Monitoring of large carnivores in Botswana for conservation management

By
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Doctor of Philosophy (Wildlife Management)

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Supervisor: Prof. M.J. Somers

DECLARATION

I, Christiaan Willem Winterbach, declare that this thesis, which I hereby submit for the degree Doctor of Philosophy (Wildlife Management) at the University of Pretoria, is my own work and has not previously been submitted by me for a degree at this or any other tertiary institution.

SIGNATURE:

DATE:

22 November 2019

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ABSTRACT

Assessing the range, population size and trend of large carnivores are required for conservation management at global to local scales. Survey techniques should be precise with low bias, detect population trends and cover large areas efficiently. Track surveys may be used to monitor populations of lion (*Panthera leo),* leopard (*Panthera pardus*), spotted hyaena (*Crocuta crocuta*), brown hyaena (*Parahyaena brunnea*), cheetah (*Acinonyx jubatus*) and African wild dog (*Lycaon pictus*) in Botswana. Call up surveys is an alternative to survey lion and spotted hyaena.

I recalibrated the published relationships between track density and true density. Linear regression through the origin was justified over linear regression with intercept for all models tested. Models through the origin fitted better and Akaike Information Criteria showed that these models were more robust.

A review of the track survey method clarified ambiguous terminology and inappropriate analyses of data. This allows others to replicate track surveys in a way that improves inter-survey comparisons of results. I calculated species-specific calibrations to estimate confidence intervals of track densities from track incidence frequencies, and thus carnivore densities, for the entire southern African large carnivore guild.

Calibration of carnivore response is required to estimate density from call up surveys. There correlation between the mean number of lions per calling station and reference density was significant ($P < 0.05$). Response probability did not differ by sex and age among surveys (call duration 90 minutes). Site-specific calibrations performed better than published calibrations. Call up surveys are unlikely to detect population trends in the Okavango Delta due to the high coefficient of variation (>200%).

I collated data from various sources and conducted track surveys to assess brown hyaena distribution and density across land uses in Botswana. Density ranged from 0 brown hyaena / 100 km^2 in parts of northern Botswana, to 2.94 (2.16–3.71) brown hyaena / 100 km^2 in the southern Central Kalahari Game Reserve. The estimated brown hyaena population is 4642 (3133–5993). Agricultural land in Botswana is important to link populations in South Africa, Namibia and Zimbabwe.

In 2005, Botswana implemented a minimum age threshold strategy to manage sustainable trophy hunting of male lions. I evaluated tooth: pulp width and area ratios to estimate age of trophy-hunted male lions in Botswana as a *post hoc* monitoring of compliance (2005 – 2007) to the 6-year minimum-age threshold. I used digital radiographs of the second premolar for measurements and cementum annuli analysis to calculate reference ages. The linear regression $y = 0.990 x + 2.512$ ($r^2 = 0.647$), between the tooth root area: root pulp cavity area ratio (R: RPA) and reference ages < 10 years ($F_{1, 23} = 42.244$, $P < 0.001$), was used to estimate age from R: RPA ratios. The 95% confidence intervals of age estimates from R: RPA ratios for the reference age classes 4 - 5 years and 6 - 7 years were mutually exclusive, showing the methods ability to distinguish between animals older and younger than the minimum age threshold.

Wildlife tourism can provide economic incentives for conservation. I evaluated tourism potential in the Northern Conservation Zone of Botswana, using wildlife biomass and diversity estimates from aerial survey data as indicators of tourism potential. Areas used for High Paying Low Volume tourism had significantly higher mean wildlife biomass and wildlife diversity than the areas avoided for this type of tourism. Only 22% of the Northern Conservation Zone has intermediate to high tourism potential. Tourists will have significantly better wildlife sightings in areas with high tourism potential compared to low potential areas. Although the largest part of the Northern Conservation Zone has low tourism potential, it has a high conservation value. Alternative conservation strategies should be developed to complement the economic incentive provided by wildlife-based tourism in Botswana.

Note on text

The layout of this thesis includes a general introduction and chapters 2-7 as stand-alone papers. Reference to figures, tables, and the literature was kept in the format as required by the different journals in which the manuscripts were published.

Supplementary data provided during publication of the manuscripts in the different journal were excluded from this text.

Chapters 2, 5 and 6 have been published as follows:

Chapter 2:

Christiaan W. Winterbach, Sam M. Ferreira, Paul J. Funston, Michael J. Somers. 2016. Simplified large African carnivore density estimators from track indices. *PeerJ* 4:e2662. 10.7717/peerj.2662

Chapter 5:

Christiaan W. Winterbach, Glyn Maude, Gosiame Neo-Mahupeleng, Rebecca Klein, Lorraine Boast, Lindsey N. Rich, Michael J. Somers. 2017. Conservation implications of brown hyaena (*Parahyaena brunnea*) population densities and distribution across landscapes in Botswana. *Koedoe* 59(2), a1441. 10.4102/koedoe.v59i2.1441

Chapter 6:

Christiaan W. Winterbach, Caroline Whitesell, Michael J. Somers**.** 2015. Wildlife abundance and diversity as indicators of tourism potential in Northern Botswana. *PLoS ONE* 10(8): e0135595. 10.1371/journal.pone.0135595

Chapter 3 clarifies the data analysis of track surveys and provides calibrations to improve confidence limits of carnivore density estimates from track indices. Chapter 4 quantifies lion response to call up surveys in the Okavango Delta and compares that to other published response calibrations. The ability to detect population trends reliably is assessed. Chapter 6 evaluates the potential of northern Botswana for high-paying, low volume tourism as a financial incentive to conserve the important range of carnivores. Chapter 7 applies the pulp-tooth ratio to estimate the age of trophy-hunted lions in Botswana as part of monitoring compliance to the age threshold for trophy lions.

Ethics Note

Research activities in Botswana were done under the guidance of permits issued by the Ministry of Wildlife Environment and Tourism, Botswana.

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The price one pays for the privilege and excitement of working in a remote area such as northern Botswana, is limited contact with family and friends. My family and friends provided support and maintained long distance friendships; this was despite visits, emails and phone calls statistically described as the rare events of a Poisson distribution. The stochastic events of sharing the bush with them made it worthwhile.

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CHAPTER 1. GENERAL INTRODUCTION

1.1 Monitoring for conservation management

1.1.1 Conservation management

Estimating abundance is a cornerstone of wildlife biology, and conservation management of species (Harris *et al*. 2013; Katzner *et al*. 2011). Reliable measures of species' distributions, and their population densities and trends are required to assess their status and to identify threats, for example in plants (Bornand *et al*. 2014; Garrard *et al*. 2008; Garrard *et al*. 2013), birds (Bart *et al*. 2004; Wintle *et al*. 2005), marine mammals (Taylor *et al*. 2007), terrestrial herbivores (Barnes 2002) and carnivores (Karanth *et al*. 2011; Long *et al*. 2008; Ripple *et al*. 2014; Weise *et al*. 2017). These data should guide policy and conservation planning, identify conservation priorities and assess the success of conservation management (Bart *et al*. 2004; Durant *et al*. 2007; Zipkin *et al*. 2014).

Biodiversity loss has accelerated globally (Durant *et al*. 2007) and requires management at different scales, from impact mitigation of local development (Garrard *et al*. 2013) to range wide conservation plans spanning international boundaries (IUCN/SSC 2006; IUCN/SSC 2007). Large carnivores as a group are highly endangered, with range loss and the loss of intact carnivore guilds impacting on ecosystem functions and services (Wolf & Ripple 2018). Large carnivores are umbrella species for conservation (Di Minin *et al*. 2016), because they are wide ranging, occur at low densities and require large areas to be conserved.

Conflict between large carnivores and humans has a major impact on carnivore conservation (Winterbach *et al*. 2013). Expected land use changes will result in important range losses of threatened carnivores and reduce their effectiveness as umbrella species (Di Minin *et al*. 2016). It is not only land use change and increasing human populations that affect species, but also activities such as trophy hunting (Whitman 2006) and even recreation in conservation areas that may impact negatively on species (Kowalski *et al* 2015).

Due to the increasing land-use pressures on African conservation areas from a growing human population, expanding agriculture and large development projects, there is a need for monitoring programs (Caro 2016). However, protected area networks in Africa are underfunded and the wildlife populations declining (Lindsey *et al*. 2014; Scholte 2011). Large carnivores are difficult to study because they occur at low densities and range over large areas

(Alibhai *et al*. 2017; Balme *et al*. 2009; Belbachir *et al*. 2015; Henschel *et al*. 2016). Assessing the status of carnivore populations is hampered by a general lack of data to evaluate population size and trends based on repeatable survey techniques (Riggio *et al*. 2013).

The IUCN status of Africa's seven large carnivores are lion *Panthera leo* (Vulnerable), leopard *Panthera pardus* (Near Threatened), spotted hyaena *Crocuta crocuta* (Least Concern), brown hyaena *Parahyaena brunnea* (Near Threatened), striped hyaena *Hyaena hyaena* (Near Threatened), cheetah *Acinonyx jubatus* (Vulnerable) and wild dog *Lycaon pictus* (Endangered). The interactions among large carnivores, and with their prey species, are important components of biodiversity (Dalerum *et al*. 2008; Mills 2005) that need to be conserved. To conserve the intact large carnivore guild, a mosaic of high and low densities of the dominant competitors, lion and spotted hyaena, are required to provide refuge areas for the subordinate species (brown hyaena, cheetah and wild dog) (Creel & Creel 1996; Creel *et al*. 2001; Hayward & Kerley 2008; Marker & Dickman 2004; Winterbach *et al*. 2013).

There are few large areas left in Africa that are suitable for the conservation of the large carnivore guild, thus the conservation of the guild has higher priority in these areas than that of the individual species (Woodroffe & Ginsberg 1998). Botswana is a key country in Africa with the potential to conserve the intact carnivore guild for the long-term conservation of large carnivores (Winterbach *et al*. 2014). Botswana set aside 38% of its surface area for conservation in the form of protected areas and wildlife management areas (Department of Surveys and Mapping 2001). Significant numbers of large carnivores occur in Botswana (Winterbach & Winterbach 2003), including lion (Bauer *et al.* 2015), cheetah, wild dog (IUCN/SSC 2007), spotted hyaena and leopard (Jacobson *et al*. 2016). The largest population of brown hyaena occurs in Botswana (Wiesel 2015).

The Northern and Southern conservation zones in Botswana (Fig. 1-1) provide the potential to conserve the intact carnivore guild (Winterbach, Winterbach & Somers 2014). The Northern Conservation Zone contains a patchwork of high, medium and low prey densities. There are high prey densities in and around the Okavango Delta and along the Kwando/Linyanti/Chobe river system (Winterbach *et al*. 2014) with high densities of lions and spotted hyaenas (Cozzi *et al*. 2013; Winterbach & Maude 2015; Winterbach & Winterbach 2003). Low prey densities have been recorded in the dry parts of the Northern Conservation Zone with low densities of lion and spotted hyaena. A mosaic of low and medium densities of wild prey for lions and spotted hyaenas occurs in the Southern Conservation Zone (Winterbach *et al*. 2014) that support low to medium densities of both carnivores (Funston *et al*. 2001; Maude & Selebatso 2012; Mills 2015).

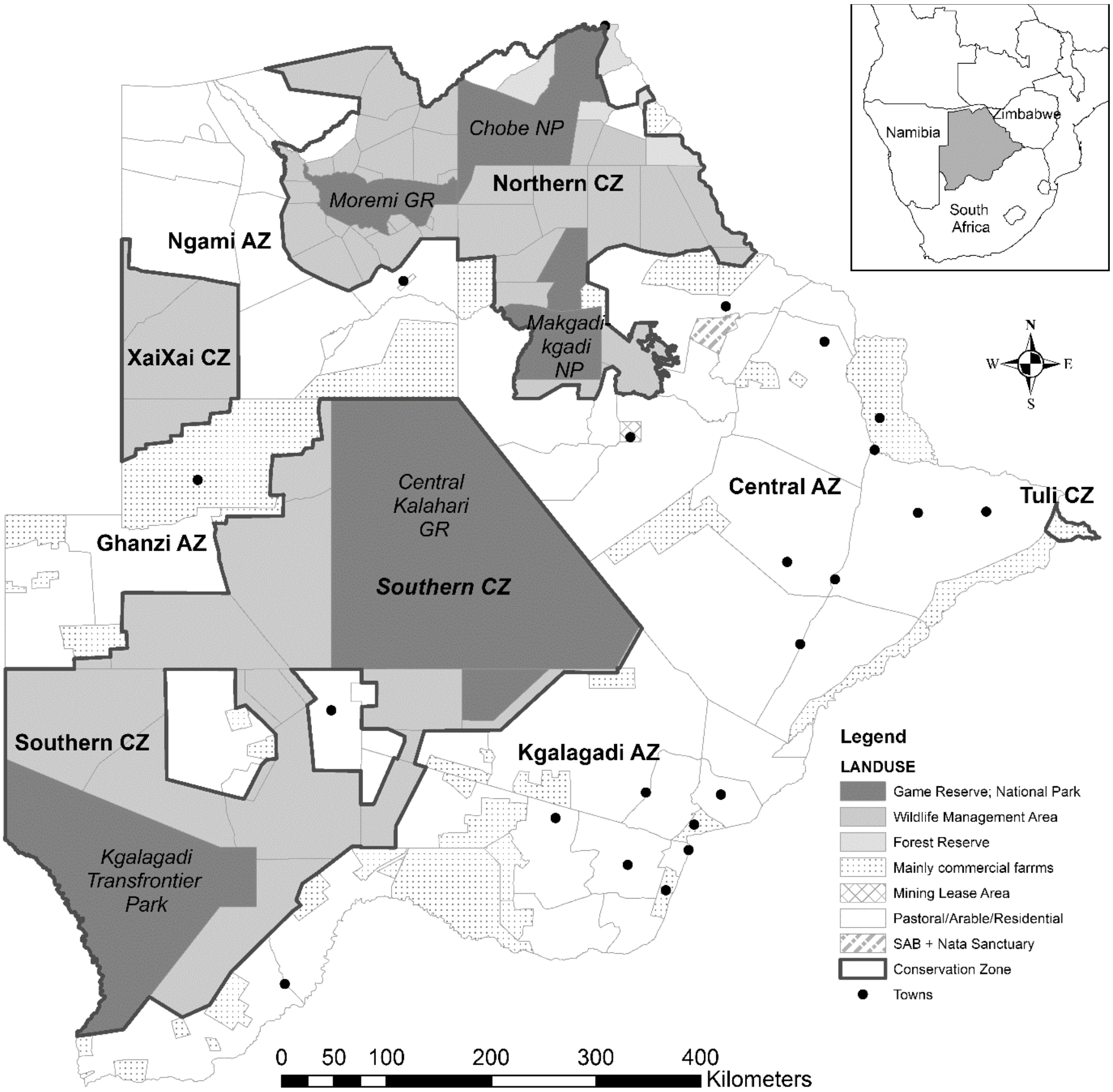

Figure 1-1 The conservation and agricultural zones in Botswana

The distribution and biomass of wild prey and livestock are important to understand the large carnivore guild (Winterbach *et al*. 2013). Lean season prey biomass determines carnivore abundance (Gittleman & Harvey 1982; Hayward & Slotow 2009; Van Orsdol *et al*. 1985) and prey base composition impacts on the composition of the carnivore guild. Aerial surveys and transect counts are monitoring techniques commonly used to estimate herbivore numbers and population trends. This provides biomass and population trend measures of the prey base for the different large carnivores. The Department of Wildlife and National Parks has conducted country wide aerial surveys since 1988, counting wildlife and livestock (Wint 2000). These data were used to predict landscape suitability for carnivores across Botswana (Winterbach *et al*. 2014; Winterbach *et al*. 2015).

The first lion surveys in Botswana were conducted on concessions in 1995 and 1996 using call up surveys (Winterbach & Vrahimis 1995; Winterbach & Winterbach 1996), followed by more extensive surveys in northern Botswana from 1998 to 2000. These surveys were partly used to guide quota setting for trophy hunting. Funston *et al*. (2001) conducted the first track surveys in the Kgalagadi Transfrontier Park.

These early surveys and data from intensive research projects (Hemson 2003; Maude 2005) contributed to the first country-wide estimates for the six large carnivores in Botswana as part of the process to draft the Predator Management Strategy for Botswana (Winterbach & Winterbach 2003). This process highlighted the paucity of density and distribution data for large carnivores required to guide conservation management on national and regional level, for example lion (IUCN/SSC 2006), and cheetah and wild dog (IUCN/SSC 2007), and to assist with the IUCN species assessments such as the brown hyaena (Wiesel 2015). The majority of subsequent studies and surveys used track counts to estimate carnivore densities (Bauer *et al*. 2014; Keeping & Pelletier 2014; Kent & Hill 2013; Maude & Selebatso 2012; Mudongo & Dipotso 2010). More recent carnivore studies in Botswana included camera trapping (Kent & Hill 2013; Rich *et al*. 2016).

To manage sustainable hunting quotas for lions from 2005 to 2007, the Department of Wildlife and National Parks implemented the minimum age threshold for lion hunting (Whitman *et al*. 2004). Field ageing criteria for lion (Whitman 2010) assist hunters in the selection of trophy animals older than the minimum threshold age. When Botswana implemented the age threshold for trophy lions, the need for post hoc monitoring of compliance became apparent. The premolar teeth from hunted specimens can be used for post hoc monitoring of compliance with the age threshold (Cheater 2006), but no calibration data for Botswana lions existed.

Conservation has costs at local, national and global scales that are partly compensated for by the benefits from nature and wildlife-based tourism (Balmford & Whitten 2003; Bookbinder *et al*. 1998; Lindsey *et al*. 2007b). This is particularly relevant in Botswana with 38% of the country dedicated to conservation. Tourism contributed approximately 15.0% to the Gross Domestic Product (GDP) of Botswana in 2011 (Statistics Botswana 2012) and provided an estimated 10.6% of all jobs in the country (WTTC 2008).

Wildlife-based tourism in Botswana occurs primarily in the form of High Paying Low Volume (HPLV) tourism and included sport hunting until 2012 (Office of the President 2012). HPLV tourism is concentrated in the Okavango Delta and Chobe National Park areas of the Northern Conservation Zone (WTTC 2008) with limited or no photographic tourism in the multiple use concessions outside the Okavango Delta. The photographic tourism industry perceived some parts of northern Botswana as not suitable for HPLV tourism. Phasing out sport hunting raised the question whether photographic tourism can replace sport hunting as an income generator in these areas. Although these areas are characterised by low wildlife densities (Winterbach *et al*. 2014), the whole area has a high conservation value. The Northern Conservation Zone is a key area to conserve the intact large carnivore guild and may be an important refuge area for wild dog (Winterbach *et al*. 2014). It also provides wet season range for elephants *Loxodonta africana* and buffalo *Syncerus caffer* (Chase 2011) and is part of zebra migrations (Bartlam-Brooks *et al*. 2011).

The presence of charismatic species and good visibility determine the suitability of areas for wildlife tourism (Goodwin & Leader-Williams 2000). The majority of international tourists to Africa are interested in seeing abundant wildlife with a strong focus on a few charismatic species like lion, leopard, elephant, buffalo and rhinoceros (*Ceratototerium simum* and *Diceros bicornis*) (Di Minin *et al*. 2013; Goodwin & Leader-Williams 2000; Kerley *et al*. 2003; Maciejewski & Kerley 2014). Although tourists' preference for charismatic species have been determined, there are no techniques to evaluate the wildlife tourism potential of an area based on species diversity and abundance.

1.1.2 *Selecting appropriate techniques for conservation management*

"If scientists wish to provide inputs into conservation management, policy, and practice, they need to understand and take into account management needs, priorities, and constraints." (Durant *et al*. 2007).

The cost of acquiring abundance data and the logistical challenges of a survey are determined by the size of the study site, accessibility of the area and the characteristics of the target species (Dénes *et al*. 2015; Harris *et al*. 2013; Karanth *et al*. 2011). The cost, efficiency of monitoring populations and logistical constraints are of concern, whether it be plants in Australia (Garrard *et al*. 2013), birds in North America (Bart *et al*. 2004), herbivores in Africa (Caro 2016), or carnivores around the world (Durant *et al*. 2007; Harris *et al*. 2013; Hines *et al*. 2010).

Protected areas in Africa are chronically underfunded, hampering wildlife monitoring and conservation management (Balmford *et al*. 2003; Caro 2016; Durant *et al*. 2007; Keeping *et al*. 2018; Lindsey *et al*. 2014; Scholte 2011). Therefore, estimates of abundance should be economical and efficient, producing appropriate results that are scientifically defensible (Harris *et al*. 2013). Intensive methods are typically not used for landscape-scale surveys due to the logistical constraints (Karanth *et al*. 2011).

Some techniques require complex statistical analysis and programming skills that limits accessibility to the broader audience (Dénes *et al*. 2015; Rich *et al*. 2019). This problem is more acute in Africa, where conservation is hampered by a lack of capacity among researchers, managers and resources (Caro 2016; Durant *et al*. 2007; Keeping *et al*. 2018; Lindsey *et al*. 2014; Scholte 2011).

The main constraint in Botswana is resources (Keeping *et al*. 2018). Based on the preceding sections, some of the needs in Botswana are:

1. Methods that are not resource intensive.
2. Surveys to monitor carnivore population distribution, density and trend.
3. Post hoc evaluation of compliance to age threshold set for trophy lions.
4. An objective evaluation of the potential for photographic safaris across the Northern Conservation Zone in Botswana is required to guide conservation strategies that will maintain conservation as the primary land use. Losing parts of the Northern Conservation Zone will compromise the ability to conserve the large carnivore guild intact.

1.2 Overview of survey techniques

A variety of techniques are available to survey mammals, each with its own requirements and assumptions to meet, strengths, weaknesses and limitations. Some are costly and time consuming (Elliot & Gopalaswamy 2017; Karanth *et al*. 2011), and some technically and computationally complex (Belant *et al*. 2016; Rich *et al*. 2019). Counting animals can be grouped in broad categories: aerial surveys, sign surveys and ground surveys consisting of line transects and point transects.

Techniques to survey African large carnivore populations developed over time. Early techniques included individual identification of lions (Pennycuick & Rudnai 1970), mass capture and marking (Smuts *et al*. 1977), and collaring of animals for intensive studies. These types of studies are expensive, invasive, and time consuming. Point transect surveys using audio calls to lure species (Mills *et al*. 2001; Ogutu & Dublin 1998; Thorn *et al*. 2010) and line transect surveys recording tracks (Funston *et al*. 2010; Keeping 2014) are techniques that may be suitable to monitor large carnivore populations at the landscape level. Recent survey developments include the use of camera traps (Karanth *et al*. 2011; Meek *et al*. 2016; Rich *et al*. 2016), track survey data (Henschel *et al*. 2016; Midlane *et al*. 2014) and sighting data (Elliot & Gopalaswamy 2017) to model imperfect detection.

1.2.1 Aerial surveys

Aerial survey is one of the standard techniques to count medium to large-bodied herbivores, especially sampling remote areas where ground surveys are not logistically feasible. Aerial surveys are expensive and may be difficult to sustain in the long term (Caro 2016; Gaidet *et al*. 2003). For example, the country wide aerial surveys in Botswana have reduced in frequency and portions covered during the past 30 years (Keeping & Pelletier 2014).

Undercounting is a major problem of aerial surveys (Jachmann 2002), and is more pronounced for the cryptic and smaller species (Jachmann 2002; Keeping *et al*. 2018). With standardization of the method, trends can be determined, but Jachmann (2002) considers aerial surveys of limited use for most species due to the variable undercounting bias of the technique. Aerial surveys are not suitable for carnivores due to their cryptic nature. Despite these shortcomings, aerial surveys are often the only data source available to assess wildlife population numbers and trends (Keeping *et al*. 2018).

1.2.2 Line transect surveys

Ground surveys are an alternative to aerial surveys that can reduce costs, equipment needs, and personnel constraints (Caro 2016; Gaidet *et al*. 2003; Keeping *et al*. 2018). Ground surveys have been conducted using vehicles, bicycles, elephants, horses and on foot (Caro 2016; Gaidet *et al*. 2003; Keeping *et al*. 2018; Wegge & Storaas 2009).

Data analysis using distance sampling (Buckland *et al*. 1993) is predominantly used to derive population estimates from line transect data and has been accepted as the standard monitoring protocol to monitor the tiger prey base in south Asia (Wegge & Storaas 2009). The alternative to distance sampling is to count all animals up to a maximum distance on either side of the transect and calculate a relative index of abundance (number of individuals/km). This eliminates the need for a rangefinder to record distance of each sighting from the transect (Caro 2016).

A minimum threshold of 60 to 80 observations per species is required for distance sampling to model the detection function (Buckland *et al*. 1993). Several studies failed to reach this minimum threshold (Ahlswede *et al*. 2019; Caro 2016; Keeping *et al*. 2018). This problem is more pronounced in dense woodland and forest habitats (Barnes 2002; Jathanna *et al*. 2003).

Carnivores are difficult to survey due to their elusive nature and low densities (Funston *et al*. 2010; Karanth *et al*. 2011). This limits the use of line transects, or other survey techniques relying on visual sightings of carnivores, to open habitats with good visibility. The studies of Durant *et al*. (2011) and Elliot & Gopalaswamy (2017) in Serengeti and the Masai Mara are examples of studies that visually recorded carnivores in open habitat. These approaches are very costly, for example Elliot & Gopalaswamy (2017) needed three vehicles, with a group of people on each vehicle, to drive around for three months in an area that is only 2200 km^2. They considered 65 cheetah sightings as a small sample size. In contrast, only two visual sightings of lions were recorded during 2018 while completing 2000 km of transect counts in the savannah woodland of northern Botswana (Winterbach, unpublished data).

1.2.3 Point transect surveys

Point transects, an alternative to line transects, are frequently used for bird surveys (Bart *et al*. 2004; Buckland *et al*. 1993). The method has been expanded to include bait and lures to attract animals to detection devices (Buckland *et al*. 2006; Omoya *et al*. 2013). Broadcasting sounds to lure target species to calling stations can be used for lions, spotted hyaenas, brown hyaenas and jackals (*Canis mesomelas*) (Ogutu & Dublin 1998; Mills, Juritz & Zucchini 2001; Thorn *et al*. 2010).

1.2.4 Sign surveys

Sign surveys of tracks and other signs are important tools assess carnivore distributions and populations, their habitat use and factors influencing their local presence at landscape scales (Karanth *et al*. 2011). Recording animal signs on line transects instead of visual observations can be more efficient and provide better data to determine density and distribution of species (Ahlswede *et al*. 2019; Fragoso *et al*. 2016). Track surveys can cover large areas, and multiple species can be surveyed on suitable substrates (Funston *et al*. 2010; Keeping & Pelletier 2014; Stander 1998).

These sign surveys include tracks, scat, track stations and camera traps with or without baits and lures (Long *et al*. 2008), but most of these methods do not address imperfect detections of animal signs (Karanth *et al*. 2011). Depending on the survey design, the data can be analysed for point or line transects. A relative abundance index can be calculated from sign surveys data (Long & Zielinski 2008) and from line transect data (Caro 2016).

Indices of abundance are generally unreliable for carnivores due to the low detection rates (Royle *et al*. 2008). Analysing presence/absence data excluding imperfect detection underestimates spatial distribution (Karanth *et al*. 2011; MacKenzie *et al*. 2002).

The use of relative indices is contentious, with the index varying in unpredictable ways with abundance (Elliot & Gopalaswamy 2017), and it is "unvalidated indices that rely on unsupported assumptions" (Hayward *et al*. 2015). In contrast, Caro (2016) considered population indices (individuals/km) for herbivores calculated from line transect counts, valid for the general purpose of detecting broad trends in savannah habitats.

Interpretation of indices are therefore not straight forward and may be disputed. This may delay implementation of conservation management, for example the debate whether dingoes can be used to reduce the impacts of foxes and cats in Australia (Hayward *et al*. 2015; Hayward & Marlow 2014; Nimmo *et al*. 2015). Long & Zielinski (2008) provide an overview of problems of abundance indices used with North American carnivores.

The track surveys following Stander (1998) and Funston *et al*. (2010) differ from general signs surveys (Allen *et al*. 1996; Evangelista *et al*. 2009; Karanth *et al*. 2011; Long *et al*. 2008) in two key aspects: data collected represents a time frame of 24 hours and the detection probability is approaching 1 (Funston *et al*. 2010; Stander *et al*. 1997). Hayward *et al*. (2015) highlighted the need to calibrate track indices against true density, in order to use it with confidence, as was done by Stander (1998) and Funston *et al*. (2010).

It is possible to estimate carnivore and herbivore densities from track indices and day ranges using the Formozov-Malyshev-Pereleshin (FMP) formula. This approach does not require calibration against known density (Keeping 2014; Keeping & Pelletier 2014). The extremely high encounter rates of mammal tracks on sandy substrates make it practical to assess the mammalian community above threshold body size and provide results that are as good and better than results obtained from distance sampling and aerial surveys (Ahlswede *et al*. 2019; Keeping *et al*. 2018).

1.2.5 Modelling imperfect detection

Results from biological surveys may be biased due to imperfect detections and thus misinform conservation management (Garrard *et al*. 2013; Karanth *et al*. 2011). Estimating detection probability requires temporal or spatial replication (Belant *et al*. 2016; Hines *et al*. 2010; Karanth *et al*. 2011). A variety of models have been developed that incorporate imperfect detection, for example, N-mixture models (Belant *et al*. 2016; Zipkin *et al*. 2014), random encounter models (Cusack *et al*. 2015) and spatially explicit capture-recapture (SECR) (Hines *et al*. 2010) that can even be used with unmarked or partially marked populations (Chandler & Royle 2013).

Recent advances in analysis resulted in a shift from hypothesis driven statistics to the use of large data sets and complicated quantitative models to investigate how ecological processes are affected by different mechanisms at landscape scales (Burnham & Anderson 2002; Garrard *et al*. 2013; Gerber *et al*. 2012; Hines *et al*. 2010; Karanth *et al*. 2011; MacKenzie *et al*. 2002; Royle & Nichols 2003; Zipkin *et al*. 2014). Despite this shift in statistical analysis, the required skills are not provided in most doctoral programs in the United States (Touchon & McCoy 2016) and the complexity of some models limits accessibility to the broader audience (Dénes *et al*. 2015; Rich *et al*. 2019).

Models can produce biased results (Cusack *et al*. 2015; Gerber *et al*. 2012) and there is a need to evaluate the limitations and reliability of the emerging models beyond simulation (Dénes *et al*. 2015; Palmer *et al*. 2018). For example, field studies provided support to estimate density of geographically open populations of black bear *Ursus americanus* with spatially explicit SECR models (Gardner *et al*. 2010; Obbard *et al*. 2010).

It is important to quantify the detection probabilities of species during flora and fauna surveys (Garrard *et al*. 2008), but this can be expensive, laborious and time consuming (Royle & Nichols 2003). Initially, this included intensive capture–recapture sampling of photographs or faecal DNA from individuals that typically are beyond the logistical constraints of landscape-scale surveys (Karanth *et al*. 2011). Recent developments to estimate detection probability include zero-inflated binomial models (Wintle *et al*. 2005), search-encounter models (Royle *et al*. 2011) and continuous time models (Borchers *et al*. 2014).

Detection probability of plant species can be estimated as time spent searching and applied to determine the minimum sampling effort (Bornand *et al*. 2014; Garrard *et al*. 2008). The multi-species approach using plant traits and observer characteristics in time-to-detection modelling may have application in other taxa to estimate detection probability (Garrard *et al*. 2013).

The use of camera trap capture /recapture data in animal surveys increased rapidly as the main data source for the above mentioned modelling approaches (Apps & McNutt 2018; Brassine & Parker 2015; Delgado *et al*. 2015; Gerber *et al*. 2012). Not only does the fitness for purpose of different camera trap brands and models vary, but camera setup also impacts on performance, introducing as yet unmeasured biases into detection probability estimates (Apps & McNutt 2018). Detection probability can be further compromised by animal responses such as trap happy or trap shy individuals (Meek *et al*. 2016).

1.3 Carnivore monitoring in Botswana

Track surveys and call up surveys have been used in Botswana to determine carnivore density and distribution (Bauer *et al*. 2014; Boast & Houser 2012; Cozzi *et al*. 2013; Funston *et al*. 2001; Keeping 2014; Mudongo & Dipotso 2010; Winterbach *et al*. 2002). Given the volume of work done with these two techniques, a detailed account of track surveys and call up surveys are presented below. This is followed by an overview of the pulp: tooth ratio method to estimate age of trophy hunted lions.

1.3.1 Track surveys

Researchers have been studying large carnivores from tracks and other signs for a long time, relying on the expertise of local trackers (Bothma & le Riche 1984; Eloff 1973). The level of expertise of Kalahari trackers was quantified (Stander *et al*. 1997) and used in a structured and replicable population survey technique about two decades ago (Stander 1998). The Kalahari trackers have a near-perfect record of identifying individual large carnivores from their tracks and the ability to reconstruct complex behaviours from the signs (Stander *et al*. 1997), and Funston *et al*. (2010) concluded “We were thus confident that our spoor indices were not confounded by observer bias”.

Stander (1998) and Funston *et al*. (2010) demonstrated strong relationships between track density and estimates of true carnivore densities for different large African carnivores.

Funston *et al*. (2010) provided the first calibrations that could be applied to large African carnivores. Substrate influenced the relationships between track density and true carnivore density. All species could be combined in a single model on sandy substrates for general application. Although Funston *et al*. (2010) provided calibrations for clay soils and sandy soils, Belant *et al*. (2016) could not estimate lion density and abundance from track data on clay soils in south-eastern Serengeti National Park, Tanzania.

More recently, researchers incorporated the daily movement ranges of animals into calibrations to estimate animal density, using the FMP formula (Ahlswede *et al*. 2019; Keeping 2014; Keeping *et al*. 2018; Keeping & Pelletier 2014). Data sets collected following Stander (1998) and Funston *et al*. (2010) (i.e. recording tracks of an individual only once in 24 hours), are incompatible with the FMP method where all track incidences of an individual crossing the line transect are recorded.

Carnivore track encounters are recorded along pre-defined line transects (typically roads or fire breaks) to measure mean distance between spoor incidences (track frequency) and to calculate track density (Funston *et al*. 2010; Stander 1998). Survey design includes spatial and temporal replication of transects which compromises the ability to derive confidence intervals for track densities (Funston *et al*. 2010).

The same individual should not be recorded more than once in a 24-hour period. Mean distance between incidences (termed “spoor frequency” by Stander (1998)) is the number of km per track incidence and track density is the number of individual tracks per 100 km of sampling (Funston *et al*. 2010; Stander 1998). Multiple animals (social groups) moving as a group are recorded as a single event (Stander 1998). Track frequency can provide a reliable estimator of confidence intervals for track density and thus population density, given appropriate species calibration for the relationship between track frequency and track density (Funston *et al*. 2010).

The general model on sandy soils (Funston *et al*. 2010) yields negative density estimates below track densities of 0.4 tracks / 100 km because it follows the conventional approach of a linear model with intercept (Eisenhauer 2003; Quinn & Keough 2002; Sokal & Rohlf 1995). Boast and Houser (2012) solved this issue by using the leopard calibration from Stander (1998), a linear model through the origin, to estimate leopard densities at low track densities. Williams *et al*. (2016) opted for the lion and wild dog model from Stander (1998) to estimate all carnivore densities, despite this model being based on only four data points.

There are circumstances when regression through the origin is appropriate. Therefore, I applied the guidelines of Eisenhauer (2003), Quinn & Keough (2002) and Sokal & Rohlf (1995) to the Funston *et al.* (2010) dataset to determine whether a linear model fitted through the origin is more appropriate than the linear model with intercept (Chapter 2).

The basic track count method described by Stander (1998) seems straightforward and simple, regardless of the calibration used to estimate carnivore density. However, the literature on track surveys shows flawed logic mistakes, perplexing terminology and erroneous data analyses. Two studies calculated true density without considering the time that individuals spent outside the study area, resulting in an over estimate of true density (Gusset & Burgener 2005; Houser *et al*. 2009). Funston *et al*. (2010) provide appropriate procedures to estimate true density that allows for the time that study animals spend outside the study area.

Track frequency (mean distance between track incidences), as defined by Stander (1998), has been calculated as the distance between track incidences, correctly treating a group of animals as one incidence (e.g. Bauer *et al.* 2014; Kent and Hill 2013). Others treated animals in the group as separate incidences, with a zero distance between detections or did not describe the way that track frequency was calculated (see: Balme *et al*. 2009; Boast & Houser 2012; Houser *et al*. 2009; Williams *et al*. 2016).

There are inconsistencies in the calculation and reporting of confidence intervals for carnivore density estimates or a lack of reporting confidence intervals altogether. Funston *et al*. (2010) proposed to estimate confidence intervals for track density from the mean distance between track incidences, and as an example provided lion calibrations of the mean distance between track incidences versus track density. The lack of calibrations describing the relationship between track density and track frequency (mean distance between incidences) for other carnivores probably contributed to the fact that none of the recent studies (Bauer *et al.* 2014; Boast & Houser 2012; Houser, Somers & Boast 2009; Kent & Hill 2013; Midlane *et al.* 2015; Williams *et al.* 2016; Winterbach *et al.* 2017) followed this proposed method of calculating confidence intervals.

Stander (1998) and Funston *et al*. (2010) plotted what they referred to as the coefficient of variation (CV) versus sample size, to determine the required number of track incidences for sampling resulting in a CV < 20%. Midlane *et al.* (2015) were the first to point out the confusion surrounding the CV in the track survey literature. This confusion originated from Stander

(1998), who used the term CV to describe the relative standard error (SE/mean) instead of the traditional CV (standard deviation/mean) (Sokal & Rohlf 1995; Quinn & Keough 2002).

SE gives the expected variability of repeated estimates of the mean, in other words, how confident you are about the mean. SD measures of the variability of the sample. Standard deviation (SD) and CV stabilise as sample size increase, while SE and relative standard error (RSE) decrease, because $SE = SD / \sqrt{n}$ (Sokal & Rohlf 1995; Quinn & Keough 2002).

This erroneous terminology is inconsequential to determine the minimum sample size of track incidences required. When you need to use CV in other calculations, such as Gerodette's inequality (Gerrodette 1987), but substitute it with RSE, the impact is massive. For example, if your RSE is 20% with a sample size of 25, the CV is 100%, five times higher.

Gerodette's inequality is a method to assess the power to detect a population trend with linear and exponential regression, when a trend is present (Gerrodette 1987). This method has been applied to population estimates of elephants, marine mammals and track surveys of carnivores (Barnes 2002; Funston *et al*. 2010; Taylor *et al*. 2007). Substituting RSE for CV will result in an overestimation of the power to detect trends using Gerodette's inequality (Gerrodette 1987).

When testing any null hypothesis, there is a risk of making Type I or Type II errors. A Type I error (α) occurs when the null hypothesis is rejected when there is no effect. A Type II error (β)wrongly accepts the null hypothesis when there is an effect. Related to Type II error is the statistical power (β-1) of a test, the confidence in detecting an effect if one existed (Quinn & Keough 2002). In terms of monitoring animal populations, the power is the probability to detect a population trend when it is present (Gerrodette 1987).

Designing a monitoring program, the number of samples required, the precision of the samples and the probability of detecting a trend are key questions to avoid wasted time and effort on a program that is unlikely to provide useful information. Interpreting results with power analysis shed light on how large a change could have been detected and the probability of detecting it. Five parameters are interlinked in the power analysis of changes in abundance that take place with constant increments (linear models) or at constant rates (exponential models) (Gerrodette 1987): power (β-1), sample size (*n*), probability of a Type I error (α), the rate of change (*r*) and the coefficient of variation of the estimate of abundance (CV).

The power is conventionally set at 0.80 (Bart *et al*. 2004; Quinn & Keough 2002) and most wildlife surveys aim for a CV < 20%; above this threshold is not considered suitable for monitoring trends (Boulanger *et al*. 2002; Keeping *et al*. 2018). The CV of line survey data increase as population abundance decrease, and thus the probability to detect the population change declines (Barnes 2002; Taylor *et al*. 2007).

Funston *et al*. (2010) concluded that most carnivore estimates derived from spoor indices had a high precision and that a 10% change in population abundance could be detected with only two surveys 1 year apart. In cases with less precise estimates, there could be 6 years of uncertainty, requiring three surveys up to 3 years apart to detect the trend (Funston *et al*. 2010). Unfortunately, these conclusions were based on substituting RSE for the actual CV, thus over estimating the power to detect a population trend.

Cumulatively, these methodological issues raise concern over the general application of track surveys for population monitoring, the power to detect population trends, while also impeding the comparability of results derived from multiple sites, surveys, and species to assess populations at national or regional scales. Clear guidelines on how to analyse track survey data and calibrations of the mean distance between track incidences versus track density to calculate confidence intervals for track density are required (Chapter 3).

Serious logical mistakes and computational stakes in track surveys have been pointed out above. Track surveys are far less complicated than occupancy modelling techniques that can require complex statistical analysis and programming skills (Dénes *et al*. 2015; Rich *et al*. 2019), skills that should be included in doctoral programs (Touchon & McCoy 2016). Therefore, it will be highly unlikely to successfully implement routine monitoring programs that require advanced modelling techniques.

1.3.2 Call up surveys

Smuts *et al*. (1977) used sound and bait to lure lions for mass capture. Lion surveys conducted in Botswana in 1995 and 1996 used sound and bait to attract lions and achieve individual recognition from reference photos using whisker spots (Pennycuick & Rudnai 1970) to establish minimum numbers. The 1998, 1999 and 2000 lion surveys in Botswana included basic corrections for the proportions of lions responding to call up surveys (Winterbach *et al*. 2002). Ogutu and Dublin (1998) developed a calibration of lion response in the Masai Mara to correct density estimates for non-response.

Two key calibrations are required to estimate carnivore density from calling station data (Cozzi *et al*. 2013; Ferreira & Funston 2010; Mills *et al*. 2001; Ogutu & Dublin 1998): i) the probability of individuals responding, and ii) the response radius covered around the calling station. Respectively, these correct for the non-response of individuals and determine the area surveyed. Only Omoya *et al*. (2013) implemented lure count analysis with point transect sampling methods (Buckland *et al*. 1993; Buckland *et al*. 2006), to estimate the effective radius of response: the radius where animals closer to the lure and not responding, equals the number of animals responding that are further away. Although researchers should test how well density estimates from call up surveys with response calibrations reflect true densities (Ogutu *et al*. 2005), it has not been done.

Response probabilities of lions and spotted hyaena decline with distance from calling station (Brink *et al*. 2013; Cozzi *et al*. 2013; Ferreira & Funston 2010; Mills *et al*. 2001; Ogutu & Dublin 1998; Omoya *et al*. 2013). In contrast to some studies (Brink *et al*. 2013; Ferreira & Funston 2010; Whitman 2006), Ogutu and Dublin (1998) found that lion response probability is constant regardless of sex and age. Other factors that can contribute to the heterogeneity in calibration results among studies include the type of call used (prey animal in distress, hyaena vocalizations, lion sounds and lion roars) (Cozzi *et al*. 2013; Mills *et al*. 2001), presence of migratory prey species (Ogutu & Dublin 1998), and thermal inversion (Garstang *et al*. 1995; Larom *et al*. 1997). In the short term lions did not readily respond more than once to calls (Belant *et al*. 2016; Ferreira & Funston 2010). This change in response probability is a concern for studies with temporal replication.

1.3.3 Threshold minimum age for trophy hunting of lions

Trophy hunting creates economic incentives for conservation over vast areas of Africa and lion is a high-value species in the trophy hunting industry (Lindsey *et al*. 2007a). Due to the potential negative impact of trophy harvesting on the long-term survival of hunted lion populations, sustainable harvest strategies are required (Becker *et al*. 2013; Croes *et al*. 2011; Loveridge *et al*. 2007; Packer *et al*. 2009).

Trophy hunting of male African lions can be managed at a sustainable level by targeting males above a minimum age threshold of 5-years old (Whitman *et al*. 2004). Botswana implemented a 6-years old minimum threshold. Professional hunters evaluate the age of male lions in the field using physical characteristics (Whitman 2010). Although conservation managers are only able to monitor hunters' compliance to the age threshold strategy after the lion has been shot, the information on the age of hunted lions can be used to inform future hunting quotas to ensure sustainability.

Two tooth characteristics, the deposition of cementum annuli and the closure rate of the pulp cavity, have been used as a proxy for age in archaeology (Jones 2012), vertebrate paleontology (Binder & van Valkenburgh 2010), forensic science (Paewinsky *et al*. 2005), and wildlife studies (Smuts *et al*. 1978; Van Aarde 1983). Cementum annuli analysis involves counting the number of growth layers (annuli) deposited in the cementum of the tooth that correlates with seasonal changes in nutrition (Asmus & Weckerly 2011; Lieberman 1994). Annual increments of cementum annuli were observed in lions (Cheater 2006; Smuts *et al*. 1978).

The pulp cavity closure rate is an alternative to cementum annuli analysis for ageing animals. Due to the deposition of secondary dentine along the wall of the pulp chamber, the pulp cavity becomes progressively smaller with age throughout the life of the tooth (Morse *et al*. 1991). The incremental deposition of dentine in lion ceases only when the dental pulp is exposed, and necrosis begin (Smuts *et al*. 1978). The pulp cavity:tooth ratio can be used to separate lions into age classes (Cheater 2006; Meachen-Samuels & Binder 2010; Smuts *et al*. 1978).

Cementum annuli analysis requires trained and experienced interpreters (Calvert & Ramsay 1998; Rolandsen *et al*. 2008) and factors such as tooth type used, the type of sectioning and staining, sample size, and interpretation of annuli and environmental factors may impact on accuracy (Gasaway *et al*. 1978; Goodwin & Ballard 1985; Hess *et al*. 2011; Landon *et al*. 1998; Medill *et al*. 2009).

Calculating the pulp cavity:tooth ratio using radiographs is fast, easy to perform, and less expensive, but differences in pulp cavity:tooth ratio may occur between upper and lower jaw teeth (Knowlton & Whittemore 2001) and between males and females (Kershaw *et al*. 2005). In both the cementum annuli analysis and pulp cavity:tooth ratio methods, the accuracy of age estimates decreased with increasing age of the animal (Gipson *et al*. 2000; Rolandsen *et al*. 2008) and with the pulp cavity:tooth ratio method (Landon *et al*. 1998; Thomson & Rose 1992). Smuts *et al*. (1978) and Cheater (2006) showed annual increases in cementum annuli for lions in southern Africa. With the lack of known-age reference material from free-ranging lions, the cementum annuli analysis method is the best age reference available to calibrate the pulp cavity: tooth ratio as an index of age for lions in Botswana.

1.4 Study area

The Republic of Botswana is a landlocked country of 582 000 km^2, bordering Namibia, South Africa, Zimbabwe and Zambia. Altitude ranges from 515 to 1491 m above sea level. Most of Botswana is arid to semi-arid, with the Kalahari ecosystem occupying approximately 82% of the country. Rainfall is erratic, and the mean rainfall ranges from 250 mm per year in the south-west to over 650 mm in the north-east (Department of Surveys and Mapping 2001). Over 90% of rainfall occurs in the summer months between November and April. Apart from the Okavango Delta and the perennial Kwando/Linyanti/Chobe river system, the only other surface water occurs in rivers and pans during the rainy season (Department of Surveys and Mapping 2001). The mean minimum temperatures range from 5°C in July to 19°C in January with the mean maximum daily temperatures ranging from 22°C in July to 33°C in January (Department of Surveys and Mapping 2001).

Vegetation over most of the country is shrub and tree savannah, which is classified as Sandveld (Department of Surveys and Mapping 2001). The Hardveld vegetation types are associated with hills and rocky outcrops in the eastern part of Botswana. The northern conservation zone comprises the wetland of the Okavango Delta, Sandveld, mopane *Colophospermum mopane* dominated vegetation types and limited Miombo woodland in the north-east. The Okavango Delta consists of a mosaic of islands, waterways and seasonal floodplains (Department of Surveys and Mapping 2001).

Seventeen percent of Botswana is fully protected as designated national parks and game reserves, and a further 21% is partially protected as designated Wildlife Management Areas (WMAs) (Fig. 1-1). Only 5% of the country is classified as urban. The balance of 57% comprises uncultivated rangeland consisting of approximately 5% freehold land, 25% state land and 70% tribal/communal grazing land (Department of Surveys and Mapping 2001). Commercial livestock production occurs on freehold, state land and tribal land.

Based on primary land use, Winterbach *et al*. (2014) identified four conservation zones and four agricultural zones in Botswana (Fig. 1-1). The conservation zones consist of the large northern conservation zone and southern conservation zone, and the two smaller conservation zones of XaiXai and Tuli. The northern conservation zone includes Chobe National Park, Moremi Game Reserve, Nxai Pan National Park, Makgadikgadi Pans National Park and the associated WMAs. The Central Kalahari Game Reserve, Kgalagadi Transfrontier Park and associated WMAs form the southern conservation zone (Fig. 1-1). The agricultural zones are Ngami, Central, Ghanzi and Kgalagadi (Fig. 1-1).

Livestock (mainly cattle) rearing forms 70% – 80% of the contribution of the agricultural sector to the GDP (Botswana Ministry of Agriculture 2011). The national herd numbered 2.6 million cattle, 1.8 million goats and 300 000 sheep in 2012. Traditional cattle posts on communal grazing land are the most common livestock production system (Botswana Ministry of Agriculture 2011). There were 109 ranches producing game or cattle and game in 2013; these cover approximately 11 500 km^2 (Boast 2014). Approximately half of the population of 2 million people live in rural villages and small settlements and are thus partially or fully dependent on livestock for their livelihoods (Central Statistics Office 2014).

1.5 Scope and objectives

The focus of this study was on the tools used for monitoring large carnivores for conservation management. This included landscape scale surveys to determine distribution range, population size and population trends of large carnivores for conservation strategies and management. Conducting call up surveys and track count surveys, and reviewing peer reviewed literature, highlighted the challenges and limitations of these survey techniques. Clarification, refinement, calibration and evaluation of these techniques were necessary to guide practitioners in conservation management. Post hoc monitoring of compliance to the minimum age threshold for trophy lions, set by the Department of Wildlife and National Parks, was important to assist setting sustainable quotas and manage compliance.

Stopping sport hunting in Botswana after 2012, raised the concern that some concessions that only conducted hunting, would not be suitable for photographic tourism. In future this may result in pressure to change the land use to livestock. Losing key parts of the Northern Conservation Zone due to a lack of economic returns will pose a significant risk to carnivore conservation in Botswana. Objective assessment of tourism potential would help to guide conservation strategies for these areas perceived as marginal tourism areas. No techniques are available to assess the tourism potential, but aerial and line transect survey data were available to develop methods.

Specific objectives:

1. To evaluate track surveys as a monitoring tool for large carnivore populations:
    a. Revise the Funston *et al*. (2010) calibration to provide simplified indices that don’t yield negative estimates at low track densities (Chapter 2).

   b. Provide a national population estimate of brown hyaena in Botswana for conservation management planning (Chapter 5).
   c. Calibrate mean distance between track incidences against track density to estimate confidence intervals of track density (Chapter 3).
   d. Provide clear guidelines and a clear analysis template for analysing track survey data (Chapter 3).
   e. Assess suitability of track surveys to monitor carnivore population trends (Chapter 3).
2. To evaluate call up surveys as a monitoring tool for lion populations (Chapter 4):
   a. Evaluate the impact of call duration on response probability.
   b. Calibrate the mean effective response distance of lions following point transect sampling with lures (Buckland *et al*. 2006).
   c. Test the published calibrations with data from lion populations with known density.
   d. Assess the suitability of call up surveys to monitor carnivore population trends.
3. To assess the photographic tourism potential of northern Botswana (Chapter 6):
   a. Create a method to assess the tourism potential in terms of diversity and biomass, using existing aerial survey data.
   b. Create a method to compare the expected tourist experience between areas with low and high potential for wildlife based tourism, using existing line transect survey data.
4. To assess the compliance with the minimum age threshold set for lion trophy hunting in Botswana, using the pulp-tooth ratio to estimate the age of trophy-hunted lions post hoc (Chapter 7):
   a. Calibrate the pulp-tooth ratio for lions in Botswana.
   b. Evaluate the compliance to the minimum age threshold.

Chapter 2 revised the Funston *et al*. (2010) calibrations with regression through the origin. The original article tested for the impact of substrates and differences between species, thus in Chapter 2 I only recalculated the established regressions through the origin instead of with the intercept. This solved the negative population estimates at low track densities.

In Chapter 5 the revised and simplified indices were applied to provide the first detailed estimates of brown hyaena across different landscapes in Botswana, outlining conservation management implications respectively. Scientific journals are often not accessible to conservation managers and scientific publications can be difficult for non-scientists to understand (Durant *et al*. 2007). The target audience was conservationists working in government and NGOs, and political decision makers. Therefore, the aim was to keep this peer reviewed publication to be accessible to non-scientists. The research hypotheses were not explicitly stated but the designs of data analysis were based on testing the following hypotheses:

H_0: Proportions of farmers reporting the presence/ absence of brown hyaena did not differ among commercial livestock farms, cattle posts and game farms in the agricultural zones.

H_1: Proportions of farmers reporting the presence/ absence of brown hyaena differed among commercial livestock farms, cattle posts and game farms in the agricultural zones.

Applying the simplified indices to brown hyaena data in Botswana yielded much higher CVs than previously published. It highlighted the confusion and need for clear descriptions of data analysis and the need for calibrations of the mean distance between track incidences against track density to derive density estimates with confidence intervals. This is addressed in Chapter 3 with species specific calibrations of the mean distance between track incidences and track densities that are required to derive density estimates with confidence intervals. Chapter 3 provides details on the analysis of track survey data with templates to assist with standardized and repeatable procedures. The impact of deviating from the appropriate analysis is demonstrated. Due to the high CV of track data, one is unlikely to detect population trends (Gerrodette 1987). I demonstrate the use of Chi-Square tests with Bonferroni intervals (Byers *et al*. 1984) to test for significant increases or decreases in population estimates among surveys as an alternative to trend analysis.

The evaluation of population monitoring techniques is extended to call up surveys for lions in Chapter 4, specifically looking at response calibration, call duration, effective response distance and power to detect population trends. Call up surveys were repeated over several years in an area with a known lion population. This provided the opportunity to test the calibrations from different studies, and compare with my calibrations.

In Chapter 7 I calibrated the pulp-tooth ratio to estimate the age of trophy-hunted lions in Botswana and assess compliance with the minimum age threshold set for lion trophy hunting in Botswana. This is an important potential tool to manage sustainable trophy hunting of lions.

Conservation has costs and benefits on local, national regional and international level. Photographic tourism has been the only generator of income from conservation areas in Botswana since the hunting ban in 2013. Northern Botswana is a key area for the conservation of large carnivores, but the perception is that parts of this area are not suitable for photographic tourism, and may not contribute benefits on local and national levels. Chapter 6 addresses the primary question of what areas in northern Botswana are suitable for HPLV tourism. I developed two techniques to assess the potential for wildlife based tourism objectively, using standard aerial survey data and line transect data. Assessing tourism potential objectively provides a tool to managers and policy makers to guide conservation planning. This evaluation is very relevant in light of the recent decision to open hunting again in Botswana. Similar to Chapter 5, I did not explicitly express the two hypotheses tested in Chapter 6:

H_0: Wildlife abundance and diversity did not differ between areas subjectively selected or avoided for photographic tourism.

H_1: Wildlife abundance and diversity were different between areas subjectively selected or avoided for photographic tourism.

and

H_0: Tourist can expect to see similar diversity and quantity of wildlife in areas with high and low tourism value.

H_1: Tourists can expect to see a higher diversity and quantity of wildlife in areas with high tourism value than areas with low tourism value.

The following chapters have been published:

Chapter 2:

Christiaan W. Winterbach, Sam M. Ferreira, Paul J. Funston, Michael J. Somers. (2016) Simplified large African carnivore density estimators from track indices. *PeerJ* 4:e2662. 10.7717/peerj.2662

Chapter 5:

Christiaan W. Winterbach, Glyn Maude, Gosiame Neo-Mahupeleng, Rebecca Klein, Lorraine Boast, Lindsey N. Rich, Michael J. Somers. (2017) Conservation implications of brown hyaena (*Parahyaena brunnea*) population densities and distribution across landscapes in Botswana. *Koedoe* 59(2), a1441. 10.4102/koedoe.v59i2.1441

Chapter 6:

Christiaan W. Winterbach, Caroline Whitesell, Michael J. Somers**.** (2015) Wildlife abundance and diversity as indicators of tourism potential in Northern Botswana. *PLoS ONE* 10(8): e0135595. 10.1371/journal.pone.0135595

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CHAPTER 2. SIMPLIFIED LARGE AFRICAN CARNIVORE DENSITY ESTIMATORS FROM TRACK INDICES

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2.1 Abstract

Background. The range, population size and trend of large carnivores are important parameters to assess their status globally and to plan conservation strategies. One can use linear models to assess population size and trends of large carnivores from track-based surveys on suitable substrates. The conventional approach of a linear model with intercept may not intercept at zero, but may fit the data better than linear model through the origin. We assess whether a linear regression through the origin is more appropriate than a linear regression with intercept to model large African carnivore densities and track indices.

Methods. We did simple linear regression with intercept analysis and simple linear regression through the origin and used the confidence interval for ß in the linear model $y = \alpha x + ß$, Standard Error of Estimate, Mean Squares Residual and Akaike Information Criteria to evaluate the models.

Results. The Lion on Clay and Low Density on Sand models with intercept were not significant ($P > 0.05$). The other four models with intercept and the six models through the origin were all significant ($P < 0.05$). The models using linear regression with intercept all included zero in the confidence interval for ß and the null hypothesis that ß = 0 could not be rejected. All models showed that the linear model through the origin provided a better fit than the linear model with intercept, as indicated by the Standard Error of Estimate and Mean Square Residuals. Akaike Information Criteria showed that linear models through the origin were better and that none of the linear models with intercept had substantial support. **Discussion.** Our results showed that linear regression through the origin is justified over the more typical linear regression with intercept for all models we tested. A general model can be used to estimate large carnivore densities from track densities across species and study areas. The formula *observed track density = 3.26 x carnivore density* can be used to estimate densities of large African carnivores using track counts on sandy substrates in areas where carnivore densities are 0.27 carnivores/100 km^2 or higher. To improve the current models, we need

independent data to validate the models and data to test for non-linear relationship between track indices and true density at low densities.

2.2 Introduction

Africa has seven large carnivores: lion *Panthera leo* (Linnaeus, 1758), leopard *Panthera pardus* (Linnaeus, 1758), spotted hyaena *Crocuta crocuta* (Erxleben, 1777), brown hyaena *Parahyaena brunnea* (Thunberg, 1820), striped hyaena *Hyaena hyaena* (Linnaeus, 1758), cheetah *Acinonyx jubatus* (Schreber, 1775) and wild dog *Lycaon pictus* (Temminck, 1820). The range, population size and trend of these large carnivores are important parameters to assess their status globally (Bauer *et al*. 2015; Wiesel 2015). These parameters are used to plan conservation strategies at different scales ranging from the entire distribution range of hyaena (Mills & Hofer 1998), cheetah and wild dog (Durant 2007), to regional conservation plans for lion (IUCN/SSC 2006) and national plans for cheetah and wild dog (Lindsey & Davies-Mostert 2009).

Methods used to estimate densities of African large carnivores include intensive studies (Smuts 1982; Maude 2010), call in surveys (Cozzi *et al*. 2013; Mills *et al*. 2001; Ogutu & Dublin 1998), camera trap surveys (Balme *et al*. 2009; Kent & Hill 2013), track counts (Funston *et al*. 2010; Keeping & Pelletier 2014; Stander 1998) and measuring track dimensions to identify individuals (Gusset & Burgener 2005). This paper focuses on the use of track indices to estimate large carnivore densities in Africa.

A previous study (Stander 1998) demonstrated a significant linear correlation between true density and track density for leopard, lion and wild dog; which can be used to estimate carnivore densities (animals / 100 km ^{2}) from track densities (tracks / 100 km). The leopard model is based on a bootstrap analysis to simulate different leopard densities using known individuals from a known population at one study area (Stander 1998). The lion and wild dog model used densities from two sites (four data points). The slope of the regression for leopard was different to that of lion and wild dog (Stander 1998), showing potential differences in the track density- true density relation for species and sites.

Funston *et al*. (2001) provided a calibration for lion in the Kgalagadi Transfrontier Park (southern Botswana and South Africa) and showed the potential to estimate large carnivore densities from track counts, using one general model. Houser *et al*. (2009) did track density-true density calibration for cheetah in southern Botswana, but unfortunately their estimate of true density was flawed and thus their calibration is invalid. Funston *et al*. (2010) did the first analysis for multiple species and localities and provided models to assess population size and trends of large carnivores from track-based surveys on sandy and clay soils. This calibration included data for five of the seven large carnivores in Africa and spanned 18 different study sites from seven study areas in Namibia, Botswana, South Africa, Zimbabwe, Kenia and Tanzania (Funston *et al*. 2010). Some recent studies used these models to assess large carnivore densities in parts of Botswana (Bauer *et al*. 2014; Boast & Houser 2012; Ferreira *et al*. 2013; Kent & Hill 2013). Refer to Funston *et al*. (2010) for the protocols to conduct track count surveys.

The formula to estimate large carnivore densities using the general model on sandy soils is $y = 3.15\,x + 0.4$, where y is track density (tracks / 100 km) and x is carnivore density (animals / 100 km ^{2}) (Funston *et al*. 2010). This formula would yield negative density estimates below track densities of 0.4 tracks / 100 km, for example carnivore density would be estimated as -0.06 animals per 100 km ^{2} from a track density of 0.2 tracks per 100 km. Boast & Houser (2012) resolved this problem for leopard by using the formula from Stander (1998) to estimate leopard densities at low track densities. Williams (2011) and Williams *et al*. (2016) opted to use the lion and wild dog model from Stander (1998) to estimate carnivore densities, although this model is based on only four data points.

Whereas Stander (1998) used linear models through the origin, Funston *et al*. (2010) followed the more conventional approach of a linear model with intercept (Eisenhauer 2003; Quinn & Keough 2002; Sokal & Rohlf 1995). Although biology may dictate that there should be no tracks if no carnivores are present (i.e. we expect $Y = 0$ when $X = 0$), the regression may not intercept at zero. Imperfect detection of tracks (Mackenzie 2006) at low densities may result in such a biological anomaly, or the relationship may not be linear with values approaching zero (Quinn & Keough 2002). In such a case, Quinn & Keough (2002) recommended using a model with intercept that fits the data better, even if it does not intercept at zero. Although there are circumstances where regression through the origin is appropriate (Quinn & Keough 2002; Sokal & Rohlf 1995), Eisenhauer (2003) described the use of regression through the origin as “a subject of pedagogical neglect, controversy and confusion”.

We used the guidelines from Sokal & Rohlf (1995), Quinn & Keough (2002) and Eisenhauer (2003) to assess whether a linear model fitted through the origin is more appropriate for the dataset from Funston *et al*. (2010) than the linear model with intercept that they used. We demonstrate the impact of using different models to estimate population size at various track densities.

2.3 Methods

We repeated the simple linear regression with intercept analysis done by Funston *et al*. (2010) and extended it to include simple linear regression through the origin. The data used by Funston *et al*. (2010) is summarized in Table 2-1. For their analysis of carnivores on sandy soils Funston *et al*. (2010) used the lion data (record 1 to 10 in Table 2-1) (Model 1) and then Model 2 “a combined model for all carnivore species on sandy soils” (record 1 to 16). Although they called it “all carnivore species” they excluded the data points they had for brown hyaena (Table 2-2). Also included in Table 2-2 are the data from Stander (1998) for leopard density, and track density for the site he labeled “Experimental”. Funston *et al*. (2010) included this leopard density without the track density in their table of mean densities for the respective large carnivores. We did an additional model for all the carnivores on sandy soils that included the data from Table 2-1 (record 1 to 16) and Table 2-2 (record 25 to 27). For clarity we will refer to this as Model 3 General Carnivores on Sand. Models 4 and 5 are Lion on Clay and lion plus Cheetah on Clay. Model 6 Low Density on Sand is a subset of carnivore densities below 1 animal / 100 km² for sandy substrates.

Forcing the model through the origin is rarely appropriate (Quinn & Keough 2002 page 110), therefore we used the criteria in Table 2-3 to assess if linear regression through origin is justified over linear regression with intercept (Eisenhauer 2003; Quinn & Keough 2002; Sokal & Rohlf 1995). There is some justification to fit a linear model through the origin if $Y = 0$ when $X = 0$, and the null hypothesis that $ß = 0$ is not rejected (Quinn & Keough 2002 page 99). This warrants further investigation using Standard Error of Estimate and Mean Square Residual (Table 2-3). We also calculated corrected Akaike Information Criteria (AIC_c) to assist model selection between intercept and through the origin models: $AIC_c = n \ln(SS\ residual/n) + 2K + (2K(2K+1))/(n-K-1)$, where n is sample size and K is the total parameters in the model including intercept and σ^2 (Burnham & Anderson 2004). A non-linear relationship with values approaching zero is possible (Quinn & Keough 2002). We added a logarithmic curve fitted as part of Model 6 to test for a non-linear relationship at low densities.

Table 2-1 Mean density and tracks density of respective carnivores used in linear regression models by Funston *et al* (2010).

Record number	Substrate	Location	Species	Density individuals / 100 km²	Density tracks /100 km
1	Sandy	Dune-north	Lion	0.67	1.6
2	Sandy	Dune-south	Lion	0.95	2.9
3	Sandy	Sesatswe	Lion	1.35	5.5
4	Sandy	Mabuasehube	Lion	1.68	4.5
5	Sandy	Mosimane	Lion	2.2	7.2
6	Sandy	Main camp	Lion	2.73	9.5
7	Sandy	Venetia	Lion	3.3	9.7
8	Sandy	El Karama	Lion	5.8	18.2
9	Sandy	Mugie	Lion	6	17.8
10	Sandy	Mpala	Lion	6.15	22.5
11	Sandy	Dune-north	Cheetah	0.54	1.7
12	Sandy	Dune-south	Cheetah	0.54	4.9
13	Sandy	Dune-north	Leopard	0.27	0.8
14	Sandy	Dune-south	Leopard	0.27	0.4
15	Sandy	Dune-north	Spotted Hyaena	0.9	4.7
16	Sandy	Dune-south	Spotted Hyaena	0.9	3.4
17	Clay	Short-grass dry	Lion	7	1.5
18	Clay	Short-grass wet	Lion	20	10.5
19	Clay	Long-grass wet	Lion	21.08	8
20	Clay	Long-grass dry	Lion	24.28	16.5
21	Clay	Short-grass dry	Cheetah	2.26	1.0
22	Clay	Long-grass wet	Cheetah	2.29	0.9
23	Clay	Short-grass wet	Cheetah	6.78	9.0
24	Clay	Long-grass dry	Cheetah	9.16	1.6

Table 2-2 Additional mean density and tracks density of brown hyaena from Funston *et al*. (2010) and leopard from Stander (1998).

Record number	Substrate	Location	Species	Density individuals / 100 km²	Density tracks /100 km
25	Sandy	Dune-south	Brown Hyaena	1.6	5.2
26	Sandy	Dune-north	Brown Hyaena	1.6	6.4
27	Sandy	Experimental	Leopard	1.45	2.62

Table 2-3 Criteria to assess the use of linear regression through origin over linear regression with intercept.

CRITERIA	SOURCE
$Y = 0$ when $X = 0$	(Eisenhauer 2003; Quinn & Keough 2002 page 99)
Null hypothesis that ß = 0 is not rejected based on P value for ß; Confidence interval for ß in the linear model $y = \alpha x + ß$ includes zero	(Eisenhauer 2003; Quinn & Keough 2002 page 99)/ (Sokal & Rohlf 1995 page 474)
Mean Squares Residual is smaller for regression through the origin than regression with intercept, indicating a better fit.	(Quinn & Keough 2002 page 99)
Standard error is smaller for regression through the origin than regression with intercept, indicating a better fit.	(Eisenhauer 2003)

We compared population estimates derived from the leopard model (Stander 1998), Model 2 Carnivores on Sand (regression with intercept) (Funston *et al*. 2010) and Model 3 General Carnivores on Sand (regression through origin). We used track densities from recently published studies to estimate carnivore density before calculating population estimates for a hypothetical study area of 10000 km². The difference between population estimates using Model 3 General Carnivores on Sand model (regression through origin) and Model 2 Carnivores on Sand (regression with intercept) were calculated as a percentage of population estimate from Model3 General Carnivores on Sand model (regression through origin).

2.4 Results

Regressions through the origin were significant ($P < 0.05$) for the six models tested (Table 2-4). Regression analyses with intercept were statistically significant at $P < 0.05$ except for Model 4 Lion on Clay with intercept and Model 6 Low Density on Sand with intercept (Table 2-4) that were not significant. Model 6 Low Density on Sand logarithmic was not significant ($t_i = 3.86 + (2.32 \times \ln(x_i))$, $F_{1,6} = 5.587$, $P = 0.056$, $R^2 = 0.482$). Eight data points, two each for lion, leopard, spotted hyaena and cheetah were used in Model 6 Low Density on Sand.

We used the criteria from Table 2-3 to assess and select between the models with linear regression with intercept and regression through the origin. Zero tracks are expected when zero carnivores are present, complying with the condition that $Y = 0$ when $X = 0$. The six models using linear regression with intercept all included zero in the confidence interval for ß (Table 2-5). The P values for ß were not significant ($P > 0.05$), thus the null hypothesis that ß = 0 could not be rejected for all linear models with intercept (Table 2-5). The Mean Square Residual and Standard Error of Estimate (Table 2-6) for the linear model through the origin were smaller than the comparative linear model with intercept, indicating that the linear model through the origin provided a better fit than the linear model with intercept for all the models.

The value of K was three to calculate AIC_c for models with intercept (one parameter plus intercept plus σ^2) and two for models through the origin (one parameter plus σ^2). Sample size for Model 4 Lion on Clay with intercept resulted in K =0 and AIC_c could not be calculated. AIC_c values are used to compare models based on the same data set (Burnham & Anderson 2004), for example different versions of Model 1, but cannot be used to compare among models 1 to 6. Models through the origin had the lowest AIC_c values and the Δ_i were between 2.28 and 5.60 (Table 2-6) for the models with intercept, indicating that none of the intercept models have substantial support ($\Delta_i > 2$) (Burnham & Anderson 2004).

We tested Model 3 General Carnivore on Sand through origin with data from a study area of 629 km ^{2}. At the time of the track survey there were 18 lions present mat a density of 2.86 lions / 100 km^2. We surveyed 294 km and recorded 13 incidences of lions consisting of 22 individuals. Track frequency was 10 ± 1.5 km / lion (CV = 14.8 %). Track density was 7.2 (95% CI 3.3 -1.1.2), resulting in a density estimate of 2.2 (1.0 -3.4) lions / 100 km^2. This was 78% of the true density and the 95% CI included the true density.

Table 2-7 provides a comparison of population estimates derived from three models. The leopard density estimates from Model 3 General Carnivore on Sand model is 58% of estimates calculated with the leopard model from Stander (1998). The difference in population estimates between Model 3 General Carnivore on Sand through origin and Model 2 Carnivore on Sand with intercept (Funston *et al*. 2010) is the largest at low densities (Table 2-7). At the lower limit for density extrapolation (0.88 tracks / 100 km) the difference is 43.5 % and converged to less than 10% difference at track density of 3.1 tracks / 100 km. Using Model 6 Low Density on Sand through origin provided more conservative estimates that are 80% of estimates using Model 3 General Carnivore on Sand through origin.

A total of 2273 km need to be surveyed at a track density of 0.88 tracks / 100 km to obtain the recommended minimum number of track incidences of 19 (Funston *et al*. 2010). The required survey distance will increase to 5000 km at a track density of 0.4 tracks / 100 km (Table 2-7).

Table 2-4 Summary of linear regression with intercept and through the origin for carnivore density (predictor) and track density (dependent) on sandy and clay soils.

Model	Description	Linear Regression	F value	Signifi-cance	R Square[a]	R Square[b]
Model 1	Lion sandy soil with intercept	$t_i = 3.3x_i - 0.31$	$F_{1,8} = 244.914$	$P<0.001$	0.972	
	Lion sandy soil through origin	$t_i = 3.23x_i$	$F_{1,9} = 819.856$	$P<0.001$		0.990
Model 2	Carnivores sandy soil with intercept	$t_i = 3.16x_i + 0.42$	$F_{1,14} = 333.281$	$P<0.001$	0.962	
	Carnivores sandy soil through origin	$t_i = 3.26x_i$	$F_{1,15} = 732.137$	$P<0.001$		0.981
Model 3	General Carnivores on sand with intercept	$t_i = 3.18x_i + 0.31$	$F_{1,18} = 356.600$	$P<0.001$	0.954	
	General Carnivores on sand through origin	$t_i = 3.26x_i$	$F_{1,19} = 850.826$	$P<0.001$		0.979
Model 4	Lion on clay with intercept	$t_i = 0.75x_i - 4.34$	$F_{1,3} = 9.998$	$P>0.05$	0.833	
	Lion on clay through origin	$t_i = 0.53x_i$	$F_{1,4} = 44.847$	$P<0.01$		0.937
Model 5	Lion and Cheetah on clay with intercept	$t_i = 0.55x_i - 0.28$	$F_{1,7} = 14.695$	$P<0.01$	0.710	
	Lion and Cheetah on clay through origin	$t_i = 0.54x_i$	$F_{1,8} = 47.940$	$P<0.001$		0.873
Model 6	Low density on sandy soil with intercept	$t_i = 4.10x_i - 0.03$	$F_{1,6} = 4.615$	$P>0.05$	0.435	
	Low density on sandy soil through origin	$t_i = 4.06\ x_i$	$F_{1,7} = 37.116$	$P<0.001$		0.841

R Square[a] measures the proportion of variation in the data described by the linear regression with intercept. R Square[b] measures the proportion of the variability in the dependent variable about the origin explained by regression through the origin. This cannot be compared to R Square[a].

Table 2-5 Coefficients for linear regressions with intercept and linear regression through origin using density (predictor) and tracks (dependent). Standard error for coefficient, coefficient of variance, t value and level of significance are shown for each model coefficient.

Model	Description	Coefficient	Value	SE of Coeffi-cient	CV (%)	t	Significance level	Lower Bound	Upper Bound
Model 1	Lion sandy soil with intercept	Constant (b)	-0.31	0.8	258.1	-0.385	P=0.711	-2.196	1.58
	Lion sandy soil with intercept	Rate of change (a)	3.3	0.21	6.4	15.65	P<0.001***	2.8	3.796
	Lion sandy soil through origin	Rate of change (a)	3.23	0.11	3.4	28.633	P<0.001***	2.971	3.491
Model 2	Carnivores sandy soil with intercept	Constant (b)	0.42	0.51	121.4	0.813	P=0.431	-0.69	1.523
	Carnivores sandy soil with intercept	Rate of change (a)	3.16	0.17	5.4	18.256	P<0.001***	2.785	3.532
	Carnivores sandy soil through origin	Rate of change (a)	3.26	0.12	3.7	27.058	P<0.001***	3	3.516
Model 3	General Carnivores on sand with intercept	Constant (b)	0.31	0.47	151.6	0.656	P=0.521	-0.68	1.29
	General Carnivores on sand with intercept	Rate of change (a)	3.18	0.17	5.3	18.884	P<0.001***	2.83	3.54
	General Carnivores on sand through origin	Rate of change (a)	3.26	0.11	3.4	29.169	P<0.001***	3.03	3.5
Model 4	Lion on clay with intercept	Constant (b)	-4.34	4.53	104.4	-0.958	P=0.439	-23.85	15.16
	Lion on clay with intercept	Rate of change (a)	0.75	0.24	32.0	3.162	P=0.087	-0.27	1.76
	Lion on clay through origin	Rate of change (a)	0.53	0.08	15.1	6.697	P<0.01**	0.28	0.79
Model 5	Lion and Cheetah on clay with intercept	Constant (b)	-0.28	2.05	732.1	-0.137	P=0.896	-5.299	4.737
	Lion and Cheetah on clay with intercept	Rate of change (a)	0.55	0.14	25.5	3.833	P<0.01**	0.2	0.904
	Lion and Cheetah on clay through origin	Rate of change (a)	0.54	0.08	14.8	6.924	P<0.001***	0.353	0.719
Model 6	Low density on sandy soil with intercept	Constant (b)	-0.03	1.30	4333.3	-0.025	P = 0.981	-3.208	3.143
	Low density on sandy soil with intercept	Rate of change (a)	4.10	1.91	46.6	2.148	P =0.075	-.570	8.769
	Low density on sandy soil through origin	Rate of change (a)	4.06	0.69	17.0	6.092	P<0.001***	2.481	5.629

Table 2-6 Evaluation of linear regression models for carnivore density (predictor) and track density (dependent) on sandy and clay soils. Smaller values of Standard Error of Estimate, Mean Square Residual and small sample corrected Akaike Information Criteria (AIC_c) indicate better fit of model.

Model	Description	Linear Regression	Standard Error of Estimate	Mean Square Residual	AIC_c	Δ_i Values	ω_i
Model 1	Lion sandy soil through origin	$t_i = 3.23x_i$	1.283	1.645	8.46	0.00	0.88
Model 1	Lion sandy soil with intercept	$t_i = 3.3x_i -0.31$	1.357	1.841	12.54	4.08	0.12
Model 2	Carnivores sandy soil through origin	$t_i = 3.26x_i$	1.381	1.907	13.12	0.00	0.76
Model 2	Carnivores sandy soil with intercept	$t_i = 3.16x_i +0.42$	1.398	1.955	15.40	2.28	0.24
Model 3	General Carnivores on sand through origin	$t_i = 3.26x_i$	1.352	1.828	15.18	0.00	0.77
Model 3	General Carnivores on sand with intercept	$t_i = 3.18x_i +0.31$	1.374	1.888	17.56	2.38	0.23
Model 4	Lion on clay through origin	$t_i = 0.53x_i$	3.063	9.379	23.80	0.00	-
Model 4	Lion on clay with intercept	$t_i = 0.75x_i -4.34$	3.105	9.642	-	-	-
Model 5	Lion and Cheetah on clay through origin	$t_i = 0.54x_i$	3.118	9.722	23.53	0.00	0.94
Model 5	Lion and Cheetah on clay with intercept	$t_i = 0.55x_i -0.28$	3.363	11.307	29.10	5.58	0.06
Model 6	Low density on sandy soil through origin	$t_i = 4.06\ x_i$	1.28	1.639	9.29	0.00	0.94
Model 6	Low density on sandy soil with intercept	$t_i = 4.10x_i -0.03$	1.383	1.912	14.89	5.60	0.06

Table 2-7 Comparison of carnivore population estimates for a reference area of 10000 km^2, using different models to estimate density (animals / 100 km^2) from track densities (tracks / 100 km). The survey distances to obtain the recommended minimum of 19 track incidences at different track densities are shown.

SOURCE	Species	Track Density (tracks/ 100 km)	Survey Distance for 19 track incidences	Leopard Model [a]		Carnivore on sand Intercept Model [b]		General Carnivore on Sand Origin Model [c]		Difference (c-b)*100/c
			(km)	Density (animals/ 100 km^2)	Population Estimate	Density (animals/ 100 km^2)	Population Estimate	Density (animals/ 100 km^2)	Population Estimate	%
Boast & Houser (2012)	Leopard	0.00		0.00	0	-0.13	-13	0.00	0	
Boast & Houser (2012)	Leopard	0.10	19000	0.05	5	-0.10	-10	0.03	3	410.5
Boast & Houser (2012)	Leopard	0.20	9500	0.11	11	-0.06	-6	0.06	6	203.5
		0.40	4750	0.21	21	0.00	0	0.12	12	100.0
Boast & Houser (2012)	Leopard	0.47	4043	0.25	25	0.02	2	0.14	14	84.6
	Lower Extrapolation Limit	0.88	2159	0.46	46	0.15	15	0.27	27	43.5
Boast & Houser (2012)	Cheetah	1.02	1863			0.20	20	0.31	31	37.1
Boast & Houser (2012)	Cheetah	2.24	848			0.58	58	0.69	69	15.0
Stander (1998)	Leopard	2.62	725	1.38	138	0.70	70	0.80	80	12.3
Bauer *et al* (2014)	Lion	3.05	623			0.84	84	0.94	94	10.1
Bauer *et al* (2014)	Lion	5.36	354			1.57	157	1.64	164	4.2
Boast & Houser (2012)	Brown hyaena	6.15	309			1.83	183	1.89	189	3.2
Boast & Houser (2012)	Brown hyaena	7.90	241			2.38	238	2.42	242	1.7

2.5 Discussion

Our results show that linear regression through the origin is justified over the more typical linear regression with intercept for the six models we tested. Adding the brown hyaena and leopard data (Table 2-2) did not alter the slope (a = 3.26). The slope of Model 3 General Carnivore on Sand model through origin (a = 3.26 ± 0.24; $r^2 = 0.98$; t = 29.169; $P<0.001$) was very similar to the slope for a small sample of lion and wild dog densities (a = 3.28 ± 0.24; $r^2 = 0.98$; t = 13.55; $P<0.01$) presented by Stander (1998). This further supports the Funston *et al*. (2010) conclusion that a general model can be used to estimate large carnivore densities from track densities across species and study areas. It is important to use the appropriate model to account for the substrate of the study area, since linear models to estimate large carnivore densities differed significantly on sandy and clay soils (Funston *et al*. 2010).

Model 3 General Carnivore on Sand through origin provided more conservative leopard population estimates than the Stander (1998) leopard model. The Stander (1998) leopard model is from a single study site and based on the assumption that the linear relationship between leopard density and track density holds below the density of 1.45 leopard / 100 km² (2.62 tracks / 100 km) in his study area. Stander (1998) simulated lower densities by randomly including different numbers of individual leopard in the analysis, but the assumption of a linear relationship between carnivore density and track density may not hold below the sample range.

The valid extrapolation range for Model 3 General Carnivores on Sand through origin exceeds that of Stander (1998), thus allowing carnivore density estimates as low as 0.27 carnivores / 100 km² (track density 0.88 tracks / 100 km). Density estimates below this should be considered with caution. Also, this model would not yield negative carnivore density estimates. We therefore conclude that the formula: *observed track density = 3.26 x carnivore density* can be used to estimate densities of large African carnivores using track counts on sandy substrates in areas where carnivore densities are 0.27 carnivores / 100 km^2 or higher. The validity of density estimates below 0.27 carnivores / 100 km^2 (< 0.88 tracks / 100 km) (Table 2-7) is questionable, but it may be the best available data to guide conservation. Estimates and trends obtained from track surveys in low density populations should be interpreted with caution.

The potential non-linear relationship with X approaching zero (Quinn & Keough 2002) adds uncertainty to estimates obtained track densities below 0.88 tracks / 100 km. The current models are based on a data set consisting predominantly of lion data points and limited or no data for other large carnivore species. We found a significant linear relationship at low carnivore densities with densities ranging between 0.27 and 0.95 carnivores / 100 km². With equal numbers of lion, spotted hyaena, leopard and cheetah, Model 6 Low Density on Sand through origin was not dominated by lion, but all data points were from the Kgalagadi Transfrontier Park. We need data, especially at lower densities, for a variety of large carnivores to improve the current models. The one independent data point for lion density we had, provided a good density estimate. More independent data for different species are required to validate the models.

We recommend that studies using track surveys to estimate carnivore densities provide a data summary with all the relevant data to facilitate recalculation of density estimates. This will ensure that results can be compared among studies that used different models and that density estimates can be recalculated in future if new calibrations become available.

Track surveys are cost effective and can cover large areas. At low carnivore densities there is a trade-off between data quality and survey effort required from track surveys. Selecting an appropriate carnivore survey technique depends on the survey objectives, resources and expertise available, the size of the survey area and expected range of carnivore densities.

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CHAPTER 3. ESTIMATING CARNIVORE DENSITY FROM TRACK SURVEYS: CALIBRATION AND REVISED ANALYSES FOR IMPROVED CONFIDENCE

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3.1 Abstract

The reliable monitoring of wildlife populations is paramount for conservation management. This presents challenges, particularly in terms of data collection and analysis. Carnivores, for example, are inherently difficult to detect as they occur at naturally low densities, cover large areas, and behave cryptically. Researchers, therefore, often rely on track surveys conducted along line transects to determine local carnivore densities and infer population trends. Different analysis paradigms have been developed to estimate densities from track indices. Yet, methods are often inconsistent, confounding the comparability of results derived from multiple sites, surveys, and species to assess carnivore populations at national or regional scales. Researchers also use different ways to calculate confidence intervals for their estimates, if at all. Here, we provide an in-depth re-analysis of track survey data gathered for the entire southern African large carnivore guild. We analyse data from 45 surveys conducted between 1998 and 2016. These surveys spanned across seven eco-regions in four countries, with a total sampling effort of 26,229 kilometres. We use these data to develop a rigorous, replicable analysis that increases our confidence in density estimates. We provide a step-wise explanation of our analysis, alongside relevant survey and calculation templates. These serve as an updated guideline to derive carnivore density estimates, including species-specific calibrations for the relationship between the mean distance between track encounters and track densities that allow us to estimate confidence intervals of track densities. The high coefficients of variation of track survey data result in low power to detect population trends reliably. However, we demonstrate that a χ^2-test with Bonferroni intervals provides a robust, yet change-sensitive, alternative to test for differences among track surveys. Despite some limitations, track surveys are a practical tool to assess large carnivore populations at different scales.

3.2 Introduction

There is a need for monitoring programs due to the increasing land-use pressures on African conservation areas and the declining wildlife populations (Balmford *et al*. 2003; Caro 2016; Durant *et al*. 2007; Keeping *et al*. 2018; Lindsey *et al*. 2014; Scholte 2011). Assessing the status of carnivore populations is hampered by a general lack of data to evaluate population size and trends based on repeatable survey techniques (Riggio *et al*. 2013). Protected areas in Africa are chronically underfunded; therefore, the cost and efficiency of monitoring programs as well as logistical constraints are of concern (Bart *et al*. 2004; Caro 2016; Durant *et al*. 2007; Harris *et al*. 2013; Hines *et al*. 2010). Intensive survey methods are rarely used for landscape-scale surveys because of logistical constraints (Karanth *et al*. 2011).

Advanced modelling techniques require complex statistical analysis and programming skills (Dénes *et al*. 2015; Rich *et al*. 2019). These skills are lacking in doctoral programs (Touchon & McCoy 2016) and are making these advanced techniques inaccessible to the broader audience (Dénes *et al*. 2015; Rich *et al*. 2019). This problem is more acute in Africa, where conservation is hampered by a lack of capacity among researchers, managers and resources (Caro 2016; Durant *et al*. 2007; Keeping *et al*. 2018; Lindsey *et al*. 2014; Scholte 2011). Therefore, it is unlikely that routine monitoring programs that are resource intensive and require advanced modelling techniques can be implemented successfully.

Large carnivores are difficult to study because they occur at low densities, range over large areas and are elusive (Alibhai *et al*. 2017; Balme *et al*. 2009; Belbachir *et al*. 2015; Henschel *et al*. 2016). However, as carnivores move through the environment, they typically leave tracks and other signs that may be the only information readily available to researchers (Bothma & Le Riche 1984; Eloff 1973).

Track-based surveys are rapid and cost-effective (Gusset & Burgener 2005), and can be used at large spatial scales (Funston *et al*. 2001, 2010; Winterbach *et al*. 2017). Track surveys are non-invasive, minimizing disturbance of focal species and can cover multiple species simultaneously (Funston *et al*. 2010; Keeping 2014), including herbivores (Ahlswede *et al*. 2019; Keeping *et al*. 2018). Consequently, carnivore track surveys, and their many possible variants, are used around the globe (Alibhai *et al*. 2017; Allen *et al*. 1996; Balme *et al*. 2009; Beier & Cunningham 1996; Edwards *et al*. 2000; Evangelista *et al*. 2009; Hayward *et al*. 2002; Isasi-Catalá & Barreto 2008; Karanth *et al*. 2011; Linnell *et al*. 2007; Miller & Jug 2001; Sanei *et al*. 2011; Silveira *et al*. 2003; Smallwood & Fitzhugh 1995; Wong *et al*. 2010).

Multiple analysis paradigms have been developed to derive carnivore density estimates from track counts at various spatial scales (Funston *et al*. 2010; Keeping 2014; Stander 1998), including refinements of analyses (Keeping & Pelletier 2014; Winterbach *et al*. 2016). Particularly in southern Africa, where limited resources and difficult terrain and habitat usually impose serious limitations on carnivore survey designs, track-based population density indices have become a popular tool for estimating large carnivore numbers, and sometimes those of medium-sized carnivores (Boast & Houser 2012; Gusset & Burgener 2005; Keeping *et al*. 2018). Funston *et al*. (2010) highlighted the need for suitable substrate and sufficiently skilled trackers.

The expertise of Kalahari trackers on sandy soils was quantified, demonstrating a near-perfect record of identifying individual large carnivores from their tracks, and the ability to reconstruct complex behaviours from the signs (Stander *et al*. 1997). Based on this ability, Funston *et al*. (2010) consider that highly skilled trackers introduce limited observer bias in the data.

The strong relationships between spoor density and true carnivore densities for different large African carnivores allow extrapolation of population estimates from track indices (Funston *et al*. 2010; Funston *et al*. 2001; Stander 1998). The substrate, being sand or clay, influenced the relationships between track density and true carnivore density. All six large carnivore species could be combined in a single model on sandy substrates for general application to estimate carnivore density (Funston *et al*. 2010).

More recently, researchers incorporated the daily movement ranges of animals into calibrations to estimate animal density, using the Formozov-Malyshev-Pereleshin (FMP) formula (Ahlswede *et al*. 2019; Keeping 2014; Keeping *et al*. 2018; Keeping & Pelletier 2014). Data sets collected following the method of Stander (1998) and Funston *et al*. (2010) by recording tracks of an individual only once in 24 hours, are incompatible with the FMP method where all track incidences of an individual crossing the line transect are recorded (Keeping & Pelletier 2014).

When designing a monitoring program, the number of samples required to reach a minimum level of the precision, and the probability of detecting a trend are key questions. Planning accordingly helps to avoid wasted time and effort on a program that is unlikely to provide useful information (Buckland *et al*. 1993; Gerrodette 1987).

Following Greig-Smith (1983), Stander (1998) used the ratio of Standard Error of the Mean (SE) divided by the sample mean to assess the variation of the mean with increasing sample size to determine the minimum number of track incidences required. Stander referred to this ratio as the *Coefficient of Variation* (CV). However, the true CV is calculated as the ratio of Standard Deviation (SD) over the sample mean (Quinn & Keough 2002; Sokal & Rohlf 1995), and can be expressed as a percentage value by SD / mean * 100.

Midlane *et al.* (2015) were the first to point out the confusion surrounding the CV in the track survey literature. For clarity, we will refer to the CV as its statistical original meaning, i.e. SD / mean (Quinn & Keough 2002; Sokal & Rohlf 1995). We define the Relative Standard Error (RSE) as SE of mean/mean, to replace Stander's (1998) "CV" used in several subsequent analyses of track surveys e.g. Bauer *et al*. (2014), Boast & Houser (2012) and Funston *et al*. (2010).

SD provides a measure of sample variation (i.e. measurement dispersion), while the SE informs us about the variation of the sample mean (Quinn & Keough 2002; Sokal & Rohlf 1995). Since SE = SD/√n, the SE progressively declines in relation to the SD by a ratio of 1/√n with increasing sample size (Greig-Smith 1983, Midlane *et al*. 2015), resulting in decreasing values of RSE.

This erroneous terminology is inconsequential to determine the minimum sample size of track incidences required. Stander (1998) and Funston *et al*. (2010) plotted RSE versus sample size, and showed that 20 to 30 track incidences mostly result in a RSE < 20%, the target level of precision they set. The corresponding CV for a RSE = 20% with sample sizes of 16, 25 and 36 track incidences, will be 80%, 100% and 120% respectively. When you need to use CV in other calculations, but substitute it with RSE, the impact is massive.

The power to detect population trends, declines with an increase in CV (Gerrodette 1987). The power is conventionally set at 0.80 (Bart *et al*. 2004; Quinn & Keough 2002) and most wildlife surveys aim for a CV < 20%; above this threshold is not considered suitable for monitoring trends (Boulanger *et al*. 2002; Keeping *et al*. 2018).

When testing any null hypothesis, there is a risk of making Type I or Type II errors. A Type I error (α) occurs when the null hypothesis is rejected when there is no effect. A Type II error (β) wrongly accepts the null hypothesis when there is an effect. Related to Type II error is the statistical power (β-1) of a test, the confidence in detecting an effect if one existed (Quinn & Keough 2002). In terms of monitoring animal populations, the power is the probability to detect a population trend when it is present (Gerrodette 1987).

Gerodette's inequality is a method to assess the power to detect a population trend with linear and exponential regression, when a trend is present (Gerrodette 1987). This method has been applied to population estimates of elephants, marine mammals and track surveys of carnivores (Barnes 2002; Funston *et al*. 2010; Taylor *et al*. 2007). Interpreting results with power analysis shed light on how large a change could have been detected and the probability of detecting it.

Five parameters are interlinked in the power analysis of changes in abundance that take place with constant increments (linear models) or at a constant rates (exponential models) (Gerrodette 1987): power (β-1), sample size (*n*), probability of a Type I error (α), the rate of change (*r*) and the coefficient of variation of the estimate of abundance (CV). Using the smaller RSE metric instead of the larger CV in Gerodette's inequality (Gerrodette 1987), will portray a false ability to detect population trends from successive surveys.

There are inconsistencies in the calculation and reporting of confidence intervals for carnivore density estimates or a lack of reporting confidence intervals altogether. Spatial and temporal replication of transects compromises the ability to derive confidence intervals for track densities (Funston *et al*. 2010). They proposed to estimate confidence intervals for track density from the mean distance between track incidences, and as an example provided lion calibrations of the mean distance between track incidences versus track density. The lack of calibrations describing the relationship between track density and track frequency (mean distance between incidences) for other carnivores probably contributed to the fact that none of the recent studies (Bauer *et al.* 2014; Boast & Houser 2012; Houser, Somers & Boast 2009; Kent & Hill 2013; Midlane *et al.* 2015; Williams *et al.* 2016; Winterbach *et al.* 2017) followed this proposed method of calculating confidence intervals.

When calculating true density to establish the relationship between carnivore density and track density, the carnivore density calculation should incorporate the time that individuals spent outside the study area (Funston *et al*. 2010). Not allowing for time that animals spend outside the study area, will result in an over estimate of true density and an erroneous calibration. Houser *et al*. (2009) worked with a known population of cheetah moving in and out of their study area, but failed to incorporate this in their true density calculation. The resulting estimates of true densities were 10 to 20 fold higher than density estimates reported by other cheetah studies in Botswana.

Using two different techniques, Gusset & Burgener (2005) identified three and four individual leopards from tracks in their 15 km^2 study area. They noted that the carnivores in their study area probably move beyond the boundary of the area. They erroneously motivated to adjust the leopard density estimate from tracks, using the Stander (1998) leopard calibration, with a factor of 1.3. This was based on a previous study in the same general area, reporting a density 1.3 times higher than in the Stander study. This resulted in a density estimate of 3.2 leopards / 100 km^2 instead of 2.46 leopards / 100 km^2. This density estimate of 3.2 leopards / 100 km^2 was then compared to the 3 to 4 individuals identified in the study area, concluding that the three methods yielded very similar density estimates. The problem is that the 3 to 4 individuals partly using 15 km^2 are not density estimates and the density estimate of 3.2 leopards / 100 km^2 is invalid. Funston *et al*. (2010) provide appropriate procedures to estimate true density that allows for the time that study animals spend outside the study area.

Track frequency (mean distance between track incidences), as defined by Stander (1998), has been calculated as the distance between track incidences, correctly treating a group of animals as one incidence (e.g. Bauer *et al.* 2014; Kent & Hill 2013). Others treated animals in the group as separate incidences, with a zero distance between detections or did not describe the way that track frequency was calculated (see: Balme *et al*. (2009), Boast & Houser (2012), Houser *et al*. (2009), Williams *et al*. (2016). Treating individuals in the same group as separate incidences result in zero inflation of the data set and over dispersion (Harrison 2014).

The basic track count method described by Stander (1998) seems straightforward and simple, regardless of the calibration used to estimate carnivore density. However, as shown above, the literature on track surveys shows flawed logic, perplexing terminology and erroneous data analyses. These methodological issues raise concern over the general application of track surveys for population monitoring, and the ability to detect population trends. It also impedes the comparability of results derived from multiple sites, surveys, and species to assess populations at national or regional scales.

We used our own and published multi-species survey data from various countries and sites to address the issues identified:

1) Develop species-specific calibrations to estimate the CIs for carnivore density estimates from track surveys, based on the species-specific relationships of mean distance between track incidences and track density;
2) Demonstrate the impact of analysis variations and inconsistencies on survey results;
3) Demonstrate power analysis to detect trends from track survey data, using Gerrodette’s inequality and the alternative of using Bonferroni intervals with Chi square test;
4) Provide comprehensive set of survey and analysis templates and R script that allow others to replicate our analysis method (Appendix 1) by following the required steps for correct analysis of carnivore tracks to obtain density estimates;

We discuss the implications, strengths and shortcomings of track surveys to assist researchers and conservation managers in considering this method to monitor large carnivore populations. We hope that this article will provide a guideline for best practices and assist researchers in the correct analysis of their track survey data.

3.3 Methods

3.3.1 Study area

We analyse large carnivore track data from a diversity of habitats, land tenure systems, and land uses in Zimbabwe, Botswana, South Africa and Namibia, collected between 1998 and 2016 (Fig. 3-1; Table 3-1). We include our surveys and published data (Funston *et al*. 2001; Kent 2011; Williams *et al*. 2016) into our analyses. These 45 surveys encompassed a total sampling effort of 26,229.3 km (along 392 unique transects) across a survey area of 131,282 km^2 that spanned seven ecoregions in southern Africa (Dinerstein *et al*. 2017). We followed the stratification of Winterbach *et al*. (2014) for surveys that covered large areas in Botswana.

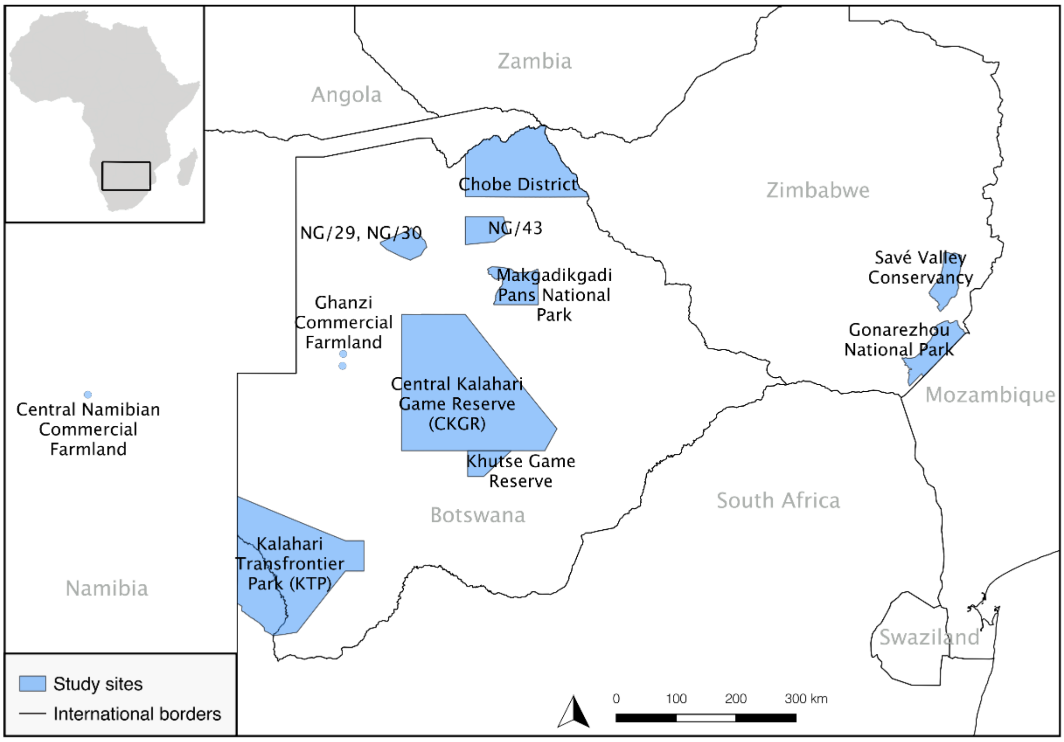

Figure 3-1 Distribution of main track survey sites used in analyses.

Table 3-1 Summary of study sites, ecoregion, habitat types, land use and tenure, substrates, focal species and sampling effort included in this study.

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Botswana	1	Chobe district	1.2.1; 1.2.3; 1.3.1; 1.3.5 & 1.4.3; 1.3.6; 1.4.1	2005	G Neo-Mapuleng	mixed none/partial/full	Mixed woodlands and shrublands	Zambezian mopane woodlands; Zambezian Baikiaea woodlands; Kalahari xeric savannah	mixed tribal and state with lease concessions	conservation + non consumptive wildlife tourism; livestock production; pastoral and residential areas; crop production	Kalahari sand	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	40	19.1 (2.5)	765.8	21551	28.1	65	1235.9	partially published
Botswana	1	Chobe district	1.2.3; 1.3.1; 1.3.2; 1.3.4; 1.3.5 & 1.4.3; 1.3.6; 1.4.1	2006	G Neo-Mapuleng	mixed none/partial/full	Mixed woodlands and shrublands	Zambezian mopane woodlands; Zambezian Baikiaea woodlands; Kalahari xeric savannah	mixed tribal and state with lease concessions	conservation + non consumptive wildlife tourism; livestock production; pastoral and residential areas; crop production	Kalahari sand	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	42	19.7 (3.3)	825.5	21551	26.1	91	1765.2	partially published
Botswana	2	Makgadikgadi Pans National Park	1.5.3	2012	G Maude & Winterbach *et al.* 2017b	full - national park	Seasonally flooded grasslands and savannahs	Makgadikgadi halophytics; Kalahari xeric savannah; Zambezian mopane woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	19	25.4 (5.0)	482.4	7549	15.6	19	482.4	partially published
Botswana	3	Central Kalahari Game Reserve North	2.1.1	2012	G Maude	full - state game reserve	Xeric shrublands; deserts	Kalahari xeric savannah	state	conservation + non consumptive wildlife tourism	sandy	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	51	22.2 (6.0)	1133.0	18850	16.6	51	1133.0	unpublished reports

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Botswana	4	Central Kalahari Game Reserve South	2.1.2; 2.1.3	2012	G Maude	full - state game reserve	Xeric shrublands; deserts	Kalahari xeric savannah	state	conservation + non consumptive wildlife tourism	sandy	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	46	22.2 (4.4)	1021.6	31088	30.4	46	1021.6	unpublished reports
Botswana	5	Khutse Game Reserve	2.1.4	2012	G Maude	full - state game reserve	Xeric shrublands; deserts	Kalahari xeric savannah	state	conservation + non consumptive wildlife tourism	sandy	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	17	23.3 (4.6)	396.9	4902	12.4	17	396.9	unpublished reports
Botswana	6	Ghanzi farmland - Hardveld	7.1.0	2008-2009	Kent 2011	none	Semi-arid bush savannah	Kalahari xeric savannah	private free-hold and lease-hold	mixed livestock and wildlife ranching	Kalahari sand interspersed with occasional outcrops of calcrete or rock	*Panthera pardus*; *Acinonyx jubatus*; *Parahyaena brunnea*	6	10.7 (4.6)	64.4	567	8.1	16	1023	pubished data re-analysed
Botswana	7	Ghanzi farmland - Sandveld	7.1.0	2009	Kent 2011	none	Semi-arid bush savannah	Kalahari xeric savannah	private free-hold and lease-hold	livestock ranching	soft loose Kalahari sand	*Panthera pardus*; *Acinonyx jubatus*; *Parahyaena brunnea*	5	9.9 (6.5)	49.5	264	5.3	20	990	pubished data re-analysed
Botswana	8	Okavango Delta - NG/29 & NG/30	1.1.7	2007	CW Winterbach	partial	Seasonal floodplains with riparian vegetation	Zambezian mopane woodlands; Zambezian flooded grasslands	leased concessions on tribal land	conservation + non consumptive wildlife tourism	Kalahari sand	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	8	20.2 (5.7)	161.8	1507	9.3	27	563.9	partially published

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Botswana	9	Masame - NG/43	1.3.2	2011	CW Winterbach	partial	Mixed mopane and *Acacia* woodland and shrubland	Kalahari xeric savannah; Zambezian mopane woodlands	leased concessions on state land	conservation + non consumptive wildlife tourism	Kalahari sand	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	19	19.9 (4.5)	377.6	3452	9.1	46	843.7	partially published
Botswana	9	Masame - NG/43	1.3.2	2012	CW Winterbach	partial	Mixed mopane and *Acacia* woodland and shrubland	Kalahari xeric savannah; Zambezian mopane woodlands	leased concessions on state land	conservation + non consumptive wildlife tourism	Kalahari sand	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	13	18.3 (4.8)	237.4	3452	14.5	31	550.4	partially published
Botswana	9	Masame - NG/43	1.3.2	2013	CW Winterbach	partial	Mixed mopane and *Acacia* woodland and shrubland	Kalahari xeric savannah; Zambezian mopane woodlands	leased concessions on state land	conservation + non consumptive wildlife tourism	Kalahari sand	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	19	20.9 (8.1)	396.3	3452	8.7	29	573.0	partially published
Botswana & South Africa	10	Kalahari Transfrontier Park - Dune (Other)	2.2.1	1998-2001	Funston *et al.* 2001	full - national park	Vegetated dunes and shrubby grassland	Kalahari xeric savannah	state	conservation + non consumptive wildlife tourism	sandy	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	35	34.1 (10.6)	1190	10043	8.4	62	2146.4	pubished data re-analysed
Botswana & South Africa	11	Kalahari Transfrontier Park - Dune (Study)	2.2.1	1998-2001	Funston *et al.* 2001	full - national park	Vegetated dunes and shrubby grassland	Kalahari xeric savannah	state	conservation + non consumptive wildlife tourism	sandy	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	21	32.5 (11.5)	672	5590	8.3	41	1334.1	pubished data re-analysed

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Botswana	12	Kalahari Transfrontier Park - Tree Mabuasehube	2.2.1	1998-2001	Funston *et al.* 2001	full - national park	Sparse to open tree-savannah with large areas of grassy plains	Kalahari xeric savannah	state	conservation + non consumptive wildlife tourism	sandy	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	16	28.2 (5.2)	451	2042	4.5	71	2002.6	pubished data re-analysed
Botswana	13	Kalahari Transfrontier Park - Tree Mosimane	2.2.1	1998-2001	Funston *et al.* 2001	full - national park	Sparse to open tree-savannah with large areas of grassy plains	Kalahari xeric savannah	state	conservation + non consumptive wildlife tourism	sandy	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	12	29.5 (2.8)	372	1836	4.9	39	1150.3	pubished data re-analysed
Botswana	14	Kalahari Transfrontier Park - Tree (Other)	2.2.1	1998-2001	Funston *et al.* 2001	full - national park	Sparse to open tree-savannah with large areas of grassy plains	Kalahari xeric savannah	state	conservation + non consumptive wildlife tourism	sandy	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	49	35.1 (21.0)	1720	14115	8.2	55	1932.0	pubished data re-analysed
Botswana	15	Kalahari Transfrontier Park - Tree Sesatswe	2.2.1	1998-2001	Funston *et al.* 2001	full - national park	Sparse to open tree-savannah with large areas of grassy plains	Kalahari xeric savannah	state	conservation + non consumptive wildlife tourism	sandy	*Panthera leo*; *Panthera pardus*; *Acinonyx jubatus*; *Crocuta crocuta*; *Parahyaena brunnea*; *Lycaon pictus*	15	34.8 (5.7)	525	2142	4.1	30	1042.7	pubished data re-analysed

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Namibia	16	Central Namibian commercial farmland	n/a	2008	FJ Weise	none	*Acacia-dominated highland savannah*	Kalahari xeric savannah	private free-hold	livestock and wildlife ranching; tourism; occasional trophy hunts	mixed sandy and rock with dusty top-soils	*Panthera pardus; Acinonyx jubatus; Parahyaena brunnea*	7	11.3 (1.6)	78.9	159	2.0	76	682.9	unpublished data
Zimbabwe	17	Savé Valley Conservancy - North	n/a	2008	Williams *et al.* 2016	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	7	21.6 (3.8)	151.0	1039	3.4	14	302.1	pubished data re-analysed
Zimbabwe	18	Savé Valley Conservancy - South	n/a	2008	Williams *et al.* 2016	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	6	20.4 (4.4)	124.8	800	3.6	11	224.7	pubished data re-analysed

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Zimbabwe	17	Savé Valley Conservancy - North	n/a	2009	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	7	21.8 (3.9)	152.3	1039	6.8	7	152.3	unpublished reports
Zimbabwe	18	Savé Valley Conservancy - South	n/a	2009	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	6	20.7 (4.3)	123.9	800	6.5	6	123.9	unpublished reports
Zimbabwe	17	Savé Valley Conservancy - North	n/a	2010	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	7	21.8 (3.8)	152.9	1039	6.8	7	152.9	unpublished reports

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Zimbabwe	18	Savé Valley Conservancy - South	n/a	2010	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	6	20.0 (4.5)	119.9	800	6.7	6	119.9	unpublished reports
Zimbabwe	17	Savé Valley Conservancy - North	n/a	2011	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	7	21.8 (3.9)	152.4	1039	6.8	7	152.4	unpublished reports
Zimbabwe	18	Savé Valley Conservancy - South	n/a	2011	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	6	21.4 (5.1)	128.1	800	6.2	6	128.1	unpublished reports

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Zimbabwe	17	Savé Valley Conservancy - North	n/a	2012	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	11	22.3 (4.2)	245.0	1039	4.2	11	245.0	unpublished reports
Zimbabwe	18	Savé Valley Conservancy - South	n/a	2012	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	6	21.3 (5.1)	127.9	800	6.3	6	127.9	unpublished reports
Zimbabwe	17	Savé Valley Conservancy - North	n/a	2015	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	7	22.4 (3.5)	157.0	1039	6.6	7	157.0	unpublished reports

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Zimbabwe	18	Savé Valley Conservancy - South	n/a	2015	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	6	21.9 (5.2)	131.3	800	6.1	6	131.3	unpublished reports
Zimbabwe	17	Savé Valley Conservancy - North	n/a	2016	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	7	22.5 (3.5)	157.3	1039	6.6	7	157.3	unpublished reports
Zimbabwe	18	Savé Valley Conservancy - South	n/a	2016	RJ Groom	partial - private conservancy	Deciduous woodland savannah dominated by *Colophospermum mopane*, *Acacia–Combretum* woodland and *Acacia tortillis* woodland	Zambezian mopane woodlands; Dry miombo woodlands	private	consumptive (predominantly) and non consumptive wildlife based land use	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	6	22.0 (5.2)	132.0	800	6.1	6	132.0	unpublished reports

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Zimbabwe	19	Gonarezhou National Park - North	n/a	2011	RJ Groom	full - national park	Mopane, Miombo and alluvial woodlands	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	11	20.1 (7.9)	220.7	1665	7.5	11	220.7	unpublished reports
Zimbabwe	20	Gonarezhou National Park - South	n/a	2011	RJ Groom	full - national park	Mixed woodlands, dominated by *Colophospermum mopane* and *Combretum spp.*	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	13	23.4 (8.3)	304.1	2121	7.0	13	304.1	unpublished reports
Zimbabwe	19	Gonarezhou National Park - North	n/a	2012	RJ Groom	full - national park	Mopane, Miombo and alluvial woodlands	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	11	20.0 (7.4)	220.4	1665	7.6	11	220.4	unpublished reports
Zimbabwe	20	Gonarezhou National Park - South	n/a	2012	RJ Groom	full - national park	Mixed woodlands, dominated by *Colophospermum mopane* and *Combretum spp.*	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	11	25.7 (7.9)	282.2	2121	7.5	11	282.2	unpublished reports
Zimbabwe	19	Gonarezhou National Park - North	n/a	2013	RJ Groom	full - national park	Mopane, Miombo and alluvial woodlands	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	10	23.0 (6.8)	229.8	1665	7.2	10	229.8	unpublished reports

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Zimbabwe	20	Gonarezhou National Park - South	n/a	2013	RJ Groom	full - national park	Mixed woodlands, dominated by *Colophospermum mopane* and *Combretum spp.*	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	12	25.2 (8.4)	301.8	2121	7.0	12	301.8	unpublished reports
Zimbabwe	19	Gonarezhou National Park - North	n/a	2014	RJ Groom	full - national park	Mopane, Miombo and alluvial woodlands	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	10	22.8 (5.0)	228.2	1665	7.3	10	228.2	unpublished reports
Zimbabwe	20	Gonarezhou National Park - South	n/a	2014	RJ Groom	full - national park	Mixed woodlands, dominated by *Colophospermum mopane* and *Combretum spp.*	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	12	24.6 (8.1)	295.0	2121	7.2	12	295.0	unpublished reports
Zimbabwe	19	Gonarezhou National Park - North	n/a	2015	RJ Groom	full - national park	Mopane, Miombo and alluvial woodlands	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	11	21.8 (5.1)	239.4	1665	7.0	11	239.4	unpublished reports
Zimbabwe	20	Gonarezhou National Park - South	n/a	2015	RJ Groom	full - national park	Mixed woodlands, dominated by *Colophospermum mopane* and *Combretum spp.*	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	12	24.8 (8.5)	297.1	2121	7.1	12	197.1	unpublished reports

Country	Map ID	Site/Stratum	Carnivore strata (Winterbach *et al.* 2014)	Year	Source	Protection status	Main habitat	Ecoregion (following Dinerstein *et al.* 2017)	Land tenure	Main land use(s)	Substrate(s)	Focal species	No. of transects	Mean transect length (SD) in	Cumulative transect length	Survey area size (km2)	Transect penetration of survey	Total no. of transects	Total sampling effort (km)	Notes
Zimbabwe	19	Gonarezhou National Park - North	n/a	2016	RJ Groom	full - national park	Mopane, Miombo and alluvial woodlands	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	11	21.3 (5.0)	234.0	1665	7.1	11	234.0	unpublished reports
Zimbabwe	20	Gonarezhou National Park - South	n/a	2016	RJ Groom	full - national park	Mixed woodlands, dominated by *Colophospermum mopane* and *Combretum spp.*	Zambezian-Limpopo mixed woodlands; Zambezian mopane woodlands; Dry miombo woodlands	state	conservation + non consumptive wildlife tourism	sandy	*Lycaon pictus; Panthera leo, Panthera pardus, Crocuta crocuta (and also Acinonyx jubatus and Parahyaena brunnea)*	12	25.0 (8.4)	299.9	2121	7.1	12	299.9	unpublished reports

3.3.2 Data collection

All surveys focussed on monitoring of the southern African large carnivore guild, i.e. lion (*Panthera leo* Linnaeus, 1758), leopard (*Panthera pardus* Linnaeus, 1758), cheetah (*Acinonyx jubatus* Schreber, 1775), spotted hyaena (*Crocuta crocuta* Erxleben, 1777), brown hyaena (*Parahyaena brunnea* Thunberg, 1820), and African wild dog (*Lycaon pictus* Temminck, 1820), and used Stander's (1998) line transect survey design. The original method involves the recording of carnivore track encounters (also called "incidences", Funston *et al*. 2010) found along pre-defined line transects (typically roads or fire breaks) in order to measure mean distance between track incidences (termed "spoor frequency" by Stander 1998); i.e. the number of km per track incidence. Multiple animals (social groups) may contribute to a single track encounter/incidence (a set of tracks), but these are recorded as a single event (Stander 1998). The researcher records the number of individuals per track incidence to calculate track density, i.e. the number of individual tracks per 100 km of sampling effort (Stander 1998).

Surveys were conducted from vehicles driven along dirt road transects at variable speeds < 20 km/h, each manned with one or two experienced local trackers. The time of sampling varied across surveys but focussed on early morning or late afternoon hours. Upon detection of large carnivore tracks, survey teams recorded the following information if tracks were ≤ 24 hours old: date, time, GPS location (latitude/longitude), km from the start, species, the total number of animals, and, where possible, the sex and age class of animals.

3.3.3 Data preparation

We provide a step-wise explanation of our analysis, which simultaneously serves as an updated guideline to derive carnivore densities (Appendix 2). We treated tracks from a group of animals as one incidence (Stander 1998; Funston *et al*. 2010). Published data were included if the method described the calculation of the mean distance between track incidences as the distance between groups (Kent 2011), not individuals (e.g. Williams *et al*. 2016). We recalculated the mean distance between track incidences from the available raw data of Williams *et al*. (2016), excluding data without distances between incidences. We excluded published raw data that we could not conclusively verify or recalculate correctly. We calculated results as follows:

We created one mega-transect per study site, by joining individual transects completed in chronological order. The location of each track incidence is expressed in km from the start of the first transect, and we calculated the distance between successive track incidences per species before calculating the mean distance between track incidences (km/track incidence) (See Stander (1998) and

Funston *et al*. (2010)). A total of *n* track incidences results in (*n-1*) distances between track incidences. We calculated track density from the total number of individuals recorded and the total distance surveyed (individuals / 100 km).

3.3.4 The mean distance between track incidences and track density relationship

We randomly allocated 10% of the screened data sets per species as independent data to validate our species-specific models describing the relationships between the mean distance between track incidences and track density (Starfield, Smith & Bleloch 1990). For each species, we fitted power curves in SPSS version 20.0.0 (IBM 2013) with track density as the dependent variable and mean distance between track incidences as the independent variable. R^2 and significance values specified the strength of the relationship between the variables.

We used the selected independent data to test species models to estimate track density with confidence limits from the mean distances between track incidences. We calculated the mean distance between track incidences with SD, CV, SE and RSE. We used bootstrap estimates with replacement (20,000 iterations) of the mean distance between track incidences, SE, RSE with 95% upper and lower confidence limits (CL) calculated with the percentile method in program R version 3.3.2 (R Development Core Team 2008). We estimated track density with a 95% CL from species-specific calibrations. We expected that the 95% CL for track density estimated from the mean distance between track incidences should include the survey track density (*number of individual tracks *100 / survey length*) calculated from survey data.

3.3.5 Density estimates and evaluation parameters

We used the general model for sandy substrates (Winterbach *et al*. 2016) to estimate carnivore density from track density (*track density / 3.26 = animals / 100 km^2*). We calculated the mean track density with 95% confidence limits and evaluation parameters in three different ways with the 2007 transect data from the Okavango Delta study site. The statistical correct way is to consider all temporal repeats of a transect as one transect (Buckland *et al*. 1993), for example, repeating a transect of 20 km five times, results in one transect of 100 km for analysis. We also treated temporal repeats of a transect as separate transects in analysis. This violates the assumption of independence, but Funston *et al*. (2010) justified this approach due to the low encounter rate of carnivore tracks and reasonable sampling intervals. We compared these results with the results calculated from the mega transect, using the distance between track incidences / track density relationships to estimate mean track density with confidence intervals.

We used the following parameters to evaluate the quality of the density estimates: CV measures sample variability and SE variability of the mean (Quinn & Keough 2002; Sokal & Rohlf 1995). We expressed SE as RSE, where *RSE = SE * 100 / mean* (called CV by Stander (1998) and Funston *et al*. (2010)). In addition, we used Percentage Confidence Limit *PCL = ((Upper limit – lower limit)/2) * 100 / mean* (Barnes 2002, Ferreira & Funston 2010) with lower values of RSE and PCL indicating higher precision. SE and confidence limits were estimated with bootstrap (Canty & Ripley 2019) in the program R version 3.3.2 (R Development Core Team 2008).

We considered samples with less than 10 distances between track incidences as small samples, because the CV has not stabilised. We used the survey track density to estimate the upper CL for carnivore density, mean carnivore density as *Upper limit / 2* and lower limit as 0.0 animals / 100 km^2.

The track survey density estimates for lion density was compared with the reference density of lions in the long-term study area. The reference density was calculated using the known lions from the long-term study and other lions present at the time of survey.

3.3.6 Detecting population trends

We used the CV to assess the ability to detect changes in population density, with power set at 80%. That is a 20% probability of not finding a population change while one is present (Gerrodette 1987). We used CV and RSE in Gerrodette’s (1987) inequality, to demonstrate the impact on the assessment of power to detect trends. We selected the following parameters: 1-tailed test at p =0.05, power at 0.8, total population decline of 50% linear or exponential over time frame. CV was set to increase with declining population. We wanted to vary the starting CV from 20% to 100% with 10% increments to assess the change in the number of years required to detect a 50% decline in a population based on these parameters.

We used annual carnivore density estimates from the NG/43 study site to compute linear regressions to test for significant population trends. As an alternative to trend analysis, we used the χ^2 test with Bonferroni intervals to test for years with observed frequencies of each species that differed significantly from the expected frequencies (Byers *et al*. 1984). We classified track observations of individuals into years independently (observed frequencies) and all years had expected frequencies of >5%. We calculated expected proportions from sampling effort (survey length in km) per year. The relative proportion surveyed each year of the total sampling effort (surveys combined in km) provides the proportion of each species tracks expected per survey.

3.4 Results

3.4.1 The mean distance between track incidences and track density relationship

We obtained high R-square values (0.723 – 0.986) and significant relationships ($P < 0.001$) between mean distance between track incidences and track densities of brown hyaena, leopard, lion and spotted hyaena (Table 3-2). For cheetah and wild dog, relationship slopes (B) were significant, but constants were not ($P > 0.05$) (Table 3-2). Figure 3-2 shows species-specific calibration curves for the relationship between the mean distance between track incidences and track density.

We tested these calibrations with independent data. The estimated track density confidence limits included all survey track densities (Table 3-3). The PCL for track density, and thus also for estimated carnivore density, decreased from 130% to 25% with increasing sample size, i.e. from <10 to >60 track incidences respectively (Fig. 3-3).

3.4.2 Mean distance between track incidences of groups vs individuals

Treating individual animals of a group as separate track incidences with zero inter-incidence distances result in zero inflation of the data, and reduced mean distance between track incidences (Table 3-4). By splitting groups into individuals, our sample size of the number of track incidences approximately doubled for lion, spotted hyaena and cheetah, and the mean distance between track incidences were in the order of 50% of the values calculated correctly, while CV, RSE and PCL were inflated (Table 3-4).

3.4.3 Density estimates and evaluation parameters

Figure 3-4 shows changes in bootstrap estimates of the CV, RSE and PCL with increasing sample size for leopard track incidences at the NG/29/NG30 study site. The bootstrap estimate of the CV stabilised after 10 track incidences, while the RSE declined below the selected threshold of 20% after 20 incidences.

The density estimates and evaluation parameters calculated from transects and the distance between method, are presented in Table 3-5. The CI for lion density estimates (distance between method) of 1.81 – 3.81 lions / 100 km^2 (Table 3-5), included the reference density of 3.42 lions / 100 km^2 and the survey density of 2.91 lions / 100 km^2 (54 lion tracks recorded on 569.3 km). The reference density consisted of the known lion population in the long-term study site with 17 adults and subadults plus 9 cubs. Six of the cubs were still hidden in a den and therefore excluded from the reference density calculation. There were five more lions present in the area at the time of the survey, resulting in a total of 22 adult and subadult lions plus three cubs old enough that they were moving around with the older lions. The long-term study site was 730 km^2. The resulting reference density was 3.01 adult and sub adult lions / 100 km^2 and 3.42 lions / 100 km^2 for all ages (excluding small cubs that are not moving around).

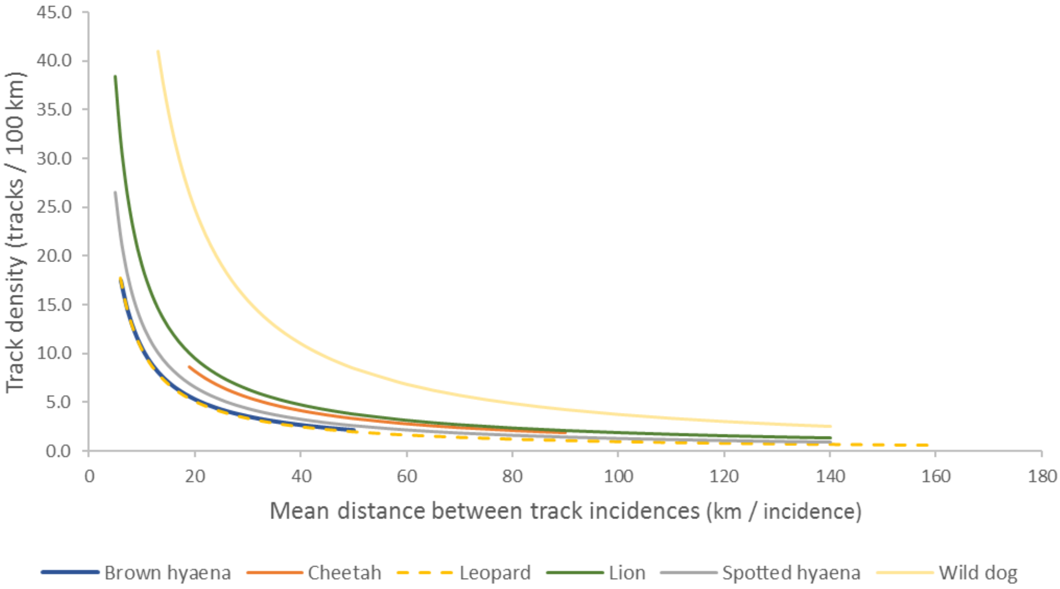

Figure 3-2 The relationship between mean distance between track incidences and track density for six large carnivores in southern Africa.

Table 3-2 Summary of mean distance between track incidences (x) and track density (y) relationships estimated for the southern African large carnivore guild. Data are from studies across Botswana, Namibia, South Africa and Zimbabwe.

Species	Equation	n	R	R Square	Adjusted R Square	SE	P value	
							B (slope)	Constant
Brown hyaena	$y=103.707x^{-0.995}$	15	0.971	0.942	0.938	0.131	< 0.001	< 0.001
Cheetah	$y=158.455x^{-0.987}$	12	0.921	0.848	0.833	0.232	< 0.001	0.067
Leopard	$y=113.13x^{-1.034}$	38	0.993	0.986	0.985	0.122	< 0.001	< 0.001
Lion	$y=193.863x^{-1.005}$	32	0.930	0.865	0.86	0.388	< 0.001	< 0.001
Spotted hyaena	$y=133.975x^{-1.006}$	43	0.978	0.957	0.956	0.177	< 0.001	< 0.001
Wild dog	$y=820.02x^{-1.168}$	22	0.850	0.723	0.709	0.494	< 0.001	0.091

Table 3-3 Testing calibrations of species-specific track density/mean distance between track incidences relationships to estimate track density with 95% confidence limits (CL) from mean distance between track incidences and bootstrap estimates of 95% confidence limits.

Independent test data for six large carnivores were derived from multiple study sites across southern Africa.

Species	Area	Survey	Track incidences	Track density (tracks / 100 km)	Mean distance between track incidences (km / incidence)	CV (%)	Estimate track density (tracks / 100 km)	Survey Density (animals / 100 km2)	Estimate Density (animals / 100 km^2)	PCL (%)
Brown hyaena	Central Kalahari GR 2.1.3	2012	62	7.3	15.3 (12.1 - 20.1)	102	6.9 (5.2 - 8.7)	2.3	2.1 (1.6 - 2.7)	25
Cheetah	Gonarezhou NP South	2014	10	6.1	28.2 (14.2 - 46.7)	92	5.9 (3.6 - 11.5)	1.9	1.8 (1.1 - 3.5)	68
Leopard	Chobe 1.3.4	2005	7	4.9	15.1 (6.9 - 33.0)	106	6.8 (3.0 - 15.4)	1.5	2.1 (0.9 - 4.7)	90
Leopard	Gonarezhou NP North	2011	31	15.4	6.6 (5.0 - 8.8)	80	16.0 (12.0 - 21.5)	4.7	4.9 (3.7 - 6.6)	30
Leopard	Masame – NG/43 1.3.2	2013	17	3.5	32.5 (20.7 - 50.6)	93	3.1 (2.0 - 4.9)	1.1	0.9 (0.6 - 1.5)	48
Lion	Chobe 1.4.1	2006	8	3.9	39.4 (16.8 - 73.9)	99	4.8 (2.6 - 11.4)	1.2	1.5 (0.8 - 3.5)	91
Lion	Gonarezhou NP South	2011	9	7.6	25.3 (10.7 - 51.3)	117	7.5 (3.7 - 18.0)	2.3	2.3 (1.1 - 5.5)	95
Lion	Savé Valley Conservancy North	2012	17	16.7	11.3 (7.2 - 16.2)	83	16.9 (11.8 - 26.8)	5.1	5.2 (3.6 - 8.2)	44
Lion	Savé Valley Conservancy North	2016	20	26.1	11.3 (7.2 - 16.2)	104	16.9 (11.8 - 26.8)	8.0	5.2 (3.6 - 8.2)	44
Spotted hyaena	Chobe 1.2.1	2006	24	48.1	3.5 (2.1 - 6.5)	138	38.1 (20.4 - 63.5)	14.8	11.7 (6.3 - 19.5)	57
Spotted hyaena	Chobe 1.3.4	2005	17	27.9	8.3 (3.7 - 17.8)	158	16.0 (7.4 - 35.5)	8.6	4.9 (2.3 - 10.9)	88
Spotted hyaena	Chobe 1.3.4	2006	44	31.7	4.7 (3.2 - 8.4)	157	28.0 (15.7 - 41.6)	9.7	8.6 (4.8 - 12.8)	46
Spotted hyaena	Masame – NG/43 1.3.2	2011	63	10.8	13.3 (10.1 - 18.0)	117	9.9 (7.3 - 13.1)	3.3	3.0 (2.2 - 4.0)	29
Spotted hyaena	Savé Valley Conservancy North	2015	27	22.9	6.0 (4.6 - 7.8)	70	22.2 (16.9 - 28.7)	7.0	6.8 (5.2 - 8.8)	27
Wild dog	Savé Valley Conservancy North	2008	13	33.8	24.0 (14.4 - 33.1)	73	20.1 (13.8 - 36.4)	10.4	6.2 (4.2 - 11.2)	56
Wild dog	Savé Valley Conservancy North	2012	22	44.9	11.5 (8.6 - 15.6)	72	47.1 (33.2 - 66.6)	13.8	14.4 (10.2 - 20.4)	35

Table 3-4 Differences in mean distance between track incidences (km/track incidence), coefficient of variation (CV), relative standard error (RSE) and percent confidence limit (PCL) when analysing track incidences at group and individual level.

Species and Site	Incidences per group									Incidences per individual								
	n	Mean	CL lower	CL upper	SD	CV	SE	RSE (%)	PCL (%)	n	Mean	CL lower	CL upper	SD	CV	SE	RSE (%)	PCL (%)
Leopard Okavango	29	18.44	12.79	25.13	16.89	91.64	3.14	17.02	33.49	31	17.25	11.71	23.49	16.96	98.32	3.05	17.66	34.13
Lion Okavango	24	22.16	14.57	31.11	20.77	93.71	4.24	19.13	37.32	53	10.03	5.63	15.15	17.74	176.78	2.44	24.28	47.45
Spotted hyaena Okavango	87	6.13	4.30	8.48	10.01	163.34	1.07	17.51	34.08	156	3.42	2.29	4.83	8.05	235.74	0.64	18.87	37.17
Cheetah Namibia	17	37.81	16.21	65.54	53.77	142.24	13.04	34.50	65.24	34	18.90	6.89	34.12	42.07	222.58	7.22	38.17	72.03

Table 3-5 Comparison of density estimates and evaluation parameters calculated from individual transects and replicates treated as individual transects in comparison to results from distance between track incidences method. Data is from the Okavango Delta site in 2007.

	Lion			Leopard			Spotted hyaena			Cheetah		
Method	Individual transect	Replicates as transects	Distance between	Individual transect	Replicates as transects	Distance between	Individual transect	Replicates as transects	Distance between	Individual transect	Replicates as transects	Distance between
Mean tracks/100 km	8.40	9.42	22.03	5.33	5.57	18.44	24.70	25.22	6.13	0.69	0.76	
SD	7.26	14.41	20.84	2.65	5.25	16.90	24.49	31.46	10.01	1.44	2.62	
SE	2.74	2.77	4.15	1.00	1.01	3.06	9.26	6.05	1.06	0.55	0.50	
n	7	27	24	7	27	29	7	27	88	7	27	2
CI	6.71	5.70		2.46	2.08		22.65	12.45		1.33	1.04	
CI minimum	1.69	3.72	15.39	2.87	3.49	13.52	2.06	12.78	4.58	-0.64	-0.28	
CI maximum	15.11	15.12	32.37	7.78	7.65	25.98	47.35	37.67	9.13	2.03	1.80	
Mean density	2.58	2.89	2.66	1.63	1.71	1.70	7.58	7.74	6.64	0.21	0.23	0.14
Minimum density	0.52	1.14	1.81	0.88	1.07	1.20	0.63	3.92	4.44	-0.20	-0.08	0.00
Maximum density	4.63	4.64	3.81	2.39	2.35	2.35	14.52	11.55	8.88	0.62	0.55	0.27
CV	86	153	95	50	94	92	99	125	163	208	344	
RSE	33	29	19	19	18	17	37	24	17	79	66	
PCL	80	61	38	46	37	34	92	49	33	192	136	

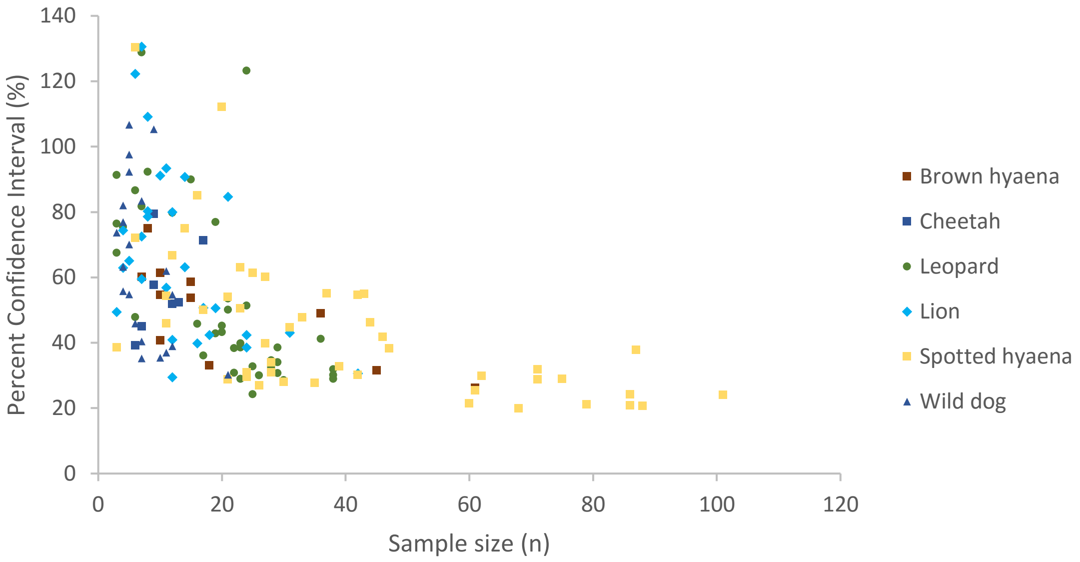

Figure 3-3 Percent confidence limits (PCL) of density estimates plotted against the number of track incidences for six large carnivores. Data represent study sites across southern Africa.

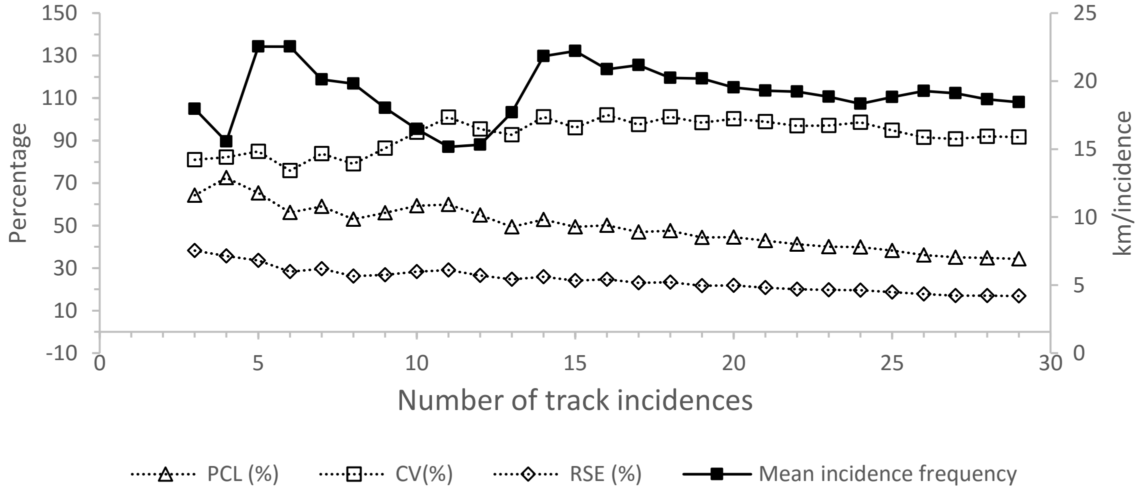

Figure 3-4 Mean distance between leopard track incidences with Coefficient of Variation (CV) and Relative Standard Error of the Mean (RSE) and Percent Confidence Length (PCL) calculated from bootstrap estimates with replacement of the of Standard Error and the 95% confidence interval. Results are shown for a sample of 3, 4, 5, …., 30 samples randomly drawn from the entire sample of 30 track incidences. Data collected at the NG29/NG30 study site.

RSE was below 20% for lion, leopard and spotted hyaena densities calculated with the distance between method (Table 3-5). Although the CV for these species were lower when calculated treating repeats of an individual transect as one extended transect, it resulted in the highest PCL. The distance between method provided the lowest PCL. Spotted hyaena and leopard had the lowest PCL, followed by lion.

Cheetah were recorded only three times on the track survey and evaluation criteria could not be calculated with the distance between method. The proposed small sample size protocol resulted in a cheetah density of 0.0 to 0.27 cheetah / 100 km^2.

3.4.4 Detecting population trends

Stander (1998) and Funston *et al*. (2010) set the target RSE at < 20% for mean distance between track incidences. Assuming that this was reached with a sample size of 25 track incidences, the corresponding CV will be 100% ($RSE = CV / \sqrt{n}$). Using the software TRENDS, we wanted to solve the time frame to detect a total population decline of 50%, declining linear or exponentially. We set a 1-tailed test at p =0.05, power at 0.8 and CV increasing with decreasing population. The intention was to resolve the time frame to detect a 50% population decline, varying the starting CV from 20% to 100% with 10% increments. The highest CV that we could use in the calculation was 39%. With a starting CV of 20% it would take 9 to 10 years to detect a 50% population decline for linear and exponential declines respectively. At 39% CV, this time frame would increase to between 31 and 33 years.

CV varied between 70% and 163% for different species and sites (Tables 3-3, 3-4, 3-5). The CV for leopard and spotted hyaena from 2011 to 2013 in NG43 varied between 88% and 112% (Table 3-6). Linear regression showed no significant population trends in NG43 from 2011 to 2013 (slope not significantly different from zero) for leopard (P = 0.977) and spotted hyaena (P = 0.664). Due to only one track incidence recorded during the 2013 survey in NG/43, we were unable to calculate a lion density; thus, a trend analysis was not possible for lion. Given the high CV for track surveys we did not expect to reject the null hypothesis, that the slope of regression did not differ significantly from 0.

Using χ^2 with Bonferroni intervals, we found that the proportion of leopard (χ^2 = 1.14, df = 2, *P* >0.05) and lion (χ^2 = 5.25, df = 2, *P* >0.05) recorded per survey effort did not differ significantly among the three years (Tables 3-7). The Bonferroni confidence intervals for leopard did not show any significant differences among years (Table 3-6), indicating a stable leopard population.

Table 3-6 Summary of leopard, lion and spotted hyaena density estimates from track surveys conducted in NG/43 from 2011 to 2013.

Species	Survey	Survey length (km)	Track incidences	Track density (tracks / 100 km)	Mean distance between track incidences (km / incidence)	CV (%)	Estimate track density (tracks / 100 km)	Survey Density (animals / 100 km^2)	Estimate Density (animals / 100 km^2)	PCL (%)
Leopard	2011	843.7	24	3.1	31.8 (21.5 - 47.7)	0.99	3.2 (2.1 - 4.7)	0.9	1.0 (0.6 - 1.5)	41.9
Leopard	2012	550.4	23	4.2	23.6 (16.8 - 34.5)	0.88	4.3 (2.9 - 6.1)	1.3	1.3 (0.9 - 1.9)	37.1
Leopard	2013	573.0	17	3.5	31.4 (19.3 - 59.6)	1.17	3.2 (1.7 - 5.3)	1.1	1.0 (0.5 - 1.6)	56.8
Lion	2011	843.7	7	1.3	N/A	0.87	N/A	0.4		
Lion	2012	550.4	4	1.3	N/A	0.58	N/A	0.4		
Lion	2013	573.0	1	0.2	N/A	N/A	N/A	0.1		
Spotted hyaena	2011	843.7	63	10.8	13.1 (9.8 - 17.0)	1.10	10.0 (7.7 - 13.4)	3.3	3.1 (2.4 - 4.1)	28.3
Spotted hyaena	2012	550.4	28	5.8	18.4 (13.2 - 25.6)	0.90	7.1 (5.1 - 10.0)	1.8	2.2 (1.6 - 3.1)	33.8
Spotted hyaena	2013	573.0	34	8.6	15.3 (10.5 - 22.4)	1.12	8.6 (5.9 - 12.6)	2.6	2.6 (1.8 - 3.9)	39.4

Table 3-7 Bonferroni intervals to test for differences in the observed and expected number of carnivores recorded during track surveys in NG/43, northern Botswana, between 2011 and 2013 (k=3, α =0.05, Z =2.6384).

Expected proportions were calculated from survey efforts of 843.7 km (2011), 550.4 km (2012) and 573.0 km (2013).

Species	Survey	Number of animals		χ^2	Proportion of animals		Pi(1-Pi)/n	Bonferroni interval (*Pi*)	Use index	Selection
		Expected	Observed		Expected (*Pio*)	Observed *(Pi)*				
Leopard	2011	29.6	26	0.44	0.4289	0.3768	0.0583	0.2229 ≤ Pi ≤ 0.5307	0.88	0
	2012	19.3	23	0.71	0.2798	0.3333	0.0568	0.1836 ≤ Pi ≤ 0.4831	1.19	0
	2013	20.1	20	0.00	0.2913	0.2899	0.0546	0.1457 ≤ Pi ≤ 0.4340	1.00	0
	TOTAL	69	69	1.14	1.0000	1.0000				
Lion	2011	8.1	11	1.00	0.4289	0.5789	0.1133	0.2801 ≤ Pi ≤ 0.8778	1.35	0
	2012	5.3	7	0.53	0.2798	0.3684	0.1107	0.0764 ≤ Pi ≤ 0.6604	1.32	0
	2013	5.5	1	3.72	0.2913	0.0526	0.0512	-0.0825 ≤ Pi ≤ 0.1878	0.18	-[*]
	TOTAL	19	19	5.25	1.0000	1.0000				
Spotted hyaena	2011	73.8	91	4.02	0.4289	0.5291	0.0381	0.4287 ≤ Pi ≤ 0.6295	1.23	0
	2012	48.1	32	5.40	0.2798	0.1860	0.0297	0.1078 ≤ Pi ≤ 0.2643	0.66	-[*]
	2013	50.1	49	0.02	0.2913	0.2849	0.0344	0.1941 ≤ Pi ≤ 0.3757	0.98	0
	TOTAL	172	172	9.45[*]	1.0000	1.0000				

[*] Significant at P = 0.05

Significantly fewer lions were recorded in 2013 than 2011 and 2012 (Table 3-7). The number of spotted hyaena differed significantly among years ($\chi^2 = 9.45$, df = 2, $P = 0.05$), with significantly fewer animals recorded in 2012 (Table 3-6).

3.5 Discussion

Funston *et al*. (2010) proposed to use the mean distance between track incidences to estimate confidence intervals for track density, and thus for carnivore density. Subsequent studies did not follow this approach (see Bauer *et al*. 2014; Boast & Houser 2012; Kent & Hill 2013; Winterbach *et al*. 2017), in part due to the lack of species-specific calibrations of the relationships between the mean distance between track incidences and track density.

We derived these calibrations for the southern African large carnivores. Testing the calibrations with field data supported this approach to estimate density confidence intervals. The estimated confidence intervals for track densities included the survey track densities for all six species. The confidence interval for the lion density included the one lion reference density we had for comparison. Our approach provides an improved, and simplified way to estimate track density confidence intervals directly from the mean distance between track incidences, instead of using transect based calculations.

Due to the low encounter rate of carnivore tracks, track surveys often entail spatial and temporal replicates of transects (Boast & Houser 2012; Funston *et al*. 2010; Stander 1998). The assumption of independence can be ignored if transects are placed randomly and a robust estimator of variance is employed (Buckland *et al*. 1993).

The threshold selected for adequate sampling in track surveys is an RSE of ≤ 20% for mean distance between track incidences, and is in general achieved with 20 to 30 track incidences (Stander 1998; Funston *et al*. 2010). CV stabilised after 10 track incidences. Estimating confidence intervals with the distance between method resulted in lower CV, RSE and PCL compared to transect based calculations.

The confidence interval for the lion density estimate at the Okavango study site included the reference density. The transect based analysis of a track survey on sandy soils in Kafue National Park (Zambia) was more accurate than call up surveys, but had a larger PCL (Midlane *et al*. 2015).

However, using track surveys on the clay soils of Serengeti to estimate lion density were problematic. Track data could not be used in N-mixture models to estimate lion density (Belant *et al*. 2016) and using the lion and cheetah on clay model (Winterbach *et al*. 2016) overestimated lion density (Belant *et al*. 2019). Linear models to estimate large carnivore densities differed significantly on sandy and clay soils (Funston *et al*. 2010). The lion and cheetah on clay model (Winterbach *et al*. 2016) consisted of four data points per species, as pointed out by Belant *et al*. (2019) and may be insufficient to provide reliable density estimates.

Although confidence in results reduces as sample size decrease, conservation managers do need population estimates. We propose that a minimum of 10 track incidences are needed to estimate confidence intervals of carnivore density with the distance between method. For samples below 10 track incidences we propose to use the survey density as the upper limit of the confidence limit, zero density as the lower limit and *survey density / 2* as the estimated population density. This provides a conservative approach when carnivores are recorded at low densities.

Not following the analysis protocol will produce erroneous results. For example, we demonstrated that treating animals within a group as individual track incidences will underestimate mean distance between track incidences by up to 50%, and as a result inflate density estimates of carnivores. We provide a step-wise protocol, analysis templates and an R script (Appendix 3) to do the analysis and produce results that can be compared among studies. We incorporated bootstrap estimates of 95% confidence limits and other parameters to improve the precision of estimates from small samples.

Applying the results to conservation planning, the practitioner should take into account the valid extrapolation range of the model used, for example, the general model on sandy substrates can be used for carnivore density estimates as low as 0.27 carnivores / 100 km^2 (track density 0.88 tracks / 100 km) (Winterbach *et al*. 2016). Density estimates below this should be considered with caution, but it may be the best available data to guide conservation.

The power to detect population trends with linear or exponential regression depends on suitably small CVs (Gerrodette 1987). The power to detect population trends is conventionally set at 0.80 (Bart *et al*. 2004; Quinn & Keough 2002) and most wildlife surveys aim for a CV < 20%; above this threshold is not considered suitable for monitoring trends (Boulanger *et al*. 2002; Keeping *et al*. 2018). The CVs of our track survey data were 93 – 163%. The CV of line survey data increase as population abundance decrease, and thus the probability to detect the population change declines (Barnes 2002; Taylor *et al*. 2007).

Several track-based studies calculated the RSE metric and called it CV, including Stander's landmark publication in 1998. The threshold RSE, (not CV), has been set at <20%, a value generally achieved after 20 to 30 samples. Funston *et al*. (2010) concluded that most carnivore estimates derived from spoor indices had a high precision and that in best case scenario a 10% change in population abundance could be detected with only two surveys 1 year apart. Unfortunately, these conclusions were based on substituting RSE for the actual CV in Gerrodette's inequality (1987). This provided a false sense of the statistical power to detect population trends. An RSE of 20% (n = 25) will be the result of a CV of 100%, a far cry from the desired CV <20%.

With the CV for distance between track incidences in the order of 100%, a trend analysis is unlikely to detect a 50% decline in population size over a ten-year period, a criterium for a species to be categorised as endangered (IUCN Species Survival Commission 1994). The time frame required to detect a trend, and the magnitude of change that would have occurred in the population at that stage, limit the conservation management value of using regression for population trend analysis with track survey data. Early detection of true population trends, however, is critical for effective conservation management. We demonstrate that the χ^2 test with Bonferroni intervals provides an alternative to test for surveys with significant more or less tracks observed than expected.

A large variety of environmental, species-specific, methodological, and analytical factors can influence the results derived from track surveys. Heinemeyer *et al*. (2008) provide a useful synopsis of the strengths and weaknesses of different track-based methodologies for carnivore surveys. Inter-observer variability in detection (Wharton 2006; Wong *et al*. 2010; Zielinski & Schlexer 2009) and fatigue can affect detection probability. Experienced trackers are critical to minimize inter-observer variability (Funston *et al*. 2010; Stander *et al*. 1997), but there are no guidelines to assess tracker expertise and skill level. Light conditions change during the course of day, impacting detection probability. Tracks are best observed at low sun angles (Liebenberg 1990).

Soto Navarro *et al*. (2012) demonstrated the strong impact of climatic variables, including temperature, humidity, wind and precipitation on track preservation and detection. Rain and wind can obliterate tracks before detection and wind also hampers the correct aging of tracks.

Survey rigour can help to reduce the potential impacts of these factors. Use experienced trackers. Do not survey during the middle of the day. Assess suitability of the substrate and, if necessary, exclude unsuitable sections of a transect from analysis. Treat sections of transects where vehicles or animals, such as elephants, obliterated tracks as unsuitable sections, or redo the transect at a later stage. Postpone the survey if unfavourable conditions such as rain or strong wind occurred during the preceding 24 hours.

Changes in conditions may also influence space use and movements of animals. Belant *et al*. (2019) suggested that short term changes in space use and movements of lions resulted in high variation among weekly density estimates in Serengeti. They assumed that the RSE (which they called "CV") should be less than < 20%, because they recorded between 35 and 80 lions per week. However, the 20% threshold is typically reached after 20 to 30 track incidences (Funston *et al*. 2010), not after recording 20 to 30 individual animals. Assuming a mean group size of four animals, their sample size of track incidents was between 9 and 20 incidences. Therefore, inadequate sample size contributed to the observed high variation in weekly density estimates.

However, short term, seasonal and location differences in space use and movement will impact on results. The FMP method that incorporates day ranges is a potential solution and does not need reference densities for calibration (Keeping & Pelletier 2014). Day range movements can be estimated with body scaling when data are not readily available (Keeping 2014). We recommend surveys to repeat transects over time to capture the variation in movement and present RSE and CV as part of the evaluation parameters.

A mixture of local substrate conditions increase variability in preservation and detection of tracks (e.g. Bauer *et al*. 2014). Detection of tracks on the hard surface of clay soils can be problematic. Dragging a transect the previous afternoon can assist with the aging of tracks (Belant *et al*. 2019), but may also obliterate tracks that should have been counted as part of the past 24-hours. Dragging may also create a fine powder surface on top of the hard clay substrate, changing the track preservation characteristics of the surface.

Supporting data should be collected. Conducting herbivore censuses simultaneously along line transects, for example using the Distance Sampling method (Buckland *et al*. 1993) or track surveys (Ahlswede *et al*. 2019; Keeping *et al*. 2018), could enhance the value of carnivore track surveys. Data on prey changes contribute important insight when interpreting the carnivore density estimates. Combining surveys, however, may slow down field work and necessitate additional resources and expertise.

Researchers have access to many tools to conduct carnivore surveys (Boitani & Powell 2012; Long *et al*. 2008; Wilson & Delahay 2001), each with its own strengths, weaknesses, advantages and disadvantages. A growing body of literature compares track surveys with other carnivore monitoring strategies (Balme *et al*. 2009; Belant *et al*. 2016; Midlane *et al*. 2015; Pirie *et al*. 2016; Silveira *et al*. 2003; Torrents-Ticó *et al*. 2017). This will enable researchers to identify whether the potential benefits of track surveys outweigh the constraints in comparison to other techniques, taking into consideration the objectives.

The application of track survey data has been expanded to include occupancy modelling. Environmental variables (e.g. land cover, water availability, precipitation, vegetation cover anthropogenic impacts) were used as covariates to determine important drivers of lion distribution in conservation areas in Mozambique, West Africa and Zambia (Everatt *et al*. 2014; Henschel *et al*. 2016; Midlane *et al*. 2014).

3.6 Conclusions

Track surveys provide a practical tool to assess large carnivore population size and distribution at different scales. Due to the high CV of track survey data, trend analysis will not detect population changes reliably. We recommend χ^2 with Bonferroni intervals to identify observed frequencies that differ significantly from expected frequencies between years. We cannot over-emphasise the importance of rigorous survey design, correct analysis paradigms, and critical interpretation of the results towards track-based density estimation of large carnivores, many of which are now highly threatened.

3.7 References

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3.8 Appendix 1 Track Survey Analysis Template

Appendix 1_Track Survey_Analysis Template.xls

3.9 Appendix 2 Track Survey Work-Flow

Chronological track survey work-flow

1. Plan transects per site, ideally in a randomised fashion. See Buckland *et al*. (1993) for planning transect and line surveys. Consider important implication/requirements. Also, consider measuring biological and non-biological variables potentially influencing the results (Soto Navarro *et al*. 2012). If necessary, consult experienced researchers and/or statisticians for help.
2. Calculate penetration (total transect length to total area) to assess survey representation in relation to the entire study area.
3. Data collection. Ensure to collect/record all required data. See Appendix 4 template for data recording in the field, and Appendix 1_Track Survey Analysis Template for data entry.

   Minimum study area data:

- Accurate size of study area and accurate size of the effective survey area;
- Individual and total transect lengths;
- GPS coordinates of all transect start and end locations (latitude/longitude);
- Substrate types and condition; and
- No. of replicates per transect and total sampling distance (i.e. survey effort).

Minimum survey data:

- Date and time;
- Observers and recorders;
- Transect ID and transect type (e.g. road, fire break, etc.); and
- Start odometer/kilometre reading, and end odometer/kilometre reading.

Minimum track incidence data:

- Date and time;
- Observers and recorders;
- Odometer/kilometre reading and GPS coordinates (latitude/longitude);
- Substrate type and condition;
- Species and total number of individuals;
- Age and sex information for all individuals (if possible);
- Age of track (<24hrs, >24hrs); and
- Direction of travel.

4. Enter, check and clean data. Use Appendix 1 Track Survey Analysis Template for data entry.
5. Map all transects and track incidences in GIS to identify and correct outliers/false records.
6. Analyse data per site.
    a. Create mega-transect with distance from start for each incidence.
    b. Calculate distances between incidences for each species separately.

The following steps are computed per species:

    c. Determine if sample size is adequate
        i. Calculate bootstrap estimate of distance between track incidences and mean group size, (with SD, SE, CV (*SD/mean*100*), CL, SE) after 2,3,4…n incidences. (Use Appendix 3 R script for analyses).
        ii. The CV indicates how data are distributed around the mean at each survey site. Lower CV indicates higher power to detect population trend.
        iii. Relative SE (RSE = (*SE/mean*100*)) (called CV in Stander 1998 and Funston *et al*. 2010) indicates how precision improves with sample size for mean distance between incidences. Stander (1998) and Funston *et al*. (2010) defined the RSE threshold arbitrarily at < 20%. Funston *et al*. (2010) demonstrated that 20 to 30 spoor incidences are generally a sufficient sample size.

d. Estimate track density (tracks/100 km) with CL

    i. Use bootstrap estimate of the mean distance between track incidences with SD, SE, CV and CL for the entire data set of n samples. Bootstrap estimate of CL provides upper and lower limits that are unequal around the mean. (Use Appendix 3 R script for analyses).

    ii. Use the appropriate species-specific curve for track density – mean distance between track incidences relationship to estimate track density with CL from mean distance between track incidences with CL.

    iii. Calculate track density from mega-transect (total animals *100/total km).

    iv. Calculate PCL, including bootstrap estimates of CL: *PCL =((Upper limit – lower limit)/2)*100/mean*

    v. Divide estimate of track density (track/ 100 km) by 3.26 to derive density estimate in animals / 100 km^2 (Winterbach *et al*. 2016).

e. Test for significant trend or significant differences between years.

    i. Compute regression of estimated density against time and test for slope significantly different from zero.

    ii. Use χ^2 with Bonferroni intervals to identify observed frequencies that differ significantly from expected frequencies between years.

7. Interpret and present results.

3.10 Appendix 3 R_script

```
################################################################################################
#   R script for running bootstrap analysis for spoor data. Published in conjunction with:      #
#                                                                                    #
#   "Estimating Carnivore Density from Track Surveys: Revised Analyses for Improved                              #
#     Confidence." by Winterbach et al, 2018.                                                       #
#                                                                                    #
#                                                                                    #
#                                                                                    #
################################################################################################


#Uncomment following two lines to install required packages if not already present on your system.
#install.packages("boot")
#install.packages("beepr")


#Load required packages
library(boot)                  #Key bootstrapping function
library(beepr)                 #Non-essential but useful for large datasets. Makes a sound when command is completed.


################################################################################################
############ Load data and reformat as required #################################################
################################################################################################

#Specify the working directory. You will need to change this to suit your setup. Raw data is read from this directory,
#and the results are written to it into a file called "Bootstrap_Results.csv".
#setwd("/Users/tawny/Documents/OLMP/SpoorCalculations/")               #MacOS formatted path.
setwd("C:/Users/Christiaan/Documents/R_scripts/Spoordata/")                                                     #Windows
formatted path
```

```
#Input data from a .csv file. Details of the format of this file are also specified in the workflow document.
#The key columns are as follows and must be named exactly as listed here, including the case. The order does not matter.
#
#"Location" = Name of the survey area.
#"Section" = Various sub-divisions/strata within the Location. If it is all one section, then enter the same value
#throughout this column.
#"Species" = 'Name of species', or 'Start' or 'Stop' to represent the beginnings and ends of each transect.
#"Km_reading" = Distance from the start of each transect to the current observation. This will be "0" for every Start,
#and the full length of that transect for every Stop.
#"Animals Total" = Number of individual animals counted at each observation.
#"Unique_Transect_ID" = A value that uniquely identifies each transect driven, and that is different for repeated trips
#along the same route. The file should be sorted before loading into R using this field first, and the "TransDist" field
#from lowest to highest.
#
#These columns must be named precisely as listed, including case.
#Please avoid including non alphanumeric characters in any of the data within these key columns.

#The following line loads the formatted .csv file with your data fro the directory specified above.
#Change the filename to suit your data.
spoorData_Raw <- read.csv("NCZ_NG41_NG43_spoor_2016.csv", header = TRUE, na.strings=c(""," ","NA","FALSE"))

#Optional commands to examine your newly loaded data. Uncomment as required.
head(spoorData_Raw)
#tail(spoorData_Raw)
#spoorData_Raw
#nrow(spoorData_Raw)


##################################################################################################
############You should not need to make any changes beyond this point.###########################
##################################################################################################
```

```
#Add first two new columns, insert zero into the first record of each

spoorData_Raw[1,"Total.previous.transect"] <- 0
spoorData_Raw[1,"Distance.megatransect"] <- 0

#Move through data using for loops to generate the required sums in "Total.previous.transect"
for (i in 2:nrow(spoorData_Raw))
{
	j <- i-1
	ifelse(spoorData_Raw[j,"Species"] == "Stop",
	spoorData_Raw[i,"Total.previous.transect"] <- spoorData_Raw[j,"Km_reading"] + spoorData_Raw[j,"Total.previous.transect"],
	spoorData_Raw[i,"Total.previous.transect"] <- spoorData_Raw[j,"Total.previous.transect"])
}

#Move through data using for loops to generate the required sums in "Distance.megatransect"
for (i in 2:nrow(spoorData_Raw))
{
	spoorData_Raw[i,"Distance.megatransect"] <- spoorData_Raw[i,"Total.previous.transect"] + spoorData_Raw[i,"Km_reading"]
}


#Create the "ANALYSIS_ID" column that will be used to divide the results up by species and section. Later on the code will re-extract these data
#for the final tables, using the underscore character as a field separator. So first ensure no extraneous underscores exist.
spoorData_Raw$Species <- gsub('_', '.', spoorData_Raw$Species)
spoorData_Raw$Location <- gsub('_', '.', spoorData_Raw$Location)
spoorData_Raw$Section <- gsub('_', '.', spoorData_Raw$Section)

spoorData_Raw$ANALYSIS_ID <- as.factor(gsub('\\s+', '.', paste(spoorData_Raw$Species, spoorData_Raw$Location, spoorData_Raw$Section, sep="_")))

#Replace instances of "&" with ".". If users follow the instructions in the attendent workflow, the following line should be redundant.
spoorData_Raw$ANALYSIS_ID <- gsub('&', '.', spoorData_Raw$ANALYSIS_ID)
```

```
#Sort the data based on the ANALYSIS_ID column.

SpeciesSort <- spoorData_Raw[order(spoorData_Raw$ANALYSIS_ID),]

#Create the crucial "Distance_Between" column.

SpeciesSort[1,"Distance_Between"] <- NA

for (i in 2:nrow(SpeciesSort))
{
	j <- i-1
	ifelse(SpeciesSort[i,"ANALYSIS_ID"] == SpeciesSort[j,"ANALYSIS_ID"],
	SpeciesSort[i,"Distance_Between"] <- SpeciesSort[i,"Distance.megatransect"] - SpeciesSort[j,"Distance.megatransect"], "FALSE")
}

#Examine if required.
#head(SpeciesSort,10)

#Calculate the total distance driven in each Section.
Transect_Length <- SpeciesSort[SpeciesSort$Species == "Stop",c("Section", "Km_reading", "Species")]
Survey_Length_Section <- aggregate(Transect_Length$Km_reading, by=list(Section=Transect_Length$Section), FUN=sum)
colnames(Survey_Length_Section) <- c("Section", "Length")
Survey_Length_Section$Section.lower <- as.factor(tolower(gsub('\\s+', '.', paste(Survey_Length_Section$Section))))

#Omit rows where the Species is either "Start" or "Stop". These are no longer needed.
SpeciesSort <- SpeciesSort[!(SpeciesSort$Species == "Start"),]
SpeciesSort <- SpeciesSort[!(SpeciesSort$Species == "Stop"),]

#Extract the columns we will need and place into a new data.frame.
SpoorData <- SpeciesSort[,c("ANALYSIS_ID", "Location", "Section", "Species", "Distance_Between")]

#Tidy up.
```

```
rm(list=c("spoorData_Raw", "Transect_Length"))

# A few options for inspecting the data. Uncomment if required.
#head(SpoorData,10)                          #Show top 10 records
#tail(SpoorData,10)                   #Show last 10 records
#dim(SpoorData)                              #Display number of rows and columns
#names(SpoorData)                     #Display column (field) names.


################################################################################################
############ Create a summary of the data to be called by subsequent functions ##################
################################################################################################


Group_Summary <- aggregate(SpoorData$ANALYSIS_ID, by=list(ANALYSIS_ID=SpoorData$ANALYSIS_ID), FUN=length)
colnames(Group_Summary) <- c("ANALYSIS_ID", "Records")

# Given that the first record in each set contains NA. Any group with <3 records cannot be analysed. In practical terms,
# we recommend a minimum sample size of n >= 10, since our analysis has shown that this is the point where CV stabilises.

Group_Summary_Trimmed <- subset(Group_Summary, Group_Summary$Records >= 10)

number_Groups <- nrow(Group_Summary)
number_Groups_Trimmed <- nrow(Group_Summary_Trimmed)

Dist_Between_Stats <- data.frame(matrix(NA, nrow = number_Groups, ncol = 8))
colnames(Dist_Between_Stats) <- c("ANALYSIS_ID", "track.incidences", "num.dist.between", "dist.between.mean",
                                                    "dist.between.SD", "dist.between.CV", "dist.between.SE", "dist.between.RSE")

Dist_Between_Stats$ANALYSIS_ID <- Group_Summary$ANALYSIS_ID
Dist_Between_Stats$track.incidences <- Group_Summary$Records
Dist_Between_Stats$num.dist.between <- Group_Summary$Records -1
Dist_Between_Stats$dist.between.mean <- tapply(SpoorData$Distance_Between, SpoorData$ANALYSIS_ID, mean, na.rm=TRUE)
```

```
Dist_Between_Stats$dist.between.SD <- tapply(SpoorData$Distance_Between, SpoorData$ANALYSIS_ID, sd, na.rm=TRUE)
Dist_Between_Stats$dist.between.CV <- Dist_Between_Stats$dist.between.SD / Dist_Between_Stats$dist.between.mean
Dist_Between_Stats$dist.between.SE <- Dist_Between_Stats$dist.between.SD / sqrt(Dist_Between_Stats$num.dist.between)
Dist_Between_Stats$dist.between.RSE <- (Dist_Between_Stats$dist.between.SE * 100) / Dist_Between_Stats$dist.between.mean


##################################################################################################
############ Do the bootstrapping ###################################################################
##################################################################################################


#Setup functions to use in bootstrap function.

boot_Combo <- function(data, i) {
         #Configure our required statistics
   mean.dat      <- mean(data[i])
   sd.dat        <- sd(data[i])
   CoVa.dat      <- sd(data[i]) / mean(data[i])
   SE.dat        <- sd(data[i]) / (sqrt(length(data[i])))
   RSE.dat       <- ((sd(data[i]) / sqrt(length(data[i]))) * 100) / mean(data[i])
   #And put them altogether.
   out <- cbind(mean.dat, sd.dat, CoVa.dat, SE.dat, RSE.dat)
   return(out)
}


#Loop through the different ANALYSIS_IDs and create separate data frames for each output. These will be combined later.
#We recommend using 20,000 bootstrap replicates, however, if you need to change it, it is set in the term below (R=20000)

for (i in 1:number_Groups_Trimmed)
{
   rawdata <- na.omit(subset(SpoorData, SpoorData$ANALYSIS_ID == Group_Summary_Trimmed[i,1]))
   rawdata <- rawdata$Distance_Between
```

```
	assign(paste(Group_Summary_Trimmed[i,1], sep=''), boot(rawdata, boot_Combo, R=20000, stype="i"), envir=.GlobalEnv)
}
beep(sound=7) #On our MacBook, 3500 samples with R=10,000 takes about 7 minutes to run.


##############################################################################################
############ Summarise the bootstrapping results ##################################################
##############################################################################################


#Create an empty data frame to store the results.
Results.all <- data.frame(matrix(NA, nrow = number_Groups_Trimmed, ncol = 21))
colnames(Results.all) <- c("ANALYSIS_ID", "track.incidences", "num.dist.between", "Species", "Species.corrected",
                                                "dist.between.mean", "dist.between.SD", "dist.between.CV", "dist.between.SE", "dist.between.RSE",
                                                "Boot.dist.between.SE", "Boot.dist.between.RSE", "Boot.dist.between.CI.Low",
                                                "Boot.dist.between.CI.High", "Boot.dist.between.PCL",
                                                "Est.Track.Density.Mean", "Est.Track.Density.Low", "Est.Track.Density.High",
                                                "Est.Animal.Density.Mean", "Est.Animal.Density.Low", "Est.Animal.Density.High")
Results.all$ANALYSIS_ID <- Group_Summary_Trimmed$ANALYSIS_ID


#Populate the newly created data frame, calculating the CIs as we go.
for (i in 1:number_Groups_Trimmed) {
	#Fill in the results for the non-bootstrapped results.
	Results.all[i,"num.dist.between"] <- length(eval(parse(text=paste(Results.all[i,1], "data", sep="$"))))
	Results.all[i,"track.incidences"] <- Results.all[i,"num.dist.between"] +1
	aaa <- eval(parse(text=paste(Results.all[i,1], "t0", sep="$"))) #Temporary container
	Results.all[i,"dist.between.mean"] <- c(aaa[1,1])
	Results.all[i,"dist.between.SD"] <- c(aaa[1,2])
	Results.all[i,"dist.between.CV"] <- c(aaa[1,3])
	Results.all[i,"dist.between.SE"] <- c(aaa[1,4])
	Results.all[i,"dist.between.RSE"] <- c(aaa[1,5])
	########
```

```
	#Insert values of each bootstrapped statistic.
	bbb <- eval(parse(text=paste(Results.all[i,1], "t", sep="$"))) #Temporary container
	Results.all[i,"Boot.dist.between.SE"] <- sd(bbb[,1]) #SD of the distribution of the means, i.e. the SE.
	Results.all[i,"Boot.dist.between.RSE"] <- (sd(bbb[,1]) * 100) / Results.all[i,"dist.between.mean"]
	########
	#Insert the output from boot.ci()
	boot.out <- eval(parse(text=Results.all[i,1]))
	ci.out <- boot.ci(boot.out, conf=0.95, type="bca")
	Results.all[i,"Boot.dist.between.CI.Low"] <- ci.out$bca[1,4]
	Results.all[i,"Boot.dist.between.CI.High"] <- ci.out$bca[1,5]
	########
	#Calculate PCL
	Results.all[i,"Boot.dist.between.PCL"] <-
			((Results.all[i,"Boot.dist.between.CI.High"] - Results.all[i,"Boot.dist.between.CI.Low"]) / 2) *
			(100 / Results.all[i,"dist.between.mean"])
}

#Tidy up.
rm(list=(Group_Summary_Trimmed$ANALYSIS_ID))


#############################################################################################
############ Calculating carnivore density from bootstrapped results ############################
#############################################################################################

#The command tolower() is included, since even the cleanest data sets can sometimes have names that
#utilise different cases. We have also found that it is not uncommon for some species names to include
#a trailing white space, which is replaced by a period in our code. This additional period is not a
#problem for the ANALYSIS_ID as it is simply required to be unique, but here the species names must
#match those that are pre-defined. We use agrep() to fuzzy match the species name to the allowed list.

Results.all[,"Species"] <- as.factor(tolower(sapply(strsplit(Results.all$ANALYSIS_ID,"_"), `[`, 1)))
```

```
#The following factors are taken from Table 1 in the main paper.
Density.factors                    <- data.frame(matrix(NA, nrow = 6, ncol = 2))
colnames(Density.factors)    <- c("A", "B")
rownames(Density.factors)  <- c(                    "brown.hyaena",                "cheetah",        "leopard",        "lion",
    "spotted.hyaena","wild.dog")
Density.factors$A                  <- as.numeric(   c("103.707",            "158.455",        "113.130",        "193.863",        "133.975",
    "820.020"))
Density.factors$B                  <- as.numeric(   c("-0.995",                    "-0.987",          "-1.034",          "-1.005",          "-1.006",
    "-1.168"))


#We use the values of CI.Low and CI.High to generate Track.Density.Low and Track.Density.High, using
#the factors stored in our Density.factors table (also see Table 1, in the main paper). These are then
#converted to actual carnivore density using the standard factor devised by Winterback et al (2016)
#of track density / 3.26 = carnivore density.

for (i in 1:number_Groups_Trimmed) {
    Results.all[i,"Species.corrected"] <- agrep(as.factor(Results.all[i,"Species"]),
            c("lion", "leopard", "brown.hyaena", "spotted.hyaena", "wild.dog", "cheetah"),
            max = 2, ignore.case = TRUE, value = TRUE)[1]

    Factor_A <- Density.factors[as.character(Results.all[i,"Species.corrected"]),"A"]
    Factor_B <- Density.factors[as.character(Results.all[i,"Species.corrected"]),"B"]
    Value_x <- Results.all[i,"Boot.dist.between.CI.Low"]
    Results.all[i,"Est.Track.Density.High"]            <- Factor_A * (Value_x^Factor_B)
    Results.all[i,"Est.Animal.Density.High"]   <- Results.all[i,"Est.Track.Density.High"] / 3.26

    Value_x <- Results.all[i,"Boot.dist.between.CI.High"]
    Results.all[i,"Est.Track.Density.Low"]      <- Factor_A * (Value_x^Factor_B)
    Results.all[i,"Est.Animal.Density.Low"] <- Results.all[i,"Est.Track.Density.Low"] / 3.26

    Value_x <- Results.all[i,"dist.between.mean"]
    Results.all[i,"Est.Track.Density.Mean"]   <- Factor_A * (Value_x^Factor_B)
```

```
        Results.all[i,"Est.Animal.Density.Mean"] <- Results.all[i,"Est.Track.Density.Mean"] / 3.26
    }

    #head(Results.all)

    #Tidy up.
    rm(list=c("Value_x", "Factor_A", "Factor_B", "aaa", "bbb", "rawdata", "ci.out"))


    ################################################################################################
    ############ Calculating carnivore density from basic data and comparing to bootstrap ###########
    ################################################################################################


    Animals_Total_BySection <- aggregate(SpeciesSort[,"Animals.Total"], by=list(ANALYSIS_ID=SpeciesSort[,"ANALYSIS_ID"]), FUN=sum, na.rm = TRUE)
    colnames(Animals_Total_BySection) <- c("ANALYSIS_ID", "Animals_Total")

    Animals_Total_BySection[,"Species"] <- as.factor(tolower(sapply(strsplit(Animals_Total_BySection$ANALYSIS_ID,"_"), `[`, 1)))
    Animals_Total_BySection[,"Section.lower"] <- as.factor(tolower(sapply(strsplit(Animals_Total_BySection$ANALYSIS_ID,"_"), `[`, 3)))

    for (i in 1:nrow(Animals_Total_BySection)) {
        Animals_Total_BySection[i,"Species.corrected"] <- agrep(as.factor(Animals_Total_BySection[i,"Species"]), c("lion", "leopard", "brown.hyaena", "spotted.hyaena", "wild.dog", "cheetah"), max = 2, ignore.case = TRUE, value = TRUE)[1]
    }

    ccc <- merge(Group_Summary, Animals_Total_BySection, by.x="ANALYSIS_ID", by.y="ANALYSIS_ID", all=TRUE) #Temporary container
    ddd <- merge(ccc, Survey_Length_Section, by.x="Section.lower", by.y="Section.lower", all=TRUE) #Temporary container

    #Select required columns and tidy up.
    Calculated_Density <- ddd[,c("ANALYSIS_ID", "Section", "Length", "Species.corrected", "Records", "Animals_Total")]
    Calculated_Density[,"Survey.Track.Density"] <- round ((Calculated_Density[,"Animals_Total"] * 100) / Calculated_Density[,"Length"], 2)
    Calculated_Density[,"Survey.Animal.Density"] <- round ((Calculated_Density[,"Survey.Track.Density"] / 3.26), 2)
    Calculated_Density[,"num.dist.between"] <- Calculated_Density[,"Records"] -1
```

```
    Key.results.combined <- merge(Calculated_Density, Results.all, by.x="ANALYSIS_ID", by.y="ANALYSIS_ID", all=TRUE)


    #head(Key.results.combined, 8)
    #names(Key.results.combined)

    for (i in 1:nrow(Key.results.combined)) {
    Key.results.combined[i,"Survey.Mean.between.CI"]           <-           ifelse(Key.results.combined[i,"Survey.Animal.Density"]          <
Key.results.combined[i,"Est.Animal.Density.High"] &&
                                                        Key.results.combined[i,"Survey.Animal.Density"]                       >
Key.results.combined[i,"Est.Animal.Density.Low"], "Y", "N")
    }

    #Preserve just the columns required for the final output.
    Final.Results <- Key.results.combined[,c(
        "ANALYSIS_ID", "Section",      "Length", "Species.corrected.x", "Records", "num.dist.between.x", "Animals_Total",
        "dist.between.mean", "dist.between.SD",      "dist.between.CV", "dist.between.SE", "dist.between.RSE",
        "Boot.dist.between.SE","Boot.dist.between.RSE", "Boot.dist.between.CI.Low", "Boot.dist.between.CI.High", "Boot.dist.between.PCL",
        "Survey.Track.Density", "Est.Track.Density.Mean", "Est.Track.Density.Low", "Est.Track.Density.High",
        "Survey.Animal.Density", "Est.Animal.Density.Mean", "Est.Animal.Density.Low", "Est.Animal.Density.High", "Survey.Mean.between.CI"
        )]

    #Change some column names
    colnames(Final.Results) <- c(
        "ANALYSIS_ID", "Section",      "Transect.Length", "Species.corrected", "track.incidences", "num.dist.between", "Animals.Total",
        "dist.between.mean", "dist.between.SD",      "dist.between.CV", "dist.between.SE", "dist.between.RSE",
        "Boot.dist.between.SE","Boot.dist.between.RSE", "Boot.dist.between.CI.Low", "Boot.dist.between.CI.High", "Boot.dist.between.PCL",
        "Survey.Track.Density", "Est.Track.Density.Mean", "Est.Track.Density.Low", "Est.Track.Density.High",
        "Survey.Animal.Density", "Est.Animal.Density.Mean", "Est.Animal.Density.Low", "Est.Animal.Density.High", "Survey.Mean.between.CI"
        )
```

```
#Insert the various statistics calculated for dist.between data for the ANALYSIS_IDs with too few data to do bootstrapping.
#Can overwrite these entire columns since the calculations are the same.

Final.Results$dist.between.mean <- Dist_Between_Stats$dist.between.mean
Final.Results$dist.between.SD <- Dist_Between_Stats$dist.between.SD
Final.Results$dist.between.CV <- Dist_Between_Stats$dist.between.CV
Final.Results$dist.between.SE <- Dist_Between_Stats$dist.between.SE
Final.Results$dist.between.RSE <- Dist_Between_Stats$dist.between.RSE

#Add "Small Sample mean, min, max" columns that will only have values for track incidences of < 10.
#This is included for instances where spoor samples were below 10 incidences, to provide a more conservative estimate
#due to the lower confidence and variability associated with small sample sizes.

Final.Results$Small.Sample.Min[Final.Results$track.incidences < 10] <- 0
Final.Results$Small.Sample.Mean[Final.Results$track.incidences < 10] <- Final.Results$Survey.Animal.Density[Final.Results$track.incidences < 10] / 2
Final.Results$Small.Sample.Max[Final.Results$track.incidences < 10] <- Final.Results$Survey.Animal.Density[Final.Results$track.incidences < 10]

#Delete everything we don't need.
rm(list=c("Animals_Total_BySection", "ccc", "ddd", "boot_Combo", "boot.out", "Calculated_Density", "Group_Summary", "Survey_Length_Section"))
rm(list=c("Group_Summary_Trimmed", "i", "j", "number_Groups", "number_Groups_Trimmed", "Results.all", "SpeciesSort", "SpoorData", "Dist_Between_Stats"))


#You may change the output filename in the following line if you wish.
write.csv(Final.Results, file="Bootstrap_Results_Test_Data.csv")


rm(list=c("Density.factors", "Key.results.combined"))


##################################################################################################
############ End of Script ######################################################################
##################################################################################################
```

3.11 Appendix 4 Track Survey Field Data Sheet

Track Survey *(vehicle)*

Location:

Section:

Date:

Transect:

Total km:

Tracker(s):

Persons:

Substrate:

First data record (time + location only) **is when you start with the transect. At this point you put the km reading at zero. Last data record** (time, km, location) **is when you finish the transect or when you have to terminate the activity for some reason.**

unique ID **e.g. CT001 = first cheetah tr track in survey**

Time	km reading	GPS Location		Track ID	Species	No. of animals				Direction		Age	Comments
		latitude	longitude			total	male	female	juv.	from	to	of track	
	0.0				Start								START of transect
													Stop of transect

Further comments:

CHAPTER 4. CALIBRATION OF CALL-UP SURVEYS FOR LIONS IN THE OKAVANGO DELTA

Christiaan W. Winterbach, Hanlie E. K. Winterbach and Michael J. Somers

4.1 Abstract

Carnivore surveys are important for conservation management. Call up surveys, luring animals in by broadcasting prey in distress and sounds of spotted hyaenas *Crocuta crocuta* and lions *Panthera leo*, have the potential to survey lions, spotted hyaenas, brown hyaenas *Parahyaena brunnea* and jackals (*Canis adustus, Canis mesomelas*) in a short time on a landscape level. Calibration of response is required to estimate density from calling station data. Environmental factors, aspects of carnivore biology and ecology, and survey parameters can influence carnivore response. Few of the underlying assumptions for call up surveys have been investigated and a linear relationship between lion abundance and abundance indices from call up surveys has not been shown.

We calculated reference densities of lions using long term home ranges in the Okavango Delta, Botswana, and calculated response probabilities from the proportion of known lions responding after 30, 60 and 90 minutes of playbacks. We estimated density with published calibrations, calibrations from this study and Generalized Linear Modelling. We compared density estimates with the mean relative bias and Coefficient of Variation of bias.

Over a period of 10 years, the reference densities of known lions declined from 7.99 to 2.33 lions/100 km^2. Mean number of lions per calling station correlated with reference density ($P < 0.05$) but The CV of lions per calling station was high (>200%). Individual identification showed that 24% of the lions responded more than once.

Response probability did not differ by sex and age among surveys when calling extended to 90 minutes (X^2 tests). Therefore, we calculated response probabilities for four surveys combined. The mean actual response distance increased less than anticipated, from 2.77 km (CV 14%) to 2.85 km (CV 14%) when extending calling from 60 to 90 minutes. Mean effective response distance was 1.84 km (CV 14%) for 90 minutes' call duration. We calculated density estimates with these calibrations for response distances and probabilities and with two Generalized Linear Models that were selected with corrected Akaike's Information Criterion.

Site specific calibrations performed better than published calibrations. Extending total time to 90 minutes reduced bias and coefficient of variation of bias for density estimates. The results of calling station surveys with high a coefficient of variation for lions/calling station are unlikely to detect

population trends and should be interpreted cautiously. Density estimates should be supporting with prey base and population structure data where available.

4.2 Introduction

Carnivore surveys provide data to plan, implement and evaluate conservation plans (Bauer *et al.* 2015; IUCN/SSC 2006; Lindsey & Davies-Mostert 2009; Wiesel 2015). There is a need for a method to survey carnivores in a short time on a landscape level to determine population size and trend (Groom, Funston & Mandisodza-Chikerema 2014; Katzner *et al.* 2011). Broadcasting sounds to lure target species to calling stations have the potential to fulfil this role for lions *Panthera leo*, spotted hyaenas *Crocuta crocuta*, brown hyaenas *Parahyaena brunnea* and jackals (*Canis adustus, Canis mesomelas*) (Mills, Juritz & Zucchini 2001; Ogutu & Dublin 1998; Thorn *et al.* 2010).

Call up surveys started with Smuts, Whyte and Dearlove (1977) describing the technique of calling lions over bait. Various calibrations have been done for call up surveys (Cozzi *et al.* 2013; Ferreira & Funston 2010; Ogutu & Dublin 1998). There are two key calibrations required to estimate carnivore density from calling station data: i) the probability that individuals respond, and ii) the response radius covered around the calling station. These provide a correction for non-response of individuals and the size of area sampled respectively, in order to calculate density. The differences in call duration, response distance and response probability among studies highlight the need for site specific calibration and an understanding of factors influencing response.

Factors that influence carnivore response can bias results (Cozzi *et al.* 2013; Kiffner *et al.* 2008; Ogutu & Dublin 1998) and include environmental factors, aspects of carnivore biology and ecology, and survey parameters. Thermal inversion and wind speed predict the calling distance and area covered by vocalizations of African elephants *Loxodonta africana* and lions (Garstang *et al.* 1995; Larom *et al.* 1997). The audible range of elephant vocalizations can extend 5-fold due to thermal inversion. Thus, thermal inversion can influence that sound travel during call up surveys (Ferreira & Funston 2010; Ferreira & Funston 2016) and needs to be investigated (Ogutu, Bhola & Reid 2005).

Response probability of lion and spotted hyaena declines with distance from calling station (Brink, Smith & Skinner 2013; Cozzi *et al.* 2013; Ferreira & Funston 2010; Kiffner *et al.* 2008; Mills, Juritz & Zucchini 2001; Ogutu & Dublin 1998; Omoya *et al.* 2013; Whitman 2006). Therefore, the response distance selected will influence the proportion of animals responding.

Call up surveys are a form of Distance Sampling using point transects (Omoya *et al*. 2013). Point transects are frequently used for bird surveys (Bart *et al*. 2004; Buckland *et al*. 1993) and the method has been expanded to include bait and lures to attract animals to detection devices (Buckland *et al*.

2006; Omoya *et al*. 2013). Omoya *et al*. (2013) applied the lure count analysis of Distance Sampling (Buckland *et al*. 2006), to calculate the effective response radius for lions. This is the distance at which the number of animals beyond the radius that respond is equal to the number closer to the lure that do not respond.

Age, sex and presence of cubs may also play a role in lion response (Brink, Smith & Skinner 2013; Ferreira & Funston 2010; Whitman 2006) although Ogutu and Dublin (1998) found that response probability is the same regardless of sex and age. Response probability of lions decreased rapidly when exposed multiple times to call ups in the short term (Belant *et al*. 2017; Belant *et al*. 2016; Ferreira & Funston 2010). This reduce the risk that the same lions will respond to more than one call up in a survey (Ferreira & Funston 2010), but it may be problematic for survey designs with temporal replication.

The type of call (prey animal in distress, hyaena sounds, lion sounds and roars) impact on response speed, distance and probability of lions and spotted hyaenas (Cozzi *et al.* 2013; Mills, Juritz & Zucchini 2001). Lions are unlikely to respond when feeding on a carcass and response is more uniform when migratory wildebeest are absent in the Mara (Ogutu & Dublin 1998).

These factors contribute to the heterogeneity in calibration results among studies. Ogutu, Bhola and Reid (2005) highlighted the need to collect data to test how well density estimates using response calibrations reflect true densities. Carnivore studies using track indices demonstrated a linear relationship between density indices and reference density (Balme, Hunter & Slotow 2009; Funston *et al.* 2010; Stander 1998) but this has not yet been demonstrated for call up indices.

The following assumptions apply to call up surveys:

1. There is a monotonic linear relationship between the number of lions per calling station and true lion density (Thorn *et al.* 2010).
2. Animals are not counted twice (Ferreira & Funston 2010; Ferreira & Funston 2016; Mills, Juritz & Zucchini 2001), although double counting is a possibility (Ferreira & Funston 2010).
3. Although not explicitly stated by any of these studies, the assumption is that response probability and response distance remain the same for surveys repeated over time within a study area.
4. Animals are evenly distributed around the calling stations (Cozzi *et al.* 2013; Graf *et al.* 2009; Kiffner *et al.* 2008), although Mills, Juritz and Zucchini (2001) made provision for spatial heterogeneity.
5. Response probability and response distance remain the same among habitats within the survey area (Cozzi *et al.* 2013; Ferreira & Funston 2016; Mills, Juritz & Zucchini 2001).
6. All lured animals are detected (Kiffner *et al.* 2008).
7. Minimal influence of diurnal and inter-diurnal variation in atmospheric conditions on the response range (Ferreira & Funston 2010; Ogutu, Bhola & Reid 2005).

8. Call duration is long enough for animals within the auditory range to respond but too short for animals from outside to enter the response area and respond (Cozzi *et al.* 2013).

These assumptions have not been tested. We tested the main assumption that there is a linear relation between number of lions per calling station and reference density, and quantified the number of lions counted more than once.

Call up surveys on land scape level were completed in northern Botswana between 1998 and 2000. Stratified sampling covered 18150 km^2 with up to 254 calling stations per survey (Winterbach *et al*. 2002). Only one calibration to estimate lion density was available at the time (Ogutu & Dublin 1998). There are several more recent calibrations (Brink *et al*. 2013; Cozzi *et al*. 2013; Ferreira & Funston 2010; Omoya *et al*. 2013). None of these calibrations have been tested in areas with known lion densities.

We used five call up surveys conducted between 1996 and 2007 in our long-term study area with a known lion population. Reference densities served as independent data to test the published calibrations and our own calibrations for call up surveys. Our results have implications for the planning, implementation and interpretation of call up surveys for lions, and potentially for other audio-acute large carnivores.

4.3 Methods

4.3.1 Study area

The Okavango Delta in north-western Botswana is a unique wetland ecosystem, spread over almost 18 000 km^2 of the semi-arid Kalahari Desert, forming one of the largest inland deltas in the world (Roodt 1998). The Delta is an intricately balanced, highly dynamic and sensitive ecosystem that supports a wide variety of life forms. The herbivore density and distribution are affected by the annual rainfall during the wet season and the annual floods during the dry season.

NG/29 and NG/30 are tourism concessions located in the south-western part of the Okavango Delta (Figure 4-1). NG/29 comprises a total area of 1,848 km^2, of which almost 60% consists of a sandveld tongue, covered by deep sandy soils, sparse grass cover and a mosaic of scrub and tall "cathedral" *Colophospermum mopane*, *Terminalia sericea* and various *Acacia* species. The remainder of the area consists of seasonally inundated areas (approx. 30%) and intermittently flooded areas (approx. 10%). NG/30 comprises a total area of 951 km^2, of which more than 70% are seasonally inundated or intermittently flooded areas. A very small area of permanent swamp occurs in the northern-most part of NG/30.

Our study area falls into the Seasonal Floodplain West stratum (Winterbach *et al.* 2015) consisting of seasonal floodplains in the western part of the Okavango Delta (Figure 4-2). The study area for the calibration of calling stations consisted of 463 km² in 1998 and 730.99 km² (1999 to 2007). We merged the long-term home ranges of our five study prides (95% MCP home ranges of the individual prides) to determine the boundary of the study area. The northern section (156.03 km²) was excluded due to seasonal flooding that limited access to conduct calling stations.

4.3.2 Call up surveys

Our original objective was to record and identify as many individual lions as possible with calling station surveys and obtain minimum counts of lions in the survey areas (Winterbach & Vrahimis 1995; Winterbach & Winterbach 1996). We selected locations of calling stations to be a maximum 5 km apart to ensure overlap between calling stations. Sampling effort is the number of calling stations per 100 km². The response area is a function of response distance (*response area* = π * *response distance*²).

The known population of lions was identified as part of a long-term study that commenced in 1997. In this study, we followed five prides to collect demographic data. We calculated reference density as the density of known lions present in the survey area at the time of each survey, using radio tracking to confirm presence or absence in the survey area. Lions that were not part of focal prides but present in the survey area were excluded from all calculations.

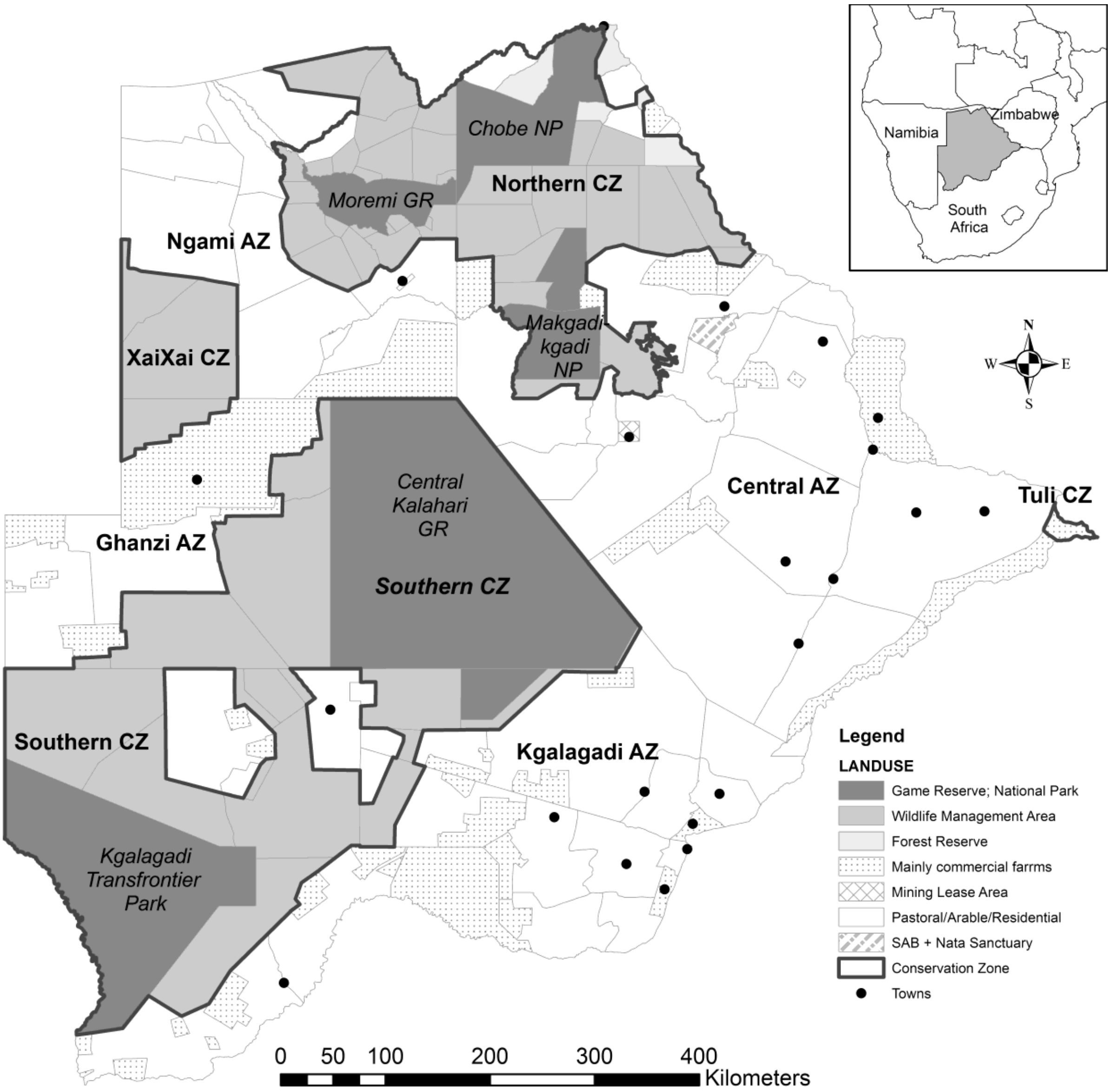

Figure 4-1 Location of study area in Botswana.

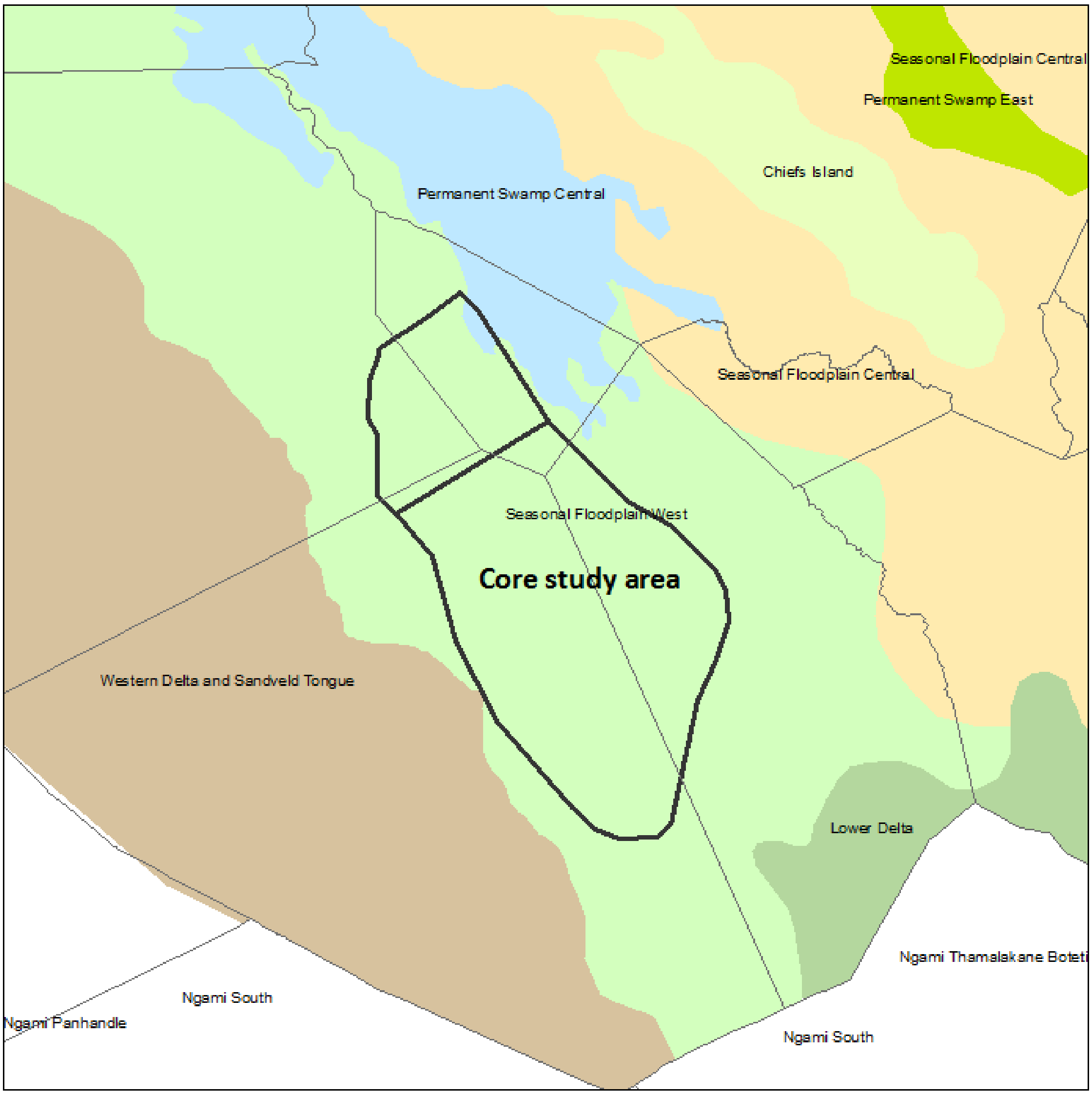

Figure 4-2 Study area for call up surveys with known lions in NG/29 and NG/30 conducted in 1996, 1998, 1999, 2000 and 2007.

We took identification photos to prevent double counting of individuals. We used ungulate bait at each calling station, collected under the relevant permit from the Department of Wildlife and National Parks in Botswana. Baiting was done to ensure enough time to photograph responding individuals for whisker spot identification, and also to reduce risk of animals not responding on subsequent calling stations because they experienced no reward. In 2007, we used a mixture of blood and glycerin splattered around the site to provide smell for animals responding to calls.

The technique of calling lions over bait is based on the method described by Smuts, Whyte & Dearlove (1977), and guidelines provided by the DWNP (Vandewalle 1995). A public address system with two 10-inch horn speakers was used for broadcasting. We played calls from a vehicle parked 15 to 20 m from the bait. Recordings were played for 5 minutes followed by a 5-minute break. The sound of a distressed buffalo calf was played twice, followed by spotted hyaenas harassing lions, an inter-clan fight between spotted hyaenas, hyaenas competing for a kill and lions competing for a kill. No recordings of lions roaring were used during the survey. Maximum call duration was 120 minutes in 1998 and 90 minutes from 1999 onwards. Calling stations were not operated when it rained or if there was a strong wind (> 2 Beaufort scale). Between two and three calling stations could be completed per night, starting after sunset and commencing with a new calling station not later than midnight.

A spotlight, in combination with a strong flashlight, was used to photograph responding lions with a 300 mm f2.8 lens or a 400 mm f5.6 lens. A film camera with ISO 800 print film was used for the 1996 to 2000 surveys and a digital camera for the 2007 survey. Data recorded at each station included the date, coordinates, weather conditions, phase of the moon, time each calling station began and ended, the arrival time of lions, and the total number of lions with their sex and age.

Lions that visited calling stations were photographed and recorded as male or female, and adult (>4 years), sub-adult (2-4 years) or cub (<2 years). The ages of lions were estimated following the guidelines from Smuts (1982). Notes and drawings were made on data sheets showing whisker spots, scars and other distinctive features and references to match notes with photos. The pattern of whisker spots, particularly those found above the top whisker row and the relative placement of these spots in reference to the top whisker row, is the most reliable identification characteristic in lions. This pattern is unique to every individual, and does not alter during an individual's life (Pennycuick & Rudnai 1970).

We tested for correlation between mean number of lions per calling station (CS) and reference densities.

4.3.3 Calibration

We calculated response probability as the proportion of known lions that responded to calling stations after 30, 60 or 90 minutes for the 1998 to 2007 surveys. This was summarized per age class (adult, sub-adult and cub) and sex (male or female), adult plus sub-adult combined and all lions combined. We used χ^2 tests with Bonferroni confidence intervals (Byers, Steinhorst & Krausman 1984) to test for differences in response proportions between sex and age groups and years.

We combined the data from the four surveys (1998, 1999, 2000 and 2007) to estimate mean response/calling station and 95% confidence interval (CI) with the bootstrap procedure in SPSS for 30, 60 and 90 minutes call durations). The results included the mean number of lions responding per calling station, standard deviation, variance and the 95% confidence intervals. Multiplying the estimated mean number of lions responded per calling station and the 95% CI limits with the total number of calling stations for each survey, provided the estimate of the number of lions that responded with a 95% CI for each survey and all surveys combined. We used these estimates to calculate response probabilities (bootstrap) with CI.

We used "response distance" as a general reference to how far lions respond to calling stations. Estimated response distance referred to response distances estimated from regressions of response speed and response time. To estimate response distance, we used SPSS to fit linear, logarithmic, power and exponential regression curves to response speed and response time (sourced from the lion response calibration data of Cozzi *et al.* (2013)). From these equations, we calculated estimated response distance (*response distance linear, response distance logarithmic, response distance power* and *response distance exponential*) for calling durations of 30, 60 or 90 minutes. We also calculated the actual response distance (*actual response distance*) from reference density, response probability and lions/calling station.

We followed the lure count analysis, a variant of Distance Sampling (Buckland *et al*. 2006), using the effective response distance: the distance where the number of individuals not responding within the response distance equals the number of individuals responding from further away, thus requiring no further correction for non-response (Buckland *et al.* 2006; Omoya *et al.* 2013). We calculated the effective response distance for each survey from reference density and number of lions/ calling station for 30, 60 and 90 minutes of calling: *effective response distance = SQRT ((Lions/calling station*100) / lions/100 km² / 3.1415)*.

4.3.4 GLM to estimate density

Count data is described by the Poisson distribution (Quinn & Keough 2002) and the call up survey data was not log-transformed (O'Hara & Kotze 2010). Therefore, we carried out Generalized Linear Modelling (GLM) (Quinn & Keough 2002), with lion reference density as the dependent variable and model parameters Lions per calling station (*Lion_CS*), mean response probability (*Mean_response*), mean response time (*Mean_time*), mean actual response distance (*mean_actual_response_distance*), and response area (*response_area*) as predictors.

We used Wald Chi-Square to test parameters for removal from the model. Parameters with significance values less than 0.05 have some discernible effect and non-significant terms were removed stepwise from the model. Smaller values of sample size corrected Akaike Information Criteria (AIC_c) indicate better fit of a model based on the same dataset. Models with $\Delta_i < 2$ have substantial support. Models with $4 \leq \Delta_i \leq 7$ have considerably less support and models having $\Delta_i > 10$ have essentially no support (Burnham & Anderson 2004).

4.3.5 Testing calibrations

We tested published calibrations with our independent calling station data and reference densities. Only the 1996 data were available as independent data to test our own calibrations. We used bias and precision to evaluate the different models. We modified the % relative bias (Balme, Hunter & Slotow 2009) as the absolute value of [(D- d)/d] * 100 where D is estimated density and d is reference density. We compared the mean relative bias and variability (Coefficient of Variation (CV)) among the four surveys for the different calibrations.

4.4 Results

4.4.1 Call up surveys

The surveys in 1996, 1998, 1999, 2000 and 2007 consisted of between 16 and 31 calling stations with a survey effort of 3.01 – 5.99 calling stations/ 100 km² (Table 4-1). The reference densities of known lions declined from 7.99 to 2.33 lions/100 km² (Table 4-2). The CV of lions per calling station exceeded 200% (Table 4-2). The number of lions per calling station correlated significantly with reference density (Table 4-3) for all call durations. The highest R square was for a call duration of 90 minutes.

The known lion population was between 57 and 66 lions from 1998 to 2000, declining to 26 lions in 2007 (Table 4-4). The number of known lions responding after 30, 60 and 90 minutes are summarised in Table 4-4.

4.4.2 Calibration

4.4.2.1. Response time

The proportion of calling stations with response times within 0 – 30 minutes, 31 – 60 minutes and 61 – 90 minutes are shown in Fig. 4-3. The general pattern was that most lions responded within the first 30 minutes, less lions in 30 – 60 minutes and least after that. All responses were within the first 30 minutes in 1996, and 71% were during the first 30 minutes in 1998. No lions responded during the first 30 minutes in 2000. There were no responses after 60 minutes during three surveys (Table 4-4).

Table 4-1 Sampling effort during call up surveys in NG/29 and NG/30 between 1996 and 2007.

Year	Core area (km²)	Calling Stations (n)	Calling Stations /100 km²
1996	267	16	5.99
1998	463	25	5.40
1999	730	27	3.70
2000	730	22	3.01
2007	730	31	4.25

Table 4-2 Reference density of lions and the number of lions per calling station in NG/29 and NG/30 between 1996 and 2007 (mean ± SD (CV)) .

Year	Reference density lions/100 km²	Lions/CS 30 min	Lions/CS 60 min	Lions/CS 90 min
1996	15.73	2.00 ± 4.24 (212%)	2.00 ± 4.24 (212%)	2.00 ± 4.24 (212%)
1998	7.99	0.72 ± 2.17 (301%)	0.76 ± 2.17 (285%)	0.76 ± 2.17 (285%)
1999	7.26	0.32 ± 1.25 (389%)	0.36 ± 1.25 (351%)	0.57 ± 1.53 (267%)
2000	6.44	-	0.45 ± 1.92 (423%)	0.68 ± 2.15 (315%)
2007	2.33	0.32 ± 1.30 (403%)	0.35 ± 1.31 (368%)	0.35 ± 1.31 (368%)

Table 4-3 Correlation between reference density and lions per calling station with 30, 60 and 90 minutes call durations in NG/29 and NG/30 between 1996 and 2007 (n=5).

Call Duration	Equation	R Square	Significance
30 minutes	$y = 0.143 x - 0.465$	0.787	p = 0.045
60 minutes	$y = 0.134 x - 0.280$	0.867	p = 0.021
90 minutes	$y = 0.129 x - 0.151$	0.931	p = 0.008

Table 4-4 Known lion numbers in the study area and the numbers responding to calling stations after 30, 60 and 90 minutes during the surveys in 1996, 1998, 1999, 2000 and 2007. (AM = Adult Male; AF = Adult Female; SAM = Subadult Male; SAF = Subadult Female; A+SA = Adult plus Subadult).

YEAR	PRIDE	AM	AF	SAM	SAF	All CUBS	A+SA	TOTAL
1996	Known lions	4	17	6	15	13	42	55
1996	Responded 30 minutes	4	14	6	8	8	32	40
1996	Responded 60 minutes	4	14	6	8	8	32	40
1996	Responded 90 minutes	4	14	6	8	8	32	40
1998	Known lions	4	23	4	6	25	37	62
1998	Responded 30 minutes	1	11	4	2	12	18	30
1998	Responded 60 minutes	2	11	4	2	12	19	31
1998	Responded 90 minutes	2	11	4	2	12	19	31
1999	Known lions	11	29	7	6	13	53	66
1999	Responded 30 minutes	1	3	5	0	1	9	10
1999	Responded 60 minutes	2	3	5	0	1	10	11
1999	Responded 90 minutes	3	7	5	1	2	16	18
2000	Known lions	5	32	5	5	10	47	57
2000	Responded 30 minutes	0	0	0	0	0	0	0
2000	Responded 60 minutes	1	4	2	3	1	10	11
2000	Responded 90 minutes	1	7	3	4	4	15	19
2007	Known lions	5	12	0	0	9	17	26
2007	Responded 30 minutes	4	6	0	0	0	10	10
2007	Responded 60 minutes	4	7	0	0		11	11
2007	Responded 90 minutes	4	7	0	0	0	11	11

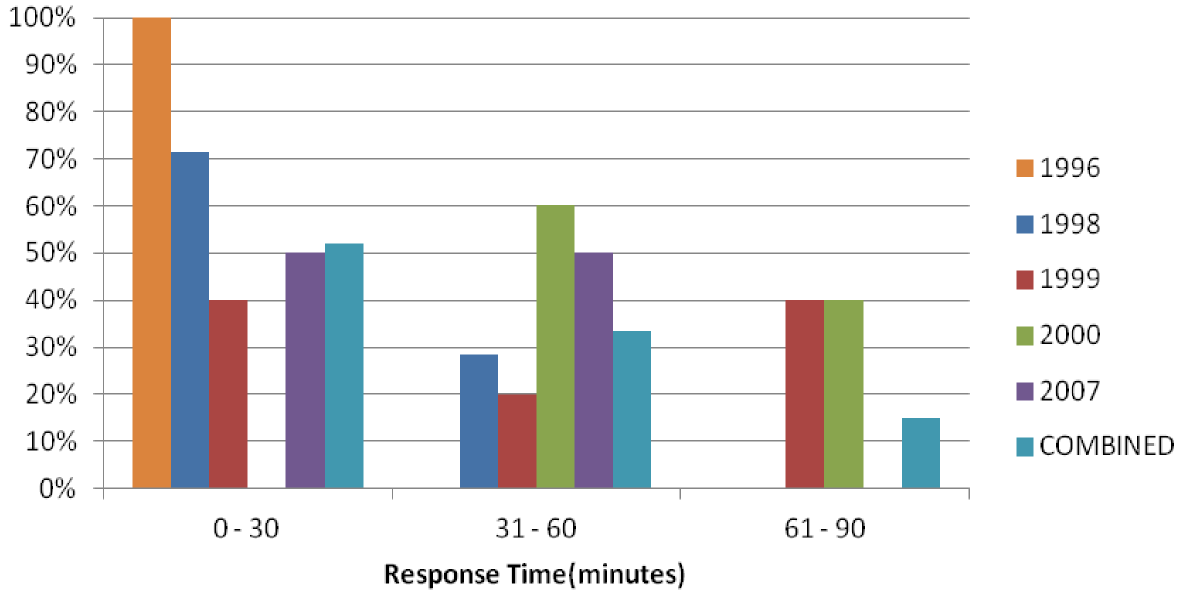

Figure 4-3 The proportion of calling stations with response times within 0 – 30 minutes, 31 – 60 minutes and 61 – 90 minutes for surveys conducted in NG/29 and NG/30 between 1996 and 2007.

4.4.2.2. Response probability

Probability of counting double

Of the 121 calling stations operated between 1996 and 2007, 10 (8%) attracted lions that were recorded at a previous calling station. Twenty-eight of 119 known lions (24%) responded more than once to calling stations. Fifty per cent of adult males (7 of 14 lions) and 15% of females (7 of 47) responded more than once. Cubs responded more than once on only three occasions.

Bonferroni confidence intervals to compare response probabilities

We calculated response probabilities (number of known lions responded / total known lions) per sex and age class, adults and sub-adults combined and for all lions combined per year and all years combined (see Table 4-4). χ^2 tests showed that the observed (known lions responded) and expected (known lions) values differed significantly in four comparisons with a 60 minutes call duration but not with a 90 minutes call duration (Table 4-5). We found significant differences with Bonferroni confidence intervals for a call duration of 60 minutes. During the 1999 survey significantly less subadult females responded (Table 4-6), and significantly less adult plus sub-adult (males and females) responded (Table 4-7). After 60 minutes of calling, significantly more lions (adults, sub-adults, cubs) responded during the 1998 survey while significantly less lions (adults, sub-adults, cubs) responded during the 1999 survey (Table 4-8). In contrast, no significant differences were found when calling extended to 90 minutes. Although the χ^2 test indicated a significant difference among AM, AF, SAM and SAF for the 1998, 1999, 2000 and 2007 surveys combined, the Bonferroni CI could not identify the differences (Table 4-9).

Response probability bootstrap

χ^2 tests showed that lion response with 90 minutes of calling did not differ per sex and age among surveys. We combined the data of the 1998, 1999, 2000 and 2007 surveys and calculated response probabilities with 95% confidence intervals for 30-, 60- and 90-minutes call duration, from the bootstrap estimates of the mean number lions per calling station, standard error and 95% confidence intervals (Table 4-10). The PCL and CV results provided further support for a call duration of 90 minutes: increased calling time resulted in lower CV and PCL (Table 4-10). The PCL and CV for adult plus sub-adult lions with 90 minutes of calling (Fig. 4-4) were the lowest for all sex and age groups and calling durations.

Table 4-5 Summary of X^2 tests for differences in lion response to calling stations in the Okavango Delta. Differences among surveys, sex and age classes were tested at p=0.05, and significant differences indicated by *.

Group	Duration	Comparison	X^2
1998	60	AM, AF, SAM, SAF	$X^2 = 2.28$, df = 3
1999	60	AM, AF, SAM, SAF	$X^2 = 12.50$, df = 3^*
2000	60	AM, AF, SAM, SAF	$X^2 = 5.51$, df = 3
2007	60	AM, AF, SAM, SAF	$X^2 = 0.26$, df = 3
98 - 2007	60	AM, AF, SAM, SAF	$X^2 = 7.85$, df = 3^*
1998	90	AM, AF, SAM, SAF	$X^2 = 2.28$, df = 3
1999	90	AM, AF, SAM, SAF	$X^2 = 4.69$, df = 3
2000	90	AM, AF, SAM, SAF	$X^2 = 6.09$, df = 3
2007	90	AM, AF, SAM, SAF	$X^2 = 0.26$, df = 3
98 - 2007	90	AM, AF, SAM, SAF	$X^2 = 6.03$, df = 3
98 - 2007	60	AM, AF, SAM, SAF, Cubs	$X^2 = 9.26$, df = 4
A+SA	60	1998,1999, 2000, 2007	$X^2 = 14.34$, df = 3^*
A+SA	90	1998,1999, 2000, 2007	$X^2 = 5.88$, df = 3
A+SA+CUBS	60	1998,1999, 2000, 2007	$X^2 = 15.49$, df = 3^*
A+SA+CUBS	90	1998,1999, 2000, 2007	$X^2 = 4.86$, df = 3
98 - 2007	60	A+SA vs CUBS	$X^2 = 0.86$, df = 1
98 - 2007	90	A+SA vs CUBS	$X^2 = 0.72$, df = 1

Table 4-6 Bonferroni simultaneous confidence intervals to compare lion response of adult males, adult females, sub-adult males and sub-adult females after 60 minutes of calling in the 1999 survey (k = 4, α = 0.05, Z= 2.4977).

60 minutes	Observed	Expected	DIFOE	SQDIFOE	CHI	Pio	Pi	Pi(1-Pi)/n	Bonferroni CI	Use Index	Significant
AM	2	2.08	0.075	0.01	0.00	0.207547	0.200000	0.126491	-0.1159 ≤ Pi ≤ 0.5159	0.96	0
AF	3	5.47	2.472	6.11	1.12	0.547170	0.300000	0.144914	-0.0620 ≤ Pi ≤ 0.6620	0.55	0
SAM	5	1.32	3.679	13.54	10.25	0.132075	0.500000	0.158114	0.1051 ≤ Pi ≤ 0.8949	3.79	0
SAF	0	1.13	1.132	1.28	1.13	0.113208	0.000000	0.000000	0.0000 ≤ Pi ≤ 0.0000	0.00	-
Total	10	10.00			12.50		1.000000				

Table 4-7 Bonferroni simultaneous confidence intervals to compare lion response of adults and sub-adults combined after 60 minutes of calling among the 1998, 1999, 2000 and 2007 surveys (k = 4, α = 0.05, Z= 2.4977).

60 minutes	Observed	Expected	DIFOE	SQDIFOE	CHI	Pio	Pi	Pi(1-Pi)/n	Bonferroni CI	Use Index	Significant
1998	19	12.01	6.987	48.82	4.06	0.240260	0.380000	0.068644	0.2085 ≤ Pi ≤ 0.5515	1.58	0
1999	10	17.21	7.208	51.95	3.02	0.344156	0.200000	0.056569	0.0587 ≤ Pi ≤ 0.3413	0.58	-
2000	10	15.26	5.260	27.66	1.81	0.305195	0.200000	0.056569	0.0587 ≤ Pi ≤ 0.3413	0.66	0
2007	11	5.52	5.481	30.04	5.44	0.110390	0.220000	0.058583	0.0737 ≤ Pi ≤ 0.3663	1.99	0
TOTAL	50	50.00			14.34		1.000000				

Table 4-8 Bonferroni simultaneous confidence intervals to compare lion response of adults, sub-adults and cubs combined after 60 minutes of calling among the 1998, 1999, 2000 and 2007 surveys (k = 4, α = 0.05, Z= 2.4977).

60 minutes	Observed	Expected	DIFOE	SQDIFOE	CHI	Pio	Pi	Pi(1-Pi)/n	Bonferroni CI	Use Index	Significant
1998	31	18.81	12.194	148.70	7.91	0.293839	0.484375	0.062469	0.3283 ≤ Pi ≤ 0.6404	1.65	+
1999	11	20.02	9.019	81.34	4.06	0.312796	0.171875	0.047159	0.0541 ≤ Pi ≤ 0.2897	0.55	-
2000	11	17.29	6.289	39.55	2.29	0.270142	0.171875	0.047159	0.0541 ≤ Pi ≤ 0.2897	0.64	0
2007	11	7.89	3.114	9.70	1.23	0.123223	0.171875	0.047159	0.0541 ≤ Pi ≤ 0.2897	1.39	0
TOTAL	64	64.00			15.49		1.000000				

Table 4-9 Bonferroni simultaneous confidence intervals to compare lion response of adult males, adult females, sub-adult males and sub-adult females after 60 minutes of calling in the 1998, 1999, 2000 and 2007 surveys combined (k = 4, α = 0.05, Z= 2.4977).

60 minutes	Observed	Expected	DIFOE	SQDIFOE	CHI	Pio	Pi	Pi(1-Pi)/n	Bonferroni CI	Use Index	Significant
AM	9	8.12	0.883	0.78	0.10	0.162338	0.180000	0.054332	0.0443 ≤ Pi ≤ 0.3157	1.11	0
AF	25	31.17	6.169	38.05	1.22	0.623377	0.500000	0.070711	0.3234 ≤ Pi ≤ 0.6766	0.80	0
SAM	11	5.19	5.805	33.70	6.49	0.103896	0.220000	0.058583	0.0737 ≤ Pi ≤ 0.3663	2.12	0
SAF	5	5.52	0.519	0.27	0.05	0.110390	0.100000	0.042426	-0.0060 ≤ Pi ≤ 0.2060	0.91	0
TOTAL	50	50.00			7.85		1.000000				

Table 4-10 Estimated mean number of lions (bootstrap) that responded to calling stations during the 1998, 1999, 2000 and 2007 surveys (all surveys combined) and proportion (%) that responded.

RESPONSE	AM	AF	SAM	SAF	CUB	A+SA	TOTAL
30 minutes	6.36 (1.06 - 13.78)	20.14 (4.24 - 42.40)	8.48 (1.06 - 20.14)	2.12 (0.00 - 6.36)	12.72 (0.00 - 38.16)	37.10 (11.66 - 69.96)	49.82 (12.72 - 97.52)
30 minutes %	25.44 (4.24 - 55.12)	20.98 (4.42 - 44.17)	53.00 (6.63 - 125.88)	12.47 (0.00 - 37.41)	22.32 (0.00 - 66.95)	24.09 (7.57 - 45.43)	23.61 (6.03 - 46.22)
30 minutes CV	48%	48%	61%	95%	87%	39%	43%
30 minutes PCL	100%	95%	113%	150%	150%	79%	85%
60 minutes	8.48 (3.18 - 16.96)	25.44 (7.42 - 48.76)	10.60 (2.12 - 22.26)	5.30 (0.00 - 13.78)	12.72 (0.00 - 38.16)	49.82 (20.14 - 85.86)	63.60 (23.32 - 117.66)
60 minutes %	33.92 (12.72 - 67.84)	26.50 (7.73 - 50.79)	66.25 (13.25 - 139.13)	31.18 (0.00 - 81.06)	22.32 (0.00 - 66.95)	32.35 (13.08 - 55.75)	30.14 (11.05 - 55.76)
60 minutes CV	41%	41%	52%	68%	87%	34%	37%
60 minutes PCL	81%	81%	95%	130%	150%	66%	74%
90 minutes	9.54 (4.24 - 18.02)	31.80 (11.66 - 58.30)	11.66 (3.18 - 23.32)	7.42 (1.06 - 14.84)	13.78 (1.06 - 39.22)	61.48 (28.62 - 99.64)	79.50 (34.98 - 133.56)
90 minutes %	38.16 (16.96 - 72.08)	33.13 (12.15 - 60.73)	72.88 (19.88 - 145.75)	43.65 (6.24 - 87.29)	24.18 (1.86 - 68.81)	39.92 (18.58 - 64.70)	37.68 (16.58 - 63.30)
90 minutes CV	38%	35%	47%	51%	81%	29%	31%
90 minutes PCL	72%	73%	86%	93%	138%	58%	62%

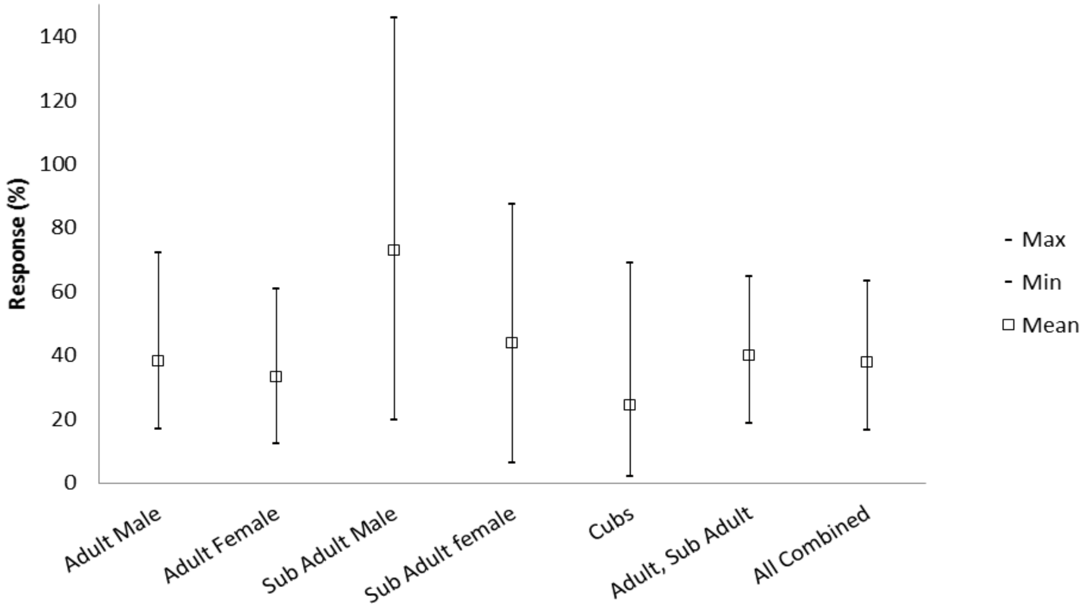

Figure 4-4 Percentage of lions responding per sex and age group after 90 minutes of calling for all surveys combined (1998, 1999, 2000 and 2007). Response percentage and 95% Confidence Interval calculated from bootstrap estimates of the mean number of lions per calling station with 95% Confidence Interval and known lion population.

4.4.2.3. Response distance

Estimate of response distance from response speed and response time

Lion response speed decreased with time to respond (Cozzi *et al*. 2013) as shown in Fig. 4-5. All four regression curves fitted to time (independent variable) and speed (dependent variable) were significant at $p < 0.05$. The exponential and power curves had the highest R square values (Table 4-11).

We used the equations (Table 4-11) to estimate response distance and response area at 30, 60, 90 minutes (Table 4-12). Extrapolation beyond 70 minutes exceeded the range of response times in the data set and is statistically incorrect (Sokal & Rohlf 1995). Estimated response distances from the linear and exponential regressions decreased between 60 and 90 minutes, while they should increase (Table 4-12). The estimate response distances with the logarithmic and power regressions increased from 60 to 90 minutes. Response distance estimates from the linear and exponential regressions were excluded from further analysis.

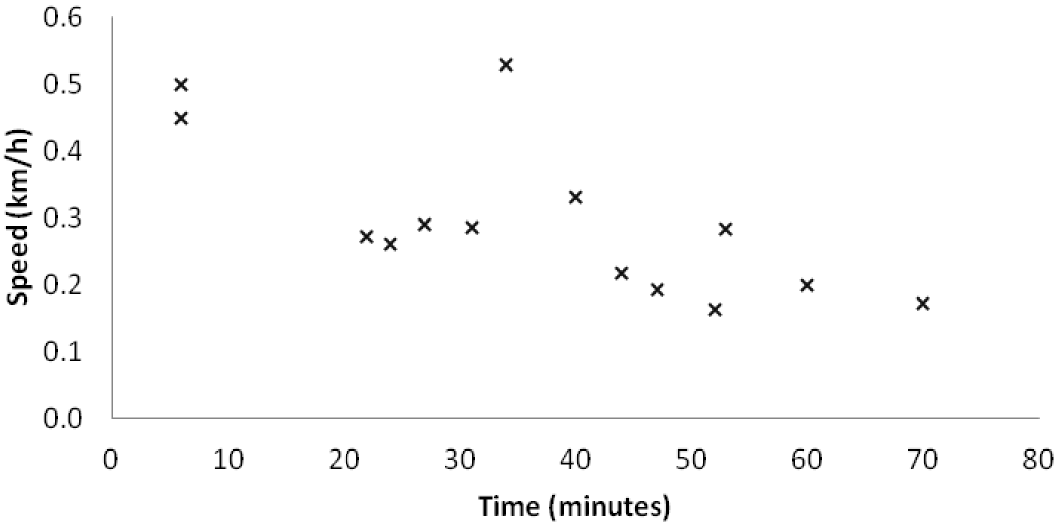

Figure 4-5 Lion response speed declined with response time in the Okavngo Delta. Data from Cozzi *et al.* (2013).

Table 4-11 Results of curves fitted to response time (independent) and response speed (dependent). Data from Cozzi *et al.* (2013).

Equation	Model Summary					Parameter Estimates	
	R Square	F	df1	df2	Sig.	Constant	b1
Linear	.508	12.369	1	12	p = 0.004	4.598	-0.045
Logarithmic	.521	13.054	1	12	p = 0.004	6.783	-1.123
Power	.536	13.868	1	12	p = 0.003	9.549	-0.364
Exponential	.572	16.058	1	12	p = 0.002	4.817	-0.015

Table 4-12 Estimated response speed, estimated response distance and response area after 30, 60 and 90 minutes of calling for the linear, logarithmic, power and exponential curves fitted to response data from Cozzi *et al.* (2013).

	Linear			Logarithmic			Power			Exponential		
	$y=-0.045x + 4.598$			$y = -1.123 \ln (x) + 6.783$			$y = 9.549x^{-0.364}$			$y = 4.817 e^{(-0.051 x)}$		
Time (x) (min)	Speed (y) (km/h)	Response distance (km)	Response area (km²)	Speed (*y*) (km/h)	Response distance (km)	Response area (km²)	Speed (*y*) (km/h)	Response distance (km)	Response area (km²)	Speed (*y*) (km/h)	Response distance (km)	Response area (km²)
30	3.248	1.62	8.29	2.96	1.48	6.90	2.77	1.38	6.02	3.07	1.54	7.41
60	1.898	1.90	11.32	2.19	2.19	15.00	2.15	2.15	14.54	1.96	1.96	12.05
90*	0.548	0.82	2.12	1.73	2.59	21.15	1.86	2.78	24.35	1.25	1.87	11.02

* Extrapolation beyond the 70 minutes response time of the data set used in the regression analysis.

Actual and effective response distance

The actual response distance calculated from reference density and response probability, differed among our surveys, ranging between 2.43 km and 3.25 km (Table 4-13). In 1998, 2000 and 2007 the calculated actual response distances within the survey remained the same regardless of call duration. In 1999 the actual response distance increased by 0.4 km between 30 and 90-minute call durations. The mean actual response distance was 2.77 km for a call duration of 60 minutes and increased to 2.85 km for 90 minutes. The effective response distances were lower than the actual response distances and covered smaller response areas (Table 4-13).

Table 4-13 Actual and effective response distances of lions to calling stations for the 1998, 1999, 2000 and 2007 calibration surveys in the Okavango Delta. Actual response distance calculated from lions/calling station, adult plus sub-adult response probability and reference density. Effective response distance calculated from lions/calling station and reference density.

Year	Actual Response Distance (km)			Effective Response Distance (km)		
	Calling 30 min	Calling 60 min	Calling 90 min	Calling 30 min	Calling 60 min	Calling 90 min
1998	2.43	2.43	2.43	1.69	1.74	1.74
1999	2.59	2.66	2.99	1.19	1.25	1.58
2000	-	3.25	3.25	-	1.50	1.84
2007	2.74	2.74	2.74	2.10	2.20	2.20
Mean ± SD	2.59 ± 0.16	2.77 ± 0.35	2.85 ± 0.35	1.66 ± 0.46	1.67 ± 0.41	1.84 ± 0.26
CV	6%	12%	12%	28%	24%	14%
Response Area (km²)	21.02	24.10	25.56	8.66	8.79	10.64

4.4.3. *GLM to estimate density*

Only one Generalized Linear Model for *reference density* had substantial support (Δ_i values ≤ 2) with a probability of 0.82 to be the correct model. One model had between substantial support and considerably less support ($4 \le \Delta_i \le 7$) (Table 4-14). Lions per calling station (*Lion_CS*) was the best predictor of adult plus sub-adult lion density (*reference density)* according to AIC_c criteria. Table 4-15 provides a summary of the parameter estimates of the Generalized Linear Models.

Table 4-14 Generalized Linear Models with lion reference density as dependent variable and model parameters lions per calling station (*Lion_CS*), mean response probability (*Mean_response*) and mean response time (*Mean_time*), mean actual response distance (*mean_actual_response_distance*), and response area (*response_area*) as predictors. Smaller values of small sample corrected Akaike Information Criteria (AIC_c) indicate better fit of model.

Description of model	AIC_c	Δ_i Values	ω_i
Model: *Lion_CS*	49.638	0.0	0.82
Model: Lion_CS, Mean_time	52.926	3.3	0.16
Model: Lion_CS, Mean_ actual_ response _distance, Mean_time	57.016	7.4	0.02
Model: Lion_CS, Mean_ actual_ response _distance, Response_area, Mean_time	60.093	10.5	0.00
Model: Lion_CS, Mean_response, Mean_actual_ response _distance, Response_area, Mean_time	71.075	21.4	0.00
Total			1

4.4.4. *Testing calibrations*

The Cozzi *et al.* (2013) calibration in the Okavango Delta performed the best of the published calibrations (Table 4-16) with a mean relative bias estimating reference density of 42% (6% - 102%) with a CV of 98%. The lure count analysis method (Omoya *et al.* 2013) was the second best with relative bias ranging between 18% and 100% (CV = 62%). The Kruger National Park calibration (Ferreira & Funston 2010) had the lowest variation among years (14% CV) but with a higher bias (77%).

Density estimates with our calibrations improved with call duration (Table 4-17). Estimating density with the mean response probability and mean actual response distance resulted in a mean relative bias of 22% and a CV of 57% (Table 4-17). Using the bootstrap estimate of response probability (Table 4-10) in combination with the estimated response distance (power) (Table 4-12) also resulted in a mean relative bias of 22% but the CV was 110%. Mean effective response distance resulted in a mean relative bias of 20% and a CV of 91%. These calibrations performed better than any of the published calibrations (Table 4-16).

The results from the GLMs to estimate reference density (Table 4-18) had lower mean proportional differences for 30 and 60 minutes' calling than other calibrations and were comparable to the best calibrations for a call duration of 90 minutes (Table 4-16) with a mean relative bias of 27% and 29%. The CV was 107% and 127% respectively.

Table 4-15 Parameter estimates with Generalized Linear Models to predict lion density (adult and sub-adult lions) from lions / calling station (*Lion_CS*), mean response probability (*mean_response*), mean actual response distance (*mean_actual_response_distance*), response area (*response_area*) and mean response time (*mean_time*).

Parameter	B	Std. Error	95% Wald Confidence Interval		Hypothesis Test		
			Lower	Upper	Wald Chi-Square	df	Sig.
Lion_CS	11.262	0.9968	9.309	13.216	127.662	1	0.000
(Scale)	3.237[a]	1.3803	1.404	7.467			
Lion_CS	8.820	2.0602	4.782	12.858	18.326	1	0.000
Mean_time	0.045	0.0338	-0.021	0.111	1.761	1	0.184
(Scale)	2.790[a]	1.1898	1.210	6.436			
Lion_CS	7.986	2.9572	2.190	13.783	7.294	1	0.007
Mean_response_distance	0.251	0.6429	-1.009	1.511	0.152	1	0.696
Mean_time	0.038	0.0378	-0.036	0.112	1.014	1	0.314
(Scale)	2.752[a]	1.1736	1.193	6.348			
Lion_CS	8.345	2.4976	3.450	13.240	11.163	1	0.001
Mean_response_distance	10.903	5.0551	0.995	20.811	4.652	1	0.031
Response_area	-1.297	0.6121	-2.497	-0.098	4.492	1	0.034
Mean_time	0.085	0.0389	0.009	0.162	4.816	1	0.028
(Scale)	1.954[a]	0.8333	0.847	4.508			
Lion_CS	8.679	2.4833	3.812	13.546	12.214	1	0.000
Mean_response	-16.915	23.3626	-62.705	28.875	0.524	1	0.469
Mean_response_distance	15.932	8.5228	-0.772	32.636	3.494	1	0.062
Response_area	-1.585	0.7179	-2.992	-0.178	4.874	1	0.027
Mean_time	0.085	0.0380	0.011	0.159	5.001	1	0.025
(Scale)	1.865[a]	0.7954	0.809	4.303			

a. Maximum likelihood estimate

Table 4-16 Relative bias of different published models to estimate lion density. Reference density of known lions and lions per calling station from NG/29 and NG/30 in the Okavango Delta.

YEAR	CALIBRATION	Cozzi *et al* 2013	Ferreira & Funston 2010	Ogutu & Dublin 1998	Brink *et al* 2012	Omoya *et al* 2013
	Duration (min)	60	60	30	40	30
	Response Distance (km)	2.00	4.30	2.50	1.50	1.56
	Response Area (km²)	12.57	58.09	19.63	7.07	7.6
	Response Probability	0.60	0.70	0.26	0.73	
1998	Lions/CS	0.76	0.76	0.72	0.72	0.72
1998	Estimate /100 km²	10.08	1.86	13.89	13.95	9.42
1998	% relative bias	26%	-77%	74%	75%	18%
1998	Reference density	7.99	7.99	7.99	7.99	7.99
1999	Lions/CS	0.36	0.36	0.32	0.32	0.32
1999	Estimate /100 km²	4.74	0.88	6.20	6.23	4.20
1999	% relative bias	-35%	-88%	-15%	-14%	-42%
1999	Reference density	7.26	7.26	7.26	7.26	7.26
2000	Lions/CS	0.45	0.45	0.00	0.00	0.00
2000	Estimate /100 km²	6.03	1.11	0.00	0.00	0.00
2000	% relative bias	-6%	-83%	-100%	-100%	-100%
2000	Reference density	6.44	6.44	6.44	6.44	6.44
2007	Lions/CS	0.35	0.35	0.32	0.32	0.32
2007	Estimate /100 km²	4.71	0.87	6.22	6.25	4.22
2007	% relative bias	102%	-63%	167%	168%	81%
2007	Reference density	2.33	2.33	2.33	2.33	2.33
	Mean relative bias	42	77	85	86	60
	SD	42	11	77	78	37
	95% CI	41	11	87	88	36
	CV	98%	14%	90%	91%	62%

Table 4-17 Relative bias of different models to estimate lion density with callup surveys in NG/29 and NG/30 in the Okavango Delta.

Year	Parameter	Mean response probability with mean actual response distance			Bootstrap response probability, response distance power			Bootstrap response probability, response distance logarithmic			Mean effective response distance		
	Duration (min)	30	60	90	30	60	90	30	60	90	30	60	90
	Response Distance (km)	2.59	2.77	2.85	1.38	2.15	2.78	1.48	2.19	2.59	1.66	1.67	1.84
	Response Area (km^2)	21.1	24.1	25.5	6.0	14.5	24.4	6.9	15.0	21.1	8.7	8.8	10.6
	Response Probability	0.31	0.39	0.45	0.24	0.32	0.40	0.24	0.32	0.40			
1998	Lions/CS	0.72	0.76	0.76	0.72	0.76	0.76	0.72	0.76	0.76	0.72	0.76	0.76
1998	Estimate /100 km^2	10.98	8.07	6.69	49.64	16.16	7.82	43.33	15.66	9.00	8.32	8.64	7.14
1998	% relative bias	37%	1%	-16%	521%	102%	-2%	442%	96%	13%	4%	8%	-11%
1998	Reference density	7.99	7.99	7.99	7.99	7.99	7.99	7.99	7.99	7.99	7.99	7.99	7.99
1999	Lions/CS	0.32	0.36	0.57	0.32	0.36	0.57	0.32	0.36	0.57	0.32	0.36	0.57
1999	Estimate /100 km^2	4.90	3.79	5.03	22.16	7.59	5.88	19.35	7.36	6.77	3.71	4.06	5.37
1999	% relative bias	-32%	-48%	-31%	205%	5%	-19%	166%	1%	-7%	-49%	-44%	-26%
1999	Reference density	7.26	7.26	7.26	7.26	7.26	7.26	7.26	7.26	7.26	7.26	7.26	7.26
2000	Lions/CS	0.00	0.45	0.68	0.00	0.45	0.68	0.00	0.45	0.68	0.00	0.45	0.68
2000	Estimate /100 km^2	0.00	4.83	6.00	0.00	9.66	7.01	0.00	9.37	8.08	0.00	5.17	6.41
2000	% relative bias	-100%	-25%	-7%	-100%	50%	9%	-100%	46%	25%	-100%	-20%	0%
2000	Reference density	6.44	6.44	6.44	6.44	6.44	6.44	6.44	6.44	6.44	6.44	6.44	6.44
2007	Lions/CS	0.32	0.35	0.35	0.32	0.35	0.35	0.32	0.35	0.35	0.32	0.35	0.35
2007	Estimate /100 km^2	4.92	3.77	3.12	22.24	7.54	3.65	19.41	7.31	4.20	3.73	4.03	3.34
2007	% relative bias	111%	62%	34%	855%	224%	57%	734%	214%	80%	60%	73%	43%
2007	Reference density	2.33	2.33	2.33	2.33	2.33	2.33	2.33	2.33	2.33	2.33	2.33	2.33
	Mean relative bias	70	34	22	420	95	22	361	89	31	53	36	20
	SD	41	27	13	341	95	24	290	92	34	39	29	19
	95% CI	40	26	12	334	93	24	284	90	33	39	28	18
	CV	57%	77%	57%	79%	97%	110%	79%	101%	105%	73%	78%	91%

Table 4-18 Relative bias of GLM models to estimate lion density. Reference density of known lions and lions per calling station from NG/29 and NG/30 in the Okavango Delta. Testing GLM reference density.

	GLM Reference density	Reference density= 11.262 (lions/CS)			Reference density= 8.820 (lions/CS) + 0.045 (mean time)		
	Duration	30	60	90	30	60	90
1998	Mean Time				18.20	25.86	25.86
1998	Lions/CS	0.72	0.76	0.76	0.72	0.76	0.76
1998	Estimate /100 km²	8.11	8.56	8.56	7.17	7.87	7.87
1998	% relative bias	1%	7%	7%	-10%	-2%	-2%
1998	Reference density	7.99	7.99	7.99	7.99	7.99	7.99
1999	Mean Time				25.00	33.33	49.60
1999	Lions/CS	0.32	0.36	0.57	0.32	0.36	0.57
1999	Estimate /100 km²	3.62	4.02	6.44	3.96	4.65	7.27
1999	% relative bias	-50%	-45%	-11%	-45%	-36%	0%
1999	Reference density	7.26	7.26	7.26	7.26	7.26	7.26
2000	Mean Time				-	42.67	57.60
2000	Lions/CS	0.00	0.45	0.68	0.00	0.45	0.68
2000	Estimate /100 km²	0.00	5.12	7.68	-	5.93	8.61
2000	% relative bias	-100%	-20%	19%	-	-8%	34%
2000	Reference density	6.44	6.44	6.44	6.44	6.44	6.44
2007	Mean Time				5.00	23.17	23.17
2007	Lions/CS	0.32	0.35	0.35	0.32	0.35	0.35
2007	Estimate /100 km²	3.63	4.00	4.00	3.07	4.17	4.17
2007	% relative bias	56%	72%	72%	32%	79%	79%
2007	Reference density	2.33	2.33	2.33	2.33	2.33	2.33
	Mean relative bias	52	36	27	29	31	29
	SD	40	28	30	18	35	37
	95% CI	40	28	29	20	35	36
	CV	76%	77%	107%	69%	111%	127%

A mean of 2.0 lions (adult plus sub-adult) responded per CS during the 1996 survey. The reference density of known lions (adult plus sub-adult) at the time of the 1996 survey was 15.73 lions / 100 km². Mean response probability with mean actual response distance estimated reference density of this independent data point with the smallest bias (Table 4-19), followed by the GLM model ranked second (AIC_C criteria) and mean effective response distance.

Table 4-19 Relative bias of different models to estimate lion density with the 1996 reference density of known lions and lions per calling station from NG/29 and NG/30 in the Okavango Delta.

	Mean response probability with mean actual response distance	Reference density= 8.820 (lions/CS) + 0.045 (mean time) AICC no 2	Mean effective response distance	Bootstrap response probability, response distance power	Reference density= 11.262 (lions/CS) AICC no 1	Bootstrap response probability, response distance logarithmic	Response probability and response distance (Cozzi *et al* 2013)
Duration (min)	90	90	90	90	90	90	60.00
Mean Time		7.50					
Response Distance (km)	2.85		1.84	2.78		2.59	2.00
Response Area (km²)	25.5		10.64	24.4		21.1	12.57
Response Probability	0.45		1.00	0.40		0.40	0.60
Lions/CS	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Estimate /100 km²	17.60	17.98	18.80	20.57	22.52	23.69	26.53
Reference density	15.73	15.73	15.73	15.73	15.73	15.73	15.73
% relative bias	12%	14%	20%	31%	43%	51%	69%

4.5 Discussion

A significant positive correlation existed between the number of lions responding to calling stations and reference density, a key assumption. Previous studies lacked reference density data to show that this relationship existed (Cozzi *et al.* 2013; Ferreira & Funston 2010; Ogutu & Dublin 1998).

4.5.1 Response time

If lions are evenly distributed around calling stations, as assumed (Cozzi *et al.* 2013; Kiffner *et al.* 2008), and their response declines with distance (Cozzi *et al.* 2013; Ferreira & Funston 2010; Ogutu & Dublin 1998; Omoya *et al.* 2013) one would expect less instances of lions responding with increasing time. This was the general pattern for survey data combined but not for individual surveys. Clumping of responding lions within the first 30 minutes or no lions responding before 30 minutes or after 60 minutes of calling in individual surveys indicated that lions were unevenly distributed across the response area. Spatial heterogeneity in distribution across landscapes have been recorded for lions (Cozzi *et al.* 2013), spotted hyaena (Graf *et al.* 2009; Mills, Juritz & Zucchini 2001) and brown hyaena (Winterbach *et al.* 2017). Ecological processes associated with seasonal flooding in the Okavango Delta result in temporal and spatial heterogeneity of prey species distribution and accessibility of parts of the landscape, contributing to the spatial heterogeneity of lions.

4.5.2 Response probability

Previous surveys spaced calling stations relatively far apart to reduce the probability of recording individuals more than once (Brink, Smith & Skinner 2013; Cozzi *et al.* 2013; Ferreira & Funston 2010; Ogutu, Bhola & Reid 2005; Omoya *et al.* 2013). Our study differed from other studies in two key aspects: we selected sites to have some overlap (maximum 5 km apart) to record as many individuals as possible, and we provided bait. Having calling stations closer to each other should increase the risk of individuals responding more than once. Identifying individuals, we could quantify multiple responses and only include the first response of individuals in our calculations.

In the short-term lions are less likely to respond a second time when exposed to calling without bait (Belant *et al*. 2016; Ferreira & Funston 2010; Omoya *et al*. 2013). Our results showed that 24% of lions responded to more than one call up; we attribute this to the bait that we provided, with responding animals finding a reward. When surveys have to be repeated in a short time span, animals recently exposed to calls have a lower response probability (Belant *et al*. 2017; Ferreira & Funston 2010; Omoya *et al*. 2013). Providing bait or blood can help to mitigate the trap shy response but can result in a trap happy response. Studies using a minimum distance of 7.5 km between calling stations (Ferreira & Funston 2010) reduce the risk of double counting the same individuals.

Ogutu and Dublin (1998) found that lions responded randomly with respect to age and sex. Our results supported this when the call duration extended to 90 minutes. Cozzi *et al.* (2013) suggested there is a more variable motivation for lions to respond when further than 1.5 km away from the calling stations. We found that CI and CV decreased by extending the call duration to 90 minutes, indicating more uniform responses than calling for 30 or 60 minutes. Ogutu, Bhola and Reid (2005) recommended a call duration of 60 minutes for lions in East Africa. We recommend a call duration of 90 minutes in the Okavango Delta. This should also be tested in other study areas.

Response probability of lions and spotted hyaenas are influenced by the type of call (Cozzi *et al.* 2013; Mills, Juritz & Zucchini 2001). Cozzi *et al.* (2013) predicted a decline in lion response probability from 0.6 (60 minutes) to 0.4 (90 minutes) when excluding lion roars from playback calls. Their prediction was marginally lower than the mean response probability of 0.45 that we calculated. Some studies have used only prey in distress calls to reduce the impact of competition between lions and hyaenas on response probability (Ferreira & Funston 2010; Ferreira & Funston 2016; Thorn *et al.* 2010). The use of different types of calls among surveys impact on response probability and response distance (Cozzi *et al.* 2013; Ferreira & Funston 2016), and therefore limit the direct compatibility of results to compare densities and determine population trends. Standardizing calls (Cozzi *et al.* 2013) or using only prey in distress calls (Ferreira & Funston 2010; Ferreira & Funston 2016) will improve comparability of results.

Response probability varies between study areas (Cozzi *et al.* 2013; Ferreira & Funston 2010; Ogutu & Dublin 1998; Omoya *et al.* 2013). We found that response probability differed significantly among surveys when calling for 30 or 60 minutes. Extending calling to 90 minutes reduced differences to non-significant levels. In contrast to the non-significant differences in response probability among surveys, the variation in the relative bias among years (29%) in our results indicated undetected variability among surveys in our study area. A once off calibration of lion response in the Okavango Delta may bias results. This raises the question whether response probability is similarly variable in other study areas.

4.5.3 Response distance

Response distance calibration methodology differed between Ogutu and Dublin (1998) and other studies. Most studies positioned the test calling stations at a pre-selected distance from the lions and called from that spot for the normal duration of a calling station (Cozzi *et al.* 2013; Ferreira & Funston 2010; Omoya *et al.* 2013). The procedure was repeated with different lions at different distances.

Ogutu and Dublin (1998) started calling from 3 km away, called for 5 minutes and waited 5 minutes. If the observed lions did not respond, they moved 500 m closer and repeated the calling.

Response distance varied considerably among studies. The type of call can influence response distance (Cozzi *et al.* 2013; Mills, Juritz & Zucchini 2001) and may partly explain some of the observed differences. An underestimate of response distance will result in an overestimate of density and vice versa. Ogutu and Dublin (1998) worked with a response distance of 2.5 km (30 minutes) and Brink, Smith and Skinner (2013) with 1.5 km (40 minutes). Cozzi *et al.* (2013) estimated a response distance in the Okavango Delta of 2 km for 60 minutes in contrast to the 4.3 km estimate of Ferreira and Funston (2010) in KNP for the same call duration.

Our estimated response distances (60 minutes), modelled with the Cozzi *et al.* (2013) calibration data, exceeded the 2 km estimated by them but were lower than the actual response distance we calculated from response probability and reference density. None of the estimated response distances in the Okavango Delta came close to the 4.3 km estimate in the Kruger National Park (Ferreira & Funston 2010), which is by far the longest response distance of any study.

Response distance should increase with call duration. We expected response distance to increase between 1.1 km and 1.4 km when extending the call duration from 30 to 90 minutes (Table 4-12). Contrary to expectations, we found that the actual response distance did not extend with longer calling, but the proportion of lions responding increased and became more uniform among different ages and sexes. A possible explanation is that carnivores will take longer to finish the carcass of a large prey than a small prey animal. Therefore, motivation to respond should increase with a longer the call duration, because it is more likely to be worthwhile for the animals to respond.

Different studies assumed minimal influence of diurnal and inter-diurnal variation in atmospheric conditions on the response distance of lions (Ferreira & Funston 2010; Ogutu, Bhola & Reid 2005). Thermal inversion can increase the audible range of elephant communication as much as fivefold, depending on the strength of the thermal inversion (Garstang *et al.* 1995; Larom *et al.* 1997). Our surveys were conducted at different times between August and November, covering the transition from cold dry season to hot dry season. The mean actual and mean effective response distances had the lowest CV among the four surveys (CV ≤ 14%) despite potential seasonal differences in thermal inversion. We lack appropriate data to assess the potential seasonal changes in thermal.

Thermal inversion may explain the longer response distance calculated in KNP (Ferreira & Funston 2010) in comparison to the Okavango (this study and Cozzi *et al.* (2013) and Serengeti/ Mara system in East Africa (Brink, Smith & Skinner 2013; Ogutu & Dublin 1998). KNP contains savanna vegetation with well-developed tree canopy that covers vast areas. In comparison, the floodplain systems in the

Okavango have limited tree cover, as do the Serengeti/ Mara system in East Africa. The tree cover helps to limit radiation and contains warmer air, thus stronger inversion should result in more woodland than grassland and floodplains. We also recommend that the impact of thermal inversion and vegetation on sound propagation needs to be investigated further (Cozzi *et al.* 2013; Ferreira & Funston 2010; Ogutu, Bhola & Reid 2005). The assumption that response probability and response distance remain the same among habitats within the survey area (Cozzi *et al.* 2013; Ferreira & Funston 2016; Mills, Juritz & Zucchini 2001; Ogutu, Bhola & Reid 2005) may not hold true.

4.5.4 Testing calibrations

Independent density data to test density estimates from indices are not generally available (Balme, Hunter & Slotow 2009; Ogutu, Bhola & Reid 2005). We used our survey data and reference lion densities as independent data to test published calibrations for call up surveys. The Cozzi *et al.* (2013) and Omoya *et al.* (2013) calibrations performed best of the calibrations we evaluated. The Cozzi *et al.* (2013) calibration was derived from sampling in the Okavango Delta, in a similar habitat to our study, and should perform well. The Kruger NP calibration (Ferreira & Funston 2010) had the lowest CV among surveys, but underestimated reference density due to the large response distance in their calibration.

We tested our calibrations with the same data, although only the 1996 data were independent. The highest ranked Generalized Linear Model required one parameter (lions/calling station) and the second highest ranked model added mean response time. The model including mean response time performed better to estimate the 1996 lion density. Lions were not uniformly distributed across the landscape, and mean time to respond incorporated this into the density estimate.

Mean response probability in combination with the mean actual response distance and the effective response distance, calculated using the “lure count” analysis method (Omoya *et al.* 2013), were the two best models to estimate lion density. Response probability is linked to the selected response distance because animals further away are less likely to respond (Ferreira & Funston 2010; Omoya *et al.* 2013). The “lure count” analysis method (Buckland *et al.* 2006; Omoya *et al.* 2013) to estimate the effective response distance (the distance where the number of animals inside the response radius that do not respond equals the number of animals that respond from outside), standardise the calculation. We recommend the “lure count” analysis method as the standard to calibrate response for call up surveys. It improves compatibility among studies to compare density estimates, response distance and response probabilities.

Although the mean relative bias improved with longer call durations, the CV remained very high (57% - 110%) at 90 minutes’ calling. It will be difficult to detect population trends with a CV of this magnitude (Gerrodette 1987; Taylor *et al.* 2007). The higher CV in our study than in KNP (Ferreira & Funston 2010) may be due to a higher spatial heterogeneity of lions in the Okavango Delta.

Results from biological surveys may be biased due to imperfect detections and thus misinform conservation management (Garrard *et al*. 2013; Karanth *et al*. 2011). Recent advances in analysis resulted in a shift from hypothesis driven statistics to the use of large data sets and complicated quantitative models to investigate how ecological processes are affected by different mechanisms at landscape scales (Burnham & Anderson 2002; Garrard *et al*. 2013; Gerber *et al*. 2012; Hines *et al*. 2010; Karanth *et al*. 2011; MacKenzie *et al*. 2002; Royle & Nichols 2003; Zipkin *et al*. 2014).

A variety of models have been developed that incorporate imperfect detection, for example, N-mixture models (Belant *et al*. 2016; Zipkin *et al*. 2014), random encounter models (Cusack *et al*. 2015) and spatially explicit capture-recapture (SECR) (Hines *et al*. 2010) that can even be used with unmarked or partially marked populations (Chandler & Royle 2013). Belant *et al*. (2016) applied N-mixture models to call up surveys for lions with temporal replication to estimate lion density in a section of Serengeti.

Estimating lion density with N-mixture models and other models relying on temporal or spatial replication may work well with the heterogeneity of lion response to call ups (e.g. Belant *et al*. 2016). These models can also produce biased results (Cusack *et al*. 2015; Gerber *et al*. 2012) and there is a need to evaluate the limitations and reliability of the emerging models beyond simulation (Dénes *et al*. 2015; Palmer *et al*. 2018). Our data, with reference densities, provide the opportunity to test models using spatial replication and compare performance with the more simplistic approaches we tested.

4.6 Conclusions

Call duration of 90 minutes provided the best results for lion in the Okavango Delta. A once off calibration to estimate response distance and response probability of a species may bias density estimates. Lion response varied between surveys, and we recommend that calibrations are repeated to improve estimates of response probability and response distance.

The high CV of lions/calling station plus the variability of response probability and response distance that we recorded, raise concerns regarding the ability to detect population trends of lions in the Okavango Delta with call up surveys. Planning surveys for other study areas and species should incorporate calibration and allow for a potential high variation. The results of calling station surveys should be interpreted cautiously and, where available, be supported with population structure and prey base data.

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CHAPTER 5. CONSERVATION IMPLICATIONS OF BROWN HYAENA (*Parahyaena brunnea*) POPULATION DENSITIES AND DISTRIBUTION ACROSS LANDSCAPES IN BOTSWANA

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Koedoe 59(2)

5.1 Abstract

The brown hyaena (*Parahyaena brunnea*) is endemic to southern Africa. The largest population of this near-threatened species occurs in Botswana, but limited data were available to assess distribution and density. Our objectives were to use a stratified approach to collate available data and to collect more data to assess brown hyaena distribution and density across land uses in Botswana.

We conducted surveys using track counts, camera traps and questionnaires and collated our results and available data to estimate the brown hyaena population based on the stratification of Botswana for large carnivores.

Brown hyaenas occur over 533 050 km² (92%) of Botswana. Our density estimates ranged from 0 brown hyaenas/100 km² in strata of northern Botswana to 2.94 (2.16–3.71) brown hyaenas/100 km² in the southern stratum of the Central Kalahari Game Reserve. We made assumptions regarding densities in strata that lacked data, using the best references available. We estimated the brown hyaena population in Botswana as 4642 (3133–5993) animals, with 6.8% of the population in the northern conservation zone, 73.1% in the southern conservation zone, 2.0% in the smaller conservation zones and 18.1% in the agricultural zones.

The similar densities of brown hyaenas in the Central Kalahari Game Reserve and the Ghanzi farms highlight the potential of agricultural areas in Botswana to conserve this species. The conservation of brown hyaenas in the agricultural landscape of Botswana is critical for the long-term conservation of the species; these areas provide important links between populations in South Africa, Namibia and Zimbabwe.

Conservation implications:

Botswana contains the core of the brown hyaena population in southern Africa, and conflict mitigation on agricultural land is crucial to maintain connectivity among the range countries.

5.2 Introduction

The brown hyaena (*Parahyaena brunnea*) has an estimated population size of <10 000 mature individuals and is therefore listed as near threatened on the IUCN Red List (Wiesel 2015). The species is endemic to southern Africa with range countries including Botswana, Namibia, South Africa, Angola and Zimbabwe (Mills & Hofer 1998; Wiesel 2015). The largest population of brown hyaena is found in Botswana (Wiesel 2015), a country that hosts one of the most diverse carnivore assemblages in Africa. The large carnivore guild includes lion (*Panthera leo*) (IUCN/SSC 2006), cheetah (*Acinonyx jubatus*) and wild dog (*Lycaon pictus*) (IUCN/SSC 2007) and significant populations of spotted hyaena (*Crocuta crocuta*) and leopard (*Panthera pardus*) (Jacobson *et al*. 2016).

The brown hyaena occurs over most of Botswana, except the Okavango Delta and sections in the north (Wiesel 2015). The range includes a diversity of human land uses such as conservation areas (Keeping 2014; Maude & Mills 2005), commercial farms (Boast & Houser 2012; Kent & Hill 2013) and communal land used for subsistence livestock farming (Muir 2009; Schiess-Meier *et al*. 2007).

The large carnivore guild in Botswana comprises a strong, interspecific dominance hierarchy including subordinate competitors (cheetahs, African wild dogs and brown hyaenas), dominant competitors (lions and spotted hyaenas) and leopards (i.e. the large carnivore least affected by interspecific competition) (Marker & Dickman 2005; Mills 2015). The interactions among carnivore guild members as well as their interactions with prey species are important components of biodiversity (Dalerum *et al*. 2008; Mills 2005). Conservation of the intact carnivore guild has a higher priority than the conservation of individual species (Woodroffe & Ginsberg 2005).

To conserve the intact guild, a mosaic of high and low densities of the dominant competitors is required to provide refuge areas for the subordinate competitors (Winterbach *et al*. 2013). Botswana has two large conservation zones with the potential to conserve the intact carnivore guild (Winterbach, Winterbach & Somers 2014). The southern conservation zone is characterised by a mosaic of low and medium densities of wild prey for lions and spotted hyaenas (Winterbach *et al*. 2014), and both carnivores occur at low to medium densities (Funston *et al*. 2001; Maude & Selebatso 2012; Mills 2015; Mudongo & Dipotso 2010). The northern conservation zone has a mosaic of high, medium and low prey densities for large carnivores, with the highest prey densities in and around the Okavango Delta and along the Kwando/Linyanti/Chobe river system (Winterbach *et al*. 2014). High densities of lions and spotted hyaenas have been recorded in the Okavango Delta and low densities in the dry parts of the northern conservation zone (Cozzi *et al*. 2013; Winterbach & Maude 2015; Winterbach & Winterbach 2003).

In addition to these conservation zones, the long-term conservation of brown hyaenas depends on the agricultural areas in Botswana (Boast & Houser 2012; Kent & Hill 2013). These areas do not have the potential to conserve the intact carnivore guild (Winterbach *et al*. 2014). They do, however, provide an opportunity to conserve the less dominant species including brown hyaenas, cheetahs, leopards and wild dogs (Boast & Houser 2012; IUCN/SSC 2007; Kent & Hill 2013; Klein 2007; Maude & Mills 2005; Winterbach *et al.* 2015).

Conservation of carnivores in the agricultural areas requires mitigation measures (Winterbach *et al*. 2013). Although brown hyaenas are mainly scavengers (Maude & Mills 2005; Mills 1990, 2015) and a low level of conflict is expected with livestock owners, human persecution is a significant threat to the species in the range countries (Mills & Hofer 1998; Wiesel 2015). In the North West province of South Africa, 40% of livestock owners regard the brown hyaena as a problem animal (Thorn 2008), and in Namibia, 72% of livestock owners believe that brown hyaenas were responsible for livestock depredations (Lindsey *et al*. 2013). Weise *et al*. (2015) confirmed conflict with brown hyaenas in Namibia, and according to farmers, conflict happens especially during peak calving or lambing seasons. Although faecal analysis and inspection of food remains at den sites showed cattle were a significant food source in the farming areas of the Gauteng and Limpopo, discussions with farmers indicated that the killing of cattle by brown hyaena was probably rare and that removing the individual responsible solved the problem (Skinner 1976). Although translocation can solve the conflict, one should consider brown hyaena sociality as part of the decision process to translocate individuals (Weise *et al.* 2015). Some cattle farmers in Botswana believe that brown hyaenas kill new born calves when the calves are left hidden while the mothers go foraging (pers. comm.). Maude and Mills (2005) reported that only 25% of cattle post owners around Makgadikgadi Pans National Park, Botswana, believed that brown hyaenas might cause livestock losses. They also found no loss of livestock because of brown hyaena predation around Makgadikgadi Pans National Park in a 5-year period, but did show that carcasses of livestock were an important food source to sustain brown hyaena populations in agricultural areas. Furthermore, brown hyaenas were a minority of the problem animal incidents reported in the Kweneng district, a livestock area south of the Central Kalahari Game Reserve in Botswana (Schiess-Meier *et al*. 2007). Boast (2014) recorded low levels of conflict with brown hyaenas and that they are the most tolerated large carnivore in Botswana.

The wide distribution of brown hyaenas on both conservation and agricultural land requires density estimates for all ecosystems and land uses in Botswana. Data to assess the large carnivore populations in Botswana are available in peer-reviewed articles and internal and unpublished reports. We reviewed articles and reports to identify some of the areas that were data deficient and conducted large carnivore surveys at multiple sites in Botswana. Here we present our data on brown hyaenas, collated with other published studies and unpublished or internal reports from the research community in Botswana to provide a countrywide population estimate of brown hyaenas. We discuss the implications of the results for carnivore conservation.

5.3 Research method and design

5.3.1 Study area

The Republic of Botswana is a landlocked country of 582 000 km^2, sharing international boundaries with Namibia, South Africa, Zimbabwe and Zambia. Altitude ranges from 515 to 1491 m above sea level. Most of Botswana is arid to semi-arid, with the Kalahari ecosystem occupying approximately 82% of the country. Rainfall is erratic and the mean rainfall ranges from 250 mm per year in the south-west to over 650 mm in the north-east (Department of Surveys and Mapping 2001). Over 90% of rainfall occurs in the summer months between November and April. Apart from the Okavango Delta and the perennial Kwando/Linyanti/Chobe river system, the only other surface water occurs in rivers and pans during the rainy season (Department of Surveys and Mapping 2001). The mean minimum temperatures range from 5°C in July to 19°C in January with the mean maximum daily temperatures ranging from 22°C in July to 33°C in January (Department of Surveys and Mapping 2001).

Vegetation over most of the country is shrub and tree savannah, which is classified as Sandveld (Department of Surveys and Mapping 2001). The Hardveld vegetation types are associated with hills and rocky outcrops in the eastern part of Botswana. The northern conservation zone comprises the wetland of the Okavango Delta, Sandveld, mopane *Colophospermum mopane* dominated vegetation types and limited Miombo woodland in the north-east. The Okavango Delta consists of a mosaic of islands, waterways and seasonal floodplains (Department of Surveys and Mapping 2001).

Seventeen percent of Botswana is fully protected as designated national parks and game reserves, and a further 21% is partially protected as designated Wildlife Management Areas (WMAs). Only 5% of the country is designated as urban. The balance of 57% comprises uncultivated rangeland consisting of approximately 5% freehold land, 25% state land and 70% tribal/communal grazing land (Department of Surveys and Mapping 2001). Commercial livestock production occurs on freehold, state land and tribal land.

Winterbach *et al*. (2014) identified four conservation zones and four agricultural zones in Botswana based on primary land use. The conservation zones consist of the large northern conservation zone and southern conservation zone, and the two smaller conservation zones of XaiXai and Tuli. The northern conservation zone includes Chobe National Park, Moremi Game Reserve, Nxai

Pan National Park, Makgadikgadi Pans National Park and the associated WMAs. The Central Kalahari Game Reserve, Kgalagadi Transfrontier Park and associated WMAs form the southern conservation zone (Figure 5-1). The agricultural zones are Ngami, Central, Ghanzi and Kgalagadi (Figure 5-1).

Livestock (mainly cattle) rearing forms 70% – 80% of the contribution of the agricultural sector to the gross domestic product (Botswana Ministry of Agriculture 2011). The national herd was 2.6 million cattle, 1.8 million goats and 300 000 sheep in 2012. Traditional cattle posts on communal grazing land are the most common livestock production system (Botswana Ministry of Agriculture 2011). There were 109 ranches producing game or cattle and game in 2013; these cover approximately 11 500 km^2 (Boast 2014). Approximately half of the population of 2 million people live in rural villages and small settlements and are thus partially or fully dependent on livestock for their livelihoods (Central Statistics Office 2014).

5.3.2 Procedure

We conducted track counts following the methodology of Stander (1998) and Funston *et al*. (2010). Existing roads and 4 × 4 trails were used for transects, covering parts of the northern conservation zone and the Central Kalahari Game Reserve in the southern conservation zone (Figure 5-2). Two trackers identified the tracks from a vehicle driven at slow speeds not exceeding 15 km/h. All fresh tracks (<24 hours old) of large carnivores were identified and recorded with the date, GPS location, species and number of individual animals. Data recording excluded roads disturbed by vehicles or rain in the previous 24 hours. Each individual should only be recorded once per day on the survey (Stander 1998). Multiple track incidences (observations of tracks) from the same species on the same transect were reviewed with the trackers to determine whether the tracks belonged to the same or new individuals.

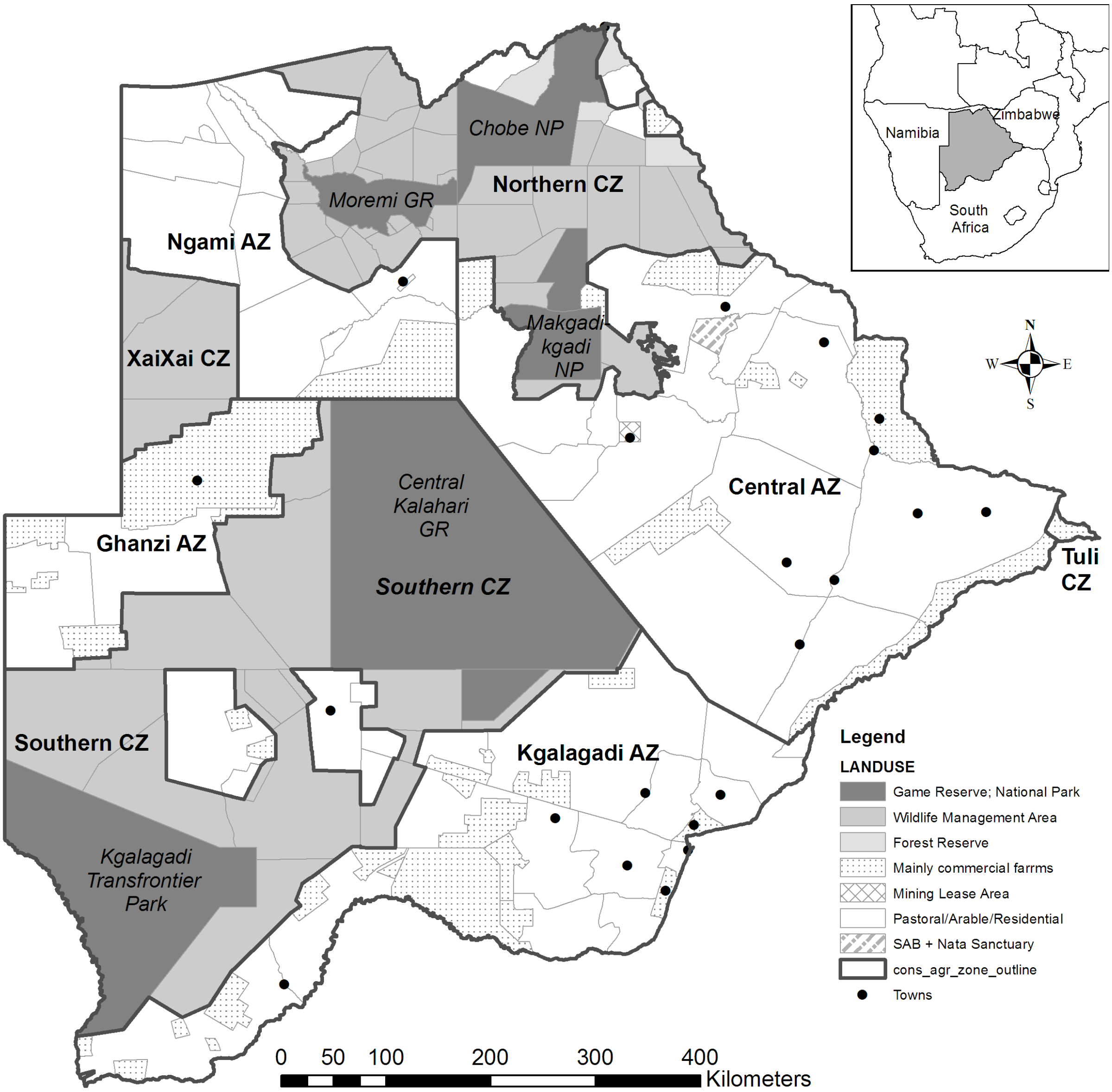

Figure 5-1 Primary land use in Botswana, including the conservation zones (national parks, game reserves and wildlife management areas) and agricultural zones (commercial farms and communal land with cattle posts). Insert shows Botswana in relation to southern Africa.

Source: Department of Surveys and Mapping (2001) and Winterbach *et al.* (2015)

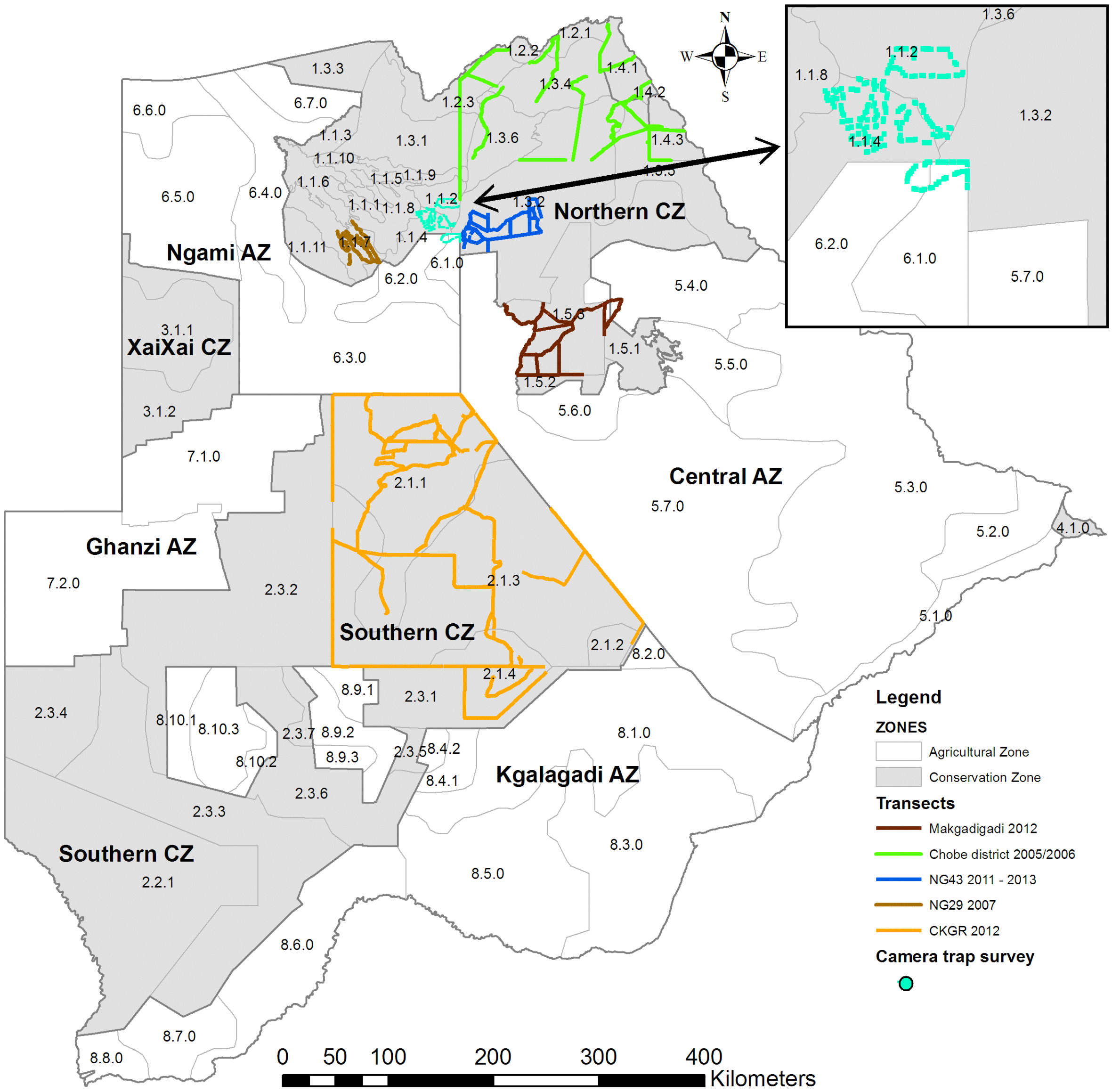

Figure 5-2 Stratification of Botswana and locations of track surveys and camera trap survey (detail on inset) conducted between 2005 and 2015 to estimate brown hyaena (*Parahyaena brunnea*). Stratification of Botswana is following Winterbach *et al*. (2014, 2015). Stratum identification numbers are shown on the map for reference.

We deployed Panthera v4 incandescent-flash and Bushnell TrophyCam infra-red camera traps at 221 locations across a 550 km^2 study area in the northern conservation zone and Ngami Agricultural Zone between February and July 2015. We used 5 km^2 grid cells to guide the placement of cameras and ensure systematic coverage of the entire study area. We deployed two camera stations within each grid cell, one on the road closest to the predetermined centre point of each grid cell and the second on the road closest to a predetermined random point within each grid cell. We placed all cameras on sand roads to increase our probability of photographing carnivores given that large predators and carrion feeders often use lightly travelled roads as movement corridors (Forman & Alexander 1998). If cameras had been placed randomly or on the extensive network of game trails, we believe our detection rates would have been prohibitively low. Each camera station included two opposing cameras mounted on trees, offset by 0.5 m–1 m. If there were no trees available, we mounted cameras on metal fence posts hammered into the ground. We secured cameras at knee height and positioned cameras to photograph flanks of passing animals. We programmed cameras to take three photos at each trigger event in the daytime with a delay of 30 seconds between trigger events. At night-time, the infra-red cameras took three photos when triggered but the incandescent-flash cameras could only take one photo every 15 seconds because of the flash having to recharge. For each station, we combined information from the two opposing cameras using the time or date stamps on the photographs.

We used a rotational system for camera deployment. We divided our study area into five sub-areas of approximately 110 km^2 each and sequentially sampled each area for 30 nights. We deployed an average of 44 camera stations (i.e. 88 cameras) within each sub-area. We checked cameras every 5–10 days to download photos, replace batteries and ensure cameras were still operational.

Distribution records of brown hyaenas consisted of track observations we recorded during our surveys, supplemented with brown hyaena observation records from publications, reports and verifiable observations between January 2005 and April 2016. Brown hyaena status, collected as part of a larger questionnaire survey conducted during 2012 and 2013, was recorded as present (visual sightings or tracks seen at least quarterly), transient (visual sightings or tracks seen less frequently than quarterly) or absent (never seen brown hyaena or its tracks). Klein (2013) surveyed cattle posts and commercial farms in the Kalahari region of Botswana, and Boast (2014) targeted primarily game ranchers and commercial livestock farmers in the game ranching regions of the Central, Ghanzi, Ngamiland and North East regional districts.

5.3.3 Data analysis

Transects were pooled per stratum for analysis, following the landscape stratification of Botswana for large carnivores (Figure 5-2) from Winterbach *et al*. (2014, 2015). The ratio of stratum size and transect length was used to calculate penetration (km^2/km of transect) as an index of sampling effort (Funston *et al*. 2010). We calculated track density (number of individual tracks/100 km sampled) per transect, treating each repetition an individual record. Following Funston *et al*. (2010), we calculated the mean and standard deviation of the distance (km) between track incidences for each stratum sampled. Funston *et al*. (2010) recommended that the coefficient of variance (CV) of distance between track incidences (standard deviation × 100/mean km per track incidence) should be less than 20% to ensure appropriate precision of the density estimate. This generally occurs after 19–30 track incidences (Funston *et al*. 2010). We provide penetration, distance between track incidences with the CV and number of track incidences as indicators of the quality of density estimates in each stratum. We estimated brown hyaena densities from track densities using the extended carnivore model for sandy substrates formula *animal density = track density/3.26* from Winterbach *et al*. (2016).

We included density data from other studies for the national population estimate (Appendix 1). We sourced articles and reports from the research community in Botswana. We recalculated the density estimates from other track surveys using the extended carnivore model for sandy substrates (Winterbach *et al*. 2016) for consistency of density calculation methods across studies. The population estimates were calculated per stratum from the density estimates, size of the stratum and the proportion of that stratum included in the brown hyaena range (*stratum population = density* × *stratum size/100* km^2).

No density estimates were available for some strata. Where we deemed strata to be similar (general habitat and general land use) to areas with reference data, we assumed similar population densities. In all other cases for strata lacking suitable reference data, we followed the same approach as the Namibia Large Carnivore Atlas (Hanssen & Stander 2004; Stein *et al*. 2012) using standardised density categories of low, medium or high:

Assumed low density: 0.085 brown hyaenas/100 km^2 (range 0.07–0.10).
Assumed medium density: 0.125 brown hyaenas/100 km^2 (range 0.10–0.15).
Assumed high density: 0.575 brown hyaenas/100 km^2 (range 0.15–1.0).

Because of the limited number of brown hyaenas recorded during the camera trap survey we could not calculate density. The camera trap records were used to indicate the presence and absence as part of mapping distribution. Locations from our surveys and questionnaires were plotted with other reliable location records collected from publications, reports and personal communications.

We used a chi-square test with Bonferroni simultaneous confidence intervals (Byers, Steinhorst & Krausman 1984) to compare frequencies that respondents reported brown hyaenas as present or transient or absent on commercial livestock farms and cattle posts compared to game farms, whose frequencies were defined as the expected values.

5.4 Results

5.4.1 Density estimates

We present data from track surveys we conducted between 2005 and 2007 (Chobe district and Okavango Delta) and between 2011 and 2013 (NG43, Makgadikgadi Pans National Park and the Central Kalahari Game Reserve) (Table 5-1). The lack of roads and 4 × 4 trails limited access to large parts of the study area and resulted in a low sampling effort in some strata as measured by the penetration index (Table 5-1). Penetrations ranged from 8.5 to 118.5 km²/km of transect. The surveys conducted during 2005 and 2006 totalled 3000 km of north-eastern Botswana (Figure 5-2). We completed 844 km in NG43 (WMAs near the Okavango Delta) during 2011, partially covering Stratum 1.3.2 (Figure 5-2). As part of long-term monitoring, we repeated surveys in NG43 during 2012 and 2013 (Table 5-1). During 2012, we completed the following transects: a total of 482 km in Makgadikgadi Pans National Park (Stratum 1.5.3), 1120 km in the northern Central Kalahari Game Reserve (Stratum 2.1.1), 1022 km in southern and eastern Central Kalahari Game Reserve (Strata 2.1.2 and 2.1.3) and 397 km in Khutse Game Reserve (Stratum 2.1.4).

The track incidences recorded in Makgadikgadi Pans National Park and the Central Kalahari Game Reserve ranged from 17 to 87 per stratum with mean distance between track incidences of 11.76 to 22.47 km (Table 5-2). The CV was between 102% and 143%, exceeding the 20% guideline (Funston *et al*. 2010). Although we completed 5550 km of track surveys north of Makgadikgadi Pans National Park, we only recorded tracks of brown hyaenas on five occasions. We could not calculate CV for the track frequency in this area because of the small sample size. The density estimates were between 0.00 and 0.14 animals/100 km². The southern Central Kalahari Game Reserve had the highest density of brown hyaenas (2.94 animals/100 km²), followed by 2.55 animals/100 km² in Makgadikgadi Pans National Park (Table 5-2).

The camera trap survey yielded only five records of brown hyaenas from 11 618 trap nights. These data were not sufficient to estimate density, but provided confirmation of the edge of the brown hyaena range in part of northern Botswana.

Table 5-1 Summary of track surveys completed between 2005 and 2013 to estimate brown hyaena (*Parahyaena brunnea*) densities in study areas across Botswana.

Survey year	Description	Stratum ID	Total distance of roads (km)	Study site (km²)	Penetration (km²/km transect)	Total distance sampled (km)	Mean transect length (km) ± SE	Number of transects (*n*)
2005–2006	Dry Woodland	S 1.1.2	19.5	1261	64.9	19.5	19.5 ± 0.0	1
2005–2006	Seasonal Floodplain North East	S 1.1.9	9.5	930	98.2	18.9	9.5 ± 0.0	2
2005–2006	Chobe	S 1.2.1	65.8	1070	16.3	285.0	21.9 ± 0.2	13
2005–2006	Kwando Linyanti	S 1.2.3	126.4	3576	28.3	732.8	18.3 ± 0.9	40
2005–2006	Kwando Delta link	S 1.3.1	58.7	6957	118.5	176.1	19.6 ± 0.1	9
2005–2006	Masame	S 1.3.2	191.5	19933	104.1	273.9	24.9 ± 3.0	11
2005–2006	Nogatsaa and Nunga	S 1.3.4 and S1.3.5	286.8	11374	39.7	894.4	18.6 ± 2.1	48
2005–2006	Savuti Mababe	S 1.3.6	80.7	3684	45.7	261.4	20.1 ± 0.3	13
2005–2006	Pandamatenga	S 1.4	127.6	4202	32.9	338.6	17.8 ± 2.2	19
2007	NG29 and NG30 (partial S1.1.7)	S 1.1.7	158.6	1506	9.5	582.7	20.1 ± 5.3	29
2011	NG43 (partial S 1.3.2)	S 1.3.2	355.3	3454	9.7	843.7	18.3± 5.0	46
2012	NG43 (partial S 1.3.2)	S 1.3.2	220.9	3454	15.6	550.4	17.8 ± 4.7	31
2013	NG43 (partial S 1.3.2)	S 1.3.2	406.1	3454	8.5	573.0	19.8 ± 7.1	29
2012	Makgadikgadi Pans National Park	S 1.5.3	482.4	7549	15.6	482.4	25.4 ± 5.0	19
2012	Central Kalahari Game Reserve North	S 2.1.1	1119.5	18850	16.8	1119.5	22.8 ± 5.2	49
2012	Central Kalahari Game Reserve South	S 2.1.3 and S 2.1.2	1021.6	31088	30.4	1021.6	22.2 ± 4.4	46
2012	Khutse	S 2.1.4	396.9	4902	12.4	396.9	23.3 ± 4.6	17

Table 5-2 Results of track surveys completed between 2005 and 2013 to estimate brown hyaena (*Parahyaena brunnea*) densities in study areas across Botswana.

Survey year	Description	Stratum ID	Track incidences: n	Mean distance between track incidences: no. km/set of tracks ± SE (CV)	Tracks/100 km: mean ± SE	Tracks/100 km: (95% CI)	Animals/100 km^2: (95% CI)
2005–2006	Dry Woodland	S 1.1.2	0	-	0	-	0
2005–2006	Seasonal Floodplain North East	S 1.1.9	0	-	0	-	0
2005–2006	Chobe	S 1.2.1	0	-	0	-	0
2005–2006	Kwando Linyanti	S 1.2.3	0	-	0	-	0
2005–2006	Kwando Delta link	S 1.3.1	0	-	0	-	0
2005–2006	Masame	S 1.3.2	0	-	0	-	0
2005–2006	Nogatsaa and Nunga	S 1.3.4 and S1.3.5	1	-	0.13 ± 0.88	-0.12–0.37	0.04 (-0.04–0.11)
2005–2006	Savuti Mababe	S 1.3.6	0	-	0	-	0
2005–2006	Pandamatenga	S 1.4	0	-	0	-	0
2007	NG29 and NG30 (partial S1.1.7)	S 1.1.7	0	-	0	-	0
2011	NG43 (partial S 1.3.2)	S 1.3.2	3	-	0.42 ± 1.63	-0.06–0.89	0.14 (-0.02–0.27)
2012	NG43 (partial S 1.3.2)	S 1.3.2	0	-	0	-	0
2013	NG43 (partial S 1.3.2)	S 1.3.2	1	-	0.15 ± 0.83	-0.15–0.46	0.09 (-0.05–0.14)
2012	MP NP	S 1.5.3	37	12.81 ± 18.37 (143%)	8.30 ± 6.70	5.29–11.32	2.55 (1.62–3.47)
2012	Central Kalahari Game Reserve North	S 2.1.1	53	20.72 ± 21.06 (102%)	5.69 ± 7.55	3.58–7.81	1.75 (1.10–2.39)
2012	Central Kalahari Game Reserve South and South East	S 2.1.3 and S 2.1.2	87	11.76 ± 13.41 (114%)	9.57 ± 8.71	7.05–12.09	2.94 (2.16–3.71)
2012	Khutse	S 2.1.4	17	22.47 ± 26.16 (116%)	4.50 ± 4.24	2.48–6.51	1.38 (0.76–2.00)

CV, coefficient of variance

5.4.2 Distribution

Brown hyaenas occur over 533 050 km² (92%) of Botswana (Figure 5-3). Previous studies recorded the presence of brown hyaenas in the Kgalagadi Transfrontier Park and surrounding WMAs (Funston *et al*. 2001; Keeping 2014; Mills 1990, Mudongo & Dipotso 2010) and Ghanzi (Boast & Houser 2012; Kent 2011; Kent & Hill 2013). These studies and the location data from our track surveys, camera trap survey, questionnaire survey and additional observation records are shown in Figure 5-3. Only a few records of brown hyaenas were recorded north of Makgadikgadi Pans National Park despite 5550 km of track surveys completed. The concessionaire reported that they only saw brown hyaena tracks in the southern half of NG43 during 15 years of operation (1999–2013) (pers. comm.). The results of the camera trap survey support that this was the northern edge of the brown hyaena range as brown hyaena were only recorded at the southern stations of the camera survey.

Eighty-two of 418 respondents did not record brown hyaenas as present, transient or absent and were excluded from further analysis. The questionnaire survey provided presence records of brown hyaenas in all four agricultural zones (Figure 5-3). Respondents recorded brown hyaenas as present (91.6%) and absent (8.4%) on game ranches (n =107). Presence of brown hyaena was 85.5% on commercial farms (n = 55) and 53.4% on cattle posts (n = 174). The observed frequency of presence and absence or transience differed (χ^2 = 331.25, df = 3, p < 0.001). Brown hyaenas were reported absent or transient more frequently and present less frequently on cattle posts than on game ranches (p = 0.001). It did not differ significantly between commercial livestock farms and game ranches (Table 5-3).

5.4.3 Population estimates

Our data and data from other sources that we used for the population estimates are summarised in Appendix 1. Large parts of the northern conservation zone are outside brown hyaena range or are part of the northern limit of their range. We calculated three density estimates north of Makgadikgadi Pans National Park, all between 0.04 and 0.14 animals/100 km². The five track records obtained along the 5550 km track survey were too low for robust density estimates and may indicate a patchy distribution towards the northern limit of the brown hyaena range.

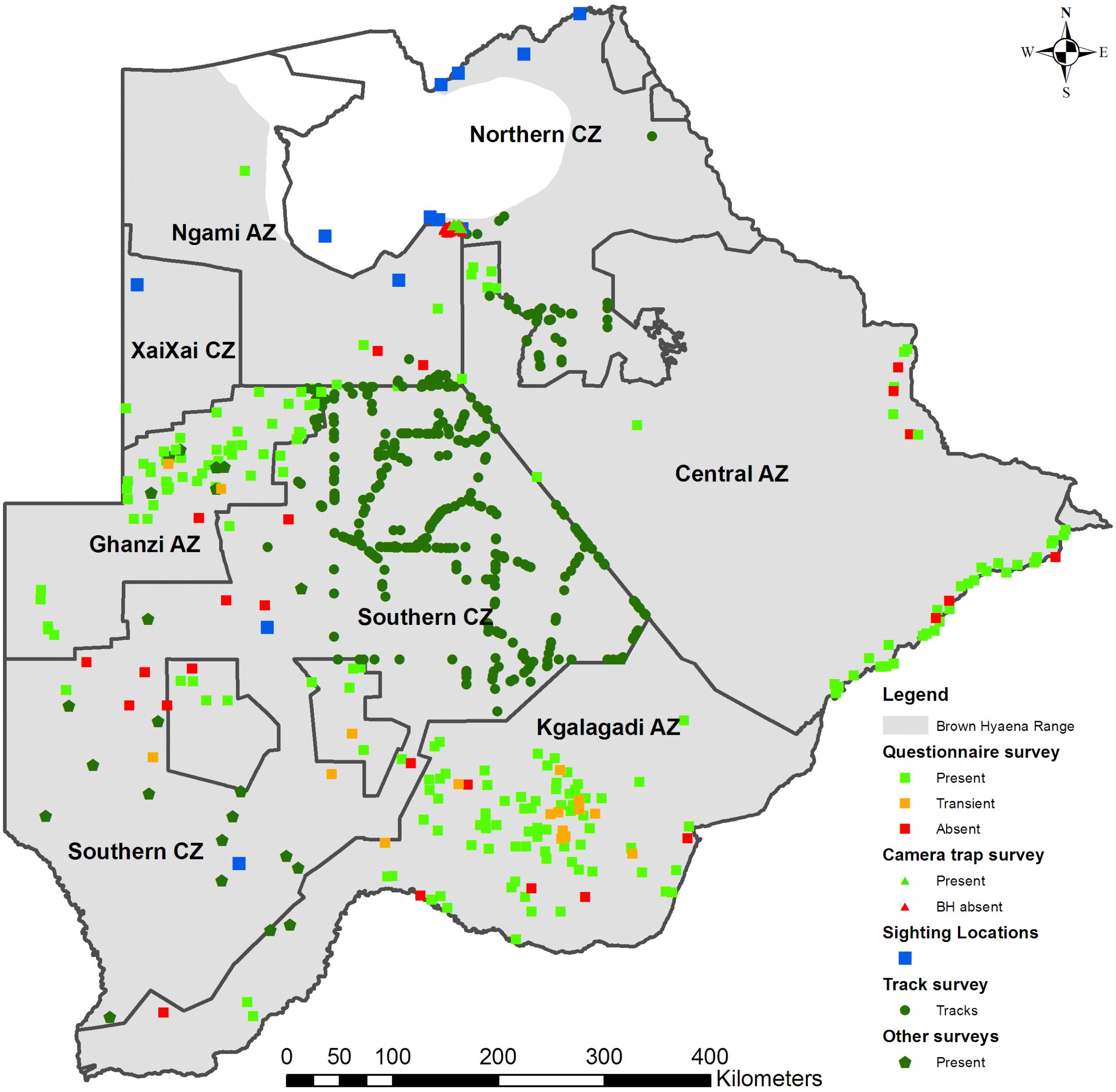

Figure 5-3 Brown hyaena (*Parahyaena brunnea*) distribution range in Botswana based on occurrences recorded between 2005 and 2016.

Table 5-3 Bonferroni simultaneous confidence intervals for the presence and absence ortransience of brown hyaena (*Parahyaena brunnea*) based on questionnaires (2008–2009 and 2011–2012) completed by farmers on game ranches (*n* = 107), commercial livestock farms (*n* = 55) and traditional cattle posts (*n* = 174) in the agricultural zones of Botswana.

Land use and status	**Expected**	**Observed**	**Chi square**	**Expected proportion Pio**	**Bonferroni intervals for observed proportion Pi**	**Use index Pi/Pio**	**Significant (α = 0.001)**
Absent + transient cattle post	14.64	81	300.93	0.0639	0.2380 ≤ Pi ≤ 0.4694	5.53	+
Present cattle post	159.36	93	27.64	0.6959	0.2873 ≤ Pi ≤ 0.5250	0.58	-
Absent + transient commercial livestock	4.63	8	2.46	0.0202	-0.0095 ≤ Pi ≤ 0.0794	1.73	0
Present commercial livestock	50.37	47	0.23	0.2200	0.1075 ≤ Pi ≤ 0.3030	0.93	0
Total	229.0	229	331.25	1.0000	-	-	-

The proportions of present (91.6%) and absent or transient (8.4%) recorded on game farms were used as the reference to calculate the expected proportions for commercial livestock farms and cattle posts (k = 4, α = 0.001, Z = 3.6623).

We, therefore, assumed (Appendix 1) that brown hyaenas occur at extremely low densities north of Makgadikgadi Pans National Park (range 0.0–0.1 animals/100 km² and mean 0.05 animals/100 km²). Recorded densities (Appendix 1) varied between 1.13 and 3.90 animals/100 km² in the southern conservation zone (Funston *et al*. 2001; Keeping 2014, Maude & Selebatso 2012, Mills 1990; Mudongo & Dipotso 2010). Brown hyaena densities were estimated between 1.89 and 3.10 animals/100 km² in the Ghanzi farms (Boast & Houser 2012; Kent 2011; Kent & Hill 2013).

No density data were available for the Central Agricultural Zone (north-east Botswana) and Kgalagadi Agricultural Zone (south-east Botswana). These two zones border on the North West province in South Africa where Thorn *et al*. (2011) estimated overall density of brown hyaenas in agricultural land as 0.15/100 km² ± SE 0.08. Based on this, we assumed a medium density with 0.125 brown hyaenas/100 km², ranging from 0.1 to 0.15 animals/100 km² in these two agricultural zones (Appendix 1).

For substrata 6.5.0 (Ngamiland) and 7.2.0 (Ghanzi), we assumed high density [0.575 brown hyaenas/100 km² (range 0.15–1.00)] based on the high densities recorded in neighbouring areas (Appendix 1). The low-density assumption [0.085 brown hyaenas/100 km² (range 0.07–0.10)] was applied to the substrata of Ngami Agricultural Zone, as this is part of the northern edge of brown hyaena distribution (Table 5-4).

We estimated the brown hyaena population of Botswana as 4642 animals (3133–5993) (Table 5-4). Seventy-three percent of the brown hyaena population (approximately 3393 animals) occurred within the southern conservation zone (Table 5-4) and 18.1% in the agricultural zones (843 brown hyaenas). The remaining 6.8% occurred in the northern conservation zone and 2% in the conservation zones of XaiXai and Tuli. The estimated and assumed densities are shown in Figure 5-4.

5.5 Discussion

Our study combined information from track surveys, camera trap surveys, questionnaires and previous studies to estimate local densities and distribution of brown hyaenas across Botswana. Information on brown hyaena populations is vital to making informed conservation decisions and to mitigating population declines, particularly as the species is listed as near threatened (Wiesel 2015). Because of the large scale of this study our data had several limitations.

Table 5-4 Population estimates per stratum of brown hyaenas (*Parahyaena brunnea*) in Botswana based on surveys conducted between 2005 and 2015.

Zone	Stratum	Size (km²)	Population estimate	Range minimum	Range maximum
Northern CZ	1.1 Okavango Delta	16 045	0	0	0
Northern CZ	1.2 Kwando/Chobe	5103	1	0	2
Northern CZ	1.3 Dry North	44 899	13	0	27
Northern CZ	1.4 Pandamatenga	4202	2	0	4
Northern CZ	1.5 Pans	11 684	298	190	406
Southern CZ	2.1 Central Kalahari GR & Khutse	54 841	1309	917	1702
Southern CZ	2.2 KTP	26 210	603	419	734
Southern CZ	2.3 KTP WMA	73 697	1481	999	1840
Xaixai CZ	3.1 Xaixai	15 597	90	23	156
Tuli CZ	4.1 Tuli GR	686	2	0	4
Central Agric	5.1 Central Tuli farms	4345	5	4	7
Central Agric	5.2 Central Agric Molalatau	5978	7	6	9
Central Agric	5.3 Central Agric East	50 837	64	51	76
Central Agric	5.4 Central Agric North	12 161	15	12	18
Central Agric	5.5 Central Agric Pans	5593	7	6	8
Central Agric	5.6 Central Agric Mopipi	4013	5	4	6
Central Agric	5.7 Central Agric West	53 688	67	54	81
Ngami Agric	6.1 Ngami East	1060	1	1	1
Ngami Agric	6.2 Ngami Thamalakane Boteti	3908	2	2	3
Ngami Agric	6.3 Ngami South	18 886	16	13	19
Ngami Agric	6.4 Ngami Panhandle	8696	4	3	4
Ngami Agric	6.5 Ngami Tsodilo	14 169	81	21	142
Ngami Agric	6.6 Ngami North West	4164	4	3	4
Ngami Agric	6.7 Ngami North East	3303	0	0	0
Ghanzi Agric	7.2 Ghanzi Agric	34 894	439	305	591
Kgalagadi Agric	8.1 Kgalagadi Kweneng	26 059	33	26	39
Kgalagadi Agric	8.2 Kgalagadi Dithopo	1057	1	1	2
Kgalagadi Agric	8.3 Kgalagadi Gaborone	18 545	23	19	28
Kgalagadi Agric	8.4 Kgalagadi Kane	2763	3	3	4
Kgalagadi Agric	8.5 Kgalagadi Molopo	17 550	22	18	26
Kgalagadi Agric	8.6 Kgalagadi Tsabong	11 970	15	12	18
Kgalagadi Agric	8.7 Kgalagadi Middelpits	4888	6	5	7
Kgalagadi Agric	8.8 Kgalagadi Bokpits	2159	3	2	3
Kgalagadi Agric	8.9 Kgalagadi Kang	6580	8	7	10
Kgalagadi Agric	8.10 Kgalagadi Hukuntsi	8915	11	9	13
	Grand Total	57 9144	4642	3133	5993

CZ, Conservation Zone; GR, Game Reserve.

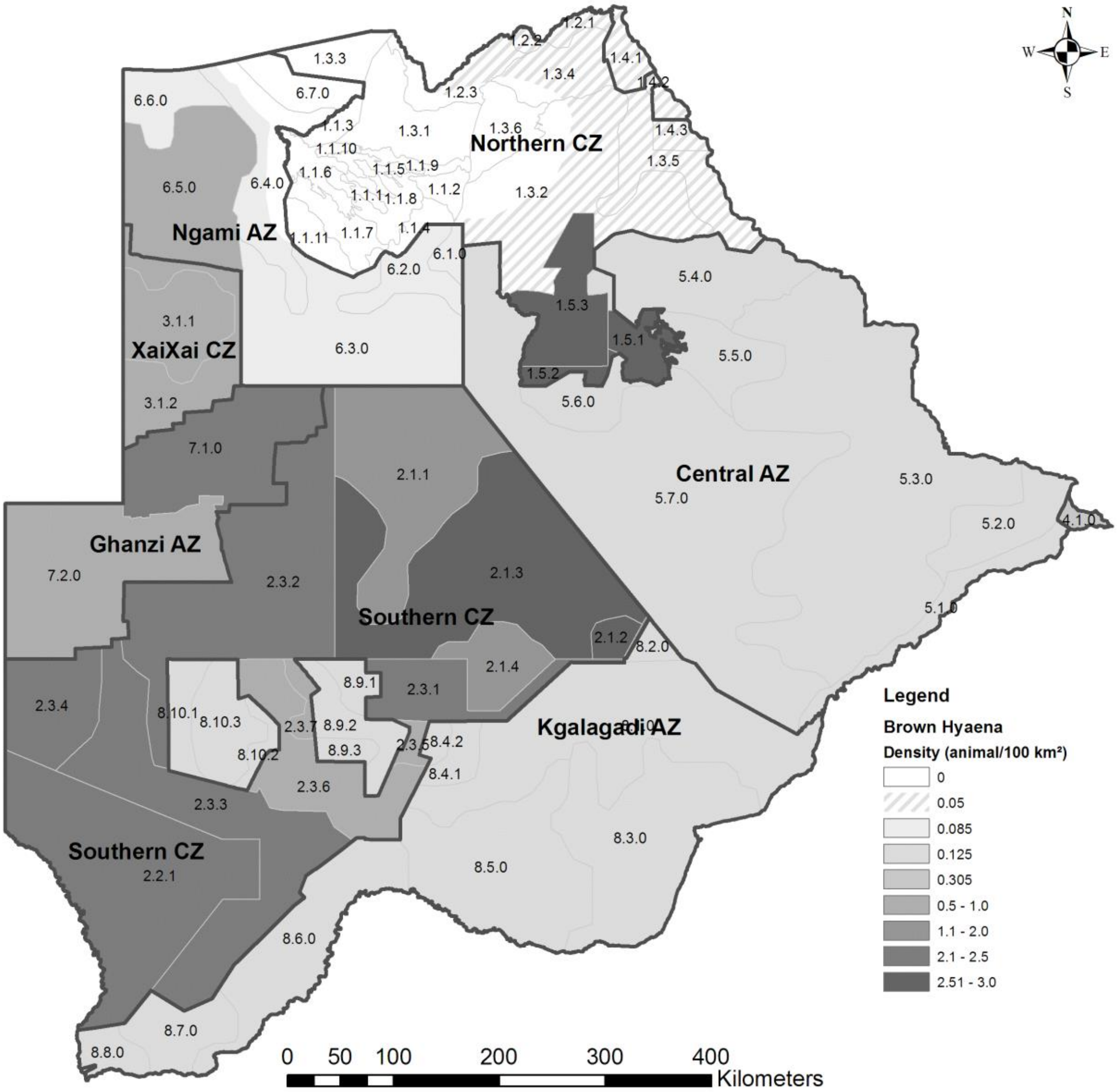

Figure 5-4 Estimated and assumed densities per stratum of brown hyaenas (*Parahyaena brunnea*) in Botswana based on surveys conducted between 2005 and 2015. Stratum identification numbers are shown on the map for reference.

The data were collected over a decade, and there is a risk that carnivore densities changed because of ecological factors or changes in land use as demonstrated in Zimbabwe (Williams *et al*. 2016). No large-scale changes in land use occurred in Botswana during the past 15 years. The brown hyaena density estimates in Kgalagadi Transfrontier Park area of Mills (1990), Funston *et al*. (2001) and Keeping (2014) were similar despite covering more than 20 years. The long-term study of Maude (2010) found no population declines in the Makgadikgadi region. Despite this long timespan, our data sets are the best indication of the national population of brown hyaena in Botswana.

The recommended sampling effort of 19–30 recorded track incidences per sampling unit should result in a CV below 20% for mean distance between track incidences (Funston *et al*. 2010; Kent 2011). Despite us recording track incidences of 37, 53 and 87 in three strata of the Central Kalahari Game Reserve, the CVs of these strata still exceeded 100%. This contradicts the results from localised surveys in the Ghanzi farms area (Boast & Houser 2012; Kent 2011) that achieved CVs of less than 20%. These studies recorded densities varying between 1.89 and 3.10 brown hyaenas/100 km² on small units within the stratum 7.1.0 Ghanzi farms. Similarly, brown hyaena densities varied among habitats within stratum 2.2.1, the Kgalagadi Transfrontier Park (1.13–2.17 brown hyaenas/100 km²) (Funston *et al*. 2001) and among locations in the surrounding WMAs (2.36–3.90 brown hyaenas/100 km²) (Mudongo & Dipotso 2010). The above mentioned studies show heterogeneity in brown hyaena densities across landscapes in conservation and agricultural zones of Botswana on a finer resolution than the stratification we used. Therefore, we should expect heterogeneity in brown hyaena densities in the large survey strata that result in high CVs.

Although our current estimate of 4642 (3133–5993) brown hyaenas is higher than the previous estimates of 3900 (3500–4500) (Mills & Hofer 1998) and 2636 (1990–3282) (Winterbach & Winterbach 2003), it probably does not reflect a population increase. Rather, it indicates that the population was previously underestimated because of a lack of comprehensive data. Although there is evidence that brown hyaenas are persecuted in Botswana (Maude & Mills 2005), we found no indication of a population decline. This is supported by a long-term study on brown hyaenas in the Makgadikgadi region of Botswana by Maude (2010). He concluded that brown hyaenas are sufficiently adaptable to live alongside people in agricultural areas and thus are not vulnerable to significant population declines under current land uses (Maude 2010).

The similar brown hyaena densities recorded in Ghanzi and the southern conservation zone (Appendix 1) highlight the importance of agricultural zones in Botswana for brown hyaena conservation (Boast & Houser 2012; Kent & Hill 2013). Furthermore, Maude and Mills (2005) showed that brown hyaenas occurred at higher densities in the communal land in the agricultural zone around Makgadikgadi Nxai Pan National Park, where home range sizes are smaller and clan numbers are higher than inside the national park. In contrast to these findings, Thorn *et al*. (2011) estimated a much lower density of brown hyaenas in agricultural land (0.15/100 km² ± SE 0.08) than in Pilanesberg National Park, North West province of South Africa. Less antagonism, lower human density, a lack of large-scale crop production and differences in livestock management practices may contribute to a more hospitable environment for brown hyaenas in Ghanzi than the North West province (Kent & Hill 2013).

The findings of Maude and Mills (2005), Schiess-Meier *et al*. (2007) and Kent and Hill (2013) indicated that we potentially could have relatively high brown hyaena densities in the Ngami, Central and Kgalagadi agricultural zones. We lacked density data in these areas that include large parts of communal land with cattle posts. Brown hyaena densities in Botswana, apart from the north, varied between 1.2 and 3.9 animals/100 km² (Appendix 1). However, our questionnaire surveys showed that brown hyaenas were more likely to be reported as transient or absent on the communal land with cattle posts than on commercial and game farms. This can be the result of observer bias, with the respondents either over- or under-reporting the presence of brown hyaena. The alternative is that brown hyaena did occur less on communal land. Our current data are not suitable to clarify this. The brown hyaena densities recorded elsewhere in Botswana might not be representative of these agricultural zones, and therefore, we opted for the conservative density assumption of 0.15 animals/100 km² based on the results of Thorn *et al*. (2011).

The communal farmlands with cattle posts in these three agricultural zones represent a major gap in our knowledge of brown hyaenas in Botswana. Brown hyaenas can thrive in agricultural areas of Botswana (Boast & Houser 2012; Kent & Hill 2013; Maude & Mills 2005). Therefore, we recommend that future brown hyaena surveys focus particularly on the Ngami, Central and Kgalagadi agricultural zones, which may be strongholds for the species.

A significant proportion (18.1%) of the brown hyaena population in Botswana occurred in agricultural areas of Botswana. For example, our estimates showed more brown hyaenas in the Ghanzi farms (Stratum 7.1.0) than all of the northern conservation zone. The agricultural zones are not only key areas for the conservation of brown hyaenas in Botswana but are also essential to maintain links with populations in Namibia and South Africa.

The stronghold for brown hyaenas in the northern conservation zone is the Makgadikgadi Pans. This area and the strata in the northern conservation zone without perennial water have low prey densities and should be refuge areas for subordinate carnivores (Winterbach *et al*. 2014). However, we recorded brown hyaena infrequently in the strata north of Makgadikgadi, despite low densities of lion and spotted hyaena (unpublished data from our surveys). Annual rainfall increases from 400 mm in Makgadikgadi to 700 mm in the north-east of the northern conservation zone (Department of Surveys and Mapping 2001). This may be a factor that limits brown hyaena in the northern conservation zone directly or indirectly, because brown hyaena occurs in areas with rainfall up to approximately 700 mm (Wiesel 2015). Also, the species does not occur in the Okavango Delta, the area around Moremi Game Reserve, an area that supports high densities of lions and spotted hyaenas (Cozzi *et al*. 2013; Winterbach & Winterbach 2003). Mills and Mills (1982) and Mills (2015) found that spotted hyaenas outcompete brown hyaenas and this interspecific competition may explain why the Okavango Delta is not part of the brown hyaenas range (Mills & Hofer 1998; Wiesel 2015). Although it is not clear from our distribution and density data whether brown hyaenas are resident in localised areas or merely transient through the northern part of their range in Botswana, the northern conservation zone provides the link between the Botswana and Zimbabwe populations.

Botswana supports the highest number of brown hyaenas of all the range countries (Wiesel 2015), and a significant proportion of this national population occurs on agricultural land. The focus of carnivore conservation in Botswana should be on maintaining the intact large carnivore guild in the conservation zones, complimented by a species conservation approach in the agricultural zones. We recommend that future surveys in Botswana specifically include density estimates for brown hyaenas on communal farmland in the Ngami, Central and Kgalagadi agricultural zones.

Less livestock owners in Botswana regard brown hyaenas as a risk to livestock than their counterparts in South Africa and Namibia (Lindsey *et al.* 2013; Maude & Mills 2005; Thorn 2008), which may be related to farming practices, that can increase the risk of brown hyaenas killing new born and weak calves (pers. comm.). Regional and individual differences in behaviour of brown hyaenas may contribute to the lower perceived threat in parts of Botswana. Although brown hyaenas may on rare occasions cause livestock losses (Skinner 1976; Weise *et al*. 2015), they are not a significant problem animal species.

Persecution of brown hyaenas in the livestock areas of Botswana may impact negatively on the long-term conservation of the species across its distribution range in southern Africa. The resident brown hyaena population in the livestock areas of Botswana is not only a significantly large population, but is also critical to maintain links among the conservation zones in Botswana and the neighbouring range countries Namibia, South Africa and Zimbabwe. The information provided here should be incorporated into conservation strategies for large carnivores in Botswana.

5.6 Conclusion

We found no indication that the estimated population of 4642 (3133–5993) brown hyaenas in Botswana declined. Brown hyaena densities were heterogeneous across landscapes in conservation and agricultural zones on a finer resolution than the stratification we used. We lack data to assess the population in large parts of the agricultural areas and recommend that future brown hyaena surveys focus particularly on the Ngami, Central and Kgalagadi agricultural zones, which may be strongholds for the species. The agricultural areas in Botswana are important to maintain population links between Namibia, South Africa and Zimbabwe.

5.7 References

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5.8 Appendix 1

TABLE 1-A1: Summary of reference data to estimate density of brown hyaenas (*Parahyaena brunnea*) in Botswana.

Zone	Sub-stratum	Range (% of unit)	Location	Reference	Method and year	Mean track dens	Density
Northern CZ	1.1.1 Chiefs Island	0	-	-	-	-	0.00
Northern CZ	1.1.2 Dry Woodland	0	-	-	-	-	0.00
Northern CZ	1.1.3 Eretsa community	0	-	-	-	-	0.00
Northern CZ	1.1.4 Lower Delta	0	-	-	-	-	0.00
Northern CZ	1.1.5 Permanent Swamp East	0	-	-	-	-	0.00
Northern CZ	1.1.6 Permanent Swamp Central	0	-	-	-	-	0.00
Northern CZ	1.1.7 Seasonal Floodplain West	0	-	C.W. Winterbach	Track survey 2007: 83 km, no tracks	0.0	0.00
Northern CZ	1.1.8 Seasonal Floodplain Central	0	-	-	-	-	0.00
Northern CZ	1.1.9 Seasonal Floodplain North East	0	-	-	-	-	0.00
Northern CZ	1.1.10 Seasonal Floodplain North West	0	-	-	-	-	0.00
Northern CZ	1.1.11 Western Delta and Sandveld Tongue	0	-	-	-	-	0.00
Northern CZ	1.2.1 Chobe	100	-	G. Neo Mapuleng	Track survey 2005/2006: 285 km, no tracks	0.0	0.00
Northern CZ	1.2.1 Chobe	100	-	-	Assume extremely low density.	-	0.05 (0.00–0.10)
Northern CZ	1.2.2 Enclave community	100	-	-	Assume extremely low density.	-	0.05 (0.00–0.10)
Northern CZ	1.2.3 Kwando Linyanti	19	-	G. Neo Mapuleng	Track survey 2005/2006: 733 km, no tracks.	0.0	0.00
Northern CZ	1.2.3 Kwando Linyanti	19	-	-	Assume extremely low density.	-	0.05 (0.00–0.10)
Northern CZ	1.3.1 Kwando Delta link	0	-	G. Neo Mapuleng	Track survey 2005/2006: 176 km, no tracks.	0.0	0.00
Northern CZ	1.3.2 Masame	76	-	G. Neo Mapuleng	Track survey 2005/2006: 274 km, no tracks.	0.00	0.00

Zone	Sub-stratum	Range (% of unit)	Location	Reference	Method and year	Mean track dens	Density
Northern CZ	*1.3.2 Masame*	*76*	*NG43*	*C.W. Winterbach*	*Track survey 2011: 844 km, 3 individuals, Track Frequency 0.42.*	*0.42*	*0.13 (-0.02–0.27)*
Northern CZ	*1.3.2 Masame*	*76*	*NG43*	*C.W. Winterbach*	*Track survey 2012: 550 km, 0 individuals*	*0.00*	*0.00*
Northern CZ	*1.3.2 Masame*	*76*	*NG43*	*C.W. Winterbach*	*Track survey 2013: 573 km, 1 individuals, Track Frequency 0.15.*	*0.15*	*0.05 (-0.05–0.14)*
Northern CZ	1.3.2 Masame	76	NG43		Assume extremely low density.	-	0.05 (0.00–0.10)
Northern CZ	1.3.3 Ngami North	0	-	-	-	-	0.00
Northern CZ	1.3.4 Nogatsaa	100	-	-	Assume extremely low density.	-	0.05 (0.00–0.10)
Northern CZ	*1.3.4 & 1.3.5*	*100*	-	*G. Neo Mapuleng*	*Track survey 2005/2006: 894 km, 1 individual, track frequency 0.13.*	*0.126*	*0.04 (-0.04–0.11)*
Northern CZ	1.3.5 Nunga	100	-	-	Assume extremely low density.	-	0.05 (0.00–0.10)
Northern CZ	1.3.6 Savuti Mababe	0	-	G. Neo Mapuleng	Track survey 2005/2006: 261 km, no tracks.	0	0.00
Northern CZ	*1.4*	*100*	*1.4.1, 1.4.2 and 1.4.3 combined*	*G. Neo Mapuleng*	*Track survey 2005/2006: 339 km, no tracks.*	*0*	*0.00*
Northern CZ	1.4.1 Kazangula	100	-	-	Assume extremely low density.	-	0.05 (0.00–0.10)
Northern CZ	1.4.2 Panda farms	100	-	-	Assume extremely low density.	-	0.05 (0.00–0.10)
Northern CZ	1.4.3 Pandamatenga	100	-	-	Assume extremely low density.	-	0.05 (0.00–0.10)
Northern CZ	1.5.1 Pans Community	100	-	Maude (2008)	Assume = Pans NP 2012 survey.	-	2.55 (1.62–3.47)
Northern CZ	1.5.2 Pans Community Area	100	-	Maude (2008)	Assume = Pans NP 2012 survey.	-	2.55 (1.62–3.47)
Northern CZ	*1.5.3 Pans NP*	*100*	*Pans*	*Maude (2008)*	*Collars*	-	*1.20 (0.90–1.50)*

Zone	Sub-stratum	Range (% of unit)	Location	Reference	Method and year	Mean track dens	Density
Northern CZ	1.5.3 Pans NP	100	Pans	G. Maude	Track survey 2012: 482.2 km, 40 individuals, Track Frequency 8.3.	8.30	2.55 (1.62–3.47)
Southern CZ	-	*100*	*Central Kalahari Game Reserve North and south combined, including Khutse*	*G. Maude*	*Track survey 2012: 2538 km, 172 individuals, track freq 7.1.*	*7.10*	*2.18 (1.73–2.63)*
Southern CZ	-	*100*	*Central Kalahari Game Reserve*	*Senior Wildlife Biologist (2000)*	*Track survey 2000: Small sample.*		*2.60 (2.00–3.20)*
Southern CZ	2.1.1 Central Kalahari Game Reserve North	100	Central Kalahari Game Reserve Central Kalahari Game Reserve North (2.1.1) and northern boundaries	G. Maude	Track survey 2012: 1119.5 km, 60 individuals, Track Frequency 5.69.	5.69	1.75 (1.10–2.39)
Southern CZ	2.1.2 Central Kalahari Game Reserve SE	100	Central Kalahari Game Reserve South (2.1.3) and south east (2.1.2) combined with boundary	G. Maude	Track survey 2012: 1021.6 km, 95 individuals, Track Frequency 9.57.	9.57	2.94 (2.16–3.71)
Southern CZ	2.1.3 Central Kalahari Game Reserve South	100	Central Kalahari Game Reserve South (2.1.3) and south east (2.1.2) combined with boundary	G. Maude	Track survey 2012: 1021.6 km, 95 individuals.	9.57	2.94 (2.16–3.71)
Southern CZ	2.1.4 Khutse	100	Central Kalahari Game Reserve Khutse including boundaries	G. Maude	Track survey 2012: 396.9 km, 17 individuals.	4.50	1.38 (0.76–2.00)

Zone	Sub-stratum	Range (% of unit)	Location	Reference	Method and year	Mean track dens	Density
Southern CZ	2.2.1 KTP	100	KTP and KTP Buffer KTP and KD1, KD2 combined	Keeping (2014)	Track survey with FMP formula, 634 km, CV 10%.	-	2.30 (1.60–2.80)
Southern CZ	*2.2.1 KTP*	*100*	*KGNP*	*Mills (1990)*	*Radio collar*	-	*1.70 (1.60–1.80)*
Southern CZ	*2.2.1 KTP*	*100*	*Dune Savanna*	*Funston, P.J. et. al. (2001)*	*Track survey 2000: 3480.6 km, 129 individuals, Track Frequency 27.6.*	*3.7*	*1.13*
Southern CZ	*2.2.1 KTP*	*100*	*Tree Savanna*	*Funston, P.J. et. al. (2001)*	*Track survey 2000: 6128 km, 433 individuals, Track Frequency 14.3.*	*7.1*	*2.17*
Southern CZ	*2.2.1 KTP*	*100*	*Tree and Dune combined*	*Funston, P.J. et. al. (2001)*	*Track survey 2000: 9608.6 km, 562 individuals, Track Frequency 5.85).*	*5.8*	*1.79*
Southern CZ	2.3.1 Central Kalahari Game Reserve Buffer	100	-	Keeping (2014)	Assume = KTP buffer.	-	2.30 (1.60–2.80)
Southern CZ	2.3.2 Western corridor	100	-	Keeping (2014)	Assume = KTP buffer.	-	2.30 (1.60–2.80)
Southern CZ	2.3.3 KTP buffer	100	KTP and KD1, KD2 combined	Keeping (2014)	Track survey with FMP formula, 634 km, CV 10%.	-	2.30 (1.60–2.80)
Southern CZ	*2.3.3 KTP buffer*	*100*	*Kgalagadi WMA Survey combined*	*Mudongo & Dipotso (2010)*	*Track survey 2010: 900 km, 86 individuals, Track Frequency 11.4.*	*9.56*	*2.93 (2.60–3.27)*
Southern CZ	*2.3.3 KTP buffer*	*100*	*Kgalagadi WMA KD 15*	*Mudongo & Dipotso (2010)*	*Track survey 2010: 300 km, 38 individuals, Track Frequency 9.1.*	*12.70*	*3.90 (3.16–4.64)*
Southern CZ	*2.3.3 KTP buffer*	*100*	*Kgalagadi WMA KD 12*	*Mudongo & Dipotso (2010)*	*Track survey 2010: 150 km, 16 individuals, Track Frequency 10.7.*	*10.70*	*3.28 (2.47–4.10)*
Southern CZ	*2.3.3 KTP buffer*	*100*	*Kgalagadi WMA KD 2*	*Mudongo & Dipotso (2010)*	*Track survey 2010: 350 km, 29 individuals, Track Frequency 12.5.*	*8.30*	*2.55 (2.15–2.94)*

Zone	Sub-stratum	Range (% of unit)	Location	Reference	Method and year	Mean track dens	Density
Southern CZ	*2.3.3 KTP buffer*	*100*	*Kgalagadi WMA KD 1*	*Mudongo & Dipotso (2010)*	*Track survey 2010: 300 km, 23 individuals, Track Frequency 14.3.*	*7.70*	*2.36 (1.91–2.82)*
Southern CZ	2.3.4 KTP buffer west	100	-	Keeping (2014)	Assume = KTP buffer.	-	2.30 (1.60–2.80)
Southern CZ	2.3.5 Eastern corridor 1	100	-	-	Assume high density.	-	0.58 (0.15–1.00)
Southern CZ	2.3.6 Eastern corridor 2	100	-	-	Assume high density.	-	0.58 (0.15–1.00)
Southern CZ	2.3.7 Eastern corridor 3	100	-	-	Assume high density.	-	0.58 (0.15–1.00)
Xaixai CZ	3.1.1 Xaixai Core	100	-	-	Assume high density.	-	0.58 (0.15–1.00)
Xaixai CZ	3.1.2 Xaixai Perifery	100	-	-	Assume high density.	-	0.58 (0.15–1.00)
Tuli CZ	4.1.0 Tuli GR	100	Mashatu, Northern Tuli Game Reserve	Snyman pers. comm.	Estimate 4 total.	-	0.31 (0.01–0.60)
Central Agric	5.1.0 Central Tuli farms	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Central Agric	5.2.0 Central Agric Molalatau	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Central Agric	5.3.0 Central Agric East	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Central Agric	5.4.0 Central Agric North	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Central Agric	5.5.0 Central Agric Pans	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Central Agric	5.6.0 Central Agric Mopipi	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Central Agric	5.7.0 Central Agric West	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Ngami Agric	6.1.0 Ngami East	100	-	-	Assume low density.	-	0.09 (0.07–0.10)
Ngami Agric	6.2.0 Ngami Thamalakane Boteti	69	-	-	Assume low density.	-	0.09 (0.07–0.10)
Ngami Agric	6.3.0 Ngami South	100	-	-	Assume low density.	-	0.09 (0.07–0.10)
Ngami Agric	6.4.0 Ngami Panhandle	51	-	-	Assume low density.	-	0.09 (0.07–0.10)
Ngami Agric	6.5.0 Ngami Tsodilo	100	-	-	Assume high density.	-	0.58 (0.15–1.00)
Ngami Agric	6.6.0 Ngami North West	100	-	-	Assume low density.	-	0.09 (0.07–0.10)
Ngami Agric	6.7.0 Ngami North East	0	-	-		-	0.00
Ghanzi Agric	7.1.0 Ghanzi farms	100	Surveys combined	Boast & Houser (2012)	Track survey 2007–2008: 3535 km, 257 individuals, Track frequency 13.76 ± 1.18 (8.6% CV)	7.27	2.23 (1.91–2.68)
Ghanzi Agric	*7.1.0 Ghanzi farms*	*100*	*East of Ghanzi (Game plus livestock)*	*Boast & Houser (2012)*	*Track survey 2007–2008: 1026 km, 81 individuals,*	*7.89*	*2.42 (1.91–3.30)*

Zone	Sub-stratum	Range (% of unit)	Location	Reference	Method and year	Mean track dens	Density
					Track frequency 12.67 ± 1.69 (CV = 13%)		
Ghanzi Agric	*7.1.0 Ghanzi farms*	*100*	*North West of Ghanzi: game*	*Boast & Houser (2012)*	*Track survey 2007–2008: 1268.8 km, 78 individuals, Track frequency 16.27 ± 2.60 (CV = 16%)*	*6.15*	*1.89 (1.44–2.75)*
Ghanzi Agric	*7.1.0 Ghanzi farms*	*100*	*South West of Ghanzi: livestock*	*Boast & Houser (2012)*	*Track survey 2007–2008: 1240 km, 98 individuals, Track frequency 12.65 ± 1.90 (CV = 15%)*	*7.9*	*2.42 (1.87–3.43)*
Ghanzi Agric	*7.1.0 Ghanzi farms*	*100*	*North West of Ghanzi: game*	*Kent (2011); Kent & Hill (2013)*	*Track survey 2008 -2009: 1023 km, 90 individuals, Track frequency 12.634 ± 0.297*	*8.795*	*2.70 (0.00–0.00)*
Ghanzi Agric	*7.1.0 Ghanzi farms*	*100*	*South West of Ghanzi: livestock*	*Kent (2011); Kent & Hill (2013)*	*Track survey 2008 -2009: 990 km, 100 individuals, Track frequency 9.970 ± 0.181*	*10.101*	*3.10 (0.00–0.00)*
Ghanzi Agric	*7.1.0 Ghanzi farms*	*100*	*South West of Ghanzi: livestock*	*Kent (2011); Kent & Hill (2013)*	*Camera trap 2009: 3187 camera trap days at 56 stations on three sites*	-	*2.30 (0.00–0.00)*
Ghanzi Agric	7.2.0 Ghanzi community	100	-		Assume high density.	-	0.58 (0.15–1.00)
Kgalagadi Agric	8.1.0 Kgalagadi Kweneng	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.2.0 Kgalagadi Dithopo	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.3.0 Kgalagadi Gaborone	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.4.1 Kgalagadi Kane 1	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.4.2 Kgalagadi Kane 2	100	-	Thorn *et al.* (2011)	Assume medium density.	-	0.13 (0.10–0.15)

Zone	**Sub-stratum**	**Range (% of unit)**	**Location**	**Reference**	**Method and year**	**Mean track dens**	**Density**
Kgalagadi Agric	8.5.0 Kgalagadi Molopo	100	-	Thorn *et al*. (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.6.0 Kgalagadi Tsabong	100	-	Thorn *et al*. (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.7.0 Kgalagadi Middelpits	100	-	Thorn *et al*. (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.8.0 Kgalagadi Bokpits	100	-	Thorn *et al*. (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.9.1 Kgalagadi Kang 1	100	-	Thorn *et al*. (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.9.2 Kgalagadi Kang 2	100	-	Thorn *et al*. (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.9.3 Kgalagadi Kang 3	100	-	Thorn *et al*. (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.10.1 Kgalagadi Hukuntsi 1	100	-	Thorn *et al*. (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.10.2 Kgalagadi Hukuntsi 2	100	-	Thorn *et al*. (2011)	Assume medium density.	-	0.13 (0.10–0.15)
Kgalagadi Agric	8.10.3 Kgalagadi Hukuntsi 3	100	-	Thorn *et al*. (2011)	Assume medium density.	-	0.13 (0.10–0.15)

CZ, Conservation Zone; GR, Game Reserve; CV, coefficient of variance; NP, National Park; SE, South East.

Data used to calculate population estimates are in normal type font and *additional data are in italic font*.

CHAPTER 6. WILDLIFE ABUNDANCE AND DIVERSITY AS INDICATORS OF TOURISM POTENTIAL IN NORTHERN BOTSWANA

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6.1 Abstract

Wildlife tourism can provide economic incentives for conservation. Due to the abundance of wildlife and the presence of charismatic species, some areas are better suited to wildlife tourism. Our first objective was to develop criteria based on wildlife abundance and diversity to evaluate tourism potential in the Northern Conservation Zone of Botswana. Secondly, we wanted to quantify and compare tourism experiences in areas with high and low tourism potential. We used aerial survey data to estimate wildlife biomass and diversity to determine tourism potential, while data from ground surveys quantified the tourist experience. Areas used for High Paying Low Volume tourism had significantly higher mean wildlife biomass and wildlife diversity than the areas avoided for this type of tourism. Only 22% of the Northern Conservation Zone has intermediate to high tourism potential. The areas with high tourism potential, as determined from the aerial survey data, provided tourists with significantly better wildlife sightings (ground surveys) than the low tourism potential areas. Even Low Paying tourism may not be economically viable in concessions that lack areas with intermediate to high tourism potential. The largest part of the Northern Conservation Zone has low tourism potential, but low tourism potential is not equal to low conservation value. Alternative conservation strategies should be developed to complement the economic incentive provided by wildlife-based tourism in Botswana.

6.2 Introduction

Nature and wildlife-based tourism provides benefits that compensate to some extent for conservation costs at local, national and global scales [1-3]. These costs can be high [4] and local communities are often burdened with most of the indirect conservation costs, including wildlife damage and restrictions on land-use and use of natural resources [1].

The majority of tourists to Africa are interested in seeing abundant wildlife with a strong focus on a few charismatic species like lion *Panthera leo*, leopard *P. pardus*, elephant *Loxodonta africana*, buffalo *Syncerus caffer* and rhinoceros (*Ceratototerium simum* and *Diceros bicornis*) [5-8]. Experienced wildlife viewers have a wider interest [3]. Due to the presence of charismatic species and good visibility, some areas are better suited to wildlife tourism than others [6] and can provide a better economic incentive for conservation.

Financial incentives for conservation are especially important in the case of Botswana where 38% of the country is set aside for conservation in the form of protected areas and wildlife management areas [9]. Tourism contributes directly and indirectly to Botswana's economy [10]: in 2011 the Trade, Hotels and Restaurants sector contributed approximately 15.0% to the Gross Domestic Product (GDP) [11] while tourism provided an estimated 10.6% of all jobs in Botswana [10].

Wildlife-based tourism in Botswana is primarily in the form of High Paying Low Volume (HPLV) tourism which is concentrated in the Okavango Delta and Chobe National Park [10], both located in the Northern Conservation Zone. The Government of Botswana lease exclusive use areas called concessions to operators. HPLV tourism is implemented through limiting the number and capacity of private lodges per concession; most lodges have a capacity of 16 to 24 beds. Low cost tourists are limited to the public camp sites in the national parks and game reserves while the HPLV tourists also use the private lodges [12]. Botswana phased out sport hunting [13], and is promoting photographic tourism to replace it. This raises the question about the potential to expand photographic tourism in the Northern Conservation Zone. The suitability and constraints for photographic tourism in this zone should be considered when reviewing recommendations to increase tourism capacity and to diversify tourism activities in Botswana [10]. The quality of game viewing will influence the willingness of wildlife-based tourists to pay for safaris [3, 14]. This in combination with the tourist capacity allocated and the annual rental required will determine the economic viability of high, medium and low budget tourism in different concessions of the Northern Conservation Zone. Increased numbers of tourist can have direct and indirect impacts on ecosystems and cultures of local people that may not be biologically or socially sustainable [15].

Our objectives were to quantify the differences in wildlife abundance and diversity between the subjectively selected sites for HPLV tourism lodges in northern Botswana and the areas with a perceived low tourism potential, and to develop criteria to evaluate the tourism potential based on wildlife abundance and diversity. We quantified and compared the tourism experiences in areas with high and low tourism potential and discuss the implications of the photographic tourism potential as an economic incentive for conservation in northern Botswana.

6.3 Methods

6.3.1 Study area

Botswana is located in southern Africa (Fig. 6-1) and has an area of approximately 582 000 km^2. The country is relatively flat with a mean altitude of 1000 m above sea-level.

Most of Botswana is arid to semi-arid, with the Kalahari occupying approximately 82% of the country. Rainfall is erratic and ranges from 250 mm per year in the southwest to over 650 mm in the north-east [9]. Over 90% of rainfall occurs in the summer months, between November and April. Apart from the Okavango Delta and the perennial Chobe/Linyanti river system, the only other surface water occurs in rivers and pans during the rainy season **[9]**.

Temperature ranges widely in Botswana. In Maun, located in northern Botswana, the mean maximum daily temperature is 35.5 °C in January and the mean minimum is 8.5 °C in July. The extreme minimum and maximum temperatures are -6 °C and 42 °C **[16]**.

Vegetation over most of the country is shrub and tree savanna of the Sandveld. The Hardveld vegetation types are associated with hills and rocky outcrops in the eastern part of Botswana. The Northern Conservation Zone has the wetland of the Okavango Delta, Sandveld, mopane dominated vegetation types and limited Miombo woodland in the north east. The Okavango Delta consists of a mosaic of islands, waterways and seasonal floodplains [9].

Conservation areas comprising of protected areas and wildlife management areas cover 38% of Botswana [9]. The Northern Conservation Zone is an area of 78911 km^2 and is part of the Kavango-Zambezi Trans Frontier Conservation Area (KAZA TFC). The Northern Conservation Zone includes concession areas, forest reserves, Chobe National Park, Moremi Game Reserve, Nxai Pan National Park and Makgadikgadi National Park (Fig. 6-1).

Some concession areas were designated exclusively for photographic safaris while most were multi-use (sport hunting and photographic safaris). The number and capacity of camps and lodges allowed per area were limited. The operators of multiple-use concessions subjectively zoned their areas for photographic and/or sport hunting use, and chose the locations of lodges. Photographic operators deemed parts of the Northern Conservation Zone not suitable for HPLV photographic tourism and thus only camps for sport hunting were established in those concessions.

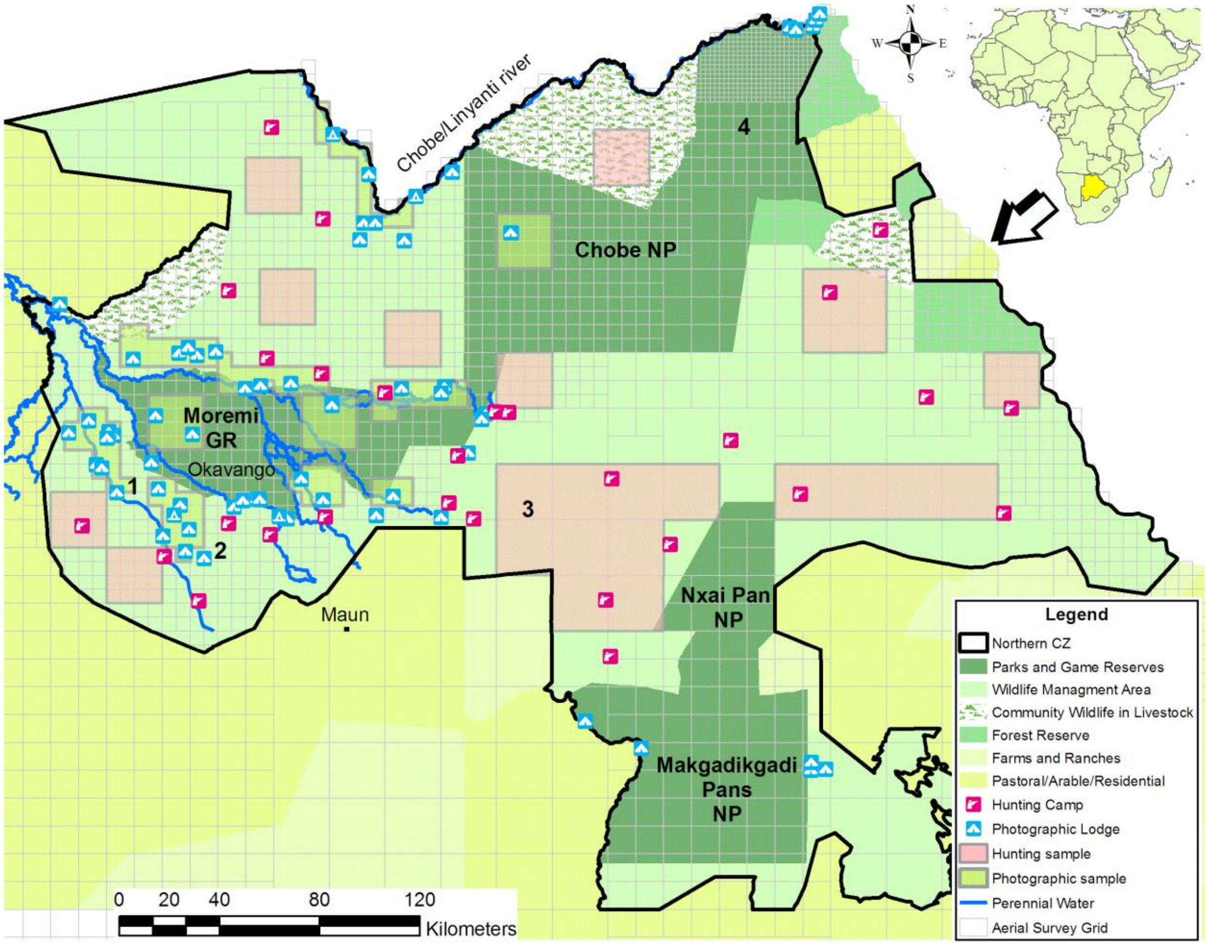

Figure 6-1 Land use and the locations of photographic and hunting camps in northern Botswana in 2005.

Sample areas to compare wildlife biomass and diversity between areas are indicated with a green overlay (High Paying Low Volume photographic tourism and red overlay (without HPLV photographic tourism, used for sport hunting). The four ground survey areas were: 1 = Macatoo, 2 = Xudum, 3 = NG/43, 4 = Nogatsaa.

The Northern Conservation Zone protects key populations of elephant, six of the large African carnivores (lion, leopard, African wild dog *Lycaon pictus*, cheetah *Acinonyx jubatus*, spotted hyena *Crocuta crocuta*, and brown hyaena *Hyaena brunnea*), rare antelope, and a variety of other herbivore species. For this study, we considered wildlife as mammals ranging in size from steenbok *Raphicerus campestris* to elephant. This encompassed the three categories that high budget tourists are the most interested in: the big-five, mammals and predators [3]

The quality and diversity of wildlife viewing in the Okavango Delta is a key feature for tourists [14]. The peak tourist season is from July to September, coinciding with the dry season concentration of wildlife along the perennial water sources of the Okavango Delta and Chobe/Linyanti river systems. These are also the two focal areas for the HPLV photographic tourism. Most (44.7%) tourists to Botswana stay for 1 – 3 three nights while 32.6% stay for 4 – 7 nights and 19.3% stay for 8 nights or longer [10]. HPLV tourists to private lodges in Botswana stay for 6 – 8 nights in total, visiting 2 – 3 lodge for 2 – 3 nights [12]. On average tourists are able to go on two game drives per day.

Ground transect data were collected as part of a monitoring program in four areas in the Northern Conservation Zone: two areas with high wildlife abundance and two with low wildlife abundance. The Xudum area of concession NG/29 (south-western Okavango Delta) and Macatoo area of concession NG/26 (western Okavango Delta) (Fig. 6-1) are examples of areas with HPLV tourism and high wildlife densities in seasonal floodplain systems with islands.

The Nogatsaa area in Chobe National Park is located approximately 70 km south of the Chobe River (Fig. 6-1). It is a typical dry woodland area that lacks natural perennial water sources. Surface water is limited to seasonal pans and eight artificial water points that are pumped during the dry season. The area is characterised by low wildlife densities during the dry season but is part of the wet season ranges of elephants and buffalo [17].

NG/43 is a dry woodland area southeast of Moremi Game Reserve (Fig. 6-1). It also lacks natural perennial surface water and has a lower long term average rainfall than the Nogatsaa area. Three artificial water points are pumped during the dry season. Wildlife densities are very low during the dry season. Elephants, buffalo, and zebra (*Equus burchellii*) move seasonally into and through the area [17].

Wildlife abundance and diversity in the Northern Conservation Zone

We used dry season aerial survey data from the Department of Wildlife and National Parks (DWNP) to quantify wildlife abundance and diversity per grid cell in the whole Northern Conservation Zone. Carnivores were excluded from the aerial survey data. The aerial survey data are presented in grid cell format in the Botswana Aerial Survey Information System (BASIS) of DWNP and cover the whole country. The grids vary in size according to the survey stratifications and the resulting spacing of the original aerial transects (1, 3, 6 or 12 minutes) and associated grids (Fig. 6-1) [18].

We calculated wildlife diversity (total number of herbivore species recorded) and mean wildlife biomass (Large Stock Units/ 100 square km) per grid cell for the dry period (1994, 1995, 1996 and 1999 surveys) and the wet period (2001, 2002, 2003, and 2006 surveys) as indicators of wildlife abundance in the Northern Conservation Zone. The dry period was characterized by below average rainfall and low flood levels. This represents conditions at the time that concession holders zoned their concessions and selected sites for camps and lodges. Rainfall and flood levels were higher during the subsequent wet period.

6.3.2 Wildlife abundance and diversity at sample sites

We compared wildlife abundance and diversity between the subjectively selected sites within the Northern Conservation Zone. The tourism operators selected lodge sites and avoided areas based on their perceived high or low tourism potential. We selected 11 sample sites consisting of 16 grid cells each to represent the main game drive areas used by the HPLV photographic lodges (Fig. 6-1). The sample sites were approximately 400 km², with one exception: the sample site on the Chobe riverfront, where the 16 grid cells covered only 44.44 km² due to the high sampling intensity of the aerial survey. We compared these sample sites with 11 sample sites around hunting camps in areas avoided by the photographic industry (non-HPLV areas) due to the perceived low photographic tourism potential. Similar to the HPLV sample sites we selected 16 grid cells with a total of 400 km² per sample sites. There were three exceptions due to lower sampling intensity of the aerial surveys in parts of the Northern Conservation Zone: one sample site had nine grid cells (900 km²); one sample site had four grid cells (1600 km²); and one sample site had nine grid cells (3600 km²) (Fig. 6-1).

We calculated the mean, standard deviation, and 95% Confidence Intervals of wildlife biomass and wildlife diversity for both HPLV and non-HPLV sample sites. One-way ANOVAs were used to compare wildlife biomass and diversity between HPLV and non-HPLV cells.

6.3.3 Tourism potential

We ranked all the grid cells into three classes, High, Intermediate and Low potential for HPLV tourism using criteria based on the 95% confidence intervals for wildlife biomass and species diversity at HPLV and non-HPLV sites. Tourism potential: 1 = low, 2 = intermediate, 3 = high. Cut off values derived from the 95% CI for wildlife biomass (18.8 -29.28; 2.47 - 6.35) and species (5.63 - 6.38; 1.21-1.64) at HPLV and non-HPLV sites respectively.

The same criteria were applied to the aerial survey data from the wet period (2001, 2002, 2003, and 2006) to assess the variability and potential changes in tourism potential as indicated by wildlife abundance and diversity under conditions of average to above average rainfall and increased flood duration and depth.

6.3.4 Tourism experience

We used herbivore transect count data that we collected during ground surveys to quantify and compare the tourism experience in areas with high and low wildlife abundance and diversity. Data were collected in the Xudum seasonal floodplain system in 1997, 1998, and 1999. We collected from Macatoo during 2011, after the subsequent decline of wildlife populations reported in the Okavango Delta [19]. The Nogatsaa and NG/43 data were collected in 1998 and 2011 respectively. Data collection included the cold dry, warm dry, and warm wet seasons.

Based on the length of stay for HPLV tourists in Botswana of 6 – 8 nights in total, visiting 2 – 3 lodges for 2 – 3 nights [12], we determined tourist experience for one game drive, four game drives, ten game drives, or more than ten game drives. The high HPLV tourism potential sites are Macatoo and Xudum, indicated in Fig. 6-1 by numbers 1 and 2 respectively. Numbers 3 and 4 represent the locations of NG/43 and Nogatsaa (Fig. 6-1). The counts were carried out with vehicles, boats, or horses. We recorded species, location and total number of animals within 200 m of the transect line for each observation. The analysis includes observations of herbivores from small antelope to elephant and ostriches (*Struthio camelus*). For each transect we calculated the number of observations per kilometre and number of species per kilometre. The number of observations refers to the number of times animals were seen and not to the total number of animals seen.

All statistics were performed with SPSS 16.0.0 and significance level was set at 0.05. We used an ANOVA to test the null hypothesis that the four sites had equal numbers of observations per kilometre. The data were fourth root transformed to meet the assumption of homoscedasticity (equality of variance), and Tukey HSD post hoc tests were done [20]. We also tested the null hypothesis that each site had equal numbers of species per kilometre. After the data were fourth root transformed, Macatoo, Xudum, and NG/43 data were normally distributed but were still heteroscedastic. Thus, the Welch t test and Dunnetts T3 post hoc test were performed [20].

For each species recorded, we determined the number of kilometres per observation. Random sub samples from the Nogatsaa and NG/43 data were included in the subsequent analysis to ensure that the total distance sampled per area were comparable.

We categorized species according to the number of game drives a tourist would need to take in order to observe each species. We assumed that a game drive lasts three hours at a mean speed of 10 km/h and covers a distance of 30 km. "Common" species were expected to be seen in one game drive, "regular" species were expected to be seen in two to four game drives, "uncommon" species were expected to be seen in five to ten game drives, and "rare" species were expected to be seen in more than ten game drives.

6.4 Results

6.4.1 Wildlife abundance, diversity and tourism potential

We found that the HPLV tourism sample sites in Northern Conservation Zone had a significantly higher mean wildlife biomass (Welch $F_{1, 258.213}$ =422.797, P=0.000) and wildlife diversity (Welch $F_{1, 833.190}$ =47.353, P=0.000) than non-HPLV tourism sample sites (Table 6-1) for the 1994, 1995, 1996 and 1999 aerial surveys. This excluded carnivores. As both wildlife diversity (Levene $F_{1,312}$=45.009, P=0.000) and wildlife biomass (Levene $F_{1,1234}$=54.988, P=0.000) were heteroscedastic, equality of means were tested with the robust Welch t test [20]. The wildlife tourism potential of each grid cell was ranked as low (1), intermediate (2) or high (3) following the criteria in Table 6-2. We derived the cut off values in Table 6-2 from the 95% CI for wildlife biomass (18.8 - 29.28; 2.47 - 6.35) and species (5.63 - 6.38; 1.21 - 1.64) at the HPLV and non-HPLV sites respectively (S1 File). Figure 2 shows the resulting tourism potential for the Northern Conservation Zone, which represents 14% of Botswana. Only 22% of the Northern Conservation Zone has intermediate to high potential for tourism while 78% has low tourism potential (Table 6-3). Twenty two concessions in Northern Conservation Zone included areas with high tourism potential while ten did not include high potential tourism areas (Fig. 6-2). There were small differences in the proportions and distribution of areas with low and intermediate to high tourism potential between the dry and wet period (Table 6-3 and Fig. 6-3).

6.4.2 Tourist experience

We analysed 224 transects covering 4,656.2 km from four sites (Table 6-4). We recorded the following species: buffalo, duiker (*Sylvicapra grimmia*), eland (*Taurotragus oryx*), elephant, oryx (*Oryx gazella*), giraffe (*Giraffa camelopardalis*), hippopotamus (*Hippopotamus amphibius*), impala

Table 6-1 Mean, standard deviation, and 95% Confidence Intervals for wildlife biomass (Large Stock Units/ 100 km^2) and wildlife diversity (number of species) for sample sites with HPLV[*] photographic tourism and without HPLV photographic tourism in the Northern Conservation Zone of Botswana.

Criteria	Cells	n	Mean	s	95% CI
Wildlife Biomass (LSU/ 100 km²)	No HPLV	576	4.41	23.71	2.47 - 6.35
	HPLV	660	24.04	68.75	18.8 - 29.28
Wildlife Diversity (number of species)	No HPLV	150	1.43	1.36	1.21 - 1.64
	HPLV	164	6.01	2.47	5.63 - 6.38

Analysis is based on aerial survey data from 1994, 1995, 1996, and 1999.
[*] High Paying Low Volume

Table 6-2 Ranking criteria based on the wildlife biomass and diversity to evaluate tourism potential of each grid cell in the Northern Conservation Zone of Botswana.

	Wildlife Biomass (Large Stock Units/ 100 km^2)		
Wildlife Diversity	≤ 6.35	>6.35 - <18.8	≥ 18.8
≤ 2 species	1	1	2
3 – 5 species	1	2	3
≥ 6 species	2	3	3

Tourism potential: 1 = low, 2 = intermediate, 3 = high. Cut off values derived from the 95% CI for wildlife biomass (18.8 - 29.28; 2.47 - 6.35) and species (5.63 - 6.38; 1.21 - 1.64) at HPLV and non-HPLV sites respectively.

Table 6-3 The size of areas in the Northern Conservation Zone of Botswana with low, intermediate and high tourism potential based on aerial survey data for the dry period (1994, 1995, 1996 and 1999 surveys) and the wet period (2001, 2002, 2003, and 2006 surveys) .

Tourism Potential	1994 – 1999		2001 - 2007	
	Area (km^2)	Area (%)	Area (km^2)	Area (%)
No data available	-	-	876	1
Low (1)	61769	78	59146	75
Intermediate (2)	8574	11	11374	14
High (3)	8568	11	7514	10
Total	78911	100	78911	100

Table 6-4 The number and length of transects conducted at two HPLV sites and two non-HPLV sites in the Northern Conservation Zone of Botswana, showing the mode of transport used to collect data.

Site	Type	Transport	Transects (n)	Length (km)
Macatoo (2011)	HPLV	Boat	9	102.6
Macatoo (2011)	HPLV	Horse	13	227.7
Macatoo (2011)	HPLV	Vehicle	16	141.3
Xudum (1997 – 1999)	HPLV	Vehicle	59	481.1
NG/43 (2011)	Non-HPLV	Vehicle	40	928.2
Nogatsaa (1997)	Non-HPLV	Vehicle	87	2775.3
Total			224	4656.2

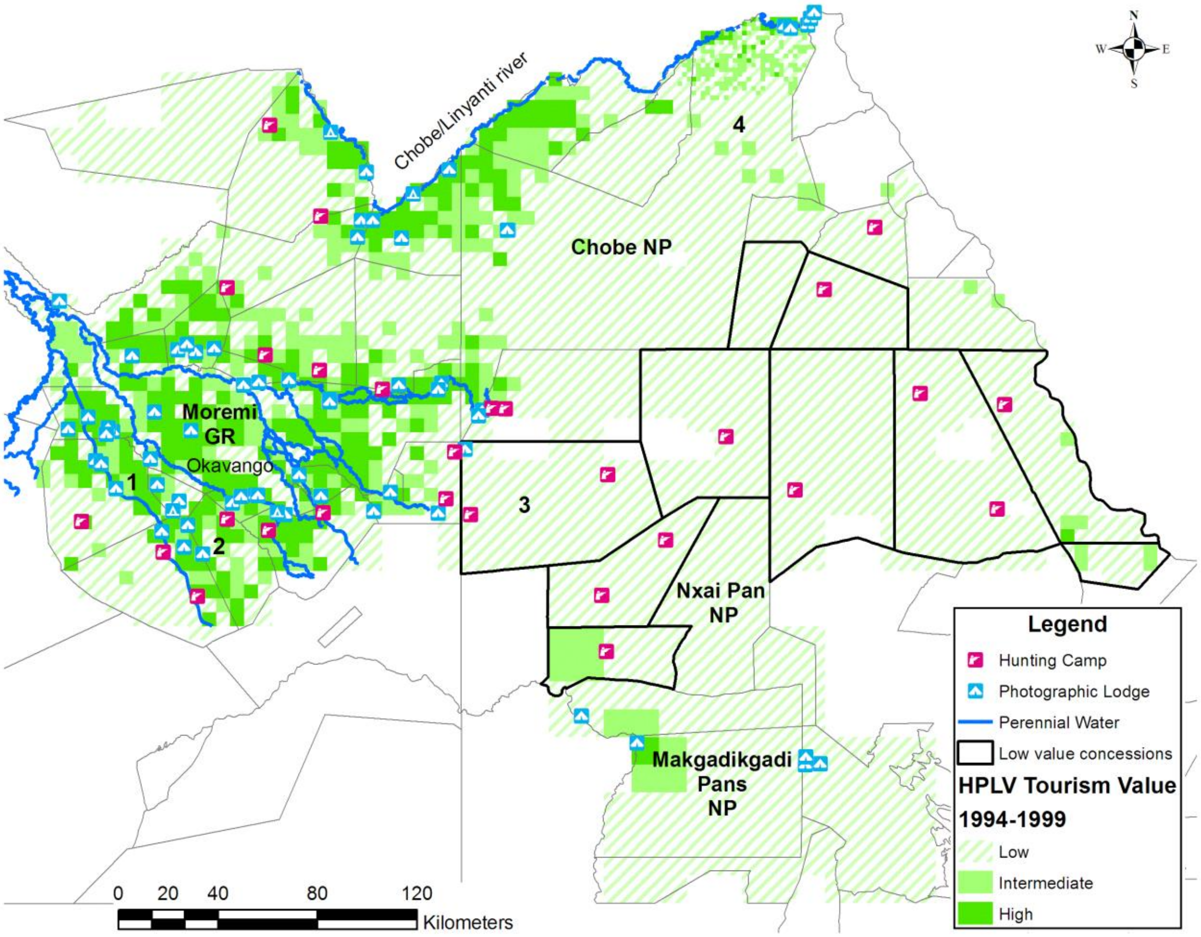

Figure 6-2 The potential for High Paying Low Volume tourism in the Northern Conservation Zone of Botswana based on wildlife biomass and diversity from 1994 to 1999.

The concessions without high tourism potential areas are demarcated by a black boundary. The wildlife biomass and diversity were calculated from the Department of Wildlife and National Parks dry season aerial surveys in 1994, 1995, 1996 and 1999.

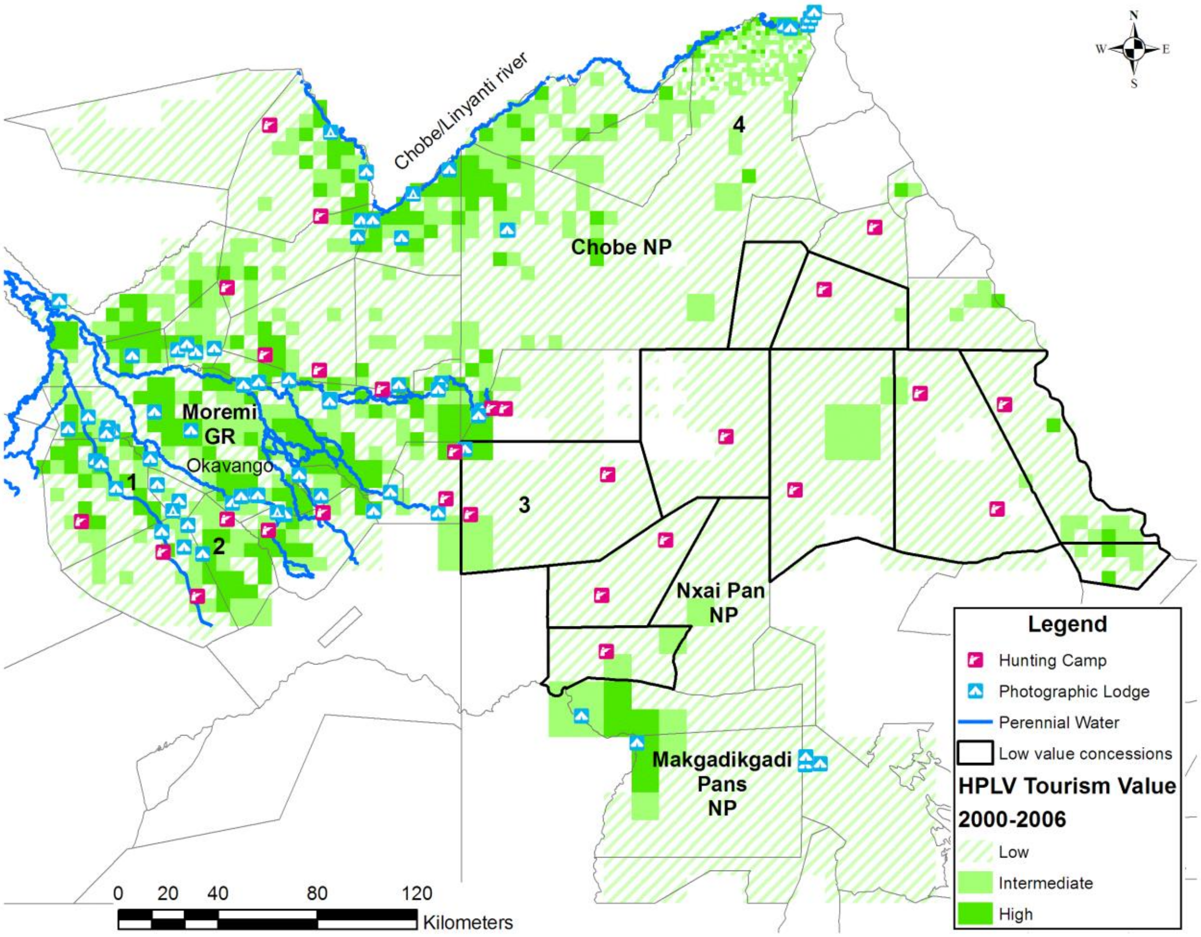

Figure 6-3 The potential for High Paying Low Volume tourism in the Northern Conservation Zone of Botswana based on wildlife biomass and diversity from 2000 to 2006.

The concessions without high tourism potential areas are demarcated by a black boundary. The wildlife biomass and diversity were calculated from the Department of Wildlife and National Parks dry season aerial surveys in 2001, 2002, 2003 and 2006.

(*Aepyceros melampus*), greater kudu (*Tragelaphus strepsiceros*), lechwe (*Kobus leche*), ostrich, reedbuck (*Redunca arundinum*), roan (*Hippotragus equinus*), sable (*Hippotragus niger*), steenbok (*Raphicerus campestris*), tsessebe (*Damaliscus lunatus*), warthog (*Phacochoerus africanus*), waterbuck (*Kobus ellipsiprymnus*), wildebeest (*Connochaetes taurinus*) and zebra.

The mean observations per kilometre and mean species per kilometre had normal distributions after being fourth root transformed. The mean observations per kilometre met the assumption of homoscedasticity (Levene $F_{3,220}$=1.799, P=0.148) but mean species per kilometre were heteroscedastic (Levene $F_{3,220}$=5.282, P=0.002).

The results of the one-way ANOVA showed a significant difference between mean observations per kilometre (fourth root transformed) for the sites ($F_{3,220}$=66.650, P<0.001). Tukey post hoc tests (α = 0.05) showed that Xudum and Macatoo were significantly different from each other and from NG/43 and Nogatsaa, but NG/43 and Nogatsaa were not significantly different from each other. The Welch t test showed significant difference between mean number of species per kilometre (fourth root transformed) for the sites (Welch $F_{3,\ 89.072}$ =98.579, P<0.001). The two non-HPLV sites, NG43 and Nogatsaa, had significantly lower mean number of species per kilometre than the HPLV sites, but did not differ significantly from each other (Dunnett T3 α = 0.05).

The Macatoo and Xudum sites (high tourism potential) had ten to twelve species seen commonly and regularly, more than the five to six species seen at the NG/43 and Nogatsaa sites (low tourism potential) (Fig. 6-4 and Table 6-5). A tourist on a typical game drive of 3 hours would expect to have 33 wildlife sightings in the Xudum area, one on average every 6 minutes. In contrast a typical game drive in NG/43 should result in 6 wildlife sightings, one every 30 minutes.

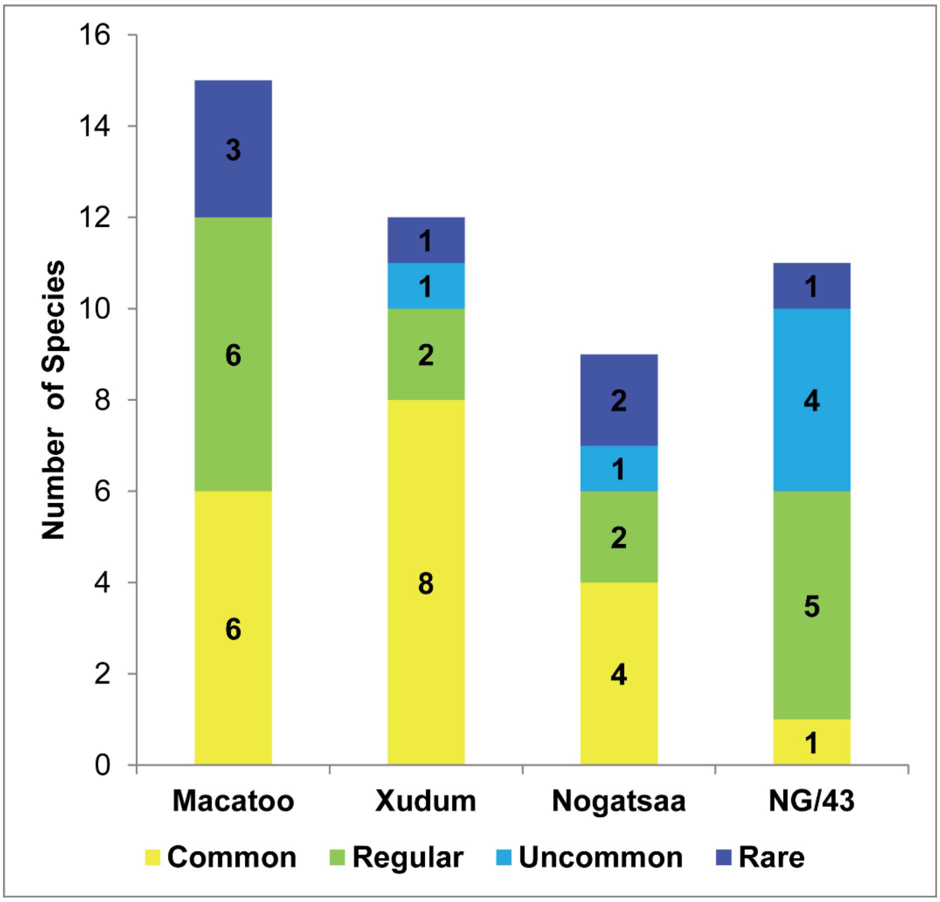

Figure 6-4 The number of species regarded as common, regular, uncommon and rare at four study sites.

Expect to see common species in one game drive, regular species in two to four game drives, uncommon species in five to ten game drives and rare species in more than ten game drives.

Table 6-5 The classification of wildlife species as common, regular, uncommon and rare at four study sites in the Northern Conservation Zone of Botswana and the mean sighting frequency (kilometres/ observation) for each species.

	Macatoo		**Xudum**		**Nogatsaa**		**NG/43**	
	Species	**km/obs**	**Species**	**km/obs**	**Species**	**km/obs**	**Species**	**km/obs**
Common	Impala	4.0	Tsessebe	3.4	Elephant	13.0	Steenbok	11.7
(1 game drive)	Elephant	12.1	Impala	4.1	Giraffe	20.3		
	Kudu	15.2	Zebra	7.8	Impala	21.1		
	Giraffe	15.7	Giraffe	8.8	Steenbok	23.1		
	Lechwe	15.7	Wildebeest	11.4				
	Tsessebe	18.9	Lechwe	14.9				
			Kudu	15.4				
			Warthog	24.6				
Regular	Warthog	39.3	Steenbok	44.7	Warthog	56.3	Giraffe	43.7
(2 – 4 game drives)	Zebra	39.3	Reedbuck	61.4	Kudu	101.4	Elephant	47.7
	Buffalo	42.9					Gemsbok	47.7
	Hippopotamus	42.9					Impala	74.9
	Steenbok	42.9					Zebra	87.4
	Reedbuck	78.6						
Uncommon	(none)		Waterbuck	245.8	Sable	253.6	Kudu	131.1
(5 – 10 game drives)							Duiker	131.1
							Eland	174.8
							Buffalo	262.2
Rare	Duiker	471.6	Duiker	491.5	Ostrich	507.1	Warthog	524.4
(>10 game drives)	Ostrich	471.6			Roan	507.1		
	Wildebeest	471.6						

A game drive is assumed to last three hours and cover up to 30 km.

6.5 Discussion

Applying our tourism potential criteria to the Northern Conservation Zone shows that limited areas have high tourism potential. The data also shows the operators of HPLV lodges have already subjectively selected sites with high tourism potential and avoided areas with low potential. The high potential tourism areas were associated with the perennial water sources of the Okavango Delta, the Kwando/ Linyanti/ Chobe system and the perennial waterholes of the Boteti River in Makgadikgadi National Park. The low tourism potential areas lacked natural perennial water.

Tourism experience as measured by the number of observations and diversity of animals a tourist could expect on a game drive differed significantly between high and low suitability areas. Xudum and Macatoo, which both have high photographic tourism potential, offered tourists an experience with many observations that included a diverse range of species. In contrast, the low potential areas offered very low diversity and frequency of wildlife sightings.

The low potential areas also lacked the charismatic species important to tourists. Elephants and buffalo are present during the wet season but move to the perennial water sources during the dry season [17], which coincides with the peak tourist season. Appropriate data on large carnivores were not available for the analysis, but the expected densities of lion, leopard and spotted hyaena are positively correlated with prey biomass [21], and higher lion densities were recorded in the part of the Northern Conservation Zone where more prey was available [22]. Tourists are more likely to see lion and leopard in the high tourism potential areas than the low potential areas. The following unpublished data (CWW) support this: Lion density in the Okavango Delta is between 5 and 20 times higher than at site 3; Leopard density at site 3 is between 50% and 75% of the density at high tourism potential areas in the Okavango Delta. Two subordinate competitors, cheetah and wild dog, may use areas with low lion and hyaena densities as refuges [23]. Although these two species are expected to occur at higher densities in the low tourism potential areas than in the high potential areas, low visibility due to dense vegetation [9, 24] limit all wildlife sightings in the low potential areas.

Although “modified high volume – mixed price” tourism has been promoted since the implementation of the Botswana Tourism Master Plan in 2000 [25], tourists to Botswana remain concentrated in Chobe National Park and the Okavango Delta and photographic tourism has not been diversified into other areas [10]. The photographic operators have not been interested in developing lodges in the concessions that lack high tourism potential. The financial viability of the only exception depends on access to Moremi Game Reserve (pers. comm.*). In personal communications (CWW) the other nine concessionaires indicated that they could not find photographic operators willing to develop photographic tourism in these concessions to complement their sport hunting safaris. All ten of these concessions were conducting sport hunting and the decision by the government to phase out sport hunting will have the biggest negative economic impact on these areas.

Mladenov *et al.* [14] showed tourists rated the quality of wildlife viewing in the Okavango Delta very high and 71% of them were not willing to pay the same price for diminished wildlife viewing elsewhere. Due to the expectations of high paying tourists, it is not a viable option to divert them to low tourism potential areas. Targeting experienced tourists with wider interests than just the charismatic species, and low budget tourists is an option. Lindsey *et al.*[3] found that low-budget tourists in South Africa were more interested in birds, plants and scenery in contrast to the focus of high budget tourists on the big-five, mammals and predators.

Most of the low tourism potential areas are characterised by large sections of monospecific dense woodland with low visibility and flat terrain lacking focal features [9, 24]. The lack of landscape variation combined with the low wildlife numbers and diversity may even be a limiting factor for budget tourism. Low budget tourists rarely used the Nogatsaa area in Chobe National Park during the two years of field work in the area (pers. obs.).

We are concerned that concessions with only low tourism potential area have very limited potential to sustain even budget photographic tourists. The Government of Botswana may consider rebates on land rental to entice tourism operators to take the risk to develop new and traditional tourism product in the areas with low tourism potential.

A lack of an economic incentive in the medium to long term may result in pressure to convert the “unproductive area” to non-wildlife land use such as livestock production. Livestock production has a high priority in Botswana: “This deliberate decision was taken to increase our national herd from 2.5 to 3.5 million…….” (Minister of Agriculture, C. De Graaff as quoted in the Ngami Times July 18 – 25, 2008). Mineral prospecting is taking place in some of these concessions and may impact future land use [24].

Despite the low tourism potential, these concessions have a very high conservation value and form a key area in the Northern Conservation Zone and the larger KAZA TFC [26]. The block of ten concessions forms the only link between the Okavango Delta, Chobe National Park, Nxai Pan/ Makgadikgadi National Park in Botswana and Hwange National Park (to the east) in Zimbabwe. It is critical wet season habitat for the largest remaining elephant population in Africa [19] and a large buffalo population. One of the longest zebra migrations in Africa moves through part of this area [27]. With the low lion density [22] it is also an important refuge area for wild dog and cheetah.

Large areas with a mosaic of low and high prey densities are important for the conservation of the African large carnivore guild [28]. The Northern Conservation Zone is such an area and includes significant numbers of lion, leopard, spotted hyaena, cheetah and wild dog [22, 29, 30]. The northern edge of brown hyaena range falls within the Northern Conservation Zone [31]. Losing part of the ten low tourism potential concessions will impact negatively on the conservation of the large carnivore guild in the KAZA TFC including carnivore species classified by the International Union for Conservation of Nature as Endangered (African wild dog), Vulnerable (cheetah and lion) and Lower Risk, Near Threatened (leopard and brown hyaena)[32].

The Government of Botswana has a good conservation track record and there is no immediate threat to these areas. However, pressure from Botswana's livestock industry for access to areas with limited tourism is likely to increase in the medium to long term to fulfil their aspirations to grow their industry. In the meantime, alternative conservation strategies should be developed to complement the economic incentive provided by wildlife-based tourism in Botswana.

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CHAPTER 7. SUITABILITY OF THE PULP-TOOTH RATIO TO ESTIMATE THE AGE OF TROPHY-HUNTED AFRICAN LIONS IN BOTSWANA

Christiaan W. Winterbach, Hanlie E. K. Winterbach, Debbie Peake, Michael J. Somers

7.1 Abstract

Several African countries implemented a minimum age threshold strategy to manage trophy hunting of male African lions (*Panthera leo*) at sustainable levels. Thus, the professional hunter must decide if an animal is above the minimum threshold age based on field aging criteria before the lion can be hunted. I evaluated the suitability of six different tooth:pulp width and area ratios to estimate animal age of trophy-hunted male lions in Botswana as a *post hoc* means to monitor compliance to a 6-years minimum-age threshold. I used digital radiographs of 42 single-rooted second upper premolars of lions hunted between 2005 and 2007, and cementum annuli analysis as reference ages for evaluation. The linear regression y= 0.990 x + 2.512 between the tooth root area:root pulp cavity area ratio (R:RPA) and cementum annuli analysis ages < 10 years ($F_{1, 23}$ = 42.244, P < 0.001) provided the best correlation (r^2 = 0.647). The 95% confidence intervals for the age estimates from R:RPA ratios in the cementum annuli analysis age classes 4-5 years and 6-7 years were mutually exclusive, showing the methods ability to distinguish between animals older and younger than 6 years old.

7.2 Introduction

Trophy hunting is of major importance in a number of African countries as it creates economic incentives for conservation over vast areas (Lindsey, Roulet & Romañach, 2007). Lions (*Panthera leo*) are one of the high-value species in the trophy hunting industry (Lindsey *et al*., 2007). However, there is increasing concern about the potential negative impact of trophy harvesting on the long-term survival of hunted lion populations (Croes *et al*., 2011; Loveridge, Searle, Murindagomo & Macdonald, 2007; Packer *et al*., 2010). Consequently, implementation of sustainable harvest strategies for lion populations in Africa is now vital (Becker *et al*., 2013; Edwards, Bunnefeld, Balme & Milner-Gulland, 2014; Packer *et al*., 2009).

Whitman *et al*. (2004) found trophy hunting of male African lions can be managed at a sustainable level if only males above a minimum age threshold of 5-years old are targeted, and several companies already implement a 6-years old minimum threshold (Anonymous, 2013). Professional hunters are only able to evaluate the age of male lions in the field by means of five physical characteristics: mane, tooth wear, leg markings, nose colour and facial characteristics; field guidelines have been provided to aid their evaluation of age before it is hunted (Whitman, 2010). Regrettably, managers are only able to monitor hunters' compliance to the age threshold strategy after the lion has been shot. Nonetheless, information on the age of hunted lions can be used to inform future hunting quotas to ensure sustainability. They therefore need the best tools to assess the ages of already hunted lions in order to manage them effectively.

Two tooth characteristics: the deposition of cementum annuli, and the closure rate of the pulp cavity have been used as a proxy for age in archaeology (Jones, 2012), vertebrate palaeontology (Binder & Van Valkenburgh, 2010), forensic science (Paewinsky, Pfeiffer & Brinkmann, 2005), and wildlife studies (Hess, Stephens, Thompson, Danner & Kawakami, 2011; Smuts, Anderson & Austin, 1978; Reig, Daniels & Macdonald, 2001; Van Aarde, 1983). Cementum annuli analysis (CAA) involves counting the number of growth layers (annuli) deposited in the cementum of the tooth that correlates with seasonal changes in nutrition (Asmus & Weckerly, 2011; Lieberman, 1994). Validation studies found CAA age estimates to be within ±1 year of actual age for ≥80% of the samples examined (Divljan, Parry-Jones & Wardle, 2006; Hamlin, Pac, Sime, DeSimone & Dusek, 2000; Harshyne, Diefenbach, Alt & Matson, 1998; Moffit, 1998; Rolandsen *et al*., 2008). The same level of accuracy was obtained in gray wolves (*Canis lupus*) (Gipson, Ballard, Nowak & Mech, 2000; Goodwin & Ballard, 1985) and in lynx (*Lynx l. lynx*) (Kvam, 1984). Annual increments of cementum annuli were observed in the Iriomote cat (*Prionailurus bengalensis iriomotensis*) (Nakanishi, Ichinose, Higa & Izawa, 2009), feral cats in Marion Island (Van Aarde, 1983) and Hawaii (Danner, Farmer, Hess, Stephens & Banko, 2010), and in lions (Smuts *et al*., 1978; Cheater, 2006). The CAA method was successfully used in estimating the age of felids both in the northern and the southern hemispheres. In the northern hemisphere, CAA was used for wildcat (*Felis silvestris*) (Reig *et al*., 2001; Krüger, Hertwig, Jetschke & Fischer, 2009), cougar (*Puma concolor*) (Cooley, Wielgus, Koehler & Maletzke, 2009; Robinson, Wielgus, Cooley & Cooley, 2008), Eurasian lynx (*L. lynx*) (Nilsen, Brøseth, Odden & Linnell, 2010), and in the southern hemisphere on cheetah (*Acinonyx jubatus*) in Namibia (Marker *et al*., 2008).

The pulp cavity closure rate is an alternative to CAA for aging animals. The pulp cavity becomes progressively smaller with age due to deposition of physiological secondary dentine along the wall of the pulp chamber throughout the life of the tooth. This incremental growth of dentine is a function of ontogeny (Morse, Esposito, Schoor, Williams & Furst, 1991), and Smuts *et al*.(1978) found incremetal deposition of dentine ceases only when the dental pulp is exposed and necrosis begins. The closure rate expressed by a pulp cavity:tooth ratio was found suitable to separate animals into age classes for gray wolves (Landon, Waite, Peterson & Mech, 1998), dingoes (Thomson & Rose, 1992), coyotes (*Canis* *latrans*) (Knowlton & Whittemore, 2001), wild dogs (*Lycaon pictus*) (Kershaw, Allen, Lisle & Withers, 2005), and lions (Smuts *et al*., 1978; Cheater, 2006; Meachen-Samuels & Binder, 2010).

Both methods have limitations in their accuracy, but factual comparison between studies is hampered mainly by differences in methodologies. Accuracy of the CAA depends on factors such as tooth type used, the type of sectioning and staining, sample size, and interpretation of annuli (Gasaway, Harkness & Rausch, 1978; Goodwin & Ballard, 1985; Hess *et al*., 2011; Landon *et al*., 1998; Medill, Derocher, Stirling, Lunn & Moses, 2009; White *et al*., 2016). In some cases the pattern of annuli deposition may differ between upper and lower jaw teeth (Kershaw *et al*., 2005; Thomson & Rose, 1992), between the right and left teeth of the same jaw (Danner *et al*., 2010; Asmus & Weckerly, 2011) or between areas of the same tooth (Medill *et al*., 2009; Landon *et al*., 1998). In addition, CAA is a laborious, complicated, and expensive method that requires trained and experienced interpreters (Calvert & Ramsay, 1998; Rolandsen *et al*., 2008) to identify false- , split- and/or compound-annuli which otherwise may lead to erroneous counts (Rice, 1980). Bingham & Purchase (2003) and Thomson & Rose (1992) found CAA unsuitable to estimate age for two Canid species; jackals (*Canis adustus* and *C. mesomelas*) and dingoes (*C. lupus dingo*), respectively, as regular cementum annuli could not be identified by the researchers. The deposition of cementum annuli is also influenced by environmental factors such as physiology and behaviour of the animal, climate conditions, and pattern of food availability the sample animals were exposed to. In white-tailed deer, *Odocoileus virginianus*, populations occurring at low densities with access to high food quality throughout the year cementum annuli in teeth were indistinguishable (McCullough, 1996). Lam (2008) attributed distinct but irregularly deposited annuli in springbok (*Antidorcas marsupialis*) from the South African Cape to be a result of their adaptable behaviour to arid environments that prevents them from experiencing distinct annual growth and non-growth periods.

In contrast to the CAA, calculating the pulp cavity as a proportion of the tooth using radiographs is fast, easy to perform, and less expensive. Differences in pulp cavity:tooth width ratio may occur between upper and lower jaw teeth (Knowlton & Whittemore, 2001) and between males and females (Kershaw *et al*., 2005). In both the CAA and pulp cavity:tooth width ratio methods accuracy decreased with increasing age of the animal (CAA: Gipson *et al*., 2000; Rolandsen *et al*., 2008; pulp cavity ratio: Thomson & Rose, 1992; Landon *et al*., 1998).

Currently, there is a lack of known-age reference material from free-ranging lions, and building up a reference database of known-age wild lions per country will be difficult and take an immense amount of time. In the light of the current pressures on lion populations across Africa, managers do not have the luxury to wait for these data in order to control the trophy hunting of lions. In this paper, I use the CAA method as the best alternative age reference currently available to evaluate whether the pulp cavity:tooth ratio can be used as an index of age for lions. Smuts *et al*. (1978) and Cheater (2006) showed annual increases in cementum annuli for lions in southern Africa. I accepted that our samples from Botswana are part of this population with annual increments of cementum annuli, and the CAA method should therefore provide good reference ages. I also improve the measurement of the pulp cavity:tooth ratio from other studies for any species by eliminating the potential effect of crown wear and the irregular shape of the crown pulp chamber on the ratio.

7.3 Methods

I collected fifty upper second premolar (PM²) teeth from 69 skulls of male lions trophy-hunted in Botswana between 2005 and 2007. I collected the teeth and measured the skulls as part of preparing the trophies for export by Mochaba Enterprises. Based on geographic location, I classed each trophy as collected in a low (< 2 animals/100 km²) or high (> 4 animals/100 km²) density lion population. All skulls were of unknown age and collected teeth were a mix of the left and right jaws. Of these, I deemed 42 single-rooted teeth (84%) as suitable to use in this study. I examined each tooth visually to asses tooth wear on a broad 4-point scale: 0=no wear (crown tip and both cusp ridges sharp), 1=slight wear (at least one cusp ridge sharp), 2=wear (crown tip and both cusp ridges rounded), and 3= heavy wear (crown tip flat and lowest cusp worn away). I measured the dimensions of each tooth (length, crown height, crown length and crown width) to the nearest 0.1 mm with a manual micrometer. I then digitally photographed the tooth using a D200 Nikon camera fitted with a 55 mm macro lens.

Each tooth was digitally radiographed by the Zurron Dental Clinic, Gaborone, using a Trophy RVG IRIX set at 0.10 t / s. I used the tooth length and crown width measurements of each tooth to adjust the radiographed image in ARCView Version 3.2 to scale, and verified the position of the enamel-cementum junction on the radiographs from the digital photos.

From the correctly scaled images I digitized lines to measure the widths of the pulp cavity and tooth at four positions (Fig. 7-1): 1) Width ratio A: at the enamel-cementum junction (Paewinsky *et al*., 2005), 2) Width ratio B: 3 mm below and parallel to the enamel-cementum junction (Knowlton & Whittemore, 2001; Kershaw *et al*., 2005), 3) Width ratio C: at the midpoint of and perpendicular to the root pulp cavity length, and 4) Width ratio D: at the midpoint between Width ratio C and the tip

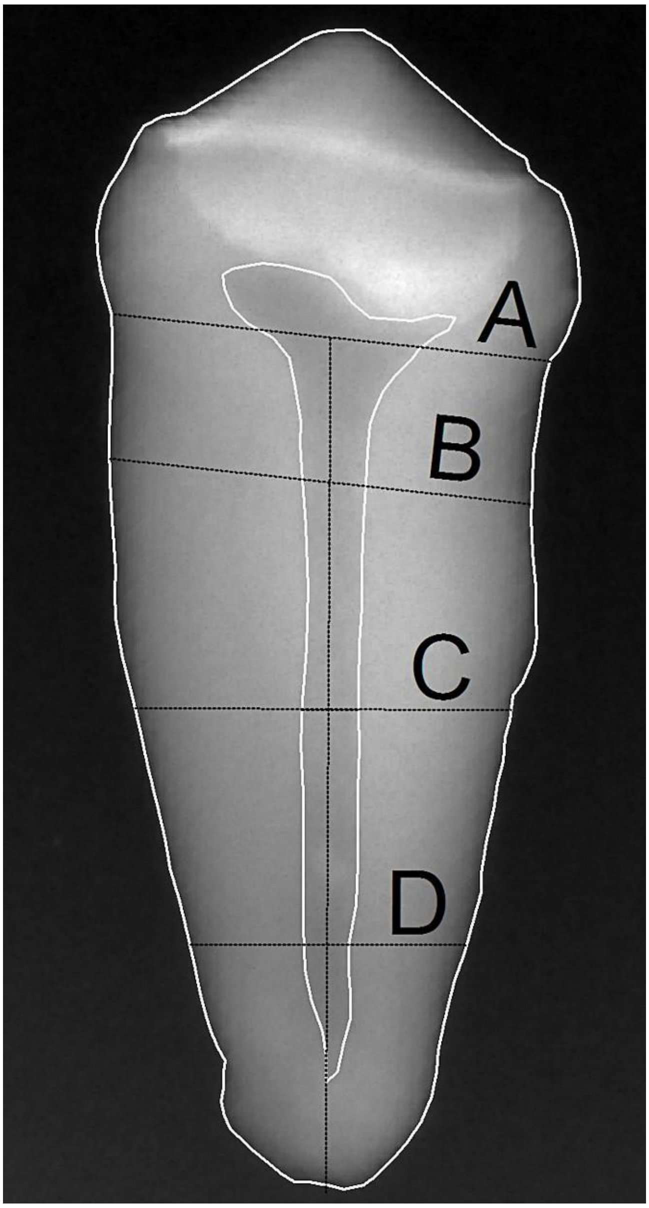

Figure 7-1 Digital radiograph of a second premolar teeth of a lion (*Panthera leo*) showing the digitized tooth and pulp cavity outlines to calculate tooth area and pulp area. A, B, C and D indicate the four positions where tooth width and pulp width were measured to calculate pulp:tooth width ratios.

of the tooth root. I measured the root pulp cavity length as a straight line from the midpoint of the enamel-cementum junction to the furthest tip of the pulp cavity, and the tooth root length as a vertical line from the midpoint of the enamel-cementum junction to the apex of the tooth. I calculated the ratios as pulp cavity to tooth width, as suggested by Kershaw *et al*. (2005).

The pulp cavity – and tooth areas were measured in two ways: 1) by outlining the entire tooth pulp cavity- and tooth areas (Cheater, 2006) and, 2) by outlining the root pulp cavity - and tooth root areas from the line 3 mm below Width ratio B and perpendicular to the enamel-cementum junction (Fig. 7-1) using ARCView. For comparison, the area ratios were calculated as tooth pulp cavity:tooth area and as root pulp cavity:root area.

The CAA was done by Matsons Laboratory LLC, USA, using the paraffin method. Teeth were decalcified with acid, rinsed with water, dehydrated in alcohol, cleared in toluene, and then treated and embedded in melted paraffin. Teeth were sectioned at 14µm using a rotary microtome, the sections mounted on microscope slides, deparaffinized, then hydrated and dried. Slides were stained with Giemsa blood stain (Ricca Chemical Company, Arlington, Texas, USA) and coverslips affixed by resin. Sections were examined microscopically using a Leica compound with transmitted light microscope at 40 x, 60 x, 100 x, and 160 x magnifications. Cementum annuli were counted using a standardized analysis model that is specific for each species and tooth type. A certainty code was given for each section analyzed: A = good reliability of age given, B = good reliability that age falls within range given, C = error is likely. The CAA ages are considered only as reasonable estimates (G. Matson, Matsons Laboratory, personal communication).

I tested for monotonic relationships between two variables using Spearman's rank order correlation (Quinn & Keough, 2002) with the CAA age and one of the width or area ratios. For significant monotonic relationships ($p < 0.01$) I fitted linear, logarithmic, exponential and logistic models using CAA age as the independent variable and the ratio as the single dependent variable. I selected the best models based on the r square value. The analysis was done on the full data set and a subset with CAA ages of < 10 years.

7.4 Results

In the CAA analysis 67% of the teeth were given an A certainty code and 33% a B code. The dark-staining cementum layer can be detected only after the next light-staining layer started forming (G. Matson, Matsons Laboratory, personal communication). Therefore, based on the assumptions that the earliest age of cementum deposition in lions is 18 months (Crowe, 1972; Kvam, 1984; Smuts *et al*.,

1978) and in Botswana the dense layer corresponds with a lean season of 6 months (June to November) then a lion aged as 5 years old by the CAA can in fact be between 5.1 years and 6.5 years, and a lion aged as 6 years can be between 6.1 years and 7.5 years old.

Some teeth were not usable for the linear and area measurements and were thus excluded from the relevant samples. Of the six variables I tested, only the Width ratio A (Pulp:tooth width ratio at enamel-cementum junction) did not have a significant monotonic relation to CAA age (Table 7-1). I excluded Width ratio A from further analysis and fitted curves to the five variables with significant Spearman's rank order correlation with CAA ages.

The linear, logarithmic, exponential and logistic curves fitted with CAA age as the independent variable and one of Width ratios B, C and D were all significant at $p < 0.05$ or higher (Table 7-2). A higher percentage of variation was explained for all models for the subset of data using only teeth with CAA ages < 10 years old, as evident from the higher r^2 values (Table 7-2). The best r^2 value ($r^2 = 0.554$) for the pulp cavity:tooth width ratio, using the all-age sample of teeth, was produced with the logarithmic curve fitted to ratio calculations measured at Width ratio D (Table 7-2). The logarithmic equation also provided the best fit ($r^2 = 0.593$) for Width ratio D with CAA age < 10 years (Table 7-2). The formula derived was: $y = -0.095\ln(x) + 0.294$ (Equation A), where y is the ratio of the pulp cavity width and the tooth width at Width ratio D and x is the CAA age in years.

Curves fitted to the area ratios and CAA ages (Table 7-3) explained more variation than the width ratios B and C (Table 7-2). The linear regression $y = 0.990 x + 2.512$ where x is CAA age < 10 years and y is the R:RPA ratio, explained 64.7% of the variation (Table 7-3). This translated to *age estimate= (R:RPA ratio – 2.512) / 0.990* (Equation B). Equation B was used to back-age the complete sample of teeth (*n*=41). The 95% confidence intervals for estimated ages by the R:RPA ratio and which have CAA ages of 4-5 years old and 6-7 years old were mutually exclusive (Table 7-4). This suggests that the minimum age with the 95% confidence interval for 6-7 years old animals can be used to identify trophy-hunted lions that are most likely younger than the set 6-years old age threshold.

Table 7-1 The Spearman rank order correlation between cementum annuli analysis (CAA) age and various tooth:pulp width ratios and tooth:pulp area ratios from the upper second premolar teeth of lions (*Panthera leo*) collected in Botswana between 2005 and 2007. A significant Spearman rank order correlation indicated a monotonic relationship between two variables. The analysis included the full data set and the subset with cementum annuli analysis ages < 10 years old.

Dependent variable	**Parameter**	**CAA age**	**CAA age**
		all years	**< 10 years**
Width ratio A	Correlation Coefficient	0.237	-0.176
	P (2-tailed)	0.147	0.422
Width ratio B	Correlation Coefficient	-0.646**	-0.635**
	P (2-tailed)	< 0.001	0.001
Width ratio C	Correlation Coefficient	-0.450**	-0.450*
	P (2-tailed)	0.002	0.024
Width ratio D	Correlation Coefficient	-0.645**	-0.663**
	P (2-tailed)	< 0.001	< 0.001
Tooth:Tooth Pulp Area ratio	Correlation Coefficient	0.712**	0.696**
	P (2-tailed)	< 0.001	< 0.001
Root:Root Pulp Area ratio	Correlation Coefficient	0.705**	0.770**
	P (2-tailed)	< 0.001	< 0.001

**. Correlation is significant at the 0.01 level (2-tailed)

*. Correlation is significant at the 0.05 level (2-tailed)

Width ratio A: Pulp:tooth width ratio at enamel-cementum junction

Width ratio B: Pulp:tooth width ratio 3 mm below enamel-cementum junction

Width ratio C: Pulp:tooth width ratio halfway down root

Width ratio D: Pulp:tooth width ratio ¾ down root

Table 7-2 Model summaries for curves fitted to cementum annuli analysis (CAA) age (independent variable) and width ratios as dependent variables from upper second premolar teeth of lions (*Panthera leo*) collected in Botswana between 2005 and 2007. The analysis included the full data set and the subset with cementum annuli analysis ages < 10 years old.

Dependent variable	Equation	R square	F	df1	df2	P	Constant	b
Width ratio B for all CAA ages	Linear	0.369	23.994	1	41	< 0.001	0.201	-0.009
	Logarithmic	0.426	30.386	1	41	< 0.001	0.256	-0.062
	Exponential	0.403	27.62	1	41	< 0.001	0.209	-0.062
	Logistic	0.403	27.62	1	41	< 0.001	4.777	1.064
Width ratio B for CAA ages < 10 years	Linear	0.49	22.127	1	23	< 0.001	0.249	-0.018
	Logarithmic	0.519	24.84	1	23	< 0.001	0.296	-0.089
	Exponential	0.501	23.06	1	23	< 0.001	0.267	-0.112
	Logistic	0.501	23.06	1	23	< 0.001	3.752	1.118
Width ratio C for all CAA ages	Linear	0.27	15.515	1	42	< 0.001	0.197	-0.009
	Logarithmic	0.319	19.69	1	42	< 0.001	0.258	-0.066
	Exponential	0.252	14.174	1	42	0.001	0.198	-0.066
	Logistic	0.252	14.174	1	42	0.001	5.053	1.068
Width ratio C for CAA ages < 10 years	Linear	0.29	9.396	1	23	0.005	0.248	-0.019
	Logarithmic	0.321	10.882	1	23	0.003	0.301	-0.095
	Exponential	0.221	6.515	1	23	0.018	0.245	-0.11
	Logistic	0.221	6.515	1	23	0.018	4.083	1.116
Width ratio D for all CAA ages	Linear	0.467	34.238	1	39	< 0.001	0.195	-0.01
	Logarithmic	0.554	48.369	1	39	< 0.001	0.26	-0.072
	Exponential	0.414	27.513	1	39	< 0.001	0.208	-0.081
	Logistic	0.414	27.513	1	39	< 0.001	4.798	1.084
Width ratio D for CAA ages < 10 years	Linear	0.541	25.899	1	22	< 0.001	0.242	-0.019
	Logarithmic	0.593	32.104	1	22	< 0.001	0.294	-0.095
	Exponential	0.495	21.535	1	22	< 0.001	0.272	-0.132
	Logistic	0.495	21.535	1	22	< 0.001	3.683	1.141

Table 7-3 Model summaries for curves fitted to cementum annuli analysis (CAA) age (independent variable) and area ratios as dependent variables from upper second premolar teeth of lions (*Panthera leo*) collected in Botswana between 2005 and 2007. The analysis included the full data set and the subset with cementum annuli analysis ages < 10 years old.

Dependent variable	Equation	R square	F	df1	df2	P	Constant	b
Tooth:Tooth Pulp Area ratio	Linear	0.514	44.453	1	42	< 0.001	3.955	0.693
All Ages	Logarithmic	0.436	32.492	1	42	< 0.001	0.750	4.384
	Exponential	0.555	52.443	1	42	< 0.001	5.281	0.068
	Logistic	0.555	52.443	1	42	< 0.001	0.189	0.934
Tooth:Tooth Pulp Area ratio	Linear	0.543	27.301	1	23	< 0.001	4.087	0.732
< 10 years	Logarithmic	0.517	24.667	1	23	< 0.001	2.501	3.390
	Exponential	0.542	27.175	1	23	< 0.001	4.693	0.096
	Logistic	0.542	27.175	1	23	< 0.001	0.213	0.909
Root:Root Pulp Area ratio	Linear	0.466	34.871	1	40	< 0.001	4.124	0.656
All Ages	Logarithmic	0.461	34.189	1	40	< 0.001	0.604	4.384
	Exponential	0.493	38.864	1	40	< 0.001	4.992	0.073
	Logistic	0.493	38.864	1	40	< 0.001	0.200	0.930
Root:Root Pulp Area ratio	Linear	0.647	42.244	1	23	< 0.001	2.512	0.990
< 10 years	Logarithmic	0.604	35.135	1	23	< 0.001	0.446	4.539
	Exponential	0.644	41.544	1	23	< 0.001	3.707	0.133
	Logistic	0.644	41.544	1	23	< 0.001	0.270	0.875

Table 7-4 The confidence intervals for the root pulp area (R:RPA) ratio age estimates for the two key cementum age classes when evaluating compliance with the 6-year age threshold strategy using upper second premolar teeth of lions (*Panthera leo*) collected in Botswana between 2005 and 2007.

	CCA age 4-5 years	CCA age 6 years
n	9	10
Mean R:RPA age estimate	4.5	6.4
SD	1.4	1.3
	4.5 ± 0.9	6.4 ± 0.8
Mean ± 95% CI	3.5 ≤ x ≤ 5.4	5.6 ≤ x ≤ 7.2)

The R:RPA ratio ages were plotted against the CAA ages (Fig. 7-2). The R:RPA minimum threshold age of 5.6 years was derived from the 95% confidence interval for the CAA age class 6-7 years (Table 7-4). There was agreement by both aging methods that nine animals were too young (<5.6 years) and 23 animals were old enough (>5.6 years) to have been hunted under the 6-years age threshold strategy. A disparity between the two methods arose with nine samples: the CAA flagged three samples as too young but which were aged as > 5.6 years by the R:RPA ratio method (A in Fig. 7-2), and six animals were estimated by the CAA to have been old enough to be hunted while the ratio method flagged them as too young (B and C in Fig. 7-2). The lion skulls were not available for inspection and I could only use the wear of the PM^2 as a secondary aging criterion. Tooth wear indicated that the three lions flagged by the CAA as too young were most likely >6 years, and supported the CAA ages in the six samples flagged as too young by the R:RPA ratio method. From the 26 lions shot in low density source populations, eight (31%) were too young to be hunted in contrast to only one out of 15 (7%) lions hunted in high density source populations.

7.5 Discussion

Our study showed that the R:RPA ratio age estimates can be used in Botswana as the first step in monitoring hunters' compliance with the 6-years minimum-age threshold strategy for trophy-hunted lions in Botswana.

I found that the PM² is suitable for determining the R:RPA ratios and is the most suitable tooth to sacrifice for monitoring purposes in trophy lions. The frequency of tooth breakage is lower for premolars compared to canines (Van Valkenburgh, 1998) and cessation of secondary dentine deposition is thus less likely. In Kruger National Park, South Africa, the first signs of wear on the PM² tend to occur only around 7 years of age (Smuts *et al.*, 1978). Its collection moreover does not detract from the aesthetics of the trophy.

The two area ratios yielded better correlations with the CAA age estimates than the width ratios. Our R:RPA ratio (Equation B), which is an adaptation of the tooth:tooth pulp cavity area ratio method developed by Cheater (2006), provided the best model. It eliminates the potential effect of crown wear and the irregular shape of the crown pulp chamber on the ratio, the latter which may also affect the precision of the area measurements during digitizing. Excluding the teeth with a CAA age of 10 years and older reduced the impact of the decrease in accuracy of age estimates with an increase in age of the animal for both CAA and pulp cavity:tooth ratios methods (Gipson *et al*., 2000; Rose, 1992; Rolandsen *et al*., 2008; Thomson & Landon *et al*., 1998). The mutually exclusive 95% confidence

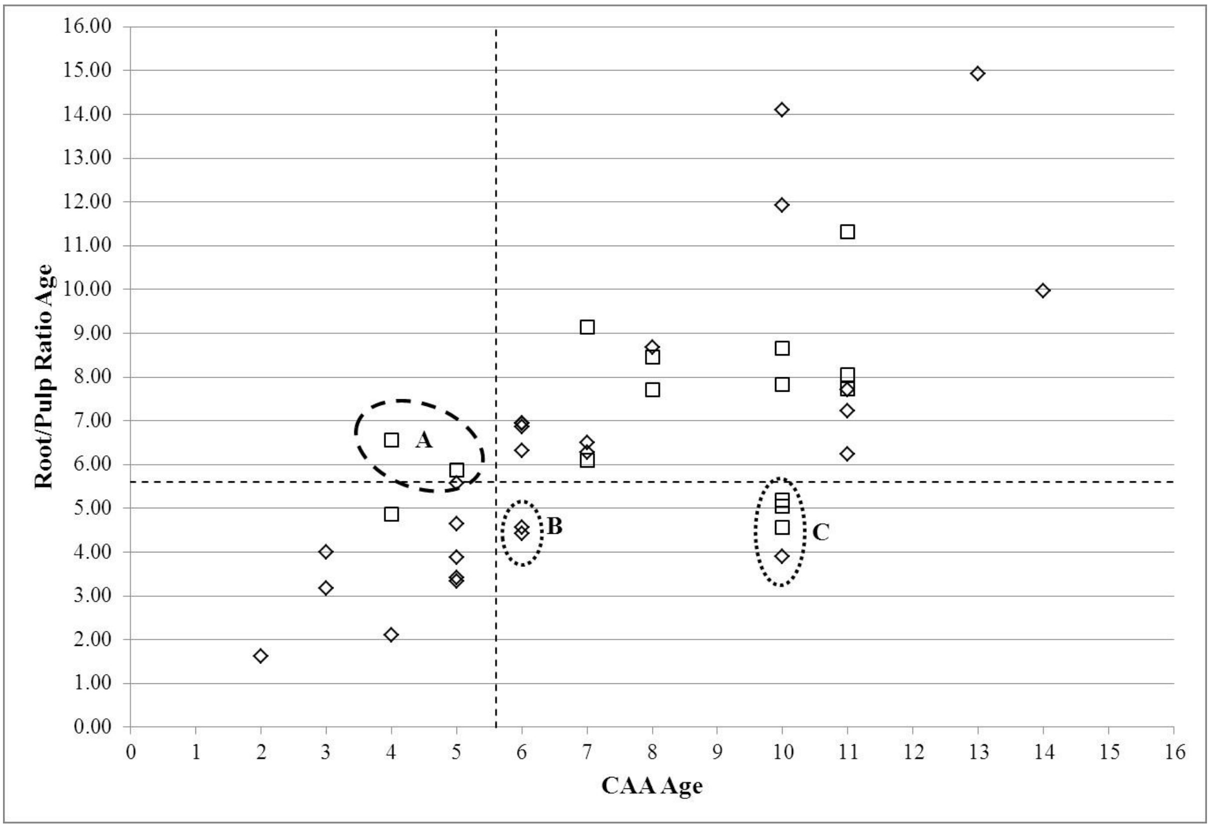

Figure 7-2 The correlation of ages between the root pulp area ratio (R:RPA) age estimates plotted against the cementum age analysis (CAA) estimates in upper second premolar teeth collected from lions (*Panthera leo*) in Botswana between 2005 and 2007. The low lion density area (◊) indicate < 2 lion / 100 km² and the high lion density area (□) >4 lion / 100 km².

intervals for the R:RPA ratios in the CAA age classes 4-5 years and 6-7 years shows the methods ability to distinguish between animals older and younger than the age threshold.

The CAA can underestimate the age of a lion by as much as 1.3 years when the most recently formed dark-staining layer is not detected. Where the R:RPA ratios aged some animals as too young and the CAA estimated the ages to be 10 years, the CAA result was supported by wear on the PM². Therefore, the ages of animals that are estimated to be < 5.6 years by the R:RPA ratios should be verified by additional aging criteria such as tooth wear, morphological characteristics, and CAA before a decision on non-compliance is made. Ideally, the accuracy of the R:RPA ratios for lions in Botswana would be improved by using known-age animals as the standard instead of the CAA age estimates. Additionally, the pattern and characteristics of the lean season for the source population of a hunted animal and the date hunted known will increase the accuracy of the CAA aging**.** With the acquisition of more known age samples, the accuracy of the ratio method can be improved.

The advantage of the 6-years minimum age threshold strategy proposed by Whitman *et al*. (2004) is that it can be implemented without quotas based on population data. I evaluated the Botswana lion trophies and found that the operators were more successful in complying with the minimum age threshold in high lion density areas than in low lion density areas. In the high-density areas 53% of the lions hunted were estimated to be 10 years and older. On the other hand, the 31% of lions hunted in the low-density areas were younger than the age threshold. With the smaller selection of available male lions in the latter areas, operators seemed more inclined to hunt under-aged males. Clearly a mechanism to enforce compliance, particularly in areas with low lion population densities, is vital for the successful implementation of the minimum age threshold strategy.

7.6 Recommendations

The mean R:RPA ratio - and CAA ages of both the left and right PM² should be used as the most probable age estimates and as part of a multi-criteria approach in estimating the ages of trophy-hunted lions. All samples estimated to be younger than the R:RPA ratio minimum threshold age of 5.6 years should be sent for CAA. Whenever the CAA- and the R:RPA ratio age estimates result in a disparate estimated age evaluation, multiple secondary aging criteria, such as wear on the PM² and other teeth, nose colouration and mane development, should be used to reach a decision regarding compliance.

Lastly, the penalty for non-compliance must outweigh the gains of hunting an under-aged lion. A good example is the Niassa Points System implemented in Niassa National Reserve, Mozambique. This system rewards ecologically sound hunting and penalizes the hunting of underage animals. A concession is awarded with an initial hunting quota of two lions. The subsequent quotas are increased or decreased according to the age of the trophies hunted (Anonymous, 2013).

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CHAPTER 8. SUMMARY AND CONCLUSIONS

8.1 Introduction

The range, population size and trend of large carnivores are important parameters to assess their status globally (Bauer *et al.* 2015; Wiesel 2015), and to design range wide conservation strategies (Durant 2007; Mills & Hofer 1998), regional conservation plans (IUCN/SSC 2006), or national plans (Lindsey & Davies-Mostert 2009). Acquiring abundance data poses logistical challenges determined by the size of the study site, accessibility of the area and the characteristics of the target species (Dénes, Silveira & Beissinger 2015; Harris *et al.* 2013; Karanth *et al*. 2011). The cost, efficiency of monitoring populations and logistical constraints are of concern (Bart *et al.* 2004; Caro 2016; Durant *et al.* 2007; Harris *et al*. 2013; Hines *et al*. 2010; Karanth *et al*. 2011).

In a conservation and wildlife monitoring environment that is chronically underfunded and hampered (Balmford *et al.* 2003; Caro 2016; Durant *et al*. 2007; Keeping *et al*. 2018; Lindsey *et al*. 2014; Scholte 2011), selecting appropriate methods to achieve the objectives are high priority. This problem is more acute in Africa, not only with a lack of resources but also a lack of capacity among researchers and managers (Caro 2016; Durant *et al*., 2007; Keeping *et al*., 2018; Lindsey *et al*., 2014; Scholte 2011).

Methods to estimate abundance should not be resource intensive, but be economical and efficient and produce appropriate results that are scientifically defensible (Harris *et al*. 2013). Methods should be appropriate for landscape-scale surveys (Karanth *et al*. 2011) and should not require complex statistical analysis and programming skills (Dénes *et al*. 2015; Rich *et al*. 2019).

The analysis and calibration problems identified in the track survey literature (Chapter 3) highlight the importance of this problem. The track survey method is supposed to be a simple method, yet experienced researchers made mistakes that survived peer review, and impacted on the interpretation of the published results. Using track survey data and call up survey data for occupancy modelling provide opportunities to gain valuable insight in the distribution of carnivores across landscapes. However, the lack of user capacity will limit the use of complex modelling in the African conservation environment. The failure of peer review to detect problems with simple track surveys, raise concerns regarding the peer review process to evaluate complex modelling applications.

How do track surveys and call up surveys measure up against the other requirements? Key requirements for track surveys are competent and experienced trackers and suitable substrate. Data recording and preparation are simple and together with the R script (Chapter 3) reduce the expertise required to produce results. No specialised equipment is necessary and track surveys are suitable at landscape level (Winterbach *et al*. 2017). However, track surveys have low power to detect population trends, due to the high CV of the data (Chapter 3). Although there are calibrations for clay soils, results indicate potential problems with track surveys on clay soils (Belant *et al*. 2019).

Call up surveys require acoustic equipment and appropriate calls. It has been used at landscape level (Ferreira & Funston 2010) and data analysis is straightforward, provided that appropriate response calibration is available. In the Okavango Delta the CV of call up data was high, resulting in low power to detect population trends (Chapter 4). In conclusion, both track surveys and call up surveys comply with most of the requirements, but lack the power to detect population trends with trend analysis.

Conservation management of large carnivores requires more than monitoring population trends. The conservation costs at local and national level can be high. Nature and wildlife-based tourism provides benefits that compensate these costs to a certain degree (Balmford & Whitten 2003; Dickman, Cooney, Johnson, Louis & Roe 2019; Hutton & Leader-Williams 2003). Furthermore, tools to measure photo-tourism potential can assist with land use planning and designation of photo-tourism versus trophy hunting (Chapter 6). Consumptive use requires management to ensure sustainable off take (Whitman, Starfield, Quadling & Packer 2004).

Maintaining the Northern Conservation Zone in Botswana is important for long term conservation of the large carnivore guild and requires monitoring of carnivore populations, land use planning and management of trophy hunting. The first part of this study focused on the methodology of monitoring large carnivore populations (Chapters 2, 3 and 4). Chapter 5 collated available survey data to assess brown hyaena (*Parahyaena brunnea*) distribution and densities across Botswana. Chapter 6 investigates the potential of photographic tourism to replace trophy hunting as an economic incentive for conservation in those parts of the Northern Conservation Zone perceived as unsuitable for photographic tourism. Chapter 7 implemented monitoring of compliance with the minimum age threshold for lion (*Panthera leo*) trophy hunting as supporting data to manage sustainable use.

8.2 Simplified large African carnivore density estimators from track indices

Stander (1998) demonstrated a significant linear correlation between true density and track density for leopard (*Panthera pardus*), lion and wild dog (*Lycaon pictus*); which can be used to estimate carnivore densities (animals /100 km ^{2}) from track densities (tracks / 100 km). While Stander (1998) used linear models through the origin, Funston *et al.* (2010) followed the more conventional approach of a linear model with intercept (Eisenhauer 2003; Quinn & Keough 2002; Sokal & Rohlf 1995). This conventional approach may not intercept with zero but may fit the data better than linear modelling through the origin. 'Forcing' the model through the origin is rarely appropriate (Quinn & Keough 2002). I, therefore, used guidelines to assess whether a linear model fitted through the origin is more appropriate to model large African carnivore densities and track indices (Eisenhauer 2003; Quinn & Keough 2002; Sokal & Rohlf 1995).

Using the track density and true density data from (Funston *et al.* 2010), I calculated simple linear regression with intercept analysis and simple linear regression through the origin and used the confidence interval for ß in the linear model $y = \alpha x + ß$, Standard Error of Estimate, Mean Squares Residual and Akaike Information Criteria to evaluate the models (Eisenhauer 2003; Quinn & Keough 2002; Sokal & Rohlf 1995).

The Lion on Clay and Low Density on Sand models with intercept were not significant ($P > 0.05$). The other four models with intercept and the six models through origin were all significant ($P < 0.05$). The models using linear regression with intercept all included zero in the confidence interval for ß and the null hypothesis that $ß = 0$ could not be rejected. All models showed that the linear model through the origin provided a better fit than the linear model with intercept, as indicated by the Standard Error of Estimate and Mean Square Residuals. Akaike Information Criteria showed that linear models through the origin were more robust and that none of the linear models with intercept had substantial support.

The results showed that linear regression through the origin is justified over the more conventional linear regression with intercept for all models I tested, supporting Funston *et al*.'s (2010) conclusion that a general model can be used to estimate large carnivore densities from track densities across species and study areas. It is important to use the appropriate model to account for the study area's substrate, since linear models used to estimate large carnivore densities differed significantly on sandy and clay soils (Funston *et al.* 2010).

The validity of density estimates below 0.27 carnivores /100 km^2 (< 0.88 tracks /100 km) is questionable, but it may be the best, or only, data available to guide conservation. Estimates and trends obtained from track surveys of low-density populations should be interpreted with caution. More independent data for different species are required to validate the models.

Recommendations

1. Studies that use track surveys to estimate carnivore densities should provide the relevant data to allow recalculation of density estimates when calibrations have improved. This will also facilitate comparison among studies that used different calibration models.

Contributions

1. Refined version of the track density / carnivore density calibration.

8.3 How to estimate carnivore density from track surveys: a guide to revised analyses improving our confidence

The strong relationships between track density and true carnivore densities for different large African carnivores enable extrapolation of population estimates from track densities (Funston *et al.* 2001; Funston *et al.* 2010; Stander 1998). Carnivore track encounters (also called "incidences" (Funston *et al.* 2010)) are recorded along pre-defined line transects in order to measure track density (number of tracks per 100 km) and mean inter-incidence distance (termed "spoor frequency" (Stander 1998)); i.e. the number of km per track incidence. Calculating confidence intervals (CIs) for track density is problematic, but track frequency can provide a reliable estimator. Appropriate species calibrations for the relationship between track incidence frequency and track density are required.

Confusion exists regarding the analysis of track survey data, e.g. the calculation of *Coefficient of Variation* (CV) (Midlane *et al.* 2015), and also inconsistencies and ambiguity in the calculation of CIs for track densities. Erroneous calculations of true or reference carnivore densities also occurred (Gusset & Burgener 2005; Houser, Somers & Boast 2009), resulting from the failure to account for only partial home range overlap of known individuals with the study area.

I review the general method, clarify the ambiguous terminology used, outline the key assumptions underlying the design and analysis of a carnivore track survey, and suggest which information has to be recorded and how. I used my own, and published, multi-species survey data from various countries and sites to clarify correct collection and analysis of carnivore track data for density estimation. Inappropriate analyses of data introduce bias in the results and impact on the perceived power to detect population trends reliably. I also derived species-specific calibrations for the relationship between track incidence frequency and track density to estimate the confidence intervals of track density for the entire southern African large carnivore guild.

The track survey method is a practical tool to assess large carnivore populations at different scales, but it has limitations. Rigorous survey design, correct analysis paradigms, and critical interpretation of the results are essential. A guideline for best practices and correct analysis of track survey data allow others to replicate track surveys in a way that improve inter-survey comparisons of results.

Recommendations

1. Prepare a detailed practical guide for practitioners to plan and conduct a track survey, and how to prepare and analyse the data.
2. Use GPS collar data from monitored individuals to assess the impact of seasonal changes in carnivore movements on existing calibrations of track density and true density.
3. Empirical determination of tracker skill in order to assess inter-observer bias regarding track detection ability, identification of carnivore species, gender, and age category and ability to age tracks accurately.
4. Assessing the impact of fatigue towards reduced detection probability (defining a maximum effective transect length);
5. Calibrate multi-site and multi-species surveys to different substrate types; and
6. Assess track survey power to detect true population trends from known populations.

Contributions

1. Provide effective way to estimate confidence intervals from the mean distance between track incidences that are narrower than confidence intervals estimated from transect based estimates.
2. Clarified several mistakes and problems and provided R script to reduce analysis problems.
3. Demonstrated lack of power to detect trends with trend analysis.
4. Provided an alternative to test for significant increases or declines in a population, using χ^2 test with Bonferroni intervals.

8.4 Calibration of call-up surveys for lions in the Okavango Delta

Call up surveys can be used to survey lions, spotted hyaenas (*Crocuta crocuta*), brown hyaenas and jackal (*Canis mesomelas*) (Mills, Juritz & Zucchini 2001; Ogutu & Dublin 1998; Thorn *et al.* 2010). Calibration of response is required to estimate density from calling station data (Cozzi *et al.* 2013; Ferreira & Funston 2010; Mills, Juritz & Zucchini 2001; Ogutu & Dublin 1998). Environmental factors, aspects of carnivore biology and ecology, and survey parameters can influence carnivore response (Brink, Smith & Skinner 2013; Ferreira & Funston 2010). Few of the underlying assumptions for call up surveys have been investigated and a clear relationship between lion abundance and abundance indices from call up surveys is lacking.

I used a known lion population that was identified as part of a long-term study in the western part of the Okavango Delta in Botswana, to calculate reference densities. I used observed response probabilities, the proportion of known lions responding after 30, 60 and 90 minutes of audio playbacks, for a site-specific calibration of response from call up surveys. I compared the bias and precision of density estimates derived from published calibrations and the calibrations from this study.

Over a period of 10 years, the reference densities of known lions declined from 7.99 to 2.33 lions /100 km². The CV of lions per calling station was high (>200%). Mean number of lions per calling station significantly correlated with reference density ($P < 0.05$). Individual identification showed that 24% of lions responded more than once.

X^2 tests showed that response probability did not differ by gender and age among surveys when calling extended to 90 minutes. Therefore, I calculated response probabilities (90 minutes call duration) for the four surveys combined. Mean actual response distance increased less than expected, from 2.77 km (CV 14%) to 2.85 km (CV 14%) when extending calling from 60 to 90 minutes. Mean effective response distance was 1.84 km (CV 14%) for 90 minutes call duration. I calculated density estimates with these calibrations for response distances and probabilities using two GLMs (selected with corrected Akaike's Information Criterion).

Site-specific calibrations performed better than published calibrations. Extending the call duration to 90 minutes reduced bias and coefficient of variation of density estimates. Due to the high coefficient of variation in the Okavango Delta survey data, this survey technique is unlikely to detect population trends and results should be interpreted cautiously.

Recommendations

1. Site-specific calibration of carnivore response is necessary to improve density estimates and our understanding of variability in response probability.
2. Density estimates should be supported with population structure data and trends in prey base.
3. Evaluate the limitations and reliability of the emerging models beyond simulation (Dénes *et al*. 2015; Palmer *et al*. 2018). Our data with reference densities provide the opportunity to test models using spatial replication and compare performance with the more simplistic approaches we tested.

Contributions

1. Tested the key assumption and showed that there is a monotonic linear relationship between the number of lions per calling station and reference lion density.
2. Improved the CI calculations to give smaller confidence intervals.
3. Provided a better understanding of factors influencing call up surveys and the limitations of the method.

8.5 Conservation implications of Brown Hyaena (*Parahyaena brunnea*) population densities and distribution across landscapes in Botswana.

The brown hyaena is endemic to southern Africa with the largest population of this near-threatened species occurring in Botswana (Wiesel 2015). However, limited data was previously available to assess distribution and density reliably. The objectives were to use a stratified approach (by land use) to collate available data and to collect additional data that would allow assessing brown hyaena distribution and density across land uses in Botswana.

I conducted track surveys (Funston *et al.* 2010; Stander 1998; Winterbach *et al.* 2016) and collated my data with data from other surveys, including track surveys, camera traps and questionnaires. Using these data I estimated the brown hyaena population based on the stratification of Botswana for large carnivores (Winterbach, Winterbach & Somers 2014; Winterbach *et al.* 2015).

Brown hyaenas occur over 533 050 km² (92%) of Botswana. Density estimates ranged from 0 brown hyaenas /100 km² in strata of northern Botswana to 2.94 (2.16 – 3.71) brown hyaenas /100 km² in the southern stratum of the Central Kalahari Game Reserve. I made assumptions regarding densities in strata that lacked data, using the best references available. I estimated the brown hyaena population in Botswana as 4642 (3133 – 5993) animals, with 6.8% of the population in the northern conservation zone, 73.1% in the southern conservation zone, 2.0% in the smaller conservation zones and 18.1% in the agricultural zones.

Similar densities of brown hyaenas in the Central Kalahari Game Reserve and the Ghanzi farms highlight the potential of agricultural areas to conserve this species. The inclusion of agricultural landscape in Botswana is critical for the long-term conservation of brown hyaenas; these areas provide important links between populations in South Africa, Namibia and Zimbabwe.

Recommendations

1. Botswana contains the core of the brown hyaena population in southern Africa, and conflict mitigation on agricultural land is crucial to maintain connectivity among the range countries.

Contributions

1. Country wide distribution and density of brown hyaenas for conservation evaluation and planning.

2. Provided important data and techniques, linked to the conservation of brown hyaena, and are useful in future studies and national population trend analysis.

8.6 Wildlife Abundance and Diversity as Indicators of Tourism Potential in Northern Botswana.

Wildlife tourism can provide economic incentives for conservation in lieu of costs at local, national and global scales (Balmford & Whitten 2003; Bookbinder *et al.* 1998; Lindsey *et al.* 2007). Due to the abundance of wildlife and the presence of charismatic species some areas are better suited for wildlife tourism (Di Minin *et al.* 2013; Goodwin & Leader-Williams 2000; Kerley, Geach & Vial 2003; Maciejewski & Kerley 2014).

Wildlife-based tourism in Botswana occurs primarily in the form of High Paying Low Volume (HPLV) tourism and included sport hunting until 2012 (Office of the President 2012). The photographic tourism industry perceived some parts of northern Botswana as not suitable for HPLV tourism. These areas are characterised by low wildlife densities (Winterbach, Winterbach & Somers 2014). Phasing out sport hunting raised the question whether photographic tourism could replace sport hunting as an income generator in these areas perceived as unsuitable for photographic tourism.

The first objective was to develop criteria based on wildlife abundance and diversity to evaluate tourism potential in the Northern Conservation Zone of Botswana. The second objective was to quantify and compare tourism experiences in areas with high and low tourism potential. I used aerial survey data to estimate wildlife biomass and diversity to determine tourism potential, while data from ground surveys quantified tourist experience.

Areas used for HPLV tourism had significantly higher mean wildlife biomass and wildlife diversity than the areas without this type of tourism. Only 22% of the Northern Conservation Zone has intermediate to high tourism potential. The areas with high tourism potential, as determined from the aerial survey data, provided tourists with significantly higher wildlife sightings (ground surveys) than low tourism potential areas. Even Low Paying tourism may not be economically viable in concessions that lack areas with intermediate to high tourism potential. The majority of Botswana's Northern Conservation Zone has low tourism potential, but low tourism potential does not equate to low conservation value. This area provides wet season range for elephants (*Loxodonta africana*) and buffalo (*Syncerus caffer*) (Chase 2011), is part of zebra (*Equus burchelli*) migrations (Bartlam-Brooks, Bonyongo & Harris 2011) and may be an important refuge area for wild dog (Winterbach, Winterbach & Somers 2014).

Recommendations

1. Alternative conservation strategies should be developed to complement the economic incentive provided by wildlife-based tourism in Botswana.
2. Monitoring of age threshold compliance is critical to ensure sustainable quotas if lion trophy hunting is reintroduced in Botswana.

Contribution

1. Two techniques to evaluate potential for photo-tourism.

8.7 Suitability of the pulp-tooth ratio to estimate the age of trophy-hunted African lions in Botswana

Trophy hunting creates economic incentives for conservation over vast areas of Africa and the lion is a high-value species in the trophy hunting industry (Lindsey, Roulet & Romañach 2007). Sustainable harvest strategies are required to reduce the potential negative impact of trophy hunting on the long-term survival of exploited lion populations (Becker *et al.* 2013; Croes *et al.* 2011; Loveridge *et al.* 2007; Packer *et al.* 2009).

Trophy hunting of male African lions can be managed at a sustainable level by targeting males above a minimum age threshold of 5-years (Whitman *et al.* 2004). Botswana implemented a 6-years minimum age threshold from 2005 to 2007. Professional hunters evaluate the age of male lions in the field by means of physical characteristics (Whitman 2010). Although conservation managers can only monitor hunters' compliance with this age threshold post hoc, age information from hunted specimens can be used to inform future hunting quotas to ensure sustainability.

Two tooth characteristics, the deposition of cementum annuli and the closure rate of the pulp cavity, have been used as a proxy for age in archaeology (Jones 2012), vertebrate paleontology (Binder & van Valkenburgh 2010), forensic science (Paewinsky, Pfeiffer & Brinkmann 2005), and wildlife studies (Smuts, Anderson & Austin 1978; Van Aarde 1983). Cementum annuli analysis (CAA) involves counting the number of growth layers (annuli) deposited in the tooth's cementum, correlating with seasonal changes in nutrition (Asmus & Weckerly 2011; Lieberman 1994). Annual increments of cementum annuli were observed in lions to guide age estimates (Cheater 2006; Smuts, Anderson & Austin 1978).

Pulp cavity closure rate is an alternative to CAA for aging animals and the pulp cavity to tooth ratio can be used to distinguish lions into age classes (Cheater 2006; Meachen-Samuels & Binder 2010; Smuts, Anderson & Austin 1978). The incremental deposition of dentine in lion ceases only when the dental pulp is exposed and necrosis begins (Smuts, Anderson & Austin 1978).

Cementum annuli analysis to estimate age requires trained and experienced interpreters (Calvert & Ramsay 1998; Rolandsen *et al.* 2008). Factors such as tooth type, the type of sectioning and staining, sample size, and interpretation of annuli and environmental factors may impact accuracy (Gasaway, Harkness & Rausch 1978; Goodwin & Ballard 1985; Hess *et al.* 2011; Landon *et al.* 1998; Medill *et al.* 2009). Calculating the pulp cavity to tooth ratio using radiographs is fast, easy to perform, and less expensive, but differences in pulp cavity to tooth ratio may occur between mandibular and maxillary teeth (Knowlton & Whittemore 2001) and between males and females (Kershaw *et al.* 2005).

In CAA, accuracy of age estimates can decrease with increasing animal age (Gipson *et al.* 2000; Rolandsen *et al.* 2008). The same is true for the pulp cavity to tooth ratio method (Thomson & Rose 1992; Landon *et al.* 1998). Smuts, Anderson and Austin (1978) and Cheater (2006) showed annual increases in cementum annuli for lions in southern Africa. Recognizing a lack of known-age reference material from free-ranging lions, the CAA method may be the best age reference available to calibrate the pulp cavity to tooth ratio as an age index for lions in Botswana.

I evaluated the suitability of six different tooth to pulp width and area ratios to estimate animal age of trophy-hunted male lions in Botswana as a *post hoc* means to monitor compliance with the 6-years minimum age threshold. Linear regression ($y = 0.990x + 2.512$) of tooth root area and root pulp cavity area ratio (R:RPA) and CAA < 10 years ($F_{1,\ 23} = 42.244$, $P < 0.001$) provided the strongest correlation ($r^2 = 0.647$). The 95% confidence intervals for age estimates from R:RPA ratios in CAA age classes 4-5 years and 6-7 years were mutually exclusive, showing the method’s ability to reliably distinguish between animals older and younger than the 6-years trophy threshold.

Operators in Botswana were more successful in complying with the minimum age threshold in high lion density areas than in low lion density areas. In high density areas, 53% of hunted lions were estimated to be 10 years and older. Conversely, 31% of lions hunted in low density areas were younger than the legal age threshold. With a smaller selection of available male lions in the latter areas, operators appeared more inclined to hunt under-aged males. A mechanism to enforce compliance is vital for the successful implementation of the minimum age threshold strategy.

Recommendations

1. Develop a multi-criteria approach for estimating the ages of trophy hunted lions.
2. The mean R:RPA ratio, and CAA ages of both the left and right PM^2, should improve confidence in age estimates.
3. All samples estimated to be younger than the R:RPA ratio minimum threshold age of 5.6 years should be submitted for CAA. Whenever CAA, and the R:RPA ratio age estimates, result in a disparate estimated age evaluation, multiple secondary aging criteria, such as wear of the PM^2 and other teeth, nose colouration and mane development, should be used to reach a decision regarding compliance.

4. The penalty for non-compliance must outweigh the gains of hunting an under-aged lion.

Contributions

1. Technique and assessment of compliance to age threshold for trophy lions hunted in Botswana.

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