficial effect of anthelmintic treatment although both factors were acting in an additive way (interaction ns). Anthelmintic treatment of the dam significantly increased effect on the birth weights of their lambs and kids. A large proportion of the females were pregnant at the start of the rains when treatment commenced, since a higher frequency of parturitions occurred between September - November (Osaer et al., 1999), and therefore direct benefits from treatment were reflected in a better production at birth. Additional beneficial effects in the treated does were seen in increased survival rates of their kids. Offspring did not receive treatment before the age of 3 months, and because of the seasonal peak of parturitions, a large number of offspring did not receive treatment until the next rainy season. The effect of treatment status in the dam on offspring growth rates until 3 months did not reveal any improvements for either species, thus, not confirming the findings of Cobon & Sullivan (1992) who observed increased milk yield in ewes receiving anthelmintic treatment and subsequently higher weight gains in their lambs. In these management conditions, other intrinsic effects (sex, litter size) and environmental effects (village) were significant factors influencing the growth rates. Trypanosome infection during late pregnancy and lactation did not significantly influence offspring growth (Osaer et al., 1999). Mean live weight productivity at 12 months, which captures the measured traits and therefore implies a good measurement of overall productivity, was considerably increased in the treated sheep and goats compared to their controls but most obviously in the latter species, with an improvement of 24% and 47%, respectively. These results are not entirely in line with those found by Tillard et al. (1992), who reported an increase of the productivity index of 33% in sheep and only 13.5% in goats in the Kolda area, using a comparable treatment scheme but with a different calculation of the index (dam's production of 3 months-old offspring). Ankers et al. (1998) reported a similar increase of 25% in the live weight productivity index (of 12 month-aged lambs) of Djallonke sheep in the Gambia following a bi-annual anthelmintic treatment. For the sheep in this study, there was no extra productivity gain derived from the three treatments versus the bi-annual scheme. Yet, the foregoing criteria may explain why results of both trials did not differ.

Packed Cell Volume

Treated groups had higher PCV levels during the rains than control groups. The effects of trypanosome infection and anthelmintic treatment on PCV level were found to act independently in the present study (interaction; ns). Apart from these 2 disease factors, seasonal effects indicated significantly lower PCV levels during the rains, attributable to the nutritional stress in small ruminants under these management conditions (Osaer et al., 1999). The erythropoiesis responding to anaemia can only be efficient when sufficient nutrients are available and will cause a metabolic shift at the cost of production. If the nutritional supply is very low, the anaemia can become non-regenerative because of erythropoietic failure. In that respect, the decline in PCV following artificial infection of *H.contortus* was more severe in lambs not supplemented with protein-rich pellets (Shaw et al., 1995). Naturally infected Djallonke sheep fed low levels of supplement had lower PCV levels than those fed high levels (Goossens, submitted, 1999). From the present study, it was clear that helminth infection combined with inadequate nutrition and trypanosome infections contributed to the lower PCV levels and consequently higher frequencies of mortality observed during the rains.

Age at off-take

The flock structure in the present study showed that male off-take at a relatively young age was high for both species, corroborating earlier observations in the Gambia (Greenwood et Mullineaux, 1989) and in Nigeria by Sumberg and Mack (1985). The proportion of females older than 4 years remained high. An abattoir survey in the Gambia revealed that the majority of small ruminants brought to slaughter were females whereas the males constituted only a small proportion, because they are rather kept for house slaughtering and religious or ceremonial feast (Goossens et al., 1998). This supports the recommendation of treating all age classes of both sexes, since the Gambian farmers not only profit from selling young males but also adult females on the local markets (lumo's). Therefore, as a follow-up, it is envisaged to calculate the cost/benefits of such prophylactic anthelmintic treatments for goats and sheep kept under low-input traditional management.

Conclusions

From the present results it can be concluded that in spite of a continuous risk of trypanosome infections and other environmental stress factors, the anthelmintic treatment scheme increased production and improved the health status of these indigenous sheep and goats. Compared with a biannual treatment scheme, extra benefits from the third treatment in sheep were observed as improved weight gains in the dry season but not on reproductive performance. A cost-benefit analysis will need to confirm whether this treatment schedule can be recommended to farmers to increase their income from small ruminant production. Nevertheless, anthelmintic treatments certainly optimise the trypanotolerance in these breeds.

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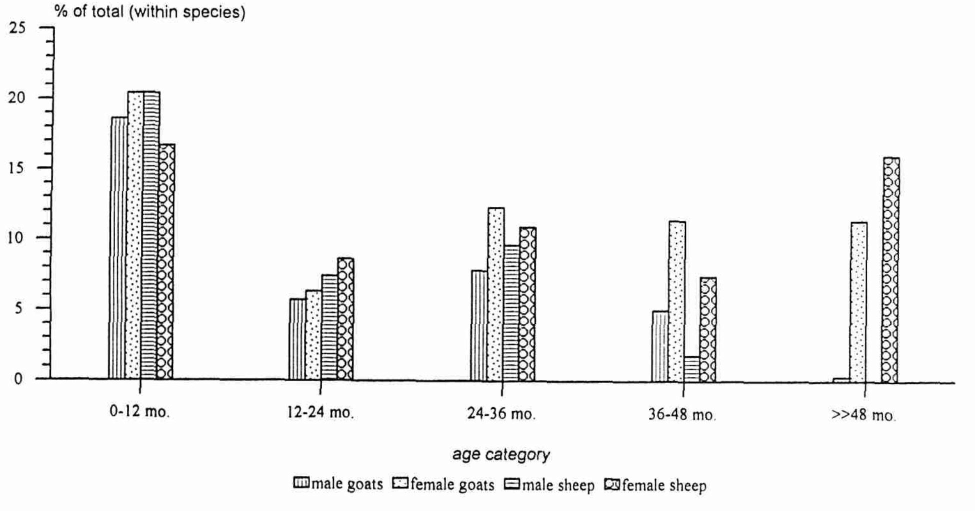

Figure 1: Average flock structure for sheep and goats: percentages for both sexes of the different age-categories within species.

CHAPTER 6

STUDIES ON THE GENETIC BASIS OF THE TRYPANOTOLERANT TRAIT IN DJALLONKE SHEEP AND THE USE OF F1-CROSSES TO INCREASE PRODUCTIVITY.

6.1

The susceptibility of Djallonke and Djallonke-Sahelian crossbred sheep to *Trypanosoma congolense* and helminth infection under different diet levels.

Goossens, B., Osaer, S., Ndao, M., Van Winghem, J. and Geerts, S. 1999.
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The susceptibility of Djallonké and Djallonké-Sahelian crossbred sheep to *Trypanosoma congolense* and helminth infection under different diet levels

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Abstract

Forty two Djallonké and 27 Djallonké-Sahelian crossbred sheep were compared during 34 weeks for their disease resistance and productivity in a multifactorial experiment including trypanosome infection, helminth infections and dietary level. Eight treatment combinations were formed in which the two breeds were balanced. Pyrexia was observed following trypanosome infection and was not different between the two breeds. However, a significant higher parasitaemia level, a shorter prepatent period and a lower antibody response in the crossbreds following infection, indicated a significant reduction of the trypanotolerance and confirmed the genetic origin of the trait. Neither helminth infection nor dietary level influenced the onset and level of parasitaemia or the level of antibody response following trypanosome infection. Trypanosome infection, helminth infection and low supplementary feeding caused independently significant reductions in PCV level and weight gain but these declines were not worse in crossbreds as compared to Djallonké. Independently, of the studied factors, crossbreds were generally heavier than Djallonké and also grew faster, especially during the second phase of the study. Crossbreds had significantly higher mean nematode egg output (epg) compared to Djallonké sheep but reduction of epg following deworming was similar in both breeds. The lower epg in the Djallonké breed indicated an innate resistance to helminths and/or more efficient immune response. Trypanosome infection tended to increase epg, confirming the immunosuppressive effect of the former. The higher body temperature in the Djallonké compared to crossbreds suggested a better heat tolerance in the former breed.

From this study it was concluded that Djallonké-Sahelian crossbred sheep inspite of a reduced trypanotolerance and lower resistance to helminth infection, posses a higher potential to intensify mutton production as compared to the pure Djallonké. However, appropriate measures should be taken to limit disease and stress factors in order to optimise production environment for this crossbred sheep. ©1999 Elsevier Science B.V. All rights reserved.

Keywords: *Trypanosoma congolense*; Sheep – Protozoa; Feeding and nutrition; Djallonké-Sahelian sheep

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1. Introduction

The fast growing human population and urbanisation in Africa stresses the need for boosting animal production. Trypanosomosis is still a major constraint for animal production in sub-Saharan Africa. Apart from attempts to reduce the vector and the use of trypanocidal drugs with the risk of inducing environmental pollution and drug resistance, the use of trypanotolerant livestock has been promoted as a more economic and justified way of combatting trypanosomosis. Trypanotolerance has been defined as the ability of certain breeds of ruminants to survive and remain productive in tsetse infested areas without the aid of chemotherapy (Chandler, 1952, 1958; Mortelmans and Kageruka, 1976; Murray et al., 1982). The origin of this trait is still not completely clear but is multifactorial. Decades of natural selection and survival of the fittest may have played an important role in appearance of trypanotolerance (Dolan, 1987). In West Africa, trypanotolerance is related to some breeds of taurine cattle (*Bos taurus*), which are humpless and of smaller size (Chandler, 1952). The mechanism of trypanotolerance in these cattle has been described as a control of anaemia and of the intensity and duration of parasitaemia (Dargie et al., 1979; Murray and Morrison, 1979). Although the N'Dama breed seem to remain productive under natural infection (Agyemang et al., 1992) they remain a very slow reproducer with high age at first calving, long calving intervals and low milk production. Efforts to increase individual production, including the production of crosses with more productive but trypanosusceptible cattle breeds such as Zebu type (*Bos indicus*), are leading to a higher demand on nutritional input and are often more susceptible to endemic diseases. In West Africa, the Djallonke sheep and West African Dwarf (WAD) goats are regarded as trypanotolerant (Mawuena, 1986; Osaer et al., 1994) although the WAD goats seem to be less tolerant than the Djallonke sheep when challenged with the same clone of *T. congolense* (Goossens et al., 1997a) or studied under natural tsetse challenge (Osaer et al., 1999).

Trypanotolerance in small ruminants has been described as an innate ability to remain productive under infection with very low mortality rather than to control the parasitaemia and anaemia (Osaer et al., 1994, 1997; Goossens et al., 1997a). Innate resistance in N'Dama cattle and Djallonké sheep has been proven by the fact that trypanotolerance is expressed already during primary infections (Paling et al., 1991a; Dwinger et al., 1992; Osaer et al., 1994). Due to their small size, Djallonké sheep are prejudged in The Gambia as less productive. Farmers therefore, have shown continuous and growing interest to import Sahel breeds because of their bigger size and expectation of better carcass yields. These trypanosusceptible Sahelian long-legged sheep are subclassified as Touabire or White Arab sheep from the North and Peul-Peul or Fulani from the central region in Senegal (FAO, 1991). The information available on comparing Djallonke and crossbred Djallonke-Sahelian sheep under different stress factors is very limited.

The main objective was to study disease resistance and productivity of Djallonké-Sahelian crossbred and pure Djallonké sheep. Therefore, both breeds were compared in a multifactorial design challenged with trypanosome infection, natural helminth infection and different diets. The responses of both breeds to trypanosome infection would in addition, further elucidate on the existence of a genetic base for trypanotolerance in the Djallonke breed.

2. Materials and methods

2.1. Site

The study was conducted on-station at the coastal site of the International Trypanotolerance Centre (ITC), The Gambia, West Africa. The climate is sub-humid with a mean annual rainfall of approximately 1000 mm. Based on entomological and pathological records, the site is generally considered to be at no risk of tsetse challenge (Rawlings et al., 1993).

2.2. Animals

Forty two pure Djallonké sheep and 27 F1-crosses (Djallonké × Sahelian sheep), either females or castrated males were selected from the ITC flocks following a breeding programme. Sahelian rams originating from northern Senegal were used for F1-crossbreeding after an adaptation period of 1 year. At the start, the experimental animals aged between 6 and 8 months. On average, the F1-crosses weighed 12.6 kg (σ 2.8) and the group of Djallonke's weighed 10.3 kg (σ 2.9) at the start of the experiment. All animals had been raised in absence of trypanosomosis risk, so the possibility of acquired resistance was non-existent. Within sex, animals were divided into blocks according to their age, followed by random allocation to eight different treatments. Assignment to groups was done separately for the two breeds to allow breed comparison. In this way, the number of Djallonke and F1-crosses present per group was balanced according to numbers available. All animals were vaccinated against Peste des Petits Ruminants (PPR-Tissue Culture Rinderpest vaccine, ISRA/LNRV, Dakar) which was repeated every 6 months. They were treated with pour-on acaricide against external parasites (Bayer, Bayticol® 1% pour-on, Flumethrin 1% at recommended dosage) and this was repeated every 4 weeks during the wet season. Prior to trypanosome infection the animals were checked for trypanosomes following examination of the buffy coat by dark ground (DG) microscopy (Murray et al., 1977) and serum was screened for antibodies against *T. congolense*, *T. vivax* and *T. brucei* by the Immunofluorescence Antibody Test (IFAT) (Katende et al., 1987).

2.3. Experimental design and sampling methods

The study started during the rainy season of 1997 (July) and had a total duration of 34 weeks. The design included the factors trypanosome infection (Tc or control), nutritional level (high or low) and natural helminth infection (Deworming or not D), resulting in eight different treatment combinations: 1 (Tc-H-Dew), 2 (C-H-Dew), 3 (Tc-H-nD), 4 (C-H-nD), 5 (Tc-L-Dew), 6 (C-L-Dew), 7 (Tc-L-nD), 8 (C-L-nD). In addition, a breed comparison was possible since the two breeds were allocated separately to treatment groups. Considering this design, 36 trypanosome-infected sheep could be compared with 33 controls, whereby all other effects are corrected for in the analyses. In a similar way, 36 high supplemented versus 33 low supplemented, 39 dewormed sheep versus 30 not treated sheep and 42 Djallonke versus 27 crossbreds were compared. Whether breeds responded differently to one of the above factors, was measured by the interaction breed and each of these factors.

A West African stock of *T. congolense* originating from a clone (SAT86/CRTA/91) from Burkina Faso was used as artificial infection. The stabilate was first expanded in a goat and thereafter the sheep assigned to infection groups (1, 3, 5 and 7) each infected intravenously with 1 ml of infected blood containing 10^5 trypanosomes. Infection took place on 17 July 1997 (Day 0) and all weeks refer to time post *T. congolense* infection. Trypanosome infection was terminated by treatment of all infected animals with trypanocidal drugs (Diminazene aceturate at 3.5 mg/kg bodyweight) at the end of Week 21. The dewormed groups (1, 2, 5 and 6) were given fenbendazole at strategic intervals during the rainy season – end July (Week 2), mid September (Week 9) and end October (Week 16) at the recommended dosage (Panacur®, 10%, Hoechst, Germany, 5 mg/kg bodyweight). The low nutrition groups (5–8) received a restricted supplement based on cotton seed, while the high nutrition groups (1–4) received a supplement based on groundnut cake, cotton seed and rice bran. The low level supplement offered about 40.5% of the required rumen degradable protein (RDP) and 20.7% of metabolisable energy (ME), based on a daily live-weight gain of 50 g/day. The highly supplemented groups received at least five times more to satisfy their demand for growth (Mcdonald et al., 1988). During daytime, all animals were grazing together on natural pastures to allow helminth infection and supplements were fed per group before grazing. Rectal temperatures were taken three times per week during the first 4 weeks. Animals were bled twice weekly between Week 0 and 3 and once weekly between Week 4 and 30. Packed cell volume (PCV) levels were assessed and parasitaemia was measured by the DG method and scored by the method of Paris et al. (1982). Serum was collected from the blood samples taken in Weeks 0, 4, 8 and 12 in order to measure the titre (1/dilution) of trypanosomal IFAT antibodies according to Magnus (1988) using Fluorescein-conjugated rabbit IgG fraction to sheep. Rectal faecal samples were collected from all animals at fortnightly intervals from Week 2 to 34. The number of strongyle eggs per gram faeces (epg) was determined using a McMaster technique with a sensitivity of 100 epg (Thienpont et al., 1979). Body weights were recorded weekly using a Salter Spring balance. After Week 22, all animals were further weighed fortnightly for another 3 months.

2.4. Statistical analyses

Statistical analyses were carried out using SAS (1998) statistical package version 6.12. Continuous parameters were analysed by the general linear model (GLM procedure) as repeated measurements using a mixed model. The model included the following main effects: infection (control versus infected), diet (low versus high), deworming (deworm versus not deworm), breed (Djallonké versus crossbred), sex, block (sex), period (acute: Week 0–5; post acute: Week 6–21; recovery: Week 22–34), week (nested within period), animal (nested within infection, diet, deworming, block, sex, breed, block) and their interactions (two-way, three-way and four-way). The interactions breed × infection, breed × diet, breed × deworming were of high interest in this study since they indicated whether breeds responded differently to the different factors. The other interactions gave evidence of interdependency of the studied factors. The animal effect was regarded as random. Parameters such as prepatent period and daily gain were analysed in a similar model but without the animal effect. Before analysis, egg output data (epg) were subjected to a logarithmic trans-

Table 1
Mortality in Djallonke and F1-crossbred sheep during a multifactorial experiment including trypanosomosis, helminthosis and high or low level of nutrition[a]

Group (total)	No died (breed)	Week	Symptoms	Post-mortem findings
1 Tc-H-Dew (10)	0	–		
2 C-H-Dew (11)	1 (Dj)	19	Respiratory problems, PCV 23	Fibrinous pleuropneumonia, hydropericard, petechial haemoraghes on pericard and endocard
3 Tc-H-nD (8)	0	–		
4 C-H-nD (7)	0	–		
5 Tc-L-Dew (11)	0	–		
6 C-L-Dew (7)	1 (Dj)	2	Lice infestation, weight loss, Diarrhoea, PCV 16	Mild hydrothorax, mild pneumonia
7 Tc-L-nD (7)	1 (Dj)	14	Treated-PCV < 15%	Survived
8 C-L-nD	1 (F1)	13	Weight loss, high epg, PCV 17	Generalised enteritis and inflamed lymphenodes
	1 (Dj)	18	Fever, PCV 28	Hydropericard, petechial haemoraghes on pericard and endocard, mild enteritis

[a] Absolute figures per group, indicate breed, week of death, clinical symptoms and post-mortem results.

formation to approximate a normal distribution. All hypotheses were tested by the *F*-test (Snedecor and Cochran, 1980). Results were regarded as statistically significant when the Type I error probability was smaller than 5%. Means of the different groups are presented as least square means ± standard error (s.e.).

3. Results

3.1. Clinical symptoms

During the observation period four animals died and one sheep needed trypanocidal treatment since its PCV level went below 15% and was consequently withdrawn from the trial. Table 1 gives an overview of groups and numbers died with symptoms and post-mortem observations. Three Djallonke and one F1 sheep died and all four originated from the youngest age blocks at the start of the experiment. All had a history of weakness and a decrease in PCV, however not below 15% and/or showed weight loss and high faecal egg counts. In one case, cowdriosis was suspected (Group 8) however, not confirmed by brain smear.

Following trypanosome infection, rectal temperature significantly increased ($P < 0.001$), with means of 39.56 ± 0.03°C for infected groups versus 39.35 ± 0.02°C for control groups. There was a breed effect ($P < 0.05$) with Djallonke sheep having a mean higher rectal temperature (39.52 ± 0.03°C) than crossbreds (39.40 ± 0.04°C). The interaction infection × breed was non significant (n.s.) indicating that both effects were additive and that two breeds were not responding differently to the trypanosome infection. Neither diet nor deworming had an effect on rectal temperature and also the interactions diet × infection and deworming × infection were not significant. There was an important variation between animals ($P < 0.001$).

3.2. Parasitaemia level and prepatent period

In all trypanosome infected animals (Groups 1, 3, 5, 7) the onset of parasitaemia or prepatent period was on average 8.8 ± 1.7 days. A longer prepatent period ($P < 0.05$) was observed in Djallonké sheep (9.4 ± 0.4 days) compared to the crossbreds (8.1 ± 0.5 days). All trypanosome infected groups had their first parasitaemic peak around Week 3 and showed subsequently a similar pattern up to trypanocidal treatment at the end of Week 21 (Fig. 1(a)). During the period of infection, crossbreds had higher parasitaemia levels ($P < 0.001$) than Djallonke sheep (score: 2.2 ± 0.1 versus 1.5 ± 0.1, respectively) (Fig. 1(b)). In non-trypanosome infected animals, no parasites were detected during the total duration of the experiment. Diet did not influence significantly the prepatent period, despite a tendency to be shorter in the low supplemented groups (8.3 days for low versus 9.1 days for high diet, pooled s.e. = 0.5). Neither the level of diet nor the deworming, influenced the parasitaemia level and none of the interactions were significant.

3.3. Packed cell volume

Mean weekly group PCV levels are presented in Fig. 2. Concurrently with the appearance of trypanosomes, PCV levels dropped ($P < 0.001$) due to infection (means of 22.5 ± 0.1% in infected groups versus 25.2 ± 0.2% in control groups). The interaction period × infection ($P < 0.001$) confirmed an important drop in PCV of 1.7 ± 0.2% due to infection between acute and post-acute phase. Low supplementary feeding also decreased PCV levels ($P < 0.001$), with overall means of 24.9 ± 0.1% and 22.8 ± 0.2% for high and low level diet groups, respectively. The groups receiving higher dietary supplements did not encounter the drop induced by infection, but both effects were acting independently since the interaction diet × infection was non-significant. Deworming had a positive effect on PCV (dewormed 24.7 ± 0.1% and not dewormed 23.0 ± 0.2%) which was most apparent in the subacute phase (interaction period × dewormed; $P < 0.001$). Between breeds, there was no significant difference in PCV level. Crossbreds did not respond differently in terms of PCV changes to the trypanosome infection, diet or deworming compared to Djallonké sheep. In addition, there were no significant interactions between infection, diet and deworming, thus all effects were additional. This was clearly demonstrated by the lowest PCV levels in Group 7 (Tc-Nd-L) (Fig. 2).

3.4. Antibody response

Following trypanosome infection, titres of trypanosomal IFAT antibodies (1/dilution) were measured in the infected groups up to 90 days post infection. Neither supplementary feeding nor deworming, influenced the antibody level significantly despite an initial higher titre for the low supplemented groups (Fig. 3(a)). However, there was a significant breed difference ($P < 0.001$) with consistently lower levels for the crossbreds during the 3 months post infection and a mean titre of 652 ± 140 (1/dilution) in crossbreds versus 1476 ± 105 (1/dilution) in Djallonké sheep (Fig. 3(b)).

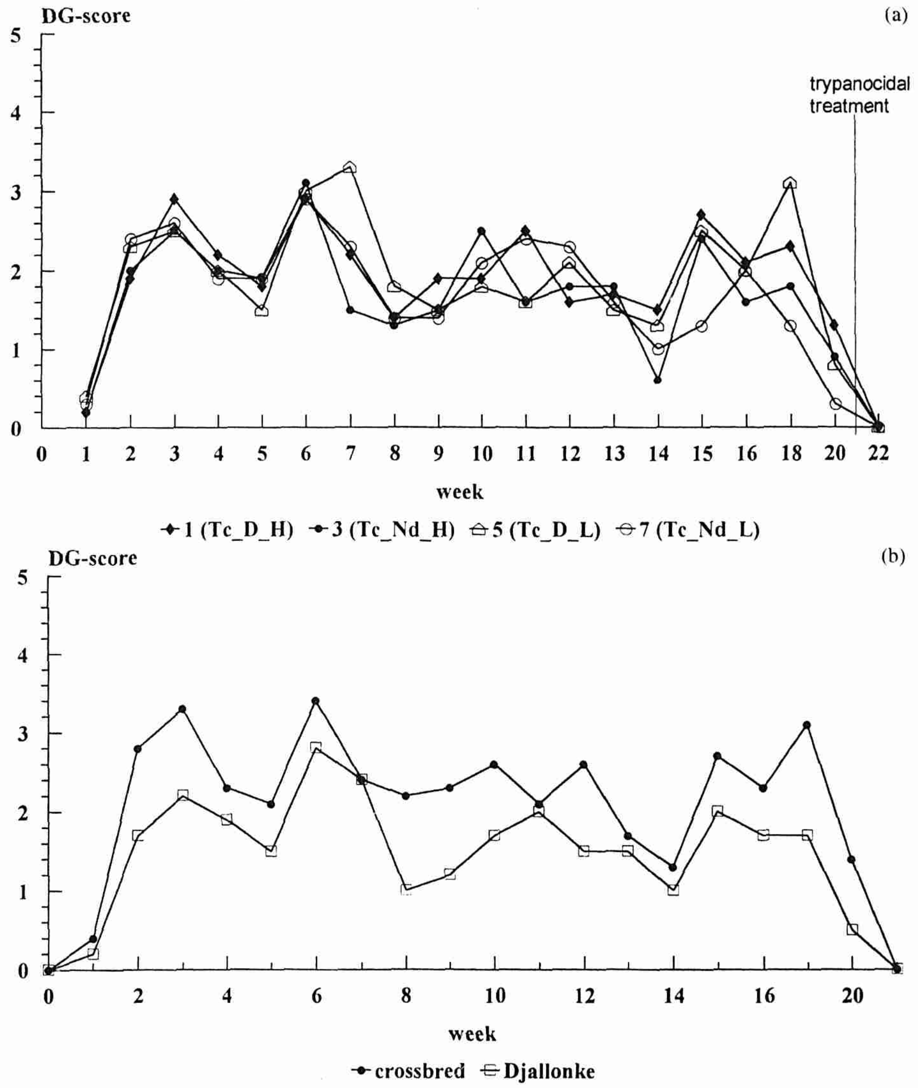

Fig. 1. (a) Parasitaemia levels of trypanosome infected sheep (comparison of four trypanosome infected groups under high or low level of nutritional supplementation and treated or not against helminths); (b) Parasitaemia levels of trypanosome infected sheep (comparsion of pure Djallonke sheep and F1-crosses with Sahelian breed).

3.5. Live weight

Mean weekly body weights are presented in Fig. 4. Trypanosome infection caused a reduction in weight gain of 0.7 kg $\pm$ 0.2 from the start till the end of the infection period (infection $\times$ period; $P < 0.01$) compared to the controls. However, over the entire study period, the difference in total weight gain between controls and infected groups was 1.6 kg $\pm$ 0.2

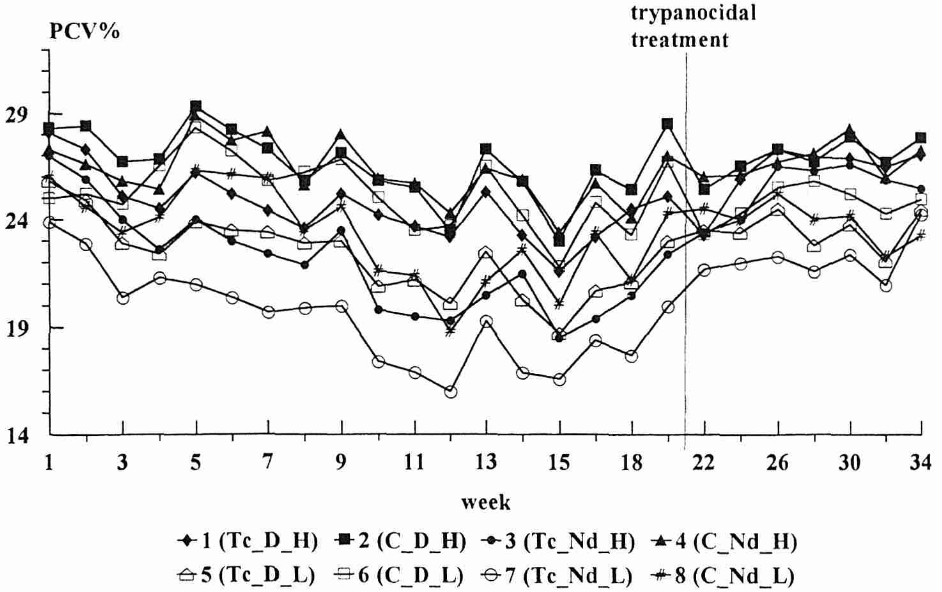

Fig. 2. Mean weekly packed cell volume % (PCV%) in sheep (comparison of eight groups infected with *Trypanosoma congolense* or controls, under high or low level of nutritional supplementation and treated or not against helminths); trypanocidal treatment in Week 21 post-infection.

Table 2
Effect of breed on individual daily weight gain (lsmeans ± s.e.; in g/day) during the period of infection and recovery: comparison between Djallonké sheep and F1-crossbreds (Djallonké × Sahelian)

Breed	Infection period (g/day)	Recovery period (g/day)
Djallonké	23.6 ± 2.4	44.5 ± 3.4
Crossbreds	27.6 ± 3.1	57.9 ± 4.3
Significance	n.s.	$P < 0.001$

($P < 0.001$). Dewormed animals had on average 1.1 kg ± 0.1 better weight gain compared to not-dewormed animals over the whole observation period (deworming × period; $P < 0.001$). High level of supplementary feeding caused a surplus in weight increase of 3.01 kg ± 0.2 compared to the low level (period × diet; $P < 0.001$). There were no significant interactions between trypanosome infection, diet or deworming, thus all effects were acting additive as is demonstrated by Group 7 (Tc-Nd-L) which had the lowest weight increase (Fig. 4). Although the two breeds differed in body weight ($P < 0.001$), the crossbreds did not respond differently in terms of weight changes than Djallonké sheep following trypanosome infection, diet or deworming. To compare growth rates between breeds, an individual daily weight gain (linear regression, $Y = b_0 + b_1$ days) was calculated for the period of infection and recovery, respectively and the estimated regression coefficient (b_1) was analysed in a similar model. During the period of infection (acute and post-acute phase), breeds did not show significantly different growth rates, whereas during the recovery phase, crossbreds had higher ($P < 0.001$) weight gains than Djallonké sheep (Table 2).

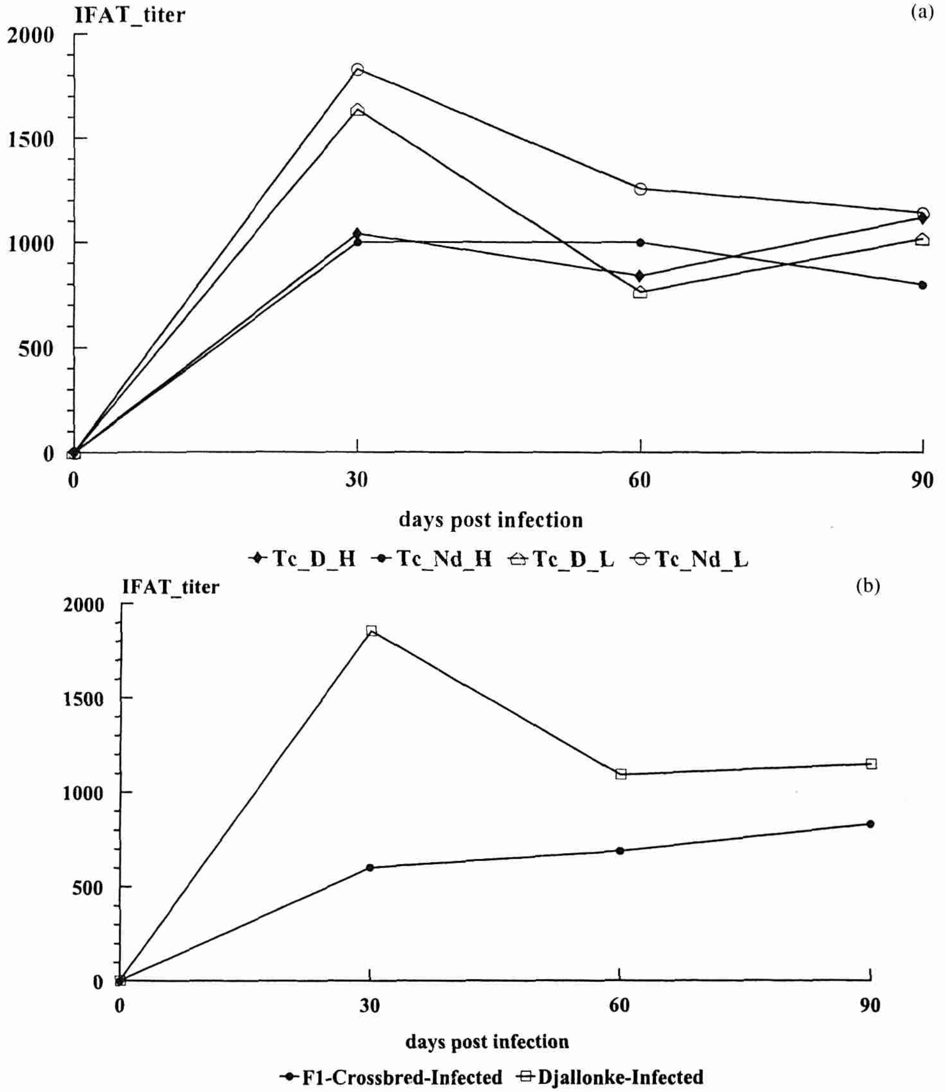

Fig. 3. (a) Mean antibody titers (IFAT) for sheep. Comparison of four groups infected with *Trypanosoma congolense*; (b) Mean antibody titers (IFAT) in sheep (comparison of Djallonke sheep with F1-crosses, with Sahelian sheep infected with *Trypanosoma congolense*).

3.6. Nematode egg excretion

The arithmetic mean weekly numbers of strongyle eggs per gram faeces (epg) are presented in Fig. 5(a). Peak egg excretion was seen for the not-dewormed groups (3, 4, 7 and 8) between Week 10 and 14, which corresponds to the period – end September–mid-October. Group 7 (Tc-Nd-L) and to a lesser extent Group 8 (C-Nd-L) had a second peak in Week 18.

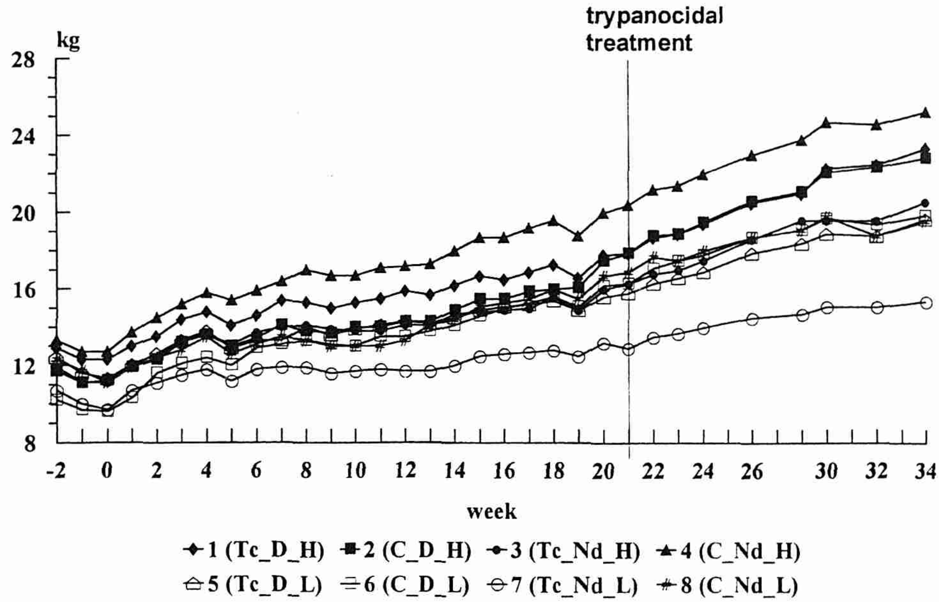

Fig. 4. Mean body weight changes in sheep (comparison of eight groups infected with *Trypanosoma congolense* or not, under high or low level of nutritional supplementation and treated or not against helminths); trypanocidal treatment in Week 21 post-infection.

Epg was significantly decreased by deworming ($P < 0.001$), with a difference in mean epg (log-transformed) of 1.94 between treated and not-treated animals for the period between Week 6 and 21. There was a significantly higher ($P < 0.01$) mean epg for the crossbreds (3.77 ± 0.15) compared to Djallonke (3.30 ± 0.13), illustrated also by the weekly arithmetic means of both breeds (Fig. 5(b)). However, crossbreds did not respond differently to the effects of deworming on epg. Diet had no effect on epg and also did not interact with the deworming. Trypanosome-infected groups tend to have higher epg levels with 3.67 ± 0.21 for infected groups versus 3.41 ± 0.15 for control groups, but the difference was not significant and there was no interference with the deworming effect (interaction; n.s.).

4. Discussion

Primarily, this experiment aimed at evaluating the resistance to the effects of a trypanosome infection in trypanotolerant sheep under basic dietary conditions challenged concomitantly with helminths. A second aim was to make a comparison between trypanotolerant sheep and their crossbreds with trypanosusceptible breeds to further elucidate a genetic basis for trypanotolerance in the former breed.

4.1. Mortality and clinical symptoms

Both Djallonké and crossbred sheep suffered from anaemia and weight losses as a result of trypanosome infection, helminths and low nutrition but mortality due to trypanosomosis

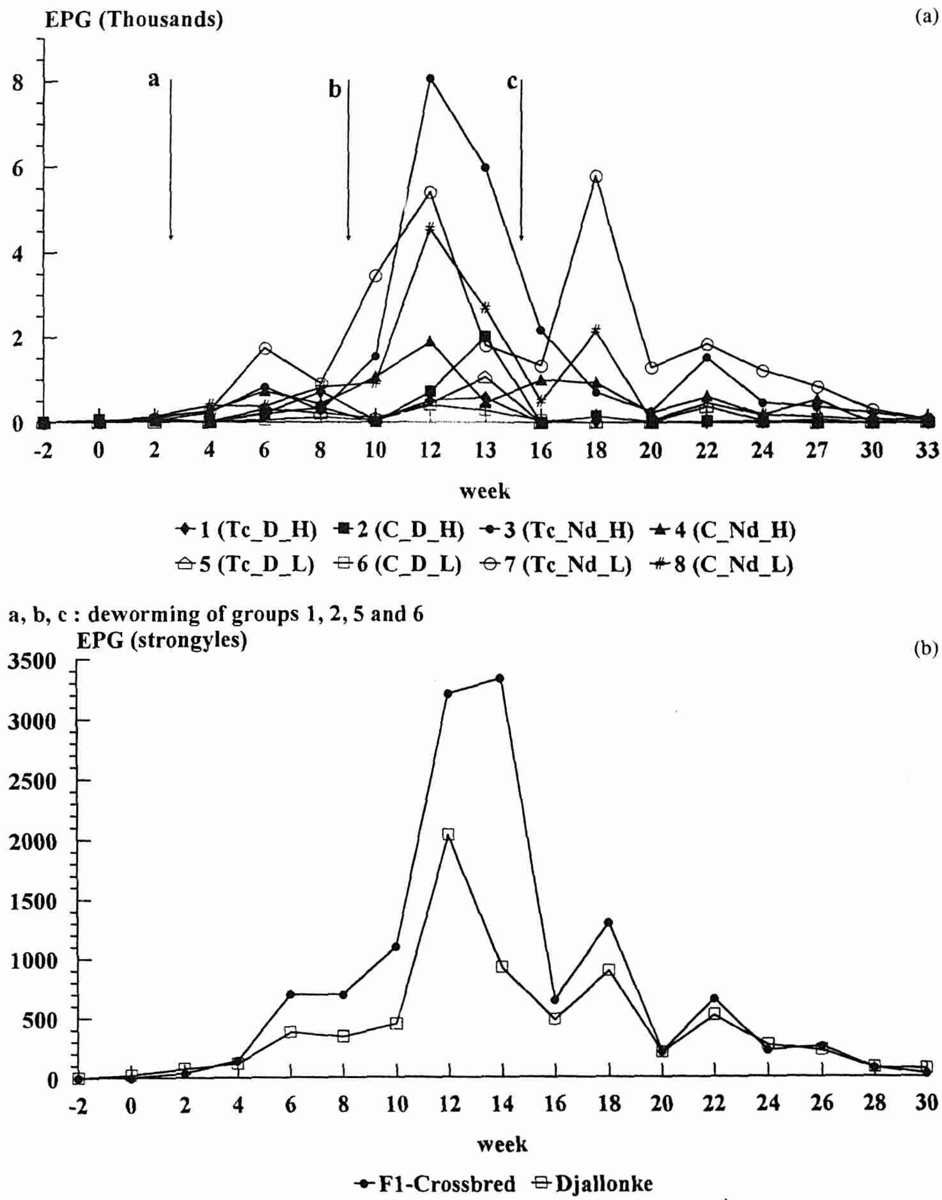

Fig. 5. (a) Mean faecal egg counts (epg) in sheep (comparison of eight groups infected with *Trypanosoma congolense* or not, under high or low level of nutritional supplementation and treated or not against helminths); (b) Mean faecal egg counts (epg) in sheep (comparison Djallonke sheep with F1-crosses with Sahelian sheep).

alone was non-existent. The sheep which died, originated from the blocks with younger age and lowest weight at the start and were kept under low nutritional conditions. The three Djallonke sheep which died, belonged to non-trypanosome infected groups and two were dewormed. Post-mortem findings gave suspicion of infection with *Cowdria ruminantium*, although this could not be confirmed by a positive brain smear. The one Djallonké sheep with PCV below 15% which was given trypanocidal treatment also originated from the block of youngest age, was not dewormed and fed low supplements. The two sheep from Group 8 (C-L-nD) which died were the youngest animals. In the F1-crossbred animal, mortality was clearly associated with clinical helminthosis, whereas for the Djallonke there was suspicion of cowdriosis. It has to be noted that the experimental animals did not have an acquired immunity since they were born during the previous dry season and this was their first contact with helminths. It was suggested earlier that acquired resistance in sheep under these climatic conditions was very low or non-existent (Ankers et al., 1994). Lambs up to 6 months of age were less able to mount a vigorous immune response or acquire protective immunity to helminth infection than older sheep (Lloyd and Soulsby, 1987). Due to the low mortality rates in general, no breed differences could be identified based on these results.

Pyrexia induced by the trypanosome infection during the first 4 weeks confirms the pathogenicity of the clone and is an important factor in the non-specific defence mechanism of the host (Kluger and Rothenburg, 1980). Pyrexia is associated with the peaks of parasitaemia waves during trypanosome infection (Losos and Ikede, 1972; Griffin and Allonby, 1979; Bengaly et al., 1993; Osaer et al., 1997; Goossens et al., 1997b). The higher body temperature found in Djallonké compared to crosses with Sahelian breeds might be due to better thermoregulatory abilities of the former. Heat tolerance, reported in trypanotolerant N'Dama cattle (Greig and Mcintyre, 1979) has also been observed in Djallonké sheep in Gambia (Osaer, unpublished observations). Another contributing factor could be the lower body weights exposed to the same environmental temperatures resulting in higher body temperatures, as was observed in Angora goats (Mcgregor, 1985).

4.2. *Trypanotolerance*

The onset of parasitaemia was delayed in Djallonké as compared to the F1-crosses. Since trypanosomes were inoculated intravenously, no localised skin reaction or chancre influenced the first appearance of parasites as observed in cattle (Murray et al., 1981). The difference in prepatent period between breeds could therefore only be due to differences in rapidity of humoral immune response. The intensity of parasitaemia was also consistently lower in the Djallonké sheep indicating that, even following primary challenge, they are capable to limit parasite multiplication better than crossbreds. Better control of parasitaemia level in trypanotolerant breeds has been shown in comparative studies with more susceptible cattle breeds (Roberts and Gray, 1973; Pinder et al., 1984, 1988; Paling et al., 1991a, b; Dwinger et al., 1992) or susceptible sheep breeds (Toure et al., 1981; Bengaly et al., 1993). One of the underlying mechanisms for better control of the parasitaemia is a superior immune response of the trypanotolerant breeds, feature mainly studied in cattle (Murray et al., 1982; Roelants and Pinder, 1984; Authié, 1994). Pinder et al. (1988) demonstrated an earlier appearance and a higher titre of neutralising antibodies in more tolerant breeds. In the present study, control of parasitaemia and the humoral immune response in general was significantly reduced in the crossbreds as compared to pure Djallonke. The significantly higher level of antibodies in the Djallonke sheep during the first 3 months of infection, explains the consistently lower level of parasitaemia and confirms the superior immune response in this trypanotolerant breed. Despite a better immune response resulting in a controlled parasitaemia, there was no tendency for self-cure as described in trypanotolerant cattle following primary infection (Wellde et al., 1981; Nantulya et al., 1986). Self-cure in Djallonke sheep following a primary infection with *T. congolense* was observed only after 24 months (Goossens et al., 1997a). Self-cure may occur when the antigenic repertoire of that particular serodeme is exhausted (Nantulya et al., 1986). The difference with trypanotolerant cattle is that this process lasts apparently much longer in sheep. Dietary supplementation did not influence the onset or intensity of parasitaemia in the present trial which accords with previous findings (Katunguka-rwakishaya et al., 1995; Wassink et al., 1997). Groups with concurrently higher worm burdens did not express higher parasitaemia levels, indicating there was no immunodepressive effect caused by helminths.

The anaemia induced by trypanosome infection was similar in both breeds. This is in contrast with previous comparative studies when Djallonké are compared with pure Sahelian (Peulh) sheep (Toure et al., 1981). Also trypanotolerant-cattle breeds develop a less severe anaemia following trypanosome infection when compared with susceptible breeds and this anaemia was used as parameter to measure trypanotolerance (Dargie et al., 1979; Murray et al., 1981; Paling et al., 1991b; Dwinger et al., 1992). Trypanotolerant-sheep differ in that respect from cattle because they do not control the drop in haematocrit to the same extent. A better parameter to measure trypanotolerance in sheep is their potential to remain productive under trypanosome challenge (Osaer et al., 1994; Goossens et al., 1997a). Although, higher supplementation increased the level of haematocrit, it could not alter the drop, induced by infection. So, effects of diet and infection were adding up as reported elsewhere (Wassink et al., 1997). Non-dewormed groups had reduced PCV levels. Diet did not alter the effects of helminths on PCV, neither did the trypanosome infection, thus all effects were additive.

4.3. Liveweight gain

Trypanosome infection depressed live weight gain as demonstrated previously in Djallonké sheep under experimental infection (Osaer et al., 1997, 1999) and under natural challenge (Osaer, unpublished results). Earlier work reported on a transient reduction of feed intake and increased maintenance requirements due to infection all resulting in a reduced weight gain (Holmes, 1987; Verstegen et al., 1991; Akinbamijo et al., 1994; Osaer et al., 1999). The weight loss induced by infection was neither altered by dietary supplements nor deworming and was also not worse in crossbreds. Crossbreds are of a larger size, thus breed differences for body weight are obvious. Daily growth rates were higher in the crosses, especially when animals aged over 12 months. However, pre-weaning daily weight gain of crossbreds versus Djallonké was not better and in addition mortality rates were much higher in crosses (43%) versus Djallonké (15%) (Osaer, unpublished results).

4.4. Resistance to helminth infections

This study was carried out during the rains and peak strongyle egg excretion occurred between mid-September and mid-October. The strongyle egg excretion (epg) was sufficiently reduced by the applied deworming scheme, despite the possibility of reinfection. A recent deworming trial carried out in sheep on farm, applying the same treatment scheme as in the present study, resulted in significantly increased growth rates in sheep of all age classes (Osaer, unpublished observations). Trypanosome-infected sheep tend to have increased epg in line with earlier observations by Goossens et al. (1997b) in Djallonké sheep and by Dwinger et al. (1994) in N'Dama cattle. The immunodepressive effect of a trypanosome infection (Mackenzie et al., 1975) may have contributed to a reduced resistance to a subsequent infection with helminths. Diet did not interact with the level of nematode egg excretion. Abbot et al. (1986a, b) observed that lambs on a low protein diet were less able to withstand the pathophysiological consequences of a single infection with *H. contortus*. The mortality in the one F1 animal in the present study was clearly associated with clinical helminthosis, aggravated by low dietary supplements but this was not the case for the one Djallonke sheep in the same group (Group 8) based on clinical symptoms and post-mortem findings. Resistance against gastrointestinal strongyles is based on a history of exposure, age of the host and genetics (Gamble and Zajac, 1992; Baker, 1995). The first two factors in this study where similar for both breeds which implies that only genetics can explain the observed low egg output. Faecal egg output is a repeatable and heritable trait which has been accepted as a quantitative method for selection on helminth resistance (Baker et al., 1992). The lower faecal egg excretion found in the Djallonké as compared to crosses could indicate a better resistance to helminths in the former breed. This study adds some evidence for helminth resistance in Djallonke sheep to the indications already available (Assoku, 1981; Baker, 1995; Goossens et al., 1997b).

5. Conclusions

The present study demonstrates that trypanosome infection, helminth infection and low dietary level cause a considerable reduction in PCV level and weight gain, however, not worse in crossbreds as compared to Djallonké. Some aspects of the trypanotolerant trait were significantly reduced in the crossbreds as shown by a higher parasitaemia level, a shorter prepatent period and a lower antibody response following infection. These observations confirm a genetic origin of the trypanotolerant trait. The higher nematode egg excretion in crossbreds as compared to pure Djallonke is an indicator of innate resistance to helminths and/or more efficient immune response in the latter breed. The higher rectal temperatures found in the Djallonke breed confirms the existence of heat tolerance, inherent to local breeds. It is also concluded that the larger size and better growth rates of crossbreds may result in a higher production potential compared to the purebred Djallonké. However, if crossbreds are to be used to intensify and increase meat production, appropriate husbandry and strict health care measures should be taken to limit stress factors and optimise the productivity.

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CHAPTER 7

NON-DISEASE FACTORS INFLUENCING SHEEP AND GOAT PRODUCTION : FORAGE LEGUMES IN INTENSIVE FEEDING GARDENS AND MANAGEMENT OF FEEDING STRATEGIES

7.1 Introduction and justification for interventions on nutritional management

7.2. Introducing forage legume trees in Intensive Feeding Gardens (IFG)

7.3. Usage of *Leucaena* spp. in strategic supplementation of sheep and goats

7.3.a. High Quality Supplement Block (HQSB) for lactating WAD goats.

7.3.b. Strategic supplementation of lactating sheep for improved lamb survival and performance.

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CHAPTER 7

NON-DISEASE FACTORS INFLUENCING SHEEP AND GOAT PRODUCTION : FORAGE LEGUMES IN INTENSIVE FEEDING GARDENS AND MANAGEMENT OF FEEDING STRATEGIES

7.1. Introduction and justification for interventions on nutritional management

Agriculture in The Gambia is characterised by mixed farming on poor resource farms. Livestock are complementary to the farming system by increasing soil fertility through manure or by providing animal traction. There is an important constraint in providing good quality feedstuffs for ruminants but also other livestock species throughout the year. Forage from rangelands and crop residues (millet, mais) are the cheapest, easy accessible and thus the main sources of feed for village animals. This forage availability fluctuates depending on season and rainfall but provides significant amounts of quality feed for ruminants during rainy and early dry season. Except for groundnut hay which is collected, stored, fed (primarily to draught animals), sold or exchanged, all other crop residues produced (mais, millet, sorhum and rice) are grazed *in situ*. A number of industrial agricultural by-products are available to feed the animals. The most important are groundnut cake, cottonseed, sesame cake and rice bran. Cottonseed is the cheapest, it is locally available, but most of it is exported. Most of the groundnut cake is also exported and although it is still available locally, the price is three times higher than cottonseed. Smaller quantities of sesame cake are available from village based oil-extracting plants. Since most of these by-products are reserved for local sale, export market or reserved for draught animals, small ruminants do not benefit from these high quality feedstuffs and are basically left with natural grazing and some kitchen waste. Green fodder is lacking at the end of the dry season, although sheep and in particular goats are capable of selecting the most nutritious of what is left and make optimal use of it. However, the "standing hay" on the pastures is low in protein, digestible energy and minerals. Therefore, it is unlikely that the nutrient requirements for optimal productivity of sheep and goats are covered by natural grazing. Furthermore, the remaining grazing areas along the river side, which are important during the dry season, are increasingly used for irrigated rice cultivation and not longer accessible for livestock. Together with increased land use for dry season-cultivation of vegetables this is competing with the grazing area for ruminants. Additional constraints are bush fires which, if uncontrolled, destroy much of natural vegetation. In spite of sufficient fodder resources production of small ruminants in terms of weight gain is very low during the rainy season, in contrast to cattle (Osaer et al., 1999; Dwinger et al., 1994). During the rains, sheep and goats are herded for a few hours a day or tethered at the same place throughout the day, because the labour is scarce and is mainly involved in crop cultivation and hence, growing crops need to be protected from animals (Osaer et al., 1999). Both these foregoing management practises lead to inadequate fodder intake, although quality and quantity is good at that time of the year.

In The Gambia, cattle numbers have reached a plateau because of limitation in available grazing lands. Moreover, it has been noted that high mortality rates, drastic reductions in body weights and low milk yields experienced during the dry season are indications that the natural feed resources cannot support the present livestock population. In certain parts of the country, there are early indicators of overgrazing, soil erosion and thus desertification, which indicates that these ecosystems have exceeded their carrying capacities (Schwartz, 1999). Livestock diversification is reflected by increasing numbers of sheep and goats which have different feeding behaviour and are kept under a different management with a lower input.

With growing populations and increased pressure on rural areas, livestock and crop production need to integrate more. Attempts to increase livestock productivity are linked to a certain level of intensification. However, this alone does not reduce grazing pressure and others such as institutional and incentive changes need to be addressed (de Haan et al., 1996). These measures include a better usage and/or distribution of agricultural by-products, such as oilseed cakes or cottonseed. In addition, proper storage and efficient use of crop residues need to be improved.

During the village based research activities it has been observed that poor nutrition and poor seasonal nutritional management are, apart from the disease constraints, major limitingfactors for small ruminant production in The Gambia (Osaer et al., 1999). This lead to the idea to establish a sustainable feeding system using intensified fodder production. One method of fodder production all-year-round in a mixed farming system could be the introduction of forage legumes in Intensive Feeding Garden (IFG). Although at present cultivation of forages for ruminant feeding is not practised by the rural farmers, it would become almost a necessity under an intensified agricultural system. These Multi Purpose Trees (MPT) can provide high quality feed for livestock, firewood and organic manure. In addition, since cultivated forage legumes fix nitrogen (N), they increase soil fertility and subsequently boost crop yields in an alley cropping system. They further increase soil organic matter and soil water holding capacity. Sustainable development of such IFG has constraints, such as scarcity of land and its communal ownership, and the competition with food and cash crops. Protection of young trees and shortage of labour at times of cultivation, planting, weeding and harvesting are additional limiting factors. However, once established, such IFG can be easily managed. Proper usage of produced fodder is subject to adaptations in husbandry to allow supplementation depending on the season and/or category of animals. Fodder produced during the rainy season may be used directly in a cut-and-carry system. Produced fodder may also be stored for usage during the following dry season. This implies proper conservation techniques with a minimum of investment and labour. Most of the investigation reported here is on the possible usage of *leucaena* spp., a forage tree legume with promising features (Shelton et al., 1998).

Changing the existing nutritional management of sheep and goats in traditional rural areas aims at increasing their productivity. Zero-grazing of small ruminants during the rainy season would reduce the labour for herding. At the same time this management reduces contact with infective nematode larvae by avoiding grazing on helminth infested pastures. At the end of the dry season fodder produced in the IFG may be used to supplement natural grazing. It would avoid animals to go further away from the village in search for pasture which brings them into tsetse infested habitat and thus at higher risk for trypanosomosis. Such adapted management implies also a better health care and observation of animals, e.g. if animals are supplemented around late pregnancy and early lactation, whereby both dam and offspring's performance would benefit.

In this chapter the results are presented of some trials to improve fodder supply, storage techniques of the products and feeding strategies at the level of the small scale rural farmer. These results are equally applicable to more intensive production systems, e.g. in the peri-urban areas.

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7.2. Introducing forage legume trees in Intensive Feeding Gardens (IFG)

Methodology

On-station IFG's were established at ITC HQ (coastal station) between 1993-1997 and at the inland station of ITC (Bansang) between 1995-1998. Both on-station fodder-banks were used as example for farmers to show establishment and management of the trees. In addition, different tree species were evaluated, conservation methods of the produced fodder were tested and supplementation trials were carried out. In the rural area where the project's programme on small ruminant health and production commenced in 1995, some villages were identified in 1997 for introduction of fodder trees in IFG. Based on the experience on-station, the concept of IFG was copied to the rural areas. The first community based village garden was established during the rainy season of 1998 and went through one complete dry season (November 1998 - July 1999). A second and third garden are being established at the start of the 1999 rainy season. Pasture improvement trials using local and exotic grasses were initiated at ITC HQ (coastal station) in 1997.

In the on-station and village based IFG, the main forage legume species used is *Leucaena spp.*. *Gliricidia sepium*, *Cajanus cajan* and *Sesbania sesban* are also planted but in lower numbers. Grasses like *Pennisetum purpureum* (Elephant grass), *Andropogon gayanus*, *Panicum maximum* and *Brachiaria mutica* are tested for pasture improvement. For both forage legumes and grasses, the main objective was to study establishment, survival throughout the dry season which is lasting from November till June, and adaptation under the local conditions. Conservation and storage methods of fodder were equally tested.

The first part of the coastal on-station IFG was established in 1993 during the rainy season. Although all species were evaluated at the start, the main species of interest in the following years remained *Leucaena* spp. *Leucaena* is a fast-growing leguminous tree/shrub without special requirements of the soil. It grows in almost every tropical climat and altitudes up to 1000 including slopes and rocky soils. It performs best on well-drained soils but withstands submersion as well as periods of drought of 4-5 months. It is capable of fixing nitrogen through its association with *Rizobium spp.* and brings water and minerals to the upper soil surface through its deep pivot root system. In addition, it provides shadow and its dropping leaves are manure for the soil. This, together with the foregoing factors, makes interplanting with *Leucaena* interesting for crop-production. *Leucaena* trees can be harvested for fodder 1-2 years after planting and can be used in a rotational system for fire wood production after 3 years. *Leucaena* leaves have a crude protein (CP) content varying from 21% to 35% (on dry matter (DM) basis; book values) and production rates of DM/Ha/Year varying from 15 to 35 ton can be expected depending on the soil structure and its fertility. Leaves can be harvested in a cut-and-carry system during most of the year. Leaves can be fed freshly or stored under different ways such as dried leaf mealor as silage. *Leucaena* contains a toxine mimosine and therefore the quantity allowed in a ration is limited. However, when the given amount is gradually increased, ruminants adapt to it.

For the first planting, seeds were collected from local trees. They were pre-treated by soaking them for 2 minutes in hot water before seeding them directly or before preparation of seedlings. Seedlings were prepared during the dry season, starting in April. Once the rains were fully established (mid July) these seedlings were transplanted. Rows were planted with a distance of 1 m between the rows and 50 cm between the different plants. After transplantation the seedlings were not longer irrigated, also not during the subsequent dry season. Expansions

of the coastal IFG with *Leucaena* were done in the rainy seasons of 1994 -1995, as well as the start of the inland station in Bansang. Harvest of twigs and leaves was done every six weeks or when twigs reached a minimum length of one meter. In the rural area, a communal IFG was initiated in the village Missira at the start of the rainy season 1998. The installation was preceeded by talks with the village community in order to explain the methodology and the expected results. A plot was identified with the participation of the villagers, who were also actively involved in the installation of the garden. Fencing wire and nails were provided by the project. A well was already present. Seedlings of *Leucaena* spp. were produced at the garden and transplanted in lines with an interspacing of 2 m at the start of the rainy season (end of June). In the same season maize was interplanted and during the following dry season women used this space to grow vegetables, for which all households in the village had been allocated a plot. This allowed a continuous supervision of the area because women came daily to irrigate the vegetables. Some fruit trees (mango, cashew, papaya) were planted at the start but are not yet fruiting.
In the pasture improvement trial on station, grass species such as *Andropogon gayanus, Panicum maximum* and *Brachiaria mutica* were used in establishing small pastures ($625m^2$) with the aim of using these in rotational grazing systems. This has been done over the past two years and evaluation is still on-going. In addition, *Pennisetum purpureum* or Elephant grass has been tested over a longer period and results are available. In establishing plots of this grass, stem-cuttings (± 25 cm long) were obtained from a commercial vegetable growing farm where it was mainly used as windbreaker between the different vegetable plots. On-station, cuttings were transplanted into different plots by putting them half into the soil at distances of 20 cm. At the coastal station a field of 2500 m^2 and in Bansang a plot of 500 m^2 was planted with this elephant grass . Irrigation was necessary during the dry season since *Pennisetum purpureum* grows normally optimal in areas with a minimum of 1500 mm rain per year. The grass was harvested in a cut-and-carry system and was chopped in parts of ten cm using a cutlass or a hand operated chaf cutter (Alvan Blanch). It was either fed freshly or used in silage as a mixture of *Leuceana* and *Pennisetum*. In IFG's and pasture improvement plots, manuring with a mixture of sheep and goats-droppings was done at regular intervals.

Results on installation, adaptation and production.

At the coastal IFG an area of 2000 m^2 was planted with a 5011 *Leucaena* trees spread over different planting years. A first harvest of leaves could be done during the same rainy season (October) and a second one shortly after the rains (December). In the second year 3 or 4 harvests were possible with the last one in March. Then the trees were allowed to recover till the next rains. Mainly *Leucaena* spp. has been used in different tests and proved to be best adapted to the local conditions. The IFG in the Bansang inland station measures 700 m^2 and contains 1691 *Leucaena* and 97 *Gliricidia.* The IFG in Missira, in the rural area measures 2500 m^2 and has 2392 *Leucaena* trees. This communal IFG in Missira is well managed by the villagers. From the maize planted between the rows of *Leucaena* during the rainy season, the stovers were used as animal feed. Vegetables were planted during the dry season and used for home consumption and surplus was sold on the local market.

From all IFG, harvested forage from the *Leucaena* spp. was fed freshly to certain categories of animals as a supplement during the rainy season. In addition, conservation was done by sun-drying the leaves on drying beds after which it was stored as *Leucaena* leaf meal for supplementing mainly pregnant and lactating females in the dry season (see 7.2). At the village IFG, farmers could obtain forage supplement at most time of the year in order to give to their animals. At the coastal station, silage techniques were tested using a mixture of fresh *Leucaena* leaves and Elephant grass.

Of the pasture plots with the grass species, *Andropogon gayanus, Panicum maximum* and *Brachiaria mutica,* which were established in 1998 rainy season, all the three species survived the long dry season. Establishment of *Andropogon* by transplantation or direct seeding gave equal chance for surviving the dry season. Grazing on these plots was possible with a limited number of sheep and goats till february. Since these plots were in the establishment fase, grazing was limited, but it was intended to use the pastures for rotational grazing in the next rainy season to test carrying capacity of the plots. Thereupon, the pastures needed to be left ungrazed till the next rainy season. These three grasses may also be used in a cut-and-carry system and may possibly be transplanted or seeded under village conditions, although more research is needed to provide details on biomass production and exploitation methods. The fourth grass species, *Pennisetum purpureum*, appears to be less adapted to survive the dry season, except when it is very well protected and irrigated throughout. It might still be an option for peri-urban production sites where there are possibilities for higher investment for continuous irrigation (e.g. solar pump for water pumping) and for mechanically harvesting and silage preparation. If water is not a constraint this species may produce large quantities of biomass year round. Under on-station conditions, *Pennisetum purpureum* grass was harvested every six weeks during the rainy season and every eight to ten weeks during the dry season under irrigation. It was fed fresh to the animals, in bundels or chopped. In addition, silage was made using a fresh mixture of *Pennisetum purpureum* and *Leucaena* spp. (ratio 3:1). The cost of labour was very high and estimated between 0.36 and 0.63 D/kg. Results on analysis of DM and CP of the different fodder (fresh and different conservation products) are presented in Table 1.

The nutritional value of this silage was tested in a small feeding trial. However, adaptation period and duration of this trial were to short to draw any significant conclusions. Consequently, in-vivo digestibility trials have been carried out but are currently under analysis. However, silage in such way is certainly not an option for low-input farming systems but remains an option for the peri-urban production sites where higher investment is possible.

Table 1 : Result on analysis of dry matter (DM) and crude protein (CP on DM basis) of different feeds and their conservation methods:

Feed	DM	CP
Leucaena leaf meal(n = 11)	92.7 %	19.6 %
Fresh *Pennisetum* (n = 15)	22.9 %	14.1 %
Silage of *Leucaena* and *Pennisetum* (n=6)	24.7 %	21.7 %
Groundnut hay - leaves (n= 9)	94.3 %	8.3 %

7.3. Usage of *Leucaena* spp. in strategic supplementation of sheep and goats

7.3.a. High Quality Supplement Block (HQSB) for lactating WAD goats.

Introduction

Strategic supplementation is targeted at selected groups of animals during limited duration according to the specific needs of the animal. Supplementary feeding should be easy to organise and include as much as possible locally available materials in order to keep the cost as low as possible. Development of High Quality Supplement Blocks (HQSB) made with locally available minerals, concentrates and roughage are a suggestion in meeting the above demands. During surveys in the rural areas it was observed that farmers often use common salt (Sodium Chloride) as solely supplement for their small ruminants. This indicates that they are aware of the importance of minerals in animal health and production. Since plants tend to be low in both sodium (Na) and chlorine (Cl), it is a common practice to give salt to herbivores. Deficiencies of Na and Cl result in loss of appetite, weight losses and lowered milk production. Also calcium (Ca) and phosphorus (P) sources are usually very limited in plant material, and therefore it is expected that animals on natural grazing may become deficient for these two minerals, specially in the tropical areas where phosphorus-deficient soils occur (McDonald et al., 1988). These minerals are considered of great economic importance in grazing livestock. Ca and P are both essential for skeleton and teeth constitution and should be provided in a balanced ratio. The former element is also essential for the activity of a number of enzyme systems and in the coagulation of blood whereas the latter element is playing a vital role in energy metabolism. Deficiencies of P have been associated with poor fertility, disturbance of the oestrus cycle but also reduction in milk yield (McDonald et al., 1988). Good sources of Ca and P are bone- and fish meal. Using urea as a high quality, non-protein nitrogen (N) source increases the rumen ammonia pool rapidly and this together with essential minerals will enhance rumen micro-organism growth, both fungi and cellulolytic bacteria. Therefore, the efficiency of cellulolytic forage digestion will increase. The latter will shorten digestion and thus stimulate feed intake. An increased cellulolytic activity results in a higher availability of nutrients from plant material. There will also be an increase in availability of microbial N (Preston & Leng, 1987).

In the present study dried *Leucaena* leaf meal was used as basic element to manufacture HQSB. This is an example of using a product from a high quality legume forage which can be grown in Intensive Feeding Gardens (IFG). *Leucaena* is a source of low fermentable proteins and insoluble carbohydrates which by-pass rumen degradation. It increases the palatability and has a positive influence on the mineral absorption. The objective was to test the feasibility of manufacturing these HQSB using dried *Leucaena* and to measure their potential benefit on animal production. These HQSB were fed to goats in late pregnancy and subsequently early lactation.

Materials and Methods

Manufacturing the HQSB

The HQSB described here contain *Leucaena* leaf meal (50 %), cassava flour (10%), bone meal (15%), urea (10%), cement (10 %) and salt (5%). The dried *Leucaena* is a source of proteins which by-passes the rumen degradation. Cassava flour is added as energy supplier. Urea is a source of non protein nitrogen. Bone meal and salt are sources of mineral supplements (Ca, P, Na, Cl). Cement is added as fixative and should be reduced to a minimum by using compression to make the blocks. *Leucaena* leaves are picked from the branches and dried. Bone meal is prepared by burning bones, collected from the butchers. All ingredients are pounded and sieved after which they are mixed and pressed together to blocks of about 1 kg. The total cost was calculated at around D. 0.95 per kg. This includes drying and pounding of *Leucaena*, burning and pounding of the bones and the cost price for cement, urea and salt. The most expensive component is the urea, which takes about half of the price per kg block. All these ingredients are available to the farmers except for urea, salt and cement which need to be purchased on the market. *Leucaena* is promoted to be grown in IFG nearby the villages. The cost for installing this IFG is not included in the cost for manufacturing the HQSB. Apart from the cost of pounding, other labour-cost in preparing the blocks are not included.

Animals

Fifteen female West African Dwarf goats were selected from the ITC Bansang flock. All animals were kept at the inland station of ITC Bansang. All does were pregnant and supplementation started three to four weeks before the expected date of delivery and continued for two months after delivery. Before the start all animals were bled, PCV was read and the blood was checked for trypansosomosis infection using the Dark Ground buffy coat technique (Murray et al., 1977).

Experimental setup

For the experiment a randomized complete block design was used. Animals were divided into three blocks based on their live weight at the start. The HQSB-**SUP** group consisted of 7 goats and in the **NS** group there were 8 goats. HQSB were fed to the SUP group early in the morning and four hours later animals were released on pasture for grazing. At night they were confined and had access to water. Supplementation of the does started in the third week of March, which is the middle of the dry season. Supplementation was done one month before and two months after delivery. Results for this observation period are hereby presented. Daily intake of supplement, birth weight of offspring and weekly body weights of offspring and does were assessed.

Statistical analysis :

Individual daily weight gains of the kids were calculated as a linear regression ($Y= b_0 + b_1$ days). The latter parameter, kid's birth weight and body weights of the dams were analysed by linear model techniques (GLM procedure; SAS, 1998). The model included the main effects HQSB-supplementation of the dam (SUP-NS), the block, sex and entry type (single, twin) of the kid and for the doe's live weight the animal effect and interaction supplementation*date. All hypothesis were tested by the F-test (Snedecor and Cochran, 1980). Means of the different groups are presented as least square means ± standard error (mean ± s.e.).

Results and Discussion

The feasibility of manufacturing these blocks using available materials is promising. The cost price per block is relatively low, technique of manufacturing is easy and transferable to the farmers. Over the total observation period, it was calculated that an animal consumes on average 100 gram per day of such HQSB. Correct proportion of the components combined with an adapted management synchronises the roughage intake with maximum microbiological activity in the rumen. This results in maximum cellulolytic activity and optimum digestibility of low quality roughage. This would be particularly beneficial at the end of the dry season when there is low quality grass on pasture. The animals received their portion early in the morning, four hours before release on pasture when urea in the blocks was at that time metabolised in ammoniac which became available for the rumen microbial flora. This stimulated their cellulolitic activity which was expected to peak when animals were on pasture, and hence these animals would digest easier the low quality grass, which is high in cellulose.

Higher birth weights of the offspring of SUP goats were noted as compared to offspring from NS goats but the difference was not significant. Mean birth weight of kids were 1.8 kg ± 0.1 vs. 1.6 kg ± 0.1, for the SUP group and NS group, respectively. Single born kids and male kids were heavier than twins and female kids. The effect of the HQSB on daily weight gain of the goat offspring was highly significant ($P< 0.001$). Offspring from the SUP does gained 98.1 ± 4.8 g/day during the first month compared to 65.3 ± 3.6 g/day for the offspring of the NS does. Disregarding the SUP effect, there was a significant block effect ($P< 0.001$) with offspring from the heaviest does (block 3) having higher daily gain than those from block 2 and 1. Mean growth rates in kids during the first month were 67 ± 5.7 g/day, 73 ± 6.0 g/day and 102 ± 4.9 g/day from does of block 1, 2 and 3, respectively. Kids 's growth rates were linked to the weights of the dam and thus indirectly to a better milk production.

The effect of HQSB influenced significantly live weight changes in the does (interaction SUP*date; $P< 0.05$). The NS goats dropped from 27.3 ± 0.3 kg one month before delivery to 23.1 ± 0.7 kg two months post partum (15.5 % loss) whereas the SUP goats dropped from 27.5 ± 0.4 kg to 25.4 ± 0.6 kg during the same period (7.6 % loss). These results indicate that the SUP does were able to liit their weight loss after delivery better than the NS does, despite producing heavier kids. Here the importance of supplementation during the last month of pregnancy should be emphasized since both post-partum weights and kids' birth weights were considerable higher in the SUP dams than in the NS dams. This is in contrast to the following study (7.3b) where strategic supplementation only started the day of delivery. In the latter experiment the SUP dams lost more weight than the NS dams, despite a better growth of their lambs.

The present results indicate a clear beneficial effect of HQSB and its timing on productivity in WAD goats. To be optimal, the supplementation should start a few weeks before the expected parturition and continue till two months after parturition. This would result in higher birth weights, better growth and survival rate of their kids before weaning. In addition, their own body reserves would be better preserved whilst milk production in early lactation would be stimulated. Supplementing these HQSB to goats implies also a change in management *e.g.* keeping animals inside around the parturition in order to increase the beneficial effect. Based on the average consumption of 100 gram per day per animal over a 90 day period this would be a total of 9 kg. This would cost the farmer D. 8.6 per goat. This cost is not high but does not cover the labour for manufacturing the blocks but only those for preparing the ingredients. Like mentioned earlier, urea is the most expensive component. Possibilities to use other sources of non-protein N such as chicken manure should be explored. This is a source of uric acid, which is also rapidly converted into ammonia in the rumen. The availability of chicken manure for the rural farmer should not pose a problem and it would reduce the cost of HQSB considerably. If an optimal effect is to be expected from these HQSB, there is also a certain time schedule to follow. This together with the organisation for manufacturing the HQSB demand training and adaptation from the rural farmer. Farmer associations at village level and creation of credit facility groups could facilitate the process of bulk purchase of ingredients and producing in group a stock of these HQSB in order to fulfill a need for selected animals at certain times. Besides usage in rural areas these HQSB could also be used at the peri-urban production sites in a different supplementation scheme and thus with a higher investment. They could be used in zero-grazing systems targeted at intensive production such as milk production in goats or male fattening schemes. Further trials are envisaged to investigate the effect of HQSB on milk production in goats and on meat production in sheep.

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7.3.b. Strategic supplementation of lactating sheep for improved lamb survival and performance.

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Abstract

Two experiments were carried out in order to test the effect of supplementation of lactating ewes and an adaptation of the management on offspring productivity. Ewes were supplemented with a ration covering half or full their nutritional requirements as from the date of delivery. Locally available concentrate supplements were offered and the roughage was either commercially available or produced in Intensive Feeding Gardens comprising fodder trees. The cost of the supplementation was also calculated. Supplementation was given in 2 phases and differed in nutrient value between phases. The first phase lasted 3 weeks during which time ewes were kept inside, the second phase lasted again three weeks but ewes were released on pasture. There was no offspring mortality in the supplemented groups. In both trials, a clear beneficial effect was seen in the offspring growth rates of the latter groups up to three months, although it was statistically significant in only one trial.

From these results it was concluded that the tested supplementation scheme of lactating ewes had a clear effect on milk production and therefore increased lamb growth rates. Strategic supplementation for sheep at rural level supported by forage legumes grown in Intensive Feeding Gardens may contribute to a better productivity of rural sheep production.

Key words : Djallonke sheep, lactation, *Leucaena spp.*, productivity, lamb performance

Introduction

Offspring mortality in village based sheep in The Gambia is very high (35 %; from 0 to 4 Mo.) and influences significantly the overall productivity of sheep (Osaer et al., 1999b). Births weight and growth rates of lambs from first parity ewes are lower than those from higher parity ewes (Wilson et al., 1985; London & Weniger, 1995; Nash et al., 1996). In addition, such offspring has decreased survival changes till weaning. In general and regardless the dam's parity, low birth weights and poor milk supply in early lactation are major factors in increased postnatal mortality of the offspring (Nash et al., 1996). High but varying pre-weaning mortality rates in Djallonke sheep were also noted during the first month in Nigeria (19 %), between birth and three months in Togo (12 %) and from birth till two months of age in Ghana (11-13 %) (Otesile and Oduye 1991; Missohou et al., 1989; London and Weniger, 1996). In Senegal, Fall et al. (1982) found higher mortality rates in offspring between birth and 4 months, varying between 14 and 54 % depending on the year of observation in a situation comparable to the Gambian village setting. A constant finding, however, was a higher mortality during the rainy season, in twin lambings and in first parity ewes (Fall et al., 1982). Main reasons for high offspring mortality are found in the extensive management system. Mating is uncontrolled. Consequently inbreeding occurs and too young animals get pregnant, resulting in abortions and premature births. Grazing or herding management of sheep is non existent during the dry season, when they roam freely on the nearby fallow land or cropping fields. With the dry season progressing fodder availability around the villages becomes scarce and sheep have to go further in the bush and in tsetse infested areas like the swamp area, increasing the host-vector interaction and thus higher risk of trypanosomosis (Osaer et al, 1999b). Losses due to predators, theft or non-return from the bush are estimated high, specially in the young age-categories (Osaer et al., 1999b). In this traditional management, there is no supplementation for sheep and no special care for ewes in late pregnancy or in lactation. Since mating is not recorded, it is difficult to predict lambing dates and lambing therefore often occurs in the bush. Housing or night shelters are basic or non exisiting. Inspite of availability of more and better quality fodder on natural pastures during the rainy season, sheep loose weight (Greenwood et al., 1989; Osaer et al., 1999b). One of the factors contributing to this is the poor grazing management during the rains: sheep are kept confined till noon, and thereafter they are tethered at the side of the cropping fields or may occasionally be herded in groups by herdsman or children (Osaer et al., 1999b). These practices are done in order to protect crops from damage or because labour is not available, but limit considerably the grazing time or acces to sufficient good quality grass. There is a possible benefit from supplementation of sheep in both the dry and rainy season. Under extensive management productivity can be improved by simple and low cost interventions in the fields of disease control, nutrition and breeding management (Symoens and Hardouin, 1988).

A possible low cost intervention for fodder supply is the use of legume forages or Multi-Purpose Trees (MPT) planted in communally owned Intensive Feeding Gardens (IFG). Based on operational IFG's the following experiments were designed to study possible usage of the forages produced. These include strategic supplementation and a better observation of sheep around the time of lambing. This may result in reduced offspring mortality and increased growth rates. These low- cost interventions should be adaptable to the rural areas and acceptable by the farmer.

Material and Methods

Two experiments were carried out in The Gambia, West Africa. The first study (expt. A) was situated on-farm in the peri-urban area (40 km from the capital, Banjul) while the second study (expt. B) was carried out on-station in Bansang (320 km from the capital). Both studies were carried out in the early and mid dry season. (December 1998 to April 1999)

Experiment A

Experiment A was carried out on a farm with an area of 100 ha. Natural pasture grazing consists mainly of *Andropogon* grass. Livestock management on this farm is extensive and comparable to low-input farming system. The tsetse challenge in this area is low with occurrence of only *Glossina palpalis gambiensis* (Rawlings et al., 1991, 1993). For this study, twenty Djallonke dams were used of zero parity,were oestrus synchronised by the 'ram effect' and mated by Djallonke rams. The ewes were divided at random into supplemented (SUP-10 ewes) and non-supplemented group (NS-10 ewes). Five ewes out of the selected group gave birth to a live lamb which died within 24 hours and were consequently omitted from the trial. Eight ewes remained in the SUP group and 7 in the NS group. The experiment started at the day of parturition which was also the start of supplementation. All ewes were kept inside the day of lambing and released for grazing the next day. Ewes of the supplemented group started to receive their supplementation on the day of lambing. The ration offered consisted of dried *Leucaena spp.*- leaf meal (LL, 150 gram) and groundnut hay (GNH, 500 gram). This combination provided the dams with 50 % of the required digestible crude protein (DCP) and

44.2 % of the required metabolic energy (ME). These requirements were calculated for a lactating dam with an average bodyweight of 23 kg (10.4 Mj ME and 83.4 g DCP). The balance was supposed to be covered by natural grazing. The LL was given in the morning before release on pasture and the GNH was given in the evening upon return from pasture. This combination was fed for 21 days after which date the GNH was stopped but LL continued for another 21 days. The latter supplement covered 22 % of the required DCP and 15 % of the required ME. The LL was obtained from an operational Intensive Feeding Garden (IFG)containing *Leucaena* trees, of which the leaves were sun dried and stored. The cost for this supplement was calculated and details are given in Table 1. For the production of one kilogram dried *Leucaena* a cost price of D. 5 (D=Dalasis; 1US $ =D. 11) was taken. This includes the collection of the leaves, spreading for drying and collecting in bags. This price does not include the installation of the IFG itself. The commercial price for groundnut hay as paid for by the research centre is D.1 per kg. Since ground nuts are grown in most parts of the Gambia, this by-product may be available for the small scale farmers at a lower cost. For the rural farmer it then involves labour for collecting and creation of a storage space. The average cost per ewe/day for supplementation was calculated at D.1. Lambs from the supplemented dams remained inside till day 21 and were then allowed out on pastures together with their dams. The non-supplemented control dams were subjected to an extensive management system. The dam was kept inside for one day and thereafter released on pasture, with the lamb free to follow the mother. No supplementary feed was given. One herdsman was taking care of the supplementary feeding and the herding on pastures.

Experiment B

This experiment was carried out at the Bansang station of the International Trypanotolerance Centre (ITC) in The Gambia. The tsetse challenge is low and animals are therefore at low risk of trypanosomosis (Rawlings et al., 1993). Eighteen Djallonke ewes were used. Four sheep had zero parity, the remaining with a parity of 2 or higher. Half of them were allocated at random to the group with supplementary feeding (SUP group), the other half was kept as control group (NS group). The dams were oestrus synchronised by the ram effect and Djallonke rams were used for mating. Three dams gave stillbirths and were not included in the further trial. There were 9 remaining dams in the SUP group and 6 in the NS group, with in each group two first parity ewes. As in experiment A, the supplementation started at the day of parturition which is the start of the trial. All ewes remained inside at the day of lambing. The ewes from the SUP group remained inside afer delivery for 21 days during which they were stal fed. The applied schedule of supplementation was similar as in experiment A, but the offered rations were different. During the first 21 days, the SUP dams received a mixture of 150 gram cotton seed, 200 gram rice bran and an amount of 1.5 kg fresh mixture of 'cut and carry' *Pennisetum purpureum* grass and *Leucaena spp.* (ratio: 65-35 %). This combined ration provided the dams with the required DCP and ME calculated for a lactating dam of 21.5 kg bodyweight (9.9 Mj ME, 79.5 g DCP). At day 22, the dams were released for grazing but concentrate (cotton seed and rice bran) continued for another 21 days. The latter supplement provided in 29.5 % of the required DCP and 33.2 % of the required ME. The cost for this supplement was calculated and details are given in Table 1. For the cut and carry fodder, a price per day was calculated (D.5), covering only the labour cost per day to cut and carry bundles of *Pennisetum* grass and *Leucaena* leaves to the animals, but not including the installation cost of the IFG. Cotton seed and rice bran are locally available at a commercial price of D.1/ kg and

D. 0.9/kg, respectively. In that way, an average cost per ewe/day for this supplementation scheme was estimated at D.0.6. Lambs of the SUP dams remained inside together with their mothers till day 21, after which they joined their dams on the pastures. The NS dams were subjected to the same extensive management as described in experiment A. As from week seven onwards, all dams and lambs were treated equally and did not receive supplementary feed any longer. One herdsman was involved to take care of stalfeeding and herding.

For both experiments a routine preventive treatment consisting of a parenteral injection of Penicilline LA (1 ml/ 10 kg of bodyweight) was given after delivery. In case of a sick animal, body temperature was recorded and a blood sample was taken to check for trypanosome infection by (DG) buffy coat technique (Murray et al. 1977), followed, if needed by the trypanocidal drug Diminazene Aceturate (Berenil®, at 7 mg/kg body weight).

Traits recorded and statistical analysis.

Offspring born were weighed within 24 hours and thereafter once weekly till three months of age. Dams were weighed monthly. Lamb body weights at 1 and 3 months, their individual daily weight gains between 0-1 month and between 1-3 months were calculated as a linear regression ($Y= b_0 + b_1$ days). These parameters as well as body weight of the dams were analysed by linear model techniques (GLM procedure; SAS, 1998). The model included the main effects supplementation of the dam, sex of lamb and entry type (single, twin) and the interactions supp*entry type and supp*sex, and for dam's live weight the animal effect. All hypotheses were tested by the F-test (Snedecor and Cochran, 1980). Means of the different groups are presented as least square means ± standard error (mean ± s.e.). Mortality rates (up to 90 days) were compared between treatment groups using Chisquare (Chi^2) tests.

Results

Experiment A

Eleven dams delivered in a 18-days period and the remaining four during the two following weeks, implying that these dams conceived in the 2nd oestrus cycle. In the NS group (7 ewes) there were eight lambs whilst in the SUP group (8 ewes) there were 11 lambs born alive. In the NS group two lambs died, a single and a twin lamb with an exit age of 53 days and 40 days, respectively. Arithmetic mean birth weights (sd) were 1.63 kg (σ=0.4) for single born lambs and 1.51 kg (σ=0.3) for twin lambs. Supplementation of the dams had a positive, but non significant effect on daily growth of offspring until 1 month, between months 1-3 and over the whole 3-month period (see Table 2). Entry type had a signifcant effect on the daily gain. Irrespective of the supplementation, daily weight gain in single lambs (91.7 ± 8.7 g/d) was much higher during the first month than in twins ($P < 0.01$) (49.8 ± 13.7 g/d). This difference due to the entry type remained for the growth between 1-3 months ($P< 0.01$) and body weight at 1 and 3 months, respectively ($P<0.05$). Differences due to sex were not significant for daily gain nor for body weight at 1 month and 3 months. All other interactions were not significant (supp*sex and supp*entry type).

Mean weight changes of the dams during the 3 months following parturition (data not shown) indicated significant differences between the NS and SUP group ($P< 0.05$): between one and three months post-partum, the mean weight loss in the SUP group was 1.4 kg and in the NS 0.6 kg. There was an important individual variation in dam's body weight ($P< 0.001$).

Table 1. : Cost of supplementation of lactating Djallonke ewes with different concentrates and roughage.

Experiment	supplement (duration in days)	daily ration per ewe	total amount (price[2]/kg or price/day[3])	total cost per ewe	cost/per ewe/day
A 8 ewes	Leucaena leaf meal (42 days)	150 g	50.4 kg (D. 5/kg)	D. 31.5	D. 1.0
	groundnut hay (21 days)	500 g	84 kg (D. 1/kg)	D. 10.5	
B 9 ewes	cotton seed (42 days)	150 g	56.7 kg (D. 1/kg)	D. 6.3	D. 0.6
	rice bran (42 days)	200 g	75.6 kg (D. 0.9/kg)	D. 7.5	
	cut and carry[1] (21 days)	1.5 kg	283.5 kg (D. 5/day)[3]	D. 11.6	

[1] : a fresh mixture of *Pennisetum purpureum* and *Leucaena* spp. in a ratio: 65%-35%.
[2] : Price in Dalasis - 1 US$ = ± 11 Dalasis
[3] : cost to 'cut and carry' one day amount of the mixture of *Pennisetum purpureum* and *Leucaena* spp.

Table 2 : Effect of supplementation of the ewe on lamb body weight (KG) at 1 and 3 months and lamb daily weight gain (DWG) between 0 and 1 months, between 1 and 3 months and between 0 and 3 months of age (g/day). Results for experiment A and B presented as LS means ± s.e.

	Experiment A			Experiment B		
TRAITS	SUP group (n = 11)	NS group (n= 8)	*sign.*	SUP group (n=10)	NS group (n=6)	*sign.*
weight at 1 month (kg)	4.04 ± 0.4	3.4 ± 0.5	*ns*	4.7 ± 0.5	4.9 ± 0.7	*ns*
weight at 3 month (kg)	7.7 ± 0.8	7.1 ± 1.2	*ns*	8.2 ± 1.1	7.9 ± 0.8	*ns*
DWG 0-1 months (g/day)	77.9 ± 9.6	63.6 ± 12.6	*ns*	69.0 ± 12.9	36.9 ± 18.9	*ns* (P=0.11)
DWG 1-3 month (g/day)	60.6 ± 11.6	52.2 ± 15.6	*ns*	63.7 ± 6.5	41.8 ± 9.5	$P < 0.05$
DWG 0-3 month (g/day)	68.0 ± 7.0	58.0 ± 9.5	*ns*	59.0 ± 8.3	31.9 ± 12.2	$P < 0.05$

Experiment B

All dams became pregnant and delivered over a time span of three weeks. Three dams gave stillbirths. In the SUP group (9 ewes) there was one twin and thus 10 lambs born alive. In the NS group (6 ewes) six single lambs were born alive of which one died at an age of 86 days. The birth weights of the lambs were considerably higher than in experiment A with arithmetic means (sd) of 2.34 kg (σ=0.5) and 2.3 kg (σ=0.1), for single and twin lambs, respectively. More obviously than in trial A, there was a positive influence of supplementation of the dams on daily growth of offspring during the first month, although this difference was not significant (see Table 2). The supplementation of the dam had a significant positive effect on daily weight between months 1-3 or between 0-3 ($P<0.05$). Moreover, there was a positive interaction between sex*supplementation effect ($P<0.05$) indicating a better effect of supplementation in female as compared to male lambs with means for female lambs of 61.0 ± 9.5 vs 19.2 ± 13.1 g/day, and for male lambs of 66.4 ± 7.6 vs 64.5 ± 10.3 g/day for SUP and NS groups, respectively. Regardless the supplementation effect, male lambs grew better than the females during months 1-3 ($P< 0.05$).

Mean weight changes of the dams during the 3 months post parturition (data not shown) indicated differences between the NS and SUP group ($P< 0.001$). The body weights in the SUP group remained stable between 1 and 3 months post parturition with a mean change of 0.02 kg, whereas a mean increase of 1.7 kg was observed for the NS group. There was an important individual variation in dam's body weight ($P< 0.001$).

Discussion

Results from both experiments indicate that supplementation of dams in the early lactation period has a beneficial effect on the offspring. This was proven by zero mortality and higher growth rates of offspring in the supplemented groups compared to those from the non-supplemented dams. However, not only the supplementary feeding, but also the improved management with better observation of animals have contributed to this improved lamb performance. Costs for supplementation are low and could be adopted in different production systems.

Djallonke sheep possess a high production potential even under extensive management. Several reports exist on lamb performance of the Djallonke sheep in different countries of West-Africa indicating some variation within the breed, which can also be influenced by different climatological conditions (Cameroun, Symoens & Hardouin, 1988;The Gambia, Osaer et al., 1999b; Ghana, London & Weniger, 1995; Senegal, Fall et al., 1982; Ivory Coast, Yapi-Gnaore et al., 1997a, b). Despite the high potential of the breed, an extensive management system is faced with problems such as high perinatal and pre-weaning lamb mortality. According to a 4-year survey on village based sheep production in The Gambia a mean lamb mortality of 35 % between 0-4 months was observed (Osaer et al., 1999b). An important proportion of these mortalities were due to disease but exits due to losses in the bush and/or predators were also found important. A study in Nigeria revealed similar high mortality rates in lambs under extensive management. Lambs were abandoned by their dams after birth in the bush because of lack of mothering ability, weakness of the lambs to follow or presence of predators (Otesile et al., 1991). The authors suggested that keeping ewes and lambs inside for one week on zero-grazing and concentrates would reduce these kind of losses.

Although overall offspring mortality in the two experiments was low, it only occurred in first parity sheep of the NS flocks. The lower birth weights in those lambs contributed probably to this mortality, together with their lower growth rates. Regardless the parity of the ewe, the effect of supplementation seems to have resulted in a positive effect on the milk production as reflected by the better survival and growth rates of the lambs in SUP groups of both experiments. In addition, the change in management whereby the lambs of SUP ewes were under continuous supervision during the first 3 weeks and not loosing extra energy by following the dams, contributed to their better performance. However, whilst their lambs grew better, the SUP dams in experiment A appeared to loose more weight than the NS dams in the 3 months following parturition. The ewes of both groups did the same walking parcours during grazing, thus the difference in weight loss could only be ascribed to their level of milk production. At the start of the lactation period animals are usually in a negative energy and nitrogen balance and if intake of nutrients is not sufficient, body reserves are catabolised resulting in weight losses. The nutrient requirements of the SUP ewes were not sufficiently covered by their intake because they had a higher milk offtake, as was reflected in the higher growth rates of the their lambs. In contrast, the NS group had a lower milk offtake as reflected in poorer lamb growth rates, but their weight loss was less. The walking exercise of the lambs of NS-ewes as from day 3 onwards was an extra energy cost, which negated their growth but made them also less fit to suckle, in contrast to those from the SUP group.

In experiment B the effects of supplementation were more prominent with significant increased lamb growth rates. There were no weight losses in the dams from both groups, but similar differences as in experiment A were noted. The NS dams showed an increased body weight during the post partum period, while the SUP ewes kept their body weight stable. In contrast to experiment A, the ration of the SUP ewes covered the needs for milk production and could prevent a weight loss. The SUP group was stal-fed in the first period, hence they did not lose extra energy for walking the grazing parcours unlike those in experiment A. Similar as in experiment A, the difference in weight changes between the NS and SUP ewes was mostly related to their respective levels of milk offtake. Apart from supplementation, different management systems may influence lamb growth (London & Weniger, 1995). Offspring from sheep under a semi-tethering/confinement system had a lower daily weight gain as compared to those under a free range management system. The former management resulted in a lower quantity and accessibility to fodder plants resulting in a lower milk production. This was also observed to a certain extent by Osaer et al (1999b). Based on these observations it would appear that a free ranging management is a better option for sheep production.

Reports on milk production in Djallonke sheep demonstrate means of 57 and 86 kg, with one or two lambs respectively, over a lactation period of 105 days of which the production in the first month is 37% of the total production (Amégée, 1984). During their first weeks of life, lambs are solely depending on milk. If milk production is optimal during the first three to four weeks, it will influence positively early growth rates and their overall productive performance later on, such as weaning weight and age at sexual maturity (London & Weniger, 1994; Osaer et al., 1999a; Berger and Ginistry, 1980). The present results have proven that supplementation increases the milk production in the dam but the availability of such for the rural farmer needs to be ascertained. In experiment A some of the supplements derived from an IFG. Ongoing trials at village level establishing IFG, mainly planted with *Leucaena spp.* are evaluating the feasibility of this intervention. The Gambian farmers have accepted the concept of IFG, which is communally owned and managed by themselves. Such interventions will demand a farmer controlled approach in order to make it sustainable. The groundnut hay is available in the rural areas and cost may be lower than estimated in this study. The concentrates used in experiment B may be bought at the local market at reasonable prices while the cut-and-carry fodder is also a product from the IFG. Only the labour cost was included in the cost of IFG-derived products . The total cost involved in the supplementation of the ewes over a limited period is very low. Since most farmers have one or two animals at the time to be supplemented, communally based credit facilities could support the farmers in organising such operations.

The present results have proven that when ewes are supplemented and/or kept confined during early lactation, it is beneficial for the growth rates and survival of the offspring, not only during the period of supplementation but at least till 3 months of age. The low cost involved and the high potential benefit makes this intervention highly recommendable for the rural farmers. Moreover, the availability of the used feedstuffs makes this scheme of supplementation easily adaptable at rural farm level. Future on-farm trials will be carried out to test the sustainability of this intervention.

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CHAPTER 8

GENERAL DISCUSSION

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TRYPANOTOLERANCE IN DJALLONKE SHEEP AND WEST AFRICAN DWARF GOATS.

The series of studies in chapter two described the effects of either a single *Trypanosoma congolense* inoculation or natural infection with trypanosomes on the health of Djallonke sheep and West African Dwarf (WAD) goats. Clinical parameters, haematological changes and body weight changes were evaluated. These studies gave evidence of the trypanotolerant trait in these indigenous West African dwarf breeds of sheep and goats.

Artificial infections with *T. congolense* (2.1- 2.2.).

The severity of anaemia and the parasitaemia level showed that the East African *T. congolense* strain (Ea-Tc) was less virulent than the West African strain (Wa-Tc) but both were highly pathogenic, as reported in susceptible Zebu cattle earlier (Dwinger et al., 1992). Goats suffered more from single trypanosome infection than sheep, especially in terms of development of anaemia (2.1). This was further confirmed in the study on haematological changes and immune response following trypanosomosis (2.2). Following primary challenge with 2 different *T. congolense* strains of different pathogenicity, all sheep and goats survived while body weight changes during the 12 weeks observation period were slightly lower but not significantly different from the uninfected controls. It was observed that Djallonke sheep expressed a higher degree of trypanotolerance than the WAD goats following artificial infection with *T. congolense*. Based on these controlled studies, it was suggested that the mechanism of trypanotolerance in sheep and goats might be different from the one described in trypanotolerant cattle.

Trypanotolerance in cattle has been described as a combination of both innate and acquired resistance, of which the latter depends on previous exposure to trypanosomosis (Murray et al., 1981, 1982; Paling et al., 1991a). Trypanotolerant cattle are also able to limit the level and duration of the parasitaemia and have the capacity to resist or control the anaemia (Dargie et al., 1979; Murray et al., 1982). The controlled studies in chapter two demonstrated that the mechanism of trypanotolerance in sheep and goats differed from those in trypanotolerant cattle.

Very low mortality rates together with the ability to maintain positive weight gain following infection were considered as good indicators of trypanotolerance in small ruminants, confirming the earlier findings of Mawuena (1986, 1987). The first study (2.1) demonstrated that the degree of resistance in both Djallonke sheep and WAD goats was innate and not acquired, since they were born and raised in a tsetse free area and had never been in contact with trypanosomes before. Since the results under chapters 2 to 5 did not provide evidence in the Djallonke breed nor in the WAD goat for controlling the duration of parasitaemia nor the severity of anaemia following trypanosome infection, trypanotolerance in small ruminants may be better described as 'resilience' to infection rather than 'resistance'. Resilience has been defined by Woolaston and Baker (1996) as the ability of the host to maintain a relatively undepressed production level under parasite challenge, whereas resistance implies that the host suppresses the establishment of the parasites or even eliminates them.

In the studies under chapter two it was demonstrated that Djallonke sheep and moreover WAD goats have great difficulty controlling the level of anaemia as compared to N'Dama cattle. Factors responsible for anaemia are haemolysis and erythrophagocytosis, however their sequence and mutual dependency are not completely clear. According to Anosa (1988a) the most important one is haemolysis resulting in extravascular erythrophagocytosis. More recently it was suggested that the process leading to haemolysis starts with intra vascular erythrophagocytosis as an auto-immune process when red cells are coated with parts of lysed trypanosomes and consequently recognised by the phagocytes as foreign bodies (Uilenberg, 1998). The development of the anaemia following trypanosome infection depends on the rate of red cell destruction and the compensation by active erythropoiesis. As long as no equilibrium between these is reached, the infected host remains anaemic. In this trial, Djallonke sheep became anaemic but responded quickly with a marked erythropoiesis, as indicated by a macrocytic, hypochromic anaemia. The leucocytosis which became apparent in the chronic phase could have been an indicator of increased marrow activity, because a neutrophilia often accompanies a regenerative anaemia (Morris & Dunn, 1992). The anaemia observed in the goats in the acute phase following infection was normocytic and normochromic, indicating a non-regenerative anaemia. Moreover, the anaemia was not accompanied with a persistent leucocytosis as was observed in the infected sheep. Whereas leucopenia is usually observed with trypanosomosis (Anosa, 1988b), leucocytosis seems to be a feature of trypanotolerant breeds (Adah et al., 1993; Ogunsanmi et al., 1994; Paling et al., 1991b). This was corroborated with the results in this study and highlighted the higher degree of resistance in sheep as compared to goats. The erythropoietic response was earlier and faster in sheep than in goats and this resulted in a less severe drop of erythrocyte values (PCV, RBC and Hb) in the former species. The activity of the haematopoietic response during trypanosomosis in breeds of sheep and goats has been related to their degree of trypanotolerance (Kaaya et al., 1977; Griffin & Allonby; 1979; Whitelaw et al., 1985; Adah et al., 1993). A weak erythropoietic response or dyshaemopoiesis has also been reported in trypanosusceptible breeds of cattle as one of the reasons of persistent anaemia (Losos et al., 1973; Valli et al., 1978; Naylor, 1971). However, trypanotolerance in N'Dama cattle has been primarily related to the control of the parasitaemia and not to a more efficient erythropoietic response (Dargie et al., 1979; Murray et al., 1982; Authié et al., 1993). In addition, there seems to be no correlation between the parasitaemia and the severity of anaemia or other parameters, indicating that both are regulated by different mechanisms (Authié et al., 1993 ; Paling et al., 1991b). Similarly as stated for trypanotolerant cattle, the present results do not indicate a correlation between parasitaemia control and development of anaemia in trypanotolerant small ruminants, feature which was confirmed in the comparison between Djallonke and Sahelian x Djallonke crosses (chapter 6). For both breeds there was a clear-cut difference in ability to control parasitaemia but no difference in severity of anaemia. Djallonke sheep control the onset and intensity of parasitaemia significantly better than the Sahelian-Djallonke crossbreds, providing evidence for a genetically controlled mechanism, similar as in cattle.

The important role of the immune response in resistance to trypanosomosis was initially thought to be the result of an earlier appearance and higher titre of neutralising antibody response to the variable surface glycoproteins (VSG) in the more resistant cattle (Chandler, 1958; Pinder et al., 1988; Dwinger et al.,1992). More recently, the role of a superior immune response (IgG1) to invariant antigens, released after antibody-mediated trypanolyse, such as

T.congolense-cysteine protease or 'congo pain' has been demonstrated during primary infection in N'Dama cattle, enabling a more efficient control of the parasitaemia in trypanotolerant cattle (Authié et al., 1993; 1994). The results of study 2.2 showed that the Djallonke sheep mounted a superior humoral immune response (Ig G) compared to the WAD goats, as measured by the indirect immunofluorescent antibody test (IFAT), using the method of Katende (1987) whereby antibodies against both VSG and invariant antigens were detected (Magnus, 1988; Ndao, 1998). In spite of their difference in antibody level, neither species showed tendency for self-cure as described in trypanotolerant cattle following primary infection (Wellde et al., 1981; Nantulya et al., 1986). However, when resistance to trypanosomosis was compared between Djallonke and Sahelian x Djallonke crosses, a superior humoral immune response linked to a better control of parasitaemia was found in the former breed (see chapter 6). Self-cure may occur when the antigenic repertoire of that particular serodeme is exhausted (Nantulya et al. 1986). The difference with trypanotolerant cattle was that in small ruminants more time was needed to clear the parasites from the host and eventually to realise self-cure.

The conclusion was that low mortality combined with positive weight gains could be used as indicators for trypanotolerance in small ruminants. Based on the haematological and antibody responses following primary challenge with trypanosomes, Djallonke sheep were shown to be more trypanotolerant than WAD goats, but in both species the control of parasitaemia is much less prominent than in trypanotolerant cattle. In addition, Djallonke sheep and WAD goats have a lower capacity to control anaemia. For sheep, there is evidence that, like in cattle, the onset and intensity of parasitaemia is genetically controlled (Djallonke sheep compared with Sahelian x Djallonke crossbreds). There is no evidence in the Djallonke breed nor the WAD goat that the duration of parasitaemia or the severity of anaemia can be controlled following trypanosome infection. Based on the above, trypanotolerance in small ruminants may be better described as a 'resilience' to infection rather than 'resistance' to infection, this in contrast to the situation in cattle.

Natural infections with trypanosomes (2.3).

The study on the village based animals enabled comparison of resistance to trypanosomosis under natural challenge. The epidemiology of trypanosomosis in sheep and goats and the impact of trypanosomosis on their health and productivity were studied in two areas. These surveys also evaluated the importance of intercurrent stress factors such as helminths, nutrition and management on the expression of trypanotolerance in rural based sheep and goats.

Mortality occurred mainly in the rainy season. This was not directly linked to the period with highest trypanosome prevalence (early dry season), but coincided with the period of lowest haematocrit levels which were mainly due to high helminth infestation and poorer nutritional condition. The low mortality associated with trypanosome infection confirmed what had been observed under conditions of experimental infection (2.1, 2.2). In contrast, offspring mortality was influenced to some extent by the infection status of the dam in sheep but not in goats. Other causes related to the extensive management (nutritional management and supplementation of the dams, predators, delivery in the bush) revealed to contribute more to the overall high offspring mortality rates, which were found higher than those reported for village-based sheep and goats in Nigeria (Sumberg and Mack, 1985; Otesile and Oduye, 1991). Anaemia was present in both trypanosome infected sheep and goats, but was more pronounced in the latter species, confirming findings after experimental infection. Irrespective of the presence of trypanosome infection, a strong seasonal effect was observed with significantly lower PCV levels during the rains. Major contributing factors were the peak of helminth infestations, predominantly *Haemonchus contortus* (Fritsche et al., 1993) and inadequate nutrition due to poor grazing management, whereby small ruminants are either tethered or herded during a restricted time. The importance of nutritional level and intercurrent helminth infections on their resistance to trypanosomosis are further discussed in the chapters 4, 5 and 7.

Trypanosome infection depressed weight gain in both species in high and/or moderate risk sites but declines were most obvious in periods with higher infection rate comparable to the findings following artificial infection (study 2.1-2.2). However, under these experimental conditions animals were adequately fed, hence, nutrition was not considered as a constraint. In contrast, the seasonal nutritional stress occurring in the rural areas was an additional constraint resulting in weight loss even during the rainy season.

The epidemiology of natural trypanosome infections in small ruminants revealed some differences between species and with cattle. In both study sites, goats had lower infection rates than sheep. In the high risk site where a resident cattle herd was present, both species had lower rates than cattle (Jaitner J. & Clifford D.J., 1997; unpublished results). Factors contributing to this include the low feeding success of tsetse on small ruminants (Snow et al., 1996) and a different interaction between vector and host than in cattle. The former may be related to their smaller body size and anti-feeding behaviour such as leg kicks and skin rippling, which is more pronounced in goats (Vale, 1977). Limitation of the vector-host interaction is the result of the specific grazing patterns of sheep and goats. However, even grazing in the vicinity of villages ensured a continuous trypanosomosis risk in the high risk site, since both village-sited traps and sentinel horses and donkeys showed high incidence rates (Faye, 1998). Further study on the grazing behaviour of village-based sheep and goats is needed to clarify the observed differences between species. Nevertheless, peak trypanosome infections in small ruminants followed peak tsetse density as predicted by a model of the epidemiology of trypanosomosis (Milligan and Baker, 1988). The observed differences between incidence and prevalence rates in the high risk area confirmed the absence of self-cure, as observed also following single *T.congolense* infection. *T.vivax* was the predominant species found, especially in the high risk-site, similar to the observations in the resident cattle. The lower infection rates measured in young animals had been observed earlier by Greenwood and Mullineaux (1989). Since young animals were subjected to the same infection risk as their dams, the lower infection rates could be explained by their smaller size and by the higher (non-specific) immunity in young animals (Stephen, 1986; Connor, 1994).

In conclusion, the trypanotolerance observed in both species is the result of both innate and acquired resistance. The difference in tolerance between goats and sheep as observed following artificial infection was less apparent under natural infection. This contradiction might be explained by the lower infection rates in goats exposed to natural challenge. It is concluded that both species are considered as trypanotolerant since they survive and remain productive under various levels of tsetse challenges. Despite trypanosomosis being an important constraint, other factors such as helminth infections and seasonal nutritional constraints equally affect the health and productivity of village-based sheep and goats. In addition, these may interfere with their resilience to trypanosomosis.

REPRODUCTIVE PERFORMANCE FOLLOWING TRYPANOSOME INFECTION

Effects in female small ruminants (3.1).

Following single challenge with *T. congolense*, reproductive performance was more affected in does than in ewes (study 3.1). Infected goats had more abortions and extended intervals to first kidding, indicating a reduced or disturbed ovarian activity and/or embryonal or early foetal death, resulting in a severe reduction in the number of weaned kids produced during the 2 years after primary challenge. Return to normal reproduction levels was not possible in the second year after infection since the infected does had a productivity index which was 50 % lower than the controls. However, survival of kids from infected and control does was similar. The reduced productivity index was mainly the result of fewer kids born. Similarly, Bealby et al. (1993) observed lower kidding rates in naturally infected goats and Ogaa et al. (1991) reported *T. congolense* as cause of abortion in goats.

The lambing rates, number of lambs born and intervals to first lambing were not significantly influenced by infection, indicating an almost unaffected ovarian activity and conception. In contrast, lamb performance was affected by infection of the dam, primarily due to increased lamb mortality but also as the result of reduced growth rates. The latter may have been attributed to lowered milk production. The infected ewes reached normal productivity indices in the second year, indicating full reproductive recovery in spite of a chronic parasitaemia. Noteworthy is that in this study the females were kept on adequate nutrition levels. At no time either before conception, during pregnancy or lactation, nutrition could have been regarded as a constraint and have interfered with their resistance to infection. The importance of nutrition on reproduction and trypanosomosis will be further discussed. These results on reproductive performance showed again that WAD goats are less trypanotolerant than Djallonke sheep.

Mawuena (1986, 1987) reported unaffected fertility in Djallonke sheep under natural challenge in Togo. Productivity of sheep and goats under natural infections in the Gambia was however affected due to trypanosomosis (study 2.3) since in both species, parturition intervals increased significantly. This corresponds to what was found in goats but not in sheep which were infected experimentally (study 3.1). The higher abortion rate primarily found in goats kept under high tsetse challenge, was in agreement with the results of the experimental infections. Natural trypanosome infection of the ewe resulted in a significantly increased lamb mortality up to four months (study 2.3; high risk area) while this was not observed in the does. This corresponds to the results obtained after a single experimental infection. In contrast, a natural trypanosome infection in the dam or doe had no effect on birth weight or growth rates of offspring.

Effects in male small ruminants (3.2).

The study on the semen characteristics of Djallonke rams following artificial challenge with *T. congolense* revealed that despite some transient changes in sperm quality, the reproductive performance was not severely affected in contrast to previous reports (Agu et al., 1986; Akpavie et al., 1987; Sekoni, 1992).

Semen quantity, measured as total sperm output, was not clearly influenced by infection. In contrast, semen quality was temporarily but not significantly affected, as demonstrated by the transient changes in mass motility, percentage live sperm cells and minor abnormalities. Return to normal values was remarkably quick, considering the duration of a spermatogenic cycle (7 weeks). The rams which had very high values for sperm cell defects following infection could have been regarded as temporarily unsuitable for breeding, since abnormal sperm morphology exceeding 20 % is associated with lowered fertility. In *T. congolense* infection in goats and *T. vivax* infection in sheep testicular degeneration has been demonstrated (Kaaya & Oduor-Okelo, 1980; Anosa & Isoun, 1980). The testicular degeneration which was induced by the *T.congolense* clone in the present experiment was mild, based on the type of sperm cell defects found (Wenkoff, 1988). Moreover, the rapid disappearance of the sperm defects in this study gave no evidence of severely damaged seminiferous epithelium, although no further histological examination could be performed. The impact of infection on libido was demonstrated by an increased rate of mounting failures, possibly due to a lowered plasma testosterone level, which could not be further confirmed in this study. However, clinical factors in the infected rams such as the onset of anaemia, parasitaemia and pyrexia may have equally contributed to the reduced libido. Compared to the course of infection in Djallonke ewes, it seemed that the rams suffered more in terms of depressed weight gain whereas the control of anaemia following infection seemed better.

The impact of the *T. congolense* infection on the clinical and reproductive performance of the Djallonke rams was modest. It was only evident in negative weight changes, some transient changes in sperm quality and a higher rate of mounting refusals. Although these are not the criteria of the trypanotolerant trait, the large individual differences in clinical and reproductive response found in this study could be used to rank for the trypanotolerant quality of Djallonke rams, based on both clinical tolerance and reproductive performance following trypanosome infection.

INTERACTION OF NUTRITION AND TRYPANOSOMOSIS ON REPRODUCTIVE FUNCTION OF DJALLONKE SHEEP.

Adequate reproductive performance is an essential component in efficient small ruminant production. Adequate nutrition is also essential for the reproductive function in ruminants, but their relationship is complex and responses are often variable and inconsistent (O'Callaghan & Boland, 1999). On the other hand, there is evidence of reduced trypanotolerance in cattle in response to nutritional stress, hence, the importance of nutrition was evaluated on the resistance to the effects of trypanosomosis in Djallonke ewes, with particular reference to their reproductive performance.

Establishment and outcome of pregnancy (4.1).

Study 4.1 showed that *T. congolense* had a detrimental effect on the establishment of pregnancy in Djallonke ewes. The luteal-phase progesterone levels were reduced by trypanosome infection and contributed to the reduced pregnancy rates in the infected ewes. However, the higher feed supplements could not counter this decline caused by infection. Progesterone plays a major role in establishment of pregnancy in the uterus (Wilmut & Sales, 1981; Lawson & Cahill, 1983) but also in the survival and the growth of the embryo (Wilmut et al.; 1985; Lawson, 1977). Indication of embryonic and early foetal death was seen in some trypanosome-infected ewes. However, cause-effect relationship between circulating progesterone and loss of pregnancy in the cases of foetal death could not be confirmed. The level of nutrition per se did not influence conception rates. This corroborates previous work of Mani et al. (1995) who found that under nutrition did not affect circulating luteal progesterone level in goats. Outcome of pregnancy was affected by infection but only in the High supplemented-Infected (HI) group. The dietary level seemed important in the maintenance of pregnancy, with clear differences between the control dietary groups only. The low diet groups were sufficiently fed according to their requirements for pregnancy, but combined with multiple pregnancies it resulted in poor pregnancy outcome and mortality. The effects of infection or diet were not reflected in the progesterone levels during gestation. Experiments to ascertain the effects of feed restriction during gestation in ewes have shown an inverse relationship between plane of nutrition and plasma progesterone concentration (Parr et al., 1982, 1987; Wilmut et al., 1985). Unlike the latter studies, the low, restricted rations in this study did not reach submaintenance and explains maybe the absence of diet effect on progesterone levels during gestation. It is clear that any impairment of luteal and/or placental progesterone production would have serious implications for pregnancy. The mechanism by which haematic forms of trypanosomes cause a reduction in both luteal and placental progesterone secretion have been well studied but were not subject of our studies (Mutayoba et al., 1988, 1995a; 1995b). In addition, haematic forms of trypanosomes have been shown to cross the placental barrier (Ogaa et al., 1991; Ikede & Losos, 1972) possibly leading to disturbance of placental progesterone production and fetal death.

In contrast to this study (4.1), the study 3.1 describing reproductive performance of Djallonke ewes infected with the same clone of *T.congolense* but with adequate nutrition level did not reveal significant differences in lambing rates nor in time intervals to first lambing. In study 3.1, the first peak parasitaemia following infection was not necessarily coinciding with oestrus for all dams, hence, acute effects on luteal production of progesterone were probably more diluted. In addition, individual variation in productive and clinical response to infection, of which the importance was already shown in Djallonke rams (3.2), could explain the different outcome of studies 3.1 and 4.1, as well as between infected dietary groups in this trial (4.1). Reynolds & Ekwuruke (1988) indicated a reduction in both perinatal mortality and ewe mortality in the better nourished trypanosome- infected ewes. In the present study, mortality was either the result of infection or of low nutrition combined with multiple pregnancy. It seems in contrast to the studies 2.1, 2.2 and 3.1, but higher mortality associated with trypanosomosis could be explained by the physiological status of early pregnancy which made the ewe more stressed and more susceptible.

It is concluded that *T. congolense* infection impaired the establishment of pregnancy in its acute phase. This could not be countered by dietary supplements. Maintenance of pregnancy was influenced by both nutrition and infection, although the effects of infection had become more chronic. However, the importance of individual variation indicates that some ewes cope better with the effects of trypanosome infection and low nutrition on their reproductive performance.

Effects of trypanosome infection and nutrition level on live weight of ewes during pregnancy, post partum weight, haematology parameters and lamb performance (4.2).

Whereas no interactions nor added benefits of energy and protein supplements on the effects of infection on pregnancy rates were observed, nutrition interacted positively with infection on some haematological and other productivity responses in Djallonke ewes (4.2). Neither severity of anaemia nor intensity of parasitaemia were influenced by nutrition confirming previous findings in sheep (Wassink et al., 1997; Reynolds & Ekwuruke, 1988) and in N'Dama cattle (Romney et al., 1997). In contrast, the erythropoietic response was enhanced by dietary supplements, measured as a macrocytic, hypochromic anaemia. Previous results (2.2) indicated that trypanosome-infected sheep when adequately fed, mounted a better haematological response in the acute phase compared to infected goats. Katunguka et al. (1993) measured a better erythropoietic response in trypanosome-infected sheep fed higher protein supplements. This proves the beneficial effects of nutrition in the control of anaemia following infection. This is an important feature in trypanotolerance, as previously observed in N'Dama cattle (Agyemang et al., 1990). The extra nutrients needed for the haematopoietic and immune responses were at cost of the ewe's productivity gain and this was clearly reflected in the live weight patterns. While infection *per se* did not affect live weight significantly, the interaction between diet and trypanosome infection resulted in a depression of the effect of supplementation in the infected groups as compared to non-infected controls. The latter was also reflected in lower post partum weights for the low dietary infected group as opposed to the low dietary control group. Lamb performance was positively influenced by the high dietary condition of their dams, suggesting a better milk production in the latter. Although the trypanosome infection in the dams became chronic, it still depressed growth rates of lambs of the dams kept on a low dietary plane considerably. Lamb mortality rates were not different between treatments, but effects were confounded by the high number of multiple births, especially in the HC group, resulting in smaller and weaker lambs. Whilst lamb performance seemed more affected by infection in the previous trial (3.1), the present results gave further evidence for declined milk production in the infected ewe, especially when nutrient allowance was restricted. Akinbamijo et al. (1994) showed more severe effects of *T. vivax* infection in late pregnancy than in mid pregnancy. In this study (4.2), the infection became chronic around parturition and beyond, with a less severe effect on lamb performance.

We can conclude that under these experimental conditions, in Djallonke ewes infected with *Trypanosoma congolense* during pregnancy and lactation, an adequate dietary level improved haematopoiesis and productivity in terms of ewe live weight and lamb growth rates to weaning.

Effects on ewe lambs : onset of puberty and age at first lambing (4.3).

T.congolense infection did not directly influence age at puberty. Negative effects of infection on attaining puberty were indirectly mediated through its effect on live weight patterns. A strong correlation was found between weight and age at puberty. In this experiment only the HC group (= high dietary level and not infected) attained puberty at the target age of 8 months and corresponding live weight of 16.3 kg as previously reported by Berger and Ginistry (1980). The remaining groups attained puberty in an order depending on attainment of the target weight. High supplementary feeding through its positive effect on body weight gain, reduced significantly the age at first cycling and at first lambing. However, under the present experimental conditions, sheep attained puberty and had their first parturition much later than in other observations on the Djallonke breed. This may be attributed to different management, climatological and nutritional conditions.

In clinical terms, the ewe lambs resisted well to infection, which is in agreement with the above mentioned results on Djallonke sheep described in chapter 2. The absence of dietary influence on the development of parasitaemia and on severity of anaemia was comparable to that observed in adult ewes (4.2) although it seemed that the degree of anaemia in the lambs was less severe. Furthermore, supplementary feeding enabled a better haematopoietic response in the young ewes similar to the adult ewes. As in the adult sheep, there were no signs of self cure nor an ability to control the level and duration of parasitaemia. The negative effect of infection on weight gain patterns can be partly explained by a reduction in dry matter intake (DMI) in the acute phase and partly by the metabolic cost of infection which resulted in increased maintenance requirements. The reduction in DMI was most noticeable in the high supplemented infected group (HI). Trypanosome-induced depression of DMI has been reported in sheep (Wassink et al., 1997; Reynolds & Ekwuruke, 1988; Akinbamijo et al., 1994) and goats (Verstegen et al., 1991). The latter studies also gave evidence of increased maintenance requirements due to infection, leading to a decreased nitrogen and energy retention and therefore reduction of the weight gain. Trypanosome infection has a metabolic cost in the host because of the induced fever (Verstegen et al., 1991), catabolism of immunological products (Nielsen et al., 1978) and haematopoiesis (Anosa, 1988a). Although the reduction in feed intake complicates the study on energy and protein metabolism in the infected host, the present results suggested also an increased metabolic cost due to infection. The retarded growth rates were mainly seen in the HI group whilst the extra nutrients in this group were used for a better haematopoiesis. This was not so obvious in the low supplemented infected group (LI) because the offered ration was too low to allow a further reduction of food intake by infection. Therefore, this minimal availability of nutrients resulted in a poorer haematopoiesis.

Since the ewes were free of trypanosomes at the time of parturition, the birth weights or growth rates of their lambs were not longer influenced by previous infection. In contrast, lamb mortality up to 3 months was still negatively influenced by previous infection and mainly arose from the previously infected ewes on low dietary plane. The absence of dietary effect on lamb growth was in contrast to the adult ewes. The severity of the effects on lamb performance seems to depend on the time interval between infection in the dam and parturition and subsequent lactation period. This was only partly supported by the previous study on breeding ewes, because although infection had become chronic, there was still an interacting effect between infection and diet resulting in depressed lamb growth rates in the low supplemented infected ewes (4.2). Under natural infection (2.3), whether infection of the ewe occurred in the first term or second term of pregnancy, or during the early post-partum period, birth weights and initial growth rates of their lambs were not affected, but mortality rates were significantly increased. It should be noted that under natural challenge the majority of infections were *T. vivax*. However, these ewes kept under natural conditions, were constantly at risk with a possible mixture of new and chronic infections. Intervals between infection and parturition or lactation are therefore not known and effects on mortality rates of offspring may be attributed to a mixture of acute and chronic infections. Summarising the results on lamb performance from the different trials, the nutritional condition of the ewe, which acted independently and/or interacted positively with infection, was found to be very important. It enabled a better milk production and mothering ability, seen either as an improved weight gain or survival of the lambs. Noteworthy is that dietary supplements were more effective in the older ewes, probably because milk production in primiparous ewes still has to develop, but similar studies are further discussed under chapter 7. In all above studies, birth weights were not affected by trypanosome infection in the dam. Apart from the effects of trypanosome infection and nutrition, many other intrinsic and environmental factors influence the lamb performance. Although a maximum of these factors were accounted for in the analyses, many others still remained uncontrolled, especially in the village situation.

In conclusion, the results show that, whereas trypanosome infection tended to delay puberty and age at first lambing directly, these effects were more indirectly mediated through depression of weight gain. Positive interaction between the effects of trypanosome infection and diet were observed for the supplemented infected group in a better haemopoietic response and better survival rate of their lambs, but were at cost of their own weight gain. The results undoubtedly showed the delaying effect of a low dietary level on the onset of the reproductive activity in young Djallonke sheep.

Biochemical changes influenced by nutrition and trypanosome infection (4.4).

The changes in biochemical parameters as observed in infected young and adult ewes provided some useful indications of the nitrogen and energy metabolism in these animals. These biochemical changes were also in agreement with results on live weight patterns and haematological changes in both age groups. Interactive effects between trypanosome infection and diet were mostly absent indicating their independency but additive action. The major changes induced by trypanosome infection were a decline in total protein level in both young and adult ewes, together with a decrease in albumin concentration, as measured in the ewe lambs only. Decrease in albumin was also observed in both single and serial infections of *T. congolense* and *Haemonchus contortus* (study 5.1). These results agree with those reported in susceptible breeds of sheep following trypanosome infection (Katunguka-Rwakishaya et al., 1993, 1995; Wassink et al., 1997). Reductions in total protein and albumin due to trypanosome infection were not countered by dietary supplements, and neither was the diet effect significant. These findings are in contrast to the work of Katunguka-Rwakishaya et al. (1993, 1995) and Wassink et al. (1997). Since levels went below minimum reference values in the young infected animals, it might have influenced tissue accretion and general protein metabolism pattern. The protein/albumin index was stable and within normal physiological range of 35-50% in the young ewes (Kaneko, 1989). Since haemodilution has been associated with trypanosome infection (see 4.3), it could explain partly the declined albumin and total protein concentrations (Anosa & Isoun, 1976; Katunguka-Rwakishaya et al., 1992). Trypanosome infection tended to increase plasma urea levels, which presumed, together with the declined total protein and albumin, a disorder in the nitrogen metabolism pattern as widely reported in the literature about trypanosomosis (Akinbamijo et al., 1994, Verstegen et al., 1991). In the ewe lambs, plasma glucose levels were lowered due to the low dietary allowances and this became emphasized at the onset of trypanosome-induced anorexia in the post acute phase (see 4.3), resulting in lowest values for the LI group. Low glucose values coupled with a metabolic cost of infection led to a state of negative nutrient balance in infected animals on low dietary regime whereby endogenous catabolism may have occurred. Evidence of the latter hypothesis was observed in the adult ewes (4.2), with transient peak values of plasma NEFA and BHBA levels in the acute phase of infection, being indicators of temporary ketogenesis. The observations on nutrient metabolism are corroborated by the live weight patterns in both young and adult ewes. Since there was a lower rate of weight gain in the infected animals in both trials, it was assumed that the exogenous energy and protein sources were sufficient to cover maintenance requirements while requirements for growth/weight gain were already negated by the increased metabolic cost of trypanosome infection and trypanosome-induced anorexia.

Finally, from the observed changes in biochemical parameters, it was concluded that in spite of their trypanotolerance, a trypanosome-induced alteration of the nutrient metabolism was present in both young and adult Djallonke ewes irrespective of the dietary status. Since the effect of nutrition was mostly independent, it conferred added benefits against the debilitating effects of trypanosomosis under the conditions of the present study.

INTERACTION OF HELMINTH INFECTION AND TRYPANOSOMOSIS

Effects in experimentally infected animals (5.1).

The interaction between *T.congolense* and *Haemonchus contortus* infection was studied in young female Djallonke sheep. Analyses of the different parameters measured clearly indicated the pathogenicity of the infections and the severity of mixed infections compared with single infections.

Mortality following single *T.congolense* infection was zero corroborating the results of other studies (2.1, 2.2 and 3.1). Mortality was only observed in dual infection groups indicating the severity of the combination of the two pathogens. Noteworthy is that 2 out of these three cases of mortality derived from the lowest weight-block. The *T. congolense* strain (Tc) used in this experiment was the main pathogen causing anaemia. The *H.contortus* infection (Hc) did not cause a significant reduction in PCV and did also not interact with the trypanosome infection, hence, severity of anaemia in the dual infected groups (TcHc and HcTc) was similar to the one in the Tc group. There were also no significant differences between single and dual infected groups for the other haematological parameters nor in the pyrexia induced by the Tc infection. In contrast, a shorter prepatent period (PPP) and higher peak EPG of the Hc infection, the lowest drop in plasma albumin and higher mortality in the TcHc group indicated that this dual infection was the most severe combination. The Tc infection superimposed on the Hc infection (HcTc) resulted in a slower recovery from the latter infection as shown by more chronic effects such as lower weight gains. Similarly, in N'Dama calves, Kaufmann et al.(1992) found the Tchr combination most harmful based on a shorter PPP and higher EPG of the Hc infection, lower PCV, more pronounced weight loss and higher mortality. Also Griffin et al. (1981a, 1981b) reached the same conclusions for goats based on a shorter PPP, a higher worm burden and higher mortality in Tchr group. Although it could not be confirmed in this study, it should be noted that when the infections (both trypanosomosis and helminthosis) would have resulted in severe anorexia, a higher EPG could also result from low faecal production. In the previous chapters, the trypanotolerance in Djallonke sheep following single primary challenge and without nutritional constraint was demonstrated by their zero mortality and ability to maintain productivity at acceptable level. In this study, the trypanotolerant trait of the Djallonke breed was again proven in the single Tc group. However, when trypanosome infection was combined with a Hc infection, the resilience to the effects of both pathogens was reduced in the Djallonke breed. The immunosuppressive effect of the trypanosome infection (MacKenzie 1975; Nantulya et al., 1982) may have contributed to the reduced resistance against the Hc infection. Dwinger et al. (1994) reported that N'Dama cattle infected with helminths have higher EPG counts when simultaneously infected with trypanosomes. In addition, the results of the present experiment on the single Hc infected group suggested some degree of natural resistance of Djallonke sheep to *H. contortus*, as had been previously indicated (Assoku, 1981; Baker, 1995), and will be further discussed in the next chapter.

In conclusion, this study has clearly shown that trypanotolerance in Djallonke sheep is not absolute and that a concurrent *H.contortus* infection may result in a reduced resilience. Since under field conditions mixed infections occur very frequently, expression of trypanotolerance may be negatively influenced by concurrent helminth infections and thus increasing productivity losses. Therefore, on-farm interventions to reduce the impact of helminth infections in small ruminants which are constantly at risk of trypanosomosis have been tested in both species and are discussed hereafter.

Effects in naturally infected animals (5.2).

On-farm anthelmintic treatment of sheep and goats has been done over two or three rainy season periods respectively. This on-farm trial demonstrated, in spite of a constant risk of trypanosome and helminth infections and other environmental stress factors, clear beneficial effects of prophylactic anthelmintic treatment on production and health parameters in both species but most clearly in goats. Considering the existing management practices and the design of this on-farm intervention trial, the overall significant reduction in nematode egg excretion (EPG) was satisfying. Since the most prevalent species during the rains is *Haemonchus contortus* (Fritsche et al., 1993) and a good correlation between worm burdens and EPG has been observed for this species (Roberts & Swan, 1981), it could be assumed that, based on the reduced EPG, adult worm burdens in both sheep and goats were considerable reduced in the treated groups. Weight gain advantages of treated versus control sheep were observed for both immature and adult sheep, whilst this trend was not observed for goats. Similar results in sheep were obtained using the same treatment scheme in the study described under chapter 6.1. Vassiliades (1984) used a biannual treatment scheme but a different approach and had more spectacular results. The early dry season treatment applied in our study (5.2) was probably the most efficient one for on-farm conditions. Since it was given at a time with low or no risk for reinfection, as previously recommended by Ankers et al. (1994) this treatment may be optimal with respect to remanence. This treatment scheme simulated existing village situations since rarely all small ruminants will be treated due to the cost involved. The kidding rates were increased due to the anthelmintic treatment and this trend was also noted in sheep. Average litter size tended to be higher and parturition interval shorter in both treated goats and sheep, however without reaching significance. Shorter observation time and thus less observations could explain in part the absence of statistical significance on the reproductive parameters in sheep. Furthermore, different trypanosomosis risk and other uncontrolled factors make it difficult to compare the results in sheep with the study of Ankers et al.(1998). The latter authors observed a significantly improved reproductive performance in village-based Djallonke ewes receiving a biannual treatment. The negative effects of natural trypanosome infection on reproduction in both species, as mentioned under chapter 2.3 were diminishing the beneficial effect of anthelmintic treatment. The presence or absence of positive effects of anthelmintic treatment on offspring performance in our study (5.2) was probably more linked to the timing of the treatments and the higher incidence of parturitions at the end of the rains. Positive effects were therefore only seen on the birth weight of both species and increased survival in the kids. The overall positive effect of prophylactic deworming was reflected in the increased productivity indices in both sheep (+ 24 %) and goats (+ 47 %), but was most prominent in the latter species. Similar results were obtained by Ankers et al. (1998) in sheep and by Tillard et al. (1992) in both species in the Kolda area in Senegal.

Anthelmintic treatment during the rains improved health conditions as measured by increased PCV levels. This confirmed the pathogenicity of mixed natural helminth infections, specially during the rainy season, causing anaemia. This was also shown in the study in chapter 6, during which the same deworming scheme improved PCV levels in both Djallonke sheep and Sahelian x Djallonke crosses. There was evidence that the effects of trypanosome infection and helminths on PCV level were additive, confirming the results from the previous experiment (5.1) and those in chapter 6. Apart from these 2 disease factors of which effects were measured, there were still seasonal effects with lowered PCV levels during the rains, attributable to the nutritional stress in small ruminants under these management conditions, as mentioned earlier

(2.3). If the nutritional supply is very low, the erythropoiesis responding to the anaemia becomes less efficient as demonstrated in chapter 4. The benefits of the effect of dietary supplements on PCV level during helminth infections in Djallonke sheep became also apparent in the study on breed comparison (chapter 6). From the present study, it was clear that helminth infection combined with inadequate nutrition and trypanosome infections contributed to the lower PCV levels and consequently higher frequencies of mortality observed during the rains.

It can be concluded that in spite of a continuous risk of trypanosome infections and seasonal inadequate nutrition, prophylactic anthelmintic treatment increased production of the indigenous sheep and goats. Although cost-benefit analyses need to confirm whether such intervention is economically acceptable for the farmers, it is certainly optimising the trypanotolerance in these breeds.

GENETIC BASIS OF THE TRYPANOTOLERANT TRAIT IN DJALLONKE SHEEP AND THE POTENTIAL OF THEIR F1-CROSSES TO INCREASE PRODUCTIVITY.

The disease resistance and productivity of young, growing Djallonke sheep and their F1- Sahelian x Djallonke crosses were compared in a multifactorial study (chapter 6). In this study animals were challenged with either single or dual infections with *T.congolense* and natural helminth infection. In addition, the influence of nutrition on the severity of the single or combined infections was evaluated in both breeds.

Trypanotolerance

One of the objectives of this study was to obtain further evidence of a genetic basis for trypanotolerance in the Djallonke breed. Results indicated a reduced resistance to trypanosomosis in the Sahelian x Djallonke crosses, as shown by a quicker appearance of parasites in the bloodstream, higher level of parasitaemia and lower antibody response. This proves that trypanotolerance in the Djallonke breed is an innate characteristic. From the previous results (chapters 2 and 3) it did not appear that the Djallonke sheep could limit the intensity of parasitaemia following infection, despite a good antibody response. However, when comparing with more susceptible breeds, the underlying mechanism of trypanotolerance in sheep seemed comparable to that in trypanotolerant cattle, but to a lesser extent. Differences in the control of parasitaemia between Djallonke and trypanosusceptible Sahelian sheep had previously been reported by Touré et al. (1981) and Bengaly et al. (1993). In contrast to trypanotolerant cattle there is no improved control of anaemia following infection in the Djallonke sheep. Nevertheless, it was generally concluded that the trypanotolerance in cattle is primarily an ability to control parasitaemia, through a more effective immune response, and not the ability to mount a better haematopoietic response (Dargie et al., 1979; Authié et al., 1993).

Surprising was that the crossbreds did no suffer more than Djallonke sheep following trypanosome infection in terms of pyrexia, mortality, severity of anaemia or depressed weight gain. It seems that development of anaemia and control of parasitaemia are determined by separate mechanisms, like discussed in chapter 2. These results do no agree completely with the comparison trials of Touré et al. (1981) and Bengaly et al. (1993). However, these authors used other strains or species, and the differences probably relate to differences in virulence of the strain. To confirm the present observations, the Sahelian x Djallonke crossbreds should be compared under *T.vivax* infection but also under natural trypanosomosis risk. Differences in body temperature between Djallonke sheep and the Sahelian x Djallonke crosses indicated better thermoregulatory abilities in the former breed. In addition, the much higher pre-weaning mortality in the crosses, in the absence of trypanosomosis, confirmed the poorer adaptation and/or higher susceptibility to helminths and other endemic diseases such as cowdriosis of the crossbred compared to the purebred animals (unpublished results).

Innate resistance to helminths

Irrespective of the anthelmintic treatment, Djallonke sheep had consistently lower nematode egg excretion than their crosses, whilst both breeds had been raised under similar conditions and had not developed any acquired resistance at the start of the trial (Ankers et al., 1994). Resistance to gastro-intestinal nematodes is based on a history of exposure, age of the host and genetics (Gamble & Zajac, 1992; Baker, 1995). Whilst the first 2 factors were similar, genetics only could explain the difference in EPG. These results are providing further evidence of innate resistance to helminths in the Djallonke breed, as was already suggested by the results of chapter 5.1, and in previous work (Assoku, 1981; Baker, 1995). Further controlled experiments, in which data should be collected on the worm burdens, could confirm these observations.

Interactions between helminth infection, dietary supplements and trypanosomosis

Another objective of this study was to evaluate the importance of helminths and dietary level on the resistance to trypanosomosis, to confirm the observations from on-farm (2.3) and on-station studies (chapter 4 and 5).

Results with respect to effects of helminth infection on PCV level and weight gain in Djallonke sheep have been discussed in the previous chapter. The impact of helminth infections on PCV level and live weight gain was similar in the Djallonke breed as in the crossbreds. The effects of helminth and *T.congolense* infection on the parameters PCV and live weight were independent, confirming what was found in the previous on-station and on-farm studies (Chapter 5) and indicating the severity of the combination of the two pathogens just because of their additive effect. However, some interaction between trypanosome infection and helminths was evidenced by a higher EPG excretion in sheep infected with both pathogens, confirming the immunodepressive effect of trypanosomosis as described in chapter 5.1. There was no evidence that helminth infections caused any immunodepression since they did not influence the antibody response to trypanosomosis.

Dietary supplementation was acting independently but beneficially on PCV level and weight gain, hence, it diminished the severity of anaemia and depressed weight gain caused by helminths and/or trypanosome infection. The independent but positive role of nutritional supplementation was confirmed in this experiment. Its importance was already indicated in on-station controlled studies (Chapter 4) and in the on-farm intervention trial (5.2).

Finally, it was observed that crossbreds, which were of a larger size and thus heavier weight, also had higher growth rates than the dwarf Djallonke sheep. Therefore, the potential of the crossbred animal, especially for meat production under semi-intensive management, will be further explored.

Based on these results it can be concluded that the trypanotolerant trait in Djallonke sheep has a genetic origin. In addition, an innate resistance to helminths was observed. Although their crosses with the trypanosusceptible Sahelian breed expressed a reduced trypanotolerance in terms of immune response and parasitaemia control, and were less resistant to helminth infection, the impact of both pathogens on PCV and body weight was not worse than in the pure Djallonke. Furthermore, the larger size and better growth rates of crossbreds are promising for intensification of meat production, but appropriate sanitary measures should be taken to optimise their production environment.

NON-DISEASE FACTORS INFLUENCING PRODUCTIVITY: FORAGE LEGUME PRODUCTION AND FEEDING STRATEGIES.

The inadequate and poor nutritional value of the dry season feed resources and the rainy season management of small ruminants underlines the need to produce high quality feeds and a change in nutritional management. Most small ruminants are kept in free-roaming village flocks with only limited management or capital inputs. The preliminary experiments described here have shown that there is interest for novel fodder producing methods and feeding strategies. These can be developed at low cost and are adaptable by the rural farmer in a sustainable manner. The introduction of Intensive Feed Gardens (IFG) at village level may be an optional way for fodder production to be used for ruminants under semi-confined management regardless the season. The forage produced in the IFG may be used or conserved in different ways. Rural farmers have shown interest both in growing forage legumes in communal IFG and in using the produced forage to supplement certain animals. What makes the concept popular is the space left for vegetable growing during the dry season and cash crop production in the rainy season. The conflict with cash and food crops is therefore limited. Moreover, both are complementary. On station, different feeding strategies and fodder conservation methods have been tested. All four grasses (*Andropogon gayanus, Panicum maximum, Brachiaria mutica* and *Pennisetum purpureum*) may possibly be grazed *in situ* but more investigations on the carrying capacity and biomass production are still needed. If land tenure systems are not a constraint in rural areas, these grasses may be optional to plant in fodder banks and mixed with grass legumes to be grazed in rotational systems by sheep and goats. An additional aspect is the planting of rows of legume trees in these plots in alley farming with grasses and also used as fodder banks. This aspect will be subject of future in depth studies. Till present, the *Pennisetum purpureum* grass appeared to be promising for peri-urban production sites. This grass can be used in a cut-and-carry system to be fed fresh or in silage.

The best results prove to be attained with the forage legume, *Leucaena*. It grows very well and easily adapts to the local conditions. It produces adequate quantities of good quality fodder supplement, which may be fed either fresh or in dried form as leaf meal. This leaf meal is easily produced and stored. Animals adapt directly to *Leucaena* either in the fresh form or in the dried form and there are apparently no problems with uptake and toxicity. *Leucaena* leaf meal can be incorporated into HQSB. The production of the high quality supplement blocks (HQSB) is another way of conserving this forage legume in combination with other valuable components. Although it needs some more training and support, this technique will also have its benefit not only for the peri-urban but also for rural ram fattening enterprises.

Supplementing certain categories of animals such as pregnant and lactating females clearly resulted in better productivity. The closer supervision of the animals equally contributed to a better animal health and production. The use of supplements in zero-grazing ram fattening is expected to have a good rate of return. Further on farm trials will explore the different ways of supplementing sheep and goats and adoption of these techniques by the farmers.

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SUMMARY

Over the last decade increasing efforts were put into investigating the possible role of breeds of livestock with a natural resistance against trypanosomosis. These breeds, most of them indigenous to West and Central Africa, are called trypanotolerant because they survive and remain productive in tsetse infested areas where other breeds do not survive without treatment. Trypanotolerant small ruminants mainly include Djallonke sheep and West African Dwarf (WAD) goats. These breeds are of a relatively small size, but have proven to be productive under difficult conditions. This indicates their potential role for a more sustainable solution to the growing demand for animal protein in much of tsetse infested Africa.

In **chapter one** some general information is given about the role of trypanotolerant small ruminants within the mixed farming systems of The Gambia. The overall productivity of livestock in The Gambia is far below its potential. The increase in livestock numbers has not followed the same rapid trend as for the human population. In the prospect of increasing overall productivity from the livestock sector, small ruminants are well positioned. Besides this approach, there is need to intensify individual animal productivity by limiting the influencing factors. There is a potential high benefit from sheep and goats in both the traditional farming systems and in more intensified production systems. Both systems, however, have their constraints. These include bio-technical factors such as disease pressure, reproductive wastage, nutrition and management besides socio-economic, traditional and institutional factors. Increased animal production will concurrently need some intensification of fodder production and novel conservation methods to limit further environmental pressure. Understanding all influencing factors and their possible interactions would lead to appropriate intervention measures with farmer community participation. This will in turn assure sustainability. Impact of the interventions and evaluation of their cost/benefits for the target population is subject of continuous socio-economic follow-up.

This explains the need for a multi-factorial approach to study influencing factors on potential improvement of small ruminant productivity for the benefit of the human population. The work reported in this thesis examines the level of trypanotolerance in the Djallonke sheep and the West African dwarf goats as well as the influence of different stress factors on their trypanotolerant quality and on the productivity in these breeds. Both experimental and on-farm studies were carried out concurrently.

In **chapter two** the phenomenon of trypanotolerance in Djallonke sheep and WAD goats has been examined. The series of studies, using either a single artificial *Trypanosoma congolense* inoculation or natural infection with trypanosomes demonstrated a certain degree of trypanotolerance in the indigenous West African dwarf breeds of sheep and goats. The general course of trypanosomosis was evaluated including clinical parameters, haematological changes, immune response and body weight changes both under artificial and natural infection. Very low mortality rates together with the ability to maintain positive weight gain following infection were considered as the main features of trypanotolerance in small ruminants. However, in these breeds the control of parasitaemia and anaemia following trypanosome infection was less prominent than in trypanotolerant cattle. Based on our observations (2.1 and 2.2), it seems more appropriate to define trypanotolerance in small ruminants as a 'resilience' to infection rather than 'resistance'. Djallonkesheep appeared to express a higher degree of trypanotolerance than the WAD goats following artificial infection with *T. congolense*. In addition, the quick haematological changes and antibody responses following artificial trypanosome infection in study 2.2. further confirmed the innate resistance in Djallonke sheep and to a lesser extent in

WAD goats. The epidemiology of natural trypanosome infections in sheep and goats in two areas of high and moderate tsetse challenge, respectively, revealed that goats have lower infection rates than sheep. Overall, both species seem to be less infected than cattle based in the same area. Lower feeding success of tsetse flies and different host-vector interaction in small ruminants than in cattle are presumable contributing factors. In spite of their trypanotolerance, trypanosomosis remained an important constraint to health and productivity of both sheep and goats, but other factors such as helminth infections, seasonal nutritional constraints and management were found of equal importance and interfered at certain times with their resilience to trypanosomosis.

The effects of trypanosomosis on the reproductive performance of male and female trypanotolerant small ruminants are described in **chapter three.** Experiment 3.1 demonstrated that a single experimental *T. congolense* infection affected the reproductive performance of does more severely than that of ewes. Prevalence of abortions and stillbirths was higher and intervals to first kidding longer, resulting in fewer kids born alive. On the other hand, kids from infected does appeared to have an equal probability of survival and similar performance as those from controls. The infected sheep had lambing rates and intervals to first lambing comparable with those of the controls. In contrast, lamb performance was significantly affected by infection of the dam, due to increased lamb mortality together with reduced growth rates. Infected goats did not attain full recovery in the second year post infection. In contrast, infected ewes reached normal productivity indices in the second year, in spite of a chronic parasitaemia in both species. In sheep as well as in goats under natural trypanosomosis risk, parturition intervals increased significantly following infection (2.3). Trypanosome infection of the dam increased mortality in the lambs and not in the kids which was in agreement with the results following single infection, but in contrast, neither birth weight nor growth rates of lambs were affected.

The study of the semen characteristics of Djallonke rams following *T. congolense* infection (3.2) revealed some transient changes in sperm quality and not in quantity. The semen quality parameters which were temporarily affected by trypanosome infection included mass motility, percentage live sperm cells and minor sperm cell abnormalities. The nature of these indicated a mild testicular degeneration, hence, reproductive performance was not irreversibly affected. The impact of the *T. congolense* infection on the Djallonke rams was also evident in negative weight changes and a higher rate of mounting refusals. Large individual differences in reproductive response to infection were found amongst Djallonke rams and could be important in ranking for trypanotolerance.

The importance of nutrition on the resistance to the effects of trypanosomosis in Djallonke ewes was studied in **chapter four**, with particular reference to their reproductive performance. It was found that *T. congolense* infection impaired the establishment of pregnancy in its acute phase. This could not be countered by dietary supplements (4.1). The luteal progesterone level was clearly depressed by infection and not by dietary level. The key role of luteal progesterone in successful conception was hereby confirmed. Maintenance of pregnancy was influenced by nutrition, which mainly was shown by the comparison of dietary control groups, but also by trypanosome infection, although its effects had become more chronic. Similar to what was found in the rams, individual variation indicated that some ewes did cope better with the combined effects of trypanosome infection and low nutrition on their reproduction performance. Adequate nutrition interacted positively with the course of infection in Djallonke ewes during pregnancy and lactation as indicated by improved haematopoiesis and better productivity in terms of ewe's live weight and improved lamb growth rates to weaning (4.2). The results in the young ewe lambs indicated no direct effects of *T. congolense* infection on attainment of puberty but age at first lambing tended to be delayed (4.3). Nevertheless, the negative correlation between weight and age at puberty confirmed that the onset of puberty was indirectly delayed through infection-induced depression of growth. In contrast, supplementary feeding reduced the onset of reproductive function independently. Similar to the adult ewes (4.2), diet interacted positively with the effects of trypanosome infection resulting in a better haematopoietic response and better survival rate of their lambs, although this was at the cost of their own weight gain. The results undoubtedly showed a delaying, independent effect of both low dietary level and trypanosome infection, although the latter acted indirectly, on onset of reproductive activity in young Djallonkesheep.

The changes in biochemical parameters as observed in both infected young and adult ewes (4.4) supported the results on live weight patterns and haematological changes and provided some useful indications of their nitrogen and energy metabolism. Interactive effects between trypanosome infection and diet were mostly absent indicating their independence but also their additive action. Trypanosome-induced reductions in total protein and albumin in the serum were not countered by dietary supplements. Trypanosome infection tended to increase plasma urea levels, indicating, together with the declined total protein and albumin, a disorder in the nitrogen metabolism pattern. The lowest glucose levels found in the infected young ewes on low diet level as well as the transient peak values of plasma NEFA and BHBA levels in the adult ewes, which were indicating temporary ketogenesis, reflected the trypanosome-induced anorexia during the acute phase of infection (4.3). A trypanosome-induced alteration of the nutrient metabolism was present in both young and adult Djallonke ewes irrespective of the dietary status. However, since the effect of nutrition was mostly independent, it conferred added benefits against the debilitating effects of trypanosomosis under the conditions of this study.

In **chapter five** the interaction of helminths and trypanosome infection was studied both under controlled and on farm experiments. The interaction between a controlled *T. congolense* and *Haemonchus contortus* infection was studied in young female Djallonke sheep (5.1). Analyses of the clinical, haematological and production parameters clearly indicated the pathogenicity of the infections and the severity of mixed infections compared with single infections. The immunosuppressive effect of trypanosome infection was clearly reflected in the course of the *Haemonchus* infection. This study showed that trypanotolerance is not absolute and that a concurrent *H. contortus* infection may result in a reduced resilience. Since under field conditions mixed infections occur very frequently, reducing the impact of helminth infections in trypanosome infected small ruminants will benefit the expression of their trypanotolerance.

The results of the on-farm intervention trial (5.2) demonstrated the beneficial effects of prophylactic anthelmintic treatment on weight gain, reproduction and health parameters in both sheep and goats, despite the constant risk of trypanosome and helminth infection. Although there was no interaction between the two pathogens, the trypanosome infection diminished the positive effects of anthelmintic treatment on their productivity and health parameters. Nonetheless, the productivity indices were considerable increased in both treated sheep and goats. Cost-benefit analyses would need to confirm whether such intervention is recommendable at rural level but it is certainly optimising the trypanotolerance in these breeds.

In **chapter six,** the disease resistance in naive Djallonke sheep and their F1- Sahelian x Djallonke crosses was compared in a multifactorial study including *T. congolense*, natural helminth infection and different levels of nutrition. The difference in parasitaemia control and immune response following a single *T. congolense* infection proved that the trypanotolerant trait in Djallonke sheep was genetically linked. In addition, previous suggestions on the existence of innate resistance to helminths were again confirmed in the Djallonke breed. Although their crosses with the trypanosusceptible Sahelian breed expressed a reduced trypanotolerance and lower resistance to helminth infection, the impact of both pathogens on PCV and body weight was similar. The effects of low dietary allowances, helminths and *T. congolense* infection were independent but additive on the parameters PCV and live weight. However, some interaction between trypanosome infection and helminth infection was seen as a higher nematode egg excretion in the trypanosome infected animals explained by the immuno depressive effect of the latter which confirms previous finding as described in 5.1. It is expected that the larger size and better growth rates of crossbreds are promising for intensification of meat production. However, appropriate sanitary measures should be taken to optimise the production environment.

The impact of nutritional management and husbandry on health and production in indigenous small ruminants at farm level is highlighted in **chapter seven**. The implantation of village based and community managed Intensive Feeding Gardens (IFG) containing forage legumes (mainly *Leucaena* spp.) is described. This intervention is aiming at improving the nutritional status of village based animals, specially targeting at the special needs of animals around pregnancy and lactation, with a minimum of cost and labour involved but a maximum of benefits, the latter to make it adaptable and acceptable by the rural farmer. The results indicated that the concept of IFG installation at village level was accepted by the farmers in the study area. Since these IFG may be combined with vegetable planting in the dry season and food crop planting in the rainy season, it avoids conflicts between farmers and livestock owners. Nutritional supplementation during late pregnancy and/or early lactation combined with a change in nutritional management clearly demonstrated positive effects on offspring survival and growth rates in both sheep and goats. High quality fodder supply from forage legume trees as grown in IFG has a possible benefit for rural based sheep and goat production. Fodder produced in these IFG can be used either directly or conserved to supplement when needed. Furthermore, it can be mixed with other components to manufacture High Quality Supplement Blocs. These are easy to manufacture and can be stored. All these interventions will be subject to further on-farm evaluation and will be tested equally in more intensified peri-urban production sites.

CONCLUSION

The main breeds of sheep and goats in The Gambia, the Djallonke sheep and West African Dwarf goats are trypanotolerant. The Djallonke sheep, however, have a higher degree of trypanotolerance than the WAD goats. This trait is genetically linked although the mechanism of trypanotolerance is different from the one in cattle. It is defined as a resilience to infection rather than resistance.

Both breeds are well adapted to the locally prevailing diseases and environmental conditions under which they live. Moreover, they feature the capacity of making good use of low quality feed resources. For the rural farmer, small ruminant production is an attractive enterprise and seems to be profitable even under a low input management system. Small ruminant production also has a large potential under more intensified peri-urban conditions. Trypanosomosis remains an important constraint to optimal productivity. In particular when combined with other diseases, poor nutritional status and poor husbandry practises there is a clear effect on productivity and thus profitability. All these factors should be regarded as equally important.

Based on the results of research and surveys presented in this thesis, some recommendations are formulated for interventions in order to reduce the disease constraint, improve the nutritional status and adapt the management of small ruminant production systems. Further research will be carried out to study the impact of the proposed interventions and their adaptation to the local conditions.

SAMENVATTING

De laaste decennia werd heel wat onderzoek gedaan naar de mogelijke rol van herkauwers met een zekere natuurlijke weerstand tegen trypanosomose. Bepaalde rassen van herkauwers, sinds lang aanwezig in West-en Centraal-Afrika, worden slaapziekte resistent of trypanotolerant genoemd omdat ze overleven en verder produceren in gebieden geinfesteerd met tseetseevliegen daar waar andere rassen niet kunnen overleven zonder medische behandeling. Trypanotolerante schapen- en geitenrassen zijn o.a. de Djallonke schapen en de West-Afrikaanse dwerggeiten (WAD). Deze rassen zijn relatief klein maar toch goed aangepast aan de lokale omstandigheden en hebben een aanvaardbare productiviteit. Ze kunnen een grote rol spelen in het duurzaam tegemoet komen aan de stijgende vraag naar dierlijk eiwit in grote delen Afrika, die met tseetseevliegen geinfesteerd zijn.

In hoofdstuk 1 wordt algemene informatie gegeven over the rol van kleine herkauwers in de landbouw in Gambia. De algemene productiviteit van de dierlijke sector in Gambia ligt ver beneden het optimum potentieel. De bevolkingsgroei werd in geen geval gevolgd door een vergelijkbare groei in de dierlijke populatie. Zowel op het platteland als rond de steden hebben dierlijke productie-systemen te kampen met verschillende moeilijkheden die de productiviteit beïnvloeden. Deze moeten ook op een verschillende manier benaderd worden bij het zoeken naar oplossingen. Factoren met invloed op de productiviteit kunnen van bio-technologische aard zijn zoals ziekten, voeding en management. Daarnaast zijn er socio-economische, traditionele en institutionele factoren. Intensifiëring en stijging van de individuele productie vraagt een bijkomende inspanning voor de productie van diervoeder en nieuwe aangepaste methoden voor diervoeder stockering. Dit is nodig om een verdere milieudegradatie tegen te gaan. Een goede kennis van alle factoren die de productiviteit beinvloeden, kan leiden tot het plannen en ontwikkelen van aangepaste interventies met inbreng en goedkeuring van de boeren zelf. Dit komt de duurzaamheid ten goede. Het meten van de impact van de interventies en de kosten/baten analyse vòor de doelgroep vereist een socio-economische follow-up. Dit alles verklaart de noodzaak van een multi-disciplinaire aanpak om alle factoren met een mogelijke invloed op de productiviteit van kleine herkauwers te bestuderen.

Het werk waarover in deze thesis wordt gerapporteerd onderzoekt de graad van trypanotolerantie in Djallonke schapen en WAD geiten alsook de invloed van verschillende stressfactoren en hun mogelijke interacties met resistentie tegen slaapziekte. Daartoe werden gecontroleerde experimenten uitgevoerd alsook dieren gevolgd in hun natuurlijk milieu. Deze leverden het bewijs van een verschillend mechanisme van trypanotolerantie in Djallonke schapen en WAD geiten vergeleken met deze in runderen. Na het onderzoek van alle beïnvloedende factoren worden een aantal aanbevelingen geformuleerd voor een meer duurzame productie van kleine herkauwers. Deze resultaten zijn, naast hun belang voor Gambia ook gelijktijdig toepasbaar in de meeste naburige landen van de regio.

In de studies beschreven in hoofdstuk twee, waarbij zowel artificiële *Trypanosoma congolense* infectie als natuurlijke infecties met trypanosomen werden gebruikt, werd duidelijk aangetoond dat de lokale West-Afrikaanse dwerg rassen van schapen en geiten een zekere graad van trypanotolerantie bezitten. In dit hoofdstuk werd het algemeen ziekte verloop van trypanosomose bij deze dieren bestudeerd waaronder klinische parameters, hematologische veranderingen, immuniteitsopbouw alsook gewichtsveranderingen na zowel artificiële als natuurlijke besmetting werden gevolgd.

De extreem lage mortaliteit en het positief gewichtsverloop na infectie werden beschouwd als de voornaamste kenmerken van trypanotolerantie bij kleine herkauwers. Deze

rassen waren echter niet in staat om na infectie, de duur noch de intensiteit van de parasitaemia te controleren. Ook konden ze de ontstane anaemia niet inperken, in tegenstelling tot wat is beschreven bij trypanotolerante runderen. Trypanotolerantie bij kleine herkauwers wordt dan ook beter gedefiniëerd als 'resilience' dan 'resistance' tegen infectie. Deze studies wezen ook uit dat de Djallonke schapen een hogere graad van trypanotolerantie bezitten dan de WAD geiten. Bovendien bewezen de snelle haematologische veranderingen alsook de immuniteitsopbouw na artificiële besmetting met trypanosomen in studie 2.2 nogmaals deze aangeboren resistentie bij Djallonke schapen en, zij het in mindere mate, bij de WAD geiten. De epidemiologie van natuurlijke trypanosoom infecties bij schapen en geiten in twee gebieden van hoge en middelmatige besmettingsdruk, toonde aan dat geiten, gevolgd door schapen, veel minder frequent geïnfecteerd worden dan runderen die op dezelfde plaatsen verblijven. Kleine herkauwers blijken minder geprefereerd door de vector, de tseetsee- vlieg. Ook een verschillende gastheer-vector interactie tijdens het grazen zou echter de verschillen in infectiegraad met runderen kunnen verklaren. Onafgezien van deze trypanotolerantie blijft trypanosomose een bedreiging voor de gezondheid en productiviteit van kleine herkauwers. Hetzelfde geldt voor andere factoren zoals wormbesmettingen, seizoensgebonden voedingstekorten en management factoren, die allen even belangrijk zijn, ook omdat ze hun weerstandsvermogen t.o.v. trypanosomose kunnen verminderen.

Het effect van trypanosomose op zowel de mannelijke als vrouwelijke voortplanting wordt besproken in hoodstuk 3. Bij vrouwelijke dieren was de reproductie capaciteit meer aangetast in geiten dan in schapen na experimentele infectie met *T.congolense*, zoals aangetoond door hogere frequenties van verwerping, neonatale sterftes en langere tijdsintervallen tot het eerste lammeren, resulterend in een sterk gereduceerd aantal geboren geitenlammeren. Echter, hadden lammeren die afkomstig waren van geinfecteerde geiten gelijke kansen op overleving en een even snelle groei als deze van de controle groep. Zowel de geinfecteerde als de nietgeinfecteerde ooien hadden een vergelijkbaar aantal lammeringen en tijdsinterval tot eerste aflammering. Echter was de productiviteit van de lammeren significant beïnvloed werd door infectie van de ooien. Dit was vooral te merken aan een verhoogde sterfte en een verminderde groei voor het spenen. Geïnfecteerde geiten konden hun normale productiviteit niet herstellen binnen de twee jaar na infectie, iets wat wel mogelijk was bij de ooien die een pre-infectie productiviteit bereikten in het tweede jaar na infectie, dit ondanks een persisterende chronische infectie. Schapen en geiten gehouden onder natuurlijke infectie druk hadden een duidelijk verhoogd interval tussen de worpen (2.3). Trypanosoom infectie van de ooi gaf ook verhoogde sterfte bij schapenlammeren maar niet bij geitenlammeren, wat in overeenstemming was met resultaten na artificiële infectie. Noch het geboortegewicht, noch de groei bij hun lammeren was beïnvloed.

Studie van de reproductiecapaciteit van Djallonke rammen na een trypanosoom infectie toonde voorbijgaande veranderingen aan in kwaliteit doch niet in kwantiteit. Tijdelijke, voorbijgaande vermindering van de zaad motiliteit, verhoging van mineure defecten van de sperma cel en een vermindering van het percentage levende cellen waren de voornaamste effecten. Deze duiden allen op een milde testiculaire degeneratie waarbij de mannelijke reproductie slechts tijdelijk en omkeerbaar aangetast was. Daarnaast was de impact van de infectie ook zichtbaar in het gewichtsverlies en een verhoogd aantal weigeringen om te dekken. Dit duidt erop dat individuele rammen tijdelijk niet geschikt kunnen zijn voor reproductie. De grote individuele verschillen in klinische en reproductieve respons, die werden gevonden bij de rammen, zouden kunnen van belang zijn om de dieren te rangschikken volgens de graad van trypanotolerantie.

De invloed van de voeding op de resistentie tegen de effecten van een trypanosoom infectie bij Djallonke ooien wordt beschreven in hoofdstuk vier. Als belangrijkse parameter wordt de reproductiecapaciteit geëvalueerd. Een *Trypanosoma congolense* infectie die acuut is op het moment van de bronst beïnvloedt duidelijk de conceptie. Dit kon niet worden verhinderd door een verbeterde voeding. Het was duidelijk dat de concentratie van progesteron, dat tijdens de luteale fase wordt geproduceerd en een sleutelrol heeft bij een succesvolle conceptie, werd verminderd door infectie. Het in stand houden van de dracht werd enerzijds beïnvloed door de voeding, wat vooral aangetoond werd bij het vergelijken van de twee controle dieet groepen. Aanderzijds werd dit ook beïnvloed door de trypanosoom infectie, alhoewel deze reeds in een chronische fase was. Net zoals bij de rammen waren ook de individuele verschillen bij de ooien groot. Dit toont aan dat sommige ooien beter kunnen weerstaan aan de gecombineerde effecten van een trypanosoom infectie en lage voeding. Een adequate voeding resulteerde in een positieve interactie met de trypanosoom infectie gedurende dracht en lactatie. Dit werd aangetoond door een betere hematopoiese en betere gewichtsaanzet van de ooi en groei van de lammeren tot aan de speen leeftijd (4.2). De resultaten bij jonge Djallonke ooien gaven geen indicatie van een direct effect van de trypanosoom infectie op de leeftijd waarop de puberteit bereikt werd t (4.3). Er was een negatieve correlatie tussen gewicht en de leeftijd bij het bereiken van de puberteit. Het begin van de puberteit was indirect nadelig beïnvloed door de negatieve effecten van een infectie op de groei bij jonge ooien. Daarentegen vervroegden voedingssupplementen duidelijk en onafhankelijk van de infectie het begin van de reproductie. Zoals bij de volwassen ooien (4.2), beïnvloedde de voeding het verloop van een infectie op positive wijze. Dit werd aangetoond door een verbeterde hematopoiese en een grotere overlevingskans van hun geproduceerde lammeren, alhoewel dit ten koste ging van hun eigen gewicht. Deze resultaten bewijzen zonder twijfel het vertragend, doch onafhankelijk effect van een laag voedingsniveau en trypanosoom infectie op het bereiken van puberteit bij jonge Djallonke ooien. De veranderingen in de biochemische parameters bij jonge en volwassen Djallonke ooien (4.4) bevestigden de voorafgaande resultaten en gaven verder nuttige informatie over stikstof en energie metabolisme. Trypanosomose reduceert het gehalte aan totaal proteine en albumine in het serum en dit werd niet verhinderd door een verbeterde voeding. Infectie met trypanosomen gaf een niet significante stijging van de ureum concentraties. Samen met de verlaagde albumine en totaal proteine zijn dit indicators van een versneld proteine metabolisme. De laagste glucose gehaltes werden gevonden in de laag gesupplementeerde en geinfecteerde schapenlammeren. Samen met voorbijgaande piek concentraties van niet-esteroide vetzuren (NEFA) en beta-hydroxy boterzuur (BHBA) die indicators zijn van voorbijgaande ketogenesis, reflecteert dit de trypanosoom-geinduceerde anorexie in de volwassen geinfecteerde ooien gedurende de acute fase van de infectie (4.3). Onafhankelijk van het dieetniveau veroorzaakte een trypanosoom infectie een verandering van het voedingsmetabolisme bij zowel de jonge als volwassen Djallonke ooien. Alhoewel de factor voeding overwegend onafhankelijk optrad, fungeerde het als een positieve buffer tegen de negatieve invloeden van de trypanosoom infectie.

In hoofdstuk 5 wordt de interactie tussen een helminth infectie en een trypanosoom infectie bestudeerd, zowel onder gecontroleerde als onder natuurlijke omstandigheden. Jonge Djallonke ooien werden onderworpen aan hetzij een enkelvoudige *Trypanosoma congolense*, hetzij een enkelvoudige *Haemonchus contortus* infectie, ofwel een combinatie van beide pathogenen in een verschillende volgorde. Analyse van klinische, hematologische en productie parameters wezen duidelijk op de pathogeniciteit van de enkelvoudige infecties met een merkbare verergering wanneer deze infecties gecombineerd werden. Er was een duidelijk immunodepressief effect van een voorafgaande trypanosoom infectie op het verloop van de *Haemonchus* infectie. Deze resultaten geven aan dat trypanotolerantie niet absoluut is en dat een gelijktijdige wormbesmetting resulteert in een verminderd weerstandsvermogen. Onder natuurlijke omstandigheden op het terrein komen menginfecties veelvuldig voor. Daarom zou het voorkomen of behandelen van wormbesmettingen de expressie van trypanotolerantie ten goede kunnen komen. Een profylactisch ontwormingsschema werd getest op het terrein bij natuurlijke besmette dieren (5.2). Hierbij werden positieve effecten aangetoond wat betreft gewichtsaanzet bij schapen, alsook reproductie en algemene gezondheidstoestand van zowel geiten als schapen, dit onder een continue infectiedruk van trypanosomen en natuurlijke wormbesmettingen. Niettegenstaande trypanosomose als een onafhankelijke factor optrad, resulteerde infectie toch in een vermindering van de positieve effecten van ontworming op productiviteits- en gezondheidsparameters. Niettemin waren de productiviteits-indexen van ontwormde schapen en geiten gevoelig verhoogd. Vooraleer dit ontwormingschema aan de boer kan aangeraden worden, moet echter nog een kosten/baten analyse uitgevoerd worden. In ieder geval komt deze interventie de trypanotolerantie in deze rassen ten goede.

In hoofdstuk 6 werd de ziekteresistentie vergeleken tussen naieve Djallonke schapen en de kruisingsproducten (F1) van Sahelian x Djallonke. Beide rassen werden vergeleken in een multi-factorieel experiment met een trypanosoom infectie, natuurlijke wormbesmetting en twee verschillende dieet niveaus. De vergelijking tussen beide rassen heeft duidelijk aangetoond dat de eigenschap trypanotolerantie in Djallonke schapen genetisch bepaald is zoals werd aangetoond door de verschillen in aanzet en intensiteit van de parasitemie en de immuniteitsopbouw na enkelvoudige besmetting. Daarnaast werden eerdere indicaties dat Djallonke schapen een aangeboren resistentie zouden bezitten tegen wormbesmettingen opnieuw bevestigd. Niettegenstaande de verminderde trypanotolerantie en verlaagde resistentie tegen wormbesmettingen van de kruisingsproducten werd duidelijk dat de negatieve impact van beide infecties op de hematocriet als op het gewicht dezelfde was als bij de Djallonke schapen. Effecten van dieet, trypanosoom infectie en wormbesmetting op de parameters hematocriet en gewicht waren onafhankelijk van elkaar en additief. Er was wel een interactie tussen beide pathogenen zoals bleek uit een verhoogde excretie van wormeieren bij met trypanosomen geinfecteerde dieren. Dit kan worden verklaard door het immunodepressief effect dat een trypanosoom infectie teweeg brengt (5.1). Het grotere formaat en de betere groei van de kruisingsproducten t.o.v. de Djallonke schapen van zuiver ras zijn veelbelovend voor intensifiering van de vleesproductie. Optimale productie van dergelijke kruisingproducten is echter onderworpen aan stricte zoo-sanitaire maatregelen en een aangepaste voeding.

In hoofdstuk 7 wordt nader ingegaan op de impact van voeding en voedingsmanagement op de gezondheid en productiviteit van Djallonke schapen en WAD geiten in rurale gebieden. Het concept van gemeenschappelijke "Intensive Feeding Gardens "(IFG) in de dorpen, die uitgebaat worden door lokale boeren, wordt besproken. In deze tuinen wordt vooral gewerkt met *Leucaena* spp. Deze interventie heeft als doel de voedingstoestand van herkauwers in de rurale gebieden te verbeteren. Speciale aandacht wordt gegeven aan drachtige en lacterende schapen en geiten. Deze interventie moet zo goedkoop mogelijk zijn met een minimum aan investering zowel van middelen als van mankracht dit om het aanvaardbaar te maken voor de rurale boeren. De resultaten tonen aan dat de boeren het concept goed accepteerden. Deze tuinen kunnen immers gecombineerd worden met het planten van groenten gedurende het droge seizoen en andere gewassen voor humane consumptie gedurende het regenseizoen. Daardoor worden tegelijkertijd conflicten tussen boeren en veehouders vermeden.

Voedingssupplementen gedurende dracht en lactatie gecombineerd met een aanpassing van het management heeft duidelijk een positief effect op overlevingskansen en groei van de lammeren van zowel schapen als geiten. Supplementering met legumineusen van goede kwaliteit kan mogelijks een positief effect hebben bij kleine herkauwers in rurale gebieden. Het voeder geproduceerd in dergelijke tuinen kan ofwel direct gebruikt worden ofwel geconserveerd en opgeslagen. Bovendien kan het ook gemengd worden in combinatie met andere bestanddelen om supplement-blokken van hoge kwaliteit (HQSB) te produceren. Deze zijn eenvoudig te fabriceren en te bewaren. Al deze interventies zullen worden getest voor verdere ontwikkeling in rurale gebieden en ook in bedrijven rondom de steden waar meer intensieve productie mogelijk is.

CURRICULUM VITAE

Sabine Osaer was born on October 14, 1963 in Torhout, Belgium. In 1987 she obtained the degree of Dr. in Veterinary Medicine (DVM) of the State University in Ghent, Belgium. Between 1987 and 1989, she worked at the Provincial Laboratory for Animal Diseases in Torhout as veterinarian, in charge of all aspects of poultry production in the province of West-Vlaanderen. Between 1989 and 1991 she was employed by the Ministry of Agriculture, Livestock and Fisheries of Surinam in collaboration with the Flemisch Technical Cooperation (VVOB). In Paramaribo, she worked as head of the Veterinary Laboratorium responsible for autopsies and routine parasitological and microbiological analysis of samples of animal origin obtained from the field or the abattoir besides providing technical assistance to the project on Artificial Insemination in Dairy cattle. From 1991 onwards she is employed at the International Trypanotolerance Centre (ITC) in The Gambia. She obtained a Masters degree in Tropical Animal Health at the 'Prince Leopold' Institute of Tropical Medicine (Antwerp) in 1995. At ITC she is involved in applied research on health and productivity of trypanotolerant small ruminants and their possible role in sustainable development of mixed farming in rural areas and meat and milk production for peri-urban areas in West Africa.

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