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ANAEMIA IN HUMAN AFRICAN TRYPANOSOMIASIS CAUSED BY *TRYPANOSOMA BRUCEI RHODESIENSE*
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ANAEMIA IN HUMAN AFRICAN TRYPANOSOMIASIS CAUSED BY *TRYPANOSOMA BRUCEI RHODESIENSE*

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ABSTRACT

Objective: **To find out if indeed anaemia is a major sign in human trypanosomiasis caused by *Trypanosoma brucei rhodensiense.***
Design: **A one year cross-sectional study of all admitted and surveyed *Trypanosoma brucei rhodensiense* infected patients (June 2001-June 2002)**
Setting: **Nkhotakota District Hospital-Central Region of Malawi.**
Results: **After survey and investigations, 28 patients (16 males and 12 females) were admitted to Nkhotakota District Hospital with a parasite positive *Trypanosoma brucei rhodensiense* infection. Twenty four (85.7%) of them were anaemic. Their mean haemoglobin was 8.96 ± 3.07 g/dl compared to controls that had a mean haemoglobin concentration of 12.17 ± 1.35 g/dl ($p < 0.000001$, 95% CI -4.342 to -2.0785) ($n = 45$). None of the trypanosomiasis infected individuals had schistosomiasis or hookworms. Two patients had malaria. One of them was an 18-year-old pregnant woman with hepatosplenomegaly, who developed ante partum haemorrhage. She was jaundiced and had haemoglobin of 10 g/dl. She died after two weeks following the diagnosis and treatment. The other was a two-year-old girl who had haemoglobin of 8.4 g/dl. She also had hepatosplenomegaly. All the other patients looked well nourished with no other signs of chronic diseases. Hepatosplenomegaly was significantly related to the severity of illness ($p = 0.011$) but not to anaemia.**
Conclusion: **Though basic, this study has shown that anaemia is indeed a complication of human Africa trypanosomiasis caused by *Trypanosoma brucei rhodesiense.* There is need for further investigation to investigate the type of anaemia that is caused by this disease.**

INTRODUCTION

Infection by haemoflagellate parasites of *Trypanosoma brucei rhodesiense* and *Trypanosoma brucei gambiense* give rise to a disease called African trypanosomiasis (sleeping sickness). This disease is exclusive to the African subcontinent. *T. brucei gambiense* gives rise to a chronic disease while that caused by *T. brucei rhodesiense* gives rise to an acute disease. There are two stages of the disease, stage 1 is when the parasites are restricted to the haematolymphatic system while stage 2 denotes central nervous system involvement. Trypanosomiasis is characterised by symptoms similar to malaria(1). Just as malaria this disease also causes anaemia in infected hosts in both humans and animals(2-4). Despite the extensive knowledge of mechanisms of malaria-induced anaemia, the molecular mechanisms associated with trypanosomiasis-induced anaemia are largely under-studied. There is some evidence that anaemia induced by trypanosomes may be haemolytic in type(2,5-7).

There is extensive information on trypanosomiasis in association with anaemia in domestic animals, with little information available on the association of this disease with anaemia in humans. However, it is well established that there is a marked variation in the susceptibility of different species to anaemia following trypanosomal infections(8-11). In the susceptible animals studied, it is clear that trypanosomal infection leads to generalised pancytopenia(8,12-14). The present study was designed to study the association of *T brucei rhodesiense* with anaemia in patients that were admitted to the Nkhotakota District Hospital, Malawi, Central Africa following both active and passive screening.

MATERIALS AND METHODS

Patients: After the ethical approval from the University of Malawi, College of Medicine Ethics committee, twenty eight patients were recruited into the study after informed consent between June 2001 and June 2002. Age ranged from 1.5 years to 55 years, the median age was 23.4 years. Twenty seven had stage one disease and one had stage two disease.

Forty five non-trypanosome infected people from the villages where active surveillance was done were recruited as controls.

Procedure: Diagnosis of Trypanosomiasis was made on the basis of finding trypanosomes in a thick blood film. The film was made by collecting a drop of capillary blood on a slide and spreading it evenly on an area 15-20 mm in diameter. The smear was allowed to completely dry and was stained using Field's rapid technique for thick blood film(15). The preparation was allowed to dry and a drop of immersion oil was spread on the film and the film was examined microscopically using 40X objective followed by 100X if the parasites were seen to confirm the diagnosis.

Sleeping sickness is staged by looking for parasites in the cerebrospinal fluids and the number of white cells that are found in the cerebrospinal fluid. Lumbar puncture was performed on all the patients as part of their clinical management for staging of the disease. Haemoglobin was estimated using Hemocue (Hemocue Ltd, Dronfield, Derbyshire, UK) according to manufacturer' s instructions. Patients were treated according to the guidelines recommended by the Ministry of Health in Malawi. Urine and stool microscopy were performed routinely and the thick films for trypanosomiasis were also used to diagnose malaria as the same technique is used for the detection of both parasites(15). For statistical analysis, multinomial logistic model was used.

RESULTS

There were 16 males and 12 females that were diagnosed with trypanosomiasis at Nkhotakota District Hospital during the study period (Table 1). None of them had palpable lymph nodes or obvious somnolence. No one had parasites either in their urine or stool. The mean haemoglobin value (Figure 1) of trypanosome -infected people was significantly lower than the controls ($p < 0.000001$, 95% CI -4.34 to -2.08). Of the 28 patients that were diagnosed with trypanosomiasis, 85.7% were anaemic while 14.3% had normal haemoglobin levels.

Table 1

The Trypanosomiasis patient profile for Nkhotakota District Hospital from June 2001- June 2002

Duration of Illness	Total cases	Anaemia	Hepatosplenomegaly	Deaths
<1	14	85.7 (12)	42.9 (6)	7.1 (1)
1-2	5	100 (5)	80 (4)	10.5 (1)
3-4	2	50 (1)	0 (0)	0 (0)
5-6	5	100 (5)	100 (5)	40 (2)
>6	2	50 (1)	50 (1)	0
Total	28	(24)	(16)	(4)

* Numbers represent percentages with number of cases in brackets

Figure 1

Mean haemoglobin values (g/dl) and SD of trypanosome infected patients and controls

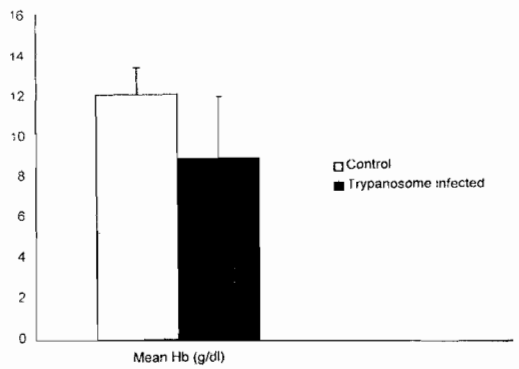

Table 1 shows proportions of trypanosome infected patients with anaemia and hepatosplenomegaly in relation to the duration of the disease. Half of the patients presented within a month following the symptoms. Most of them were anaemic and 42.9% had hepatosplenomegaly. The only person in this group who died was an 18-year old pregnant woman who had both trypanosomiasis and malaria. She developed antepartum haemorrhage. It was during the investigation for malaria that trypanosome parasites were also detected. She was jaundiced and had haemoglobin of 10g/dl. 17.6% of patients presented more than one month but less than two months of their illness. All of them were anaemic and 80% had hepatosplenomegaly. One patient in this group died. This patient was a two-year-old girl with both trypanosomiasis and malaria. She presented with fever, haemoglobin of 8.4g/dl with hepatosplenomegaly, had weight loss, failure to thrive, was pale and had silky hairs. 7.1% of patients presented within three to four months of the disease. They had no organomegally while one was anaemic. 17.9% of patients presented within five to six months of the symptoms. All these patients were anaemic and also had hepatosplenomegaly. Fourty per cent of them in this group died. One of them was a 42-year-old man with haemoglobin of 6.5 g/dl and the other was of a 9-year-old boy with haemoglobin of 7.4g/dl. Two people (7.1%) presented after six months. In this group one was anaemic while the other had splenomegally.

DISCUSSION

The present study though basic concurs with other studies that have demonstrated anaemia in patients with human trypanosomiasis(4-7). However, most of previous studies have attributed anaemia to other parasitic infections and malnutrition rather than to human trypanosomiasis. Hepatosplenomegaly is usually associated with chronic malaria infection in general. In this study it has been shown that while hepatosplenomegaly is associated with general poor health, it plays little or no role on the anaemia of sleeping sickness infected individuals.

There are various arguments that can be put forward to show that indeed trypanosomiasis was the major cause of anaemia in the present study. The patient cohort that was studied had enough sources of protein because Nkhotakota district is one of the lakeshore districts with fish and game animals as staple diets. In addition, none of the people was detected with either hookworm or schistosomes in the present study, which are some of the major causes of anaemia in the tropics. The effect of chronic malaria as a cause of anaemia cannot be ruled out in the present study. However, the high haemoglobin levels found in the control population some of whom had hepatosplenomegaly and malaria makes it unlikely that malaria plays a major role in anaemia in the population studied. In addition, splenomegaly, which is one of the major signs of chronic malaria infection, was common even in people that were not anaemic ruling out the impact of malaria on anaemia in the population studied.

One cannot rule out the effect of chronic diseases such as human immunodeficiency virus (HIV) that is endemic in the area on anaemia, but most of the people that were presented to the study were rural people with low prevalence rates of HIV infections. If there were people with HIV infection, they did not present to the hospital in the terminal acquired immune deficiency syndrome stage (AIDS). Two deaths that occurred within two months of disease could be attributed to combination of trypanosomiasis and other pathological conditions. The other two deaths that occurred were of patients that presented within five to six months of their illness with low haemoglobin levels. One might therefore argue that trypanosomiasis left untreated can lead to anaemia, that may contribute to poor health outcomes as shown by the above two cases. It is unlikely that hepatosplenic schistosomiasis contributed to the pathological outcomes in this study as no parasites were found in both stool and urine in trypanosome infected individuals. While not everyone became anaemic following trypanosomiasis infection, the ones that delayed in presentation had worst anaemia with extremely low haemoglobin. The other trypanosomiasis infected patients had mild anaemia and were discharged with a well-recovered haemoglobin level after finishing the course of treatment in the hospital. These observations while they do not by themselves rule out other causes of anaemia in trypanosomiasis infected-patients, they however make them very unlikely as major contributors of anaemia.

The mode of anaemia-induced trypanosomiasis is largely unresolved. However most studies both in humans and animals have contributed this anaemia to haemolysis(10,14,27). One study in mice showed that the haematopoietic stem cell compartment might be involved in anaemia caused by trypanosomes(12). The present study was not designed to show the association between stem cell proliferation and severity of anaemia in a trypanosome infected individual as resources were not adequate to conduct a thorough haematological investigation on sick people. A full blood count screen would have been ideal in order to answer the question of the type of anaemia. Other studies have clearly shown that some cytokines are raised in trypanosomias infected patients such as IL-10 in human African trypanosomiasis caused *Trypanosoma brucei rhodesiense*(16). IL-10 is well known to inhibit the production of cytokines such as IL-1, IL-6 GM-CSF that recruit haematopoietic stem cell into proliferation(17). This study may serve as a base for further investigations into anaemia induced by trypanosomes in human and animal models on the cascade of events that takes place at the haematopoietic stem cell compartment.

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