## Foraging behaviour and health status of Red-billed Oxpeckers (*Buphagus erythrorhynchus*) in the Kruger National Park, South Africa

by

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#### **DECLERATION**

I declare that the Master's Degree research dissertation that I herewith submit for the Degree of Master of Science at the University of the Free State is my independent work, and that I have not previously submitted it for a qualification at another institution of higher education.



(Signature of Candidate)

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at Windhoek, Namibia

#### **ABSTRACT**

Red-billed Oxpeckers (Buphagus erythrorhynchus) are tick feeding birds that reduce ectoparasite loads on African ungulates. However, little is known about their feeding ecology, seasonal abundance and health wellbeing. All these attributes contribute towards their conservation. I studied the Red-billed Oxpecker feeding ecology and health status in the southern regions of Kruger National Park by documenting their seasonal abundance, infection prevalence, body condition and foraging behaviour (host preference and foraging location on host). No significant difference in Oxpecker abundance was observed between the three seasons. Nine potential ungulate host species were recorded and birds were observed feeding on eight of the present species. White rhinoceros (Ceratotherium simum), Cape buffalo (Syncerus caffer) and giraffe (Giraffa camelopardalis) were the most preferred hosts whereas waterbuck (Kobus ellipsiprymnus) were the least preferred host. Birds preferred sitting and foraging from the back, head and neck of the host ungulate – where they appeared more tolerated by the host. No wound feeding activity was recorded during this study. In total, 30 Red-billed Oxpeckers were caught and blood and feather samples from them were screened for parasites. Ectoparasite prevalence on birds was highest during the summer months, with the majority found on the flight wing feathers. It was found that birds with ectoparasites seemed to have a lower body condition index compared to those with no ectoparasites. The most common Haemoparasites found in the Oxpeckers were Leucocytozoon. It was also the only haemoparasite found during the dry season.

# What you do makes a difference, and you have to decide what kind of difference you want to make

 $\square$  Jane Goodall  $\square$ 

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#### **CHAPTER ONE**

#### **General Introduction**

#### **Background**

In nature, organisms rarely survive/live in isolation. The majority of species to some degree offer and obtain a service from other species in what can be termed a symbiotic relationship (Yu et al. 2017). Symbiotic relationships are relied on by most organisms for some form of energy, protection or metabolic processes (Werner et al. 2015). The concept of symbiosis refers to the coexistence of two dissimilar organisms, usually in an intimate association, to the benefit of at least one of the two organisms (Relman 2008). In the animal context, this relationship may take the form of one organism using the other as a mode of transport, feeding habitat or source of food (Thomas et al. 2013; Cheng and Prayago 2014). Three types of symbiosis relationships are recognised, namely commensalism, parasitism and mutualism. Ecological interactions like commensalism and parasitism only benefit one of the two involved in the association (Brown et al. 2012), whereas mutualism is a type of relationship between two organisms where both derive a benefit from the association (Cheng and Prayogo 2014; D'Angelo and Sazima 2014). One type of mutualism that is well known amongst animals is cleaning symbiosis. This is where one species removes ectoparasites and or damaged tissues from another species. In terrestrial ecosystems of the African Savanna a well-documented example of this type of symbiosis involves the relationship between Oxpeckers (*Buphagus* spp) and ungulates (Poulin 1993; Thomas et al. 2013; Farrell et al. 2014).

#### **Oxpecker-ungulate relationship**

The Oxpecker-ungulate relationship is a key example of a mutualistic association between terrestrial vertebrates that is subject to spatial and temporal variation in sub-Saharan Africa (Mooring and Mundy 1996; Plantan et al. 2012). Both the Red-billed (Buphagus erythrorhynchus) and the Yellow-billed (Buphagus africanus) Oxpeckers are avian species from the family Buphagidae that are entirely confined to the Afro-tropical region, with both species occurring in South Africa and in some areas occurring in sympatry (Ndlovu and Combrink 2015). Interspecific competition has been observed between the two species, where the larger-bodied Yellow-billed Oxpecker outcompetes the smaller Red-billed Oxpecker at preferred feeding sites (Stutterheim et al. 1988; Koenig 1997; Jubber 2014; Ndlovu and Combrink 2015). These two species can be found over a vast range, but due to their dependence on large ungulates (both wild and domestic), their distribution appears to be patchy and in close association with the ungulate host counterparts (Stutterheim and Brooke 1981). A symbiotic relationship exists between Oxpeckers and ungulates where birds obtain their main food source, ticks, from the ungulates and in turn their feeding behaviour reduces the ectoparasite loads on host species (Mooring and Mundy 1996, Ndlovu and Combrink 2015) and also minimises the risk of ungulates contracting vector borne diseases (Weeks 2000).

During the first half of the 20<sup>th</sup> century, populations of both the Red-billed and Yellow-billed Oxpeckers became threatened in South Africa (Plantan *et al.* 2009; Ndlovu and Combrink 2015). The birds suffered range and population declines due to their close association with domestic and wild ungulates (Mellanby *et* al. 2009; Spies *et al.* 2012). Farmers were treating their livestock with acaricides which proved to be poisonous to Oxpeckers. Additionally, the over-hunting of large wild ungulates also limited the number of host species available in the wild. The rinderpest epidemic of 1896 – 1897 also played a devastating role in significantly reducing the number of available host species, especially domestic livestock, and

suitable tick species. During this time, the remaining highly fragmented populations of Oxpeckers were now restricted to national parks and protected areas (Plantan *et al.* 2014; Ndlovu and Combrink 2015). Following the incorporation of Oxpecker-friendly acaricides during the mid-1950s, the Red-billed Oxpecker population started to show signs of recovery (Plantan *et al.* 2014). The steady increase in Oxpecker population size and distribution resulted in both species being moved from "Threatened", to a category of "Least Concern" by the International Union for Conservation of Nature (Spies *et al.* 2012). However, this species still requires protection in South Africa and there are current ongoing efforts to relocate and reintroduce birds to former range areas in South Africa (Plantan *et al.* 2014).

Although ticks are reported to be the main food source for Oxpeckers (Grobler 1980; Weeks 1999; Plantan *et al.* 2012), analyses of the stomach contents showed that birds also feed on dung, earwax, insects, mites, lice, hair, scruff cells, and secretions (eyes and nose) from their hosts (Plantan *et al.* 2012; Weeks 1999). There have also been instances were birds have been recorded feeding on wounds and blood on hosts, meaning that the association is not always mutualistic (Nunn *et al.* 2011). Wound-feeding can inflict negative effects on the host, such as delayed, healing which increases the risk of infection for the host (Plantan *et al.* 2012).

In the wild, it has been recorded that from the total range of species available, Red-billed Oxpeckers show preference for only a selected number of ungulate host species (Mooring and Mundy 1996, Ndlovu and Combrink 2015). Large ungulates such as Cape Buffalo (Syncerus caffer), White Rhinoceros (Ceratotherium simum), Giraffe (Giraffa camelopardalis), Plains Zebra (Equus quagga burchelli), Greater Kudu (Tragelaphus strepsiceros) and Hippopotamus (Hippopotamus amphibius) were generally the preferred Red-billed Oxpecker host to forage for ectoparasites, particularly ticks. While Impala (Aepyceros melampus), which is not regarded as a large ungulate, also appeared to be preferred. In northern regions of the Kruger National Park, where both Red-billed and Yellow-billed Oxpeckers occur

in sympatry, the larger (in terms of body size) Yellow-billed Oxpecker prefers to forage on the large ungulates such as Cape buffalo, and appeared to restrict the smaller body-sized Red-billed Oxpeckers to foraging on smaller ungulates like impala (Ndlovu and Combrink 2015). In the absence of interspecific foraging competition and where ungulate host variety is wide, both Oxpecker species prefer to forage on larger bodied ungulates that present a greater foraging surface area. However, if competition exists, then the next preference will be to select for the most abundant host species regardless of body size (Ndlovu and Combrink 2015).

Oxpecker host preference is governed by intrinsic host characteristics such as body size, hair length as well as herd size and host abundance, which seem to play an important role in determining the attractiveness of the host to the birds (Mooring and Mundy 1996). Besides these factors, tick abundance and quality i.e. species and developmental stage, may also affect the foraging behaviour of Oxpeckers. In an optimal foraging scenario, Oxpeckers will minimise time spent searching for food, while maximising food intake. This is thought to be the reason why during periods of tick scarcity, it is more cost effective for Oxpeckers to feed on wounds present on their hosts rather than comb the skin for ectoparasites (Mooring and Mundy 1996; Plantan *et al.* 2012). However, this seasonal foraging shift from symbiotic to parasitic is yet to be studied in great depth.

#### Oxpecker health

Beside food availability and age of the bird, the foraging behaviour of Oxpeckers, like any other bird, is likely to be influenced by the wellbeing i.e. health of the bird (Bonter *et al.* 2013). Few records are available for parasites affecting Oxpeckers. To date, no blood parasites have been screened from Oxpeckers, neither through microscopy (Valkiunas 2005) nor molecular means (Bensch *et al.* 2009). To my knowledge only a single feather mite (*Montesauria buphagid*), was described from Oxpeckers (Doña *et al.* 2016), but there are no published

records of feather lice, ticks or other ectoparasites found on Oxpeckers. Parasites feeding on living bird tissues can affect the birds' health severely and in different ways, ranging from anaemia to reduced reproduction and survival (Proctor and Owens 2000). Besides the use of bird-friendly acaricides in livestock farming (Samish *et al.* 2004; Plantan *et al.* 2009; Plantan *et al.* 2014), current conservation practises have also prioritised the relocation of birds from Kruger National Park to areas where Oxpeckers once occurred (Plantan *et al.* 2009). Most of these conservation practices have overlooked the disease and parasite burden found in the "founder" Oxpecker populations. There is therefore a high potential and unknown risk that the translocated birds may harbour and introduce novel infections to new landscapes and animal populations. On the other hand, there is also a risk of exposing founding populations to novel infections endemic to the translocation destinations. A disease and parasite surveillance on Oxpeckers populations from the Kruger national park (the founder population for translocations in South Africa) will therefore inform the translocation efforts aimed at increasing Oxpecker population range and numbers.

At an ecological context the seasonal monitoring of Oxpecker body condition coupled with feeding behaviour can also indirectly be used to understand the seasonal variation in the bird's food abundance, forage effort and behaviour (Van Gils *et al.* 2007; Powell *et al.* 2015). The general body condition of a bird, as measured from mass and structural dimensions, is closely tied to it's health i.e. parasitic disease prevalence and immune response (Merila and Svensson 1995; Moreno *et al.* 1998; Galvan *et al.* 2012).

#### Study design

Red-billed Oxpeckers were once on the brink of extinction due to an increase in usage of vertebrate poisonous acaricides and the over-hunting of large ungulates (Plantan *et al.* 2009; Ndlovu and Combrink 2015). Since the introduction of Oxpecker friendly acaricides, the

reintroduction of Oxpeckers into their former range became possible (Plantan *et al.* 2014). Organizations like Endangered Wildlife Trust (EWT) started to reintroduce Oxpeckers from national parks to areas where they previously occurred (Plantan *et al.* 2009, Kalle *et al.* 2017). An important part of Oxpecker conservation in the South African context is to understand the bird's current host and feeding preferences as well as the health status (disease and body condition) of the founder populations so as to optimise Oxpecker reintroduction efforts and limit the potential risk of disease spread.

Given that Red-billed Oxpeckers are numerous and more widely distributed than the Yellow-billed Oxpeckers, most reintroduction efforts have focused on the Red-billed Oxpeckers. I therefore studied the common Red-billed Oxpecker to understand its feeding ecology in the southern region of Kruger National Park, where they occur in allopatry, and assessed how season and drought affects their health, i.e. body condition, parasite (ecto- and haemoparasites) prevalence and immune response. The study specifically addressed three objectives.

The first objective was to document Oxpecker feeding behaviour in terms of host selection and foraging location on host, while also accounting for seasonal variation and the effect of the recent 2015 – 2016 drought period. I hypothesised that Red-billed Oxpeckers would prefer to feed on hosts with larger body size and mass, since these hosts will harbour a greater number of ticks and other ectoparasites (Mooring and Mundy 1996). It was also predicted that Oxpeckers would prefer to feed on the back of the host where it is less likely to disturb the host and hence remain tolerated.

In the second objective, I monitored the local Oxpecker population fluctuations by monitoring the seasonal bird abundance along transects in relation to the available ungulate host species. The hypothesis tested was that the highest abundance of Oxpeckers would be found on host in the summer wet season compared to dry winter period, since ungulates tend

to have a higher tick load in late wet summers when grass availability is at its peak (Mooring and Mundy 1996). The last objective was to assess the seasonal body condition, and ecto- and haemoparasite prevalence on Oxpeckers caught at the study site. I hypothesised that Oxpecker body condition will be better during the summer months, when ungulate hosts harbour more ticks. I further expected birds to have higher ecto- and haemoparasites during the wet summer seasons when conditions are suitable for insect survival and potential haemoparasites vectors like mosquitoes and midges are most abundant (Perez-Rodriguez *et al.* 2015). Furthermore, given that the study area is located at a subtropical Lowveld region suspected to be highly diverse in tropical diseases, this study also presented an opportunity to document the diversity of avian parasites found in South African Oxpeckers.

#### **Study species**

The present study focuses on the widely distributed and abundant Red-billed Oxpecker as a model species to understand the obligate gleaning ecology of Oxpeckers in an African savanna setting. The Red-billed Oxpecker is one of two Oxpecker species that belongs to the family Buphagidae, previously classified as a subfamily Buphaginae within the Sturnidae family (Stutterheim *et al.* 1988; Lovette and Rubenstein 2007; Jubber 2014). The species is entirely confined to the Afrotropical region with a range extending from Central African Republic eastward to Ethiopia and all the way south to South Africa, occurring in countries such as Zimbabwe, Namibia, Botswana and South Africa (Stutterheim 1982a; Mellanby *et al.* 2009). The former range of Red-billed Oxpeckers in South Africa stretched over the Northern and Eastern Cape, Gauteng, North West, Limpopo, Mpumalanga and KwaZulu-Natal areas (Stutterheim and Brooke 1981; Stutterheim 1982a).

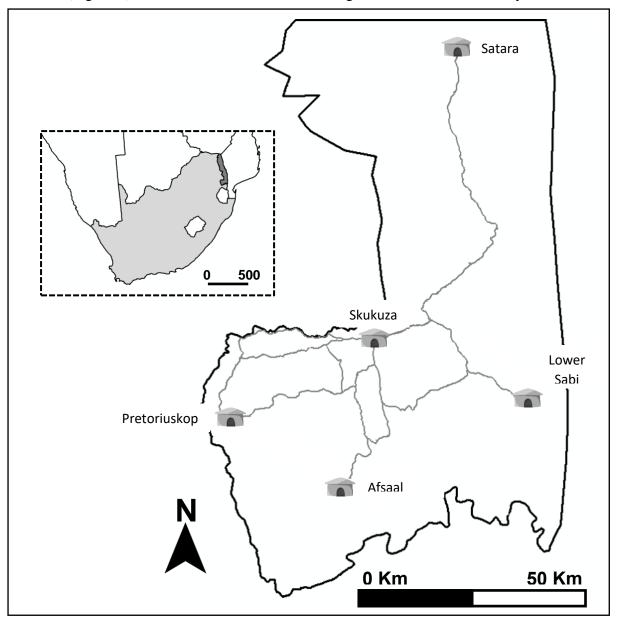
Red-billed Oxpeckers are savanna species, largely confined to areas with an annual rainfall higher than 500 mm, although they have also been recorded in drier areas where the

mean rainfall ranges between 250 – 500 mm (Stutterheim 1982a; Stutterheim and Panagis 1985). They are absent from open deserts and closed evergreen forests with their habitat selection being further constricted due to their preference for particular tick and host species (Robertson and Jarvis 2000; Mellanby *et al.* 2009; Plantan *et al.* 2014). Breeding of this species occurs during the summer months from October to March (Stutterheim 1982b; Stutterheim 1982c; Koenig 1997). Nests are built in tree cavities lined with dry grass and hair from their ungulate hosts. Sometimes rootlets and animal dung can also be used as nesting material (Koenig 1997; Plantan *et al.* 2014). Red-billed Oxpeckers can successfully raise three broods in a single breeding season (Stutterheim 1982b; Stutterheim 1982c), given the right conditions.

The main morphological feature used to distinguish the Red-billed from the Yellow-billed Oxpeckers is the colour and size of the bill. The base of the bill of the Yellow-billed Oxpecker is yellow and has a red tip, whereas the bill of the Red-billed Oxpecker is entirely red. Other distinct morphological features between the two species is the pale rump of the Yellow-billed Oxpecker, while the rump and body of the Red-billed Oxpecker has no colour difference (Jubber 2014). The Red-billed Oxpecker is also slightly smaller in body size (Average weight 50 g and average length of 20 cm) compared to the Yellow-billed Oxpecker (Average weight 60 g and average length of 20 cm) and has a laterally flattened beak used to remove adult and nymph ticks from their ungulate hosts in a scissoring like action (Hockey et al. 2015). The Yellow-billed Oxpecker on the other hand has a thicker, wider and less scissor-like bill (Hart et al. 1990; Koenig 1997). Red-billed Oxpeckers display a series of highly derived adaptations for life with large ungulates. These include short, sharp claws that help them to cling to the hides of animals and long stiff tails that provide support while they cling onto the bodies of the ungulates. They also have laterally flattened beaks that have a sharp cutting edge making it possible for them to handle ticks (Koenig 1997).

#### Study area

The study was carried out over a two-year period, with fieldwork conducted at the end of the rainy season (April), the height of the dry winter season (July) and beginning of the rainy season (November). Surveys was concentrated within an approximately 80 km radius around Skukuza (Figure 1), in areas confined within the Kruger National Park boundary.



**Figure 1:** Map of the southern section of Kruger national park, showing the routes used during observations.

Skukuza is situated in the southern part of the park. The area consists of granite rocks with moderately undulating plains (Venter *et al.* 2003). The areas south of Skukuza are dominated by the Lowveld bushveld zone, which consists of broad-leaved vegetation in the uplands and fine-leaved bushveld in the bottomlands (Venter *et al.* 2003). The area between Skukuza and Satara falls under the flat plains and is mainly composed of savanna grasses with fine-leaved trees. The average rainfall in the study area ranged between 500 – 700 mm per year and there is a notable decreasing rainfall gradient from Malelane on the southern end of the study area, and Satara which lies in the northern extant of the study area. Generally, the area has high mean temperatures during the wet summer season (mean = 27 °C) and mild, frost-free (mean = 16 °C) dry winters (Venter *et al.* 2003). A variety of animals are found within the Kruger national park including 517 species of birds and 147 mammal species (Ferreira and Harmse 2014). Among the mammal species, some of the larger ungulates that occur within the study area includes Black Rhino (*Diceros bicornis*), White Rhino, Elephants (*Loxodonta Africana*) Cape Buffalo, Greater Kudu, Giraffe and Sable antelope (*Hippotragus niger*) (Chirima *et al.* 2012).

#### **Dissertation outline**

This dissertation consists of four chapters where this first chapter is a general introduction to the study. Chapter two focuses on the first two objectives of the study that investigates the seasonal feeding behaviour and populations trends of Red-billed Oxpeckers. Here I looked at the host selection behaviour of Red-billed Oxpeckers, to see if there are specific species of ungulates that the birds preferred. Foraging behaviour on the host was documented, to determine if Oxpeckers had a preferred feeding location on hosts. Seasonal and daily variations were considered to assess if there were any differences in bird feeding behaviour at different sampling periods. I also looked at both Oxpecker and host abundance and determined how they differed between three sections in the southern area of the park during different seasons. In

chapter three I dealt with the last objective that assessed the overall health of the Red-billed Oxpeckers in terms of seasonal body condition changes, parasite (ecto- and haemoparasite) prevalence and immune status. Chapters two and three were written as stand-alone research manuscripts (article format) to ease the passage of publication. In the last chapter I collectively discuss and synthesise all the findings from the preceding chapters and suggest areas for further research.

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#### **CHAPTER TWO**

## Host selection and foraging behaviour of Red-billed Oxpeckers (*Buphagus* erythrorhynchus) in the Kruger National Park.

#### Abstract

Red-billed Oxpeckers (Buphagus erythrorhynchus) have a symbiotic relationship with African ungulates in the savanna. Oxpeckers mainly feed on ticks found on most herbivores and therefore reduce ectoparasite load and the potential for vector-borne diseases on hosts. These birds nearly went extinct and since then, numerous efforts have been made to reintroduce Redbilled Oxpeckers into their former range. An important aspect that aids in their conservation is having a good understanding of their foraging behaviour. An observational study was conducted in the southern section of the Kruger National Park in order to get a better understanding of Red-billed Oxpecker distribution and foraging behaviour during three different consecutive seasons. The abundance and distribution of Oxpeckers was similar during all three seasons. Oxpeckers showed a marked preference for larger ungulates namely White Rhino (Ceratotherium simum), Giraffe (Giraffa Camelopardalis) and Cape Buffalo (Syncerus caffer), with a slight difference in host preference amongst seasons. The preferred Oxpecker foraging location on hosts differed slightly amongst different ungulate host species, but the most preferred locations were the back, head and neck. Oxpeckers were generally tolerated well by the hosts. Highest rejection rate was observed in November at the beginning of the wet summer season. These findings support the hypothesis that Oxpecker abundance and feeding behaviour is driven by both seasonal and host species availability and therefore should be considered when reintroducing these species to their former range.

#### Introduction

Several bird species have been reported to glean ticks from larger animals, including Cattle Egrets (Bubulcus ibis), Domestic Chickens (Gallus gallus) (Dreyer et al. 1997; Samish and Rehacek 1999; Samish et al. 2004) and Magpies (Pica pica) (Found 2017), but a few are known to feed primarily on ticks (Samish et al. 2004). Two of the most known tick gleaning birds are the Red-billed (Buphagus erythrorhynchus) and Yellow-billed (Buphagus africanus) Oxpeckers which are endemic to the sub-Saharan regions of Africa (Hockey et al. 2005). The Red-billed Oxpecker is more abundant and widely distributed in the southern African region than the Yellow-billed Oxpecker (Robertson and Jarvis 2000). Oxpeckers and herbivores share a mutualistic relationship, where birds feed on ectoparasites found on their host, obtaining their major food source, while herbivores benefit from a reduced (1) ectoparasite load, (2) exposure to tick-borne diseases and (3) other negative tick effects such as tick toxicosis, anaemia, metabolic disturbances and tick worry (Mooring and Mundy 1996a; Tomazzoni et al. 2005; Plantan et al. 2012; Ndlovu and Combrink 2015). Oxpeckers have also been known to consume lice, mites, insects, dung, ear wax, secretions from the nose, eyes and mouth, as well as pieces of their hosts' skin (Samish and Rehacek 1999; Plantan et al. 2012). Wound feeding has also been reported by some authors (Samish 2000; Weeks 2000; McElligott et al. 2004; Ndlovu and Combrink 2015) however, it is a rare sight in the wild (Bishop and Bishop 2013). Oxpeckers use several feeding methods to obtain ectoparasites from their host. They either catch insects flying around their hosts, pluck or peck at parasites from their hosts or use the scissoring method, which involves the sweeping of their heads along the hosts' body while opening and closing their bill (McElligott et al. 2004). Even though Oxpeckers are visual predators (Samish et al. 2004), they spend up to 94 % of their feeding time using the scissoring method (Samish and Rehacek 1999; Weeks 1999).

Oxpecker feeding behaviour and distribution are highly dependent on the availability and distribution of their foraging host, mainly large ungulates (Galetti *et al.* 2017). Besides domestic animals, these birds have been documented foraging on a total of 21 wild ungulate species (Stutterheim 1981). Mooring and Mundy (1996a) further observed that within these suitable host species, there are certain species which are preferred, while others are rarely selected. Thus, it was predicted that in order to maintain an Oxpecker population, "key hosts" need to be present in an area. Furthermore, the foraging effort must be cost-effective for the Oxpeckers (Hart *et al.* 1990) i.e. food intake has to be maximised, while search time is minimised (Mooring and Mundy 1996a; Plantan *et al.* 2012). Prevailing environmental conditions, host phenotypic characteristics, tick species and abundance will thus affect the suitability of a foraging host for Oxpeckers (Mooring and Mundy 1996a).

One of the most significant host characteristics is probably host body size. Large-bodied hosts have a greater surface area and are able to support and tolerate a higher abundance of ticks. The more ticks per single host, the less time Oxpeckers have to spend searching for food (Hart et al. 1990; Mooring and Mundy 1996a; Mooring and Mundy 1996b; Robertson and Jarvis 2000; Nunn et al. 2011; Plantan et al. 2012). Another characteristic that makes a host more attractive to Oxpeckers is hair length. Longer hair might increase the Oxpeckers search and ectoparasite retrieval time, while shorter hair could make it easier for Oxpeckers to locate and remove ticks (Mooring and Mundy 1996a; Mooring and Mundy 1996b; Plantan et al. 2012). The herd size of a suitable host would also influence the Oxpecker host selection. Large herds would mean more individuals to forage from, especially from smaller bodied-ungulates, making it cost effective for the birds (Mooring and Mundy 1996a; Mooring and Mundy 1996b). The last host characteristic that is probably the most important is the hosts' behavioural response to the Oxpeckers. Red-billed Oxpeckers have sharp claws specifically adapted for clinging to their hosts (Hockey et al. 2005). Thus, hosts behavioural response to these birds

does not only refer to the nuisance of the birds flapping around, but it also refers to the tolerance of the Oxpeckers sharp claws while they are perched on the host. Hosts can either reject the birds by swinging their head or jumping around, or the host can tolerate them and expose tick infested areas to the birds (Hart *et al.* 1990; Mooring and Mundy 1996a; Mooring and Mundy 1996b).

Tick species and abundance on hosts also affect the Oxpecker's host selection. Oxpeckers prefer to feed on certain type of tick species such as the blue ticks, *Boophilus decoloratus*, and the brown ear tick, *Rhipicephalus appendiculatus*, and would therefore select hosts that carry these species (Robertson and Jarvis 2000; Weeks 2000; Hockey *et al.* 2005). There is a drastic spatio-temporal fluctuation in the abundance of ticks between seasons, years and sites (Mooring and Mundy 1996a). Tick densities are generally higher during the wet summer seasons, compared to the dry winter period (Hart *et al.* 1990; Mooring and Mundy 1996a; Robertson and Jarvis 2000; Plantan *et al.* 2012). Environmental factors can also have a significant effect on Oxpecker host selection. Factors such as fire, proximity to water and visibility of hosts can play a direct role in Red-billed Oxpecker host selection.

Ticks are blood-sucking ectoparasites that can have severe negative effects on their hosts (Goodenough *et al.* 2017). In National Parks and wildlife reserves, the most practical method to control tick populations is by burning. According to Goodenough *et al.* (2017), most studies have shown that after a fire, there is an immediate decline in the tick population. This is either due to direct mortality or because tick refugia within the field layer has been reduced or eliminated. A decline in the tick population directly affects Oxpeckers foraging. Proximity to water sources is an important environmental factor to consider when looking at host selection. Hosts usually congregate in high densities when at water sources, which increases their visibility to Oxpeckers. The easier it is for these birds to find suitable hosts, the more cost efficient it is for these birds (Tarakini *et al.* 2017). This is also why Oxpeckers are more likely

to select hosts that occur in areas that increase the visibility of the ungulates like grasslands, compared to closed woodlands where the birds would have to spend more time searching for hosts and less time foraging (Mooring and Mundy 1996a).

Red-billed Oxpeckers nearly became extinct during the first half of the 20<sup>th</sup> century because of the decline in wild ungulate numbers and the widespread usage of poisonous acaricides by farmers, not only poisoning the tick feeding birds, but also reducing the available amount of ticks to feed on. The population of Red-billed Oxpeckers, as well as their home range, drastically decreased to a point where they were mainly restricted to game parks and relatively underdeveloped communities (Bezuidenhout and Stutterheim 1980; Robertson and Jarvis 2000; Samish *et al.* 2004). In 2002, organizations like the Endangered Wildlife Trust, started to reintroduce Red-billed Oxpeckers into areas where they used to occur. (Plantan *et al.* 2009, Kalle *et al.* 2017). However, it is important to have a good understanding of the foraging behaviour and host selection of the birds by studying them in their natural setting, so as to gain a better insight into their ecology and be better informed when selecting suitable reintroduction areas.

The aim of this study was to document Red-billed Oxpecker seasonal distributions and foraging behaviour in the southern region of the Kruger National Park, an area very suitable for this study because it supports a large number of ungulatesspecies (Chirima *et al.* 2012) and Red-billed Oxpeckers are also frequently observed in this area (Hockey *et al.* 2005). The distribution of the birds was determined relative to the available foraging hosts, while feeding behaviour was accessed in terms of host selection and foraging location on host. I used the most abundant Red-billed Oxpecker to test the hypothesis that Oxpecker abundance and feeding behaviour was driven by both seasonal and host species availability. I predicted that bird numbers in the park will be stable since Oxpeckers are resident species (Hockey *et al.* 2005), however, their host foraging preference will be expected to change as a response to the

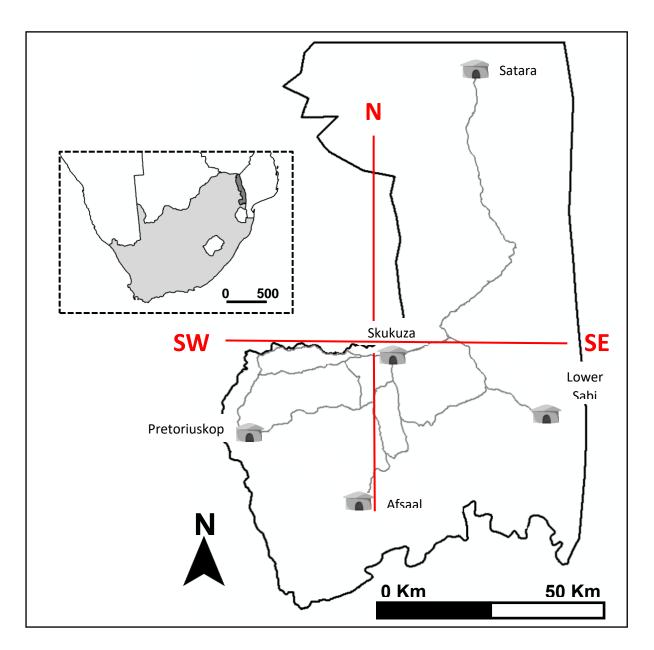
availability of suitable host species i.e. ungulate abundance, diversity and herd size which fluctuates seasonally. I hypothesized that Oxpeckers will prefer to feed on large ungulates, since these hosts will harbour a greater number of ticks and tend to be more tolerable to the birds (Ndlovu and Combrink 2015) i.e. the thick hide makes these ungulates tolerate the "stabbing" Oxpecker claws when perching and foraging on them. Red-billed Oxpeckers and possible ungulate host species abundance were repeatedly monitored during three seasons in a year on several road transects with a combined distance of approximately 431 km.

#### **Materials and Methods**

Study area

The study was carried out in the southern region of the Kruger National Park (KNP), South Africa. Fieldwork was conducted in 2017 during three distinct seasons of the year, namely: (1) end of the wet summer season in April; (2) mid dry winter period in July; and (3) beginning of the wet summer season in November. Count surveys were done along already existing park roads found within an approximately 80 km radius around Skukuza rest camp (24°59'45.66" S, 31°35'30.96" E), in areas confined within the park boundary. The sampling location was divided into three areas, namely the north, southeast and southwest sections, to be able to get better insight regarding the distribution and abundance of Red-billed Oxpeckers throughout the southern section of the park. The north section included routes between Skukuza and Satara (24°23'34.66" S, 31°46'47.53" E), the southeast section was made up of roads found in between Skukuza, Lower Sabie rest camp (25°07'08.93" S, 31°54'58.02" E) and Afsaal day visitor camp (25°17'03" S, 31°31'54" E), while the southwest section encompassed roads found in between Pretoriuskop rest camp (25°10'10.19" S, 31°16'07.05" E), Phabeni entrance gate (25°01'22" S, 31°14'03" E) and Skukuza (Fig. 2.1). Skukuza is located in the Lowveld bushveld zone, which includes broad-leaved vegetation in the uplands and fine-leaved

bushveld in the bottomlands. The average rainfall for this area ranges between 500 mm - 700 mm per year with high mean temperatures during the summer (mean = 27 °C) and mild, frost-free winters (mean = 16 °C; Venter *et al.* 2003). A variety of animals occur within Kruger National Park including 517 species of birds and 147 mammal species (Ferreira and Harmse 2014).



**Figure 2.1:** A map of the southern section of Kruger national park, showing the survey routes (roads) used for Oxpecker and ungulate observation transects. The study area was divided into

three study sections, North (Satara), Southwest (Pretoriuskop and Phabeni gate) and Southeast (Lower Sabi and Afsaal).

#### *Fieldwork*

Transect observation and counts were done following the methods of Grobler (1980) and Ndlovu and Combrink (2015). Data collection took place in the morning (07h00 – 10h00) and late afternoon (15h00 – 17h30), as these times coincides with Oxpeckers peak feeding period (Ndlovu and Combrink 2015). One vehicle, with two to four occupants, was used during observation times. Non-overlapping routes were used, travelling at speeds not exceeding 40 km/h within the sampling site area, looking for potential hosts with or without Oxpeckers. The distance covered for each survey was recorded to the nearest kilometre. A pair of binoculars (16x magnification) were used to count and positively identify the behaviour of the birds within a 150 m distance from the counting vehicle. The following observations was made each time a potential host was encountered: (1) host species; (2) number of hosts; (3) number of Oxpeckers; (4) wound presence on host individual; (5) position of Oxpecker on host; (6) behaviour of Oxpecker; and (7) host reaction to Oxpecker. The behaviour of the birds was categorised as either wound-feeding, non-wound feeding or non-feeding and the host reaction to the Oxpecker was either marked as tolerating or rejecting the Oxpeckers.

#### Statistical analysis

To determine Red-billed Oxpecker abundance per kilometre, the total number of birds observed in each section (North, Southeast and Southwest) during each season (April, July and November) was divided by the total distance of that section. The same method was used to calculate host abundance. Kruskal-Wallis Test was used to compare Oxpecker and host abundance between the different seasons. A series of Mann-Whitney U Test was used to test for differences in Oxpecker and host abundance between the two sampling time slots. The

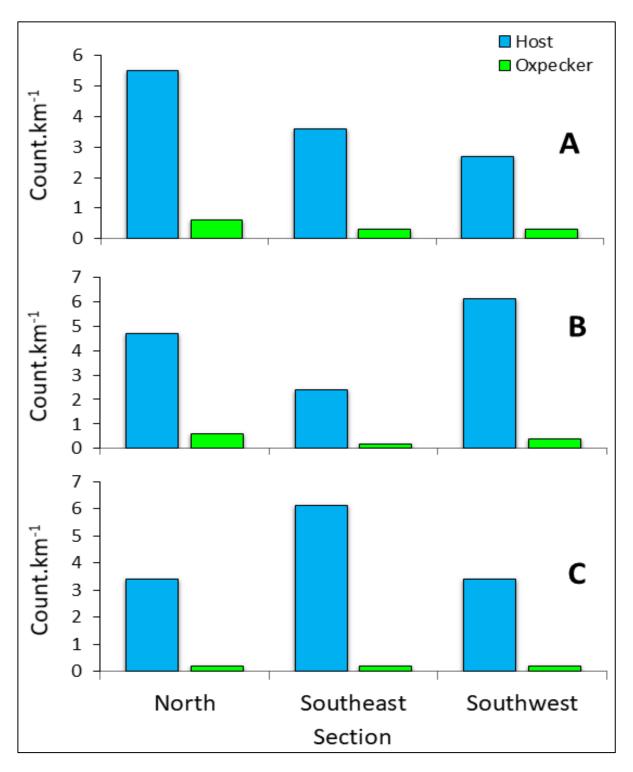
Oxpecker-Host preference index (PI) was calculated following Stutterheim and Stutterrheim (1980) method for each host ungulate species (total number of birds divided by the number of host animals counted) in order to determine Red-billed Oxpecker host preference.

$$PI = \frac{Total\ number\ of\ birds\ seen\ on\ given\ hosts}{Number\ of\ hosts\ seen\ of\ that\ species}$$

Simple arithmetic was used to determine Oxpecker host body location preference.

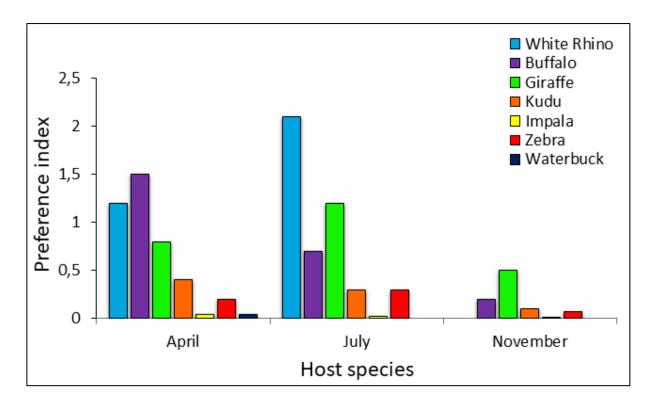
#### **Results**

A total of 5 240 host ungulates and 389 Red-billed Oxpeckers were counted during the study (See Appendix 1). Host abundance and Oxpecker abundance was similar for the three sections and the three seasons. There was no significant difference in total host abundance (numbers regardless of species composition) for the entire study site amongst the three seasons ( $H_{2, 15} = 0.573$ ; p = 0.751). Oxpecker abundance was also similar amongst the three sampling seasons ( $H_{2, 15} = 2.538$ ; p = 0.281). (Fig. 2.2) There was also no significant difference in numbers of counted hosts between the two daily sampling periods i.e. morning vs afternoon ( $U_{16} = 33$ ; z = -0.234; p = 0.818). Oxpecker count numbers also did not differ significantly between the sampling periods ( $U_{16} = 32$ ; z = 0.328; p = 0.741).



**Figure 2.2:** Host and Red-billed Oxpecker abundance (count per kilometre) at the three sections of southern region of Kruger National Park during (A) end of the wet summer season in April; (B) mid dry winter period in July; and (C) beginning of the wet summer season in November.

Overall, the most preferred ungulate host species for the Red-billed Oxpecker were the White Rhino (*Ceratotherium simum*, PI = 1.3), the Giraffe (*Giraffa camelopardalis*, PI = 0.8) and the Cape Buffalo (*Syncerus caffer*, PI = 0.5). Oxpeckers were also found on Greater Kudu (*Tragelaphus strepsiceros*), Impala (*Aepyceros melampus*), Plains Zebra (*Equus quagga*) and Waterbuck (*Kobus ellipsiprymnus*), although these were not the preferred host species (Fig. 2.3). Host preference did seem to differ slightly between seasons. White Rhino was the preferred species during April and July, but no Oxpeckers were seen on them during November, even though, White Rhinos were recorded in the study area during that time. Impala, though seemingly not significant, were an important host species: they represented 85% of the host species population recorded and also made up 26% of the total observation of Oxpeckers-Host interactions..



**Figure 2.3:** Host preference of the Red-billed Oxpecker compared between three different times of the year.

A total of 152 preferred foraging location instances were observed, with Red-billed Oxpeckers most frequently observed on the back (40 %; n = 62), head (26 %; n = 41) and the neck (23 %; n = 35) of the host. Preferred foraging location of the birds seems to differ amongst ungulate host species. The most preferred foraging location for Oxpeckers on the White Rhino (50 %, n = 3), Buffalo (66 %, n = 8), and Plains Zebra (57 %, n = 8) was the back, while for the Giraffe (38 %, n = 10) and Kudu (39%, n = 13), Oxpeckers were observed more on their necks. For the Impala, Oxpeckers were observed the most on their backs (42 %, n = 26) and their heads (36 %, n = 22). For some host species (Giraffe, Kudu and Impala), Red-billed Oxpecker were also seen foraging on the legs and around the endogenital areas (Fig 2.4).

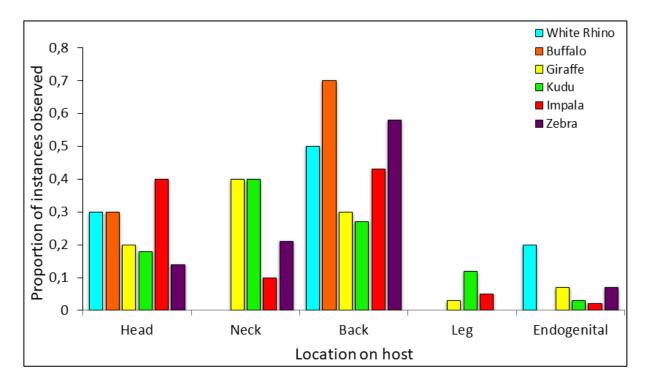


Figure 2.4: Red-billed Oxpecker foraging location on preferred ungulate host species.

Oxpeckers were mostly tolerated by their hosts throughout the year, with the only sign of rejection by hosts observed during November which falls in the summer season. Buffalo was the only host species that did not show any signs of rejection towards the birds (Table 2.1). No

wound feeding by the Oxpeckers was observed. The host ungulate species were observed to have good body condition with only three (out of a total number 5 240 of ungulate species counted during the study period) individual ungulate species, Giraffe, observed to have body wounds during the course of this study.

**Table 2.1:** Tolerant and rejection instances of the Red-billed Oxpecker observed on their preferred host species during the end of the wet season (April), the dry season (July) and the beginning of the wet season (November).

Host species	Tolerant	Rejected	Proportion Tolerant	
April				
White Rhino	11	0	1	
Cape Buffalo	3	0	1	
Giraffe	36	0	1	
Greater Kudu	9	0	1	
Impala	46	0	1	
Plains Zebra	12	0	1	
Waterbuck	2	0	1	
July				
White Rhino	17	0	1	
Cape Buffalo	33	0	1	
Giraffe	37	0	1	
Greater Kudu	21	1	0.955	
Impala	33	0	1	
Plains Zebra	13	0	1	
Waterbuck	0	0	0	
November				
White Rhino	0	0	0	
Cape Buffalo	7	0	1	
Giraffe	21	2	0.913	
Greater Kudu	7	1	0.875	

Impala	15	7	0.682
Plains Zebra	5	3	0.625
Waterbuck	0	0	0

## **Discussion**

The aim of the study was to provide insight on the seasonal distribution and foraging behaviour of the Red-billed Oxpecker in the southern region of Kruger National park. Overall, there was no statistically significant difference in Oxpecker abundance between the three seasons or sites. This is similar to what Mooring and Mundy (1996a) observed during their study. One aspect that can explain this result is the relative tick abundances during these seasons stayed similar. According to Stutterheim (1982), there are several environmental factors that can affect tick distribution and abundance within a particular area. The most significant ones affecting ticks are vegetation, rainfall and temperature. Oxpeckers prefer to feed on specific species of ticks. The presence or absence of these tick species can significantly affect the distribution of Oxpeckers. Since Oxpecker abundance stayed similar throughout the study period, it can be assumed that the environmental conditions were favourable, not only for the birds, but also for the preferred tick species.

White Rhino, Giraffe and Cape Buffalo where the most preferred hosts species of Redbilled Oxpeckers during this study. These results are similar to those observed by other authors (Ndlovu and Combrink 2015; Tarakini *et al.* 2017) and supports the hypothesis that Oxpeckers show preference to larger bodied ungulates. Since the three most preferred hosts are large bodied animals and were visited by Oxpeckers frequently, my results support the theory that larger ungulates have a higher surface area ratio and can support a higher tick/ectoparasite load (Tarakini *et al.* 2017) making them more attractive as foraging options to Oxpeckers compared to smaller ungulates. The Cape Buffalo, Giraffe and White Rhino also usually forage and travel in larger groups compared to other suitable host species, which supports the notion that Oxpeckers show preference to hosts that move in larger herds.

As seen in the results, Impalas were one of the least preferred host species of Red-billed Oxpeckers. However, even though they were not one of the preferred hosts, Impalas are still an important host species just because of the sheer numbers they occur in. Impalas are one of the smallest ungulates utilised by Red-billed Oxpeckers. Koenig (1997) and Hart et al. (1990) suggested that since Oxpeckers are often observed foraging on this species, it is possible that these ungulates harbour a greater tick load per unit body surface area ratio compared to some of the larger ungulates. Although Hart et al. (1990) also mentions that there is a possibility that Impalas may not harbour as many adult ticks as other host species, however, they might have a higher abundance of larval and nymphal stage ticks. Oxpeckers forage on both adult ticks and larvae, which could explain why Red-billed Oxpeckers were also seen foraging on impalas, even though, they are not large bodied ungulates. The habitat where Impalas are often found may also play a big role. Impala prefer mixed grass and shrub lands, which provides suitable conditions for the success of tick developmental cycles (Hart et al. 1990; Mooring and Mundy 1996b; Tarakini et al. 2017). It has also been suggested that Impalas are one of the thin-skinned ungulates, along with the Greater Kudu. Thin skin makes it easier for ticks and other ectoparasites to attach to the skin of the host and hence Impalas may possibly harbour a greater number of ticks (Tarakini et al. 2017). This and the fact that Impalas occur in large herds could be a significant factor that makes them attractive as a suitable foraging host for Red-billed Oxpeckers.

In contrast to what was observed by Bishop and Bishop (2013), the Waterbuck was not observed as a preferred host species for Red-billed Oxpeckers in this study. In fact, birds were only observed foraging once on the Waterbuck out of 65 observations. Waterbuck are generally not very tolerant of Oxpeckers according to observations made by Bishop and Bishop (2013).

They carry out vigorous resistance behaviour, either by running or head tossing, which deter the foraging of Red-billed Oxpeckers. No resistance behaviour was observed from the Waterbuck towards the birds, but then again Oxpeckers were only observed on this particular host in one occasion. This could mean that there were enough other suitable host species in the area for Oxpeckers to forage on. However, this can also mean that this vigorous resistance behaviour of Waterbuck might be the reason why Oxpeckers were only observed on this particular host species on one occasion.

It has been previously suggested by Koenig (1997) and Tarakini et al. (2017) that Redbilled Oxpecker host preference is influenced by how the host responds to the presence of these birds. Large ungulates are generally more tolerant of Oxpeckers inadvertently due to the limited mobility of these hosts (Weeks 1999; Ndlovu and Combrink 2015). Results obtained regarding host tolerance supports this. Majority of the time, the hosts showed no resistant behaviour towards the birds, with only a few instances recorded where the hosts rejected the Oxpeckers. This can be because of limited mobility or it is also possible that the hosts tolerated the birds since Oxpeckers reduce their parasite loads. Similar to what Bishop and Bishop (2013) observed, buffaloes showed no signs of resistant behaviour toward the presence of Oxpeckers and their foraging during the course of the study. This can be partly attributed to the limited ability of Buffaloes to groom themselves. Oxpecker foraging behaviour and therefore, the cost and benefits, varies significantly between different species of hosts (Koenig 1997; Weeks 1999). Some hosts may use resistance behaviour to redirect Oxpecker foraging to areas of high tick densities, potentially benefiting both host and bird. In other instances, this relocation may result from a failed attempt of the host to remove the nuisance birds (Bishop and Bishop 2013). Ndlovu and Combrink (2015) noted that Oxpeckers generally do not feed on areas of the hosts' body which are easily self-groomed. This coincides with results obtained in this study. Redbilled Oxpeckers showed preference to the head, neck and back areas of their hosts. These are all areas which are not easy for the host to groom themselves.

Previous studies have reported that Oxpecker wound feeding on wild ungulates is rarely observed and furthermore animals with fresh wounds do not tolerate the birds (Bishop and Bishop 2013, Ndlovu and Combrink 2015). This might explain why no wound feeding was observed during this study, even though a few of the hosts did have visible wounds. These findings are in contrast to those of Weeks (2000), who concluded that Oxpeckers prefer to feed on wounds, rather than on ticks that are clearly visible on the host species. Nunn *et al.* (2011) and Bishop and Bishop (2013) reported that in the wild, Oxpecker have no need to parasitize hosts, since there is usually an abundance of ticks to feed on. Since no wound feeding was observed in this study, it is reasonable to assume that these ungulates had sufficient tick abundance to meet the foraging needs of attending Oxpeckers without resorting to wound feeding. Another parsimonious explanation could be that most animals did not have open wounds. As seen in the results, more host resistant behaviour was observed during November (early wet season) when adult tick abundance was higher. It is possible that Oxpeckers tried to feed from wounds made by adult ticks and thus the hosts started to reject the birds more.

In summary, a range of factors seem to play a role in Red-Billed Oxpecker host selection. These findings seem to indicate that host size and tick abundance are some of the most important factors that play a role when it comes to host selection. Oxpeckers were mostly seen on the large ungulates, with the exception of the Impala. It was also noted that season does not seem to have a direct effect on Oxpecker abundance as long as suitable host and tick species are available. However, what was noticeable from the data gathered is that Oxpecker distribution is influenced by host distribution and thus, when new areas are considered for Oxpecker relocation, it is important to make sure the preferred host species are present in that area along with the preferred tick species.

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#### CHAPTER THREE

# Health status of the Red-billed Oxpecker (*Buphagus erythrorhynchus*) in Kruger National Park, South Africa.

#### **Abstract**

Wild birds are known to harbour a wide range of ecto- and haemoparasites which can either be beneficial or detrimental to their hosts. The objective of this study was to assess seasonal body condition, as well as, ecto- and haemoparasite prevalence on Red-billed Oxpeckers during the dry and wet seasons. Mist nets were set up at the animal enclosures at Skukuza in Kruger National Park. All birds captured were processed immediately with feather and blood samples collected and screened for any parasites present on these birds. A total of 30 adult Red-billed Oxpeckers were caught, with 21 birds found to be infected with either ecto- or haemoparasites. No significant difference was found in body condition index of Oxpeckers compared between the two seasons and there was also no significant difference in body condition between birds infected with parasites and those not infected. Oxpeckers were found to be infected with three haemoparasites with only *Leucocytozoon* present during both seasons. Feather mites was also the only ectoparasites detected on these birds. Even though, more research needs to be done in regards to Red-billed Oxpeckers and their parasites, this study provides a good basis of which parasites are carried by these birds.

## Introduction

Most free-living organisms host a variety of other species in associations that range from mutualistic to parasitic (Masello *et al.* 2006). Parasites, be it internal or external, derive resources from a suitable host for their own survival, growth and reproduction (Tschirren *et al.* 

2007). Wild birds are hosts to a range of different parasites, including intestinal parasites, ectoparasites and haemoparasites (Dolnik *et al.* 2010; Bernotiene *et al.* 2016). These have the potential to alter the general ecology, behaviour and survival of birds. Hence, in the context of unravelling the foraging ecology of Oxpeckers and their potential reintroduction in landscapes where they suffered local extinction, it is also important to understand the factors that affect their health status. As Oxpeckers recolonise their former range so will their parasites, and to date information on Oxpecker parasites is very limited.

Avian haemoparasites have a worldwide distribution (Waldenstorm *et al.* 2002; Mantilla *et al.* 2013) and have been found in approximately 4 000 bird species to date (Ots and Horak 1998; Mantilla *et al.* 2013). The three most common genera of avian haemoparasites are *Plasmodium*, *Haemoproteus* and *Leucocytozoon* spp. (Farrell *et al.* 2007). These three genera consist of a diverse group of vector-transmitted parasites that infect red-blood cells and other organs within their vertebrate hosts (Hellgren *et al.* 2004). Insects from the order Diptera are usually the vectors of these avian haemoparasites (Mantilla *et al.* 2013). Mosquitoes (Genera: *Aedes, Anopheles, Culex, Aedeomyia, Mansonia and Culiseta*) are the main vectors that transmit *Plasmodium* parasites. Biting midges (Diptera: Ceratopogonidae genus: *Culicoides*) and louse flies (Diptera: Hippoboscidae) are the primary vectors of *Haemoproteus* parasites. While blackflies (Diptera: Simuliidae) are the primary vectors that transmits *Leucocytozoon* (Silva-Iturriza *et al.* 2012).

In contrast to the intimate living associations that endoparasites have with their hosts, ectoparasites have free-living stages where they spend part of their life cycle in close association with the host. Direct contact with the host can range between the intermittent feeding of fleas to the prolonged attachment of ticks (Heeb *et al.* 2000; Clayton *et al.* 2010). Birds can be hosts to a great number and variety of different ectoparasites (Boyd 1951). The two major groups of ectoparasites found on birds include insects (four orders, including flies

(Diptera), fleas (Siphonaptera), lice (Phthirapera), true bugs (Hemiptera), mites (many families from the taxon Acari) and ticks (Poulin 1991; Clayton *et al.* 2010). Other less well known ectoparasites that can also be found on avian host includes leeches (four families), fungi (keratinophilic and cellulose decomposing forms) and bacteria (several unrelated groups that decompose feathers) (Clayton *et al.* 2010). The distribution of these parasites on their hosts varies significantly between different species. Some parasites have no habitat preference, whereas others are confined or restricted to certain areas on their host body (Boyd 1951). Nesting cavities of birds also provide a key microhabitat for a wide variety of free-living ectoparasites where the nesting material provides a place where these parasites can find refuge and reproduce (Heeb *et al.* 2000).

Due to the exploitation of resources from suitable hosts, parasites can have a negative impact on their hosts' health (Marzal *et al.* 2005). Studies have shown that parasites can affect their hosts' morphology by directly affecting their growth and maintenance, causing reduced weight gain. The behaviour of the host is also affected which can have a negative impact on the hosts fitness components causing lower reproductive success, smaller clutch sizes and ultimately affecting their long-term survival. Lastly, parasites can also have negative effects on the hosts immune response (Szép and Møller 2000; Marzal *et al.* 2005; Masello *et al.* 2006; Hamstra and Badyeav 2009; Dolnik *et al.* 2010). Sanz *et al.* (2001) mentioned that knowing exactly the effects that parasites have on free living birds is not easy to document without manipulating parasite load. During an experiment, they found that birds that were experimentally infected with haemoparasites developed myopathy, had reduced growth and some of the birds died.

Since the abundance of parasites negatively affects birds, the birds in turn, should have evolved a variety of anti-parasite behaviour, physiological and immunological defences (Masello *et al.* 2006). The first line of defence birds use against parasites is behaviour. Birds,

like many mammal species, engage in grooming behaviour as a defence mechanism to rid themselves of ectoparasites. Preening is the main form of grooming used and is effective against ridding birds from flies, ticks, lice as well as other types of ectoparasites (Villa *et al.* 2016). The development of physiological and immunological defence mechanisms requires birds to be in good condition since these defence mechanisms can be costly to the hosts. To be able to effectively deter parasites, these two defence mechanisms might include a combination of mechanical mechanisms (which includes the modification of the skin thickness and blood viscosity) and immunological mechanisms (which is the production of specific immunoglobulins or local hypersensitivity reactions). A combination of these mechanisms can increase the costs associated with the digestion of the host blood for parasites and it can also negatively impact the efficiency with which ectoparasites feed (Tschirren *et al.* 2007).

The objective of this study was to assess the seasonal body condition as well as the ecto- and haemoparasite prevalence on Red-billed Oxpeckers sampled in the southern region of Kruger National Park (KNP) during the dry winter and wet summer seasons. I hypothesised that Oxpecker body condition will be better during the summer months, when ungulate hosts harbour more ticks. I further expected birds to have higher ecto- and haemoparasites loads during the wet summer seasons, when conditions are suitable for insect survival and potential haemoparasites vectors, like mosquitoes and midges, are likely most abundant (Pérez-Rodríguez *et al.* 2015).

## MATERIALS AND METHODS

Study site

Red-billed Oxpeckers were caught at the animal enclosures (bomas) located at Skukuza (24°59'45.66"S, 31°35'30.96"E) in the KNP. The enclosures housed several White Rhinos (*Ceratotherium simum*) of various age groups, that together with their water troughs served as

a source of attraction for the ectoparasite gleaning Oxpeckers. Rhinos are also a highly preferred host ungulate for Oxpeckers (Ndlovu and Combrink 2015)

## Bird capture

Red-billed Oxpeckers were caught at the end of the dry season (July) and beginning of the wet season (November) to assess their body condition and parasite load. A series of mist-nets were set up along the elevated walkways of the animal enclosures. Nets were opened to catch birds from 06h00 - 11h00 in the morning and checked at regular intervals (every 5 mins) for any bird captures.

All captured birds were processed immediately at a ringing station set up close to the nets. The following morphometric measurements were recorded: body mass (to the nearest g), lengths of the head, culmen, tarsus and wings (to the nearest mm); and primary flight-feather moult status. Feather and blood samples were collected and screened for any parasites present on the bird. Each sampled bird was ringed with a uniquely marked South African Ringing Scheme (SAFRING) band.

## Haemo- and ectoparasite assessment

Haemoparasite assessments was done from blood samples collected from each bird caught. The bird was poked with a sterile needle, then blood was collected from the brachial vein using a capillary tube. A drop of blood was used to prepare thin blood smears. The smears were air dried, immediately fixed with absolute methanol and stained later on with 10 % Giemsa solution. Using a compound microscope, blood smears were screened for blood parasites. An area of the film approximately one third from the end of the slide was selected for examination, beginning with low magnification x100, x400 and then x1000 under immersion oil. Each slide was examined by moving two fields along the edge, two fields up and then two down observing and recording any parasites present. Photographs of all parasites and any abnormalities were

captured using analySIS getIT software (Version 5.1). Detected parasites were identified to genus level using Campbell (2015). Appendix 3 has photos of the haemoparasites detected.

Feather samples collected from the wing, back and chest of the bird were screened for feather ectoparasites. All ectoparasites on the section of the feather were counted and the area of the screened feather was measured to the nearest cm<sup>2</sup> using a leaf area meter (LI-300C area meter), and thereafter used to calculate ectoparasite loads (parasites per cm<sup>2</sup>). Photos of ectoparasites are in Appendix 4.

# Statistical analysis

The Body Condition Index (BCI) for each Oxpecker was calculated according to the formula from Ndlovu *et al.* (2010):

$$BCI = \frac{\sqrt[3]{body \ mass}}{\sqrt[2]{tarsus*culmen}} * 100$$

A T-test (two sample assuming unequal variance) was used to test for any significant differences in BCIs of Oxpeckers between the dry and wet seasons. This test was also used to test for significant differences in the body condition index of Oxpeckers between birds infected with haemoparasites and birds with no infection. Parasite prevalence was also compared between different seasons. Descriptive statistics tests were used to calculate the mean and standard deviation of the morphometric measurements recorded. Pearson's r test was used to determine if there was any significant difference in body condition between birds with and without ectoparasites. Chi-square test was used to assess for any differences in ectoparasites prevalent between seasons.

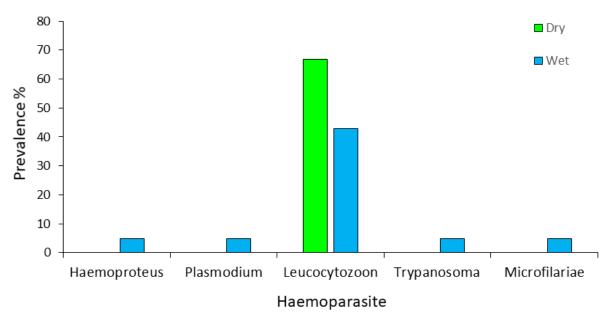
#### RESULTS

30 adult Red-billed Oxpeckers were caught during the course of this study, nine Oxpeckers in the dry season and 21 during the wet season (no recaptured birds between seasons). Out of the 30 Oxpeckers caught, 21 were found to be infected with either ecto- or haemoparasites or both. The mean ( $\pm$  SD) body mass of the caught Red-billed Oxpeckers was 47.23  $\pm$  4.17 g. Morphometric measurements revealed a small variation in body lengths of the sampled birds (Table 1).

**Table 1**: Mean (± Standard Deviation) morphometric measurements (n = 30) of the Red-billed Oxpeckers from the southern region of Kruger National Park.

	Body mass (g)	Culmen (mm)	Tarsus (mm)	Head (mm)
Mean	47.23	16.09	21.03	39.99
SD	4.17	1.05	0.85	1.32

There were no significant differences ( $t_{19} = -0.568$ ; p = 0.577) in BCIs of Oxpeckers sampled in the dry (mean BCI = 12.394) and wet (mean BCI = 12.49) season. 16 of the 30 Oxpeckers were found to be infected with haemoparasites. *Haemoproteus*, *Plasmodium* and *Leucocytozoon* spp. were among the haemoparasites present (Fig 3.1). *Leucocytozoon* was the most prevalent haemoparasite (66.7 % for dry season; 42.9 % for wet season) observed. Considering all haemoparasites, only *Leucocytozoon* had prevalence in the dry season. While screening for haemosporidian parasites, I also detected Trypanosoma and Microfilariae, although in very small numbers. There was no significant differences ( $t_{19} = 0.032$ ;  $t_{19} = 0.032$ ) in body condition (BCI) of Oxpeckers between birds infected with haemoparasites and birds not infected with haemoparasites.



**Figure 3.1**: Prevalence of haemoparasites found in Red-billed Oxpeckers sampled during the dry and wet season at Skukuza.

A total of 12 Oxpeckers were found to be infected with ectoparasites, with the majority of the parasites observed on the wing flight feathers of the birds. Only feather mites were detected (Appendix 4). It was found that Oxpeckers with more feather mites tend to have a lower body condition, although this trend was not statistically significant (Pearson's r = -0.23, p = 0.42). Also, although more Oxpeckers were infected with ectoparasites during the wet season (66%) than during the dry season (44%), this difference was not significant either ( $x^2_{1.1} = 1.29$ , p = 0.25).

# **Discussion**

The aim of this study was to gain a better understanding of the general health status of Redbilled Oxpecker, especially in terms of ecto- and haemoparasite prevalence between two seasons. No significant difference in BCI of the Oxpeckers was observed between the dry season (July) and the wet seasons (November), meaning that the overall body condition was similar for the birds between the two different seasons. Based on the results obtained it can be said that the original hypothesis that Oxpecker body condition would be better during the wet season was rejected, however, due to the small sample size this cannot be said with certainty. One factor that could influence this is the abundance and quality of the available food sources. Red-billed Oxpeckers prefer to feed on certain tick species, the blue ticks (Boophilus decoloratus) and brown ear tick (Rhipicephalus appendiculatus) (Robertson and Jarvis 2000; Weeks 2000). According to Horak et al. (2011), these tick species are present in the study area all year round. Nonetheless, Horak et al. (2011) further mentioned that adult tick numbers were low in Skukuza between February and July and only starting to increase from September again. However, they further explain that during the winter months, when adult tick numbers are low, there is an increase in questing larvae. Horak et al. (2011) speculated that larvae are active during this time because of the high average winter temperature (16 °C – 17 °C) of Skukuza. This high winter temperature is well above the threshold temperatures needed for larval development. Since Red-billed Oxpeckers feed on both adult ticks and their larvae, there seems to have been enough food available to keep up their body condition even during the dry months. Oxpeckers not only feed on ticks, they also feed on other ectoparasites found on their hosts' body (Weeks 2000; Ndlovu and Combrink 2015). An overall abundance of ectoparasites during the winter months could explain why Red-billed Oxpecker body condition did not decrease during the winter months.

Around 70 % of bird species worldwide are infected by haemoparasites from the genera *Plasmodium, Haemoproteus* and *Leucocytozoon*, negatively affecting these birds' reproductive success and survival rate (Grieves *et al.* 2018). It is likely that these species can be pathogenic in their natural hosts (Atkinson and Van Ripper 1991), however, most studies recording negative effects of these parasites to their avian hosts were performed under laboratory conditions and it is difficult to document the exact negative effects these parasites have on their wild avian hosts (Sanz *et al.* 2001). During this study it was found that there was no significant

difference in body condition between Oxpeckers infected with haemoparasites and those not infected. Townsend *et al.* (2018), explained that haemoparasite infection occur in two stages, the acute phase and the chronic phase. The acute phase is the stage where the host is initially infested with the parasite and the parasite numbers are likely to be the greatest, exerting the greatest pressure on the host. If the bird survives this phase, it enters the chronic phase where parasites occur in low numbers. The acute phase has stronger pathological effects on the host than the chronic phase. This could possibly explain why the body condition of Oxpeckers infected with heamoparasites did not differ significantly from those not infected.

Since parasite-host dynamics are dependent on seasonal variations, parasites have to adjust their lifecycles so that they reduce the negative effects on hosts during periods of minimal transmission. Consequently, many parasites enter a dormant stage during the dry season, only reappearing when conditions are favourable (Pérez-Rodríguez *et al.* 2015). Numerous studies have shown that parasites time their own reproduction so that it coincides with that of their hosts, which typically falls between spring and summer. This also coincides with the time when vectors have a high abundance (Møller *et al.* 2003; Pérez-Rodríguez *et al.* 2015). This will cause an increase in parasite abundance. This is in contrast to what was observed during this study. Even though, more Oxpeckers were infected with ectoparasites during the wet season, the difference between the two seasons was not significant. This could possibly be due to the relatively warm winter temperatures of the study site (Horak *et al.* 2011). Mild temperatures during the winter means that conditions could be favourable for the parasites which could explain why there was little difference in Oxpeckers infected between the two seasons.

Some ectoparasites, like feather lice, can have a dire impact on the fitness levels of their hosts when they occur in large numbers. These parasites cause damage to the feathers, which leads to an increase in stress for these birds. The result of these high stress levels in the avian

hosts causes a decrease in body mass of the hosts and also affecting their overwintering survival rate (Moyer *et al.* 2002). This is similar to what was observed during this study. There was a slight difference in body condition between birds infested and those not infested, where infested birds had a slightly lower body condition. My analysis show that these findings were not significant, however, due to the small sample size it is difficult to draw definite conclusions. However, the slight difference in body condition could also be an indication that Red-billed Oxpeckers have started to build up immunity against these parasites and, therefore, barely had a negative reaction towards the presence of the mites.

In conclusion, there is still more information to be gathered regarding Red-billed Oxpeckers and their parasites, but this study gave a good indication of which parasites are carried by Oxpeckers and how they affect these birds. Red-billed Oxpeckers serve as a host for a wide variety of ecto- and haemoparasites and these parasites each play their own role in how they affect the behaviour and morphology of their hosts. Since haemoparasites did not have severe effects on the Oxpeckers, it is possible that these birds build up immunity against these parasites. However, it is difficult to draw definite conclusions, since no previous published work has been done on Oxpecker health. These results, however, do provide a crucial baseline that will aid in the conservation and ecology of Red-billed Oxpeckers.

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#### CHAPTER FOUR

# **Synthesis**

In the last few decades, there has been a growing interest in the conservation of both the Redbilled and Yellow-billed Oxpeckers (Kalle et al. 2017). Since Oxpeckers nearly became extinct in southern Africa during the first half of the 20th century, numerous studies have focused on efforts to conserve these birds (Mellanby et al. 2009; Plantan et al. 2009; Ndlovu and Combrink 2015; Kalle et al. 2017; Tarakini et al. 2017; Diplock et al. 2018). Oxpeckers play an important role in removing ectoparasites from ungulates (Kalle et al. 2017). Hence, some authors have highlighted the possibility of using Oxpeckers as part of an integrated pest management program to reduce tick loads on ungulates, especially on wild animals (Bezuidenhout and Stutterheim 1980; Samish et al. 2004). Oxpeckers can feed on up to 12 500 larvae or 100 engorged adult ticks per day (Samish and Rehacek 1999; Samish 2000; Hockey et al. 2005; Jubber 2014), making them a useful tool in biological control of ticks. Some reintroduction efforts have been carried out and researchers have noted that areas where Oxpeckers have been introduced, there has been a notable decrease in tick numbers (Samish et al. 2004). In areas where Red-billed Oxpeckers were reintroduced, tick abundance on hunted ungulates decreased and farmers also reported a reduction in livestock calf mortality caused by tick borne diseases (Samish and Rehacek 1999; Samish et al. 2004). When reintroducing Red-billed Oxpeckers, it is not only important to look at the habitat requirements of these birds, but also their foraging behaviour and health status. Thus, the main aim of my study was to understand the feeding ecology of the Red-billed Oxpecker in the southern region of the Kruger National Park. I also assessed how season affect their health, i.e. body condition, parasite (ecto- and haemoparasites) prevalence and immune response.

The first objective was to document Red-billed Oxpecker feeding behaviour in terms of host selection and foraging location on the host, while also looking at seasonal variation (Chapter two). This involved looking at different ungulates found in the southern section of the Kruger National Park, specifically those that Oxpeckers forage on. I also assessed the foraging location on the host to see if Oxpeckers fed on different locations of the body of different host species. These findings were also compared and contrasted amongst three different seasons, to determine whether seasons influenced foraging behaviour of Red-billed Oxpeckers. Numerous studies have looked into the feeding behaviour of these birds (Hart et al. 1990; Plantan et al. 2012; Ndlovu and Combrink 2015), however, most of these studies were conducted over a single season. My study took it a step further and evaluated their foraging behaviour in three consecutive seasons of the same year. Findings revealed that Oxpeckers foraged from a wide range of suitable hosts, and where present, they preferred to forage on White Rhinos (Ceratotherium simmum), Giraffes (Giraffa camelopardalis) and Cape Buffalo (Syncerus caffer). These results were similar to those of previous studies done in summer in the southern regions of the Kruger National Park a few years ago (Ndlovu and Combrink 2015). There was however a slight difference in host selection amongst seasons. Oxpeckers were not seen on White Rhino during November (early wet summer season), whereas this host was one of the most preferred ungulates during the previous dry winter season. It could possibly be due to tick abundance or host tolerance behaviour or it could also be due to the fact that Red-billed Oxpeckers breed during the summer (Hockey et al. 2005). Another important factor that should be considered is the number of White Rhinos recorded. In April only nine White Rhinos were recorded and similary in July eight animals were recorded. However, during November only four White Rhinos were recorded. This could also possibly be the reason why Oxpeckers were not seen on this particular host species. There seems to also have been a slight difference between foraging location on the hosts' body between different host species. Some ungulates,

like the White Rhino and Cape Buffalo, had more Oxpeckers observed on the back of these hosts, whereas for the Giraffe and Kudu, most birds were seen foraging around the neck area. Previous studies observed similar behaviour and explained that this difference is due to the self-grooming ability of the host (Weeks 1999; Bishop and Bishop 2013, Ndlovu and Combrink 2015).

The second objective looked at fluctuations in local Oxpecker population by monitoring the seasonal bird abundance (Chapter two). I hypothesised that Oxpecker abundance would be higher during the wet summer seasons when tick abundance was higher, but no significant difference was found for Oxpecker abundance amongst the different seasons, thus the hypothesis was rejected. Oxpecker abundance was stable throughout the year, which means that Red-billed Oxpeckers are resident species in this area and may as well have sufficient food sources throughout the year. Oxpecker abundance and distribution is also dependent on ungulate abundance and distribution (Stutterheim and Brooke 1981) and as long as their numbers are stable, little fluctuations would be observed in Oxpecker numbers.

Lastly, I focused on Red-billed Oxpecker health status. Assessing the seasonal body condition, immune status and ecto- and heamoparasite prevalence on Oxpeckers (Chapter three). To my knowledge, no published work has been done regarding the health status of Oxpeckers, neither for the Yellow-billed nor the Red-billed Oxpeckers. One article mentioned blood being collected from Red-billed Oxpeckers as part of the Endangered Wildlife Trust reintroduction program (Spies et al. 2012). However, the blood samples were used for DNA sequencing and not for parasite infection assessments. Parasites dispersal is of concern among communities, especially if there is a possibility of spreading infectious diseases from one area to another during bird translocations and reintroductions. Haemoparasites (*Plasmodium*, *Haemoproteus* and *Leucocytozoon*) were detected in the blood of the Red-billed Oxpeckers caught and thus relocating these birds increases the possibilities of also spreading previously

undescribed parasites to new areas. The introduction of avian haemoparasites into naïve avian hosts in Hawaii caused a massive die out of local bird populations (Braga *et al.* 2011). There are also fears that isolated birds, like penguins, are especially vulnerable to such new infection (Piersma 1997, Clark *et al.* 2009, Braga *et al.* 2011). Hence, the current Oxpecker relocations without a prior haemoparasite screening of the moved birds may pose a serious disease risk to the existing bird populations where Oxpeckers are introduced.

Parasites that lack free-living stages, can only be spread from one area to another through their vectors or their hosts (Ricklefs *et al.* 2016). *Leucocytozoon* was the dominant avian haemoparasite detected in Oxpeckers. It was also the only haemoparasite that was found during the dry season. In contrast to what was hypothesised, haemoparasites did not seem to adversely affect the body condition of the birds. An indication that the birds have perhaps developed immunity towards these infections. This is expected with Afro-tropical species that have co-evolved with endemic haemoprotozoan infections.

# Limitations

One of the limitations of the study arise from drawing generalised inferences about Oxpeckers in the sub-Saharan region, since my sample sizes were limited both in spatio-temporal distribution and in the numbers of birds screened. It was not logistically possible to expand this study within the time constraints of an MSc study to a much larger geographic range with repeated seasonal sampling. Even though the sample size of birds caught was relatively small, a good indication (though not exhaustive) of some of the haemoparasites that infect Oxpeckers and how these affect their body condition was certainly established. The last problem could have been in the detection of haemoparasites using the traditional microscopic method. In any case, my reported prevalence is a conservative estimate and further results up to species level will be confirmed when these same blood samples I collected are screened using molecular

PCR methods. That screening aspects is still ongoing as it was hampered by lack of funds during my study. However, I am confident that my study did elucidate and confirm some of the previously unknown feeding behaviours, seasonal abundance and health status of birds. I therefore recommend that follow up studies should collect data over multiple years during both drought and wet periods so as to be able to make definitive assessments of the Red-billed Oxpecker biology.

Given that the Kruger National Park has a wide variety of ungulate species (Chirima *et al.* 2012) which could be possible foraging hosts for Oxpeckers. Not all possible host species were encountered and observed during my road transects. Hence the Oxpecker preference for elusive ungulates (Those animals that may avoid roads) could have been omitted.

#### **Future Research**

An important part of Oxpecker conservation is the reintroduction of these birds into areas where they used to occur. Successful reintroduction of these birds ensures the long-term survival of this species. Having a good understanding of Oxpecker feeding behaviour and health status will be an important aspect in conserving these birds. My study covered an extensive part of Oxpecker behaviour, however, there is still a lot to be learned such as their behaviour in new landscapes so as to ensure a successful relocation. It is also important to observe their behaviour under different environmental conditions i.e. droughts, floods, fire, so as to see how this will impact their abundance and distribution. It would also be important to screen for other parasites prevalences such as ecto- and endoparasites which will undoubtedly affect their survival in different environments. Since numerous Red-billed Oxpecker translocations have been done, it would be good to go to these areas and study how they establish as well as adapt to different environments and foraging host species. With time it will also be helpful to look at their foraging behaviour and health status to see if it differs from the

source population in the Kruger national park. Such information will be useful in ensuring successful translocations and conservation programs.

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# **APPENDICES**

# Appendix 1

Table 1: The Oxpecker and host abundance for each day sampled during the three seasons in southern section of the Kruger national park sampled during the three seasons, late wet (April), dry (July) and early wet (November).

Season	Time	Day	Oxpecker abundance	Host abundance	
Late wet	AM	9 April 2017	0.298	2.536	
Late wet	PM	9 April 2017	0.182	2.491	
Late wet	PM	10 April 2017	1.347	5.859	
Late wet	AM	11 April 2017	0.262	6.452	
Late wet	PM	11 April 2017	0.138	2.203	
Late wet	PM	13 April 2017	0.554	5.569	
Dry	AM	9 July 2017	0.438	5.479	
Dry	PM	9 July 2017	0.786	7.431	
Dry	AM	10 July 2017	0.141	2.518	
Dry	AM	11 July 2017	0.255	2.324	
Dry	AM	12 July 2017	0.170	2.576	
Dry	AM	17 July 2017	0.578	4.706	
Early wet	AM	14 November 2017	0.080	6.773	
Early wet	PM	14 November 2017	0	3.591	
Early wet	AM	15 November 2017	0.256	5.500	
Early wet	AM	16 November 2017	0.208	3.380	
Early wet	AM	17 November 2017	0.313	3.346	
Early wet	AM	20 November 2017	0.174	5.073	

# Appendix 2

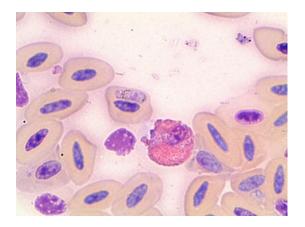
Table 2: Haemoparasites, ectoparasites count per cm<sup>2</sup> and BCI of Red-billed Oxpeckers in the southern section of Kruger National Park during the dry season (July) and the wet season (November.

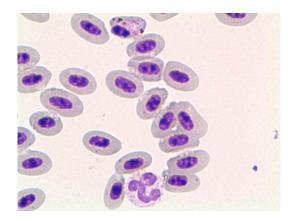
Season	Bird ID	Haemoparasites					Ectoparasite count per cm <sup>2</sup>			BCI
Season	BIIQID	Haemoproteus	Plasmodium	Leucocytozoon	Trypanosoma	Microfilariae	Back	Chest	Wing	БСІ
Dry	SKU390			1					0.643	12.722
Dry	SKU413			1						12.01
Dry	SKU418			1						12.302
Dry	SKU419			2						11.581
Dry	SKU420						1.553			12.502
Dry	SKU421			1						12.406
Dry	SKU427			1				1.31	12.802	12.951
Dry	SKU433							6.382		12.603
Dry	SKU441									12.472
Wet	SKUA011									12.038
Wet	SKUA012			1				1.748	38.975	10.634
Wet	SKUA013							3.226	0.572	12.188
Wet	SKUA014						0.649		0.385	12.688
Wet	SKUA016			1					18.524	12.882
Wet	SKUA017								81.686	12.46
Wet	SKUA019								2.044	12.725
Wet	SKUA028								6.167	12.602
C	Bird ID			Haemoparasites			Ectoparasite count per cm <sup>2</sup>			BCI
Season	Bird ID	Haemoproteus	Plasmodium	Leucocytozoon	Trypanosoma	Microfilariae	Back	Chest	Wing	
Wet	SKUA029			1						13.266
Wet	SKUA030									12.55
Wet	SKUA033								0.326	12.642
Wet	SKUA034		2	1			0.833			12.793
Wet	SKUA035			1					27.293	12.471
Wet	SKUA036	1		1					112.944	12.548
Wet	SKUA041							1.486	0.326	12.611
Wet	SKUA042									12.055
Wet	SKUA043			1						12.706
Wet	SKUA051									12.32
							I			

		Haemoparasites						Ectoparasite count per cm <sup>2</sup>		
Season	Bird ID									BCI
		Haemoproteus	Plasmodium	Leucocytozoon	Trypanosoma	Microfilariae	Back	Chest	Wing	
Wet	SKUA052			1		2	2.611			12.454
Wet	SKUA053			1					0.103	13.052
Wet	SKUA054				1					12.687

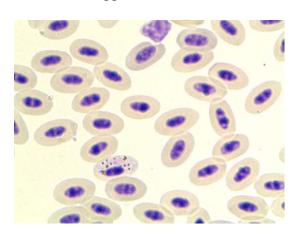
# Appendix 3

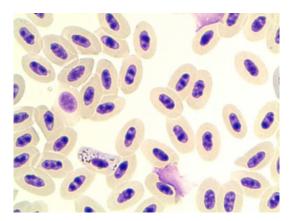
Photos of haemoparasites detected in the blood smear of Red-billed oxpeckers that are found in the southern section of the Kruger National Park



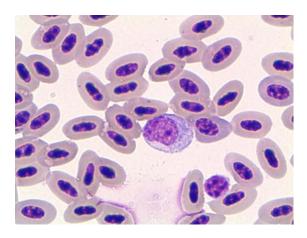


Plasmodium spp found in a blood smear of Red-billed Oxpecker.





Haemoproteus spp found in the blood smear of Red-billed Oxpeckers.

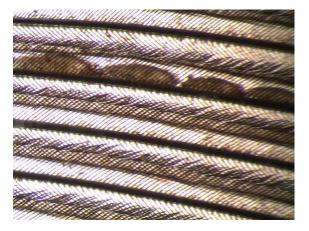


Leucocytozoon spp found in the blood smear of Red-billed Oxpeckers.

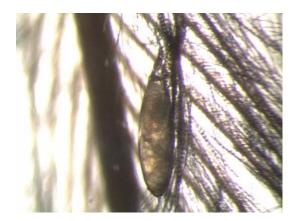
# Appendix 4

Photos of the ectoparasites detected on the wing, chest and back feathers of Red-billed Oxpeckers found in the southern section of Kruger National Park.

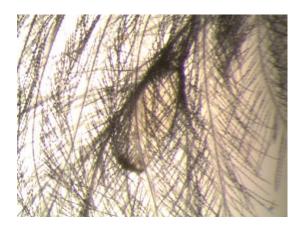




Feather mites found on the wing feather of Red-billed Oxpecker.



Feather mites found on the chest feather of Red-billed Oxpecker.



Feather mites found on the back feather of Red-billed Oxpecker.