Journal of Coastal Life Medicine

journal homepage: www.jclmm.com

Document heading doi:10.12980/JCLM.3.2015JCLM-2014-0103

Classical clinical signs in rats experimemtally infected with *Trypanosoma brucei*

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PEER REVIEW

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Comments

The authors made an effort to investigate the clinical manifestations of trypanosomosis in rats, and the present manuscript propose that clinical signs of *T. brucei* infection could be diagnostic. The topic is an interesting one, and the authors present some interesting results.
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ABSTRACT

Objective: To investigate clinical signs in *Trypanosoma brucei* infection in albino rats.
Methods: Fourteen rats grouped into 2 with 7 rats in each group were used to determine classical clinical manifestation of *Trypanosoma brucei* infection in rats. Group A rats were uninfected control and Group B rats were infected with *Trypanosoma brucei*.
Results: Parasitaemia was recorded in Group B by (3.86±0.34) d and the peak of parasitaemia was observed at Day 5 post infection. Classical signs observed included squint eyes, raised whiskers, lethargy, no weight loss, pyrexia, isolation from the other rats, and starry hair coat.
Conclusions: These signs could be diagnostic or aid in diagnosis of *Trypanosoma brucei* infection in rats.

KEYWORDS
Classical, Trypanosomosis, Clinical signs, *Trypanosoma brucei*, Rats

1. Introduction

Trypanosomiasis is a collective term for a group of diseases caused by one or more of the pathogenic trypanosomes species[1]. Various species of trypanosomes affect different species of animals, producing varying disease conditions. There is a range of variation from the very acute disease in pigs caused by *Trypanosoma simiae* to the usually mild conditions and *Trypanosoma brucei (T. brucei)*/ *Trypanosoma evansi* infections in cattle[1]. *Trypanosoma vivax* induces a hyperacute

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Article history:
Received 11 Agu 2014
Received in revised form 13 Agu 2014
Accepted 25 Agu 2014
Available online 15 Sep 2014

haemorrhagic form of the disease in cattle, sheep and goats[1]. In dogs, *T. brucei* induces a severe disease condition[2], while *T. brucei* and *Trypanosoma rhodesiense* induce a fatal central nervous system disease comparable to that seen in man[3]. *Trypanosoma cruzi* infection in dogs causes chronic myocarditis, dilation of the heart, electrocardiographic abnormalities, cardiac complications and death[4]. The disease in horses is generally characterized by infiltration of subcutaneous tissues causing swelling of the eyelids, lips and skin beneath the lower jaw as what is seen in dogs[1]. Despite the variations in the disease conditions caused by different species of trypanosomes, infected animals generally manifests clinical signs of pyrexia, weight loss, pale mucous membrane, anaemia, ocular discharges and lymphadenopathy[2]. The aforementioned salient signs of trypanosomosis and the species specific signs may be diagnostic in animals[5]. However, not much attention has been paid on the peculiarity of clinical manifestations of trypanosomosis in rats. Hence, the objective of this study was to investigate the clinical signs in *T. brucei* infection in albino rats.

2. Materials and methods

Fourteen 9-week old pathogen-free albino rats of both sexes weighing 150-300 kg were used in this study. The rats were breed in the labouratory animal house of Department of Veterinary Medicine, Michael Okpara University of Agriculture, Umudike. The rats were fed and watered *ad libitum* prior to commencement of the study. Each mouse was identified with picric acid stain.

The *T. brucei* parasite used in this study was a Federe strain obtained from the National Institute of Trypanosomosis and Onchocerciasis Research, Vom, Plateaue State, Nigeria. The parasites were cryopreserved in liquid nitrogen, from where donor rats were initially infected. The parasites were maintained by serial passage in rats at the Department of Veterinary Medicine, Michael Okpara University of Agriculture, Umudike.

About 2.5×10^6 trypanosomes suspended in 1 mL of normal saline was used to infect each experimental rat with the aid of 1 mL tuberculin syringe (*i.p.*). The quantity of parasite was estimated by using the rapid matching method of Herbert and Lumsden[6].

Fourteen albino rats were randomly devided into 2 groups with 7 rats in each group. Group A was the un-infected control; Group B was infected with *T. brucei*.

Parasitaemia was determined by using two methods: the wet bloods mount technique and the haematocrit buffy coat method described by Woo[7].

The clinical signs were evaluated by using the scoring method essentially described by Jensen *et al*[8]. Briefly, the range of degree of the lesions was divided into ordinal classes, *viz.*, absent (0), mild (+), moderate (++) or severe (+++).

Data obtained from this study were presented as mean±SE. Statistical significance were analyzed by using One way analysis of variance and Duncan's multiple range test with statistic package for social science version 16. The level of significance was accepted at $P\leqslant0.05$[9].

3. Results

T. brucei infection induced an acute form of the disease in rats with some mortality. The parasitaemia was detected in Group B on (3.86±0.34) d. The peak of parasitaemia was recorded in most of the rats on 5 d post infection and on Days 9, there was 80% mortality recorded in the infected group.

As seen in Table 1, most of the infected rats isolated themselves from the members. Almost all the infected rats had starry hair coats and were anaemic. Majority of the infected rats had squinted eyes and showed dullness (Figure 1), raised whiskers and starry hair coat (Figure 2). There were elevated body temperature but with no significant ($P<0.05$) change in weight (Figure 3) in infected group rats when compared with the control. On Days 5, the peak of parasitaemia was observed in the infected group (Table 2), and Figure 4 showed the presence of trypanosomes in the blood of an infected rat. By Days 9, there was 80% mortality in the infected rats.

Table 1
Clinical manifestations in rats infected with *T. brucei.*

Clinical signs	Group A	Group B
Isolation	0	++
Starry hair coat	0	+++
Raised whiskers	0	++
Lethargy	0	++
Flabby body	0	++
Anaemia marked by scanty or no blood from the tail vein	0	+++
Squint eyes	0	++
Pyrexia	0	+++
Weight loss	0	0

+: Mild; ++: Moderate; +++: Severe; 0: Absent.

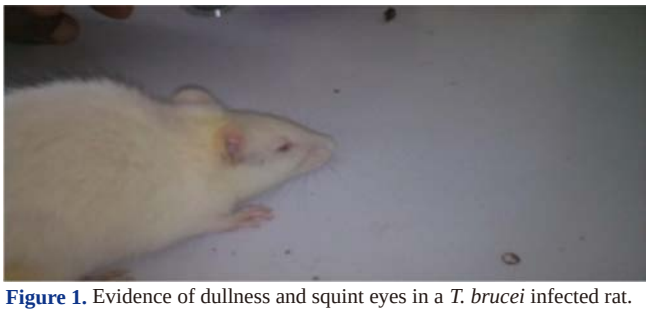

Figure 1. Evidence of dullness and squint eyes in a *T. brucei* infected rat.

Figure 2. Evidence of starry hair coat and raised whiskers.

Table 2

Weight (kg) of rats experimentally infected with *T. brucei*.

Experimental period (days)	Group A	Group B
0	2.21±1.49[a]	2.21±2.14[a]
1 [#]	2.00±1.54[a]	2.30±1.50[a]
2	2.00±1.02[a]	2.10±1.34[a]
3	2.00±1.34[a]	2.30±1.44[a]
4	2.00±1.24[a]	2.10±1.04[a]
5[+]	2.00±1.54[a]	1.93±1.70[a]
6	2.00±1.34[a]	2.00±1.04[a]
7	2.00±1.04[a]	1.93±1.70[a]
8	2.00±1.54[a]	2.20±1.20[a]
9[*]	2.00±1.00[a]	2.00±1.04[a]
10	2.00±1.04[a]	2.10±1.24[a]
11	2.20±1.24[a]	2.00±1.04[a]
12	2.10±1.00[a]	2.20±1.34[a]

Superscripts a represents the homogeneity between the experimental groups at $P \leqslant 0.05$. Data were expressed as mean±SE.

#: Day of *T. brucei* infection; +: Day of peak of parasitaemia; *: Day of 80% mortality.

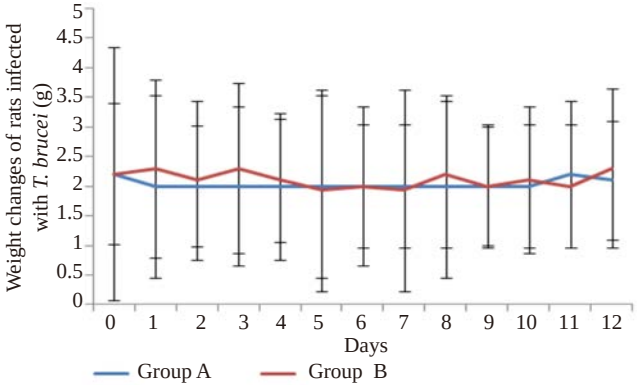

Figure 3. Graph of clinical weight changes of rats with *T. brucei* infection.

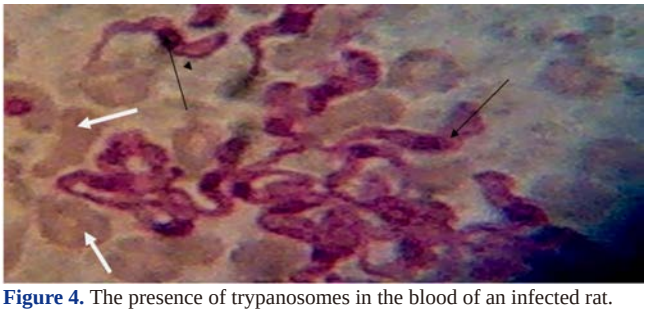

Figure 4. The presence of trypanosomes in the blood of an infected rat. The black arrow points at trypanosome parasite and the white arrow points at red blood cells.

4. Discussion

In this study, establishment of parasiatemia on (3.86±0.34) d was in line with previous works done in *T. brucei* infection in dogs[2,10]. In trypanosomosis, anaemia is seen as the cardinal sign of the disease in infected animals[11,12]. Anaemia is commonly indicated in infected animals by evidence of paleness of the mucous membrane[2]. In this study, anaemia in the infected rats was indicated by reduction or scanty blood from the tail vein. In trypanosomosis, anaemia is exacerbated at the peak of parasitaemia which was recorded at Days 5 post infections[13]. This is in line with previous findings in *T. brucei* infection in animals[14,15]. Also in this study, there was no significant change in the weight of the infected rats compared to the control. This could be due to the acuteness of the disease in the rats which inhibits apparent reduction in weight. This was similar to that recorded in *T. brucei* infection in dogs[10,16]. The isolation of infected rats from the other rats, starry hair coat, raised whiskers, flabby body and squint eyes were the classical clinical signs of *T. brucei* infection observed in the infected rats. The squint eyes and raised whiskers are indicators of pain as observed in a study on assessment of pain in rats (unpublished article). Other clinical signs such as pyrexia and lethargy were similar to what recorded in previous works on trypanosomosis in animals[2,17]. It can be concluded that, clinical signs of *T. brucei* infection could be diagnostic or aid in diagnosis of *T. brucei* infection.

Conflict of interest statement

We declare that we have no conflict of interest.

Comments

Background

Trypanosomosis is a collective term for a group of diseases caused by one or more of the pathogenic trypanosomes species. Various species of trypanosomes affect different species of animals producing varying disease conditions. The salient signs of trypanosomosis and the species specific signs may be diagnostic in animals.

Research frontiers

Not much attention has been paid on the peculiarity of clinical manifestations of trypanosomosis in rats. Hence, the present study investigated clinical signs in *T. brucei* infection in albino rats.

Related reports

There are other reports related to trypanosomosis in animals such as dogs. Also, other clinical signs such as pyrexia, and lethargy were recorded in other works on trypanosomosis in animals.

Innovations and breakthroughs

The present study was conducted due to the paucity of information on the classical clinical signs of *T. brucei* infection in rats. Authors proposed that these clinical signs could be diagnostic in case of *T. brucei* infection in rats.

Applications

Classical signs observed such as squint eyes, raised whiskers, lethargy, pyrexia, isolation from the other rats, and starry hair coat could aid in diagnosis of *T. brucei* infection in rats.

Peer review

The authors made an effort to investigate the clinical manifestations of trypanosomosis in rats, and the present manuscript propose that clinical signs of *T. brucei* infection could be diagnostic. The topic is an interesting one, and the authors present some interesting results.

References

[1] Food and Agriculture Organization. Clinical signs. Italy: Food and Agriculture Organization; 2014. [Online] Available from: www.fao.org/ag/againfo/programmes/en/paat/d_signs.html [Accessed on 5th September 2013]

[2] Nwoha RIO, Anene BM. Clinical signs and pathological changes in dogs with single and conjunct experimental infections of *Trypanosoma brucei brucei* and *Ancylostoma caninum*. *J Vet Parasitol* 2011; **25**(2): 97-102.

[3] Aquino LPCT, Machado RZ, Alessi AC, Santana AE, Castro MB, Marques LC, et al. Hematological, biochemical and anatomopathological aspects of the experimental infection with *Trypanosoma evansi* in dogs. *Arq Bras Med Vet Zootec* 2002; doi: 10.1590/S0102-09352002000100002.

[4] Companion Animal Parasite Council. Current advice on parasite control: vector-borne diseases-Trypanosomiasis. Species: *Trypanosoma cruzi*. Salem: Companion Animal Parasite Council; 2014. [Online] Available from: http://www.capcvet.org/capc-recommendations/trypanosomiasis [Accessed on 9th June, 2013]

[5] Luckins AG. Methods for diagnosis of trypanosomiasis in livestock. Italy: Food and Agriculture Organization. [Online] Available from: http://www.fao.org/livestock/agap/frg/FEEDback/War/u6600b/u6600b0a.htm. [Accessed on 5th September, 2013]

[6] Herbert WJ, Lumsden WHR. *Trypanosoma brucei*: a rapid "matching" method for estimating the host's parasitemia. *Exp Parasitol* 1976; doi: 10.1016/0014-4894(76)90110-7.

[7] Woo PT. The haematocrit centrifuge technique for the diagnosis of African trypanosomiasis. *Acta Trop* 1970; **27**: 384-386.

[8] Jensen JC, Nielsen LH, Arnason T, Cracknell V. Elimination of mange mites *Sarcoptes scabiei* var. *suis* from two naturally infested Danish sow herds using a single injection regime with doramectin. *Acta Vet Scand* 2002; **43**(2): 75-84.

[9] Snedecor GW, Cochran WG. *Statistical method*. 6th ed. Amess: Iowa state University press; 1968.

[10] Akpa PO, Ezeokonkwo RC, Eze CA, Anene BM. Comparative efficacy assessment of pentamidine isethionate and diminazene aceturate in the chemotherapy of *Trypanosoma brucei brucei* infection in dogs. *Vet Parasitol* 2008; **151:** 139-149.

[11] Ekanem JT, Yusuf OK. Some biochemical and haematological effects of black seed *Nigella sativa* oil on *Trypanosoma brucei*-infected rats. *Afr J Biomed Res* 2008; **11**: 153-157.

[12] Akanji MA, Adeyemi OS, Oguntoye SO, Sulyman F. *Psidium guajava* extract reduces trypanosomosis associated lipid peroxidation and raises glutathione concentrations in infected animals. *EXCLI J* 2009; **8**: 148-154.

[13] Anosa V, Jennings FW, Urquhart GM. The effect of splenectomy on the anaemia of *Trypanosoma brucei* infection of mice. *J Comp Pathol* 1977; **87**: 569-580.

[14] Egbe-Nwiyi TN, Igwe NC, Igbokwe IO, Nwosu CO. Effect of oral copper sulphate supplementation on the severity of *Trypanosoma brucei* and *Trypanosoma congolense* infections in rats. *Niger Vet J* 2005; **26**: 8-17.

[15] Egbe-Nwiyi TN, Aliyu MM, Igbokwe IO, Effect of oral supplementation with manganese chloride on the severity of *Trypanosoma brucei* and *Trypanosoma congolense* infections in rats. *Afr J Biomed* 2010; **13**: 27-31.

[16]Nwoha RIO, Anene BM. Clinical signs and pathological changes in dogs with single and conjunct experimental infections of *Trypanosoma brucei brucei* and *Ancylostoma caninum*. *J Vet Parasitol* 2011; **25**(2): 97-102.

[17] Eloy LJ, Lucheis SB. Canine trypanosomiasis: etiology of infection and implications for public health. *J Venom Anim Toxins Incl Trop Dis* 2009; doi: 10.1590/S1678-91992009000400002.