

Prevalence of trypanosomosis and associated haematological changes among hunting dogs in Abeokuta, Nigeria

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Abstract

African animal trypanosomosis (AAT) is one of the major constraints to the development of effective livestock production systems. Dogs are human companion and are believed to be sentinels for infection with the human species. This study was to detect subclinical and clinical infection of trypanosomes among hunting dogs in Abeokuta and its environs using molecular technique. A total of 87 dogs comprising of 49 males and 38 females were randomly screened for trypanosomes by polymerase chain reaction technique. Among 87 dogs screened, 17.2% were positive for Trypanosoma congolense and Trypanosoma brucei. Prevalence of trypanosomosis in males was 14.3% while the females accounted for 21.1%. Hematological examination revealed a significant increase ($p < 0.05$) in mean white blood cells (20.9 ± 2.11) and monocyte counts (5.9 ± 0.62) of the infected dogs compared to uninfected dogs. Packed Cell Volume (36.0 ± 3.73) and haemoglobin concentration (13.9 ± 2.10) decreased insignificantly, while, red blood cells (7.1 ± 0.87), lymphocyte (60.9 ± 9.63), neutrophil (33.3 ± 9.16) and eosinophil (1.4 ± 0.42) counts increased insignificantly ($p > 0.05$) in infected dogs compared to uninfected dogs. In conclusion, canine trypanosomosis is prevalent in hunting dogs, in Abeokuta.

Keywords: Trypanosomosis, prevalence, hunting dogs

Introduction

Trypanosomosis is a debilitating insect-borne protozoan disease of both animal and humans which continues to cause immeasurable losses in tropical Africa. It is a serious constraint to livestock production and economic development in many parts of sub-Saharan Africa (Ilemobade, 2009). In tsetse-infested sub-Saharan African countries, pathogenic trypanosomes are transmitted to a wide range of susceptible mammalian hosts, including dogs, through infective tsetse fly (*Glossina*) bites when taking blood meals (Brum *et al.*, 2010). The epidemiology of the disease is determined by the ecology of its vector, the tsetse fly. Canine trypanosomosis is a devastating disease leading to anaemia, infertility,

abortions and death if not treated (Losos and Ikede, 1972). In Nigeria, trypanosomosis in dogs is commonly caused by *Trypanosoma congolense* (Anene *et al.*, 1999). The disease has undergone a dramatic and devastating resurgence in recent years, especially in Sub-Saharan Africa (Welbum *et al.*, 2001). Trypanosomosis in dogs could be acute or chronic in nature. The acute course is mainly characterized by fever, a rapidly deteriorating general condition with anaemia, leucopenia, thrombocytopenia, circular and respiratory symptoms, multiple bleeding and oedema, conjunctivitis and opacity of the cornea. In almost all societies, dogs are widely used for protection, hunting, guarding, companionship, with the main one being security (Wells, 2007). In

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Nigeria, there is increased breeding of dogs as a result of the economic value. Although, dogs pose minimal risk for human infection, they are however sources of zoonotic parasites (Dantas-Torres, 2008; Nonaka *et al.*, 2011). The zoonotic *Trypanosoma bruceirhodesiense* has been reported in dogs in Zambia (Lisulo *et al.*, 2014).

In Nigeria, detection of trypanosomes in dogs has been mainly by microscopy and serology with less accuracy. Accurate detection of trypanosomes both in vectors and in host blood depends on molecular techniques (polymerase chain reaction). Such techniques have proven to be effective in the characterization of trypanosomes (Lehane *et al.*, 2000; Malele *et al.*, 2003), typing new trypanosomes (Gibson *et al.*, 2001), in the collection of epidemiological data (Solano *et al.*, 1999) and in animal treatment. This study used a highly sensitive technique (polymerase chain reaction) to detect the presence of trypanosomes and also determined some haematological changes associated with the infection in hunting and pet dogs in Abeokuta and its environs.

Materials and methods

Blood samples were randomly collected from hunting dogs using the cephalic vein into sample bottles containing Ethylene Diamine Tetra Acetic Acid (EDTA) after seeking owners' consent. The non-coagulated blood samples were transported inside cooler containing ice pack to Molecular laboratory, College of Veterinary Medicine, Federal University of Agriculture, Abeokuta.

Study area

This survey was carried out in three Local Government Areas of Ogun State (Abeokuta North, Abeokuta South and Odeda). Ogun State is a transitional zone between the tropical rain forest and derived savanna zone in the south-west of Nigeria and is located between latitude 7° 3' 39" N and longitude 3° 20' 54 E. Abeokuta, the capital of Ogun State is situated on the east bank of the Ogun River, around a group of rocky outcroppings that rise above the surrounding wooded savanna. Abeokuta is between Latitude 7° 9' North and Longitude 3° 21' East, and 68 meters (223ft) elevation above the sea level. Abeokuta has the humidity of 81.1% and wind of 2.6 m/s west.

Study design

A cross sectional survey was used in this study. The dogs sampled were grouped into sex and study areas.

Study population

A total of 87 dogs of both sexes were randomly screened for the presence of trypanosomes. These comprised of 49 male and 38 female dogs.

Hematological analysis

Blood was collected from the dogs through the cephalic vein into sample bottles containing EDTA and were analysed using auto-haemoanalysing machine (Mindray, North America)

DNA extraction

DNA was extracted from the blood in EDTA bottle using Quick-gDNA™ MiniPrep (Zymo Research Corporation, Irvine, CA 92614, U.S.A) as described by the manufacturer. Briefly, 400

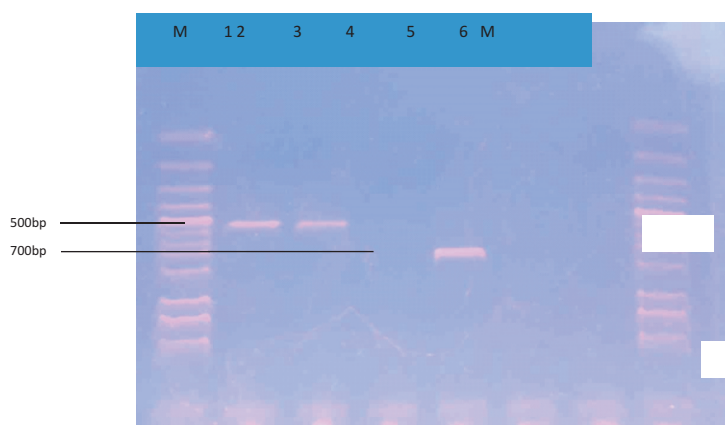


Plate 1: Gel electrophoresis showing bands of Trypanosomes

Key: M = Molecular marker, Lanes 1 and 2 = Trypanosoma brucei, Lane 4 = Trypanosoma congolense, Lanes 3, 5 and 6 = Negative samples

Table 1: Prevalence of canine trypanosomiasis based on activities and sex of the dogs

	Number of Samples	Number of Positives	Prevalence (%)
Hunting dogs	87	15	17.2
Male dogs	49	7	14.3
Female dogs	38	8	21.1

Haematological parameters of non-infected and naturally trypanosome infected dogs

The mean packed cell volume and the HB counts of the infected dogs decreased insignificantly ($p > 0.05$) compared to the uninfected dogs (Table 2). There was a

significant increase ($p < 0.05$) in the mean monocyte and white blood cells counts of the dogs with subclinical infection as compared to the uninfected ones (Table 2). However, the monocyte count was within the normal range.

Table 2: Haematological parameters of dogs uninfected and naturally infected (subclinical and clinical) with trypanosomes

Clinical Parameters	Uninfected	Infected	Reference range (Khan, 2005)
PCV (%)	43.5 ± 2.35	36.0 ± 3.73	35 – 55.00
RBC ($10^{12}/L$)	6.3 ± 0.27	7.1 ± 0.87	5.5 – 8.50
HB (g/dl)	15.3 ± 0.92	13.9 ± 2.10	12 – 18.00
WBC ($10^9/L$)	11.8 ± 0.87 ^a	20.9 ± 2.11 ^b	6 – 17.00
Neutrophil (%)	28.9 ± 2.25	33.3 ± 9.16	60 – 70.00
Lymphocytes (%)	59.9 ± 2.29	60.9 ± 9.63	20 – 30.00
Monocytes (%)	1.6 ± 0.98 ^a	5.9 ± 0.62 ^b	3 – 10.00
Eosinophil (%)	0.4 ± 0.52	1.4 ± 0.42	2 – 10.00

^{a, b} Mean values with different superscripts in the same row are statistically significant ($p < 0.05$) while those with the same superscript are insignificant at $P > 0.05$.

Discussion

African trypanosomes infect both humans and animals in sub Saharan Africa, causing both disease and economic hardship.

Different species and subspecies of the trypanosomes exist in the field, exhibiting different virulence with important epidemiological consequences. In the study

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area (Abeokuta), there are no previous prevalence studies of canine trypanosomosis in hunting dogs, rather than few case reports (Abakpa *et al.*, 2013; Mshelbwala *et al.*, 2015). The diagnosis of trypanosomes infection remains a challenge, particularly since the parasitemia is often very low in a majority of infections (Masake *et al.*, 1997). Molecular technique of identifying trypanosomes provides succour to the problem of detecting subclinical infections due to its sensitivity. In this study, the prevalence of trypanosomosis in hunting dogs in Abeokuta based on the molecular detection was 15 (17.2%). This was considerably higher when compared to 2.08% reported in Zuru, Kebbi State (Tono *et al.*, 2015), 8.8% in Anambra States (Omagebe *et al.* 1984) and 10% in Enugu (Anene *et al.*, 1997). This could be attributed to use of molecular technique in this study which detects the DNA of the parasites rather than the parasites as in the traditional microscopy, as well as their hunting activities which exposes them to the vectors of the disease. The higher prevalence of *Trypanosoma congolense*, 13 (86.7%) compared to *Trypanosoma brucei* 2 (13.3%) showed that *T. Congolense* was the predominant species that infected dogs in this environment, this was in contrast to the reports of (Omagebe *et al.* 1984; Abenga *et al.*, 2005; Anene *et al.*, 2005) that *T. brucei* is the predominant species of trypanosomes infecting dogs in Nigeria. **The low prevalence (13.3%) of *T. brucei* in this study suggested that the species was not the major trypanosome challenge in dogs in Abeokuta.**

The significant increase in white blood cells might have resulted from the increase observed in its components. The slight elevation of neutrophil count might have

been triggered by the invasion of the host's body by the trypanosomes stimulating inflammatory process in response to the infection, while that of eosinophil count might have resulted from the presence of the parasites. The monocytosis seen was an indication of prolonged inflammatory process. Though, elevated levels of white blood cells and monocytes were in contrast to Mario *et al.*, (2000), who reported fluctuations in leukocyte and monocyte counts in dogs infected with *Trypanosoma evansi*. The inconsistencies observed by the different studies might have resulted from other existing infections which were not included in the investigations.

Conclusion

Based on the findings from this study, canine trypanosomosis was prevalent among hunting dogs in Abeokuta, Ogun state, with *T. congolense* being the predominant species. The high prevalence of trypanosomosis among hunting dogs was as a result of their hunting activities which exposed them to the vectors of the disease. There is a need to screen a larger population of dogs in Ogun state for trypanosomosis and molecularly characterize the detected *Trypanosoma* species for prompt treatment of infected dogs and reduction of zoonosis.

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Assessment of Newcastle disease vaccines from different veterinary outlets in Abeokuta

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Abstract

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Newcastle disease (ND) remains one of the major diseases ravaging the poultry industry in Nigeria. Vaccination of birds is carried out to protect birds against the disease. Despite vaccination against ND, birds still come down with the disease. This study was conducted to determine the potency of ND vaccines sold at different veterinary outlets in Abeokuta. Newcastle disease vaccines were purchased from three veterinary outlets (I, II and III) in Abeokuta over a period of 3 weeks and the haemagglutination (HA) titre determined. A total of 50 broiler chicks were also purchased and divided into 4 groups A-D. Groups A-C had 12 birds each and vaccinated against ND while Group D (Control) had 14 birds and were not vaccinated against ND. Groups A-C were vaccinated with ND vaccines with different HA titre and the antibody response determined using haemagglutination inhibition (HI) assay. Varying haemagglutination titre was recorded for all the vaccines purchased from the three outlets. The first batch of vaccines had haemagglutination titre of $0\log_2$, $6\log_2$ and $0\log_2$ for outlets A, B and C respectively. The second batch had $6\log_2$, $7\log_2$ and $5\log_2$ while the third batch had $4\log_2$, $3\log_2$ and $3\log_2$ for outlets A, B and C respectively. Antibody titre stimulated in vaccinated birds by the second batch of vaccines for groups A, B and C birds were $1536\log_2$, $1792\log_2$ and $768\log_2$ respectively, while the control birds had HI titre of $5\log_2$. It is recommended that veterinary outlets improve the storage of vaccines, vaccine potency test be carried out on vaccines regularly and seromonitoring for humoral immune response in vaccinated chicken flocks be carried out for successful control of Newcastle disease.

Keywords: Vaccination, seromonitoring, potency, haemagglutination

Introduction

Newcastle disease (ND) is an acute infectious and highly contagious disease (Ohore *et al.*, 2002) with the potential of causing 100% morbidity and mortality in unvaccinated poultry (Chakrabarti *et al.*, 2006). The disease has both epizootic and enzootic patterns in different flocks or population but it is an epizootic in intensive poultry and is responsible for most economic losses associated with poultry production (Diel *et al.*, 2012). The occurrence of highly virulent ND infections are recognized as notifiable disease reportable to the Office of International Epizootics (Miller *et al.*, 2015) and is one of

the main sanitary barriers for the free trade of poultry and poultry products (Chitate and Gutal, 2011). Newcastle Disease in Nigeria has been a limiting factor to poultry meat and egg production (Saidu *et al.*, 1994). The severity of the disease is dependent on the host age, immune status of the birds, environmental stress and on the virulence of the strain of the Newcastle disease virus (McFerran *et al.*, 1988). In the control of this disease bio-security and hygiene measures are very essential in the prevention of the introduction or the spread of the disease.

In addition to bio-security measures, the