The Journal of Veterinary Science

Pathological Changes Associated with Experimental *Trypanosoma congolense* Infection in Pigs

Victor I.\textsuperscript{a*}, Num S.M.\textsuperscript{b}, Ogbaje C.I.\textsuperscript{c}

\textsuperscript{a} Department of Veterinary Medicine, University of Agriculture P.M.B. 2373, Makurdi, Benue State, Nigeria
\textsuperscript{b} Department of Veterinary Pathology and Microbiology, University of Agriculture, Makurdi, Benue State, Nigeria
\textsuperscript{c} Department of Veterinary Parasitology and Entomology, University of Agriculture, Makurdi, Benue State, Nigeria


Article history:
Received: 20 April, 2014
Accepted: 25 April, 2014
Available online: 27 July, 2014

Keywords:
Pathological, changes, *Trypanosoma congolense*, Infected, Pigs, and Zaria

Corresponding Author:
Victor I.\textsuperscript{*}
Email: vicshallom@yahoo.com

Abstract
Trypanosomosis is infection with protozoan haemoparasite of the genus *Trypanosoma*. The gross and histopathological changes due to experimental *Trypanosoma congolense* in infection in Pigs was evaluated. Pigs infected developed parasitaemia, but no mortality was recorded. Post mortem findings were rough hair coat, emaciation and paleness of carcass, splenomegaly, slightly enlarged and haemorrhagic kidneys, lymphadenitis and congested heart. Cytopathologically, there was depletion of lymphocytes at the germinal centers of the spleen; some seminiferous tubules of the testis were devoid of spermatids, lungs and liver showed massive mononuclear cellular infiltrations with lobular boundaries wider than normal. Infected parasitaemic pigs were able to spontaneously clear the parasites from circulation without chemotherapy by day 38 post-infection. In conclusion, *T. congolense* infection in pigs causes trypanosomosis evident by penile protrusion, haemorrhages and inflammation of the prepucial sheath, splenomegaly, haemorrhages on the kidney and other clinical signs that include; no mortality, mild changes in haematological and serum biochemistry, variable gross and cytopathological changes in some organs and apparent spontaneous recovery without chemotherapy. Infection in pigs with the parasite seems to be self limiting and hence does not pose a threat to pig production and health status.

Citation:

1. Introduction

The pathology of trypanosomiasis has been reviewed (Fiennes, 1970; Murray, 1974). Proliferative changes in the lymphoid organs of *T. vivax* (Masake and Morrison, 1981) and *T. brucei* (Moulton and Sollod, 1976) infected cattle have been reported with little information as regards to infection with *T. congolense*. Trypanosomiasis is characterized by chronic debilitation. In the advanced form, gross lesions, anaemia, oedema of subcutaneous tissues and lymphoid organs and serous atrophy of fat are commonly observed (Fiennes, 1970; Biryomumaisho et al., 2003). Subcutaneous oedema is particularly prominent. The liver may be enlarged and oedema of lymph nodes is often seen in the acute disease condition but they may be reduced in size in the chronic disease state. The spleen and lymph nodes may be swollen, normal or atrophic in the course of the disease. Moulton (1986) found accumulation of plasma cells and lymphocytes in medullary sinuses and cords of lymph nodes as well as haemolymph nodes of the spleen of *T. congolense* infected cattle. Necrosis of the kidneys and heart muscle and sub serous petechial haemorrhages commonly occur. Gastroenteritis is common and focal polioence-phalomalacia may be seen and this probably results from ischaemia (occlusion of
arteries) due to enormous build-up of the parasites in the terminal capillaries of the brain. A remarkable lesion is anaemia but with cellular invasion of various organs, inflammation, degeneration and necrosis become pronounced. Most body tissues show marked proliferative changes (Moulton, 1986). This study reports on the histological findings of the liver and kidney of rats infected with *T. congolense*.

1.1 Objective of research
To evaluate the pathogenicity of *T. congolense* in experimentally infected indigenous pigs, monitor the clinical signs, to evaluate blood chemistry and carry out post mortem on experimental animals.

1.2 Justification of study
Trypanosomosis is an important protozoan disease causing a major constraint in livestock production in sub-Saharan Africa. The impact of *T. congolense* on reproduction is immense causing severe effect on spermiogenesis and sperm quality in male. Trypanosomosis is still a major limiting factor to large-scale and profitable livestock production in the tsetse-infested areas of Africa. This is not only due to apparent clinical disease and mortality but also to unapparent production losses, especially in commercial farming systems. *T. congolense* has been listed among top animal parasites capable of mutation with all domestic animals including pigs at high possibility of being infected. A strain of *T. congolense* that exhibited different virulence and pathogenicity in animals has been reported.

2. Experiment

2.1 Materials and Methods
Fifteen land race pigs (3 males and 12 females) aged between eight and twenty weeks old purchased from two piggeries in Zaria. They were housed in clean, fly-proofed pens at the Faculty of Veterinary Medicine, Ahmadu Bello University Zaria, Nigeria. The pigs were acclimatized for 4 weeks prior to infection. During the acclimatization period, they were ear-tagged for identification and screened for endoparasites, ectoparasites, and hemoparasites and then were treated appropriately against helminths and ectoparasites using Ivermectin (Ivomec®) at the dose rate of 200 µg/kg body weight given sub-cutaneously. They were also intramuscularly injected with oxytetracycline® at 200 mg/kg body weight against possible bacterial infections. Within this period of acclimatization also, their baseline (hematological, body temperature, body weight) data were obtained for a period of 2 weeks.

The pigs were fed on wheat offals, milled, maize, and groundnut cake, with water provided *ad libitum* throughout the study period.

2.2 Trypanosomacongolense
The field *T. congolense* stock used in this study was obtained from the National Institute for Trypanosomiasis and Ochocerciasis Research (NITOR) Vom, Nigeria. It was inoculated intraperitoneally into five albino rats and transported to the Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria, Nigeria.

2.3 Experimental Design
The 15 pigs were randomly distributed into two groups designated A and B. Group A was assigned three pigs which were uninfected with *T. congolense* and served as control group while Group B was assigned twelve pigs to be infected with *T. congolense* organism.

2.4 Inoculation of Animals
The detection and determination of the level of parasitemia in the rats were carried out using the method described by WOO (1969). The blood of the infected rats was collected when parasitemia was +3 (30-40 parasites per microscopic field of view) and then pooled into sterile bottles containing ethylene diamine tetra.acetic acid (EDTA) as anticoagulant and was used to inoculate the pigs in Group B. Each of the 12 pigs was inoculated with 2 ml of the infected rat blood containing 1.8x10^6 parasites via the anterior vena cava.

2.5 Evaluation of Gross and Histopathological Lesions
Post mortem examinations were conducted on 10 sacrificed pigs (all pigs in the control group and 7 infected pigs). All the major organs (heart, liver, kidney, lungs, spleen, brain, testicles, ovaries and gastro-intestinal tract) were examined and gross lesions were identified and recorded. Impression smears were prepared from the lymph nodes, brain, lungs, heart, ovaries, testis and kidneys of each necropsied pig and stained with Giemsa as described by (Luntrypanosomes and other cytopathological findings).

Samples of the same organs were fixed in 10% formalin solution and later transferred to buffered formalin after 24 hours. These were later processed into slides first by initially
embedding them in paraffin wax and then sectioned into 6 µm thickness. The slides were stained with Haematoxylin and Eosin stain (H&E) as described by (Luna et al., 1960) and examined under the light microscope at x100 magnification with oil immersion for histopathological changes.

3. Results

Below: The carcass of a *T. congolense* uninfected (Figure 1) and infected (Figure 2) pig showing normal coloration and paleness of organs respectively.

**Figure 1:** (Uninfected)

**Figure 2:** (Infected)

Figure 3: Enlargement of spleen (top) in *T. congolense* infected pig and normal spleen (bottom) in a non-infected control pig.

Below: Haemorrhagic kidneys (arrows) from a *T. congolense* infected pig (Figure 4) and normal kidney in a control group (Figure 5).

**Figure 4:** Infected

**Figure 5:** Uninfected
Below: Photomicrograph of the spleen from uninfected pig (Figure 6) showing normal splenic architecture and distribution of lymphocytes and infected pig (Figure 7) showing congestion and depletion of lymphocytes (arrow) (H&E x40 magnification).

**Figure 6: Uninfected pig**

**Figure 7: Infected pig**

Below: Photomicrograph of the histopathological section of the liver of uninfected pig (Figure 10), Note the normal liver architecture devoid of mononuclear cell infiltrations and infected pig (Figure 11) Note increased mononuclear cell infiltration at the perilobular vascular areas (light arrow) of the liver. There is also a widening of the lobular boundary (thicker arrow) H&E stain, x100 magnification

**Figure 10: Uninfected pig plate**

**Figure 11: Infected pig Plate**

Below: Photomicrograph of a histopathological section of the testis in un-infected pig showing normal testicular architecture (Figure 8) and infected pig (Figure 9) Note some seminiferous tubules are devoid of spermatids (light arrow) while others contain spermatids (thicker arrow). H&E stain, x100 magnification.

**Figure 8: Uninfected pig**

**Figure 9: Infected plate pig**

Below: Photomicrograph of the histologic section of the lung of a un-infected (Figure 12) pig showing normal architecture and *T. congolense* infected (Figure 13) pig showing
mononuclear cell infiltrates (arrow) H&E stain x100 magnification.

**Figure 12:** Uninfected pig

**Figure 13:** Infected pig plate

4. Discussion

4.1 Gross and histopathological lesions

There was no mortality recorded during the course of the experiment. However, groups representatives were salvaged and examined for any gross pathologic lesions. The hair-coat of the infected pigs was ruffled; they were slightly emaciated, and the carcass were pale (Figure 1 and 2). Other changes observed in the infected pigs, which did not occur in the control pigs included splenomegally (Figure 3), slightly enlarged and hemorrhagic kidneys (Figure 4 and 5), lymphadenitis, especially involving the mesenteric lymph nodes. Variable histopathological changes were observed in organs of the infected pigs. The spleen of the infected pigs showed depletion of lymphocytes at the germinal centers (Figure 6 and 7), some seminiferous tubules of the testis were devoid of spermatids (Figure 8 and 9) liver (Figure 10 and 11) and lungs (Figure 12 and 13) showed massive mononuclear cell infiltrations with lobular boundaries wider than normal.

The NITOR stock of *Trypanosoma congolense* used in this study resulted in mild clinical trypanosomosis in the infected pigs, with no mortality recorded during the course of the experiment. The mean rectal temperature was slightly higher in the infected pigs throughout the period of the study. Increase in temperature (pyrexia) observed in trypanosomosis in this study were consistent with the findings of (Akpa et al., 2008; Ezeokonkwo et al., 2012). Pyrexia in trypanosome infection has been reported to arise from metabolism of tryptophan to tryptophol by trypanosomes (Taylor and Authie, 2004; Takeet and Fagbemi, 2009). The tryptophol tends to exert its effects on the thermoregulatory center of the hypothalamus so that the thermostatic level of the body is raised. The accumulation of tryptophol in pharmacological doses has been reported by (Tizard et al., 1978) in trypanosome infected animals. Fluctuation in the level of parasitaemia was observed throughout the study, a finding which is consistent with the report of (Allam et al., 2011) in *T. brucei* infected pigs. Anemia which was characterized by a drop in PCV values was observed during the course of this study and agrees with the reports of (Anosa, 1988; Allam et al., 2011) in *T. vivax, T. congolense* and *T. brucei* infection in gilts, respectively.

The mild infection didn’t have any significant effect on the weight of the infected pigs. This is in conformity with the reports of (Ilemobade and Balogun, 1981; Deckers, 2004) of mild disease due to *T. Congolense* infection in pigs.

The histopathological changes found in the liver were in agreement with previous reports on trypanosome infection in various animals. The major changes in the liver of the infected pigs were increased mononuclear cell infiltration and relative widening of the lobular boundary, similar report made on *T. congolense* infection in pigs (Mohammed, 1992).

The depletion of lymphocytes observed in the germinal centres of the spleen in the pigs could be due to the body requirement of this cell to combat the parasites in circulating blood and this agrees with the findings of (Chaudhari and Iqbal, 2000) in camels infected with *T. evansi*.

Some of the seminiferous tubules of the testis in the infected pigs were devoid of spermatids, a finding also reported by (Omeke et al., 2000), who explained that trypanosomes primarily exert effects on the tender germ cells involved in meiosis and maturation of sperm and spermatogenesis thereby causing the affected tubules to be devoid of spermatids.
The observed mononuclear cell infiltrations in the lungs of the infected pigs, agrees with the findings of (Uche and Jones, 1992; Ngererawa et al., 1993; Damayanti et al., 1994; Debjani et al., 2001).

**Conclusion**

The results of these gross and histological findings indicate that there is severe organ involvement, massive macrophage and lymphocyte proliferation thus disrupting the structural integrity of the cells of these organs. The observation revealed continuous massive proliferation of macrophages and lymphocytes.

**Research Highlights**

Experimental *T. congolense* infection in pigs produces mild clinical signs, no mortality was recorded, and disease is self-limiting even though infected animals were parasitaemic at certain stage of experiment. Gross and histological findings indicate that there is severe organ involvement, massive macrophage and lymphocyte proliferation thus disrupting the structural integrity of the cells of these organs. The observation revealed continuous massive proliferation of macrophages and lymphocytes.

Highlight the main points of your research

The self-cure phenomenon is a possibility in *T. congolense* infection in pigs.

Not all infected pigs became parasitaemic during the period of experiment.

Gross and histopathological changes are associated with organs in *T. congolense* infection in pigs.

Reproductive organ involvement in male was observed in *T. congolense* infection in pigs.

**Limitations**

Other parameters would have been analyzed if not for limited fund, and the time duration for the work would have been extended if there was enough fund to manage and sustain the animals.

**Funding and Policy Aspects**

The research is self funded.

**Author’s Contribution and Competing Interests**

We declare there is no competing interest.

**Acknowledgement**

I wish to sincerely acknowledge the technical help rendered by staff of the Pathology Department, the Head Pathology Department of Ahmadu Bello University (ABU) Zaria who read my slides and interpreted same. My sincere gratitude goes to Maniru the animal attendant and Kate Adeyani, Mallam Lawal and Hussaini all of the Department of Veterinary Pathology and Entomology ABU, Zaria for their technical aid.

**References**


Akpa P.O., Ezeokonkwo R.C., Eze C.A., Anene BM., 2008. Comparative efficacy assessment of pentamidine isomethamidium and diminazene aceturate in the chemotherapy of *T. brucei* infection in dogs. Veterinary Parasitology, (151), 139-149.


