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SPATIO-TEMPORAL ANALYSIS OF DOG ECOLOGY AND RABIES EPIDEMIOLOGY AT A WILDLIFE INTERFACE IN THE LOWVELD REGION OF SOUTH AFRICA

by

Michael Grover

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Dedication

This thesis is dedicated to God for the strength to carry on and to my family for all the understanding and support you provided me throughout my thesis.

Declaration

I hereby declare that this dissertation, which I submit in fulfilment of the degree of Master of Science at the University of Pretoria, South Africa, is my own work. This work has not been previously submitted by me for a degree at another university.

……………………………………………………………

Michael Grover

This dissertation emanates from Protocol V002/14 approved by the Research Committee of the University of Pretoria on 25th June 2014 and the Animal Ethics Committee of the University of Pretoria on 5th October 2014.

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Spatio-temporal analysis of dog ecology and rabies epidemiology at a wildlife interface in the Lowveld region of South Africa

Candidate:	Michael Grover
Student no.:	U13418875
Supervisor:	Prof Darryn Knobel
Co-supervisors:	Dr Anne Conan
	Dr Paul Bessell
Department:	Veterinary Tropical Diseases
Degree:	MSc (Veterinary Science)

Abstract

It is estimated that free-roaming dogs comprise on average 75% of dog populations. Interactions between free-roaming dogs and wildlife occur across the globe but little is known about these interactions in the Lowveld region of South Africa, where wildlife areas are fenced off from surrounding communities. Even with extensive fences dogs are still entering the reserves. The study site, a private reserve in eastern Mpumalanga Province on the western boundary of the Greater Limpopo Transfrontier Park, has a high density of human settlements on its boundary. These communities own dogs, many of which are free-roaming. Between January 2009 and March 2014, 170 stray dogs were destroyed inside the reserve and 65.3% of the samples returned a positive result for rabies. Dogs are not limited to the reserve edges and have been documented several kilometres into the reserve.

Eleven geographical factors were used in the spatial analysis: (i) camps or lodges in the reserve, (ii) fence line of the reserve, (iii) water points within the reserve, both natural and man-made (excluding rivers but including pans, dams and waterholes which hold water for most of the year), (iv) access roads from gates to camps and lodges, (v) access gates into the reserve, (vi) pickets (field ranger accommodation) and general staff accommodation, (vii) rivers in stream order from 1 to 6, (viii) vulnerable points for erosion along the fence line, and (ix) villages bordering the reserve. GPS locations of dogs shot were used to obtain nearest distance to each factor. Generalized linear models (GLM) were then used to analyse the spatial data of distance of dogs shot to the nearest factor.

Dogs were significantly more likely to be shot further away from pickets and closer to minor rivers. There was a significant interaction between these two factors ($p < 0.0001$). Dogs that were shot further from villages (odds ratio 1.42, 95% confidence intervals 1.18 – 1.71, $p = 0.0002$) and closer to water (odds ratio 0.41, 95% confidence intervals 0.21 – 0.81, $p = 0.009$) were more likely to test positive for rabies. A univariate GLM, with distance to fence as the only explanatory variable, showed a significant association between this and rabies test result: for every 1 km further away from the fence the odds of a dog testing rabies positive increases by 1.68 (95% confidence intervals 1.20 – 2.36, $p = 0.002$). However the fence is likely not to be the influencing factor but rather other factors close to the fence. To remove the effect of the fence an analysis of the subset of dogs found further than 200m into the reserve found a positive association between distance from a village and a positive rabies test result (odds ratio 1.58, 95% confidence intervals 1.18-2.32, $p = 0.007$).

Temporal analysis of the data shows a higher average monthly number of dogs shot during the wet season (Oct-Mar) as well as a higher variance although the cycle is not strictly seasonal. An upgrade of the reserve fence for security reasons coincided with a decrease in number of dogs destroyed in the reserve in 2012-1014. Home range analysis of most susceptible predators was collected from daily sightings data and overlaid with rabies hotspots, which gives management an indication of the need to vaccinate predators with home ranges in close proximity to the fence.

The spatial results gives management an indication to increase efforts to destroy free-roaming domestic dogs further into the reserve as the likelihood of a positive rabies result is greater. Ultimately efforts should be focused outside the reserve in the communities to eradicate rabies from the host (domestic dogs) before there is a significant transfer of rabies to wildlife.

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List of Abbreviations

ACF	Auto-correlation function
CDV	Canine distemper virus
CI	Confidence interval
GLM	Generalised linear model
GME	Geospatial modelling environment
MPVS	Mpumalanga Veterinary Services
N/A	Not applicable
OR	Odds ratio
OVI	Onderstepoort Veterinary Institute
RABV	Rabies Virus
SWRA	Sengwa Wildlife Research Area
VIF	Variance Inflation Factor
WHO	World Health Organisation

1. INTRODUCTION

1.1. Overview

Free-roaming domestic dogs (*Canis lupus familiaris*) are defined as "those that may not have an owner or are not permanently restrained or under human control" (Hughes & Macdonald, 2013). It is estimated that they comprise on average 75% of dog populations (Hughes & Macdonald, 2013). Lembo et al. (2010) state that, "the issue of roaming dogs seems not to be one of a lack of ownership, but rather an inability or unwillingness by owners to confine their dogs". Interactions between free-roaming dogs and wildlife occur across the globe (Gompper, 2014; Young et al., 2011). According to a recent review (Hughes & Macdonald, 2013), dog-wildlife interactions have been documented 69 times in published peer-reviewed articles. These interactions may take different forms, such as hybridization e.g. with Ethiopian wolves: (Laurenson et al., 1998; Young et al., 2011), competition for prey (Butler, du Toit & Bingham, 2004), and disturbance through harassment of wildlife (Young et al., 2011). However, the largest impact is arguably from the transmission of diseases such as mange, canine distemper and rabies (Butler et al., 2004; Laurenson et al., 1998; van Heerden et al., 2002). Cleaveland et al. (2000) concluded that a single strain of canine distemper virus (CDV) caused mortality in a range of species in the Serengeti ecosystem, Tanzania between 1991-1994, and that this strain was transmissible between domestic dogs and wild carnivores. Woodroffe (1999) reports that rabies and canine distemper were at least partly responsible for the extinction of African wild dogs (*Lycaon pictus*) in areas of the same ecosystem. As reservoirs of rabies, dogs may pose a greater threat to larger predators such as leopards, lions and hyaenas than to other wildlife, as these species have been documented to prey on dogs (Butler, du Toit & Bingham, 2004).

Although interactions between domestic dogs and wildlife have been documented globally, there are few data on these interactions in the Lowveld region of South Africa, where wildlife areas are fenced off from surrounding communities. Even with extensive fences dogs are still entering the reserves. The study site, a private reserve in eastern Mpumalanga Province on the western boundary of the Greater Limpopo Transfrontier Park, has a high density of human settlements on its boundary. These communities own dogs, many of which are free-roaming. Rabies virus re-emerged in the area in 2008, and is maintained in populations of free-roaming dogs. Although perimeter game fences meet Mpumalanga State Veterinary Standards and are continuously maintained and patrolled daily, 170 stray dogs were destroyed inside the reserve between January 2009 and March 2014. Dogs are not limited to the reserve edges and have been documented several kilometres into the reserve. The spread of rabies from dogs to wildlife is therefore of significant concern in the reserve. Contacts between dogs and predators are known to have occurred; however to date no predators have tested positive for rabies. This project was undertaken to improve understanding of the risk of rabies transmission from dogs to wildlife, through spatio-temporal analysis of existing data from the reserve.

1.2. Literature review

1.2.1 Overview of rabies

Rabies is an acute, typically fatal, encephalomyelitis caused by infection with members of the *Lyssavirus* genus in the Rhabdoviridae family, the most important to this study being rabies virus (RABV). Rabies affects the central nervous system of most mammals and is generally fatal. Developing countries are the most affected by the virus and the World Health Organisation (2012) estimates that more than 55 000 people die each year from rabies in Africa and Asia alone. The diagnosis of rabies cases in these developing countries is often not made, as patients may not report to medical facilities; laboratory testing for rabies is limited; and clinical cases may not be reported to the central authorities (Knobel, 2005). Therefore the actual number of rabies fatalities in humans could well be much higher. Efficacious pre-exposure and post-exposure prophylaxis are available to reduce the risk of death due to rabies but often as a high cost, however despite the preventability and high mortality rate if untreated rabies has in recent years become a neglected disease (Nel, 2013, Akerele 2014). Dogs remain the reservoir for Rabies in sub-Saharan Africa and are responsible for majority of cases in humans (Akerele, 2014).

1.2.2 Overview of rabies in Southern Africa

In Southern Africa there are two variants of RABV, namely canid RABV, mainly associated with dogs, jackals, bat-eared foxes, and herpestid (formally known as viverrid) RABV found in mongooses (Davis et al., 2007; Kloeck, 1997; Mkhize et al., 2010). In his review, Wandeler (1993) identifies that Carnivora and Chiroptera populations of a number of species can maintain independent rabies epidemics , categorizing the principal maintenance hosts in the order Carnivora as "small to medium size (0.4-20kg) omnivores, scavenging, and foraging on small vertebrate, invertebrates, fruit and refuse produced by humans". The maintenance host's physiology and biochemistry are key to ensuring the effective transmission of the virus as maintenance hosts are usually "extremely sensitive to their variant but relatively resistant to rabies virus variants of other species" (Wandeler et al., 1994).

In South Africa there are four main maintenance hosts of RABV: yellow mongooses *Cynictis penicillata* (mongoose variant), dogs, black-backed jackals *Canis mesomelas* and bat-eared foxes *Otocyon megalotis* (canid variant). Figure 1 shows the distribution of laboratory-confirmed rabies cases in these four species from 1993 through 2012. Other species may be infected following transmission of the virus from these maintenance hosts. These species are 'spillover' hosts, incapable of sustained intraspecies maintenance of specific variants. The following is a list of species in which RABV infection has been diagnosed, and which are found in the study area (Barnard & Hassel, 1981; Bishop et al., 2010; Hughes & Macdonalds, 2013):

Civet (*Civettictis civetta*)
Small-spotted genet (*Genetta genetta*)
Lion (*Panthera leo*)
Leopard (*Panthera pardus*)
African wildcat (*Felis lybica*)
Caracal (*Felis caracal*)
Serval (*Felis serval*)
Honey badger (*Mellivora capensis*)
Black-backed jackal (*Canis mesomelas*)
Side-striped jackals (*Canis adustus*),
Wild dog (*Lycaon pictus*)
Brown hyaena (*Hyaena brunnea*)
Spotted hyaena (*Crocuta crocuta*)
Ground squirrel (*Xerus inauris*)
Tree squirrel (*Paraxerus cepapi*)
Greater cane rat (*Thryonomys swinderianus*)
Cape hyrax (*Procavia capensis*)
Chacma baboon (*Papio ursinus*)
Duiker (*Sylvicapra grimmia*)
Steenbok (*Raphicerus campestris*)
Warthog (*Phacochoerus aethiopicus*)
Impala (*Aepyceros melampus*)

1.2.3 Rabies in Mpumalanga Province, South Africa

Mpumalanga Province (indicated on Figure 1 by the green square) occupies 6% of the land area of South Africa and contains approximately 8% of the human population (Statistics South Africa, 2011). The majority of the province's population (59.7%) lives in rural areas (Statistics South Africa, 2011). Large areas of the province are made up of commercial agriculture and protected wildlife areas, including the southern portion of the Kruger National Park. High dog populations are found in communal areas as dog population density correlates with human population density (Mkhize et al. 2010). Although both Southern African RABV variants have been recorded in Mpumalanga Province, the mongoose variant is restricted to the western parts of the province, with the closest recorded outbreaks over 200 km away from the study site (from Figure 1). Until 2007, canid rabies was endemic only to the southern Nkomazi district of the Province, adjacent to Swaziland and Mozambique. Sporadic outbreaks were limited to Mbombela and the south-eastern parts of Mpumalanga (Figure 2a). Spread of rabies was largely limited by the geographical landscape and physical barriers. According to the Skukuza State Veterinary Office, from 1977-2006 no rabies was reported within the Kruger National Park despite the park bordering the northern part of the Nkomazi area and occasional reports of stray dogs in the park (Kloeck, 1997; Mkhize et al. 2010).

From 2008 rabies spread northwards, entering Bushbuckridge Local Municipality on the study reserve's western boundary. Figure 2b indicates the probable temporal spread of dog rabies to the area. The areas of spread had populations of susceptible dogs that favoured transmission of the virus. Mkhize et al. (2010) highlight that the "the identification of rabies cases in close proximity to the [Kruger National] park poses a threat to wildlife." Mkhize et al (2010) indicate that the virus originated in KwaZulu-Natal Province, and entered via Swaziland. The arrows on the maps show the threat to wildlife, with the spread of rabies virus north from Nkomazi district in 2008 (Figure 2b). Rabies entered the dog population in the study area in early 2009 (Mkhize et al., 2010). The main threat to wildlife relates to the possible spillover of RABV from dogs and its establishment in the diverse and abundant wildlife communities in the adjacent reserves.

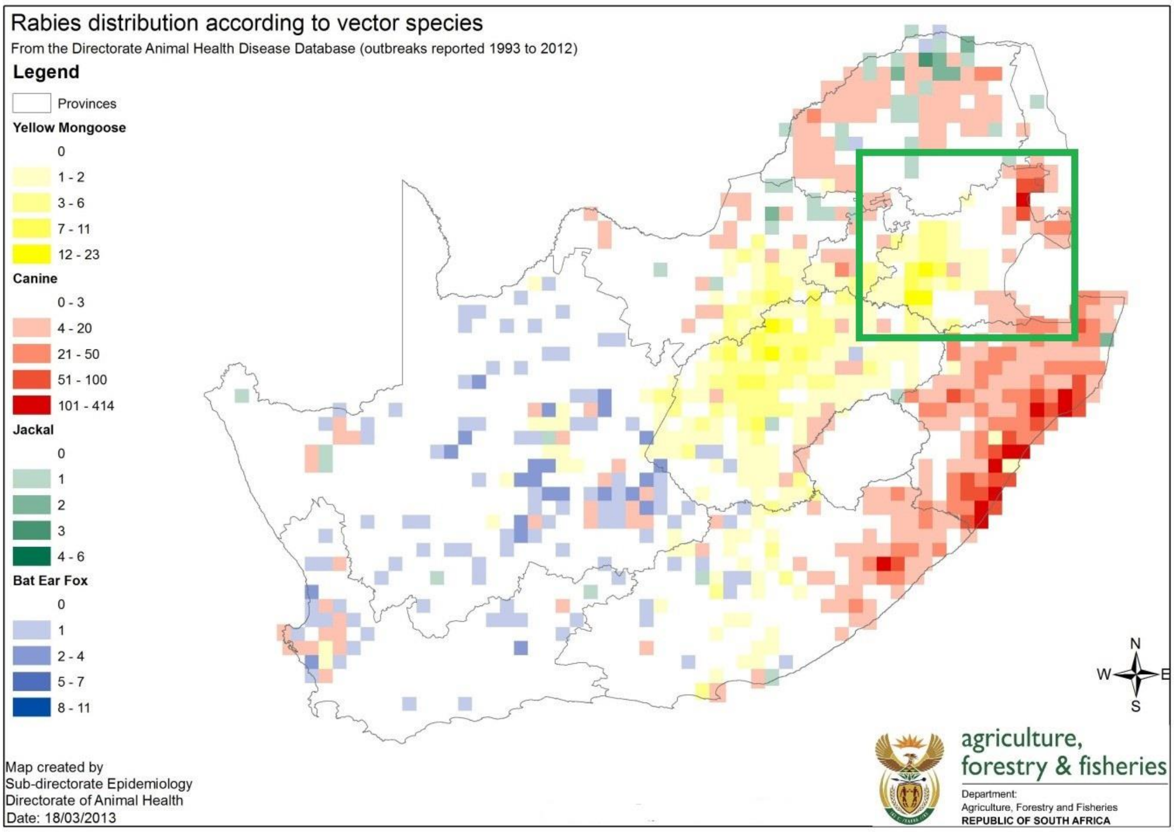

Figure 1. The distribution of rabies in South Africa by major host species. From: Rabies Guide for the Medical, Veterinary and Allied Professions, 2nd edition (2010). Reproduced with permission from the Directorate Animal Health, Department of Agriculture, Forestry and Fisheries

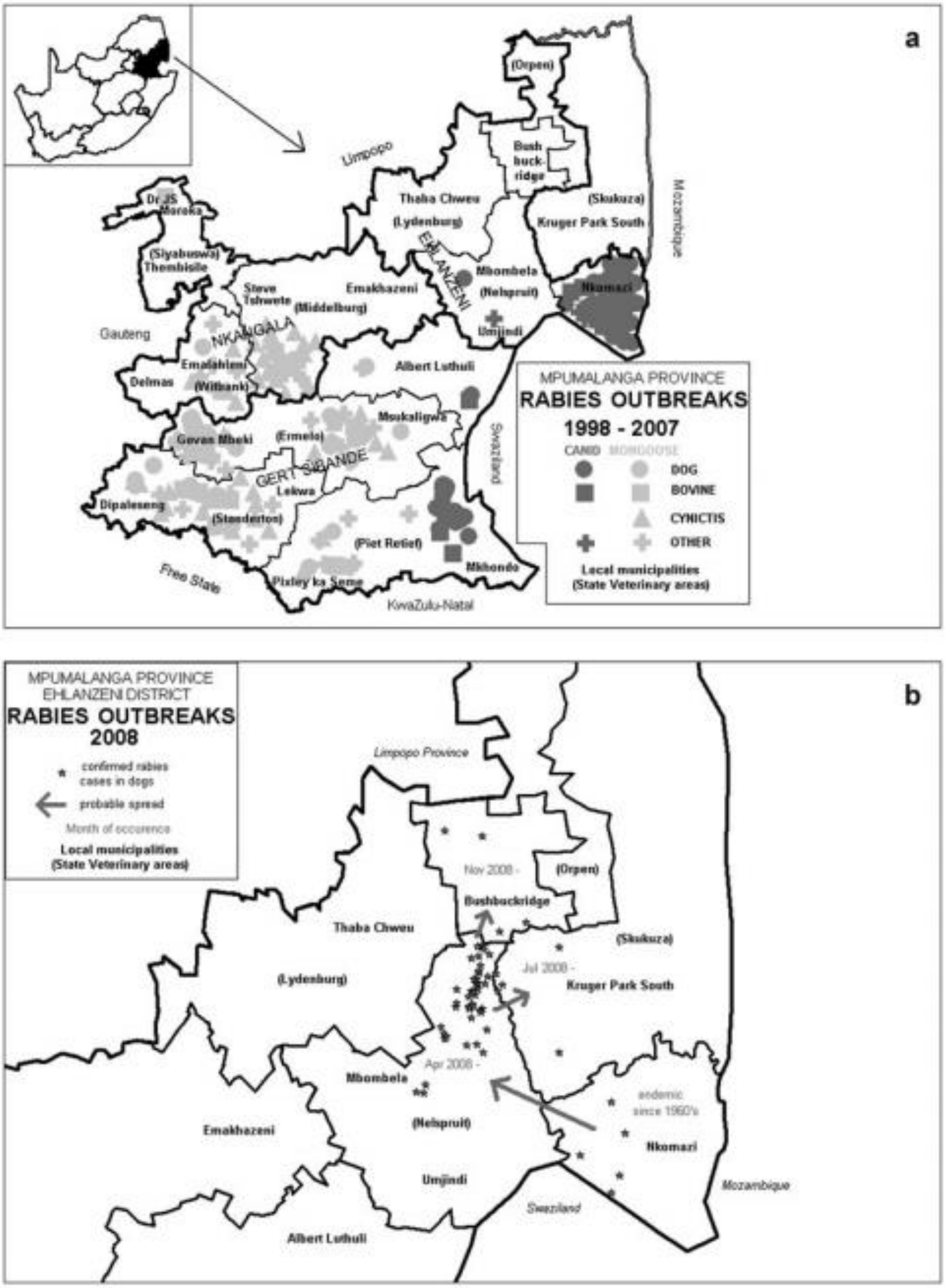

Figure 2. Maps from Mkhize et al. (2010) indicating (a) the prevalence and distribution of rabies cases in both biotypes between 1998 and 2007, and (b) the distribution of dog rabies outbreaks in Mpumalanga Province in 2008.

1.2.4 Rabies in wildlife

A study by Lembo et al. (2008) of RABV reservoir dynamics in a complex multi-host community (the Serengeti ecosystem) concluded that domestic dogs were the only population essential for persistence of the virus, and that populations of wild carnivores did not maintain the virus. On the other hand, studies have shown that RABV can persist in populations of certain species of wild carnivores: Zulu et al. (2009) showed that under appropriate ecological conditions, black-backed jackals are capable of sustaining rabies infection cycles independently of domestic dogs. Cases of rabies in black-backed jackals in South Africa occur predominantly in the northern and north-western parts of Limpopo Province, and rarely in the eastern Lowveld region of the province (Figure 1). No cases of rabies were reported in jackals from Mpumalanga Province between 1993 and 2012, although three confirmed cases in black-backed jackals and four in African wild dogs were reported in a reserve in Limpopo Province 90 km away from the study site in August 2015 (personal communication, Grant Beverly EWT). Persistence of RABV variants in host communities may depend on the density of those hosts to which the variants are adapted (e.g. jackals). In Limpopo Province, jackals can occur at relatively high densities on commercial cattle farms and wildlife ranches from which other carnivores are excluded, while in the diverse wildlife communities of eastern Mpumalanga, jackal densities may be held in check through competition with or predation by other carnivores. Opportunities for spillover from infected dogs will also be fewer in lower-density populations.

Other than jackals very little literature is available for potential wildlife hosts of the virus in the Mpumalanga area. The study area forms part of an extensive open system and therefore the movement of animals with large home ranges such as African wild dog is important. Mills (1993) postulates that differences in social systems and behavior of two important rabies hosts, African wild dogs and spotted hyaenas, could lead to the differences in the epidemiology of the disease in the two species. Hyaena clan structures are more loose and clan members interact less frequently than the highly social wild dogs that have constant contact with each other but rarely have inter-pack interactions. Mills (1993) highlights that should a "wild dog contract rabies, it is likely that the entire pack will become infected". Due to the social nature of the pack introduction of RABV will spread rapidly within a pack but will most likely not influence other packs, as pack dispersal is rare (happening normally once a year) and distances between packs are usually large. Hyaenas' interactions with other scavengers (such as jackal) at carcasses increases the likelihood of spillover to these other species and subsequent inter-clan transmission (Mills, 1993).

The behaviour of infected animals differs greatly and this influences the spread of the disease. Tepsumethanon et al (2004) showed that dogs did not survive more than nine days after the clinical onset of rabies, although we cannot assume that wildlife react similarly to dogs. Hofmeyr et al. (2000) documented the effect of rabies on a wild dog pack that was released into Madikwe Game Reserve. Although the three males of the pack were vaccinated at translocation the females were not. Through a sequence of events that included intra-pack aggression and killing of pups by a female, rabies was confirmed in three adult females and two yearlings, as well as six black-backed jackals exhibiting abnormal behavior (Hofmeyr et al., 2000). The question still remains as to what the behavioural patterns might be of a wild animal with rabies.

As per communication with field rangers within the study area on confirmation of the state veterinary records used in the study, 6 confirmed cases of predator-dog interactions resulted in the dogs being killed by the predator. All 6 of these dogs were tested and were found to be rabies positive (personal communication, Dr Bjorn Rheininghaus, 2014).

1.2.5 Dog ecology in rural areas

Throughout their range, domestic dogs are strongly associated with humans, directly or indirectly. Humans provide food and shelter for dogs even if those dogs are not owned or cared for (Gompper, 2014). Dog in communal areas of Zimbabwe obtain 87% of their food independently, through scavenging carcasses of domestic animals and consuming human faeces and waste food (Butler, 1998). In Machakos District of Kenya, which is a rural area with large areas adjacent to wildlife (similar to the study site), dogs were recorded at densities of 6-21 dogs per km^2 (Kitala et al., 2001). Although this density is not extremely high compared to other rural areas, it is far higher than similar-sized wild canid populations: densities of jackal in semi-arid savanna range from 4-7 jackal per km^2 (McKenzie, 1993). A recent study in the Mnisi area adjacent to the study site showed that the density of dogs increased from 77 per km^2 in 2011 to 84 per km^2 in 2013 with a population increase of 5.9% in a year (Konink, 2014). Konink (2014) states that this density far exceeds the threshold density for canine rabies to be maintained, estimated by the World Health Organisation (WHO) to be 4.5 dogs per km^2 (WHO, 2012).

1.2.6 Vaccination in rural areas

Demographic data on the dog population in 2011 and 2013 for the Mnisi area identified the rabies vaccination coverage to be 68.2 % in 2011 and 59% in 2013 (Akerele, 2014). The recommended 70% coverage by the WHO (World Health Organisation, 2006) is often a misconception of the critical threshold for herd immunity. Research done by Hampson et al., (2009) in Tanzania showed that the figure of 70% is recommended as the target coverage for annual vaccination campaigns as it ensures long term elimination of rabies in almost all demographic settings, including those with high population turnover. The data from the Tanzania did show successful control of the disease at low levels of coverage (30-50%) for short term control however turnover of dogs in rural areas is high and therefore where vaccinations are carried out in pulses the turnover reduces the herd immunity (Hampson et al., 2009). Documented reasons for dog vaccination failures that are pertinent to the study area include (Akerele, 2014)

- Lack of adequate strategies that account for the ecology of dog populations,
- Insufficient economic and human resources to reach required vaccination levels,
- Socio-political factors, and
- Limited knowledge of dog populations and therefore incorrect planning in both timing and geographic area

Free-roaming dogs may not remain at the households all day and often follow owners on daily routes as observed by Butler (1998) in Zimbabwe; dogs may also walk unaccompanied on the same routes. The timing of the vaccination programs may also be an area for exploration, as most campaigns happen mid-morning to early afternoon (personal communication, Dr Bjorn Rheininghaus, State Veterinarian, Bushbuckridge East): this is when many dog owners will be away from their households and therefore their dogs may be with them.

Opportunity in the Mnisi area for alternative vaccination programs or population control is explored by Konink (2014) in her baseline study for the use of a gonadotropin-releasing hormone vaccine that can be used in combination with a rabies vaccine, ultimately aiding in vaccination as well as contraception, and therefore in a lower turnover rate of the population. This addresses the concerns raised by Hampson et al. (2009) of the higher turnover rate and may allow for a more successful elimination at a lower percentage coverage. Tanzania data suggests that 60% or more is logistically and economically feasible in rural communities (Hampson et al, 2009). Rabies control in dogs results in reduced costs of human post exposure prophylaxis and therefore savings to the public health sector.

1.2.7 Dog movement

Little is known of the movements of domestic dogs in rural areas of Africa. The social ecology and movement of dogs from homesteads into adjacent wildlife areas has been documented in north-western Zimbabwe (Butler, 1998; Butler, du Toit & Bingham, 2004). Using VHF radio collars on 14 free-roaming dogs in his study site, Butler (1998) found that 24.1% of the total fixes on dog locations were recorded while the dogs were independent of their owners. Butler (1998) also recorded movements of dogs up to 17 km away from their homes. Mean independent home range area for females were 33.5 ha (+- 2.5, range 0.3-76.0) while males showed a much larger independent home range of 97.2 ha (+- 104.6, range 0.3-316.0) (Butler, 1998). Focusing on the adjacent wildlife area Butler, du Toit & Bingham (2004) recorded sightings of dogs up to 6 km inside the wildlife area when dogs were accompanied by human, but only up to 3 km when alone.

Data on the movements of individual rabid dogs is reported by Butler in Knobel et al. (2014). Movements of 24 suspected rabid dogs were recorded. Fifteen (63%) of these cases left their homestead following the onset of clinical rabies, while the remaining nine stayed at home and died. Ten of the 15 that travelled were adult dogs, and of those adults seven were males. Six dogs (five males) travelled more than 2 km (individual distances of ≥ 2.8 km, ≥ 3.7 km, 4.4 km, ≥ 4.6 km, ≥ 8.7 km, 11.1 km)

Spread of the disease from rural areas into adjacent wildlife areas may be limited by geographic and seasonal factors. In his study, Butler (1998) observed that routes taken by rabid dogs were not governed by geographic factors such as game fence lines, homesteads or riverbeds, although seasonal flooding rivers could be a temporary restriction. Observed crossings of dogs by fence-line patrols indicated little seasonal variation of dogs crossing into wildlife areas, except a once-off increase in 1995 after a season of below-average rainfall (Butler, du Toit & Bingham, 2004). The difference in behaviours between symptomatic or asymptomatic rabies dogs is not well documented. The study site of North west Zimbabwe has the most similar ecological factors to the study area due to the summer rainfall season (October-March), Mean annual rainfall of 622 mm and mean annual temperature of 22^{0}C as well as Savannah woodland with similar wildlife (Butler, 1998).

1.3. Problem statement

Rabies has been reported in domestic dogs in the communities surrounding the reserve since 2008. From January 2009 to March 2014 a total of 170 stray dogs were destroyed inside the reserve. These incursions occurred despite the presence of fences that separate wildlife areas from surrounding communities. There is thus a clear risk of rabies transmission from dogs to wildlife, which raises the possibility of establishment and persistence of the virus in wildlife hosts like the black-backed jackal or other carnivores. Ecotourism makes up the largest economic activity in the surrounding area and the viewing of predators is a major selling point for commercial lodges. Tourism hinges around Big Five game viewing and opportunities to photograph apex predators on safari. These apex predators include leopards, lions, wild dogs, cheetahs and hyaenas. Recorded interactions of rabies-positive domestic dogs and these predators is a cause for concern, as the management interventions of unvaccinated animals needed to comply with legislation and reduce disease risk could disrupt game viewing and heavily influence economic opportunity.

The study looks to learn from the well-documented dataset through temporal and spatial analysis to try and identify if there are any ecological factors that influence or are directly related to the presence of dogs and, more importantly, the rabies status of those dogs. The information gathered can then be communicated to reserve management to assist in decision making with regard to protocols that may reduce dog interactions with wildlife.

1.4. Aims and objectives

The aim of this study was to analyze spatial and temporal data on domestic dogs inside the conservation area and to link this to predator home range information, to identify areas of key concern that can be prioritized for management intervention. The focus of eco-tourism in this area is on large mammals, and this study therefore will examine management implications of relevance to these species.

The objectives of the project were to:

1. Describe the temporal variation in dog distribution and rabies infection status.
2. Describe the spatial distribution of dogs with respect to landscape features (water points, rivers, fences, roads, human settlements)
3. Determine the association between rabies infection status of dogs and their spatial distribution
4. Determine the home ranges of several key predator species inside the reserve, and relate these to dog spatial distribution to identify areas of risk.

2. MATERIALS AND METHODS

2.1. Study location

The study site is a private reserve on the unfenced western boundary of the Kruger National Park (KNP) in the Lowveld region of eastern Mpumalanga Province, South Africa (-24.928578 S, 31.479274 E). The Lowveld Region is characterised by the gradient in topography and climate from west to east. East of the base of the Drakensberg escarpment the lowveld rainfall gradient varies from 1 200mm to 550mm in mean annual rainfall over a linear distance of 100 km. Once descended from the escarpment the land is flat and undulating with mostly savanna grasslands (Shackleton, 2000).

The reserve forms part of the 3.5 million ha Great Limpopo Transfrontier Park. The reserve comprises 49 417 ha of wildlife protected area owned by 58 private land owners and managed by an association and its management team. An additional 13 162 ha of community/state-owned land falls within the boundaries of the protected area, making the total study area 62 579 ha. The area shares an unfenced boundary with an Mpumalanga provincial reserve in the north, and the eastern and southern boundaries are open to the KNP. The southern boundary of the reserve stretches to the Sabie River (Figure 3). The area is named according to old farm names (described in the study as 'Farms') and owned by both private land owners and 33 commercial tourism lodges. These high-end lodges are purely eco-tourism based and rely on game viewing and photography as their major sources of income. The western fence line is 70 km long and is of provincial veterinary specifications for disease control (mainly to contain African buffalo *Syncerus caffer*, the reservoir hosts of foot-and-mouth disease virus). Movement of buffalo and elephant through the fence is extremely rare due to well-maintained electric strands on the fence and the high number of security patrols on the fence line ensures any damages are quickly fixed and large openings are not available for dogs to move in or out of the study area.

During the study period the fence was significantly upgraded for security purposes, while maintaining the veterinary specifications. Community land of the Mnisi and Ama-Shangaana tribal authorities in Bushbuckridge Local Municipality borders the reserve's western and north-western boundary. There are 27 villages in the Bushbuckridge Local Municipality adjacent to the reserve. Eight of these lie in close proximity to the fence, ranging from 20m to 2.5 km from the fence line. A local non-profit organisation, the Reserve Trust, estimates the population of people living in close proximity to the reserve to be 120 000 people, about 20% of the Bushbuckridge population (personal communication, Reserve Management, 2013). This land is used regularly by herdsman, local villagers, hunters (often using dogs) and wood collectors.

Samples taken outside the study area were all done by Mpumalanga State Veterinary Services and included roadkill, dead animals from the Hluvukani Animal Clinic, animals found dead in the communal rangelands as well as dogs shot in housing estates and farms surrounding the study site.

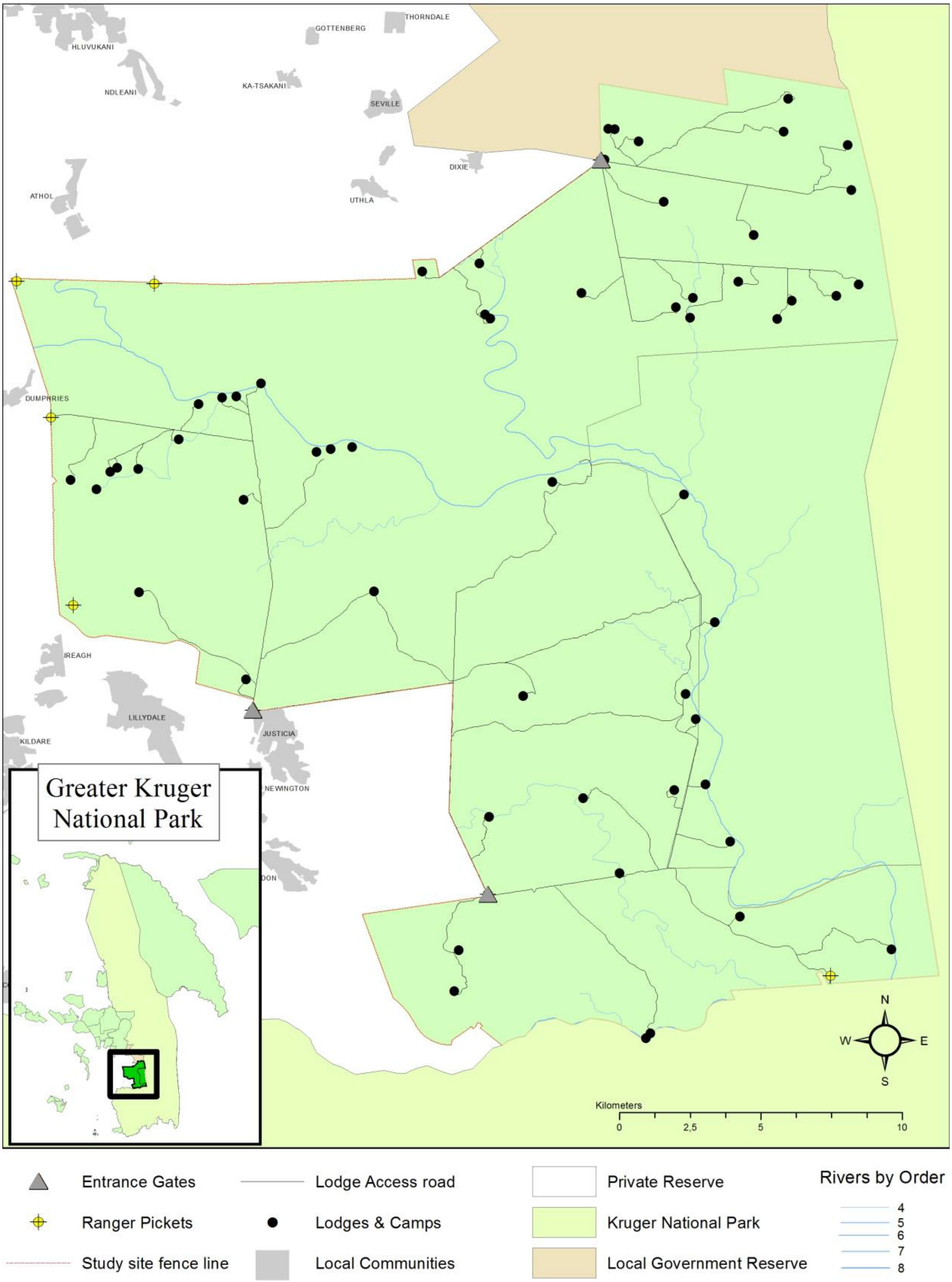

Figure 3. The study site and surrounding reserves and communities in 2013, showing infrastructure such as fence lines, access roads and camps as well as main rivers

The study area has a large population of ungulates as well as large and small carnivores, as found in the Greater Kruger National Park. Although the small mammal population may be important in the epidemiology of rabies, it is not the focus of this study. Small antelope such as grey duiker and steenbok do occur in the community land, along with scrub hare, baboons and vervet monkeys. These mammals are targeted by local hunters, sometimes using dogs.

Movement of animals from inside the reserve to the outside is restricted predominately by the fence and the size of the Bonnox fencing, or their ability to climb over or under the fence. Bonnox fencing (Bonnox (Pty) Ltd, Centurion, South Africa) is a prefabricated, equally-spaced horizontal line wire “hinge-joint” and “ring-locked” to ensure no movement of the wire in the fence, therefore ensuring a consistent spacing in the wire that cannot be pushed wider by an animal. The entire 70 km stretch of the study site fence was upgraded over a 24-month period beginning in January 2010 and ending at the end of November 2011. Fence sections were replaced in 100 m strips at a speed of roughly 8 km of fence replaced per month. No sections of fence were left open overnight and old fence was replaced with new fence to ensure no exit of wildlife or entry of dogs was possible.

The original Bonnox fence gap size was 200mm equally spaced squares, however the upgraded new fence used Bonnox “Kombi-Fence” with graduated spacing starting with 150mm vertical spacing at the base and increasing to 300mm at the top of the fence, to assist with prevention of dogs entering through the fence. Movements of large mammals out of the reserve did occur, but at a frequency of less than 20 a year. Sections of damaged fence were usually repaired within a few hours (personal communication, Reserve Management, 2013).

The fence does restrict movement of small mammals including dogs; however, due to the sandy soils on which the fence is constructed, erosion and digging can allow areas to become vulnerable. At the small non-perennial drainage lines that run under the fence, ground is often soft due to the sandy soil and additional stakes and fencing is added to vulnerable areas; however in heavy rainfall events these additional stakes and fences can be washed away.

Daily fence patrollers do close up or fill in any areas that may be vulnerable, but during heavy rain periods or periods of less frequent fence patrols, small mammals could move under the fence using small ditches and gullies. Major flood events can remove larger stretches of fence at larger drainage sections and these can remain vulnerable for a longer period. During flooding periods reserve management places additional resources on fence maintenance and vulnerable areas are closed as accessibility allows. This can range from a few hours to a few days, depending on where the fence is broken.

2.2 Study design and data collection

The study was conducted from the 1st January 2009 through to the 18th March 2014, in the period that the author worked for the reserve management. The reserve has a best practice management guideline for disease control. Within this guideline is the authority for reserve management to destroy any stray dogs found on the reserve, in line with the Mpumalanga Provincial Veterinary Services (MPVS) animal disease control policy. Surveillance on the reserve occurs through daily foot patrols on the fence line conducted by between 5-9 field ranger groups. Due to the high risk of rhino poaching in the area, the patrols are frequent and cover the entire extent of the 70 km fence in a 24-hour period. The field rangers are based at four pickets (field ranger outposts) along the western boundary fence as well as at each of the three gates along the fence line. Over a 24-hour period the field rangers return to the pickets, and therefore surveillance around the pickets is higher than on areas of the fence line away from the pickets. In addition to the foot patrols, two vehicles drive the fence line on a daily basis, each covering a 35 km stretch. Field rangers on foot and in the vehicles report any sightings of stray dogs and may destroy dogs using their standard-issue firearm. Reported dog sightings are followed up by management patrols and dogs destroyed whenever possible.

The fence line is the highest surveillance area due to the current poaching crisis. Thirty kilometers of fence line is also used as access roads to commercial lodges. Along the 70 km fence line firebreaks are burnt annually during the winter period (June - July), parallel to the fence line, and are cleared from the fence to the inner road. To estimate the actual view distance, the fire break was digitized according to tracer fire lines (inner road) and the fence line from geo-referenced SPOT spatial imagery in ArcMap 10.1 (ESRI, 2013). The total area was calculated to be 4 969 960 m^2. Dividing by the 70 km fence line gives the mean width of the fire break of 71 m. Using the imagery it was confirmed that the firebreak extends past the 100 m buffer in no more than four areas, each of which are less than 2 km in total out of the full 70 km. Therefore we can safely assume that a 100m clear view across the fire break is possible from the fence line road when looking into the reserve.

Lodge occupancy data could not be retrieved from each lodge during the study period; however based on an occupancy of 60%, an average of 50-70 game drive vehicles traverse the interior roads of the reserve during game drive times, usually 05h00 – 09h00 and 16h00-19h00 each day. Any sighting of a dog during these drives is reported and again followed up by management using vehicles. A large majority of dogs reported were destroyed, although there were some cases of dogs not being located once reported; these are assumed to either have been killed by predators or to have moved back outside the fence line. These numbers were not recorded; however, management estimated it to be less than 5% of the total dogs reported (personal communication, Reserve Management, 2013)

Those tasked with destroying the dogs had been trained to do so using a standard-issue rifle and a body placement shot to avoid the head, ensuring the best opportunity for sample collection for rabies diagnosis. Once shot, dogs were collected using gloves, placed into a black plastic bag and taken away from the area to a collection point. Reserve management contacted the MPVS and a representative was sent to sample the dog. Brain samples were collected from all dogs by the State Veterinarian or veterinary technicians of the MPVS, or by the veterinarian at the Hluvukani Animal Health Clinic, Faculty of Veterinary Science, University of Pretoria. All brain samples were sent to the Rabies Laboratory at the Onderstepoort Veterinary Institute (OVI), which is an OIE (World Organisation for Animal Health) rabies reference laboratory. Samples were tested using the gold-standard direct fluorescent antibody test (FAT) and results were sent back to the State Veterinarian. Dog carcasses were disposed of in an incinerator at Skukuza State Veterinary Offices or at the Hluvukani Animal Health Clinic.

Field rangers and reserve management captured data on dogs, including the time, date, location, sex and any additional notes, on a mobile device or smart phone using a custom data collection app designed by the author (www.gocanvas.com). This data was sent to the State Veterinarian as a report. The result of the FAT was sent back to the State Veterinarian and was matched to the data through the use of an OVI reference code. Data was then collated into a Microsoft Excel file by the management and circulated to all parties involved.

The high number of game drive vehicles traversing the reserve affords the opportunity to collect regular sighting data of key predator species such as leopards, lions, African wild dogs and cheetahs. Field guides conducting the game drives use photographs and prior knowledge of the area to individually identify most leopards and lions by name. Hyaena and jackals are not individually identified, but periodic data of hyaena den sites and resident pairs of side-striped jackal and black-backed jackal are recorded. The data are collected by means of GDM, a digital sightings map designed by PeakPi in 2011 (www.PeakPi.com). Daily sightings information is captured by field guides on return from the game drives, and the location, individual information (age, sex and name) and behavior are recorded.

Observation data of any wildlife interactions with dogs are reported and documented immediately by reserve management and distributed to the State Veterinarian. Opportunistic sampling of any dead animal found by management is also conducted, using the same methods described above for dogs. These data included both predators and larger mammals and was done throughout the study period.

2.3 Data analysis

2.3.1 Dataset

Data from dogs and other species tested for rabies were handled in Microsoft Excel 2010 using a spreadsheet format provided by the MPVS, recording the following variables:

Date, Species, Farm, Reserve, State Vet Officers reference, OVI Reference, OVI Result, GPS Position (of where the animal was found dead or shot), Age, Sex, Human contact, Human bite contacts, Additional remarks.

The original data set of all species (including dogs) sampled for rabies from all reserves in the district and their surrounding communities was supplied by the MPVS for the period of January 2009 through March 2014. In conjunction with the reserve data for the study period this dataset comprised 344 records that returned a confirmed OVI rabies test result (positive or negative).

A subset of this dataset was used to show the number of different species sampled and their rabies result and was arranged in a table format according to species and result. The variables included were: Date, Species, Reserve, GPS position, and OVI result. Analysis of 236 dogs sampled in the area was separated from that sampled in the surrounding reserves and community and analyzed according to the rabies result. . If more than one dog was destroyed in the same incident, all dogs tested were treated as individual samples, with the exception of nine puppies that were found tied up in a bag on the 15th November, 2013; this was treated as one incident as all puppies tested negative and had clearly been placed there by someone. Of these, 170 records that occurred inside or within 10 metres of the boundary of the study site were labelled "Study site", while the remaining 66 records from surrounding areas of Bushbuckridge which includes community land, farms and surrounding reserves were labelled "Other". A Chi-squared with Yates correction was done to investigate the relationship between rabies results inside the study area and in the surrounding area. An odds ratio with 95% Confidence Interval was use to on the same data.

2.3.2 Temporal analysis

Data for the temporal analysis in the study site was summed according to the month in which the dog was sampled. To investigate any monthly or seasonal trends in the data over the full study period, all data from January 2009 to December 2013 were combined to investigate data in the five calendar years. Nine samples collected from January through March 2014 were excluded as they fell outside a full year, leaving 161 observations for the temporal analysis.

Rainfall data for 2009 to 2013 was received from the South African National Parks, Scientific Services Department for Skukuza town (-24.992767 E, 31.587696 S). This data was supplemented by rainfall figures from Tshokwane area (-24.784096 E, 31.856873 S) for the period January 2013-June 2013 due to a fault with the Skukuza rain gauge. Skukuza lies on the south-eastern boundary of the study site. Two distinct rainfall periods occur in the area: October to March is considered the wet season and April to September is considered the dry season.

R statistics program (R Development Core Team, 2015) was used to test the number of dogs shot per rainfall season (wet vs. dry), using a Wilcoxon rank sum test. Time series for the total number of dogs shot per month, starting in January 2009 and ending in December 2013, was done. The time series was aggregated into months and a box plot used to show variation and median in dogs shot per month.

Correlations in the time series data were tested using the autocorrelation function (acf) in R for monthly dogs shot time series and monthly rainfall time series. Serial correlation (as it is also known) is the similarities between observations as a function of lag time between each observation. It is used in this instance to look for the presence of periodic signals that might be hidden by noise in the data. This was plotted in R and represented as two graphs.

To investigate seasonality, a seasonal decomposition analysis of the time series data of dogs shot was done, using Loess in R. This is an algorithm that was developed to divide up time series into three different components. These components along with observed data as a reference were plotted. The three components of the algorithm are trend (which describes the fluctuations that are repeated but are non-periodic by relating measurements to the times at which they occur, seasonal (reflecting cyclical variation) and random data (white noise).

2.3.3 Spatial analysis

Data collected without GPS locations was matched using the OVI Reference and prior reports, and allocated an approximate GPS point using the descriptions from the report and ArcMap 10.1 (ESRI, 2013). Twenty reported samples containing no location data and which could not be referenced were removed from the dataset for the spatial data analysis, resulting in a dataset of 150 observations for the spatial analysis. GPS co-ordinates were converted to decimal degrees as the standard for all of these points. The spatial distribution of these samples is shown in Figure 4 below.

Records in table format were imported in the WGS 84 coordinate reference system into ArcGIS 10.1, thus creating shapefiles of all sampled dogs. Existing shapefiles of the following geographic features were collected from reserve management files: (i) camps or lodges in the reserve, (ii) fence line of the reserve, (iii) water points within the reserve, both natural and man-made (excluding rivers but including pans, dams and waterholes which hold water for most of the year), (iv) access roads from gates to camps and lodges, (v) access gates into the reserve, (vi) pickets and staff accommodation, (vii) rivers in stream order from 1 to 6, (viii) vulnerable points for erosion along the fence line, and (ix) villages bordering the reserve. These shapefiles are ground-truthed shapefiles that originated from the Department of Rural Development and Land Reform's National Geo-spatial Information. Details of these features are provided in Table 1.

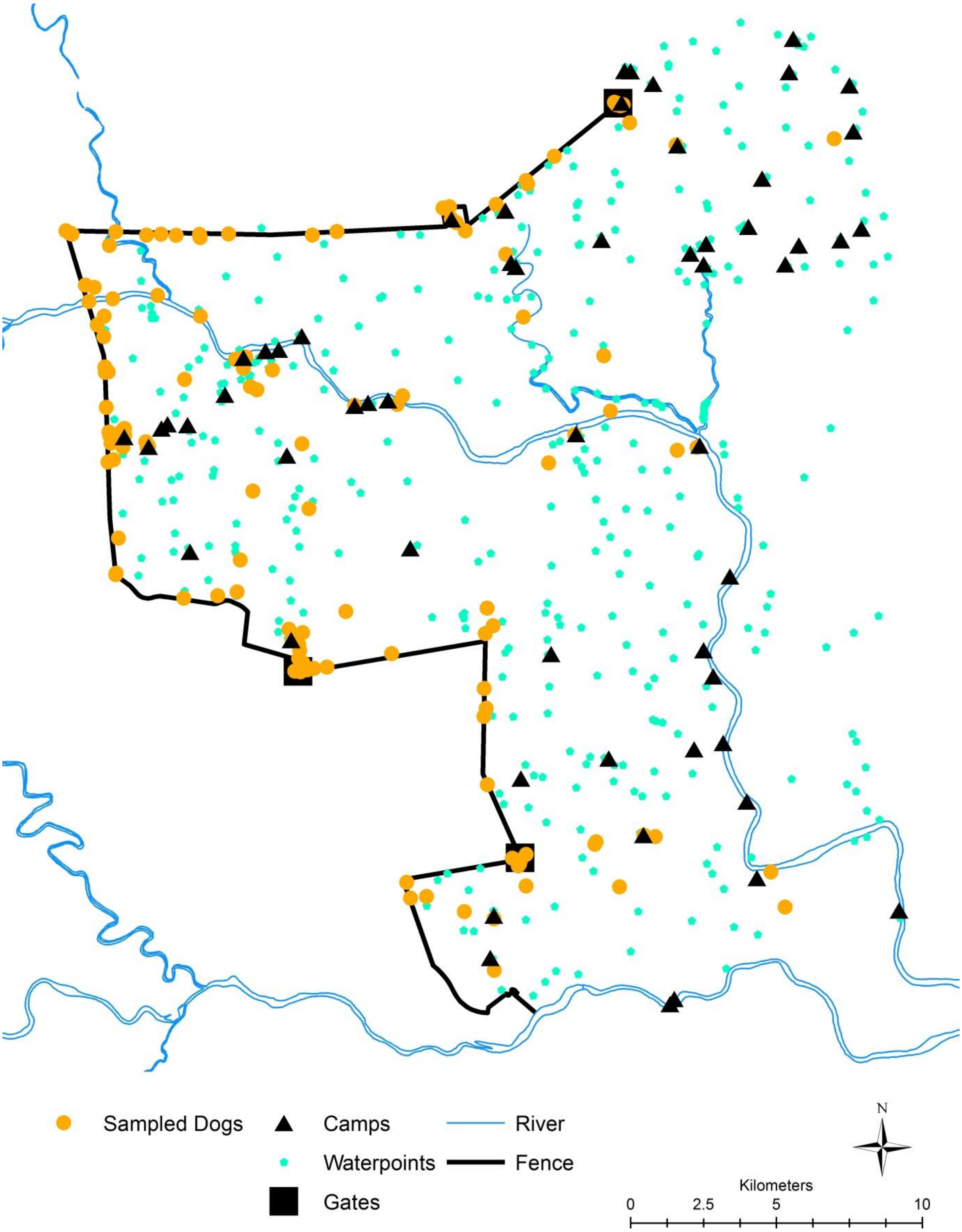

Figure 4. Map of the study area indicating the locations of 150 dogs shot from January 2009 through March 2014.

Table 1. Description of the heading names for all attributes in the main data sheet

Heading Name	Description of Feature
Chronol	Order for processing
Date	Full Date
Year	Separated year
Month	Separated month
Day	Separated day
Species	Species sampled
Farm	Farm on which the dog was shot
Result	Rabies test result from OVI
Latitude	GPS latitude location in decimal degrees
Longitude	GPS longitude location in decimal degrees
Age	Age estimate of dog (juvenile, sub-adult, adult)
Gender	Male, female or unknown
Contact	Contact through biting with any humans and if so, who the person was
Contact_with	Contact through biting with an animal
Access	Access roads from gates to all camps/lodges. These roads have highest vehicle volumes and therefore greatest surveillance
Camps	All camps/lodges in the reserve, regardless of occupation
Fence	Fence line that runs 70 km along western boundary of reserve
Gates	Gates into reserve; these all have security guards on duty from 5am to 11pm daily and staff accommodation at the gates
Pickets	Small out posts where field rangers and security personal live; higher surveillance
River_1	Non- perennial river order 1: mostly dry except in heavy downpours in mid-summer, when they only hold water for a few days
River_2	Non-perennial river order 2: mostly dry but holds water for a few weeks during peak rain season
River_3	Non-perennial river order 3: dry but holds water for approximately a month during peak rain season
River_4	Non-perennial river order 4: holds water for most of the wet season
River_5	Non-perennial river order 5: holds water almost year-round
River_6	Perennial river order 6: holds water year-round
Villages	Villages on the outside of the reserve along the entire boundary
Vulnerable	Vulnerable points on fence, identified as areas where drainage lines cross the fence, causing erosion where dogs could dig under the fence
Water points	All water points on the reserve; most hold water year round

The six river orders were merged into two data sets according to water retention, due to the event-based flow of the rivers. River orders 1 to 4 were combined to create Minor_rivers, representing all rivers that only hold water during high rainfall periods (December to March), and River orders 5 and 6 were combined to create Rivers, representing rivers where water would be available almost all year round.

Using the "Generate near table" tool, run as a batch in the analysis toolbox of Arc Map 10.1, the distance in kilometers was calculated from each sample to the closest geographical feature per shapefile. DBF files were created per geographical feature and using each sample's chronological number as a reference, a database with all dog samples and distances to the nearest geographical feature was created. This file was saved as a comma-separated value (.csv) file to be used in R. All analyses were carried out in the statistical software R, version 2.15.2 (R Core Development Team, 2015).

A bootstrap approach was used to create a random dataset of points falling within the perimeter of the study site. The random data points were created using a convex hull polygon in ArcMap 10.1. This polygon was created around existing data, which together with the fence data for the study site, was merged into a polygon constraining where dogs could occur. The shapefile was created in ArcMap 10.1. Using the R packages maptools (Bivand, 2014), splancs (Rowlingson, 2013), MASS (Venables, 2002) and spatstat (Baddeley, 2005), R was able to read the polygon using the "readShapeLines" function. The polygon created the coordinates of the bounding box of the convex hull, inside which 200 000 random points were created which adequately covered the entire area. From these, 10 000 points were randomly selected for use as generated points inside the study site convex hull. The distance to each geographical feature was created for each of the 10 000 random points, as well as for the 150 actual points of dogs shot. The sample data was bound to the randomly-generated data, creating a binary response variable of randomly-generated point = 0 and actual dog shot = 1.

Possible spatial collinearity could exist between features, therefore correlations between features were assessed using correlation of the bootstrapped random points. The results were represented in a correlation matrix and any correlation >0.75 was flagged as possible collinearity. This is of importance when using variables that have collinearity can result in relatively unstable regression coefficients.

These bootstrap data were analysed by logistic regression using a generalized linear model (GLM) with binomial error distribution and *logit* link function in R, to determine an association between the response variable and the geographical features in terms of distance in kilometres. This is done to test whether there is an association between the location the dog was shot and any landscape variable, independent of the rabies result. Firstly, a univariate analysis of each factor was carried out on all variables and variables with significant p values (<0.05) independently were selected for a step-wise backward elimination approach in the multivariable GLM. Collinearity between variables was checked using a variance inflation factor (VIF) test in the *car* (Fox, 2011) package in R. The VIF takes the R^2 value (from an auxiliary regression of factors and inflates the R^2 through the equation VIF = $1/1-R^2$. Therefore a R^2 greater than 0.8 results in a VIF of 5 or more. Those factors with scores greater than 5 were removed from the model as they will cause the model to be unstable. All remaining features were included in the multivariable GLM. The option of a stepAIC using the MASS package in R as a multivariable GLM is also an option as R works systematically through each GLM and removes the least significant variable, the lower the AIC the better the model. The stepAIC for all variable showed the same results as the manual sequential removal however the stepAIC continued till there was only one variable . Therefore the stepAIC was just used to test different combinations of variables to see if there were any variables being overshadowed; however results were not included. By removing non-significant terms sequentially in a manual process, the model was reduced to include any variables that was significant at $p < 0.05$. Interactions between the main effects were then assessed by including an interaction term in the model.

The results of the two significant factors in the multivariable GLM were plotted in an interaction scatterplot using a smoothed scatterplot of all values from the bootstrap as distance from each variable in kilometres. They were plotted using the control and cases then as a combined in a standard scatter plot.

The association between rabies test status (positive vs. negative) of the 150 records and geographical features was tested with a GLM with binomial error distribution and *logit* link function in R, using the same explanatory variables and model building process outlined above of the univariate GLM and then step-wise backward elimination multivariable GLM.

A Wilcoxon rank-sum test in R was used to test for an association between rabies test status (positive vs. negative) and shortest straight-line distance to the fence from the point in the reserve where the dog was shot. To illustrate the sample bias of the regular patrols on the fence, a univariate analysis was done to investigate the association between rabies test result and the distance from the fence, using a GLM with one factor (distance to fence) as an explanatory variable.

To support the investigation of sample bias on the fence due to the firebreak discussed in Section 2.2, the view shed (the distance of cleared firebreak where dogs would be clearly visible to the human eye) is not likely to exceed 100 m from the fence road. To indicate this distance from the data, a histogram of distance from fence, with equal bin sizes of 50 m each for the first 1000 m was created using Microsoft Excel. The number of points within each bin was graphed by accumulation as a percentage to see the relationship between number of dogs and distance from the fence. This was plotted against frequency within the bin classes. A Pearson's Chi-squared test with Yates continuity correction was run for rabies test results relating to distance from the fence > 0.1 km as well as distance > 0.2 km from the fence. Following this, a subset analysis was performed to examine the association between rabies test results and distance from the fence, using only those samples further than 0.2 km from the fence. The significant variables from the univariate GLM were used in the stepwise backward elimination multivariable GLM to establish statistically significant factors that influence points further than 0.2km from the fence in an attempt to negate fence line patrol sample bias.

2.3.4 Predator home range analysis

Predator home range analysis was done using sightings data recorded between August 2012 and March 2014 for known lion prides and hyaena dens in the reserve. Data points were collated per unit group (prides of lions or hyaena den site). Lion home ranges were presented by each pride in that area as 90% kernel home range (Seaman et al., 1999) using the Geospatial Modelling Environment (Beyer, 2012), while hyaena dens were represented by a single point of the den site. The isopleth polygon created by GME was overlaid with the data on dogs shot and rabies test result. A density heat map of all dogs shot was used to create a visual indicator for predators in a vulnerable area for positive rabies dog interaction through the overlay of sightings information.

2.4 Ethics

Clearance for this study was obtained from the University of Pretoria Animal Ethics Committee (protocol no. V002-14; Appendix). Destruction of domestic dogs is part of the management mandate in the protected area, and sampling was done opportunistically on carcasses of dogs shot, as well as on other dead wildlife in the area.

The Animal Ethics Committee Certificate is added in the appendix on page 64

3. Results

3.1 Results from Bushbuckridge area

In the period from January 2009 through March 2014, 344 animals of 28 different species from the study site and surrounding area were tested for rabies virus (Table 2). In that period, 146 (42.4%) samples tested positive for rabies. Of the 344 samples collected, 236 were from dogs. Of the dogs tested, 59.7% (141/236) were positive for rabies. Twelve samples were taken from other domestic animals: four cattle, seven stray cats and one road-kill cat. Three samples from cattle tested positive. Of the 96 samples from wildlife in the Bushbuckridge area, only two tested positive: one from a small spotted genet and one from a baboon. The latter was the only sample from a wild animal in the study reserve that tested positive. This baboon was found dead in a camp situated close to the fence. The local troop of baboons is known to regularly cross into community land over the fence.

Table 2. List of all species in the study site and surrounding areas that were tested for rabies, and their test result

Species	*Scientific name*	Negative	Positive	Total
African buffalo	*Syncerus caffer*	5	0	5
African Civet	*Civettictis civetta*	2	0	2
African Wild dog	*Lycaon pictus*	2	0	2
Baboon	*Papio ursinus*	7	1	8
Banded mongoose	*Mungos mungo*	1	0	1
Blue Wildebeest	*Connochaetes taurinus*	2	0	2
Bushbuck	*Tragelaphus scriptus*	1	0	1
Caracal	*Caracal caracal*	1	0	1
Common Duiker	*Sylvicapra grimmia*	2	0	2
Dwarf mongoose	*Helogale parvula*	3	0	3
Elephant	*Loxodonta africana*	1	0	1
Giraffe	*Giraffa camelopardalis*	2	0	2
Honey badger	*Mellivora capensis*	1	0	1
Hyaena	*Crocuta crocuta*	12	0	12
Impala	*Aepyceros melampus*	6	0	6
Jackal (Unknown species)	*Canis* spp.	1	0	1
Kudu	*Tragelaphus strepsiceros*	2	0	2
Leopard	*Panthera pardus*	8	0	8
Lion	*Panthera leo*	25	0	25
Nyala	*Tragelaphus angasii*	1	0	1
Road kill (unidentified)		1	0	1
Slender mongoose	*Galarella sanguinea*	1	0	1
Small spotted genet	*Genetta genetta*	2	1	3
Vervet monkey	*Chlorocebus pygerythrus*	1	0	1
White rhinoceros	*Ceratotherium simum*	2	0	2
Zorilla	*Ictonyx striatus*	1	0	1
Cattle	*Bos taurus*	1	3	4
Stray cat	*Felis catus*	7	0	7
Stray dog	*Canis familiaris*	95	141	236
Total		**198**	**146**	**344**

3.2 Study site results

In the study site itself, 170 samples were taken from dogs from January 2009 through March 2014, with 111 (65.3%) of the samples returning a positive result for rabies. These include 20 samples for which no GPS point data was available.

Table 3 shows the number of rabies test results of dogs sampled within the study reserve, compared to those sampled from areas in Bushbuckridge Municipality adjacent to the reserve, which includes community land, farms and surrounding reserves and is labelled 'Other'. A significantly greater proportion of dog samples inside the reserve were positive for rabies, compared to those sampled from adjacent areas (Chi-square test with Yates correction, X^2 = 6.978, d.f. = 1, two-tailed P-value = 0.008). The odds of a Study Site result being positive is 2.26 more likely than a sample from other areas outside the study site being positive

Table 3. Chi-square test results for rabies results for dogs collected inside and outside the study area

		Rabies test result			
		Positive		**Negative**	
	Total samples	**n**	**%**	**n**	**%**
Study Site	170	111	65%	59	35%
Other	66	30	45%	36	55%
Odds ratio of Positive sample in Study Site is 2.26 with C/I of 95% (1.27, 4.03).					

3.3 Temporal analysis

The number of dogs shot in the study area decreased over time (Figure 5): 52 dogs were recorded in 2009, 47 in 2010, 25 in 2011, 28 in 2012, and 9 in 2013. The proportion of shot dogs that were rabies-positive also decreased significantly over time (Chi-squared test for trend in proportions, X^2 = 9.6234, df = 1, p-value = 0.002)

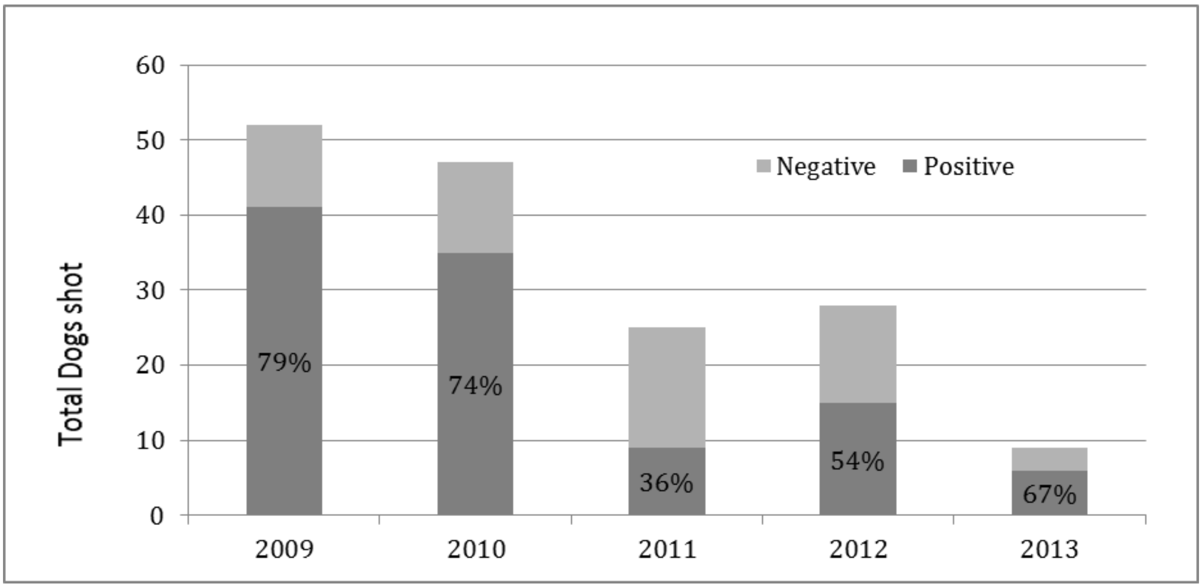

Figure 5. Total dogs shot per year with a colour variation in dark grey showing the percentage of positive rabies results.

The number of dogs shot by month is shown in Figure 6. Fences in the reserve are regularly broken by flooding drainage lines if rainfall exceeds 150-200mm in a month and therefore it would be expected that dog numbers would increase following these periods. To investigate the effect of seasonality (not necessarily related to actual rainfall events), the seasons were split into wet (October through March) and dry (April through September) and analyzed. Of the 161 dogs shot in this period, 105 were shot in the wet period (October to March) and 56 shot in the dry period (April to September). The highest numbers shot occurred before the fence upgrade.

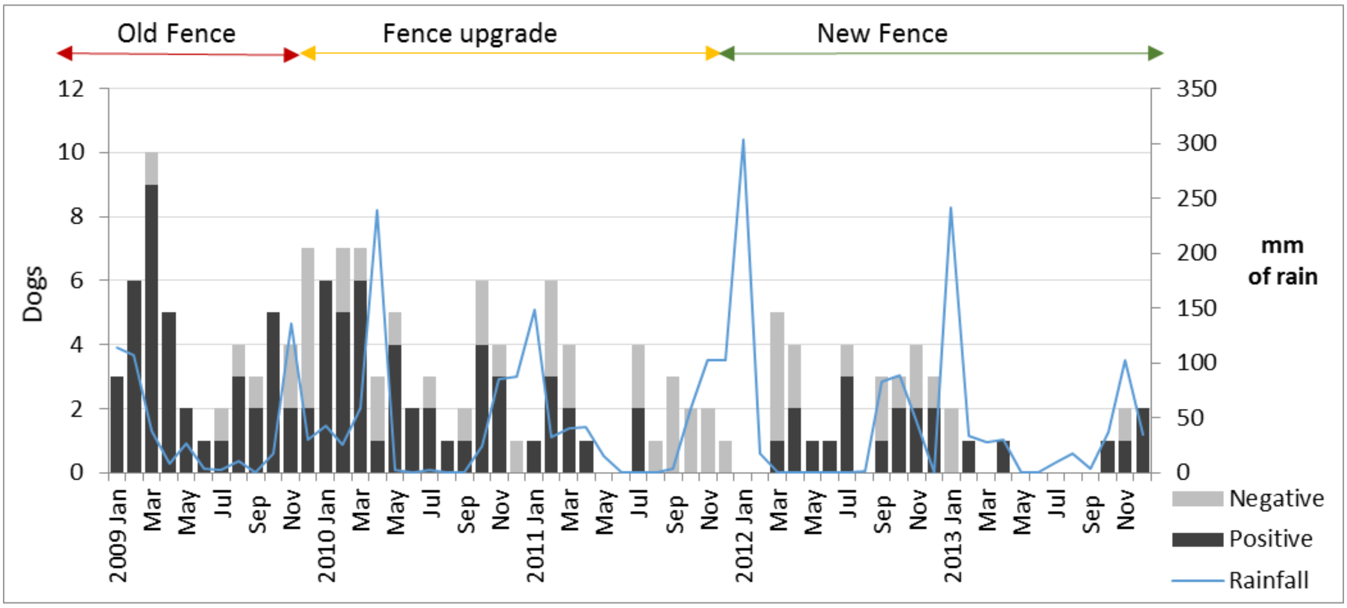

Figure 6. Number of dogs shot by month within the study site showing rabies test results, January 2009 - December 2013. The blue line and the secondary axis shows monthly rainfall totals. Indicators at the top of the graph show the status of the fence.

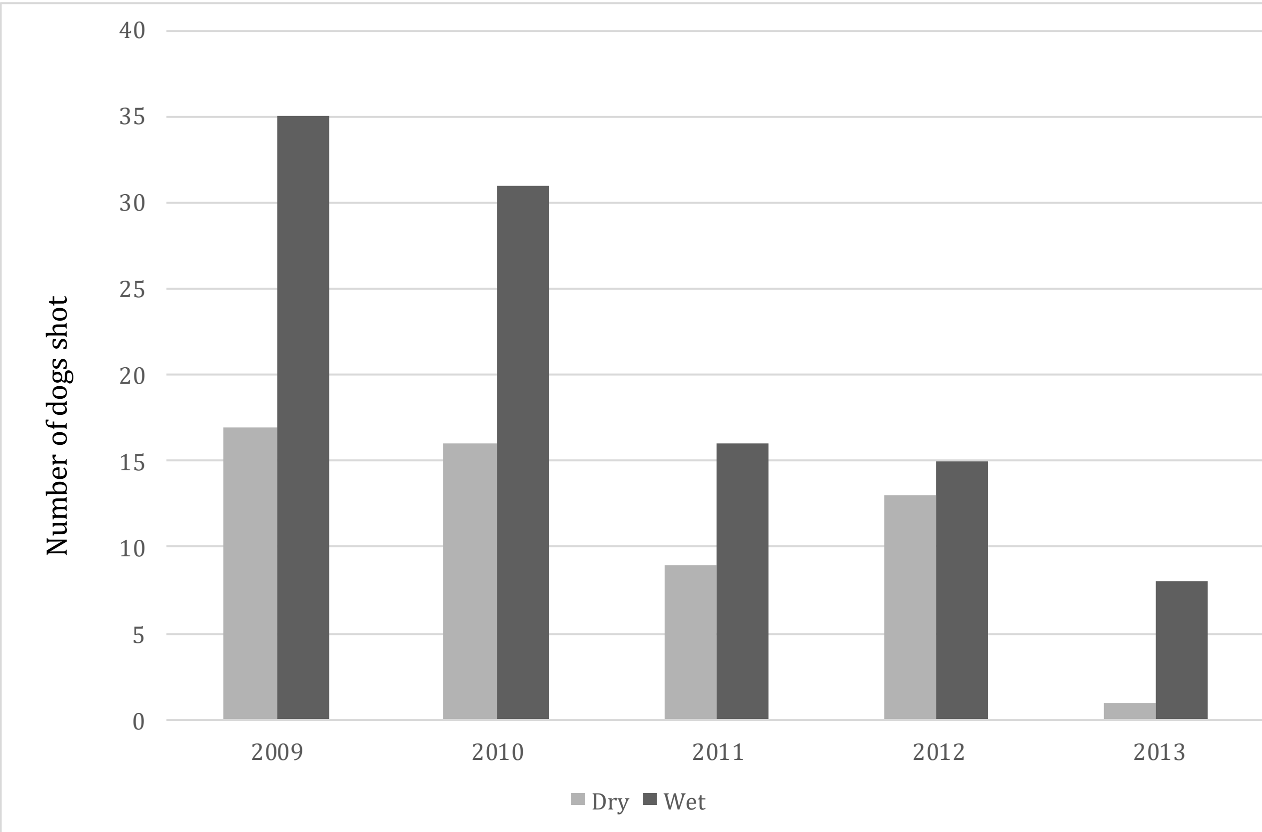

Figure 7. Total of dogs shot per year per season (wet vs. dry)

Figure 7 shows a plot of the number of dogs shot by season per year. The number of dogs shot in the wet season was significantly higher than in the dry season across the five years of the study

(Wilcoxon rank sum test with continuity correction, W = 279, p-value = 0.011) and overall the numbers declined over time except for a small increase in the 2012 dry season.

A fence upgrade took place over the study period, starting in November 2009 and concluding at the end of November 2011. Figure 8 shows the monthly average of dogs shot inside the study area per period of the fence upgrade. The old fence period of 11 months had 45 dogs at an average 4.09 dogs per month (standard deviation [s.d.] =2.47, 87% positive for rabies), during the upgrade 78 dogs were shot over 24 months at an average of 3.25 (s.d. = 2.31, 59% positive) and after the upgrade was completed 47 dogs were shot over 28 months at an average of 1.68 (s.d. = 1.66, 55% positive for rabies).

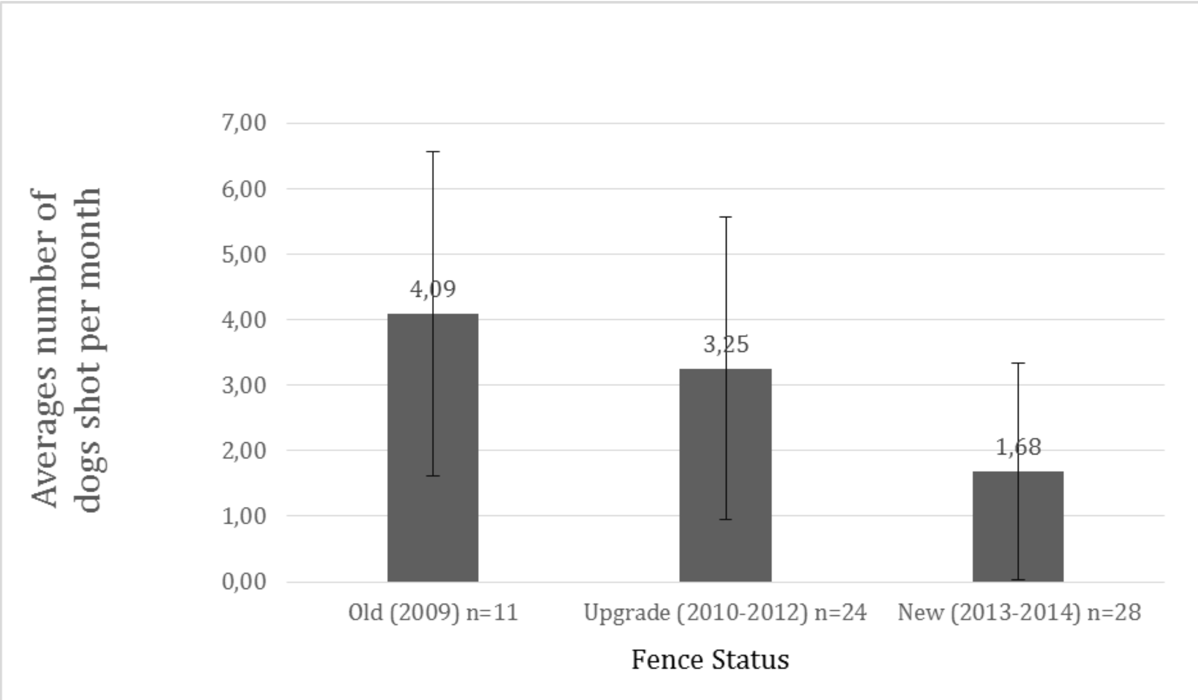

Figure 8. Bar graph for monthly mean numbers of dogs shot per time unit of the fence status. Vertical lines show the standard deviation.

To investigate the seasonal relationship further, a graph with dogs shot by month indicated by box plots is presented in Figure 9. Wet season months (October - March) show a higher inter-quartile range of dogs shot showing more variation and dry season (April - September) show lower median and less inter-quartile range variation.

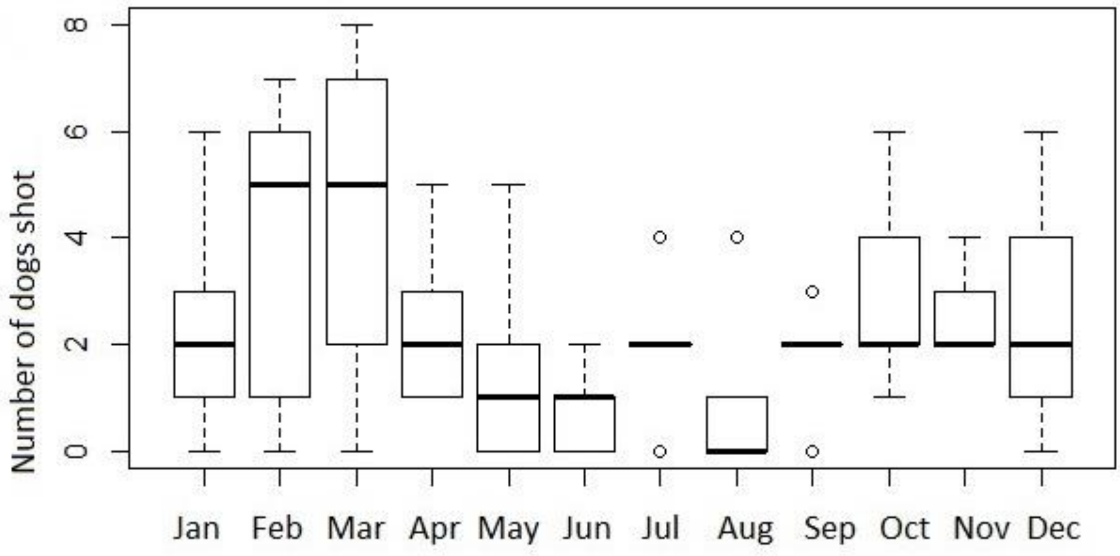

Figure 9. Time series box plots of number of dogs shot by month

Figure 10 shows plots of the autocorrelation of time series data for dogs shot and rainfall by month. These show individual autocorrelation of (a) dogs shot and (b) rainfall. For (a) dogs shot, the residuals shows positive lags following each other over 2-3 months with the first 3 lags above the dotted line indicating significant correlation or similarity to points close to each other. Monthly rainfall (b) is variable with both positive and negative lags and seasonal. The first positive lag time is above the significant bars and a few negative lags below the significance line, which are the dry month, indicating more correlation between rainfall observations. This correlation also shows the seasonality of rainfall (6 month variation). If we consider that the dogs shot times series was over the exact same time period if there was seasonality in dogs shot we would expect it to be similar to rainfall. Although there was correlation there is little significant seasonality in dogs shot.

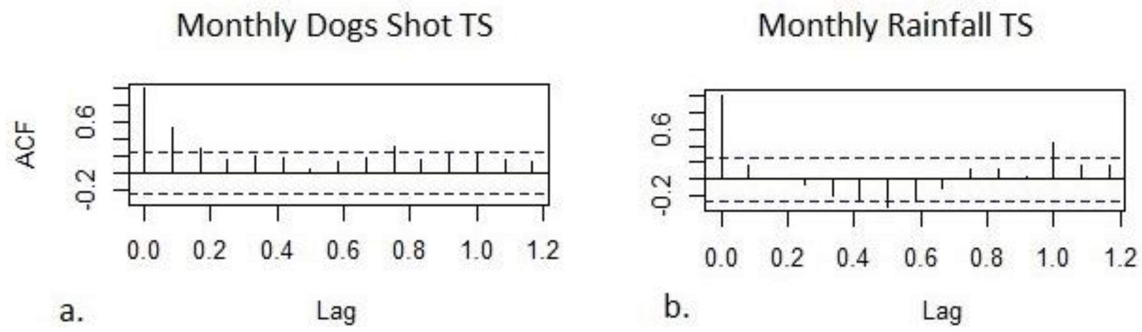

Figure 10 Autocorrelation for lag plotted for time series of monthly dogs shot and monthly rainfall independently, with 95% confidence levels indicated by dashed lines.

Therefore to test for seasonality a seasonal decomposition analysis of the time series in R was done and was plotted in Figure 11. The three components from the algorithm are plotted below the observed time series. Graph (a) this is actual data plotted over time and is used as a reference, (b) the trend data over the time series, observing the fluctuations that are repeated by relating them to time, (c) the seasonal data from the time series, plotted as a seasonal pattern, showing what the data would look like if it was recurrently seasonal. Graph (d) is random data (white noise) from time series model, plotted to show no seasonal variation. The trend data (b) shows a general decrease in dogs shot from 2009-2014. Comparing the observed data (a) to that of modelled seasonal (c) and modelled random (d), it appears that there is a slight seasonal pattern initially in 2009 and 2010 but this is not maintained over the five years, becoming more random after 2011.

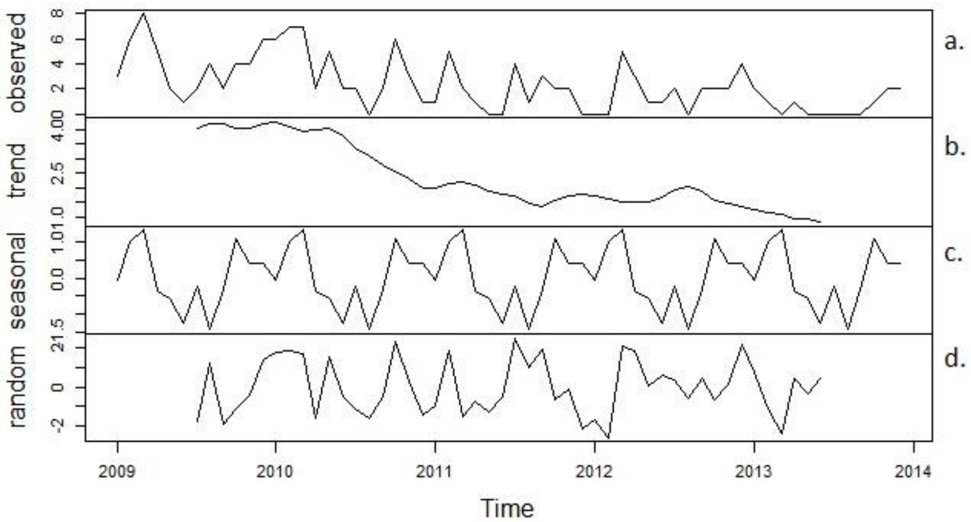

Figure 11. Decomposition of time series for dogs shot monthly from 2009-2013, (a) actual observed times series plotted (b) generalised trend data over time (c) seasonal modelled data of time series (d) modelled random time series data.

3.4 Spatial analysis

Table 4 shows the correlation coefficient for geographic variables included in the analysis; figures >0.75 are highlighted in red. Vulnerable points (points that are most likely to be breached by dogs) have a correlation to the fence as they are situated on the fence where possible dog incursions may happen; these places where dog incursions may occur are also related to minor rivers as most minor rivers passing under fence create gaps for dogs to enter. Villages are situated outside along the fence line and therefore have a correlation to both fence and vulnerable points. Minor rivers are correlated to the distance from vulnerable points, the fence and villages. Minor rivers are widely distributed throughout the landscape and therefore are correlated to other spatial factors that are widely distributed throughout the study area. A correlation between minor rivers and the gates is interesting as although statistically they correlate there is no logical explanation for this correlation. River refers to perennial rivers that contain water for majority of the year. There are very few of these rivers within the study area and most flow in the central part of the reserve far away from the fence and vulnerable points, thus a negative correlation is found.

Table 4. Correlation coefficients of distances between geographic factors in the study site

	Access	Camp	Fence	gates	Pickets	minorRiver	River	village	vulnerable	Waterpoint
Access	**1**									
Camp	0.68	**1.00**								
Fence	0.16	-0.04	**1.00**							
gates	0.21	0.12	0.76	**1.00**						
Pickets	-0.03	-0.04	-0.05	0.05	**1.00**					
minorRiver	0.19	0.10	0.78	0.95	-0.14	**1.00**				
River	-0.26	-0.07	-0.54	-0.48	-0.01	-0.47	**1.00**			
villages	0.05	-0.07	0.92	0.72	-0.06	0.75	-0.47	**1.00**		
vulnerable	0.16	-0.04	1.00	0.76	-0.04	0.78	-0.53	0.92	**1.00**	
Water points	0.36	0.33	0.27	0.15	-0.02	0.14	-0.15	0.22	0.26	**1.00**

Table 5 shows the univariate results of the bootstrap generalized linear model of spatial features associated with the location of shot dogs. Camps was not significant and therefore was excluded from the multivariable GLM. All other individual predictors were significant at $p < 0.05$ and were run in the model. Results of the multivariable GLM are shown in Table 6. Dogs were significantly more likely to be shot further away from pickets and closer to minor rivers. There was a significant interaction between these two factors ($p < 0.0001$) for the bootstrap data.

Table 5. Univariate GLM results for the bootstrap approach for random dog locations indicating factors related to dogs shot (n=10150)

Predictor Distance to:		Estimate	Std Error	p value	Significance
Access roads to camps	Log Distance (km)	-0.24168	0.09209	<0.01	**
Camps	Log Distance (km)	-0.07986	0.08347	0.34	
Fence	Log Distance (km)	-0.59761	0.05631	<0.001	***
Manned entrance Gates	Log Distance (km)	0.04974	0.02358	0.03	*
minor Rivers order 1-4	Log Distance (km)	-6.1854	0.5578	<0.001	***
Security Pickets (accommodation)	Log Distance (km)	4.2073	0.3501	<0.001	***
major Rivers	Log Distance (km)	0.24284	0.03427	<0.001	***
Villages outside study site	Log Distance (km)	-0.53199	0.03985	<0.001	***
Vulnerable points on fence	Log Distance (km)	-0.56302	0.05424	<0.001	***
Water points	Log Distance (km)	-0.5503	0.2246	0.01	*

Table 6. Multivariable binomial GLM for dogs samples and the distance to spatial features as well as the interaction of the factors with each other

Predictor	Unit	Estimate	Std error	p value	OR (95% CI's)
(Intercept)		-0.2172	0.2791	0.44	
Pickets	Log Distance (km)	1.6923	0.3043	<0.001	5.432 (2.992 - 9.862)
minorRiver	Log Distance (km)	-4.8293	0.5999	<0.001	0.008 (0.002 - 0.026)

Interaction	Estimate	Std error	p value	Significance
Pickets:minorRiver	-0.0205156	0.000926	<0.001	***

This interaction is plotted as a smoothed scatter plot in Figure 12. In the bootstrap, control data, the interaction is driven by a large group at 4-5 km from the pickets, but close to the minor rivers between 0-0.2 km. In interaction between the factors for the cases of actual dogs shot shows a large grouping 0-0.1 km from pickets and 1-1.5 km from minor rivers. The combination of the two datasets of control and cases is combined into one in the final graphic in Figure 12. This final graphic shows the interaction between pickets and minor rivers is different from controls to cases.

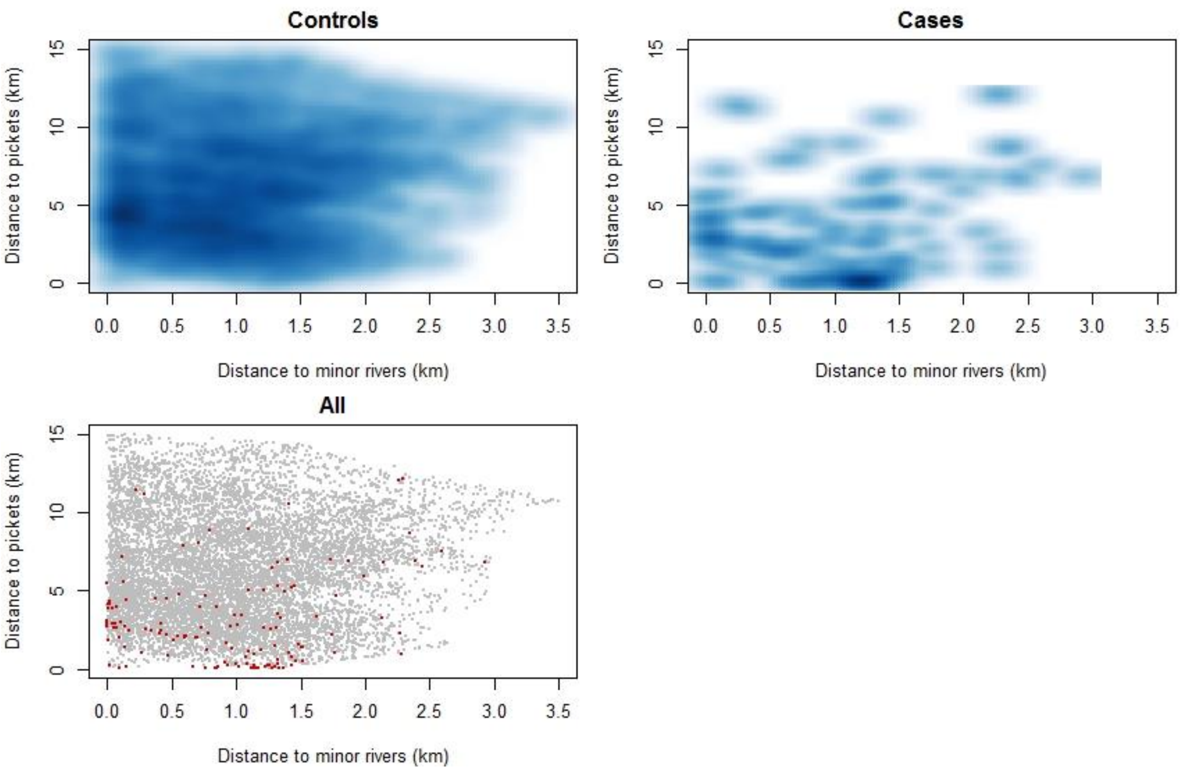

Figure 12. Interaction scatter plots of distance to pickets and distance to minor rivers, firstly for the control of bootstrapped data for dogs shot, next the cases of actual dogs shot and then combined of all data with control in grey and cases in red.

Table 7 shows the results of the univariate analyses between rabies test results and geographical features. These factors are influenced by the occurrence of rabies positive result. Access, Camp, Fence, Villages, Pickets, Vulnerable and Water point distances were all significantly associated with rabies positive test results and were therefore included in the multivariable GLM. Table 8 shows the results of the multivariable analysis with distance to water and distance to villages as the significant factors associated with rabies positive results. All combinations of different factors were tried using a stepAIC to test the rigorousness of the step-wise backward elimination and villages and water are always the two significant factors. Dogs that were shot further from villages and closer to water were more likely to test positive for rabies.

Table 7 Univariate GLM of distance to spatial features and rabies test results (n = 150)

Predictor	Unit	Estimate	Std error	p value	Significance
Access	Log Distance (km)	-0.375	0.118	<0.01	**
Camp	Log Distance (km)	-0.277	0.09532	<0.01	**
Fence	Log Distance (km)	0.519	0.1715	<0.01	**
Gates	Log Distance (km)	-0.0212	0.03263	0.52	
River	Log Distance (km)	0.0054	0.00446	0.90	
MinorRiver	Log Distance (km)	0.1873	0.2698	0.49	
Pickets	Log Distance (km)	0.2488	0.0811	<0.01	**
Villages	Log Distance (km)	0.3527	0.0934	<0.01	**
Vulnerable	Log Distance (km)	0.4243	0.1452	<0.001	***
Water points	Log Distance (km)	-0.844	0.316	<0.01	**

Table 8 Multivariable GLM of distance to spatial features and rabies test results (n = 150)

Predictor	Unit	Estimate	Std error	p value	OR (95% CI's)
(Intercept)		0.16411	0.37512	0.661767	
Villages	Log Distance (km)	0.3513	0.09332	<0.001***	1.421 (1.182- 1.709)
Water points	Log Distance (km)	-0.89168	0.3433	<0.01**	0.410 (0.208 - 0.808)

*OR - odds ratio; CI - confidence interval

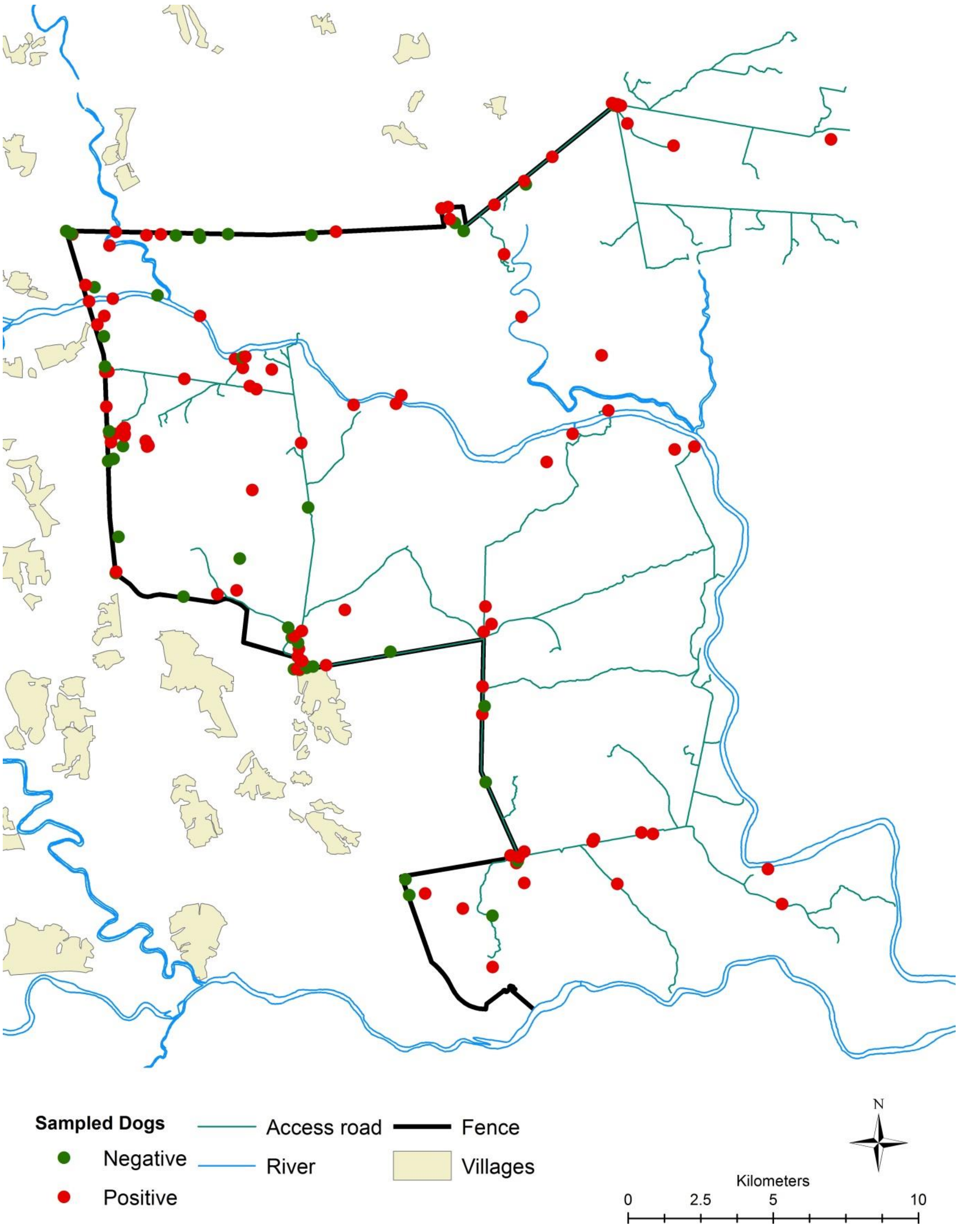

Figure 13. Map of spatial features and rabies test results for dogs shot.

Figure 13 shows the higher number of red points, indicating rabies positive dogs shot, in the interior region of the reserve compared to the many green points, indicating rabies negative shot on the fence line. However, visually the number of rabies positive dogs is also high along the fence line. The previous multivariable GLM for rabies results did not present the fence as a factor associated with rabies results as it had many influencing factors. Statistics showing the distance to the fence for rabies positive and negative differed significantly for the two groups of dogs (rabies positive vs. rabies negative; Wilcoxon rank sum test, W = 3311, p = 0.0013). The sample bias due to regular patrols on the fence is most likely the reason for the high number of dogs shot close to the fence. However we cannot neglect the fact that the fence does have a statistically significant association to rabies results; therefore we did the univariate GLM, with distance to fence as the only explanatory variable. This showed a significant association between the fence and rabies test result: for every 1 km further away from the fence the odds of a dog testing rabies positive increases by 1.68 (95% confidence intervals 1.20 – 2.36, p = 0.002).

To select data that is not affected by the view shed of the firebreak and the fence patrol bias we use a histogram, Figure 14, showing the number of dogs shot per 50m, for the range 0 – 1000m. The highest three values of all dogs shot are within the first 3 bins (0-150m) and this decreases with distance from the fence. Using the cumulative percentage of total dogs shot the graph rises steeply from 0-0.2 km and then begins to level off at the 50% mark. This 0.2 km mark is significant in the GLM models if one looks to remove the influence of the high surveillance effect of the fence on the number of dogs shot and their rabies result.

To further examine the threshold effect of the distance from the fence a Pearson's Chi-square test was performed, using 0.1 km and 0.2 km as thresholds. The 0.2 km threshold was significantly associated with rabies test results (X^2 = 12.8, p = 0.003), with more dogs positive further from the fence, while a similar effect was seen with the 1 km threshold (X^2 = 4.2, p = 0.04).

Therefore a subset analysis was performed to examine the association between rabies test results and distance from the fence, using only those samples further than 0.2 km from the fence. Table 9 shows the univariate test results, where Fence, Villages and Vulnerable points were all significantly associated with rabies result. The subset multivariable model was run with the above three factors. In the final model, only Village remained as a significant factor, with a positive association between distance from a village and a positive rabies test result (odds ratio 1.58, 95% confidence intervals 1.18-2.32, p = 0.007).

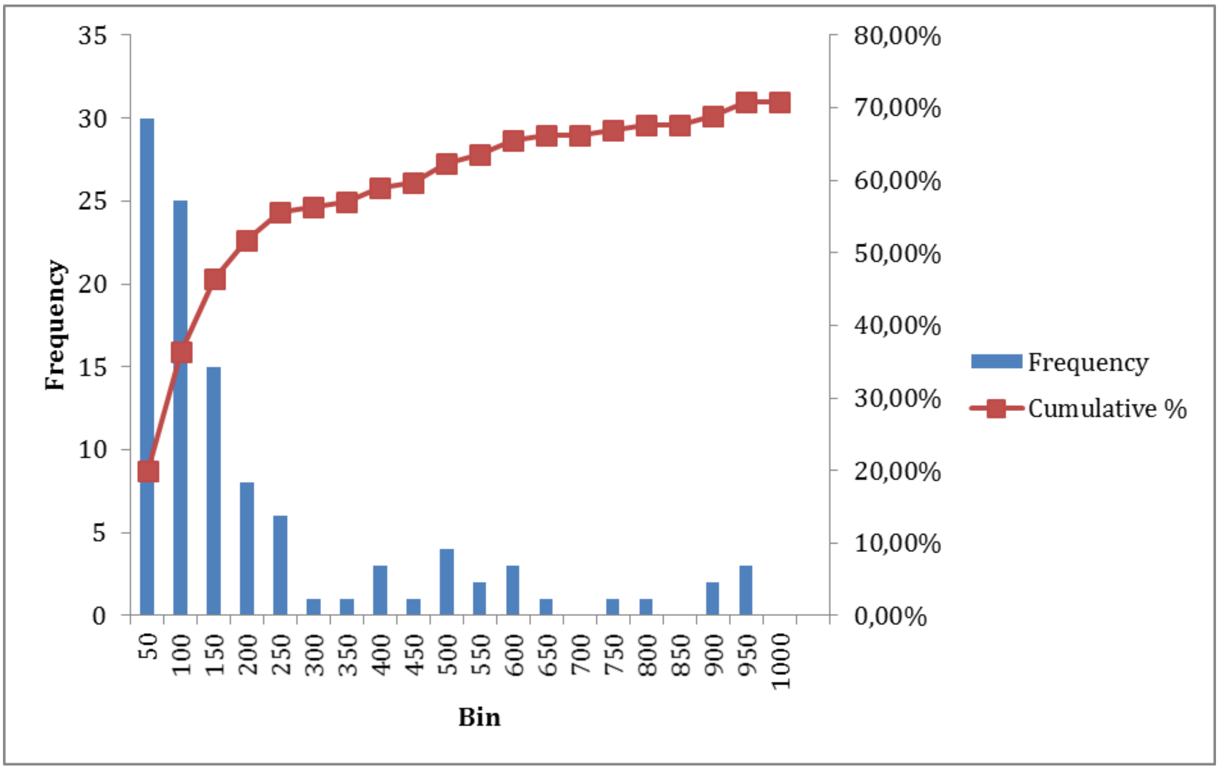

Figure 14. Histogram of dogs shot in bin of 50m increments from 0-1000m. The cumulative count is plotted as a red line graph showing percentage on the secondary axis.

Table 9. Univariate GLM of distance to spatial features and rabies test results, for dogs shot >0.2 km from the fence

Predictor	Unit	Estimate	Std error	z value	p value	Significance
Access	Log Distance	-0.2849	0.3139	-0.908	0.364	
Camp	Log Distance	-0.07951	0.22496	-0.353	0.723752	
Fence	Log Distance	0.3868	0.1897	2.039	0.0415	*
Gates	Log Distance	0.05085	0.0752	0.676	0.499	
Pickets	Log Distance	0.2009	0.1321	1.521	0.128	
Major River	Log Distance	0.009998	0.071492	0.14	0.8888	
MinorRiver	Log Distance	0.4987	0.4451	1.121	0.2625	
Villages	Log Distance	0.4562	0.1693	2.695	0.00704	**
Vulnerable	Log Distance	0.3848	0.1906	2.019	0.0435	*
Water points	Log Distance	0.4239	0.9336	0.454	0.6498	

3.5 Predator home range analysis

The kernel density of home ranges for all recorded lion sightings for the 19-month period gives a 90% estimation of home ranges for nine regularly-sighted lion prides in the study area (Figure 15). The overlap of different pride home ranges is due to the long time period and also the use of a high percentage home range of 90% unlike the commonly used 50% core home range. The reason for using this larger home range is to identify the extremes of the lion prides home range and thus the likelihood of interaction with a dog. Figure 16 indicates four prides (Othawa, Nkahuma, Southern, Ximungwe) with more than 10 rabies-positive dogs shot over the period within their 90% home range, with the highest being 39 dogs (22 positive) for the Ximungwe pride. This pride's home range lies on the western boundary fence of the reserve (Figure 15).

Density of dogs shot indicates areas of highest historical dog locations (Figure 17), which can act as a heat map for probable dog incursions in the future. The highest density shown by the dark colour is between 0.47-0.82 dogs found per 5 km radius. Hyaena den sites located on the heat map allow for quick reference probability of possible interaction between dogs and hyaenas.

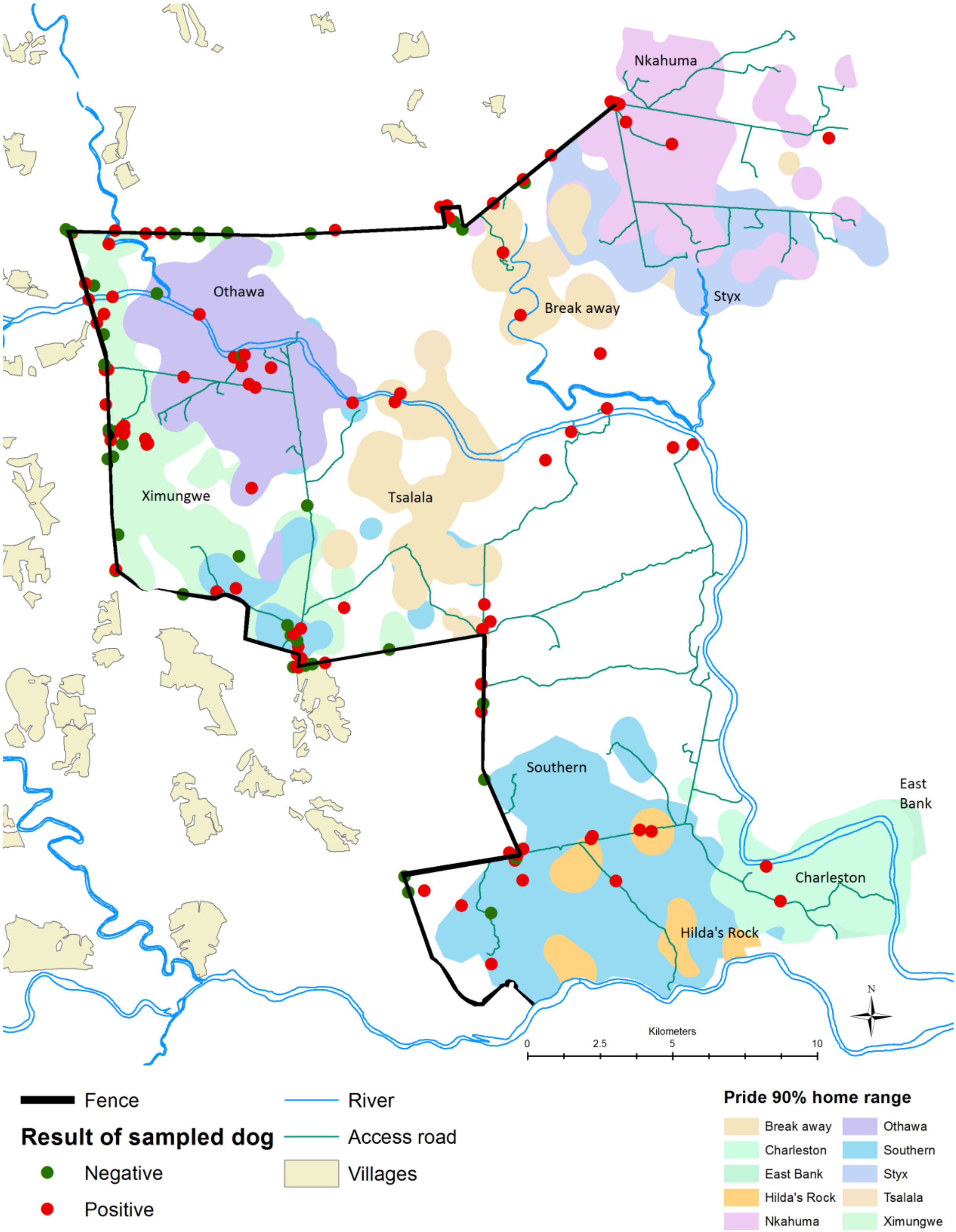

Figure 15 Kernel density 90% home range of lion prides in the study area, over laid with sampled dogs

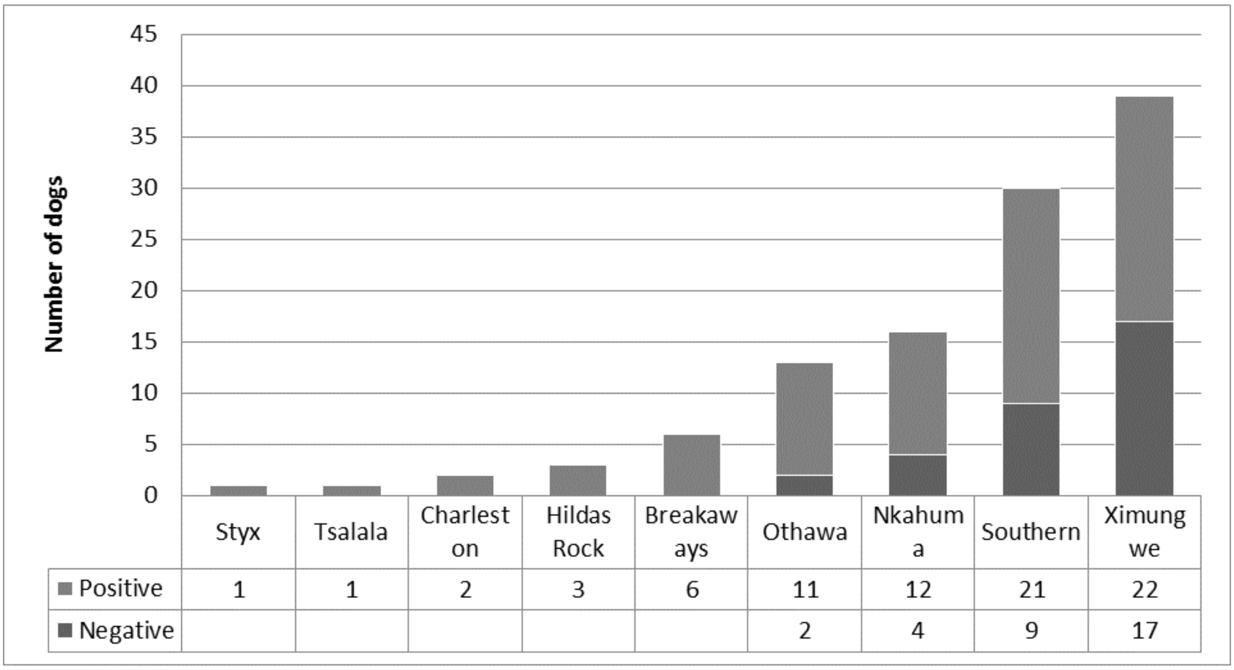

Figure 16. Number of dogs shot and rabies test result within the 90% home range of lion prides

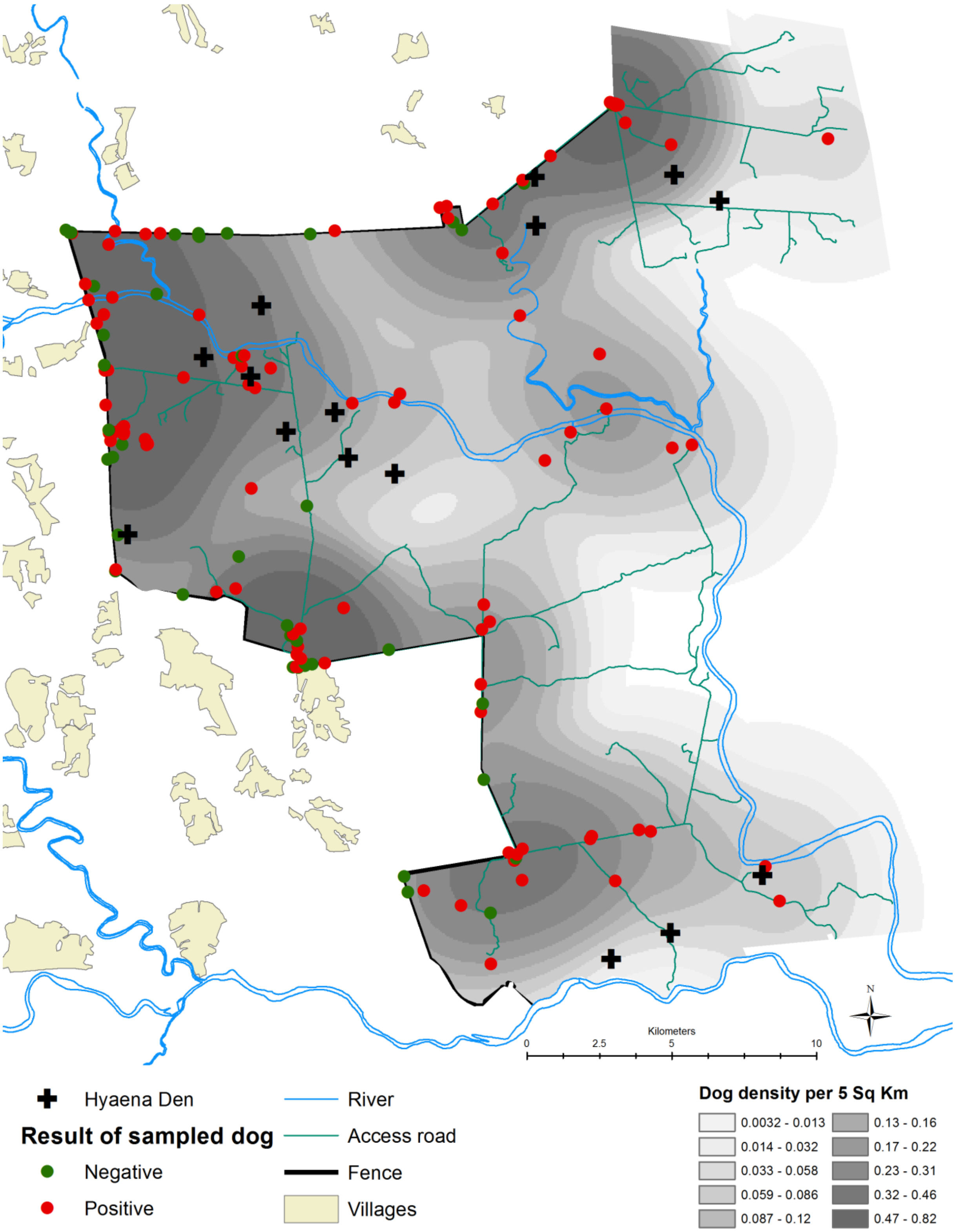

Figure 17. Density per 5 Km radius of dogs shot overlaid with dog results and hyaena dens sites

4. Discussion

The high number of 236 dogs sampled in the broader area with 59.7% positive for rabies raises concerns, not only for the reserve itself. Of the 170 samples taken in the study site itself, the percentage positive for rabies (65%) was significantly higher than in those samples taken outside. Disease monitoring by sampling animals that have not died from predation in the study area is highly effective due to the large number of commercial game drive vehicles across the reserve and the efficient communication channels with reserve management. With only one sample out of the 90 wild animals sampled between 2009 and 2014, returning a positive result for rabies, we can suggest that the virus has not established itself in wildlife in the study area; however a wildlife estate 50km north-east of the study site had three confirmed rabid jackals and four confirmed rabid wild dogs in August 2015 (personal communication, G Beverly EWT). This indicates that even though the highest number of dogs recorded in 2009 and 2010 coincided with an increase of rabies in the surrounding communities the disease is still a threat in the area surrounding the study reserve. The upgrade of the fence and an increased effort by reserve management of better fence maintenance and surveillance due to the increase in poaching activities in the area may have contributed to the decrease in stray dogs found in the reserve, however the number of dogs recorded as well as the percentage of rabies positive dogs in the study site is exceptionally high. This may be as a result of higher surveillance efforts and better resources in this private reserve compared to government reserves and outside areas that are lacking in resources.

Dog movement distances of 25 km and 35 km were recorded in Zimbabwe communal lands (Foggin, 1988). Butler et al. (2004) recorded shorter distances of dog movements as far as only 6 km into wildlife areas in Zimbabwe, however many of the long distances travelled by collared dogs were part of a route they had previously followed, often while accompanying people. The furthest distance into the wildlife area of an independent dog was 3 km (Butler, 1998). While the movement of people with dogs inside the study area is restricted due to highly effective anti-poaching units, the movement of dogs on the periphery is extensive. On many occasions these dogs are part of a hunting pack, as seen by photographic evidence and communication with the community. This communication with community members in 2012 raised the point that community members may use the reserve as a way of disposing of sick or dogs perceived to be unhealthy that may carry disease, “throwing dogs over the fence so they do not run in the community” (personal communication, Dixie Community member, 2012). Human interaction with dogs displaying signs of rabies was not observed in this study but through personal communication with cattle farmers on the reserve boundary it was reported that aggressive dogs may be chased out of villages or even thrown over the fence into wildlife areas (personal communication, Utah cattle farmers 2014). Those owners that use dogs for hunting showed reluctance for dogs to receive vaccinations or sterilization as it may affect their hunting abilities. These discussions were very much anecdotal and should be explored further with social projects in the community, but may explain the relatively high proportion of rabies-positive dogs in the reserve, compared to surrounding areas.

The increased patrol times in 2011 of two 24-hour patrols each covering the perimeter for anti-poaching (personal communication, Reserve management 2012) compliments the observation made regarding the fire breaks along the perimeter fence allowing for better visual patrolling as on the fire break (burnt area), grass volume is minimal for a large majority of the year and due to the frequency of the burning the fire breaks have very little bushy vegetation. Therefore the visibility and detection of a dog running on the fire break is increased. Using a visual representation of rabies result (Figure 17) one can see not only the high number of dogs shot close to the fence, due to detection, but also the high number of rabies positive cases further into the reserve.

The contribution of the fence upgrade to the decline in the number of dogs shot over time was not tested statistically in this study and needs to be explored further for this area. Even after completion of the entire fence in 2011, dogs still got into the reserve, therefore there is still concern of dog and wildlife interaction regardless of the quality of the fence. Further research can be done on the cost benefit analysis if prevention of dogs is the sole reason for the upgrade of the fence.

Sampled dogs on the outside of the reserve stayed consistent in 2010, 2011 and 2012 (24, 16, 20) but then dropped steeply to only 2 dogs sampled outside in 2013. Sample effort for the outside could not be recorded and therefore we cannot be sure this is due to a decrease in rabies in the area, however it is consistent with the drop in numbers found within the reserve. The statistically significant temporal factor was peak rainfall season only in the first two years. 2011 onward showed little seasonal variation in dog numbers. The actual rainfall events surprisingly don't influence the number of dogs as much as thought at the outset of the project therefore it may be to do with the availability of food as seen in Zimbabwe by Butler (1998), when the dog crossing into wildlife areas increased dramatically after a failed wet season or drought.

Experience from working on the fences suggests that heavy rainfall events would cause washing away of soil along the fence line. These washed-away areas would allow easier access for the dogs into the reserve under the fence. For this reason the vulnerable points were located on the fence using a GPS point and used as a variable in the models, however these were found not to be statistically significant to either the presence of a dog or to the rabies result. The data suggests that the wet summer rainfall period does initially show a higher number of dogs but this is not spread across the season and does not follow high rainfall events or even seasonality. During the wet season the fire-break grass regrows and vegetation thickens up, and while the area is still less dense than other bushveld, detection of dogs becomes more difficult as the wet season goes on. This may explain the drop in numbers during the latter part of the wet season and therefore suggest that dogs still enter the reserve in high numbers but are less likely to be detected. Temporal correlations should be investigated further to better understand the seasonality of the data as well as the effect of sample bias during periods of better visibility.

From the bootstrap model scatter plot the association between a dog and the distance to a picket showed most dogs further away from pickets (4-5 kms). Although pickets are placed along the fence for detection there are some pickets scattered across the reserve and therefore this large distance from most dogs makes sense. Minor rivers however are found throughout the reserve and dogs are more likely to be found closer to minor rivers. This is opposite for actual cases of dogs shot which showed a grouping at 0-0.1 km from pickets and 1-1.5 km from minor rivers. This could be related to dogs have higher detection rates due to the increased time spent there by field rangers and reserve staff or it may be symptomatic of rabies infected dogs. This highlights the need for further research to be done the sample effort versus the detection of dogs.

Some spatial factors are more prevalent than others. Those located along the fence line such as pickets, vulnerable points and to a lesser extent villages show a degree of collinearity. Spatial factors such as camps and to a lesser degree access roads are found mostly in the interior of the reserve and did not show any significant influences on dogs. Perennial rivers (labeled Rivers) run through the centre of the reserve but results indicate that they too play no role in the presence of dogs or in the rabies result.

Rabies-positive dogs show a tendency to be shot closer to water points. The reason for this could be a multiple number of factors, from disease-related effect on the dog's condition or possible higher surveillance efforts at water points. The association of the positive result with water points may be as a result of the dog looking to quench its thirst after long distances of travel. Although it cannot be explained it should not be ignored. The distance the dog travels from the village is directly linked to the rabies positive results and is a key finding for this research. Rabid dogs may "wander", leaving the community and coming into the reserve, or they may be chased from villages. Rabid dogs may be disorientated and cover vast distances for no apparent reason. This is supported by data collected by Butler (1998) which suggested that rabies dogs were not affected by geographic factors such as fences or dry river beds. Butler noted 10 of 12 adult dogs moved away from their homesteads when infected with rabies and this could be a similar avoidance of humans (Butler 1998). Statistics indicate that although the fence is significantly linked to rabies result, this may be a result of sample bias as the fence is patrolled more frequently than other roads. The removal of the element of sample bias from high detection zones statistics were done on all points greater than 0.2 km from the fence. This analysis indicates the distance from villages as the main influence on rabies positive results. Findings also highlight the urgency that is needed if a dog if a dog is found deeper into the reserve, as the likelihood of it having rabies is higher. The opportunity to contact more wildlife also increases away from the well-patrolled fence line.

The results from the presence data supported the probability of dogs being close to drainage lines (minor rivers) however when the rabies result is used in the model minor rivers are no longer significant, suggesting that rabies positive dogs begin to act irregularly. This falls in line with the distance from villages, as all dogs would have started the journey from a village. Those dogs not infected with rabies would attempt to return to the village or stay in proximity to an area they know especially due to the reliance of dogs on humans presence for scavenging of food and inability to be a successful hunter outside of a pack (Butler, 1998). Butler, du Toit & Bingham (2004) suggest that dogs follow a pattern or known route, however when affected by rabies this may cause them to wonder off that known route and into the reserve where they have not been before.

Although competition for prey, hybridization and harassment of predators is highlighted in the literature review of interaction of dogs with wildlife, the key interaction for wildlife managers is the spread of disease between dogs and predators. As pointed out by Hofmeyr et al. (2000) and Mills (1993), rabies has the ability to destroy local communities of predators and therefore is the focus of this study. The high prevalence of rabies positive results is a concern to management even though only one wildlife sample has returned a positive rabies result. To date there have been no positive cases of predators contracting rabies in the study area; however the distribution of rabies on the map from the Department of Agriculture, Forestry and Fisheries (Figure 1) indicates the distribution of canine and jackal rabies surrounding the study area. Therefore the need to prevent this spread into wildlife is important. The study area has a low density of jackals and with jackals being monogamous (Mckenzie, 1993), the social interaction is limited when densities are low; this limits the ability of jackal to become a host species or maintain rabies.

The detection of dogs relies on the reporting of dogs to management by rangers and can be a limiting factor as reports may often be delayed or not sent and therefore dogs could go undetected in the reserve. Searching for the dogs is done predominantly using a vehicle and locations of a dog is therefore more likely to be along main roads and busy areas. Insufficient data were collected on dogs reported and not shot and is therefore excluded. Sampling efforts outside of the reserve are limited due to lack of resources and therefore the data is not directly comparable to outside the reserve. Many ecological influences that cannot be quantified spatially will also affect the movement of dogs. Therefore this study is primarily focused on spatial/environmental factors that can be quantified.

While the results from the study are not inclusive of all possible factors (as the measurement of all environmental factors that may influence dog ecology is not possible), they do give management a good indication of the following protocol that can be put in place to limit the incursion of dogs, rabid dogs in particular. Dogs found further into the reserve have a higher likelihood of being rabies positive and therefore require more effort in removal. The association of rabies positive dogs with waterholes increases the ability of management to search for dogs or dog tracks in the right place and once receiving reports of a dog they can start following up at waterholes in the immediate area. All dogs have an association to minor rivers (dry river beds) and this to can direct management to search in river beds for tracks of the dog gaining the direction of movement.

Using updated data sets management can overlay lion home ranges and identify prides at highest risk and vaccinate them accordingly. Additionally using the density map of previous dog incursions, stationary predator locations such as hyaena dens, territorial jackal pairs and wild dog dens can be included in the vaccination and monitoring program. Vaccination of predators in conservation areas should not be done in isolation but rather simultaneously with vaccination efforts of the reservoir hosts, free-roaming dogs, in the local community. The question of why rabies in dogs persists can be answered by the lack of resources as well as lack of participation by the community in vaccination programs (Akerele, 2014; Butler, 1998). As mentioned in the literature review Hampson et al (2009) indicated that 70% coverage is sufficient to ensure long-term elimination of rabies in a host population. The reduction in rabies cases after 2011 may be a combined result of better fencing and an increase in vaccination programs. Personal communication with the Mpumalanga State Vet (B Rheininghaus, 2014) indicated that when the study started in 2009 the organisation and resources for vaccination campaigns was limited, but since that time efforts have increased. Vaccination campaigns in surrounding Mnisi area achieved a 68.2% coverage in 2011 and 59% in 2013 (Akerele, 2014). Konick (2014) highlighted the variability of campaign success due to additional workloads that state health technicians face such as disease outbreaks especially foot-and-mouth disease. Further steps should be followed for education of managers of conservation areas to fully understand the current status of rabies both within and outside reserves and to enable them to assist in vaccination programs to ensure the 70% coverage is maintained.

Additionally the education should include modeling and predictive analysis of implications rabies may have in wildlife areas. Conservation areas can play their part outside of the reserves by assisting veterinary departments in the vaccination campaigns, the conservation areas will be addressing the problem proactively rather than reactively. This too will reduce the spread of rabies further into the wildlife areas north of the study site.

In conclusion, the important factors for management is the fact that the further into the reserve a dog is found the more likely it is to be rabies positive. When searching for these dogs, once reported, management should check water points and minor drainage lines or dry river beds. There is a temporal pattern to the influx of dogs, during the wet season the number of dogs is higher than in the dry season. However the effect of rainfall events should be explored in future studies. Additional analysis on other ecological factors can be investigated especially looking into the effects of rabies on behavior and the relation to ecological factors. A limitation to this study is the inconsistent collection of data outside the study area. A further limitation is the lack of data for dogs reported in the study area but not shot which would give a better indication of dog incursions. This data would allow for a better understanding of seasonal detectability of dogs due to the increased vegetation growth in the latter part of the wet season and is an opportunity for further research. Despite the above-mentioned limitations in data collection and needs for further research in the surrounding communities, other reserves and farms, the study provides the baseline for comparison in areas where rabies prevalence is increasing or management is looking for proactive ways to combat domestic dog incursions and more importantly the interaction of those dogs with wildlife.

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6. Appendix

UNIVERSITEIT VAN PRETORIA
UNIVERSITY OF PRETORIA
YUNIBESITHI YA PRETORIA

Animal Ethics Committee

PROJECT TITLE	**Spatial analysis of dog ecology and rabies epidemiology at a wildlife interface in the Southern African Lowveld**
PROJECT NUMBER	**V002-14**
RESEARCHER/PRINCIPAL INVESTIGATOR	**Mr. M Grover**

STUDENT NUMBER (where applicable)	**1341 8875**
DISSERTATION/THESIS SUBMITTED FOR	**MSc**

ANIMAL SPECIES	**Canine**	
NUMBER OF ANIMALS	**Data already collected**	
Approval period to use animals for research/testing purposes		**2009-December 2014**
SUPERVISOR	**Prof. D Knobel**	

KINDLY NOTE:

Should there be a change in the species or number of animal/s required, or the experimental procedure/s - please submit an amendment form to the UP Animal Ethics Committee for approval before commencing with the experiment

APPROVED	Date	5 May 2014
CHAIRMAN: UP Animal Ethics Committee	Signature	[signature]