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Full Length Research Paper

Nutritional alternatives on the haematological and biochemical changes associated with experimental trypanosomiasis in rats

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The influence of improved nutrition on the development of pathophysiological effect of trypanosome in rats fed with two different levels of protein was investigated. The intensities of parasitaemia indicated that there was a tendency for animals receiving the high protein to sustain more parasite numbers than those receiving low protein diets. The infected animals on the two diets showed similar degree of anaemia. The PCV and MCV values were drastically lowered in the two groups of rats with regards to control groups. The albumin concentration showed dietary influences and decreased significantly in the test group. Following treatment, the infected rats on high protein diets recovered and gained weight faster than those on low protein diet. It is therefore, concluded that improved nutrition in the form of higher protein diet (soya bean supplement) intake ameliorates the adverse effect of trypanosome infection and also enhances the rate of recovery following chemotheraphy.

Key words: *Trypanosome brucei*, high protein diet, low protein diet, parasitaemia, PCV, MCV, albumin, *Rattus rattus*.

INTRODUCTION

In many tropical countries, protozoan diseases are of major importance. Parasites particularly gastro-intestinal helminths and protozoa are a major constraint to animal productivity throughout the world. One of the greatest protozoan diseases is the animal trypanosomiasis and posed serious threat to animal production in the sub-saharan Africa (Onditi et al., 2007).

It has been suggested that the nutritional status of the host can influence the pathogenesis of parasitic infection and other diseases in general and it is generally accepted that well nourished animals withstand parasitism better than those less adequately fed (Whitlock, 1949; Gibson, 1963). It has also been frequently reported from the field that during the dry season trypanosomiasis becomes more severe when the quality and quantity of nutrition is particularly low. It is possible that the diet may not only influence host resistance to either the initial infection or re-infection but may also affect the ability of the host to withstand the pathophysiological consequences of infection. The studies that examine these interactions were however replete because of inadequate controls or poorly formulated diets. More recently, attempts have been made to overcome these difficulties and a clearer picture of interaction between the host nutrition and the pathophysiological consequences of parasite infection has started to emerge (Katunguka-Rwakishaya, 2005). The special and approved diet for experimental animals is the mouse cube, since it contains all the known nutritional requirements and metabolisable energy.

However, with the increased production in livestock and subsequent increased demand and with economic situation of the third world coupled with the high cost of maintaining these animals without which research will be seriously affected. With the availability of soya beans grown everywhere around us, these present study was designed to investigate the influence of host nutrition on

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Abbreviations: SBM, Soya bean meal; **PBS,** phosphate buffered saline; LP, low protein; HP, high protein

the pathogenesis of trypanosome infection and attempt is made to compare the effect of soya beans and normal diet on growth and haematological parameters in rat.

MATERIALS AND METHODS

Experimental animals, feeding and housing

Wister rats, aged 4 - 5 months were purchased from the animal house of the Department of Vetinary Anatomy, University of Ibadan. The basic diet consisted of a mixture of shredded sugar-beet pulp (sbp), barley siftings and minerals/vitamins trace elements mixtures. Soya bean meal (SBM) was added to the high protein diet (HP) only.

The soya bean was soaked in water over night boiled dried and ground; the powder was hand moulded with water before use. They were fed for a period of four months after which weight gain/or/loss was assessed and parasitological, haematological and biochemical studies were carried out.

Chemical analysis feed

Representative samples of experimental diet were analysed periodically using standard procedures (MAFF et al., 1981).

Experimental infection

The rats were infected with *Trypanosoma brucei brucei* collected from the pathology laboratory of the Department of Vertinary Pathology, University of Ibadan. The trypanosomes were obtained from irradiated mice during the first rising parasitaemia. Mice were bled by cardiac puncture and their blood was pooled. An estimation of parasitamia was made on the pooled sample, which was then diluted with phosphate buffered saline (PBS) containing 1.5% glucose at pH 8.0 to give 1 x 10^5 trypanosomes in 1 ml of PBS. Each rat received 1 ml of the inoculums intraperitonealy.

Parasitological, haematological and biochemical techniques

The standard techniques used to collect blood samples for parasitological, haematological and biochemical examination have been described previously (Katunguka- Rwakishaya et al., 1992a) briefly, trypanosomes were detected by the dark ground buffy coat method according to Murray et al. (1977) and the intensity of parasitaemia was graded from 0 to 5 as described by Paris et al. (1982).

Experimental design

Eighteen Wister rats were involved in this study. They were divided into two groups of nine each, on the basis of their live weights and introduced to either a low protein (LP) or a high protein (HP) diet. After 4 weeks on their respective diets, six animals from the low protein (LPI group) and six animals from the high protein group (HPI group) were each infected with *T. brucei brucei* ,while the remaining three rats from the each dietary group acted as uninfected controls (LPC and HPC). Twenty-one days after infections, three animals from the HPI group (HPIT) and three rats from the LPI group (LPIT) were treated with a standard trypanocidal drug (isometadium chloride) at a dose rate of 0.1 mg/kg by deep Intraperitoneal injection and the animals were monitored for a further 14 days.

Statistical methods

Comparison between groups were achieved by one way analysis of variance followed by Newman-Kevls multiple range test intensities of parasitaemia were evaluated by the non- parametric Mann-Whitney test. These statistics were performed using Animal designs 1- P values < 0.05 were considered significant.

RESULTS

The prepatent period in both groups was the same, that is, 5 - 7 days after infection. Following patency, parasitemia fluctuated considerably with a tendency to be higher in HPI than in LPI group. Infection was associated with grater retardation of growth in the LP group compared to the LPC group (Figure 1). The gain in the live weight of the LPC group was significantly lower than that of the LPC and HPI groups. However, the HPI and HPC groups grew at similar rates. The weights of the control animals were not significantly different.

There was a significant decrease in PCV values of both infected groups compared with their controls (Figure 2). The decline in PCV values commenced with the appearance of trypanosomes in circulation and both groups of infected animals showed similar degrees of anaemia. The PVC values in control animals fluctuated between 0.38 and 0.42 litres/litre. There were no nutritional influences.

Following infection, both infected groups showed significant increases in MCV compared to their uninfected controls, however, the increase was significantly greater in HPI group than in LPI group. In the HPI group, the mean MCV increased from 31.2 ± 0.3 flat to 0 day after infection (DAI) to 38.5 ± 1.2 flat at 14 days after infection. In the LPI group the MCV values increased from 31.0±07 flat ODA1 to 35.0 ± 0.6 flat 16 DAI before decreasing to 32.3 ± 0.7 at 21 days after infection. In control groups, the MCV fluctuated between 30.0 ± 0.6 and 33.0 ± 0.6 flat with no significant difference between LPC and HPC groups.

The mean plasma albumin concentration decreased significantly in both groups of infected rats and also showed dietary influences. In the HPI group, plasma albumin decreased from 36.2 ± 0.4 g/litre at 0 DAI to 28.7 ± 0.9 g/litre at 21 DAI. In the LPI group, it decreased from 30.8 ± 0.7 to 26.3 ± 0.6 g/litre and tended to recover thereafter. In the control groups, plasma albumin concentration fluctuated between 28.3 and 32.0 g/litre in the LPC and between 32.7 and 36.3 g/litre in the HPC group. The values in the HPC groups were significantly higher than those in LPC group.

Following the treatment of the infected groups HPIT and LPIT, both groups gained weight. The LPIT group gained 0.3 kg while the HPIT group gained 0.9 kg during the three weeks after treatment. The LPC and HPC groups increased groups increased by 1.2 and 1.8kg, respectively, in the same period. Both groups of infected rats showed improvement in their PCV values following treatment;

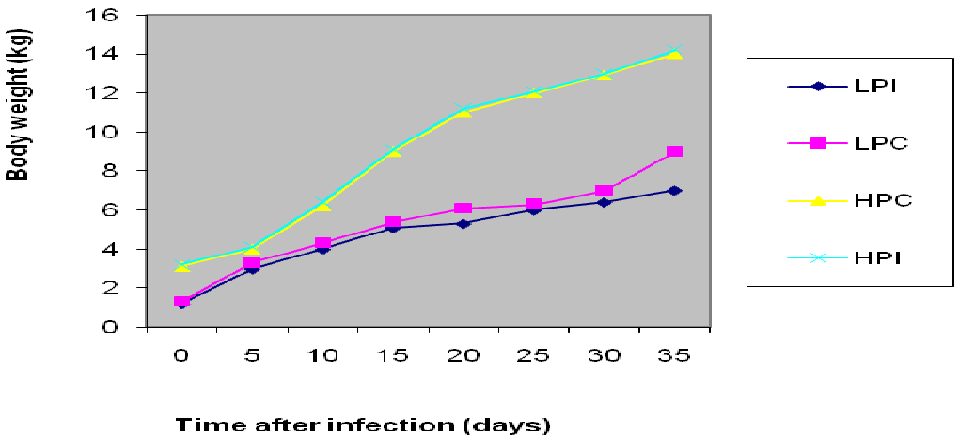

Figure 1. Body weights of rat infected with *T. brucei brucei* and given either high protein or low protein and of the respective controls

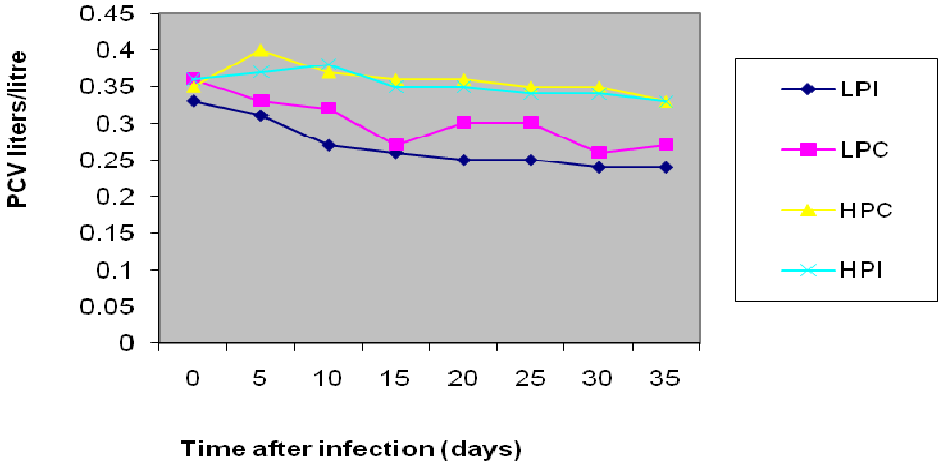

Figure 2. Packed cell volumes (PCV) of rat infected with *T. brucei brucei* and given either a high or low protein diet or the respective uninfected controls.

however, the rate of recovery was moderately faster in HPIT group. By 10 days after treatment, the PCV values of the HPIT groups were 0.32 ± 0.01litres/litre. The values in the LPIT group were 0.28 ± 0.01 while controls were 0.32 ± 0.01 litres/litre.

DISCUSSION

In the present study, it was shown that improved nutrition in the form of increased protein intake with soya bean meal supplements had a marked influence on rates of growth, intensity of parasitaemia, blood biochemical changes and rate of recovery from anaemia following administration of trypanocidal drug.

It was discovered in agreement with the findings of Little et al. (1990) that the improved nutrition did not affect the prepatent periods. The increase in parasitaemia in HPI group immediately, the parasite appears in circulation may be due to availability of proteins and lipids, but these claims have not been substantiated. The retardation of growth observed in the LPI group while HPI and HPC groups grew at smaller rates. These agrees with the report of Hecker et al. (1991) and Agymang et al. (1990) for Djallonke sheep and N'Dama cattle exposed to natural fly challenge. Versegen et al. (1991) and Zwart et al. (1991) reported the development of fever associated with heat production and increased metabolisable energy for maintenance hence, the proportion of feed left for growth is reduced; therefore low weight in the animals. This is more manifest in the rats on Low Protein diet (Katunguka-Rwakishaya, 2005).

Similar degrees of anaemia were recorded in both groups of infected rats. Little et al. (1990) had reported that the rate of development of anaemia in cattle inoculated with *Trypanosome congolese* and supplemented with

groundnut cake was slower than in unsupplemented cattle. In contrast to the report presented in this study, improved nutrition does not influence trypanosome establishment and the rate of development of anaemia (Kalunguka-Rwakishaya, 2005). The anaemia in HPI was macrocyte while it was normocytic in the LPI group. These findings suggest enhanced erythropoietic activity as a response to infection in the LPI group.

The observed hypoalbuminaemia in the infected dietary groups may occur because of the uptake of albumin-bound fatty acid and lipoproteins (Vickerman and Tetley, 1979) and haemodilution (Katunguka-Rwakishaya et al., 1992b) in trypanosome infected animals. In conclusion, the study here reported that an improvement of host nutrition is important in moderating the severity of patho-physiological consequences in trypanosome infection and also influences the rate of recovery from anaemia following chemotherapy.

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