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Effect of Tetracycline on Late-stage African trypanosomiasis in Rats

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Abstract

The effect of tetracycline on late stage African trypanosomiasis was examined in an *in vivo* experiment using rats infected with *Trypanosoma brucei brucei*. Infected rats were treated on the 5th day of infection with 10mg/kg and 20mg/kg rat weight of tetracycline and tetracycline hydrochloride. Tetracycline at 10mg/kg extended the life-span of infected rats from 6 days to 11 days, but with 20mg/kg the rats died on day 6. Tetracycline–HCl at 10 and 20mg/kg extended the life-span to 9 and 12 days respectively. The trypanocidal effect of tetracycline on late stage trypanosomiasis is probably due to its ability to penetrate the blood brain barrier and its probable role in inhibiting ribonucleotide reductase through iron chelation. We suggest that tetracycline-HCl and low concentrations of tetracycline can be used in the clinical management of African Trypanosomiasis.

Key words: African trypanosomiasis, Tetracycline, Late Stage

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INTRODUCTION

Human African trypanosomiasis is a fatal disease which affects some 36 developing African countries south of the Sahara. It exist in the rural areas and about 300,000 new cases are reported annually (WHO, 1998). Upon infection through the bite of a tsetse fly the parasite proliferate rapidly by binary fission. Lymph and blood are invaded and the organism progressively disseminate to the bone marrow and tissue fluids, and in the second stage (late-stage) it penetrate the central nervous system and cerebrospinal fluid (Masake et al, 1997).

Therapy of human African trypanosomiasis in the early stage relies on pentamidine, suramin, or diminazene aceturate (Atouguia and Costa, 1999), none of which could cross the blood brain barrier in sufficient quantity to prevent relapses of late-stage cases of the disease (Keiser et al, 2001). Melarsoprol was introduced in 1949 and is used in the treatment of late-stage trypanosomiasis but severe adverse events frequently occur. DFMO which is the most recent of the drugs is not effective against *T. b. rhodesiense* and is very expensive (Seebeck et al, 1999). Furthermore there is a constant threat that production of the currently used drugs will be interrupted because it is not profitable. New cost efficient and easy to use drugs are therefore urgently needed (Keiser et al, 2001).

Tetracycline, an antimicrobial drug with iron-chelating property (Grenier et al, 2000) have been found in a previous experiment to be effective against the early stage of the infection and the trypanocidal action was suggested to be as a result of iron-chelation (Ekanem et al, 2002) which could inhibit ribonucleotide –reductase, the iron-requiring cell cycle-dependent enzyme that plays a central role in cell division and proliferation. In this study we have examined the effect of tetracycline and tetracycline-HCl which is the commercially available form of the drug on the parasitaemia and life-span of rats infected with *T. b. brucei.*

MATERIALS AND METHODS

Lafia strain of *T. b. brucei* was collected from the veterinary and Livestock Studies Department, Nigerian Institute for Trypanosomiasis Research (VSL-NITR), Vom near Jos in Nigeria. Wister rats weighing approximately 200g were obtained from the Animal House, Department of Biochemistry, University of Ilorin. Tetracycline and Tetracycline hydrochloride were products of Sigma Chemical Company, Poole, England. The parasite was passaged intraperitoneally into rats and parasitaemia was observed on a daily basis by counting parasites in thin blood smears under light microscope. Drugs were administered intraperitoneally twice daily. 10mg/kg and 20mg/kg tetracycline and tetracycline–HCl were administered to infected rats from the 5^{th} day of infection until the rats died. Control rats were infected but not treated with any drug.

RESULTS AND DISCUSSION

10mg/kg tetracycline extended the life span of infected rats from 6 days for the control to 11 days, but the rats died on day 6 when 20mg/kg tetracycline was administered (fig 1). This could be as a result of the high volume of water used for dissolving it, since tetracycline is only sparingly soluble in water (Sande and Mandell, 1980) 10 and 20mg/kg tetracycline-HCl reduced the parasitaemia and extended the lifespan of infected rats to 9 and 12 days respectively (fig 2). The fact that low concentration of tetracycline effectively extents the life span of infected rat (fig 1) is probably because tetracycline is a more effective trypanocide

than tetracycline–HCl as previously determined (Ekanem et al, 2002).

The effect of Tetracycline on late stage trypanosomiasis in this experiment may be attributed, firstly, to its antimicrobial activity.

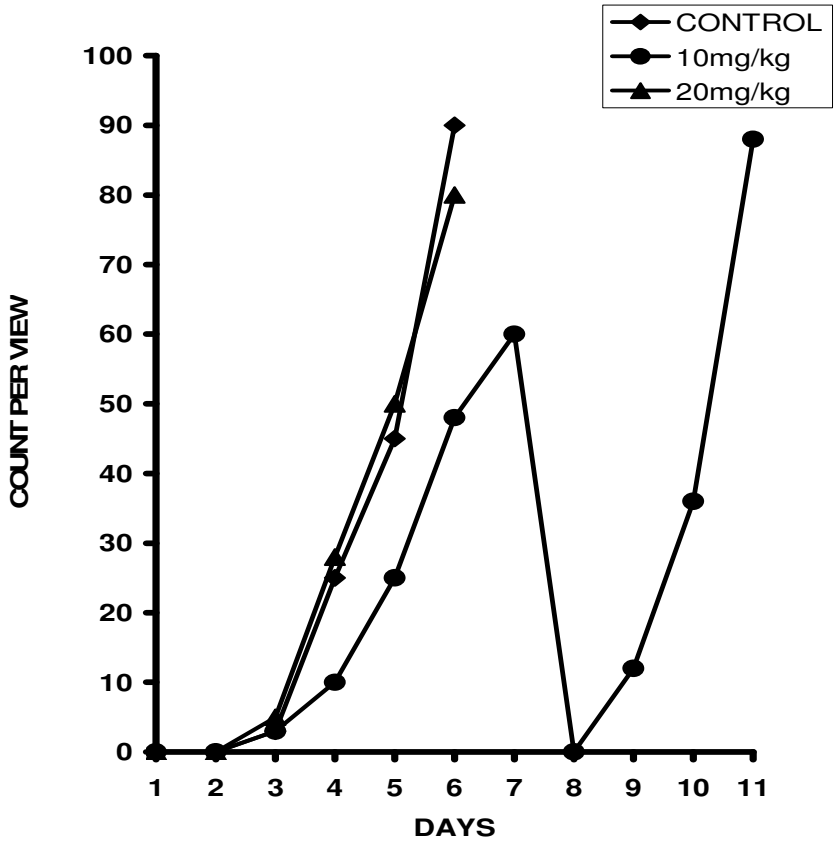

Fig. 1: Parasite count in T. brucei infected rats treated with tetracycline twice daily as the infection progressed until death. Treatment of rats started with 10mg/kg and 20mg/kg on the 5th day of infection.

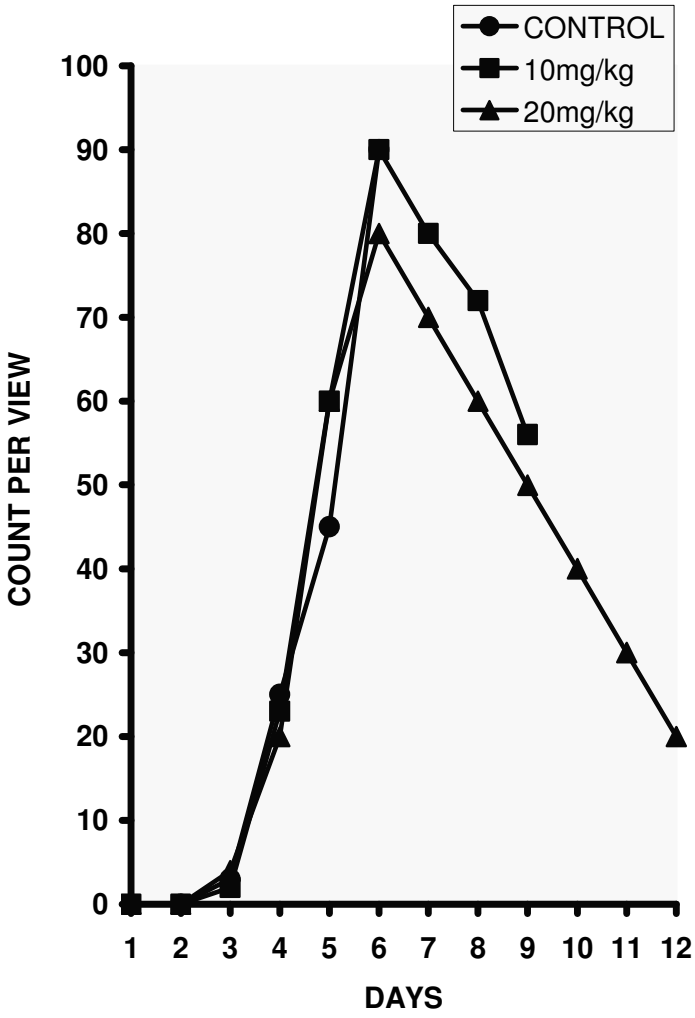

Fig. 2: Parasite count in T. brucei - infected rats treated with Tetracycline - HCl twice as the infection progressed until death. Treatment of rats started with 10mg/kg and 20mg/kg on the 5th day of infection.

It has the ability to suppress the growth of bacteria, as it does to periodontopathogenic gram-negative anaerobic bacteria, in the treatment of periodontitis, a disease affecting the tooth-supporting tissues (Rifkin et al, 1993). Secondly, the iron-chelating activity of tetracycline have been suggested to contribute to its antimicrobial activity (Grenier, et al, 2000), and it has been shown in a previous experiment that the trypanocidal action of tetracycline is related to this property (Ekanem et al, 2002). Another factor is that the distribution of tetracycline in most body fluids and tissues is excellent (Sande and Mandell, 1980). Depending on route and duration of treatment, tetracycline penetrates the cerebrospinal fluid (Sande and Mandell, 1980). The intravenous injection of the drug results in the gradual appearance of the drug in the spinal fluid over a period of 6 hours. Oral therapy yields very low spinal fluid concentrations. The rickettsial disorders which are diseases involving the central nervous system are treated effectively with tetracycline-derivatives (Bleck, 1999). We have earlier suggested the possible use of tetracycline for early stage of African trypanosomiasis (Ekanem et al, 2002). From the results of this experiment we also suggest that tetracycline at low concentration and tetracycline HCl can extend the life span of infected rats and may therefore be considered as a possible drug for the management of late stage African sleeping sickness.

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