

# Seroprevalence of canine leishmaniasis in Kwara, Oyo and Ogun states of Nigeria

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**Abstract** Leishmaniasis is an important tropical disease that is gradually gaining attention in Nigeria. The canine species which include domestic dogs have been named the reservoir host for the zoonotic form of leishmaniasis. The present study was carried out to determine the seroprevalence of canine leishmaniasis in three selected states of Nigeria using indirect enzyme linked immunosorbent assay (ELISA). Two hundred and seventy-three canine sera were tested for *Leishmania* IgG antibodies. Dogs sampled were grouped into young (<1 year) and adult (>1 year). Total prevalence recorded was 4.40 % (12/273). There was no significant difference ( $P > 0.05$ ) between sera of hunting (4.83 %) and companion dogs (3.03 %). Furthermore, there was no significant difference ( $P > 0.05$ ) between young and adults dogs. There was no correlation between sex and prevalence of canine leishmaniasis. However, significant difference ( $P < 0.05$ ) was observed in the prevalence of each state with Kwara, Oyo and Ogun having 14.63, 3.33 and 1.32 % respectively. The result of this study established the presence of canine leishmaniasis in Oyo, Ogun and Kwara States of Nigeria making the zoonotic form of the disease a possibility.

**Keywords** Leishmaniasis · Sero-prevalence · Reservoir host · ELISA

## Introduction

Leishmaniasis is a major public health problem as identified by the World Health Organisation (2010). They are a

complex of diseases caused by at least 17 species of the protozoan parasite *Leishmania* (Croft and Coombs, 2003) which belong to the Family Trypanosomatidae and cause high levels of varying clinical syndromes (Vannier-Santos et al. 2002). The *leishmania* parasite's life cycle incorporates Diptera flies of the family Psychodidae, the phlebotomine sandflies (Mazloumi Gavgani et al. 2002; da Costa-Val et al. 2007), and the presence of domestic dogs (*Canis familiaris*) cohabiting with humans aids the human disease thus making leishmaniasis a zoonotic infection. The disease affects around 12 million people worldwide, with an annual incidence of approximately two million new cases and 350 million are living at risk of being infected (World Health Organisation 2010). Traditionally, leishmaniasis has been classified into three clinical forms; cutaneous (CL), mucocutaneous (ML) also known as espundia and visceral (VL) also known as the Kala-azar leishmaniasis. The effects of leishmaniasis range from simple cutaneous ulcers to massive destruction in CL and subcutaneous tissues in ML. A recent addition to the classification is the post Kala-azar dermal leishmaniasis (PKDL). However, clinical features of leishmaniasis are highly variable due to the interplay of numerous factors in the parasites, vectors, host and environments involved (Murray et al. 2005).

Although leishmaniasis is endemic in West Africa (Hamed and Velema 1999) it is one of the less recognized or under-reported parasitic infections in this region (Desjeux et al. 1981). In Nigeria, the first report of leishmaniasis in human was carried out by Dyce-Shar (1924) and since then reports on this disease have been scarce. A number of human cases have been reported in the Northern part of Nigeria (Elmes and Hall 1944; Obasi 1991; Badmos et al. 2005; Igbe et al. 2009). Uloko et al. 2010 reported a co-infection of cutaneous leishmaniasis, with Human Immuno-deficiency Virus (HIV).

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Other African countries with reported cases of leishmaniasis include Senegal, Cameroon, Burkina Faso and recently Ghana in 1999 (Baneth 2006). Apart from the case of canine leishmaniasis reported in a dog in The Gambia in the 1980s, there has been paucity of information on canine leishmaniasis in Africa. Outside Africa, endemicity of canine leishmaniasis has been reported in Mediterranean Europe, the Middle East, Asia and Central and South America (Bettini and Gradoni 1986, Ferrer et al. 1988; Quinnell and Courtenay 2009).

In Nigeria today, most rural dwellers engage in hunting as a means of livelihood or for fun, while urban dwellers keep dogs as guards and pets. There has also been a surge in the co-infection between *Leishmania* and acquired immune deficiency syndrome thus complicating leishmaniasis (Singh 2004) and making it an AIDS defining illness. This is the first investigation of canine leishmaniasis in the three states in South Western Nigeria namely Kwara, Oyo, and Ogun States. This study also identified the risk posed to humans by the infection due to close contact with dogs since some dogs are asymptomatic even when infected and may be a source of transmission to sand flies (Cavaliero et al. 1999; Reis et al. 2006 and Moshfe et al. 2008).

## Materials and methods

### Study area

The investigation was carried out in hunting communities in Oyo (8°N, 4°E of Nigeria, average daily temperature of between 25 and 35 °C almost throughout the year), Ogun (7°N, 3°35'E of Nigeria, with similar daily temperature as that of Oyo) and Kwara (8°30'N, 5°E, average daily temperature of between 30 and 35 °C) States of Nigeria. The choice of these locations was based on accessibility as there were limitations due to the fact that most rural dwellers in Nigeria are superstitious and convincing them to have blood samples taken from their dogs was difficult; there were some who refused outrightly. Companion dogs used for the study were from the Veterinary teaching Hospital of the University of Ibadan and the State Veterinary Hospital in Ibadan, Oyo State both of which are referral Veterinary Hospitals for the south western states of the country.

### Sample collection and dog's demographic information

The study was carried out on 273 dogs (207 rural hunting and 66 urban companion dogs). Demographic factors considered during sampling were age (young: <1 year, adult: >1 year), sex (male or female), purpose of dog

keeping (hunting or companion), and location (rural or urban). Samples were collected between December 2012 and April 2013. Blood (3 ml) was collected via the cephalic vein of each dog into plain bottles without anti-coagulant, this was allowed to clot by sitting it undisturbed on the laboratory bench for 30 min then centrifuged at 1,500 revolution per minute for 10 min and the separated sera were put in micro centrifuge tubes and stored at –20 °C until analysed (Henry 1979; Thavasu et al. 1992).

### Serological analysis

The prevalence of leishmaniasis was determined using indirect enzyme linked immunosorbent assay (ELISA) technique to detect antibody against the disease. A commercial kit (GenWay, *Leishmania* Canine IgG-ELISA by GenWay Biotech, Inc. San Diego, CA 92121) was used and tests carried out according to manufacturer's protocol. Calculating results in the GenWay Units, cut-off was set at 10NTU while >12NTU was considered positive as prescribed by the kit manufacturer.

### Statistical analysis

Statistical analysis was done using Graph pad prism (version 5) with a P value of <0.05 as statistically significant. Chi square tests ( $\chi^2$ ) were conducted to test for significant difference in prevalence of leishmaniasis among hunting dogs in the three states. Fisher's exact test was used to examine the relationship between sex, age, use of dog (hunting or companion) and prevalence of canine leishmaniasis.

## Results

### Demographic information

The collation of qualitative variables showed that 207 hunting dogs and 66 companion dogs were used for the study. All the companion dogs were from Oyo state (referral veterinary hospitals in Ibadan). Of the 207 hunting dogs, 90 were from Oyo state, 76 from Ogun State and 41 from Kwara State. In all there were 152 females (134 adults, 18 young) and 121 males (93 adults, 28 young).

### Serological evaluation

Total seroprevalence rate was 4.4 % (12/273). Of the 207 hunting dogs sampled, 4.43 % (10/207) was found to be sero-positive while 3.03 % (2/66) companion dogs were sero-positive with no significant difference ( $P > 0.05$ ) (Table 1). Percentage sero-positivity was significantly

**Table 1** Sero-prevalence of companion and hunting dogs for leishmaniasis

Use	No of dogs sampled	No of positives	Prevalence rate
Hunting	207	10	4.83
Companion	66	2	3.03
Total	273	12	4.40

**Table 2** Sero-prevalence of canine leishmaniasis in hunting dogs in the states

State	No of dogs sampled	No. of positives	Prevalence
Oyo	90	3	3.33
Ogun	76	1	1.32
Kwara	41	6	14.63
Total	207	10	4.83

**Table 3** Sero-prevalence of canine leishmaniasis in the sexes

Sex	No of dogs sampled	No of positives	Prevalence
Female	152	7	4.61
Male	121	5	4.13
Total	273	12	4.40

different ( $P < 0.05$ ) for hunting dogs in Oyo, Ogun and Kwara States and was 3.33, 1.32 and 14.63 % respectively (Table 2). Of the 46 young dogs sampled, none was found to be sero-positive, while 12 of the 227 adult dogs sampled were sero-positive. However there was no significant difference ( $P > 0.05$ ) between the two groups. No significant difference was observed between the sexes, ( $P > 0.05$ ), 4.61 % in all females and 4.13 % in all males sampled (Table 3). Ten hunting dogs (4.8 %) and two companion dogs (3.0 %) were seropositive (Table 4). There was no significant difference ( $P > 0.05$ ) between sero-positivity in male and female in hunting and companion dog category (Table 4).

## Discussion

This study provides serological evidence of canine leishmaniasis in Nigeria where information on the disease have

been scanty and the few reports available in humans are from the Northern part of the country (Elmes and Hall 1944; Obasi 1991; Igbe et al. 2009). In most endemic countries, domestic dogs (*canis familiaris*) are the reservoir host of the disease (Gavvani et al. 2002; Mohebali et al. 2005), the presence of antibodies in dogs as we found will suggest that dogs in Nigeria are suspect as reservoir hosts. Although antibodies may not expressly be indicative of current infection, it however reveals that there has been some exposure to the causative agent. Serological methods for the diagnosis of leishmaniasis have been found to be comparatively more suited for diagnosing visceral leishmaniasis in endemic regions (Alborzi et al. 2007), thus improving the reliability of the results in the absence of physical symptoms.

Sand flies the insect vector of the disease thrives in an environment of warm climatic conditions such as is found in the states sampled and with the recent climatic changes and global warming, determination of the prevalence of canine leishmaniasis is necessary in order to set control measures against the zoonotic form of the disease (Tesh 1995). Average daily temperature in Kwara state may be as high as 38 °C compared to 35 °C in Oyo and Ogun states and it has been suggested that climatic factors may have an effect on the prevalence of canine leishmaniasis especially visceral (Heidarpour et al. 2014) with higher prevalence occurring in the warm climatic region while lower prevalence occurs in the cold climatic region. This coupled with the abundance of rodent burrows in Kwara state (personal communication) which has been identified as breeding site of the phlebotomine flies (Mascari et al. 2013) may be responsible for the higher seroprevalence obtained in dogs in the state. Lifestyle and exposure to sandflies are the most important factors in predisposition of dogs to *leishmania* infection (Ferrer 1992), hence the higher prevalence of 4.83 % in hunting as against 3.03 % in companion is probably due to the fact that hunting dogs are more exposed to sand flies because of their activities in the bush. However, there was no significant difference between hunting and companion dogs. This may be explained by the fact that some companion dogs in Nigeria are not completely confined hence they have access to bushes and forest harbouring sand flies around

**Table 4** Seroprevalence of canine leishmaniasis in different age groups

Category	Hunting	No positive	Prevalence (%)	Companion	No positive	Prevalence (%)
Adult female	114	6	5.3	20	1	5.0
Adult male	70	4	5.7	23	1	4.4
Young female	13	0	0.0	5	0	0.0
Young male	10	0	0.0	18	0	0.0
Total	207	10	4.8	66	2	3.0

the neighborhood. Although more females were sampled in this study, result obtained was similar to other studies (Pozio et al. (1981); Abranches et al. (1991); Sideris et al. (1996), Bokai et al. (1998) and Mohebalı et al. (2005), as there was no significant difference in prevalence between male and female dogs. The higher number of females encountered were probably due to the bias of dog keepers in Nigeria for female dogs used for breeding. Although all sero-positive dogs in the study were in the adult category, there was no significant difference between the two age groups, however only 23 of the 207 dogs sampled were under one year old. On the other hand, having all leishmania sero-positive dogs come from the adult age group could be attributed to the fact that they are more active in terms of mobility, possibly trained and are more likely to be used for hunting and other purposes with increased risk of exposure to the vector.

Leishmaniasis contributes significantly to the propagation of poverty, because treatment is expensive, often unaffordable, imposes economic burden and thus increasing loss of wages (World Health Organisation 2010). Its recent association with acquired immuno deficiency syndrome (AIDS) calls for a strategic control measure especially in a country where HIV/AIDS is on the high side (second largest number of people living with HIV) according to Central Intelligence Agency, The world factbook 2009. The seroprevalence rate of 4.40 % in canine coupled with the reported cases in humans is an indication for the country to pay more attention to the disease. The result of this study therefore brings to fore the presence of canine leishmaniasis in Nigeria especially in the southwestern part and a need to bring workable control strategies into play to forestall the possibility of increasing incidence in human population.

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