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RESEARCH ARTICLE
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Prevalence and seasonal abundance of ticks on dogs and the role of *Rhipicephalus sanguineus* in transmitting *Babesia* species in Maidugiri, North-Eastern Nigeria

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Abstract

Aim: An investigation on ticks and haemoparasites of dogs that belonged to Maiduguri metropolis (Borno State) was conducted.

Materials and Methods: Survey of ticks and haemo-parasites on 400 stray dogs was conducted in Maiduguri from December 2009 to November 2011, using a stratified random sampling technique from different locations of the town. The town was divided into 10 districts out of which a total of 40 dogs were sampled at random from each district.

Results: Four genera of ticks were identified on the infested dogs 384 (96.0), all of which belonged to the family *Ixodidae* (hard ticks). The genus *Boophilus* was predominant with a prevalence of 88.0%, *Rhipicephalus* 10.8%, *Hyalomma* 0.9% and *Amblyomma* 0.3% at (p ? 0.05). Dogs within the age-group of 6-12 months were the most infested, while those within the age-group of 24 –120 months were the least infested. Sex appeared to have a less significant influence (p > 0.05) on the prevalence of ticks among the dog population as females were more infested than the males. The perineum and the ear were the most commonly infested areas, with 328 (85.4%) and 252 (65.4%) respectively, while the scrotal and abdominal regions were the least infested areas 12 (3.1%) each. The month of August showed the highest mean tick burden of 462.5±3.2 ranging from 450-475, while the month of February showed the least number of ticks with a mean of 244.5±3.8 ranging from 239-250. Dogs found to be infected with *Babesia canis* are all harboring ticks of the genus *Rhipicephalus*. Female dogs were more infected (66.7%) with *Babesia canis* than their male counterparts (33.3%) (p ? 0.05) and dogs within the age group of 1-6 months were more infected.

Conclusion: Prevalence of ticks on dogs in this study area is relatively high and the occurrence of *Babesia* species is vector dependent, with ticks of the genus *Rhipicephalus sanguineous* being the most common vector in transmission of the parasite. Seasons also play a vital role in the prevalence of ticks and their associated haemoparasites.

Keywords: prevalence, distribution, seasonal abundance, ticks, *Babesia canis*, dogs, Borno, Nigeria.

Introduction

Ticks, regarded as the largest non-microscopic ectoparasites, are the most important vectors that infest animals, transmit a wide range of pathogens from infected to non-infected hosts. Ticks of the family *Ixodidae* are considered as vertebrate's vectors and they constitute the largest tick genera with about 235 different species described worldwide [1]. *Rhipicephalus sanguineus*, also known as the brown dog tick is the most cosmopolitan specie among the families, transmitting a wide range of pathogens to dogs and other animals, including humans [2-5]. Tick-borne haemoparasites are one of the most important vector-borne infections of dogs. They are numerous and are caused by several etiological agents such as bacterial, protozoan, and rickettsial organisms [6]. They are cosmopolitan in distribution, but are most numerous and exert their greatest impacts in the tropics and subtropics [7-9]. Tick-borne haemopathogens such as *Babesia, Ehrlichia, Anaplasma, Borrelia and Hepatozoon* are of major health concern to dogs and cause severe economic damage to dog owners, some of which are of zoonotic significance [1,10-12].

Dog keeping habit in Borno State is mainly for hunting and herding, to a minimal extent, for security purposes; in which most of them were owned but, not confined. This lack of confinement exposes them to a variety of hazards and infections which include parasites and their vectors [12]; at the same time serving as reservoirs for transmission of infections to non-infected ones and the animals they herd. Maiduguri, located in the Sahel region of Nigeria, is endowed with a favorable climate suitable for the proliferation of ticks and tick-borne parasites. Thus, this research focuses on the common ticks and tick-borne haemoparasites, their distribution based on predilection sites and seasonal abundance among dog population in the study area.

Materials and Methods

Ethical approval: All procedures were performed with the approval of the Animal Ethics Committee of the University of Maiduguri, Nigeria.

Study area: Borno State with Maiduguri as its capital city lies approximately between latitude 10.20^{N} and 13.40^{N}, and longitude 9.80^{E} and 14.40^{E}. Maiduguri town lies around latitude 11.300^{N} and 11.450^{N} and longitude 13.50^{E}. Borno State has an area of 69,436sqkm and it is the largest State in the Federation in terms of geographical area. Located in the north-eastern corner of Nigeria, the State occupies the greatest part of the Chad basin and shares border with the Republic of Niger to the North, Chad to the North-east and Cameroon to the east. Within the country, its neighbors are Adamawa to the South, Yobe to the West and Gombe to the Southwest [13]. The State has a climate which is hot and dry for a greater part of the year in the northern and central parts, while the southern part is slightly milder. The period of wet season varies from place to place due to the influence of the various climatic factors such as the direction of the rain bearing winds and topography, but generally, the rainy season is normally from July to September in the north, up to early October in the south with a relative humidity of about 49% and evaporation of 203mm per year. The State has two major vegetation zones viz: Sahel in the North and Sudan Savannah in the South [13].

Sampling methods: Survey of ticks and haemoparasites on 400 stray dogs was conducted in Maiduguri from December 2009 to November 2011, using a stratified random sampling technique from different locations of the town. The town was divided into 10 districts out of which a total of 40 dogs were sampled at random from each district. Each sub-population (district) was divided into different strata based on age and gender.

Selection criteria: inclusion criteria 1) a dog must be a stray dog or unconfined, 2) must be apparently healthy, 3) no recent treatment against haemoparasites.

Collection of ticks: A total of 400 dogs were physically examined for ticks; ticks present were detached using forceps or hand picking. The collection procedure was carefully performed to avoid destroying the mouth-parts and the ticks were preserved in 2% formalin for further identification [14].

Identification of ticks: Ticks collected from individual dogs were transferred into petri dishes and examined under a dissecting microscope, and identified using the classification keys of [15,16]; and taxonomic characteristics as described earlier [17].

Hematological examination: Blood samples were collected directly from the ear veins using heparinized micro-haematocrit capillary tubes and then centrifuged (1200 x g, 5', and room temperature). The PCV's were determined by haematocrit reader and the color of the plasma was simultaneously checked and recorded. The capillary tubes were then cut using a diamond pencil 1 mm below the buffy coat and the contents of the capillary tube were transferred onto clean glass slides, mixed and covered with cover slip. Thin smears were prepared directly from the ear vein and also from the buffy coat and fixed with methanol and stained with Giemsa. Both the wet and stained smears were examined for the presence of haemoparasites as described earlier [15].

Statistical analysis: Data collected were subjected to student's t-test and categorical data like age group were subjected to one-way analysis of variance (ANOVA) at $p \leq 0.05$ regarded as statistically significant [18]. Microsoft excel was used for presentation of the results.

Results

The results of this investigation revealed that a total of 4,216 ticks were collected from the dogs examined and that *Boophilus* species (fig.1a) were

Table-1. Prevalence of tick species infesting dogs in Maiduguri.

Tick genera identified	No. (%) Collected
Boophilus spp.	3712 (88.0)[a]
Rhipicephalus sanguineus	456 (10.8)[b]
Hyalomma spp.	36 (0.9)[c]
Amblyomma variegatum	12(0.3)[d]
Total	4216

Values with different superscripts are statistically significant (p<0.05).

Table-2. Prevalence of ticks infesting dogs based on age and sex of dogs.

Parameters		No. of dogs examined	No. (%) infested	No. (%) infested with			
				Boophilus	*Rhipicephalus*	*Hyalomma*	*Amblyomma*
Overall		400	384(96.0)	319(83.1)	78(20.3)	16(4.2)	4(1.0)
Age (months)	1 – 6	213	208(97.7)[a]	164(78.8)	39(18.8)	12(5.8)	4(1.9)
	6 – 12	146	144(98.6)[a]	123(85.4)	27(18.7)	4(2.8)	-
	24-120	41	32(78.0)[a]	32(100)	12(37.5)	-	-
Sex	Male	146	132(90.4)[a]	112(84.8)	39(29.5)	8(6.1)	-
	Female	254	252 (99.2)[a]	207(82.1)	39(15.5)	8(3.2)	4(1.6)

Values with similar superscripts are not statistically significant (p>0.05).

Table-3. Distribution of ticks based on predilection sites examined.

Predilection sites	No. (%) of dogs infested (n = 384)	No. (%) of ticks collected (n = 4216)	Mean ± SD (range) of ticks collected	confidence limit
Abdominal region	12 (3.1)	52(1.2)[a]	4.30± 3.20 (1-8)	(4.22, 4.38)
Ear	252 (65.6)	1068(25.3)[b]	4.20± 1.80 (2-11)	(4.15, 4.25)
Facial region	24 (6.3)	120(2.8)[c]	2.40± 1.20 (2-10)	(2.37, 2.43)
Inguinal region	124 (32.3)	448(10.6)[d]	3.70± 1.90 (2-9)	(3.65, 3.75)
Interdigital space	40 (10.4)	160(3.80)[e]	4.00± 3.70 (1-5)	(3.91, 4.09)
Perineum	328 (85.4)	1416(33.6)[f]	4.20± 1.80 (2-13)	(4.15, 4.25)
Scrotal region	12 (3.1)	52(1.2)[a]	4.30± 2.30 (2-5)	(4.24, 4.36)
Thigh	28 (7.3)	76(1.8)[h]	2.70±1.10 (1-3)	(2.67, 2.73)
Thoracic region	144 (37.5)	512(12.1)[i]	3.00±1.10 (1-3)	(2.97, 3.03)
Mammary region	80 (20.8)	312(7.4)[j]	3.90±1.60 (1-3)	(3.86, 3.94)

Values with different superscripts are statistically significant (p<0.05).

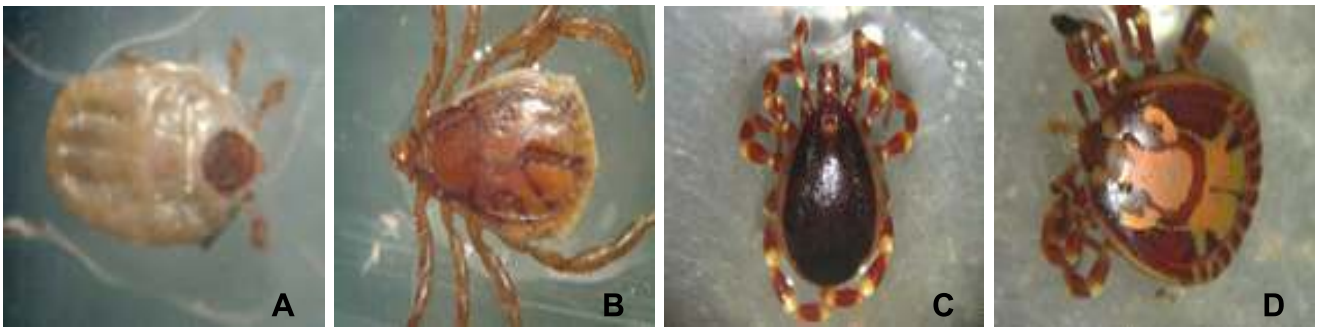

Figure-1. A. *Boophilus*. spp. B. *Rhipicephalus sanguineus,* C. *Hyalomma* spp., D. *Amblyomma variegatum*

most prevalent 3712 (88.0%) compared with *Rhipicephalus* (fig.1b) 456 (10.8%), *Hyalomma* (fig.1c) 36 (0.9%) and *Amblyomma* (fig.1d)12 (0.3%) (Table-1).

Out of the 400 dogs examined, 384 (96.0%) were infested, dogs within 6-12 months of age were more infested (98.6%) compared to those of 1-6 months and 24-120 months age which had 97.7% and 78.0%, respectively (p >0.05) (Table-2).

The distribution of ticks based on the predilection sites showed that the perineum and ears were the most preferred sites with a prevalence of 328 (85.4%) and 252 (65.6%), respectively, while the scrotal and abdominal region had the least with 12 (3.1%) each (Table-3).

The months of August and September (rainy season) had the highest mean ± SD tick values of 462.5±3.2 (450-475) and 468.5±3.1 (458-471) respectively (p>0.05), while the dry months of January through May had lowest mean ± SD tick distribution (Table-4).

The ages of 1-6 months had the highest *B. canis* (fig.2 and3) infection rate of 28 (58.3%) while 6-12 and 24-120 months had 6 (12.5%) and 14 (29.2%), respectively. Female dogs were more infected 32 (66.7%) than the male dogs 16 (33.3%)(Table-5). It is important to note that, in all evaluated cases the parasites were found on *Rhipicephalus sanguineus* infested dogs.

Table-4. Cumulative monthly distribution of tick infestation on dogs in Maiduguri.

Month	Number (%) of ticks collected (n = 4216)	Mean ± SD	confidence limit
December	370(8.8)	360±3.1 (350-370)[a]	(359.9, 360)
January	270(6.4)	368.5±3.4 (367-370)[b]	(367.9, 368.1)
February	250(5.9)	244.5± 3.8 (239-250)[c]	(244.4, 244.6)
March	272(6.5)	261±4.7 (250-272)[d]	(260.9, 261.1)
April	290(6.9)	285±5.9 (280-290)[e]	(284.9, 285.2)
May	310(7.4)	305.5±3.2 (301-310)[f]	(305.4, 305.6)
June	358(8.5)	353.5±3.8 (349-358)[g]	(353.4, 353.6)
July	350(8.3)	332.5±3.4 (315-350)[h]	(332.4, 332.6)
August	475(11.3)	462.5±3.2 (450-475)[i]	(462.4, 462.6)
September	471(11.2)	468.5±3.1 (458-471)[j]	(468.4, 468.8)
October	410(9.7)	412.5±5.0 (410-415)[k]	(412.4, 412.6)
November	390(9.3)	400±4.2 (390-410)[l]	(399.9, 400.1)

Values with different superscripts are statistically significant (p<0.05).

Table-5. Prevalence of *Babesia canis* based on the sex and age of the dogs examined.

Parameters		No. (%) of dogs infected (n = 48)	p = 0.05
Age (months)	1 – 6	28 (58.33)[a]	
	6 – 12	6 (12.50)[a]	p > 0.05
	24-120	14 (29.17)[a]	
Sex	Male	16 (33.33)[b]	
	Female	32 (66.67)[b]	p > 0.05

Values with different superscripts are statistically significant (p<0.05).

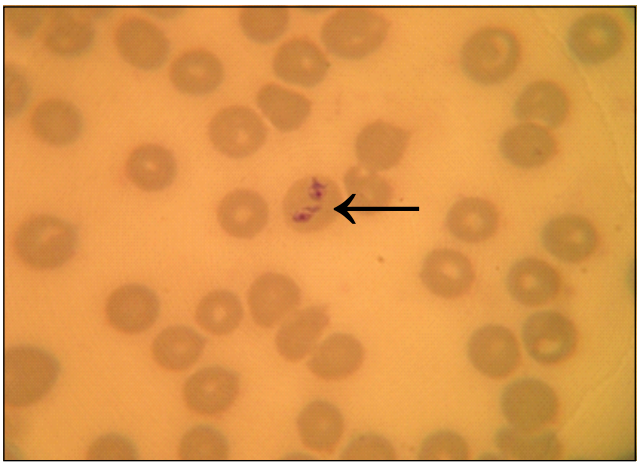

Figure-2. Photomicrograph of a Giemsa's stain thin blood smear showing a red blood cell infected with B. canis (arrow) appearing as a pair of pyriform shaped bodies with basophilic cytoplasm and a reddish chromatin (x100).

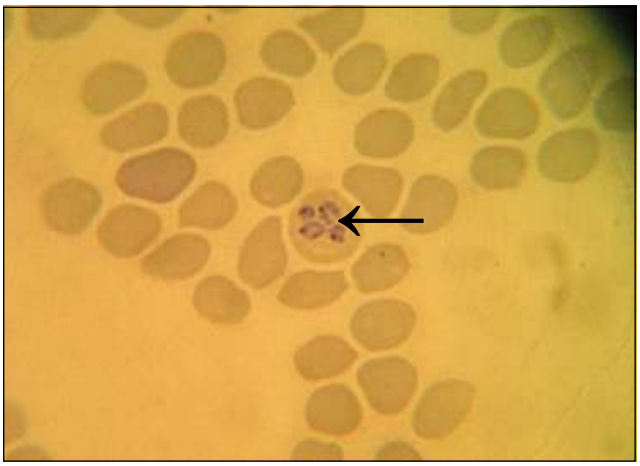

Figure-3. Photomicrograph of *Babesia canis* infected red blood cell (arrow) showing multiple infections (x100).

Discussion

The ticks collected in this study were *Boophilus spp., Rhipicephalus sanguineus, Hyalomma spp.* and Amblyomma *variegatum*, all belonging to the family *Ixodidae* are the common ticks found in Nigeria. [19-27].

With respect to sex and age, female dogs were more infested than their male counterparts. This might be due to the fact that female dogs usually form a sedentary habit while nursing their offspring; as a result, they easily get infested by ticks. This finding also conforms to the reports of Tanwia [21,24,25,28]. The infestation rate was insignificantly (p>0.05) higher in the age-group of 6-12 month old dogs. This finding contrast to the report by Shitta et al [25] and Jame-Rugu [29] who reported that adult and young (puppies) dogs were more infested by ticks than the adolescent ones; even though, the difference as seen in this study is not significant. The insignificance difference in the rate of infestation amongst sexes and age-group may be due to congregation of males and females during mating and close proximity of the young animal's body to the ground than the adult ones. Furthermore, intimacy between the older ones and their offspring helps in the spread of transmission.

The perineum was the most infested predilection site on the dogs examined. This shows that preference of ticks to the various predilection sites could be due to accessibility of blood vessels, difference in the thickness of the skin in the different parts of the body, temperature or variation in microhabitat [30]. The perineum and ears happen to be the most prevalent areas infested on the dogs examined.

Cumulative monthly distribution of ticks in this study area revealed that infestation was higher during the rainy season than during the dry season. This might be due to availability of vegetation cover which provides a favorable environment for the ticks during the rainy season, as they are noted to drop off the host to moult and quest on blades of vegetation to infest hosts that come in contact with them during grazing or movement [25]. Also the optimum temperature and high humidity may encourage high fecundity in ticks. This finding is in conformity with the work of Iwuala and Okpala [19], Iwuala and Okpala [20], James-Rugu, N.N. and Jidayi, S [21], James-Rugu, N.N. and Idu, M.E [22] Biu, A.A. and Konto, M [26], James-Rugu, N.N. and Iwuala, M.O.E. [31].

Parasitological examination of the blood samples collected during the survey work revealed *Babesia canis* (fig.2 and 3) as the most common haemoparasites of dogs in the study area, which coincided with the report [32]; and in all evaluated cases they were found on tick infested dogs. This shows how closely related are the parasites with their vectors in transmission of the disease agent. *Rhipicephalus sanguineus* is the most important vector in the transmission of the parasite (*B. canis*) among dogs as a result of transovarian persistence of *B. canis canis*, observed in *Rhipicephalus sanguineus* suggesting possible implication in transmission [33]. The absence of other haemoparasites like *Ehrlichia canis* and *Anaplasma platys* may be a result of common usage of antibiotics like Doxycycline in most of the veterinary clinics in the study area which are highly effective against most of these bacteria. It may also be likely due to the fastidious nature of the parasites having different developmental stages located in different areas of the body or insensitivity of the diagnostic technique used in detecting the parasites.

Conclusion

In order of predominance, *Boophilus, Rhipicephalus, Hyalomma and Amblyomma* species all belonging to the class hard ticks are the most prevalent ticks on dogs in the study area but, the species *Rhipicephalus sanguineus* is the most infective, and that *Babesia canis* is the predominant haemoparasite of dogs in this study. Ticks preferred the perineum to other predilection sites. Female dogs are more infested than their male counterparts, dogs within the age group of 6-12 months are more prone to infection than other age groups and the month of August has the highest tick burden while the month of February showed the lowest tick burden among dogs in the study area and the haemoparasites of the genus *Babesia canis* is the most common tick-borne haemopathogens found in the study area.

Recommendation

It is highly recommended that a modern molecular diagnostic technique like PCR be employed to detect tick-borne haemoprotozoans in the area so as to give a more accurate and comparative result between the two techniques.

Authors' contributions

AAB conceived, designed and supervised the experiment; MK performed the research and drafted the manuscript; MIA supervised, edited and type-set the work and SC analyzed the data. All authors read and approved the final manuscript.

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Competing interests

The authors declare that they have no competing interests

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