Effects of dexamethasone on the infectivity of *Trypanosoma vivax* Y486 and the haematological changes in Nigerian domestic chickens (*Gallus gallus domesticus*)

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**ABSTRACT**

The study was carried out to investigate how the infectivity of *Trypanosoma vivax* Y486 could be affected by steroid administration to Nigerian domestic chickens and to observe the haematological and serum biochemistry changes in these chickens infected with *Trypanosoma vivax* Y486. Results showed that *T. vivax* Y486 does affect the haematological parameters of Nigerian domestic chickens (*Gallus gallus domesticus*) as there may be development of anaemia as in large animals. However, there was no patent parasitaemia. In addition, steroid administration when applied at high doses did affect the haematological parameters of the local chickens. It thus showed that serial passaging of *T. vivax* Y486 can result in infectivity of the Nigerian domestic chickens with trypanosomosis.

**Key words:** trypanosomosis, *Trypanosoma vivax* Y486, dexamethasone, haematology, *Gallus gallus domesticus*

**Introduction**

*Trypanosoma congolense*, *T. vivax*, *T. simiae* and *T. brucei* are among the major trypanosomes affecting livestock in Nigeria. Economic losses resulting from stunted growth, debility, poor reproductive performance or death in affected animals...
are unquantifiable (LOSOS and IKEDE, 1972; HENSON and NOEL, 1979; ANOSA, 1988; AGYEMANG, 1989; JAWARA, 1990; KALU et al., 1991; KALEJAIYE et al., 1995; KALU and LAWANI, 1996; KALU et al., 1996; ONYIAH, 1997; ENWEZOR and LAWAL, 2003).

One approach to the control or eradication of trypanosomosis is immunology. A major obstacle is the ability of the trypanosome to undergo antigenic variation wherein the organism develops different antigens on their surface. It thus results in the host not being able to destroy organisms with antigenic variation (VICKERMAN and LUCKINS, 1969; SEED, 1974; VICKERMAN, 1978; GRAY, 1985; MURRAY et al., 1982; MURRAY et al., 1984; PINDER et al., 1988; KAMANGA-SOLLO et al., 1991; EZEOKONKWO et al., 2003).

Another approach to the study of this disease is its study in abnormal hosts (DESOWITZ, 1963; GOEDBLOOD, 1972; JOSHUA, 1979). Nigerian domestic chickens (Gallus gallus domesticus) infected with a stock of Trypanosoma brucei exhibited a chronic infection that was spontaneously terminated by self cure. Local chickens were then found to be immune to challenge with derivatives of the same trypanosome stock. It was suggested that the ability of the birds to cure the infection and retain immunity might be due to the particularly efficient nature of their immune responses. These results reaffirm the view that it may be possible to design effective trypanosome vaccines for use in animals (JOSHUA, 1983).

DINA and AROWOLO (1988) also carried out a study involving inoculation of local chickens with T. brucei brucei 8/18 and T. vivax Y58. These workers showed that the chickens did not develop a clinical disease but rather they all continued to feed and increase in body weight. The study also revealed that chickens infected with both trypanosomes did not show parasitaemia for 4 weeks post-inoculation.

This study hopes to evaluate the infectivity of Trypanosoma vivax Y486 in Nigerian domestic chickens using dexamethasone to suppress cell-mediated immunity. The blood profiles of local chickens will also be examined to perceive the effects of both the trypanosomes and dexamethasone on the haematological parameters of Nigerian local chickens.

**Materials and methods**

*Experimental animals.* Forty-four local chickens aged 10-14 weeks were bought from Molete market, Ibadan, Nigeria and were housed for 5 weeks at the experimental animal house of the Faculty of Veterinary Medicine, University of
Ibadan, Nigeria. During this period the animals were given antibiotic (neoterramycin) and anthelmintic (piperazine) treatment for 3-8 days.

Forty-four albino mice were also purchased for xeno-diagnosis of trypanosomosis. Infected blood containing \( T. vivax \) Y486 was collected from parasitaemic mice.

A strain of \( Trypanosoma vivax \) Y486 was obtained from the experimental animal house of the Faculty of Veterinary Medicine, University of Ibadan, where it has been passaged in laboratory mice and rats. The strain was first isolated in Yakewada, Nigeria on November 15, 1973 but was actually acquired from International Laboratory for Research on Animal Diseases (ILRAD) from where it was brought to the University of Ibadan, Nigeria.

Drugs. 10 ampoules of 4 mg/ml of Dexamethasone were used in the course of this study and were given through the intramuscular route.

Piperazine, an anthelmintic and neoterramycin (antibiotic) was given separately to local chickens during the period of acclimatization.

Experimental design. Four experiments were carried out in the course of this study. Prior to the experiments all the chickens were bled in order to determine basal haematological and biochemistry values.

Experiment I. Twelve local chickens were divided into 3 groups (A, B and C) of 4 birds per group. Dexamethasone (0.2 mg/ml) was administered for 5 days to the animals in group B through the intramuscular route. On day 6, approximately \( 2.5 \times 10^4 \) trypanosomes were inoculated into each chicken in groups A and B through the intraperitoneal route. Group C served as control experiment.

From day 6-14 post-inoculation (p.i.) of trypanosomes, blood was collected from the wing veins of all the birds and examined for parasitaemia. Also, at day six, p.i. of the chickens with the trypanosomes, the blood samples collected from all groups of chickens were administered intraperitoneally into the laboratory mice. The blood of the laboratory mice was in turn examined for parasitaemia 3-6 days P.I. with chicken blood. Post-treatment blood samples were collected from all the chickens at the end of the experiment.

Experiment II. The procedure was similar to that of experiment I except that:

a) 0.4 mg/ml of dexamethasone was administered with group B for 7 days.
b) \( 5.0 \times 10^4 \) trypanosomes were inoculated into all the chickens in groups A and B.
Experiment III. The procedure was also similar to the preceding experiments except that:
a) 1 mg/ml of dexamethasone was administered for 10 days to birds in group B, after which blood samples were collected from this group to assess the effect of dexamethasone on the haematology and serum biochemistry of the birds.
b) Appropriately 5.5×10⁵ trypanosomes were inoculated into chickens in groups A and B.

Experiment IV. Eight chickens were used in this experiment. Approximately 1×10⁶ trypanosomes were inoculated into all the animals through the jugular vein. From day 7-20 p.i. of the birds, blood was collected through the wing vein and examined for parasitaemia. Also at days 7, 14 and 20, p.i. of the chickens, the blood samples were collected and administered into mice; these were in turn examined for parasitaemia 3-6 days p.i. with chicken blood.

Determination of haematological parameters. Red blood cell counts (RBC), packed cell volume (PCV), haemoglobin (Hb) concentration, white blood cell (WBC) counts and the differential leukocyte count were done by the standard procedure described by Jain (1986). From the values of the PCV, Hb and RBC counts, the mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and the mean corpuscular haemoglobin concentration (MCHC) were estimated.

Statistical analysis. The levels of significant differences between the mean values of the treated and control stages were determined using the Student’s t-test at P<0.05 (Steel and Torrie, 1982).

Results
The results of this study showed that the blood samples of all chickens and laboratory mice revealed no parasitaemia. Even when the dosage of dexamethasone was increased and the duration of administration prolonged no parasitaemia was revealed. In fact, the increase in the concentration of trypanosomes with each experiment still did not reveal any parasitaemia, clinical or sub-clinical disease in the chickens or in the laboratory mice.

The results of this study with respect to the effects of T. vivax Y486 on the haematological parameters of Nigerian local chickens showed that the levels of PCV, Hb, RBC, and eosinophils in the 4 experimental groups experienced
significant reduction (P<0.05) when compared with the control. MCV and MCH experienced a significant increase (P<0.05) which monocytes experienced significant decrease. WBC only experienced significant reduction in experiment IV when compared with the control (Table 1).

Table 1. Effects of T. vivax Y486 on the haematological parameters of Nigerian local chickens

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Treatments</th>
<th>Preinfection</th>
<th>Control</th>
<th>Expt. 1</th>
<th>Expt. 2</th>
<th>Expt. 3</th>
<th>Expt. 4</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCV (%)</td>
<td></td>
<td>26.4 ± 1.2</td>
<td>29.9 ± 1.4</td>
<td>24.4 ± 3.8 *</td>
<td>25.8 ± 1.3 *</td>
<td>23.1 ± 1.6 *</td>
<td>23.0 ± 1.5 *</td>
</tr>
<tr>
<td>Hb (g/l)</td>
<td></td>
<td>7.5 ± 0.3</td>
<td>8.6 ± 0.4</td>
<td>7.0 ± 1.1 *</td>
<td>6.8 ± 1.2 *</td>
<td>6.7 ± 0.9 *</td>
<td>6.9 ± 1.1 *</td>
</tr>
<tr>
<td>RBC (×10^6/µl)</td>
<td></td>
<td>1.5 ± 0.2</td>
<td>3.4 ± 0.7</td>
<td>2.3 ± 0.5 *</td>
<td>2.2 ± 0.2 *</td>
<td>2.1 ± 1.1 *</td>
<td>2.4 ± 0.6 *</td>
</tr>
<tr>
<td>MCH (fl)</td>
<td></td>
<td>176 ± 8.9</td>
<td>106.1 ± 21</td>
<td>108.2 ± 6.5 *</td>
<td>110 ± 11.1</td>
<td>95.8 ± 9.1</td>
<td></td>
</tr>
<tr>
<td>MCHC (%)</td>
<td></td>
<td>28.4 ± 0.1</td>
<td>28.8 ± 0.2</td>
<td>28.7 ± 3.6</td>
<td>28.6 ± 2.5</td>
<td>29 ± 1.5</td>
<td>30 ± 2.3</td>
</tr>
<tr>
<td>MCHC (pg)</td>
<td></td>
<td>50 ± 2.5</td>
<td>25.3 ± 4.2</td>
<td>30.4 ± 6.1</td>
<td>30.9 ± 2.1 *</td>
<td>31.9 ± 1.8 *</td>
<td>28.8 ± 1.5</td>
</tr>
<tr>
<td>WBC (×10^3/ml)</td>
<td></td>
<td>30.2 ± 3.4</td>
<td>26.4 ± 1.7</td>
<td>23.4 ± 6.7</td>
<td>24.1 ± 3.2</td>
<td>24.6 ± 1.7</td>
<td>23.8 ± 1.2 *</td>
</tr>
<tr>
<td>Lymphocytes (×10^3/ml)</td>
<td></td>
<td>19.9 ± 2.4</td>
<td>16.5 ± 1.3</td>
<td>16.6 ± 4.6</td>
<td>16.8 ± 2.1</td>
<td>17.1 ± 1.5</td>
<td>16.5 ± 1.6</td>
</tr>
<tr>
<td>Heterophils (×10^3/ml)</td>
<td></td>
<td>10.4 ± 1.3</td>
<td>8.5 ± 1.7</td>
<td>6.1 ± 2.1</td>
<td>6.6 ± 1.8</td>
<td>6.9 ± 0.8</td>
<td>6.5 ± 2.2</td>
</tr>
<tr>
<td>Monocytes (×10^3/ml)</td>
<td></td>
<td>0.5 ± 0.1</td>
<td>0.8 ± 0.2</td>
<td>0.6 ± 0.2</td>
<td>0.5 ± 0.2 *</td>
<td>0.4 ± 0.1 *</td>
<td>0.6 ± 0.2</td>
</tr>
<tr>
<td>Eosinophils (×10^3/ml)</td>
<td></td>
<td>0.2 ± 0.04</td>
<td>0.4 ± 0.1</td>
<td>0.1 ± 0.07</td>
<td>0.2 ± 0.1 *</td>
<td>0.2 ± 0.1 *</td>
<td>0.2 ± 0.1 *</td>
</tr>
</tbody>
</table>

Note: The experiments involved group A animals. The number of animals for experiment IV was 8, i.e. n = 8. The superscripts indicate significant values at P<0.05.

The results of this study also showed that dexamethasone caused a significant reduction in the levels of PCV, RBC, MCV, MCHC and MCH when compared with the control in all the experimental groups. It was in experiments I and II that dexamethasone caused a significant reduction in the levels of haemoglobin, while there was a significant reduction in the level of lymphocyte in experiment II. It was in experiments II and III that monocytes exhibited a significant reduction. Eosinophils only experienced a significant reduction in experiment III when compared with the control (Table 2).

The effects of dexamethasone and T. vivax Y486 on the haematological parameters of Nigerian local chickens showed that it was only in experiment III that PCV, Hb and RBC experienced a significant reduction when compared with the controls. While MCV experienced a significant increase in its level for experiment II, it was the reverse for MCHC in the experimental group. However, WBC experienced a significant reduction in its levels for all the experimental groups. The result was similar for eosinophils. In the case of lymphocytes, it was in experiments II and III that a significant reduction was experienced. For
monocytes, a significant reduction in level was recorded in experiments I and III (Table 2).

**Table 2. Effects of Dexamethasone on the haematological parameters of Nigerian local chickens**

(n = 4)

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Preinfection</th>
<th>Control</th>
<th>Expt. 1</th>
<th>Expt. 2</th>
<th>Expt. 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCV (%)</td>
<td>26.4 ± 1.2</td>
<td>29.9 ± 1.4</td>
<td>27.1 ± 1.5</td>
<td>26.8 ± 1.3</td>
<td>27.5 ± 2.3</td>
</tr>
<tr>
<td>Hb (g/l)</td>
<td>7.5 ± 0.3</td>
<td>8.6 ± 0.4</td>
<td>7.9 ± 0.3</td>
<td>8.2 ± 1.1</td>
<td>7.7 ± 0.7</td>
</tr>
<tr>
<td>RBC (×10⁶/µl)</td>
<td>1.5 ± 0.2</td>
<td>3.4 ± 0.7</td>
<td>1.6 ± 0.4</td>
<td>1.8 ± 0.2</td>
<td>1.9 ± 0.6</td>
</tr>
<tr>
<td>MCV (fl)</td>
<td>176 ± 8.9</td>
<td>87.9 ± 14.5</td>
<td>109.4 ± 6.2</td>
<td>189.9 ± 2.5</td>
<td>144.7 ± 28.7</td>
</tr>
<tr>
<td>MCHC (%)</td>
<td>28.4 ± 0.1</td>
<td>28.8 ± 0.2</td>
<td>29.2 ± 0.4</td>
<td>30.6 ± 0.3</td>
<td>28.0 ± 0.2</td>
</tr>
<tr>
<td>MCH (pg)</td>
<td>50 ± 2.5</td>
<td>25.3 ± 4.2</td>
<td>49.4 ± 1.3</td>
<td>45.6 ± 3.2</td>
<td>40.5 ± 7.3</td>
</tr>
<tr>
<td>WBC (×10⁹/ml)</td>
<td>30.2 ± 3.4</td>
<td>26.4 ± 1.7</td>
<td>25.1 ± 0.2</td>
<td>24.8 ± 2.1</td>
<td>23.1 ± 3.5</td>
</tr>
<tr>
<td>Lymphocytes (×10⁹/ml)</td>
<td>19.9 ± 2.4</td>
<td>16.5 ± 1.3</td>
<td>14.8 ± 2.3</td>
<td>13.2 ± 1.8</td>
<td>15.3 ± 2.6</td>
</tr>
<tr>
<td>Heterophils (×10⁹/ml)</td>
<td>10.4 ± 1.3</td>
<td>8.5 ± 1.7</td>
<td>9.2 ± 1.2</td>
<td>10.6 ± 1.4</td>
<td>7.1 ± 1.8</td>
</tr>
<tr>
<td>Monocytes (×10⁹/ml)</td>
<td>0.5 ± 0.1</td>
<td>0.8 ± 0.2</td>
<td>0.6 ± 0.1</td>
<td>0.5 ± 0.2</td>
<td>0.4 ± 0.1</td>
</tr>
<tr>
<td>Eosinophils (×10⁹/ml)</td>
<td>0.2 ± 0.04</td>
<td>0.4 ± 0.1</td>
<td>0.4 ± 0.1</td>
<td>0.5 ± 0.1</td>
<td>0.3 ± 0.01</td>
</tr>
</tbody>
</table>

Note: The experiment involved group B animals.

**Table 3. Effects of Dexamethasone and *T. vivax* Y486 on the haematological parameters of Nigerian local chickens**

(n = 4)

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Preinfection</th>
<th>Control</th>
<th>Expt. 1</th>
<th>Expt. 2</th>
<th>Expt. 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCV (%)</td>
<td>26.4 ± 1.2</td>
<td>29.9 ± 1.4</td>
<td>29.0 ± 2.7</td>
<td>28.6 ± 1.5</td>
<td>26.1 ± 2.0</td>
</tr>
<tr>
<td>Hb (g/l)</td>
<td>7.5 ± 0.3</td>
<td>8.6 ± 0.4</td>
<td>8.2 ± 0.8</td>
<td>7.9 ± 0.7</td>
<td>7.5 ± 0.5</td>
</tr>
<tr>
<td>RBC (×10⁶/µl)</td>
<td>1.5 ± 0.2</td>
<td>3.4 ± 0.7</td>
<td>2.9 ± 0.7</td>
<td>2.7 ± 0.3</td>
<td>2.6 ± 0.4</td>
</tr>
<tr>
<td>MCV (fl)</td>
<td>176 ± 8.9</td>
<td>87.9 ± 14.5</td>
<td>100 ± 23.3</td>
<td>106 ± 6.2</td>
<td>100.4 ± 3.2</td>
</tr>
<tr>
<td>MCHC (%)</td>
<td>28.4 ± 0.1</td>
<td>28.8 ± 0.2</td>
<td>28.3 ± 0.2</td>
<td>27.6 ± 0.3</td>
<td>28.7 ± 1.1</td>
</tr>
<tr>
<td>MCH (pg)</td>
<td>50 ± 2.5</td>
<td>25.3 ± 4.2</td>
<td>49.4 ± 1.3</td>
<td>45.6 ± 3.2</td>
<td>40.5 ± 7.3</td>
</tr>
<tr>
<td>WBC (×10⁹/ml)</td>
<td>30.2 ± 3.4</td>
<td>26.4 ± 1.7</td>
<td>21 ± 4.0</td>
<td>20.7 ± 1.2</td>
<td>20.4 ± 1.1</td>
</tr>
<tr>
<td>Lymphocytes (×10⁹/ml)</td>
<td>19.9 ± 2.4</td>
<td>16.5 ± 1.3</td>
<td>13.2 ± 3.3</td>
<td>12.8 ± 2.1</td>
<td>12.6 ± 2.1</td>
</tr>
<tr>
<td>Heterophils (×10⁹/ml)</td>
<td>10.4 ± 1.3</td>
<td>8.5 ± 1.7</td>
<td>6.2 ± 2.0</td>
<td>7.0 ± 1.2</td>
<td>7.1 ± 1.1</td>
</tr>
<tr>
<td>Monocytes (×10⁹/ml)</td>
<td>0.5 ± 0.1</td>
<td>0.8 ± 0.2</td>
<td>0.5 ± 0.1</td>
<td>0.7 ± 0.1</td>
<td>0.6 ± 0.1</td>
</tr>
<tr>
<td>Eosinophils (×10⁹/ml)</td>
<td>0.2 ± 0.04</td>
<td>0.4 ± 0.1</td>
<td>0.2 ± 0.01</td>
<td>0.2 ± 0.1</td>
<td>0.1 ± 0.01</td>
</tr>
</tbody>
</table>

Note: The experiment involved group B animals.
Discussion

The results of this study showed that *T. vivax* Y486 produced a significant reduction in the level of haematological parameters of Nigerian local chickens, especially considering the fact that PCV, RBC and haemoglobin concentration all experienced significant changes when compared with the control. The implication is that the reduction of these parameters may lead to anaemia, which may be functionally defined as a decreased oxygen-carrying capacity of the blood. The easiest and most accurate laboratory indication of anaemia is reduction of the PCV or haematocrit below the normal range for the species (ANOSA, 1988; DUNCAN et al., 1994). There are 3 pathophysiologic mechanisms for the development of anaemia: blood loss, increased erythrocyte destruction (haemolysis) and inadequate erythrocyte production. Since *T. vivax* exert their effect mainly by causing severe anaemia (IKEDE, 1986; ANOSA, 1988; HUNTER and LUCKINS, 1990; OGUNSANMI et al., 1994), it is probable that the pathophysiological mechanism for development of anaemia is through erythrocyte destruction (haemolysis).

It has also been shown that animals infected with *T. vivax* are immunosuppressed and frequently have intercurrent bacterial, viral or other parasitic infections that may mask or complicate the basic clinical syndrome. In addition, the immune responses to bacterial and some viral vaccines are known to be depressed in infected animals but are restored if trypanocidal therapy is given at the time of vaccination (MWANG et al., 1990). This observation may be responsible for the significant reduction in the level of WBC in experiment IV at Table 1. The monocytes level also experienced a particularly significant reduction. This may further support the immunosuppressive attributes of this organism because the blood monocytes and tissue macrophages constitute the mononuclear phagocyte system known as the reticuloendothelial system (JAIN, 1986; DUNCAN et al., 1994). The functions of tissue macrophages include sustained phagocytic activity to remove dead and damaged tissue; microbicidal action against some bacteria, viruses, fungi, and protozoa; regulation of the immune response in both afferent and efferent limbs; tumour defence; regulation of haematopoiesis; tissue repair and remodelling; and secretion of monokines, lysosomal enzymes and other substances such as coagulation factors that have widely resulting biologic importance. It should be noted that monocytes are produced in the bone marrow and differentiate into either myeloblasts or monoblasts. Monocytes once released into blood transform into macrophages when they enter into body cavities and

*A. Adedapo et al.: Effects of dexamethasone on T. vivax and haematological changes in chickens*
tissues (AKINBAMIJO et al., 1998). The reduction in the monocytes level recorded in this study showed that Nigerian local chickens are susceptible to trypanosomal infection and all the functions of monocytes enumerated above will therefore be compromised. Eosinophils level was also significantly reduced in this study. In fact, all experimental groups experienced this. Eosinophils, while having a role as phagocytes, also have more specific functions that include providing a defence against metazoan parasites and modulating the inflammatory process. Eosinophils also respond chemotactically to histamine, immune complexes, and eosinophil chemotactic factor of anaphylaxis, a substance released by degranulating mast cells (OGUNSANMI et al., 1994). The study thus showed that *T. vivax* infection in local chickens adversely affects the functions of eosinophils as enumerated above.

Dexamethasone, a glucocorticoid, inhibits the release of inflammatory mediators from macrophages and eosinophils but do not inhibit the release of granules from mast cells. Glucocorticoids also decrease synthesis of prostaglandins, leukotrienes, and platelet-activating factor, which play important roles in the pathophysiology of respiratory tract diseases. Glucocorticoids are also known to have immunosuppressive effects and hence are generally avoided in infectious respiratory diseases (DOWLING, 1998). Glucocorticoids also suppress both inflammatory and immunological responses and thereby attenuate associated tissue destruction and fibroplasia. Inhibition of a number of lymphocyte functions forms part of the basis for immunosuppression (OGUNSANMI et al., 1994). In this study, the lymphocyte levels of local chickens in experiment II were significantly reduced. Monocytes and eosinophils levels were also significantly lowered. Further, it is interesting to note that haematological parameters such as PCV, Hb, and RBC were also significantly reduced in this study. This means that dexamethasone is able to precipitate anaemia in local chickens.

Administration of both dexamethasone and *T. vivax* Y486 produced a significant reduction in the levels of white blood cells, lymphocytes, monocytes and eosinophils. This therefore showed that the combined administration of the glucocorticoid and the protozoan produced immunosuppression in local chickens as already described. Experiment III animals also experienced a significant reduction in haematological parameters, such as PCV, Hb, and RBC. It is interesting to note that these changes only occurred when the glucocorticoid was administered at a high dosage rate and when the protozoan was also inoculated at high concentration.
References


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SAŽETAK

Ključne riječi: tripanosomoza, Trypanosoma vivax Y486, deksametazon, hematologija, Gallus gallus domesticus