THE EFFECTS OF ROADS ON PUFF ADDER (*BITIS ARIETANS*) MOVEMENT

BIANCA FIZZOTTI

A Dissertation submitted to the Faculty of Science, University of the Witwatersrand, Johannesburg, in fulfilment of the requirements for the degree of Master of Science.

JOHANNESBURG

3 May 2018

DECLARATION

I declare that this dissertation is my own unaided work. It is being submitted for the degree of Master of Science at the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination at any other University.

3rd day of May in the year 2018

ABSTRACT

The movement ecology of a species can dictate their survival and success. Animals are motivated to move depending on resource priorities and in turn their movements are influenced by the surrounding environment. Natural habitats are being increasingly disturbed by anthropogenic changes which often creates several implications for the existing wildlife. One of these changes include extensive road networks that may fragment landscapes and increase mortality risks for local fauna. This current and global issue is often researched with the aid of road kill statistics which can create a biased view. My study highlights a more realistic method for understanding how organisms interpret and react to road infrastructure by using the movement patterns of 109 telemetered Puff Adders (*Bitis arietans*). To assess the effect of roads on these snakes within Dinokeng Game Reserve (Gauteng, South Africa) over eight years, Puff Adder lie-up locations (to the nearest road) and movement paths were analysed. Sex, season, years, and the types of roads within the reserve were included to evaluate the motivation for movement and if roads were inhibiting or aiding movement patterns. Compared to random, Puff Adders were closer too, and crossed roads more often. Males crossed roads more frequently than females and during non-mating season, both sexes were closer to roads. Narrow roads with low levels of traffic were also crossed more often. Overall, the roads in Dinokeng Game Reserve do not inhibit movement and patterns suggest that roads may be advantageous for Puff Adders. Roads appear to be foraging hotspots because of micro habitats created along the verges. However, these positive attributes could result in a potential risk when road usage is intensified by traffic. Understanding how species interact with changes to their environment is essential for appropriate management and mitigation and this study illustrates the necessity for unbiased research to properly evaluate these changes and reactions.

In memory of Nonna & Nonno

1925 – 2017 · 1935 – 2017

ACKNOWLEDGEMENTS

I would like to thank Prof. Graham Alexander for his guidance and supervision throughout this dissertation. Your support and direction have been invaluable, and I sincerely appreciate every interaction we have had.

This dissertation would not have been possible without the hard work from both Graham Alexander and Xavier Glaudas and for that I am grateful. Thank you for sharing your work with me and allowing me to dissect and analyse what you have spent years collecting; along with Bryan Maritz who guided me with the concept of this study and set me off in the right direction.

The original research was conducted in accordance with ethical standards approved by the Animal Ethics Screening Committee of the University of the Witwatersrand (2007/68/1; 2007/69/3; 2012-42-04)

Mark Keith, Francesca Parrini, Sze-Wing Yiu, Contour Projects, Jeanette Greyvensteyn and the South African Weather Service assisted with shapefiles and data that have been used from the onset of this dissertation and have saved me hours of tedious mapping. Thank you.

Funding was provided by the University of Witwatersrand’s Post Graduate Merit Award; the National Research Foundation; the Herpetological Association of Africa and the East Texas Herpetological Society. Thank you to all of these organisations for supporting this research.

Thank you to the members of the Alexander Herp Lab that listened and gave insight and advice when discussing my project. I greatly appreciate the statistical help from Mariette Pretorius, Kelsey Glennon and Max Popescu. Without you all I would have lost my sanity in an R Studio console. A special thank you to Shivan Parusnath and Ash Miller for being my support system and encouragement throughout my scientific career.

I am greatly appreciative of my parents that have supported my studies and allowed me to change career paths. I know you would prefer it if I had studied kittens, so thank you. Lastly, thank you to Dino Fizzotti and Justin Popescu, for all of your encouragement and motivation. The two of you are my role models.

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CHAPTER ONE

CONCEPTUAL FRAMEWORK

The movement of an organism is an essential and defining aspect of its nature as its movement characteristics impact directly on survival and reproductive success (Carfagno & Weatherhead 2008; Glaudas & Alexander 2017). Mobile animals have life time tracks and in order to understand the influence of movement on their life history, these tracks can be measured at different resolutions in time and space (Nathan *et al.* 2008). An important goal of movement ecology is to fathom what motivates organisms to move and how the movement is adapted to a dynamic landscape. The internal and external processes that induce movement in organisms creates a continual consequential loop. Having the ability to alter movements to a changing landscape can impact an organism's life-track (Nathan *et al.* 2008) and resultingly their survival or reproductive success. The avoidance or use of modified landscapes by a species directly influences their space use and dispersal, and indirectly alters community dynamics, population dispersal and survival (Jeltsch *et al.* 2013).

The increase in human population and urbanisation changes and encroaches on natural habitats and the movement of organisms should be taken into consideration in conservation ecology. Land transformation is affecting species globally and very few populations will be undisturbed in the future (Sala *et al.* 2000). The general negative challenges that wildlife is faced with, because of anthropogenic changes, leads to the trend of overall biodiversity loss (Forester & Machlist 1996). A primary result of land transformation is that of fragmentation (Sanderson *et al.* 2002) which can create challenges for populations that would have historically tracked suitable habitats during periods of environmental change (Chevin *et al.* 2010). Taxa that can adapt to gradual changes are now faced with drastic and unprecedented anthropogenic changes (Sanderson *et al.* 2002) and altering their behaviour or physiology is not as feasible at current rates of change. How a species reacts to these changes (avoidance, adaptation or dispersal) directly relates to their movement patterns (Bowler & Benton 2005).

Many species adapt to changes in their environment, while others rely on continuity of habitat in a landscape to move and survive due to their size or modes of locomotion. For example, the movement of snakes is heavily influenced by accessibility to appropriate habitats (Brito 2003) that provide resources such as prey, mates and secure locations for breeding or brumation which are all important for survival (Ford & Burghart 1993). The required resources differ from species to species (Gregory *et al.* 1987) and the perception of the movement pattern depends on the resolution at which it is measured (Alexander & Maritz 2015). The movement pattern of an individual and the state of the individual (e.g. reproductive, digestive, health and social) can affect its habitat use, home range and dispersal (Reinert 1993). Studying movements of wildlife is of current importance and as technology advances, the measurement of movement has become increasingly possible (Nathan *et al.* 2008). This is especially true for snakes, which have previously been neglected due to their elusive nature (Slip &

Shine 1988). With a clearer insight into the patterns and ranges of animal movements, it becomes possible to evaluate the impact of urbanisation on populations.

The increasing need to alter natural habitats due to urbanisation is a key driver in loss of biodiversity, and many snakes have an increased extinction risk as a result (Brito 2003). Road infrastructure fragments habitats and impedes dispersal by creating physical boundaries or, in some cases, increases mortality rates due to increased human-snake interactions (Andrews & Jochimsen 2007). On the contrary, some studies have documented how roads create new microhabitats or movement corridors for organisms (Ashley & Robinson 1996; Donaldson & Bennett 2004; Andrews & Jochimsen 2007). Consequentially, the expansion of urban areas has been impactful to reptilian species (Jochimsen *et al.* 2004). However, the influence of urban development on wildlife has been little-studied in South Africa (Collinson *et al.* 2015). There are more than 420 species of reptiles in South Africa, Lesotho and Swaziland (Bates *et al.* 2014) and understanding wildlife reactions to an urbanising environment can inform appropriate conservation practices and policies, especially for reptiles, which usually receive little attention.

My study assesses the impact of roads on the movement ecology of Puff Adders (*Bitis arietans*). The Puff Adder is a common, ambush-foraging snake with a wide distribution throughout South Africa (Alexander & Marais 2007) and thus its exposure to paved and unpaved roads is extensive. Also, its stout body form means that it is relatively slow moving, making it especially vulnerable when traversing roads. Most previous studies on the effect of roads on animals focus on road kill statistics (Bernardino & Dalrymple 1992, Bonnet *et al.* 1999, Andrews & Jochimsen 2007, Langen *et al.* 2008, Collinson *et al.* 2015). My study used snakes as model organisms for understanding the effect of roads on wildlife by modelling movement patterns in relation to road networks. This is the first such study of any squamate in South Africa.

1.1 The movement ecology concept

From birth to death, most animals have a series of movement patterns which change over time (Nathan *et al.* 2008). These patterns scale up from diel movement steps through to lifetime tracks. The movement characteristics of an organism are moulded by evolution as much as any other trait that relates to survival and reproduction (Swingland & Greenwood 1983). Mobility can therefore be seen as a life trait that promotes evolutionary fitness (Hansson & Åkesson 2014).

Movement patterns are modulated by many factors (Nathan 2008). These include: the congenital instigation for movement; the capacity of the individual to engage in motion and navigate effectively; and the external environment which may impact the motivation to move (Nathan *et al.* 2008). The internal state of an organism motivates movement which can then be guided by motion and navigation (Åkesson *et al.* 2014). The resulting movement path is then influenced by both biotic and abiotic factors. This conceptual framework portrays a feedback loop between the mechanisms. New challenges are created as the surrounding environment of the organism changes with its movement. These factors influence the internal state of the organism as it moves through its surroundings, which rely on the motion and navigation capacity of the individual (Nathan *et al.* 2008).

Movement may be motivated by the need to forage (energy gain) or disperse (Jeltsch *et al.* 2013). Foraging is often the primary motivation for movement as speculated in numerous studies (Greenwood & Swingland 1983, Pyke 1983, Madsen & Shine 1996). To optimise survival through foraging, a maximum net rate of energy gain is needed. The concept that foraging to increase net rate of energy gain drives movement is not isolated from other behaviours such as predator avoidance, searching for mates, thermoregulation (Greenwood & Swingland 1983, Reinert 1993) and various extrinsic pressures (Pyke 1983). These behaviours are individual-specific and interpreted at fine resolutions, from diel patterns to seasonal patterns. Dispersal is generally interpreted at a much broader scale and is defined as movement from an origin or home range to a new area (Shields 1983). Both energy gain and dispersal are important drivers for movement that can impact an organism's success.

1.2 Snake movement

There are numerous variables that influence the movement of snakes and rarely have these been cumulatively taken into consideration in studies (Gregory *et al.* 1987, Macartney *et al.* 1988). These variables may be associated with acquisition of resources and the movement of some snakes increases with increasing energetic requirements (Carfagno & Weatherhead 2008). This is not always the case (Adams 2001), such as in ambush foraging snakes. The motivation for movement may depend on the sex, reproductive state, health, size and foraging mode of the snake (Webb & Shine 1997, Brown *et al.* 2002) as well as seasonal variations (Madsen & Shine 1996, Brito 2003).

Lifetime tracks have been shown to be strongly influenced by seasonal changes in the movement phases (Shine & Lambeck 1985, Huey 1991, Reinert 1993, Blouin-Demers & Weatherhead 2001). Slip and Shine (1988) observed the movements of 15 Diamond Pythons (*Morelia spilota spilota*) over 32 months and noted that the snakes made fewer movements in winter and that the majority of the movements during this period were motivated by basking. These individuals not only changed the length of their movement phases during the different seasons but also their habitat selection. In winter months, the pythons were found in rocky areas with less vegetation, suitable for basking, whereas in warmer months they were found closer to disturbed habitats (Slip & Shine 1988). The importance of resources is clearly revealed in this study where thermoregulatory behaviour dominates in winter and foraging behaviour in warmer months in terms of habitat selection. Male Lataste's Vipers (*Vipera latastei*) too selected habitats based on season (Brito 2003). The vipers hibernated in forests in late autumn and early spring, and then moved to open forest patches and areas of dense bush, which could aid as protection from predators as the snake is an ambush forager. Ectothermic species are especially vulnerable to the thermal conditions of the habitat in which they occur (Huey 1991) and temperate-zone snakes often choose habitats carefully as a result (Reinert

1993, Blouin-Demers & Weatherhead 2001). Movement patterns that change seasonally are consequentially linked to the seasonal change in priorities (e.g. forage, reproduction, or protection) for many taxa.

Snakes show sexual dimorphism in their behaviour (Madsen 1984, Reinert 1993) due to differences in their physiology which can influence their movement ecology. A study by Secor (1995) on Sidewinders (*Crotalus cerastes*) showed that males travelled approximately 200 m further per day than the non-gravid females. Male snakes travelling greater distances in the mating season in search of females appears to be a pattern common to many species of snakes (Brown & Parker 1976, Reinert & Zappalorti 1988, Slip & Shine 1988, Collinson *et al.* 2015, Glaudas & Alexander 2017). However, there are exceptions to this pattern: Webb and Shine (1997) showed that male and female Broad-headed Snakes (*Hoplocephalus bungaroides*) travelled similar, long distances compared to gravid females of the same species; and the sexes of Black Ratsnakes (*Elaphe obsoleta*) travelled similar distances (Blouin-Demers *et al.* 2007). The influence of sex and reproductive condition on movement patterns is variable depending on the species.

At a finer resolution, diel movement patterns are affected by foraging mode (Henderson *et al.* 1976). Snakes are generally divided into two foraging modes which have been correlated with a number of morphological (Fizzotti 2015), physiological (Miller *et al.* 2015) and behavioural traits (Secor 1995). The two modes are: ambush foraging, in which individuals ‘sit-and-wait’ for mobile prey to pass by; and active foraging, in which individuals move about their environments in search of prey (Vitt & Caldwell 2013). Secor (1995) observed the active-foraging Coachwhip (*Masticophis flagellum*) moving much greater distances and moving more frequently than the ambush-foraging Sidewinder. Madsen and Shine (1996) and Brown *et al.* (2002) observed rat specialist Water Pythons (*Liasis fuscus*) moving seasonally due to the migration of their prey. The pythons moved closer to water sources (which provided suitable habitats for rats) as the seasons changed and water availability decreased (Madsen & Shine 1996). The pythons move to areas where their prey would be more commonly found and relatively easily accessible for daily ambush activity. These diel movement patterns dictated by foraging relate to the availability and distribution of resources.

Movement patterns of some snakes mimic the general movement of their prey (Madsen & Shine 1996, Brown *et al.* 2002) which could be a necessity in acquiring appropriate energy intakes. Henderson *et al.* (1976) recorded the Common Lancehead (*Bothrops atrox*) moving in diel patterns in accordance with the activity pattern of its prey items (frogs and small mammals). Lanceheads moved up into vegetation at night to ambush prey and then retreated under logs in the morning. The differences in surface movement patterns and times of ambush and active foraging snakes is correlated directly to energy intakes (Carfagno & Weatherhead 2008). Snakes which move more often have higher energy demands and must therefore eat more frequently, creating a feedback loop (Secor 1995). Although ambush foraging snakes move less frequently, they may still mirror the general diel prey movement patterns by foraging during the time of day when their prey items are active (Henderson *et al.* 1976) and thus optimising energy intake.

My study focused on the movement ecology of the Puff Adder (*Bitis arietans*). Diel movement patterns for vipers generally consist of moving from an area which offers protection when inactive, to suitable foraging sites (Nilson *et al.* 1999), where they remain in ambush for a substantial portion of the diel cycle (Secor & Nagy 1994). Seasonal patterns document males travelling farther distances than females in some viperid species (Reinert & Zappalorti 1988, Nilson *et al.* 1999, Brito 2003, Maritz & Alexander 2012; Glaudas & Alexander 2017). However, relative to active foragers, ambush foragers have a stocky build and move less frequently overall (Secor 1995) which could also be in response to increased susceptibility to predation during moves; and ambush foragers have been shown to be greatly affected by habitat fragmentation (Webb & Shine 1997). Bonnet *et al.* (1999) suggested lower anthropogenic mortality rates due to less frequent movements for ambush foragers in comparison to active foragers. My study provides insight into how habitat transformation from road infrastructure may influence the natural movements of an ambush foraging species.

1.3 Wildlife and road infrastructure

The increase in human population and natural resource use has led to environmental change worldwide (Vitousek 1994; Sanderson *et al.* 2002). Specifically, land-use change has been proposed to supersede climatic change in its effects on the terrestrial ecosystem (Sala *et al.* 2000). The consequences of land transformation such as urbanisation and infrastructure are overall unfavourable for the natural environment leading to a loss in biodiversity globally (Forester & Machlist 1996; Sanderson *et al.* 2002; Benítez-López *et al.* 2010) and there are very few places and species that remain unaffected by humans. As a result, animals are faced with new challenges because of changing habitats. Some species can alter their behaviour or physiology as a means of coping with the change (Lindström *et al.* 2014). However, some species have decreased survival or extirpation of populations in response to these changes (Hamer & McDonnell 2010; Lindström *et al.* 2014). New infrastructure in a landscape can create barriers to dispersal and fragment populations (Breininger *et al.* 2011; Smith *et al.* 2014). This can cause the disruption of gene flow, having a detrimental impact on wildlife populations (Reh & Seitz 1990; Trombulak & Frissell 2001; Clark *et al.* 2010). These changes to natural environments have been documented as disastrous for many animals and snakes globally (Rudolph *et al.* 1999; Andrews & Gibbons 2005; Shepard *et al.* 2008). Infrastructure is constantly altering and permeating natural environments, thus it would be beneficial to understand how organisms react to this, so guiding the implementation of appropriate conservation strategies.

Roads tend to be viewed as a disturbance to ecosystems as they are seen as unnatural and fragment the environment. Roads generally have different properties to the surrounding environment depending on the materials used for construction and the nature of the surrounding habitat (Mader 1984; Jochimsen *et al.* 2004). The change in surface structure and material creates open spaces in landscapes, which are often associated with biodiversity loss due to landscape fragmentation, habitat loss or degradation, increased mortality and population isolation (Forman & Alexander 1998; Hamer & McDonnell 2010; Chapman *et al.* 2014).

The type of road along with other characteristics can also be influential to wildlife movement. For example, Robson and Blouin-Demers (2013) recorded Eastern Hognose Snakes (*Heterodon platirhinos*) avoiding paved roads but not unpaved roads. Similar results were observed in a study with cougars (*Puma concolor*) in southern California, as highways were avoided, and dirt roads appeared to facilitate movement (Dickson *et al*. 2005); and with hedgehogs (*Erinaceus europaeus* L.) which crossed wide roads with a lower frequency in comparison to narrow roads (Rondinini & Doncaster 2002). The variance in road avoidance due to road type correlates with other characteristics of the road. For example, paved roads generally support higher traffic volumes and have very different physical properties to unpaved roads. As a result, paved roads may create a more inhospitable environment for wildlife, and avoidance is higher in these situations (Leblond *et al.* 2013). The effects of road type on animal movement is something that needs to be considered when disrupting a landscape with road networks. If road type constrains movement, mitigation measures for surrounding wildlife should be implemented to facilitate movement at certain points.

In some cases, roads may have positive impacts on organisms, such as creating a novel and advantageous microhabitat in the environment. For example, Coffin (2007) describes instances in which roads are used as pathways, although this may be dependent on road type, and the animals that tend use them for this purpose are generalists (Forman & Alexander 1998). A novel pathway through the landscape can provide unobstructed movement opportunities. Some snake species have been recorded as using roads for their thermal benefits (Bernardino & Dalrymple 1992; Rudolph *et al.* 1999; Andrews & Gibbons 2005) or as foraging grounds. Roadside edges have been shown to provide appropriate microhabitats for some prey species which can attract snake predators (Andrews & Jochimsen 2007). Water runoff and a combination of nutrients from the surrounding environment creates patches of resources alongside roads (Coffin 2007). These patches of resources attract small mammals (Oxley *et al.* 1974) and amphibians (Jochimsen *et al.* 2004) which result in concentrations of prey sources for generalist feeders such as ambush-foraging snakes. For example, Terrestrial Garter Snakes (*Thamnophis elegans*) foraged on high numbers of Western Toad (*Anaxyrus boreas*) tadpoles found in roadside edge microhabitats in Idaho (Andrews & Jochimsen 2007). Roads clearly have an ecological impact which should be assessed in existing circumstances where roads infiltrate natural habitats.

South Africa has an extensive road network of approximately 750 000 km and almost 80% of these roads are gravel (Arrive Alive 2017). The impact of these networks on the natural environment is poorly understood and my study provides insight into understanding the effects of roads on wildlife. Puff Adders are commonly found throughout most of the South African landscape and the species may be at a higher risk of effects of road infrastructure relative to other, less widely distributed, species (Fig. 1).

The aim of this research is to assess the effect of roads on reptile wildlife without the bias and emphasis of road kill data. The results of this dissertation are presented in two chapters. Chapter Two will focus on the effects of roads on the spatial ecology of Puff Adders. How do roads influence the proximities of Puff Adder localities? Chapter Three will focus on the effect of roads on movement of Puff Adders (Chapter 3). Do roads influence the movement patterns of Puff Adders? Data in both of these chapters take variables such as road characteristics, Puff Adder sex, season and year into consideration; and are also compared to random models (which provide data for a ‘roadless’ environment). These two chapters are presented as stand-alone papers.

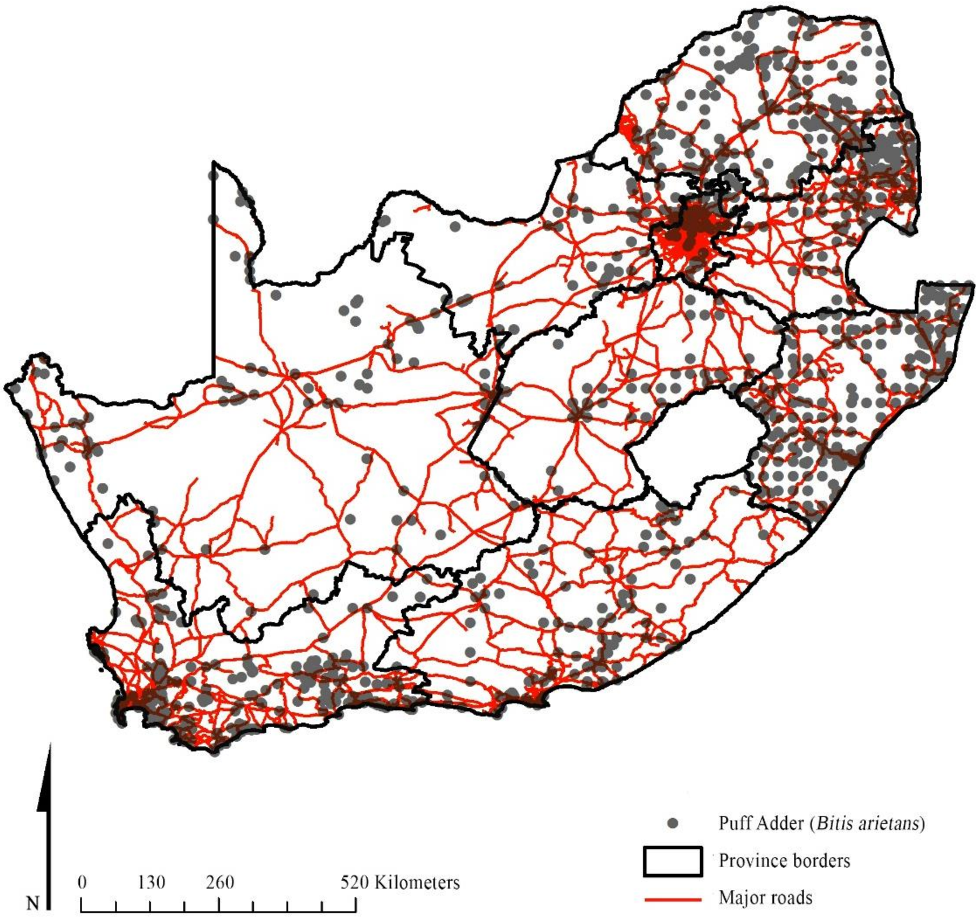

Figure 1. Distribution of Puff Adders (*Bitis arietans*) within South Africa and the major national networks.

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CHAPTER TWO

DO ROADS ATTRACT OR DETER SNAKES? A CASE STUDY ON PUFF ADDERS (*BITIS ARIETANS*) IN DINOKENG GAME RESERVE

ABSTRACT

Road networks are increasingly infiltrating natural habitats as human population and habitat change increase. Information on the response of wildlife to their changing environments is often biased towards road kill statistics and experimental setups, which focus only on the negative impacts of roads. My study used recorded locations of 109 telemetered Puff Adders (*Bitis arietans*) recorded over an 8–years period in the Dinokeng Game Reserve (Gauteng, South Africa). I measured proximity of lie-up locations of snakes to the nearest road and compared these measures to a null distribution. Puff Adder proximity measures were also analysed across seasons, sexes, years and road classes. Compared to random localities, Puff Adders lay-up closer to roads than predicted by the null distribution and were also closer to roads in seasons when foraging was a priority. These patterns suggest that roads can be advantageous to Puff Adders, which appear to use them as foraging hotspots. These findings reveal that there are positive aspects to the presence of roads to Puff Adders.

2.1 INTRODUCTION

Transformation of natural habitat elicits several responses from wildlife populations and in turn can influence both their behavioural and spatial ecology. The main responses of animals to changing environments are adaptation, dispersal or extinction (Tuomainen & Candolin 2011). Road infrastructure is an aspect of urbanisation that is a priority for economic growth (Queiroz & Gautam 1992) and is infiltrating and changing natural habitats to achieve this. Road networks directly fragment habitats and can affect the dispersal and movement of animals as the permeability of the landscape is compromised (Spellerberg 1998). These differing reactions of wildlife towards urbanisation are related to the benefits and costs that changing habitats generate.

The relationship between road infrastructure and wildlife has been extensively researched with specific emphasis on road kill data (Coffin 2007; Meek 2009; Santos *et al.* 2015; Kang *et al.* 2016). Road mortalities are one of the main direct negative effects that habitat change has on wildlife (Seiler & Helldin 2006), however there are several other adverse factors that roads can create in an environment that are not obvious from road kill statistics. Some of these extrinsic factors include pollution from vehicles or the substrate of the road (Andrews & Jochimsen 2007). Light, noise and chemical pollution are disturbances to fauna that are linked to traffic volumes and can alter the behaviour of animals (Outen 2002; Parris & Schneider 2008; Leblond *et al.* 2013). Substrate type can affect pheromone trails (Shine *et al.* 2004) as well as influence roadside areas with respect to runoff and further increase chemical pollution into the surrounding areas (Andrews & Jochimsen 2007). These negative effects can lead to road avoidance which inhibits dispersal (Vos & Chardon 1998).

Understanding how these factors may negatively influence local fauna can only be achieved by shifting the focus from road kill data to detailed observations of where animals are situated in relation to roads.

Although many species are negatively impacted by roads, there are instances where roads and their effects on the surrounding environment can create benefits for the local fauna. Roads are sometimes used as sites for thermoregulation (Sullivan 1981; Bernardino & Dalrymple 1992; Brehme *et al.* 2013) as substrate properties combined with an open space may have increased temperatures (Sartorius *et al.* 1999). These open spaces can also facilitate movement for some species that prefer unobstructed pathways (Brown *et al.* 2006; Di Bitetti *et al.* 2006). Providing movement corridors is not only limited to the road itself, as some small mammals utilise road verges for dispersal (Redon *et al.* 2015). The microhabitats which are created along road edges also allow for suitable breeding grounds for many species, including small mammals and amphibians (Bellamy *et al.* 2000; Andrews & Jochimsen 2007). Such taxa are prey items for many species of snake, and an abundance in their populations can relate to increased foraging opportunities along road edges (Andrews & Jochimsen 2007). The benefits of roads for local wildlife illustrate how some species can adapt to anthropogenic changes in their environment.

How wildlife responds to habitat change depends on the resulting consequences from changes to the natural environment. The position of an animal in relation to roads can give insight into how roads impact the life history and movement patterns of species. However, even if species are adapting by utilising the assorted opportunities that roads provide, the effects can be counteractive as an increased attraction to roads may result in the main direct negative effect of road mortality. Irrespective of whether wildlife avoids or utilises road networks, it is important to be able to distinguish the responses in order to appropriately understand and manage the effects that urbanisation and habitat change will inevitably create. Avoidance of, or attraction to roads is less-well documented (Andrews *et al.* 2008) than are road kill statistics, and the aim of my study is to provide quantifiable data to assess the response of Puff Adders (*Bitis arietans*) to a road network by analysing the proximity of individual lie-ups to roads.

2.2 METHODS

This study used 7 744 point localities recorded using telemetry of 109 Puff Adders in Dinokeng Game Reserve (DGR) from July 2009 to January 2016 using standard telemetric techniques (see Alexander & Maritz 2015; Glaudas & Alexander 2017). Prior to this study, Puff Adders had radiotransmitters surgically implanted into their body cavities according to established procedures (Alexander 2007). They were then released at capture location and located, on average every three days, with a two-element Yagi antenna (Africa Wildlife Tracking, Pretoria, South Africa) and Communications Specialists (Communications Specialists, Pretoria, South Africa) R1000 telemetry handheld receiver

(Alexander & Marais 2015; Glaudas & Alexander 2017). Point localities were recorded with a handheld GPS.

2.2.1 Study site

Dinokeng Game Reserve (25° 23′S, 28° 20′E) is a 18 500 ha reserve situated in the Gauteng province (Dinokeng Game Reserve – The Official Site 2017) in the Savanna Biome (Driver *et al.* 2005) with wet warm seasons from October to April and dry cool months (van Rooyen 2013; Fig.1). The area consists of multiple farms where some regions are open to the public for ecotourism. Large predatory mammals and indigenous ungulates are found within the DGR boundary (Alexander & Maritz 2015). Throughout the reserve there are roads of varying widths and types, with the majority being unpaved.

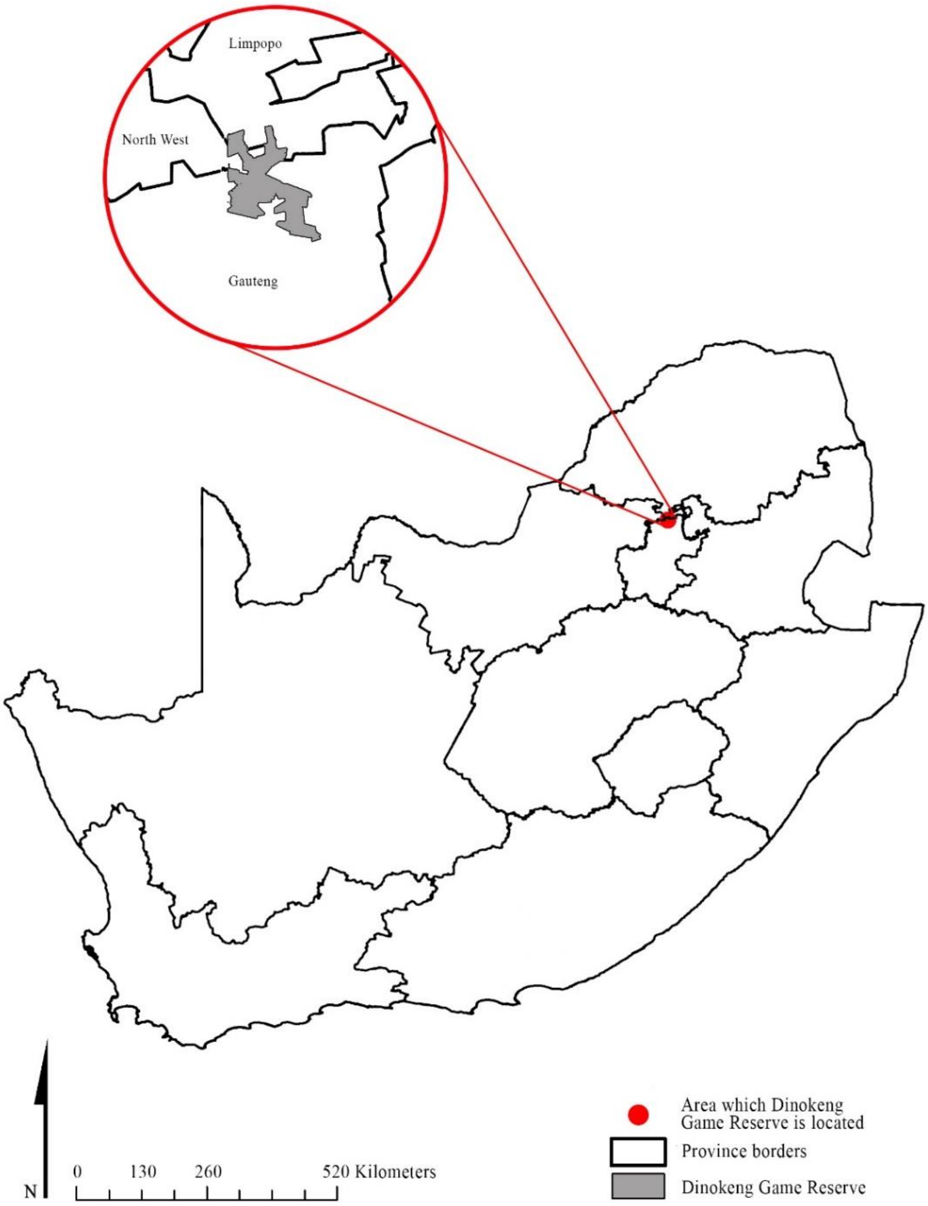

Figure 1. Dinokeng Game Reserve is situated in the Gauteng province in South Africa.

2.2.2 Study species

The Puff Adder (*Bitis arietans*, Merrem 1820) is a common, widespread member of the Viperidae. It occurs throughout South Africa apart from dry deserts and high mountainous regions; and individuals have often been observed under bush cover (Bates *et al.* 2014). These primarily-nocturnal snakes are medium sized with stout bodies and generally use sit-and-wait foraging tactics to capture prey items (Glaudas and Alexander 2017). They are generalist feeders foraging on a variety of different sized terrestrial vertebrates (Glaudas *et al.* 2017). During the mating season (March through to May) males actively search for females (Alexander & Maritz 2015). The species is currently listed in the IUCN threat status as 'Least Concern' (Bates *et al.* 2014).

2.2.3 Quantifying proximity to roads

A geodatabase was made in ArcMap 10.2.2 (ESRI, California, U.S.A.) using the GPS point locality of each Puff Adder along with a digitised map of DGR and the roads within the reserve. The geodatabase was set to the Geographic Coordinate System of 'WGS_1984' and the Projected Coordinate System of 'WGS_1984_UTM_Zone_35S' for South Africa. The point locality shape files contained coordinates, identification number, sex, date, and general notes for each individual. To compare the proximity of Puff Adders to the road network with a null model, 7 744 random points were generated and placed within the minimum convex polygon of all Puff Adder localities combined (Fig. 2). I applied the 'Spatial Join' analysis tool on the locality and road layers to calculate the distance of each point locality to the closest road.

2.2.4 Proximity to roads and the possible influence of weather

I used the climate data for the area to assess if external climatic factors affected differences in proximities of Puff Adders over the years. Average annual rainfall measures from two closest weather stations (Wonderboom Airport and Warmbad Towoomba) were used as a proxy for rainfall data in DGR**.** Wonderboom Airport is 32 km south-southwest of the DGR and Warmbad Towoomba station is 53 km north of the DGR.

2.2.5 Proximity to roads and road type

The roads within DGR were categorised into classes using a combination of Google Earth imagery and ground-truthing to interpret the influence of road type on Puff Adder localities. Initially Google Earth (version 7.1.8.3036) imagery was compared across the years 2009 through to 2016 to identify any changes to the road network. Five preliminary classes were identified using imagery and depended on surface type and width. Five roads from each preliminary class were sampled within DGR to calculate the mean width for the class and to recorded substrate and intensity of vehicular use. Intensity of vehicular use was estimated from observations of vehicle tracks and presence or absence of an island in the middle of the road. Width was measured from outer vegetation edge to outer vegetation edge. After ground-truthing the five preliminary classes were observed as five separate classes (Table 1; Fig. 3; Fig. 4).

Figure 2. The minimum convex polygon (MCP) constructed from all 109 Puff Adder (*Bitis arietans*) individuals within Dinokeng Game Reserve (DGR presented as smaller map in left hand corner). Black outlined circles with various shades of grey represent different individuals and their localities during the tracking periods. Small black dots represent random points placed within the MCP boundary. Lines of different shades and patterns represent the various road classes identified in the reserve.

Table 1. Road classes identified in Dinokeng Game Reserve. Average road widths taken from sampled roads.

Road class	**Average road width (m)** Mean width ± std deviation	**General observations**
1	2.46 ± 0.37	Grass islands almost continuously along the middle of the dirt road, very little traffic zone (Fig. 4a)
2	3.34 ± 1.45	Semi-continuous grass islands along dirt road, light traffic zone (Fig. 4b)
3	4.22 ± 1.13	Dirt road with generally no grass islands, medium traffic zone (Fig. 4c)
4	12.14 ± 3.67	Wide dirt road, high traffic zone (Fig. 4d)
5	10.13 ± 1.33	Tarred road, very high traffic zone (Fig. 4e)

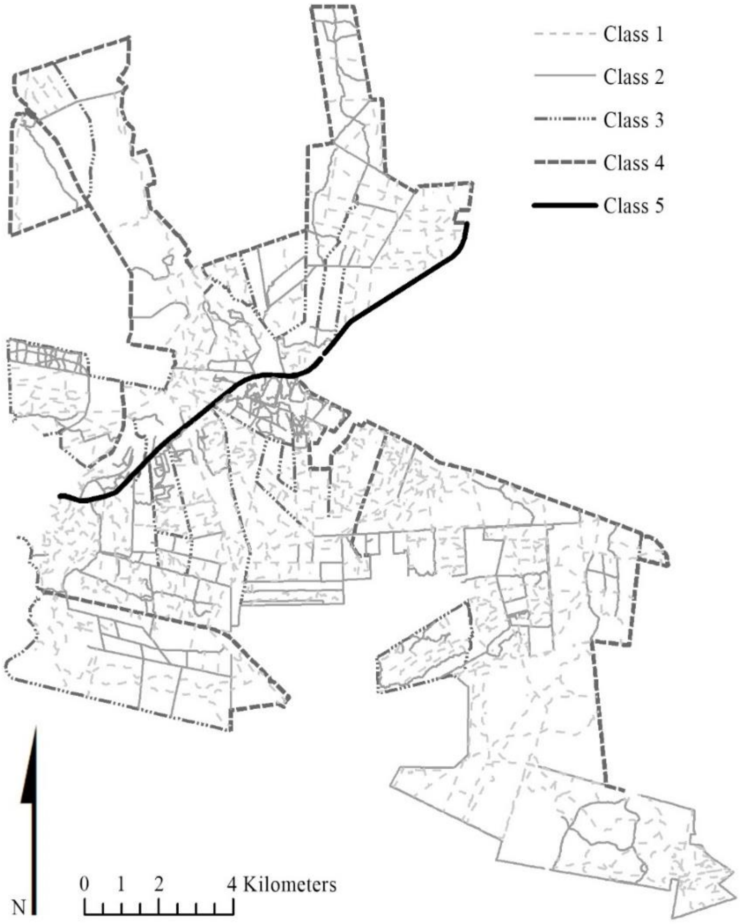

Figure 3. The five different road classes identified in Dinokeng Game Reserve.

Figure 4. Examples of the various road classes identified in the Dinokeng Game Reserve. Letters ‘a’ to ‘e’ corresponds to the classes 1 to 5 respectively.

2.2.6 Statistical analyses

Differences between the mean distribution of proximity measures of Puff Adders to roads and the null distribution of random points to roads were compared, along with differences between sexes, with a Wilcoxon signed-rank test with continuity correction. Generalised linear models (GLM) with quasi-poisson distributions were used to compare non-parametric data. Each model looked at a different aspect (mating season, year, and road classes) across the average proximities for each individual within the population along with interactions of Puff Adder identification to account for bias across the population. A general linear hypothesis test with a Tukey contrast or Fisher's least significant difference (LSD) test were applied where post-hoc analyses were appropriate. Rainfall data were correlated with the proximity of Puff Adders to roads across years to identify any climatic influences and a Pearson correlation coefficient was calculated between the two variables. All data were analysed in RStudio version 1.0.136 (RStudio Team 2016) and all distance values are rounded up to the nearest meter due to GPS locality accuracy. Measures are presented as mean ± standard deviation unless stated otherwise. Significant differences were identified with p values ≤ 0.05.

2.3 RESULTS

The mean distance of proximity measures of Puff Adders to roads was 62 ± 30 m which differed from the random point null model mean of 74 ± 13 m (Wilcoxon signed-rank test: W = 3 894; $p < 0.001$; Fig. 5). Puff adders were located, on average, closer to roads than random points across the DGR (Fig. 6). Distances from roads ranged from 0 m to 398 m for all Puff Adder localities combined. This range of distances was smaller than that of the null model which had an extent of 562 m.

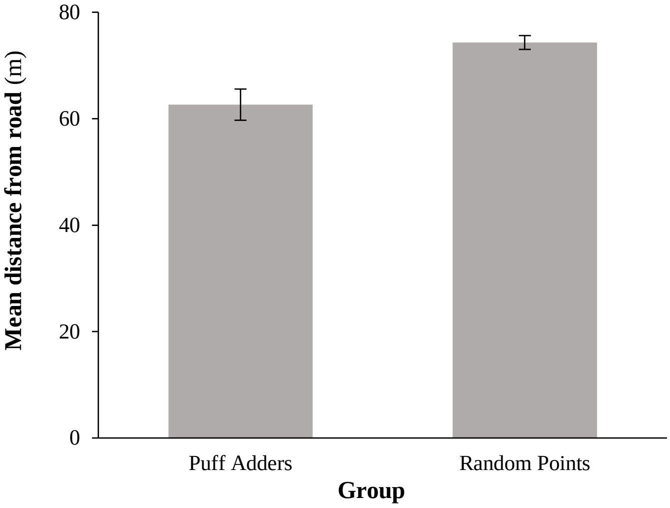

Figure 5. Mean distance of Puff Adders (*Bitis arietans*) and random points to the closest road within Dinokeng Game Reserve. Sample sizes for groups: $n_{\text{Puff Adders}} = 109$; $n_{\text{Random points}} = 109$.

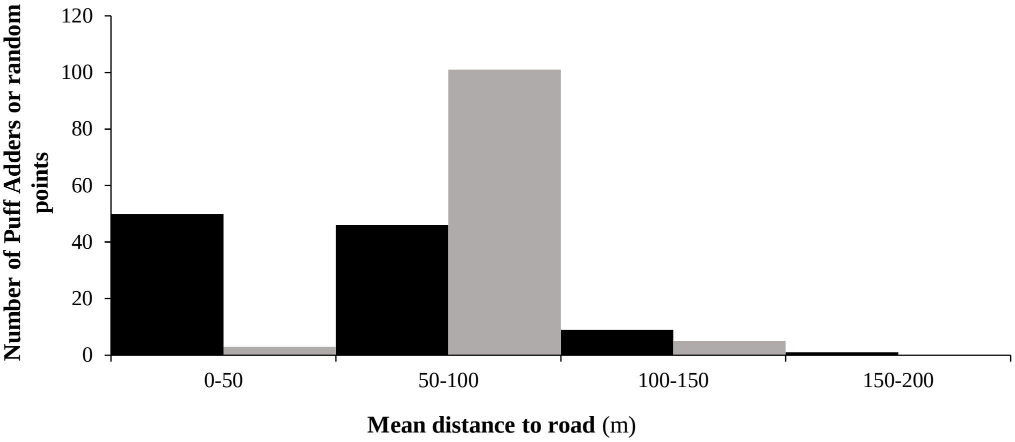

Figure 6. The frequency distribution of mean distance to roads in Dinokeng Game Reserve for Puff Adder (*Bitis arietans*) individuals (black bars) and random points (grey bars).

There was no significant effect on the mean distance to a road between male and female Puff Adders (Wilcoxon signed-rank test: W = 1 557; p = 0.66; Table 2.). However, mating season did show an effect and Puff Adders (across both sexes) were closer to roads in non-mating seasons when compared to mating seasons (GLM with quasi-poisson distribution: $F_{2,196} = 1014$; $p < 0.001$; Fig. 7). Non-mating season had a larger average range of distances to roads (range = 192 m) compared to the range of distances to roads in mating season (range = 179 m).

Table 2. Proximities to closest roads in DGR for male and female Puff Adders. Each Puff Adder's distances were averaged, and the average of each individual was then used to perform statistical analyses.

Sex (n)	**Mean distance** (m) ± std error	**Minimum distance** (m)	**Maximum distance** (m)
Male (55)	63 ± 4	18	135
Female (54)	62 ± 4	11	175

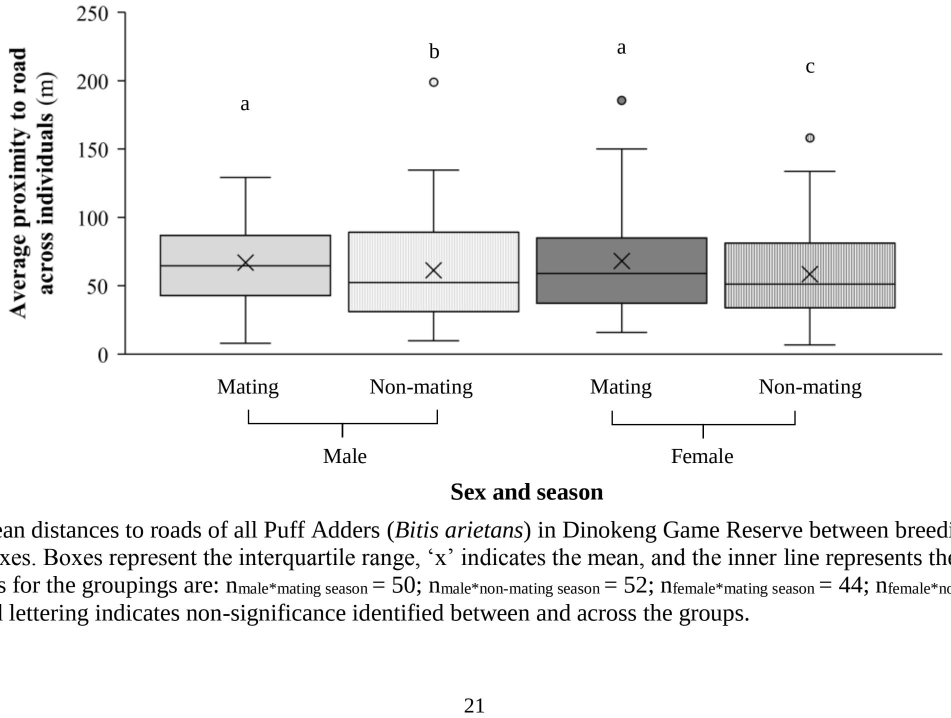

Figure 7. Mean distances to roads of all Puff Adders (*Bitis arietans*) in Dinokeng Game Reserve between breeding seasons and across sexes. Boxes represent the interquartile range, 'x' indicates the mean, and the inner line represents the median. Samples sizes for the groupings are: $n_{\text{male*mating season}} = 50$; $n_{\text{male*non-mating season}} = 52$; $n_{\text{female*mating season}} = 44$; $n_{\text{female*non-mating season}} = 54$). Shared lettering indicates non-significance identified between and across the groups.

Proximity to roads differed significantly across the eight years that the Puff Adders were monitored (GLM with quasi-poisson distribution: $F_{8,194} = 646$; $p < 0.001$; Fig. 8). In 2009 Puff Adders were the closest to roads in comparison to their average distance in 2015. These data did not show a significant correlation to rainfall data from the DGR area (Pearsons' correlation: $n = 8$; $r = 0.58$; $p = 0.13$; Fig. 9). Observed patterns in years of drought showed the highest single distance of 398 m for an individual. This was the greatest distance from the road network in DGR across all years and may show an emerging pattern. The maximum single measure distance of an individual Puff Adder from a road in 2009, which was the year with the most rainfall, was only 172 m.

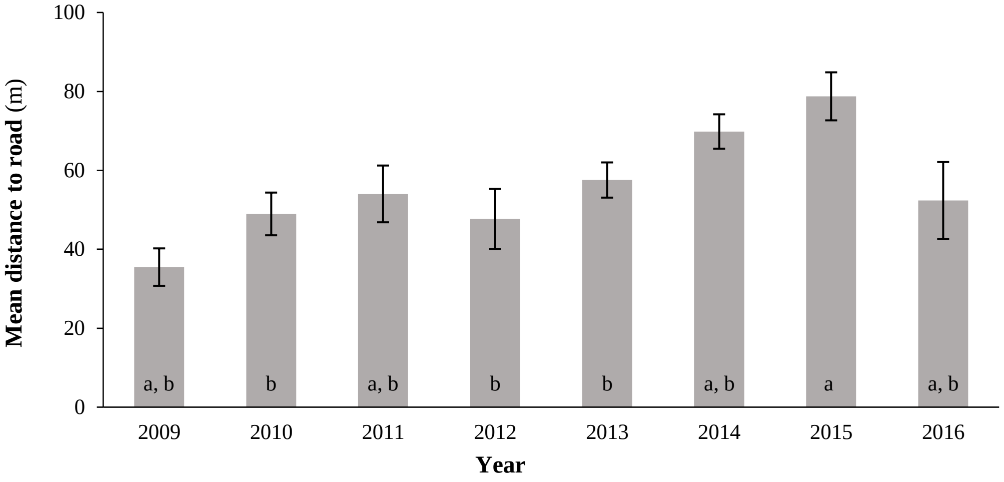

Figure 8. Mean distance to road for Puff Adders (*Bitis arietans*) in Dinokeng Game Reserve across the years snakes were tracked. Sample sizes for the years: $n_{2009} = 6$; $n_{2010} = 22$; $n_{2011} = 18$; $n_{2012} = 15$; $n_{2013} = 43$; $n_{2014} = 91$; $n_{2015} = 40$; $n_{2016} = 8$. Shared lettering represents non-significance between the mean distances.

Figure 9. Mean proximity to roads for Puff Adders (*Bitis arietans*) in relation to the mean annual rainfall for Dinokeng Game Reserve across the various years the snakes were tracked.

Road type was shown to influence the proximity to nearest road with Puff Adders being located, on average, closest to road class 3 (medium width dirt road) (GLM with quasi-poisson distribution: $F_{5,224} = 776$; $p \leq 0.001$; Table 3). Puff adders were recorded the farthest away, on average, to road class 4 which was the widest dirt road with a high level of traffic disturbance in DGR. However, when comparing the distribution of proximity values across the classes, road class 5 had the highest minimum distance and a relatively low maximum distance value.

Table 3. The average proximities of Puff Adders (*Bitis arietans*) to roads and descriptive statistics for the various road classes within Dinokeng Game Reserve. These values were calculated from the average of each individual's proximities per each road class. Group lettering identifies which classes are significantly different from each other. Shared lettering represents non-significance.

Class (n)	**Mean distance** (m) ± std deviation	**Minimum distance** (m)	**Maximum distance** (m)	**Group** LSD post hoc
1 (100)	63 ± 33	11	178	a
2 (93)	65 ± 45	9	318	b
3 (17)	48 ± 28	17	130	c
4 (15)	84 ± 55	8	178	d
5 (4)	53 ± 5	47	58	c

2.4 DISCUSSION

The road network in DGR influences the spatial ecology of Puff Adders as my analyses documented their localities as being closer to roads relative to a null model of random localities. This result suggests that Puff Adders use roads for resources and thus benefit from the road network in this regard. This effect of roads on position did not differ between sexes overall, but rather between mating seasons across both sexes. The bias of mating individuals towards having a greater distance from road networks than in non-mating season could be a response to the shift in priorities for both males and females during this time. Proximity to roads also differed across years, suggesting that some extrinsic environmental pressures may have caused individuals to move closer to or farther from roads. In addition to annual effects, the characteristics of the roads themselves were observed as influential for the distance of a Puff Adder's position to the road network. Overall the effects of roads on the surrounding environment alters both the behavioural and spatial ecology of Puff Adders.

The benefits that roads present are often overshadowed by the disturbances they create but there are some species that have adapted to these anthropogenic changes in the landscape (Vermeulen 1994; Ruiz-Capillas *et al.* 2013). For Puff Adders to be closer to roads than random there must be some advantage that the network is providing for these snakes. The location or movement of an individual is often indicative of their current priority to promote survival (Carfagno & Weatherhead 2008; Tetzlaff *et al.* 2017), and the need to forage is one of these essential priorities (Mueller *et al.* 2007). Being an ambush forager entails waiting for active foragers to pass by (Huey & Pianka 1981) and finding an appropriate site to optimise this is of importance (Glaudas & Alexander 2017). The closeness of individuals to the road network in DGR could be indicative of prey availability and indirectly indicative of how roads promote abundance in some species due to suitable road edge microhabitats that facilitate movement and breeding (Andrews & Jochimsen 2007). Although roads may have negative connotations, the findings in this study suggest that the open spaces that roads create, and the microhabitat of road verges, provides an advantageous foraging site for ambush predators.

The difference between average proximity to roads across mating seasons could also be linked to the shift in life history priorities during this time. Both male and female Puff Adders were recorded farther from roads during the mating season relative to proximities during the non-mating season. For males, the farther localities could be related to their extensive dispersal in search of females (Greenwood 1980; Alexander & Maritz 2015) and finding suitable prey capture sites is of less importance. During mating seasons, female snakes are receptive and experience significant costs in preparation for breeding (Aldridge & Duvall 2002). These costs require females to reduce forage time (Shine 1979; Charland & Gregory 1995) and concentrate their reserves on physiological responses that optimise mating and breeding, such as vitellogenesis and pheromone production and shedding (which has been proposed to aid pheromone release during mating season) (Kubie *et al.* 1978; King & Turmo 1997; Aldridge & Duvall 2002). During this period of focussed efforts on mating or breeding, energy reserves can be depleted and once mating season commences snakes have to replenish and increase their energy intake (Charland & Gregory 1995). The breeding period overlaps with winter months and some species have shown to brumate during these cooler months, which may also explain why foraging efforts are decreased (Brito 2003). This cycle of energy gain and breeding follows the shift from resource priority and could explain the pattern of increased proximity to roads during non-mating seasons due to the possible foraging opportunities provided.

The trend for Puff Adders to be near roads suggests that roads affect forage resources. Extrinsic pressures influence the distribution of resources and my study shows a difference between the proximity to road across years. Although there was no statistical correlation between annual rainfall and proximity to road, emergent patterns are evident. The year with the highest average rainfall (2009) was also the year that Puff Adders were closest to roads and in contrast, 2015 was a year of drought and when Puff Adders were recorded farthest from roads. Resource availability can be strongly affected by climatic seasons and indirectly rainfall and temperature have shown to alter movement patterns in some mammalian species (Brinkman *et al.* 2005; Loretto & Vieira 2005). Amphibians and small mammals make up a significant proportion of Puff Adder diet (Glaudas *et al.* 2017) and the distribution and breeding of some of these prey populations have also shown to flourish in microhabitats created by roadside ditches (Ashley & Robinson 1996; Andrews & Jochimsen 2007). The patterns depicted in the climatic and proximity data further support the notion that Puff Adders are adapting and exploiting the effects of roads on the natural environment.

In contrast to the positive response towards roads, some research indicates that roads with higher disturbance characteristics are avoided by certain species (based off relative proximity) (Reijnen *et al.* 1995; Forman & Alexander 1998). In DGR, road class 5 (wide tarred road) would be the most intense disturbance being a high traffic zone with a contrasting substrate to the natural environment. This class showed the highest mean minimum distance of Puff Adders to the road, possibly indicating that the disturbance from this road class may affect Puff Adder lie-ups. However, there was no evidence that Puff Adders were found closer to road classes with less disturbance. The lack of an obvious pattern across road classes could be due to the unique and specific combination of the costs and benefits to snakes from the road type. For example, road class 4 may provide appropriate foraging opportunities (because of the microhabitat and vegetation along the verge) but the negative effects of traffic for the same road class could outweigh these benefits. The avoidance of or attraction to roads to the various road classes may depend on the balance between risk and reward.

My study used more than 7 000 localities of Puff Adders in relation to a road network, collected over multiple seasons and years and has given insight into the behavioural and spatial ecology for the species. This data set is larger than other telemetry-road research datasets that have previously been studied globally (Shine *et al.* 2004; Brown *et al.* 2006; Fortney *et al.* 2012; Siers *et al.* 2014) and the first of its kind in South Africa. The natural set up of this study provided a realistic view on a topic that has been biased with road kill data or experimental setups. However, to better understand the effect of road type on proximity to road, further detailed analyses of the road and road verge characteristics should be incorporated. This would highlight the potential risks and rewards associated with each class. Surveying the spatial ecology in a system where there are fewer roads or very contrasting road types may better reveal effect of road type.

The proximity of an individual to a road can reveal its response to a changed environment and in this study Puff Adders appear to utilise the road network when foraging is a priority. The importance of understanding how roads affect the spatial and behavioural ecology of a population or species is less emphasised in the literature (Andrews *et al.* 2008) in comparison to the main negative effects of roads. Observing how wildlife are attracted to roads is further evidence for the reasons behind possible road mortalities or, in contrast, the avoidance of roads because of specific characteristics or extrinsic pressures supports the effect of fragmentation. My study has shown that not all impacts of roads are negative and gives insight into why there are particular patterns in road ecology. The understanding of these movement patterns can aid in effective management where roads infiltrate natural habitats.

2.5. REFERENCES

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CHAPTER THREE

DO ROAD NETWORKS CHANGE THE MOVEMENT OF SNAKES? A CASE STUDY ON THE PUFF ADDER (*BITIS ARIETANS*)

ABSTRACT

Human development has caused road networks to multiply and permeate natural habitats which in turn impact the surrounding biodiversity. The main negative effects of roads on animals is habitat fragmentation and increased mortality. These effects are often highlighted since most data are in the form of road kill statistics, whereas this study investigated the relationship between movement patterns of Puff Adders (*Bitis arietans*) and roads. Telemetry data from 109 individuals over several years were recorded and analysed. The majority of the snakes tracked in this study crossed at least one road in the Dinokeng Game Reserve (Gauteng, South Africa) and road crossings were sex biased. Males in mating seasons crossed roads the most often which relates to their increased dispersal for reproduction. Not only are there physiological drivers that influence movement patterns and road crossings, but there are also the characteristics of the roads that influence movement patterns. Narrow roads with islands of vegetation down the middle and very low traffic density are crossed, on average and per kilometre, six times more often than wide busy roads. However, when compared to a correlated random walk model, Puff Adders were crossing roads more often than random and, in general, the roads in Dinokeng Game Reserve are not a barrier to movement. These results show how roads may not always have negative hindering effects on wildlife movement, although this lack of avoidance of roads may have the potential to increase the risk of road mortalities. It is essential to understand how road infrastructure is affecting wildlife as well as the potential future effects so that appropriate management and mitigation can be carried out.

3.1 INTRODUCTION

Increased road infrastructure is a significant issue with complex, often unfavourable impacts on local fauna. The major impacts of roads on wildlife movement include habitat fragmentation, barriers to movement and increased mortality due to road kill (Bissonette 2002; Evink 2002; van der Ree *et al.* 2015). Dispersal is a common behaviour in many animals and may be motivated by the need to forage, reproduction or safety, and is heavily influential for the biodiversity of a region (Jeltsch *et al.* 2013). Roads decrease the continuity of habitat in the environment and they create obstacles for dispersal and movement. How animals react to these obstacles can have consequences on populations and the overall biodiversity. Most research on road ecology has focused its efforts on analysing road kill data (Enge & Wood, 2002; Rolandsen *et al.* 2011; Garrah *et al.* 2015) and experimental setups (for example, collecting individuals and placing them next to roads while monitoring their movements). Evaluating the impact of road networks on natural movement patterns is greatly understudied. Understanding the movement patterns of wildlife in response to the presence of roads is necessary to successfully mitigate the effects of roads within a habitat.

Movement patterns are influenced by the location of resources (Bowler & Benton 2005) and the fragmentation of habitats. Road networks act as barriers to animal movement, and depending on the resource and its spatial occurrence, there may be different risks associated with the movement of an individual in search of the resource. For example, movement from daily foraging patches could be limited to a single fragment of habitat and of minimal risk, compared to individuals which must travel relatively great distances to locate prey items (Brown & Parker 1976; Madsen & Shine 1996; Glaudas & Alexander 2017). The dispersal of food resources is also connected to extrinsic variables and some research has shown that variation in climatic seasons alters movement patterns as a result of this (Brinkman *et al.* 2005; Loretto & Vieira, 2005). Siers *et al.* (2016) showed that Brown Treesnake (*Boiga irregularis*) movement and rate of road crossings was influenced by foraging opportunities. Locating suitable resources is essential for survival and in foraging seasons, the benefits of dispersal outweigh the associated risks (Tetzlaff *et al.* 2017). This is also the case in mating seasons where optimizing breeding opportunities is a priority as some males disperse extensively in search of females (Greenwood 1980; Alexander & Maritz 2015). Breininger *et al.* (2011) observed male Eastern Indigo Snakes (*Drymarchon couperi*) in fragmented habitats to have a diminished home range. Depending on the motivation of an individual, for example forage or reproduction, its movements will change in accordance to the resource which is a priority

Some animals avoid roads due to the material of the road surface and traffic intensity (Leblond *et al.* 2013; Robson & Blouin-Demers 2013). Paved roads have greatly decreased surface permeability in comparison to the natural landscape. This change from one substrate to another alters movement patterns and many species have shown to be especially reluctant to cross tar roads (Brock & Kelt 2004; Brehme *et al.* 2013; Robson & Blouin-Demers 2013). Intense disturbance such as in areas of high traffic volume or stretches of roads where speed limits are high have also been shown to impact wildlife road crossings, and areas with these disturbances are avoided or have higher rates of road kill (Farmer & Brooks 2012; Leblond *et al.* 2013). Whether a road is avoided because of its properties or if it is crossed, resulting in increased mortality, the effect is a decrease in genetic cohesion for an area (Strasburg 2006).

Avoidance of roads can be due to several factors which include pollution in the form of noise, light and chemicals, and the general open space (increasing vulnerability) that is created (Andrews & Jochimsen 2007; Masterson *et al.* 2008). These along with road mortalities, are always going to be threats to wildlife where roads infiltrate natural habitats and the need to mitigate these threats is an urgent current concern (Jacobson *et al.* 2016; Teixeira *et al.* 2016). I assessed the movement patterns of Puff Adders (*Bitis arietans*) in the Dinokeng Game Reserve in Gauteng, South Africa in order to understand how road networks impacted their movement patterns. I used long-term telemetry data to investigate whether roads are avoided in movement paths. In order to understand effects of habitat change and appropriately mitigate threats to populations and biodiversity I identified drivers of interactions between movement paths and road networks.

3.2 METHODS

I used track data collected using telemetry from 109 Puff Adders (55 males and 54 females) in Dinokeng Game Reserve (DGR) from July 2009 to January 2016. The dataset includes records from all seasons and data were collected using standard telemetric techniques (see Alexander & Maritz 2015; Glaudas & Alexander 2017). Snakes were surgically implanted with radiotransmitters in accordance with established procedures (Alexander 2007); released near their capture site and tracked on average every three days. The Puff Adders were tracked using a two-element Yagi antenna (Africa Wildlife Tracking, Pretoria, South Africa) and Communications Specialists (Communications Specialists, Pretoria, South Africa) R1000 telemetry handheld receiver (Alexander & Marais 2015; Glaudas & Alexander 2017). Location data were recorded with a handheld GPS.

3.2.1 Study site

Dinokeng Game Reserve is located within the Gauteng province in the Savanna Biome (Driver *et al.* 2005) which experiences wet warm seasons from October to April and dry winter months with temperatures ranging from 0°C to 40°C throughout the year (van Rooyen 2013; Chapter 2: Fig. 1). The region is used primarily for ecotourism and includes large predatory mammals and many indigenous ungulates (Alexander & Maritz 2015). The reserve is a combination of private and public properties, some of which are fenced off. Most of the roads within this 18 500 ha area (Dinokeng Game Reserve – The Official Site 2017) are unpaved and vary in widths and traffic intensity.

3.2.2 Study species

Bitis arietans, Merrem 1820, (Puff Adder) is a common, widespread viperid occurring throughout South Africa with the exception of dry deserts and high mountain tops (Bates *et al.* 2014). Individuals have been recorded in most habitats but prefer bush cover (Bates *et al.* 2014). Puff Adders are medium-sized heavy-bodied ambush predators which are generalist feeders (Alexander & Marais 2007; Glaudas *et al.* 2017). Currently the species is listed as 'Least Concern' under the IUCN threat status (Bates *et al.* 2014). Mating season include March through to May within this area (Alexander & Maritz 2015).

3.2.3 Quantification of road crossings

The location of each individual snake was georeferenced in ArcMap 10.2.2 (ESRI, California, U.S.A. 1999 - 2014) on a digitised map of DGR creating a geodatabase in which analyses were performed. The geodatabase was set to the World Geodetic System 29184 ('WGS_1984') and 'UTM_Zone_35S' for the Universal Transverse Mercator projection for South Africa. The shapefile for each individual consisted of locality co-ordinates, identification number, sex, date, season, and general notes. The successive point localities of each Puff Adder were joined with straight lines using the ‘Path Tool’ in the ArcMET 10.2.2. v3 (Wall 2014) extension to create movement paths. Straight line distance, albeit an underestimate of Puff Adder movement (Alexander & Maritz 2015), is the standard technique used to conduct movement analyses due to lack of information from one locality to the next (Madsen 1984, Secor 1995, Brito 2003). These movement paths, along with the road network layer were used in combination with the ‘Intersect’ tool in ArcMap 10.2.2 (ESRI, California, U.S.A. 1999 - 2014) to identify point intersection localities (road crossings).

The data for the years 2009, 2012, 2016 were eliminated when looking at the difference between road crossings per year due to Puff Adders only being tracked for a relatively brief period during these years. Annual climate data and number of individuals tracked within a year were used to better interpret the effect of year on road crossings. Rainfall data from two weather stations, Wonderboom Airport 32 km south-southwest and Warmbad Towoomba 53 km north from the DGR region, were used in the analysis. Measures from the two stations of average annual rainfall were used to represent rainfall data within DGR.

3.2.4 Road crossings and road type

To assess the importance of road type on road crossings, I categorised roads within DGR into five classes depending on surface type, width, and traffic volumes with the aid of Google Earth (version 7.1.8.3036) imagery and ground-truthing. Ground-truthing involved checking each road class identified from imagery at five locations throughout the reserve. Data collection consisted of measuring the width of the road from outer vegetation edge to outer vegetation edge (thus grass islands were included in average width), as well as recording substrate and intensity of vehicular use. Intensity of traffic was identified by substrate type, width, and presence of vehicles. In total, five road classes were identified within DGR (Chapter 2: Table 1; Fig. 3; Fig. 4).

3.2.5 Road crossings and correlated random walks

The Geospatial Modelling Environment (GME) (Beyer 2012) software was used to create correlated random walks (CRW) with the “movement.simplecrw” tool (Beyer 2012) to produce a null model for road crossings. The CRW provided a realistic random model of movement based on metrics of actual movements (Byers 2001). Two CRW models were used in this study, each with different criteria for the distribution of crossings. The initial null assessed each individual Puff Adder using number of positions, step lengths and turning angles based on the individual’s actual movement metrics (Bartumeus *et al.* 2005). However, since these metrics for the null were based on Puff Adder movements in areas where there were roads, a second null model was produced based on the step lengths and turning angles of 85 individuals whose home ranges (defined by a minimum convex polygon) only included road classes 1 and 2. Exploratory analyses with the data suggested that these roads may be interpreted as natural open spaces in the landscape due to their short widths and grassy islands which grow down the middle. This second CRW model represented step lengths and turning angles that are more likely ‘random’ and more appropriately indicative of movements not influenced by a road network. Both models were restricted to a boundary, which was defined by the DGR border and each Puff Adder was run through 100 iterations.

3.2.6 Statistical analyses

Snakes were tracked over varying periods and crossings per month per each individual were calculated. Since data were not normally distributed, non-parametric tests were used. To test for a difference between sexes, I compared crossings per month per Puff Adder for males and females using a Wilcoxon signed-rank test. To test the influence of year, season and road class on crossings per month per individual I applied a generalised linear model (GLM) with a quasi-poisson distribution. For road class analyses, crossings per kilometre were used to account for the varying total lengths of road class. In each analysis, Puff Adder ID was used as an interaction to account for individual effect. Where significant differences across factors were apparent, the Fisher’s least significant difference post hoc test was used to identify were significant differences lay. The effect of weather was analysed in correlation with the number of crossings per month tracked of each year using a Pearson correlation coefficient. To test number of actual crossings to the two null models, I compared crossings per month per Puff Adder against the CRW road crossings per month per Puff Adder with Wilcoxon signed-rank tests. Data were analysed in RStudio version 1.0.136 (RStudio Team 2016) and measures are presented as mean ± standard deviation unless otherwise stated. A p value ≤ 0.05 defined a statistical significant difference.

3.3 RESULTS

Altogether 97 Puff Adders made a total of 2 225 road crossings in DGR. Males crossed roads 1 786 times in total ($n = 55$; $x_{\text{crossings per month}} = 3.35 \pm 1.95$), more than four times the number of crossings that female Puff Adders made ($n = 54$; $x_{\text{crossings per month}} = 0.70 \pm 0.83$) (Wilcoxon signed-rank test: $W = 2\,715$; $p \leq 0.001$; Fig. 1). The highest number of crossings per month for the total tracking period for one individual was 8.5, which was a male Puff Adder. The interaction between sex and season showed that males in mating season crossed roads more often than other combinations of sex and season (GLM with quasi-poisson distribution: $F_{4,196} = 115.15$; $p < 0.001$; Fig. 2). In contrast, females in non-mating season crossed, on average, the fewest number of times per month.

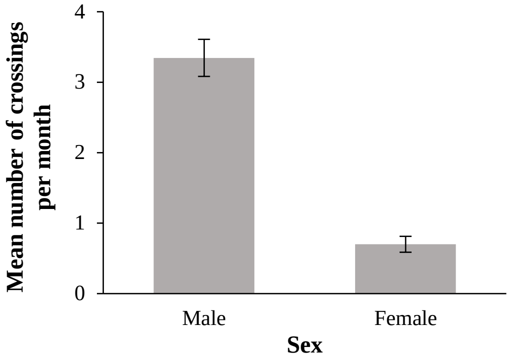

Figure 1. Mean number of crossings per month per individual for male and female Puff Adders (*Bitis arietans*) in Dinokeng Game Reserve. Sample sizes for the groups are: $n_{males} = 55$; $n_{females} = 54$.

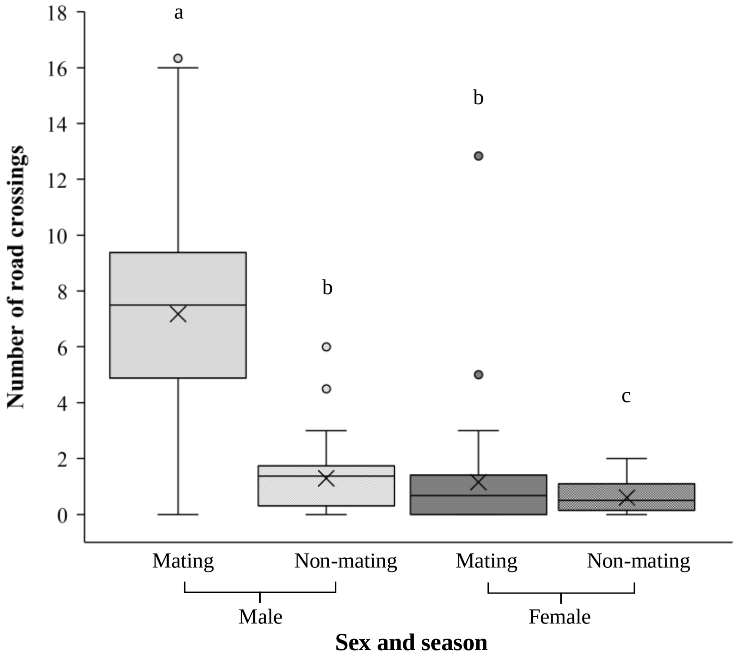

Figure 2. Number of crossings per month per individual in the respective seasons for male and female Puff Adders (*Bitis arietans*) in Dinokeng Game Reserve. Boxes represent the interquartile range, 'x' indicates the mean, and the inner line represents the median. Shared lettering indicates non-significance identified between and across the groups. Samples sizes for the groupings are: $n_{male*mating\ season} = 50$; $n_{male*non\text{-}mating\ season} = 52$; $n_{female*mating\ season} = 44$; $n_{female*non\text{-}mating\ season} = 54$.

There was a difference between the number of crossings per month across the years 2010 through to 2015 with the highest mean documented in 2014 (GLM with quasi-poisson distribution: $F_{5,168} = 11.80$; $p \leq 0.001$; Fig. 3). The GLM showed a general trend of significance, but post-hoc pairwise comparisons failed to show where the significant differences lay. When comparing mean road crossings per month to rainfall, there was no correlation across the years (Pearsons' correlation: n = 5; r = 0.40; p = 0.51).

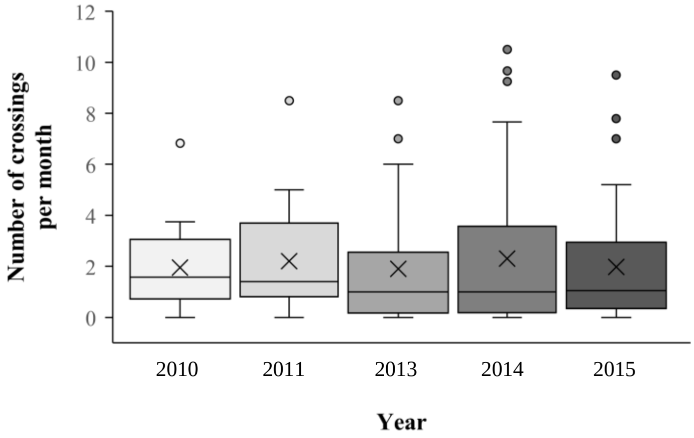

Figure 3. The number of road crossings per month per Puff Adder (*Bitis arietans*) across years they were tracked in Dinokeng Game Reserve. Years 2009, 2012 and 2016 were removed from this analysis because of insufficient data. Boxes represent the interquartile range, ‘x’ indicates the mean value and the inner line represents the median. Sample sizes for the years: $n_{2010} = 22$; $n_{2011} = 18$; $n_{2013} = 43$; $n_{2014} = 50$; $n_{2015} = 40$.

Road class had an influence on the number of road crossings (GLM with quasi-poisson distribution: $F_{5,184} = 23.3$; $p \leq 0.001$; Fig. 4), with significantly higher number of crossings per kilometre occurring on road classes 1 and 2 compared to the other road classes. Roads from classes 1 and 2 had relatively narrow widths and were not often used by vehicles. The fewest number of crossings by Puff Adders were on road classes 4 and 5 which were the widest roads in the reserve with the high traffic intensity (Pearsons’ correlation: $n = 5$; $r = 0.87$; $p = 0.05$; $y = -0.2523x + 2.9809$; Fig. 5).

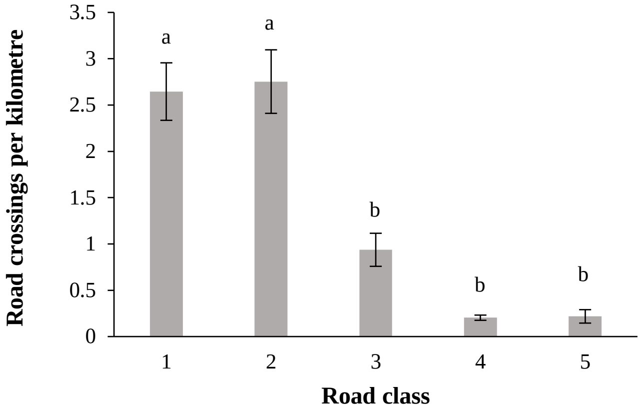

Figure 4. Mean number of road crossings per kilometre for Puff Adders (*Bitis arietans*) in Dinokeng Game Reserve across the five different road classes. Shared lettering indicates non-significance between and across the groups. Sample sizes for each class: $n_1 = 87$; $n_2 = 75$; $n_3 = 15$; $n_4 = 10$; $n_5 = 2$.

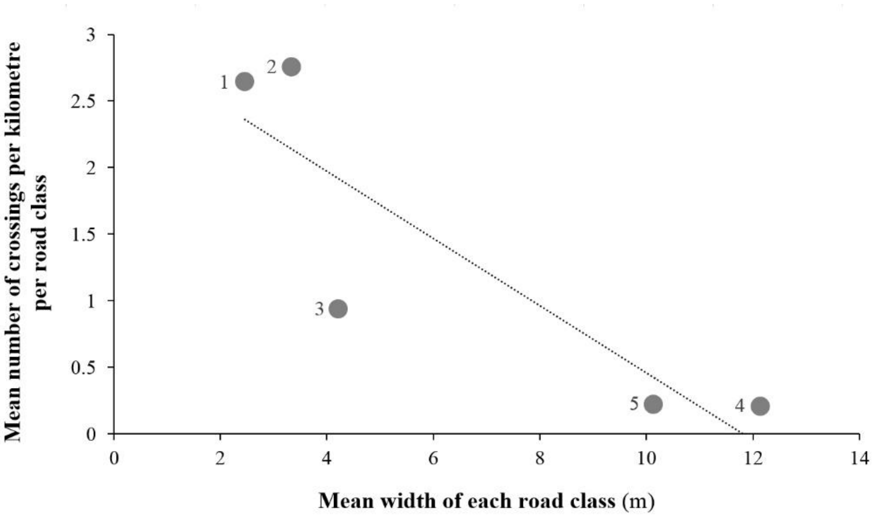

Figure 5. The relationship between the mean number of crossings per kilometre per road class for Puff Adders (*Bitis arietans*) and the mean widths of the road classes in Dinokeng Game Reserve. Road classes are identified as numbers to the left of their respective markers.

There was no difference between the mean road crossings per month of Puff Adders and the mean crossings per month from the initial CRW model which was based on the movement metrics of all 109 individuals (Wilcoxon signed-rank test: W = 5 585.5; p = 0.45; Table 1). However, there was a difference between Puff Adder road crossings per month per individual (x = 3.81 ± 2.11) when compared to the second CRW model (x = 2.47 ± 0.75) which used the 85 individuals that only had road classes 1 and 2 in their home ranges for its criteria (Wilcoxon signed-rank test: W = 300, p ≤ 0.001; Fig. 6).

Table 1. Descriptive statistics for Puff Adder (*Bitis arietans*) road crossings per month tracked in Dinokeng Game Reserve and the initial correlated random walk model road crossings per month.

	n	Mean ± std deviation	Standard error	Minimum	Maximum
Puff Adder	109	2.03 ± 2.00	0.19	0	8.50
CRW	109	2.06 ± 1.80	0.17	0	7.62

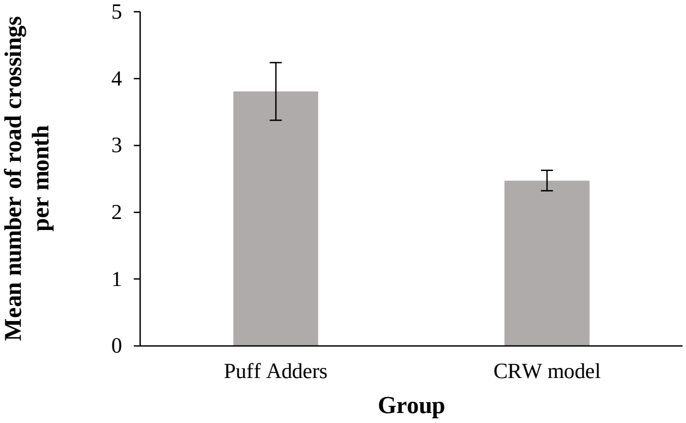

Figure 6. The mean number of road crossings per month per individual for Puff Adders (*Bitis arietans*) and the second correlated random walk (CRW) model. This second CRW model is based off of 85 Puff Adders which home ranges' exclusively included road classes 1 and 2. Sample sizes: $n_{\text{Puff Adders}}$ = 24; n_{CRW} model = 24.

3.4 DISCUSSION

Puff Adder movements were influenced by several aspects pertaining to their demographics which directly affects road crossings and these movements are not inhibited by the road network in DGR. In total, 98% of the Puff Adders in the dataset crossed at least one road in DGR while being monitored, and roads thus form an integral part of their habitats. Sex and season do influence how often roads are crossed and this is in direct relation to life history traits of Puff Adders. The difference in rates of crossings between years suggests that variability in extrinsic environmental factors have an important impact on movement patterns, and probably also life history traits of Puff Adders. The drastic significant difference in number of crossings across road classes influenced the design of the second CRW model. The results from the CRW based on movement metric for Puff Adders whose home ranges included only class 1 and 2 represented a more appropriate null model and Puff Adders crossed roads more frequently than the null model (representing movements in a roadless landscape) which suggests that roads do no inhibit Puff Adder movement in the DGR. Understanding the different drivers for movement is essential to mitigate the often negative effects that may occur by increased road crossings.

The impact of sex and season, along with the interaction of the two, was an expected finding as research has shown male snakes generally travel extensively in search of mates (Brown & Parker 1976; Madsen 1984; Carfagno & Weatherhead 2008; Alexander & Maritz 2015). In the mating season, male Puff Adders crossed roads more often than females, in either mating or non-mating season, and males in non-mating season. This is related to the breeding behaviour of the species in which the males go out in search of females, often in a straight-line movement to increase mating success (Duvell & Schuett 1997; Glaudas & Alexander 2017). Females lay pheromonal trails (Mason *et al.* 1989) and reproductive males often increase movements and distance to increase their mate-searching success (Shine 2003). These multiple straight-line movements result in approximately triple the number of road crossings made in the non-mating season by male Puff Adders. This extended movement increases the risk of exposure to predators and road crossings which can result in road mortality. Females have different priorities in the breeding season and once gravid, movement is reduced due to physiological changes (Macartney *et al.* 1988). This results in fewer road crossings by female Puff Adders compared to males across all seasons and especially mating season.

The increased dispersal during breeding seasons by male Puff Adders is not restricted to the Serpentes and many mammalian species show similar male-biased movement patterns (Greenwood 1980; Killeen *et al.* 2014). Wattles (2015) showed that male moose (*Alces alces*) increased their daily movements in the rutting season which corresponded to a peak in moose-vehicle collisions. Increased daily movement of males to find females in polygynous species results in reduced chances of inbreeding (Greenwood 1980; Perrin & Mazalov 1999). However, there are often risks associated with extended ranges such as with an increase in road-movement path intersections (Wattles 2015).

Bonnet *et al.* (1999) observed adult male Aesculapian Snakes (*Elaphe longissima*) and Whip Snakes (*Coluber viridiflavus*) had significantly higher accidental mortality rates during mating season, and Macdonald *et al.* (2008) recorded that male badgers (*Males meles*) had more wounds than females because of their frequent movements. The benefits of successful matings outweigh the costs of dispersal in many instances, which is supported by the increased number of road crossings by males in the mating season.

There are multiple variables that are related to the factor 'year', and although annual climatic changes do not explain differences in number of road crossings, other environmental extrinsic pressures could. Snakes were caught opportunistically throughout the years with relatively similar sample efforts (*pers. comm.* G. J. Alexander; Glaudas & Alexander 2017). The variation in crossing per month from each year could have been influenced by several factors. Over the years, since 2009, the DGR has experienced many ecological changes: the introduction of large herbivores and carnivores; the establishment of self-drive routes for tourists; road alterations; and construction of tourist recreation areas (Dinokeng Game Reserve – The Official Site 2017). Large mammals greatly impact the structure and organisation of the ecosystems that they inhabit, and other organisms found in the same environment are most likely to be influenced by their presence (McNaughton *et al.* 1988). Along with the addition of new species, the increase in urbanisation and habitat change has been shown to strongly alter populations (Ditchkoff *et al.* 2006; Hamer & McDonnell 2010) due to the intrusive changes which are made to a natural habitat. Finally, movement patterns of Puff Adders have been influenced by how much they consumed (Glaudas & Alexander 2017) and if there are extrinsic effects on prey populations then this too could impact movements. Any of these attributes could alter the environment and influence the behaviour of the species within the habitat, which could have affected the number of road crossings occurring per month each year.

The type of road was another extrinsic influence on the movement of Puff Adders. The narrow, low traffic volume roads were crossed much more frequently than wider roads with heavier vehicle traffic within DGR. Road classes 1 and 2 were grouped together and road classes 3, 4 and 5 could be grouped. Road class 4 was the widest road within a high traffic zone and this was the least crossed road per kilometre of road. Oxley *et al.* (1974) observed the effect of road width on medium-sized mammals and found the influence to be as detrimental as the effect of a body of water twice as wide. In DGR road classes 3, 4 and 5 result in higher levels of noise and air pollution, and heavy vibrations from high vehicular use as well as the creation of a large exposed area that requires traversing in comparison to classes 1 and 2. Although the majority of the roads in DGR are unpaved, there is one main paved road which fell within the overall range for Puff Adders. This road had the second fewest number of crossings per kilometre despite being within the home ranges of several individuals. The contrast of substrate is one possibility for road avoidance (Robson & Blouin-Demers 2013), and the intensity of disturbance is another. Not only are individuals at risk of being hit by vehicles, but they are also susceptible to predators when crossing. In contrast, narrower roads with islands (and less vehicular disturbance) pose a safer traversing area.

Puff Adders avoided crossing wide roads with increased traffic volumes. The two road classes that were crossed the fewest number of times per kilometre were similar in their widths and vehicular usage but differed in substrate material. The change from natural terrain to a different material for some animals could promote aversion and is one of the consequences of a paved road network. Robson and Blouin-Demers (2013) observed Eastern Hognose Snakes (*Heterodon platirhinos*) avoid crossing paved roads but commonly crossing dirt roads in Ontario, Canada. Similarly Garter Snakes (*Thamnophis sirtalis parietalis*) deliberately avoided crossing gravel roads and in instances when crossings were made, snakes utilised the shortest route (perpendicular to the road) (Shine *et al.* 2004). Small mammals such as mice and rats also avoid paved roads (Brock & Kelt 2004; Brehme *et al.* 2013), and road avoidance by large herbivores correlates with disturbance intensity in the form of traffic (Leblond *et al.* 2013). Despite the notion that paved roads relate to higher levels of traffic, McGregor *et al.* (2008) reported that small mammals avoid the road regardless of the traffic intensity and suggested that the road itself is recognised as a barrier. General road avoidance could be due to a combination of effects from the road characteristics and the change in natural vegetation to an open ‘risky’ space.

Puff Adders crossed roads more frequently compared to a null model (representative of movements in a roadless environment) within the DGR boundary and this identifies with the benefits associated with roads. These results concur with a study by Row *et al.* (2007) in which Black Ratsnakes (*Elaphe* obsoleta) crossed roads more often than a random model, suggesting that roads may not be a significant barrier to some species. Roads can be used to facilitate thermoregulation for animals (Camacho 2013) as they can serve as basking sites (Andrews & Jochimsen 2007). Open disturbed areas can have higher environmental temperatures relative to the surrounding natural landscape and these basking sites may then aid in foraging activity (Sartorius *et al.* 1999). The road edges also serve as foraging opportunities as in some cases they provide optimal breeding habitats for anurans (Andrews & Jochimsen 2007) and movement corridors for small mammals (Redon *et al.* 2015). The increased numbers in these prey items can lead to increased foraging along roadsides and possibly increased road crossings in search of these prey hotspots (Andrews & Jochimsen 2007). The indirect effects of roads on the surrounding ecology may be beneficial for certain fauna. However, the increased allure of roads is potentially adverse and can have negative effects on populations (Row *et al.* 2007).

There are two main negative direct effects that roads have on wildlife that are often highlighted in the literature: the creation of a barrier for physical dispersal, and the increased mortality rates from road kill (Spellerberg 1998; Bissonette 2002; Andrews & Jochimsen 2007; Coffin, 2007). Overall the roads in DGR are crossed more often than random by Puff Adder, however the type of road does influence how often roads are crossed. The combination of substrate-type, width, and traffic creates a discontinuity in a once continuous landscape. This alteration to the natural landscape can have severe repercussions by hindering natural movement patterns, and in turn, gene flow (Reh & Seitz 1990; Holderegger & Di Giulio 2010). Clark *et al.* (2010) documented how roads had significantly lowered the genetic diversity of Timber Rattlesnakes (*Crotalus horridus*) in New York State as a direct result of impeding male movement patterns in the mating season. Small-bodied animals are not alone in facing obstructions to gene flow, as the genetic diversity in Desert Bighorn Sheep (*Ovis canadensis nelsoni*) has decreased by 15% over the past 40 years due to road networks and urbanisation infiltrating natural habitats (Epps *et al.* 2005). The barrier to movement patterns and the impact on the gene flow is detrimental to many species.

The effect of roads on the genetic diversity of a population is not only a result of habitat fragmentation, but also due to road mortality (Jackson & Fahrig 2011). The increased risk of crossing roads of high disturbance can reverberate through populations and declines in number of individuals, or reductions in mating, is of high concern. Fortunately, during the years data were collected for this study, no telemetered Puff Adders died as a result of crossings roads. However, in areas where crossing roads of high disturbance is the only option for dispersal to find resources, many different taxa fall victim to traffic (Bissonette 2002). Road kill in the form of several taxa have been document in unprecedented numbers worldwide as habitat change and road networks increase (Underhill & Angold 2000; Coffin 2007; Collinson *et al.* 2015; Kang *et al.* 2016). In cases where males are travelling further in search of females, as is the case with Puff Adders, the more mobile sex is at a higher risk of road mortality. This could skew sex ratios and diminish populations (Steen & Gibbs 2004; Gibbs & Steen 2005).

The effect of roads on wildlife is heavily concentrated around road kill data (Coffin 2007) or experimental set-ups and very few studies have observed movement patterns and the influence of roads on these patterns. My study has added to the few that integrate natural movement patterns and the implications of road infrastructure. The substantial number of telemetered individuals used in my study provides for a realistic interpretation of natural movement patterns and the effects of roads on these. However, only point localities were recorded in this study and straight-line movements were thus assumed. This could have underestimated the number of road crossings and studies which track movement on a smaller temporal scale would be advantageous. The CRW is modelled from existing movement paths and the majority of Puff Adders in this study had roads intersecting their home ranges. Although the walks are random due to using frequency distributions from movement paths and numerous iterations, it would be valuable to use movement patterns from Puff Adders that live in roadless habitats to use as a more appropriate null model.

Roads are essential for human movement but the effect on wildlife needs to be better understood. Overall my study provides a novel interpretation of wildlife-road relationships. The factors that affect road crossings in Puff Adders give insight into movement patterns of many other animals and the consequences of road crossings can be adverse for a variety of populations within an area. Through analysing the movement ecology of species appropriate management and conservation techniques can be applied. Mitigating the effects of roads on movement patterns can promote genetic diversity and overall biodiversity within an area. With the knowledge of drivers for increased movement, wildlife can be successfully conserved if appropriate culverts, fencing, and overall mitigation management are implemented in systems where road networks effect the natural environment.

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CHAPTER FOUR

A SYNTHESIS: THE IMPACT OF ROADS ON THE SPATIAL ECOLOGY OF PUFF ADDERS (*BITIS ARIETANS*)

The effect of roads on Puff Adder movement is not always negative, contrary to much of the literature on wildlife-road interactions. Being closer to, and crossing roads more often than random shows how these snakes exploit this change to their environment. Puff Adders appear to be using roads when foraging as road verges and roads may be providing predation opportunities for these ambush foragers. Roads have affected the behavioural and spatial ecology of this species and understanding and acknowledging the effect of roads on wildlife movement patterns can lead to effective management and conservation efforts. However, the wildlife-road interaction is complex and has several dynamic variables that create indirect and direct effects. The lack of correlation between number of crossings and proximity to roads overall for Puff Adders could also be a result of this complex interaction (Pearsons' correlation: $n = 109$; $r = 0.11$; $p = 0.24$). Each of the dynamic variables need to be taken into consideration when trying to accurately assess the effect of roads on movement patterns.

The movement of Puff Adders differed significantly from random data that were modelled off a 'roadless' environment. Individuals were found, on average, to be closer to roads with the majority of individuals being within 0 to 50 m from the road edge compared to random points being mostly within the 50 to 100 m range. This, along with the increased number of road crossings compared to a random walk model, illustrates how the road network within Dinokeng Game Reserve (DGR) is not a barrier to movement of Puff Adders. These relatively unique findings may be a result of many of the roads within the DGR being narrow and infrequently disturbed by vehicles as much of the wildlife-road literature is focussed around frequently used roads such as highways or paved roads which results in high avoidance numbers (Rudolph *et al.* 1994; Ashley & Robinson, 1996; Andrews & Gibbons 2005; Herrmann *et al.* 2017). Row *et al.* (2007), similarly to my study, radio tracked 105 Black Ratsnakes (*Elaphe obsoleta*) over 8 years and compared these movements to random and found that roads were not avoided either. However, they extrapolated road crossing data and the few mortalities that occurred to project future effects which showed to have a significant effect on population viability in the long term. Although there were no road mortalities from telemetered snakes in my study, the findings from Row *et al.* (2007) suggest that, even if roads are not avoided, there are still potential risks. The positive relationship demonstrated in my study illustrates how roads are not always fragmenting or hindering wildlife movement patterns in the short term and that there should be more diversity when researching this current and global topic because of the numerous aspects involved.

Sex in conjunction with season are some of the aspects that are influential for many species' behaviours and the same was found for Puff Adders. Males in mating seasons had higher number of road crossings as they increase their range in search of females. The movement of an individual is influenced by a variety of factors and begins with the motivation to move (Nathan 2008), and in the mating season, finding a mate and reproducing is priority and foraging efforts decrease (Shine 1979). In contrast, both male and females in non-mating seasons were closer to roads than in mating seasons, as forage took precedence over reproduction. The lack of crossings in non-mating seasons combined with the close proximity to roads during this period could be related to suitable foraging sites created along road edges. These sites could provide sufficient resources so there is no need to depart and cross to another foraging habitat (Siers *et al.* 2014). A combination of intrinsic factors influences the motivation to move, concurrently with extrinsic variables which effect these movements too.

Both proximity to roads and number of crossings were significantly different across the years that the Puff Adders were tracked. The variability in these data did not statistically correlate to climatic patterns, however a pattern between mean distance to road and rainfall data may be emerging. It would be advantageous to have accurate climatic data for the exact region to further test if there is a possible correlation. When observing the descriptive statistics, years with low rainfall were the same years in which snakes were farthest from roads, conversely years with the highest average rainfall were the same years where Puff Adders had the closest proximity to roads. Road verges have been observed as novel and attractive microhabitats for Puff Adder prey items, such as amphibians and small mammals (Glaudas *et al.* 2017), as they create suitable breeding and movement corridors for these taxa (Andrews & Jochimsen 2007; Redon *et al.* 2015). Road sides may create a profitable foraging site for ambush predators when environmental conditions are favourable. For example, high rainfall promotes breeding in amphibians (Marsh 2000) and these snakes have shown to decrease distance travelled in response to increased feeding (Glaudas & Alexander 2017). The combination of these factors supports the idea that Puff Adders would be attracted to foraging ‘hot spots’, especially when there are favourable conditions. The differences across mean number of crossings per year may also be affected by a myriad of extrinsic variables such as other anthropogenic changes to the ecosystem or even introduction of new species, both of which have occurred over the period that snakes were tracked and within the DGR (Dinokeng Game Reserve – The Official Site 2017). Consequently, any environmental change may alter and influence wildlife patterns.

Finally, the road characteristics were also influential to movement patterns. The effects of different road classes on proximity was less obvious than the effect on number of crossings. This could be due to proximity data being influenced more so by road edges and their specific characteristics rather than the road itself. In contrast, there was clear support that Puff Adder movement is not hindered by narrow roads with homogenous characteristics to the surrounding environment. Snakes crossed narrow roads, on average, nearly six times more often than their average crossings for road classes 3, 4 and 5. The latter classes have much higher disturbance levels in terms of traffic and are wider than road classes 1 and 2. The reduced number of crossings of Puff Adders across roads classes with increased vehicular usage and wider, and potentially risky, spaces shows how influential different types of roads can be on movement patterns. These results concur with most of the research on the effect of road types on different animals. Roads that are wider are more challenging to traverse and combined with increased traffic, and the added noise, chemical, and light pollution that accompanies this, are generally avoided by wildlife (Reijnen *et al.* 1995; Forman & Alexander 1998; Brock & Kelt 2004; Robson & Blouin-Demers 2013).

The effects of roads on Puff Adder movement depends on an array of dynamic variables. Overall, there are intrinsic motivations that alter movement patterns which then are in turn affected by extrinsic factors. The wildlife-road interaction has been a prominent topic as human population increases and will continue to grow in complexity with the variety of landscapes that road networks infiltrate along with the diversity of species' habitats they change. My study highlights the positive relationship wildlife may have with roads and demonstrates the importance of researching different taxa and sites to create a more informative and realistic approach to a continually growing topic in conservation.

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