Global Veterinaria 13 (3): 378-384, 2014
ISSN 1992-6197

DOI: 10.5829/idosi.gv.2014.13.03.84303

Comparative Haematological Changes in West African Dwarf Goats Experimentally Infected with *Trypanosoma vivax and Trypanosoma brucei*: Changes Caused by Treatment with DiminazeneAceturate (Diminaze®)

[1]U.I. Osuagwuh and [2]O.O. Okore

[1]Department of Veterinary Surgery and Theriogenology, College of Veterinary Medicine
[2]Department of Zoology and Environmental Biology, College of Natural Science,
Michael Okpara University of Agriculture, Umudike, Nigeria

Abstract: A comparative study of haematological changes in West African Dwarf bucks experimentally infected with *Trypanosoma brucei* and *Trypanosoma vivax* and changes caused after treatment with (Diminaze®) was carried out using twenty three (23) bucks for a period of 18 weeks. Eight bucks were infected with *T. brucei,* another eight with *T. vivax* and seven kept as control. They were divided into three groups and housed in separate fly proof pens. All animals were fed commercial concentrate at 12-14% crude protein, fresh grasses, water and salt licks*ad-libitum*. Data on parasitaemia, rectal temperature, weight and haematological investigations were determined before infection, during infection and after treatment. *T. brucei*infected bucks developed chronic trypanosomiasis while *T. vivax* infected bucks developed an acute phase of trypanosomiasis. Clinical disease was characterised by intermittent pyrexia, slight drop in body condition and irregular parasitaemia and these was more severe in the *T. vivax* infected bucks.There was a significant difference ($P<0.05$) in the mean values of PCV, Hb concentration, RBC, MCH and MCV but a non significant difference ($P>0.05$) in MCHC post infection (PI) in all infected bucks when compared with the control bucks. However, treatment with (Diminaze®) led to a remarkable improvement with all parameters investigated. The results of this investigation showed that *T. vivax* was more pathogenic than *T. brucei* and caused more severe pyrexia, parasitaemia and anaemia (As indicated by a greater decline of PCV, Hb concentration and RBC counts). Treatment with Diminaze® improved blood components and restored cellular functions to the pre-infection state by reversing the negative effect of infecting trypanosomes.

Key words: *Trypanosoma brucei* · *T. vivax* · Treatment · Bucks

INTRODUCTION

Several reports in ruminants have demonstrated that trypanosome infection induce various changes in host metabolic system [1-3]. The severity of infection is variable and depends on many factors; virulence of different species of trypanosomes, age, nutritional status and breed [2, 4, 5]. These changes have been associated with the level of parasite in the blood [2, 6, 7] and tissue/organ damage [8, 9].

However, these changes alongside the need by the host to destroy the parasite are apparently responsible for the symptoms of trypanosomiasis. A report on Savannah brown goats experimentally infected with *Trypanosoma brucei and Trypanosoma vivax*showed acute form of trypanosomiasis which was associated with high rectal temperature, decrease weight, anemia and hypoproteinmia and that the severity of the disease was more in *T. brucei*infected goats [10]. Nevertheless, the use of anti-trypanosomal drugs has been the most widely practiced means of controlling the disease in livestock, either as curative or prophylactic drugs.

This study was specifically designed to compare the severity of some of the haematological changes in West African Dwarf (WAD) bucks infected with *T. vivax* and *T. brucei*. The West African Dwarf goat is indigenous to Western Africa and its importance in livestock agriculture of this region is underscored by the continual focus on

Corresponding Author: U.I. Osuagwuh, Department of Veterinary Surgery and Theriogenology College of Veterinary Medicine, Michael Okpara University of Agriculture, Umudike, Nigeria.

trypanosomiasis in cattle thus neglecting these goats maybe due to the conception that WAD goats are trypanotolerant.

MATERIALS AND METHODS

Animals and Feeding: A total of 23 adult ($1^{1/2}$ and 2 year old) apparently healthy WAD goats, weighing between 8 and 12kg were used in this study. The goats were purchased from two different goat markets in Imo State and Abia State respectively, Nigeria. They were housed in various pens at the College of Veterinary Medicine, Michael Okpara University of Agriculture, Umudike for a 21 days for acclimatisation. During this period, clinical examination and screening of haemoparasites was carried out on all animals. Furthermore, all the animals were treated intramuscularly with Diaminazeneaceturate at 3.5mg/kg body weight, vaccinated against PPR (Obtained from Nigeria Veterinary Research Institute,Vom) and oxytetracyclin L.A (Global organies Ltd) at 20mg/kg. A broad spectrum anthelmintics; ivermectin (HebeiYanuzhengPharmaceuticals co. Ltd) at a dose of 20mg/kg body weight was also administered subcutaneously. Body weight was recorded weekly. Animals were later transferred to the experimental building with fly proof pens were they were randomly selected into threeexperimental groups (A, B and C) containing eight, eight and seven bucks respectively. Each group kept in three separate pens for a period of 112days. All animals were fed commercial concentrate at 12-14% crude protein. Fresh grasses, water and salt licks were also provided *ad-libitum.*

Parasites: The parasite *T. brucei* (Federe CT/28 strain) was obtained from the National Institute for Trypanosomosis Research Vom, Plateau State, Nigeria and inoculated into mice for multiplication and maintained until required. *T.vivax* (Field strain) was obtained from a local abattoir and inoculated into a goat for multiplication and also maintained until required. Parasitaemia was estimated according to the method described by Murray *et al.* [11]

Infection of the Animals: The animals were conditioned for 2 weeks during which they were examined clinically as well as for the presence of trypanosomes after which, they were infected. To infect the designated bucks (day 0), blood was obtained from the infected mice and goat and diluted in a phosphate buffer solution and 2ml of the latter containing 10^4 parasites was then injected through the jugular vein. Group A buckswas infected with *T.brucei*, whileGroup B bucks with *T.vivax.* Group C bucks were kept as control.

Clinical Examination and Sample Collection: Clinical examination began 2wks before infection and continued throughout the experiment in all the bucks. Weight and rectal temperature were taken daily using electronic weighing balance and digital thermometer respectively. Blood samples were collected from all bucks on weekly basis throughout the experiment through the jugular vein with vaccutainer tubes containing EDTA. Samples were labelled legibly and transported to the laboratory for analysis.

Treatment: All Infected bucks were treated intramuscularly with Diaminazeneaceturate (Diminaze®) at 3.5mg/kg body weight. Group A bucks were treated at day 70PI while Group B at day 28 PI. Treatment decision was carried out based on the mean level of the packed cell volume (14%) of the various groups of infected animals in other to avoid death as described by Anosaand Isoun. [12]. Clinical and haematological parameters were continually monitored for six weeks post treatment to ascertain the pattern of recovery.

Parasitology and Haematology: Presence of trypanosome was done by the buffy coat method [11]. The packed cell volume (PCV) was determined by the standard microhaematocrit method described bySchalm *et al.* [13], the red blood cell (RBC) by the haemocytometer method and haemoglobin (Hb) concentration by the cyanomethaemoglobin method. Mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCHC) and mean corpuscular haemoglobin concentrations (MCHC) were calculated based on the method [13].

Statistical Analysis: Data were subjected to Student't' test using the Statistical Analysis System software package (SAS) [14]. Data are presented as mean±SE. The level of significance was detected at $P<0.05$.

RESULT

All the bucks infected with *T. brucei* and *T. vivax*developed chronic trypanosomiasis. Their blood samples were all positive for trypanomsomes within 8 days of infection. Mean parasitaemia ranged between 0 and 3 in the two groups of bucks infected with either

trypanosome parasite. Parasitaemia was higher and more chronic in the *T.vivax* infected bucks. No trypanosome parasites were found in the blood of the control bucks. As the level of parasitaemiain the blood rose, there was a steady rise in rectal temperature in the first week PI. The mean rectal temperature of bucks infected with *T.brucei*and *T.vivax* was 40°C and 41.5°C respectively, 14 days PI. The control bucks had normal rectal temperature throughout the duration of the experiment. The pyrexia fluctuated daily during the period of infection. The clinical sign observed includes the following: fluctuating parasitaemia, weight loss, weakness, lethargy, rough hair coats, pale mucous membrane, diarrhoea and alopecia in some goats. The severity of the clinical disease was pronounced in the *T.vivax* infected bucks especially the rapid decrease in weight observed at day 21 PI. No deaths was recorded throughout the period of infection. However, treatment with Diminaze®) showed remarkably improvement within 7 days post treatment.

The mean haematological values and changes of all infected bucks are shown in figures 1-6. All baseline data was recorded within 14 days pre-infection, experimental infection was done on day 0 and treatment on day 70 for the *T. brucei* group while the *T. vivax*group was done on day 28. However, day of treatment for the *T. vivax*group was done earlier than the *T.brucei*group due to the rapid health deterioration of the bucks infected with *T. vivax* and mean percentage PCV level with minimum level kept at 14% in other to avoid death.There was a decrease in the PCV level of all infected bucks, which was more severe and rapid in the *T. vivax*infected bucks. The lowest PCV recorded in the *T. vivax*group occurred in three bucks with a PCV of 12% at day 21 PI, while the lowest PCV recorded in the *T. tbrucei*infected group occurred in two bucks with a PCV of 16% at day 56 PI. The mean PCV values for *T. bucei*-infected, *T. vivax*-infected and control bucks ranged between 17 and 24, 13 and 18 and 23 and 30%, respectively. The mean PCV values of all infected bucks deferred significantly ($P<0.05$) from the control bucks throughout the experiment. The *T. vivax*-infected group had the lowest mean PCV values, which differed significantly ($P < 0.05$) from those of the *T. brucei* infected group (Figure 1).

There was a decrease in the Hb concentration of all infected bucks which corresponded with the significant increase in the population of the trypanosome parasites in the blood. The lowest recorded values amongst the bucks infected with *T. brucei*and *T. vivax* were 27.4 and 43.7% respectively. The mean values for *T. brucei*, *T. vivax* infected and control bucks ranged between 46 and 54, 30 and 40 and 52 and 61% respectively. Overall mean values were 51, 38 and 58% respectively. The decrease in Hb concentration was more severe in the *T. vivax*infected bucks than the *T. brucei*infected group. Mean Hb concentration values of all infected bucks deferred significantly ($P<0.05$) from the control bucks and *T. vivax*-infected group had the lowest mean Hb concentration values, which differed significantly ($P < 0.05$) from those of the *T. brucei* infected group (Figure 2).

The changes in the RBC count values of infected and control bucks are shown in figure 3. With the onset of parasitaemia, all infected bucks developed anaemia with a drop in the mean value of the RBC count. The severity was observed in the *T. vivax* infected group in which the lowest value was recorded at 2.6 in one buck 14 days PI. The overall mean values in the *T. brucei, T. vivax*infected and control bucks were 4.4, 3.5 and 5.5 PI, respectively (Figure 3). All mean values of infected buck differed significantly ($P < 0.05$) from the control. There was also a significant difference ($P < 0.05$) between the *T. brucei* and *T. vivax*infected groups.

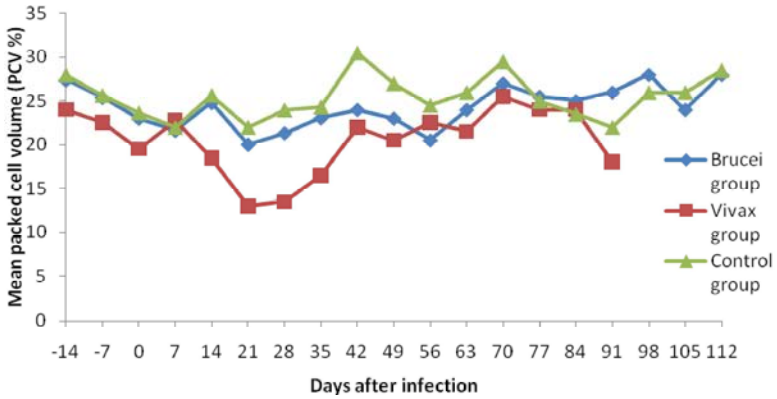

Fig. 1: Mean PCV (%) levels in of *T. brucei* (Day 0-70 post infection and day 70-112 post treatment), *T. vivax* (Day 0-28 post infection and day 28-112 post treatment) and control bucks

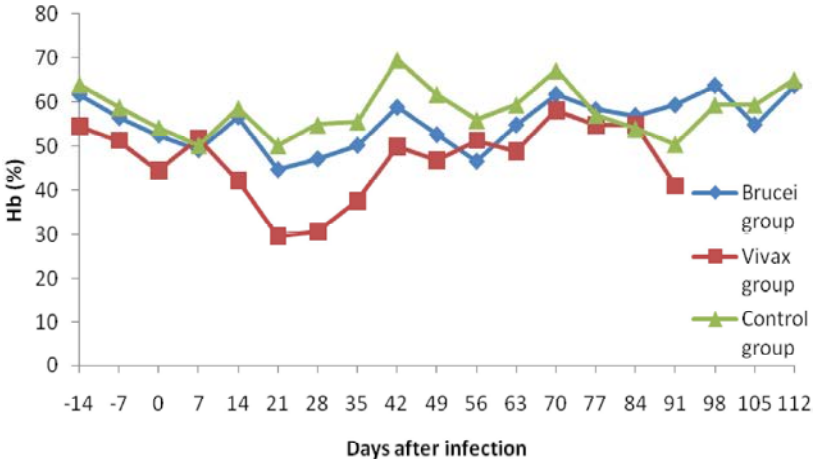

Fig. 2: Mean Hb (g%) in *T. brucei* (Day 0-70 post infection and day 70-112 post treatment), *T. vivax* (Day 0-28 post infection and day 28-112 post treatment) and control bucks

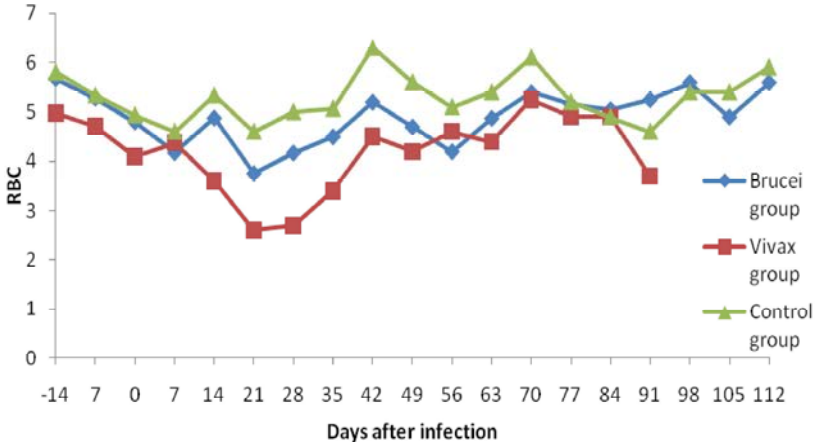

Fig. 3: Mean RBC ($_{x}10^{6}$/µl) counts level in *T. brucei* (Day 0-70 post infection and day 70-112 post treatment), *T. vivax* (Day 0-28 post infection and day 28-112 post treatment) and control bucks

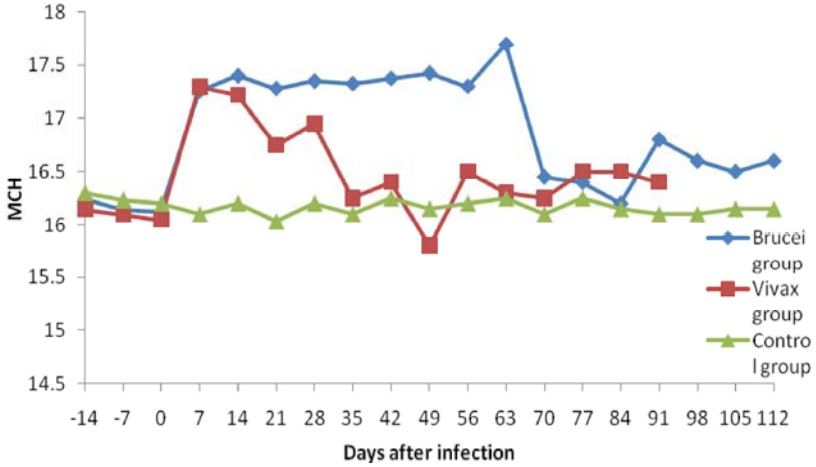

Fig. 4: Mean MCH (pg) in *T. brucei* (Day 0-70 post infection and day 70-112 post treatment), *T. vivax* (Day 0-28 post infection and day 28-112 post treatment) and control bucks

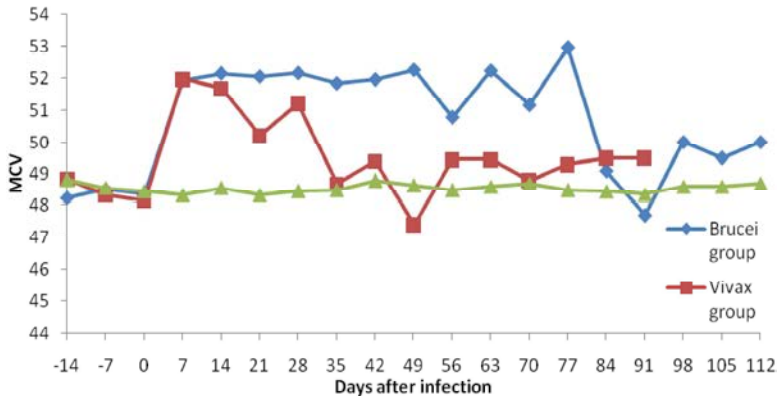

Fig. 5: Mean MCV (fl) in *T. brucei* (Day 0-70 post infection and day 70-112 post treatment), *T. vivax* (Day 0-28 post infection and day 28-112 post treatment) and control bucks

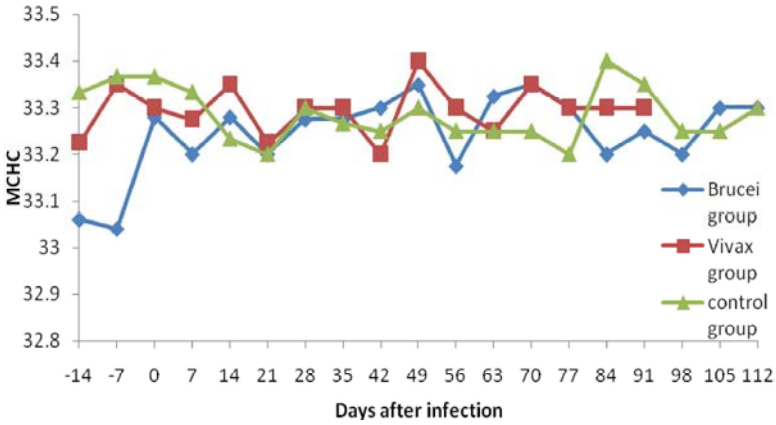

Fig. 6: Mean MCHC (g/dl) in *T. brucei* (Day 0-70 post infection and day 70-112 post treatment), *T. vivax* (day 0-28 post infection and day 28-112 post treatment) and control bucks

There was a significant increase (P<0.05) in the MCH and MCV values of infected bucks during the infective phase from the mean values for the control bucks (Fig. 4 and 5). A sharp increase was recorded 7 days post infection for the *T. brucei*and *T. vivax*infected group. The mean MCH and MCV values of infected bucks increased and remained elevated until treatment in both groups. Changes in the MCH values followed similar pattern with that of MCV. The mean MCHC values of infected bucks fluctuated throughout the course of the experiment; however, there was no statistical significant difference (P?0.05) when compared to the control bucks (Fig. 6).

There was no significant difference (P?0.05) in all the haematological values of the infected animals post treatment when compared with values of the control bucks (Fig 1-6). Treatment with Diminaze® showed rapid improvement to its initial status prior to infection within 7 days post treatment.

DISCUSSION

The results from this experiment showed that *T. vivax* was more pathogenic than *T. brucei* in WAD goats. The infection due to *T. brucei* was relatively mild than infection due to *T. vivax*. This disagrees with result of Adeiza *et al*. [10] who observed that *T. brucei*infection was more pathogenic than *T. vivax*infection in Savannah brown goats. The increase in the population of parasites corresponded with changes observed in the clinical parameters within 7 days PI. This agrees with similar works in *T. brucei*infected ewes [6], goats [10] and in *T. vivax*infected goats [10,12, 15]. The fact that the apparent self cure indicated by the disappearance of clinical signs associated withtrypanosomiasis which happened in the *T. brucei*infected bucks further shows that *T. vivax*was more pathogenic.

This investigation also shows that bucks infected with *T. brucei*were more able to cope with infection than

those infected with *Tvivax*. The changes in the haematological values observed were associated with trypanosome infection and may be due to the onset and severity of anaemia to which these values vary from normal, correlate closely with the intensity and duration of parasitemia[12]. A report ascribed acute anaemia to trypanosomosis to proliferating parasites;however, *T. vivax* induced an acute phase of infection with a short prepatent period, while*T. brucei* induced a chronic phase ofinfection [16]. This is demonstrated by the facts that *T. vivax* infected bucks had higher temperature, higher parasitaemias and greater changes in erythrocyte (PCV, Hb, RBC count, MCV and MCH) values than bucks infected with *T. brucei.* The rapid and spontaneous decline of PCV, Hb concentration and RBC count values in the *T. vivax* infected bucks than in the *T. brucei* infected bucks further testifies that *T. vivax*was more pathogenic. The MCHC values of all infected bucks were within normal range during the infection phase, this may be attributed to the fact that the anaemia was normochromic as reported in sheep [6]. However, it was observed that following treatment with Diminaze®, there was a remarkable but gradual improvement in the clinical condition and haematological characteristics to its initial status prior to infection within one week PI of all the infected bucks. Furthermore, novel diagnostic techniques to monitor the spread of infection in large areas should be explored for developing nations [17-19]. Active epidemiological study should be carried out at the time of new outbreaks to understand the origin, efficacy of current drugs used and control strategies based on the diagnosis and probably, the eradication of trypanosomiasis

The present study demonstrated that *T. brucei* and *T. vivax* infection in WAD goats caused significant changes in the haematological values. It further shows clearly that *T. vivax*was more pathogenic than *T. brucei.* However, treatment with Diminaze® restored the cellular functions of these animals to their pre-infection state.

REFERENCES

1. Seed, J.R., J.B. Sechelski and J.E. Hall, 1985. Multiple alpha-ketoaciduria in Microtusmontanus chronically infected with *Trypanosoma bruceigambiense*. Comp. Biochem. Phys., (B), 82: 73-78.
2. Anosa, V.O., 1988. Haematological and biochemical changes in human and animal trypanosomiasis. Part 1. Revue Elev. Med. Vet. Pays trop., 41: 65-78.
3. Sekoni, V.O., D.I. Saror, C.O. Njoku, J. Kumi-Diaka and G.I. Paluwa, 1990. Comparative haematological changes following *Trypanosoma vivax* and *Trypanosomacongolense* infections in Zebu bulls. Vet. Parasitol., 35: 11-19.
4. Murray, M., W.I. Morrison and D.D. Whitelaw, 1982. Host susceptibility to African Trypanosomiasis: trypanotolerance. Advances in Parasitology. 21.ed. by J.R. Baker and R. Muller Academic Press. London, pp: 1-68.
5. Awobode, H.O., 2006. The biochemical changes induced by natural human African trypanosome infections. Afr. J. Biotechnol., 5(9): 738-742.
6. Ogunsanmi, A.O., S.O. Akpavie and V.O. Anosa, 1994. Haematological changes in ewes experimentally infected with *Trypanosoma brucei*. Revue Elev. Med.vet. Pays trop., 47(1): 53-57.
7. Taiwo, V.O., M.O. Olaniyi and A.O. Ogunsanmi, 2003. Comparative plasma biochemical changes and susceptibility of erythrocytes to *in vitro* peroxidation during experimental *Trypanosome congolense* and *T. brucei* infections in sheep. Israel J. Vet Med., 58(4).
8. Nok, A.J., K.A.N. Esievo, I.A. Ukoha, O.C. Ikediobi, I.J. Baba, Z.B. Tekdek and I.S. Ndams, 1992. Kidney Na+- K+- ATP-ase: A kinetic study in rats during chronic infection with *Trypanosomacongolense*. J. Clin. Biochem. Nutr., 13: 72-79.
9. Akanji, M.A., O.S. Adeyemi, S.O. Oguntoye and F. Sulyman, 2009. Psidiumguajava extract reduces trypanosomosis associated lipid peroxidation and raises glutathione concentrations in infected animals. EXCLI J, 8: 148-54.
10. Adeiza, A.A., V.A. Maikai and A.I. Lawal, 2008. Comparative haematological changes in experimentally infected Savannah brown goats with *Trypanosoma brucei* and *Trypanosoma vivax.* African J. of Biotechnology, 7(13): 2295-2298.
11. Murray, M., P.K. Murray and W.I.M. McIntrye, 1977. An improved parasitological technique for the diagnosis of African trypanosomiasis. Trans. R. Soc. Trop. Med. Hyg., 71: 325-326.
12. Anosa, V.O. and T.T. Isoun, 1980. Haematological studies on *Trypanosoma vivax* infection of goats and intact and splenectomised sheep. J. Comp. Path., 90: 155-168.
13. Schalm, O.W., N.C. Jain and E.J. Carrol, 1975. Veterinary haematology. 3rd ed. Philadeiphia, Lea and Febiger, pp: 15-81.

14. Statistical Analysis Systems (SAS), 2004. SAS user guide, Version 9.13, 9th Edition, Cary, NC, SAS Institute Inc.
15. Murray, M. and T.M. Dexter, 1988. Anaemia in bovine trypanosomiasis: A review. Actatropica., 45: 389-432.
16. Saleh, M.A., M.A. Bassam and S.A. Sanousi, 2009. Oxidative stress in blood of camels (Camelus dromedaries) naturally infected with *Trypanosomaevansi*. Vet Parasitol., 162: 192-9.
17. Liu, X., K. Yang, A. Wadhwa, S. Eda, S. Li and J. Wu, 2011. Development of an AC electrokinetics-based immunoassay system for on-site serodiagnosis of infectious diseases. Sensors and Actuators A: Physical, 171(2): 406-413.
18. Cui, H., H. Li, Q. Yuan, A. Wadhwa, S. Eda, S. Chambers, R. Ashford, J. Jiang and J. Wu, 2013. An AC electrokinetic impedance immunosensor for rapid detection of tuberculosis. Analyst, 138(23): 7188-7196.
19. Li, S., H. Cui, Q. Yuan, J. Wu, A. Wadhwa, S. Eda and H. Jiang, 2014. AC electrokinetics-enhanced capacitive immunosensor for point-of-care serodiagnosis of infectious diseases. Biosensors and Bioelectronics, 51: 437-443.