BVA Student Travel Grant Final Report

Prevalence of the Brown Dog tick, *Rhipicephalus sanguineus*, in a population of dogs in Zanzibar, and its role as a vector of canine tick-borne disease.

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Introduction

As a result of human activities, *Rhipicephalus sanguineus* is one of the most widespread ticks throughout the world, and it is most prevalent in tropical and sub-tropical regions (Walker et al., 2005). It lives in close association with dogs and humans, where the burden of infestation in dogs is heavy, especially in dogs that inhabit confined areas and are not treated with ectoparasiticides (Lorusso, et al., 2010). *R. sanguineus* is significant to both human and animal health, due to its role as a vector for pathogens such as *Babesia, Ehrlichia, Hepatozoon canis, Anaplasma platys* and *Rickettsia.* Furthermore, heavy infestations can cause clinical illness, such as anaemia, skin abscesses and tick paralysis (Otranto, et al., 2012).

Vector-borne diseases are becoming an increasingly significant area of research in the UK and Europe due to factors such as increased pet travel (partially due to the PET travel scheme) and increased spread of vectors due to climate change and global warming. For example, there was a 61 per cent increase in the number of dogs entering or re-entering Great Britain between 2011 and 2012 (Vet Record , 2013). However, in order to understand the potential spread of vector-borne diseases and the risk they pose, we need to establish prevalence of infection in areas where vector-borne infection rate is unknown, such as Sub-Saharan Africa.

Over 13,000 people visited Zanzibar in 2013 and this is increasing every year as eco-tourism and exotic travel become more popular (Zazibar Association of Tourism Investors, 2014). People also increasingly want to take their pets on holiday with them, which is a potential risk for the spread of infectious diseases, such as vector-borne disease.

Overall, there is a distinct lack of research detailing canine tick populations and their role as vectors for disease in Zanzibar, despite it being known that tick infestation is very high in Sub-Saharan Africa (Walker, et al., 2003). This research is indicated due to the health risk that *R. sanguineus* poses to both human and animal health, and the increasingly close contact that dogs and humans live in emphasizes this.

The primary aim of this study is to establish levels of tick infestation in a population of dogs in Zanzibar, and determine what proportion of the tick burden *R. sanguineus* makes up. We wanted to establish prevalence of certain vector-borne infections in this population, namely those caused by infection with *Ehrlichia canis* and *Anaplasma platys.* We then looked at whether there is a correlation between dogs that test positive for these and dogs that present with tick infestation.

Lastly, we aimed to get an understanding of how informed local dog owners are about ticks and the potential implication of infection in their dogs.

Methods

Dogs

Our sample population was made up of fifty-six dogs originating from many areas of the island, providing us with a sample representative of the whole island. They ranged in age from young puppies (under 6 months) to adult dogs. In most cases exact ages were unknown, so dogs were grouped into three categories: under six months, young adults, adults. Of the dogs sampled, 27 were female and 29 were male. On initial examination, all dogs appeared to be in good health. Dogs that were excluded were: dogs that had tick treatment within two months, aggressive dogs where attempt to restrain could cause risk of injury to staff or compromise the animal's welfare, and dogs in a poor condition where inclusion may compromise welfare and/or health.

Tick Removal and Identification

All ticks were removed using forceps and macroscopically identified as *Rhipicephalus sanguineus*, 'other', or as 'can't tell'. We identified *R. sanguineus* using detailed photos of the tick as reference (see Appendix 1). Ticks in the 'other' category were those that were not identified as *R. sanguineus.* Ticks in the 'can't tell' category were too engorged to enable accurate identification.

Snap Tests

A SNAP 4Dx test was performed from a blood sample from each dog according to IDEXX protocol, which allowed identification of infection with *Ehrlichia canis, Ehrlichia ewingii, Anaplasma phagocytophilum, Anaplasma platys, Borrelia burgdorferi* and *Dirofilaria* (IDEXX, 2016).
Results regarding *Ehrlichia* spp. and *Anaplasma* spp. were recorded, as both of these pathogens are spread by *R. sanguineus* ticks (Little, 2010). A positive result for *Ehrlichia* spp. was assumed to be *Ehrlichia canis,* as *Ehrlichia ewingii* is not documented in Sub-Saharan Africa (Esemu, 2011). Similarly, a positive result for *Anaplasma* spp. was assumed to be *Anaplasma platys* as *Anaplasma phagocytophilum* is not documented in Sub-Saharan Africa, as it is spread by *Ixodes ricinus* ticks (Ehounoud B. Cyrille, 2016)

Client Questionnaire

A questionnaire was conducted verbally with every owner in order to gather information on variables such as ownership, age, neuter status, medical history, and perception of ticks by local dog owners. Where language difference was a communication barrier, the local Vet accompanying us translated.

Results

Tick Collection

Fifty-six dogs were included in the study; 27 dogs were female and 29 were male. All dogs included were infected with *R. sanguineus* ticks, varying in number from two to 162 ticks, with a mean of 20 ticks per dog (see Table 1).

Out of the 56 dogs sampled, 33 were also infected with ‘other’ species of ticks. A proportion of ticks found, often ticks that were heavily engorged, were not able to be identified macroscopically and were recorded as ‘can’t tell’ for the purpose of this study.

Overall, 90.9% of all ticks removed were identified as *R. sanguineus,* 7.25% were identified as ‘other’ species, and 1.87% were unidentifiable due to being too engorged (see Table 2).

Snap Test

A Snap 4Dx test was performed using blood sampled from the cephalic vein of each dog, which tested for *Ehrlichia canis, Ehrlichia ewingii, Anaplasma phagocytophilum, Anaplasma platys, Borrelia burgdorferi* and *Dirofilaria* (see Appendix 2).

Thirty-three out of 56 dogs (58.9%) were positive for *Ehrlichia* spp. (see Table 3). Out of 27 female dogs that were sampled, 18 were positive for *Ehrlichia* spp. (66.7%). Out of the 29 male dogs that were sampled, 15 were positive for *Ehrlichia* spp. (51.7%).

Ten out of 56 dogs (17.9%) were positive for *Anaplasma* spp. nine of which were also positive for *Ehrlichia* spp. (see Table 3). Out of 27 female dogs that were sampled, 7 were positive for *Anaplasma* spp. (25.9%). Out of the 29 male dogs that were sampled, 3 were positive for *Anaplasma* spp. (10.3%).

Ehrlichia was much more prevalent than *Anaplasma* in this study, as 59% of dogs tested positive for *Ehrlichia,* whereas only 18% of dogs tested positive for *Anaplasma.*

Questionnaire

Ownership was categorised into 3 different groups: privately owned, ZAASO rescue dogs and hunting dogs. Privately owned dogs were those kept as 'pets' by one family; ZAASO rescue dogs included previously stray dogs from various parts of the island that are now kept at a local rescue shelter called ZAASO; hunting dogs included dogs kept by members of the local community for the purpose of hunting, owned by many different families that share the dogs. See Table 4.

Nineteen out of 56 dogs were neutered; 13 female, 6 male. No hunting dogs were neutered. Eight out of the 16 privately owned dogs were neutered (50%). Eleven out of the 18 dogs from ZAASO were neutered (61%).

For most of the dogs sampled, exact ages were unknown, so dogs were grouped into three categories: under six months, young adults, adults. Out of the 56 dogs sampled, there were 16 young adults, 31 adults and 9 less than 6months (see Table 5)

Out of the 56 dogs sampled, 33 had received one or more vaccinations. Almost all rescue dogs and privately owned dogs received vaccinations for Distemper, Infectious Hepatitis and Parvovirus (DHP), and Rabies. No hunting dogs were vaccinated. See Table 6.

All owners thought it was important to remove ticks from dogs and all said they knew how to remove them. All owners said they would remove ticks when seen, however owners of the pet dogs did not actively remove ticks daily, unlike the owners of the hunting dogs who did. Dogs at the rescue shelter had ticks removed at least once a week.

Discussion

The aim of this study was to establish levels of *Rhipicephalus sanguineus* infestation in a population of dogs in Zanzibar, and establish prevalence of certain vector-borne infections in this population, namely those caused by infection with *Ehrlichia* spp. and *Anaplasma* spp.

This study suggests that *R. sanguineus* is the most common species of tick in the canine population in Zanzibar, with 91% of ticks identified as *R.*

sanguineus. Whilst no other papers have been published specifically looking at canine tick population in Zanzibar, this is in agreement with Walker et al. (2003), who report *R. sanguineus* as being the most common canine tick in Sub-Saharan Africa (Dantas-Torres, 2008).

The study showed that the rescue dogs were the most likely population of dog to test positive for *Ehrlichia* spp. (72%). This is expected, as the rescue dogs were once stray dogs, during which time they were very unlikely to ever have ticks removed from them or have received any tick prophylaxis. Hunting dogs were least likely to be positive for *Ehrlichia* spp., with only 41% testing positive. Pet dogs were more likely to have *Ehrlichia* spp. than hunting dogs, with 69% testing positive for *Ehrlichia* spp. A possible reason for this is that owners of the hunting dogs were very vigilant in removing ticks from their dogs, and did so every day, whereas owners of the pet dogs were less likely to spot ticks and were more reluctant to remove them everyday.

The study shows that dogs less than 6 months are much less likely to test positive for *Ehrlichia* spp. (33.3%), compared to young adults (68%) and adults (61%). This is expected as young dogs have had less exposure to tick infestation (Baneth Gad, 1996).

Although 59% dogs tested positive for *Ehrlichia* spp., no dogs appeared to have clinical ehrlichiosis such as lameness, pyrexia, weight loss, suggesting they have the disease in a subclinical form (Shawa E. Susan, 2001) (Little, 2010).

Similarly, 18% of dogs tested positive for *Anaplasma* spp., but no dogs appeared to have clinical anaplasmosis such as joint pain, pyrexia, lethargy, vomiting and diarrhea (Little, 2010). This also suggests the dogs have a subclinical form of anaplasmosis (Little, 2010).

This study only looked at a small sample size of dogs, and hence a larger study would provide more accurate data on tick infestation and levels of vector-borne disease. It would also be useful to investigate the prevalence of *Babesia* spp. infection in the dog population, as *R. sanguineus* is also a vector for this. Investigating prevalence of tick infestation and tick-borne disease in mainland Tanzania and other surrounding mainland East African countries would be useful to compare this data with, to see if being an island affected the prevalence of *R. sanguineus* and levels of tick-borne disease in Zanzibar compared to local, mainland countries.

Conclusions

The results of this study indicate that *Rhipicephalus sanguineus* is the most common species of tick infesting dogs in Zanzibar. It has also shown that the prevalence of *Ehrlichia canis* antibodies in the dog population is very high. Prevalence of *Anaplasma platys* antibodies is also high, but not as high as *E. canis,* suggesting *E. canis* is a more prevalent pathogen within Zanzibar.

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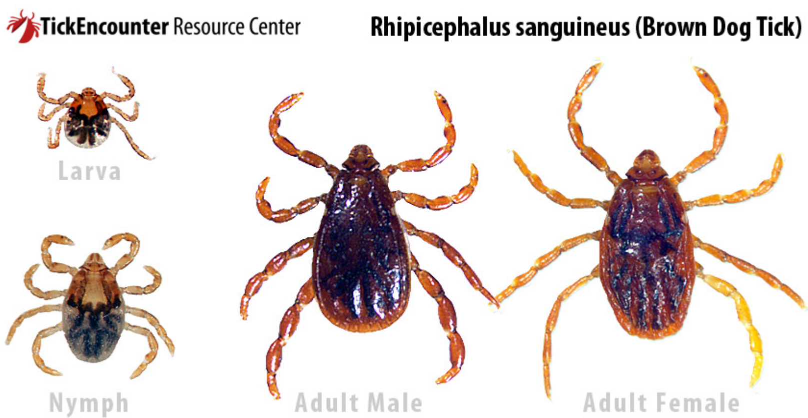
Appendix

Appendix 1



Appendix 2

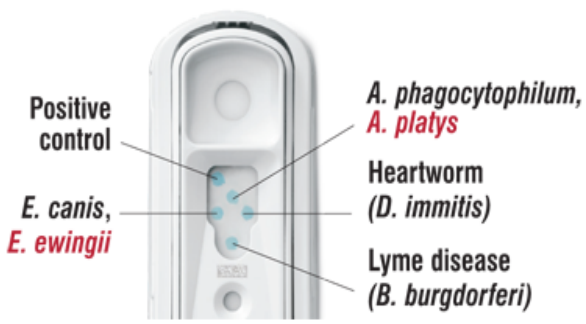


Table 1: Table to show the number and identification of ticks found on each dog.

Table 1: Tick species identified			
Dog	Rhipicephalus sanguineus	Other	Can't tell
1	9	2	0
2	39	3	0
3	5	0	0
4	22	0	3
5	104	5	3
6	60	3	0
7	21	2	1
8	11	0	0
9	43	1	0
10	162	13	3
11	16	1	0
12	50	6	0
13	80	4	0
14	58	4	1
15	2	0	0
16	12	1	0
17	15	0	0
18	21	3	0
19	11	1	0
20	15	0	0
21	10	1	0
22	12	0	0
23	9	6	0
24	5	1	0
25	10	2	1
26	23	4	0
27	13	2	0
28	9	0	0
29	5	0	0
30	7	1	0
31	6	1	0
32	17	0	0
33	12	0	0
34	9	1	0
35	11	0	1
36	13	2	0
37	8	3	0
38	10	0	1
39	15	3	1
40	7	0	0
41	4	0	0
42	13	1	1
43	23	4	1
44	15	0	1
45	6	0	0
46	8	0	0
47	10	2	1
48	9	3	0
49	9	0	1
50	3	0	0
51	6	1	0
52	7	0	0
53	9	1	1
54	2	0	1
55	14	1	1
56	10	0	0

Table 2: Table to show the percentatges of ticks identified as *Rhipicephalus sanguineus*, other or can't tell from all the ticks removed from the dogs.

Table 2: Tick species percentage	
Tick species	**Percentage of the removed ticks (%)**
Rhipicephalus sanguineus	90.9
Other	7.25
Can't tell	1.87

Table 3: Table to show the results of the SNAP 4Dx test for *Anaplasma* spp. and *Ehrlichia* spp.

Table 3: Snap test results				
Ownership of dog	**Positive result for only *Ehrlichia* spp. [1] (%)**	**Positive result for only *Anaplsama* spp [2] (%)**	**Positive result for both *Ehrlichia* spp. and *Anaplasma* spp. (%)**	**Negative result for both *Ehrlichia* spp. And *Anaplasma* spp. (%)**
Privately owned	56.3	6.3	12.5	25
ZAASO rescue dog	38.9	0	33.3	27.8
Hunting dog (shared ownership)	36.6	0	4.5	59.1
1= *Ehrlichia canis and Ehrlichia ewingii, 2=Anaplasma phagocytophilum and Anaplasma platys*				

Table 4: Table to show the ownership statuses of dogs included in the study

Table 4: Ownership status of sampled dogs	
Ownership of dog	**Percentage of dogs (%)**
Privately owned	28.6
ZAASO rescue dogs	32.1
Hunting dogs (shared ownership)	39.3

Table 5: Table to show the age distribution of dogs included in the study.

Table 5: Age of sampled dogs	
Age category	**Percentage of dogs (%)**
Under 6 months	16.1
Young adult	28.6
Adult	55.4

Table 6: Table to show the vaccination status of dogs included in the study.

Table 6: Vaccination status of sampled dogs				
Ownership of dog	**Only DHP* (%)**	**Only Rabies (%)**	**Both DHP and Rabies (%)**	**No Vaccinations (%)**
Privately owned	0	69	25	6
ZASSO rescue dogs	0	0	100	0
Hunting dogs (shared ownership)	0	0	0	100
*DHP = Distemper, Parvo virus and Infectious Hepatitis Vaccine				