BIOLOGGING AS A METHOD TO REMOTELY DETECT ORIENTATION TO SOLAR RADIATION IN BLACK AND BLUE WILDEBEEST

By

Arista Botha

A dissertation submitted to the University of the Witwatersrand in fulfilment of the requirements for the degree of Master of Science in Medicine

Supervisors:

Andrea Fuller

Robyn S. Hetem

Hilary M. Lease

Johannesburg 2014

Declaration

I, Arista Botha, declare that this dissertation is my own work, with all assistance acknowledged. It is being submitted for the degree of Master of Science in Medicine in the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination at this or any other University.

……………………..

(Arista Botha)

……………… day of ………….. 2014

The procedures used in this dissertation were approved by the Animal Ethics Screening Committee of the University of the Witwatersrand (Clearance number: 2011/09/04).

Conference presentations arising from this study

Poster Presentations (local):

Van Staden, A., Lease, H.M., Fuller, A., Hetem, R.S. Validation of a novel bio-logging method to remotely detect orientation to solar radiation. *Royal Society of South Africa Spring Festival, September 2012*, Delta Environmental Centre, Johannesburg, Gauteng (2nd place).

Van Staden, A., Lease, H.M., Fuller, A., Hetem, R.S. Validation of a novel bio-logging method to remotely detect orientation to solar radiation. *Faculty of Health Research day, September 2012*, Medical Campus, University of the Witwatersrand, Johannesburg, Gauteng.

Van Staden, A., Lease, H.M., Fuller, A., Hetem, R.S. Validation of a novel bio-logging method to remotely detect orientation to solar radiation. *Cross-faculty Graduate Symposium, October 2012,* Postgraduate Development Hub, University of the Witwatersrand, Johannesburg, Gauteng (3rd place).

Oral Presentations (local):

Botha, A., Lease, H.M., Fuller, A., Mitchell D., Hetem, R.S. Biologging as a novel technique to detect and quantify body orientation to solar radiation in black and blue wildebeest. *Zoological Society of Southern Africa, July 2013*, Tshipise Forever Resorts, Musina, Limpopo.

Botha, A., Lease, H.M., Fuller, A., Mitchell D., Hetem, R.S. Biologging as a novel technique to detect and quantify body orientation to solar radiation in black and blue wildebeest. *Cross-faculty Graduate Symposium, August 2013*, Postgraduate Development Hub, University of the Witwatersrand, Johannesburg, Gauteng.

Abstract

Animals can shift their orientation to solar radiation to adjust the amount of body surface area that is exposed to solar radiation, thereby manipulating the amount of radiant heat they absorb from their environment. This behaviour is especially important in animals that need to graze out in the open during the day, such as wild ungulates. All previous studies of orientation to solar radiation in animals have relied on visual observations. The problem with visual observations is that animal behaviour, including animal orientation, can be affected by human presence. Therefore I set out to develop a remote technique to detect and quantify orientation to solar radiation in wildebeest to eliminate the need of a human observer. I hypothesised that if an animal was orientated perpendicular to solar radiation, the side facing the sun would be hotter than the opposite side. In contrast, if the animal was orientated parallel to solar radiation I hypothesised that both sides will have a similar temperature.

To test my hypothesis, temperature-sensitive data loggers were implanted subcutaneously into free-ranging black (*Connochaetes gnou*) and blue wildebeest (*Connochaetes taurinus*) from Mokala National Park and their orientation to solar radiation was determined visually. I found that when wildebeest were orientated perpendicular to solar radiation, there was a greater difference between the left and right subcutaneous temperature than when wildebeest were orientated parallel to solar radiation (t_7=2.5, p=0.04). However, using subcutaneous temperature difference on its own to predict orientation to solar radiation could not account for how the previous orientation to solar radiation of wildebeest affected subcutaneous temperature patterns. Therefore, I designed a prediction model incorporating both subcutaneous temperature difference and rate of change in subcutaneous temperature difference to determine orientation to solar radiation.

The prediction model was accurate more often than expected by chance (60 %), but there were many factors other than solar radiation that influenced subcutaneous temperature, which reduced the accuracy of the remote technique. Further research is necessary to improve the remote technique before it can be successfully used to study orientation to solar radiation. However, my study shows, for the first time, the potential of using subcutaneous temperatures to remotely detect orientation to solar radiation in ungulates. A remote technique to study orientation to solar radiation will be a great advantage for future studies on thermoregulatory behaviour. Because behavioural responses are likely to be an animal’s first defence against increased heat loads resulting from climate change, studying behavioural thermoregulation could provide important information for conservation and management decisions.

Acknowledgements

I would like to thank all the people who made this project possible:

South African National Parks (SANParks), thank you for allowing me to use Mokala National Park as my study site. Thank you to the following Mokala staff members: Deon Joubert and Tess White, for always being friendly and helpful on the organisational side of things, and all the game rangers who helped me in the field: Abel Ramavhale, Stephan Prins, Jared, Richard and George.

The excelent capture team of SANParks Veterinary Wildlife Services: Charles Thompson, Cathy Dreyer, Craig Williams, Rudi Williams, Dave Zimmerman, Markus Hofmeyr and Angela Bruns, for the capture of the animals and your assistance during the surgery.

Everybody who was part of the surgery: The veterinarians, Leith Meyer and Anna Haw, the veterinary nurse, Mary-Anne Costello, and the people who assisted, Robyn Hetem, Benjamin Rey, Zipho Zwane, Claire Costello and Ian Murray. I appreciate your hard work.

To everyone who assisted with the field observations: Gerhard Vermaak, Theunis Botha, Ian Murray, Piet and Annie Rossouw, and Petrus and Tinkie van Staden. Thank you for giving up your time to help me. Your assistance and company was greatly appreciated.

My supervisors: Hilary Lease, Robyn Hetem and Andrea Fuller. Thank you for all your hard work and for everything that you have taught me. To Duncan Mitchell, thank you for your advice and intellectual input. To Richard McFarland, thank you for your help and advice on statistical analyses.

I would like to acknowledge the financial assistance of the National Research Foundation and the Faculty Research Committee of the Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, South Africa.

And finally, to my husband, Theunis Botha, and my parents Petrus and Tinkie van Staden. Without your emotional and financial support this would not have been possible. Thank you for your love and encouragement.

Contents

List of figures

List of tables

Chapter 1

Introduction

1 Introduction

Orientation to solar radiation is a very effective behavioural thermoregulatory mechanism used by animals to reduce or increase the heat load they experience. By altering the orientation of their body relative to solar radiation, animals can regulate the amount of surface area of their body exposed to solar radiation, in order to manipulate the amount of radiant heat they absorb from their environment. Orientation to solar radiation has been studied in a wide variety of ectotherms (Kevan and Shorthouse, 1970, Muth, 1977, Adolph, 1990, Coe, 2004, Munoz et al., 2005) as well as in endotherms (Bartholomew and Dawson, 1979, Gonyou and Stricklin, 1981, Kennedy and Day, 1989, Fortin et al., 2000, Maloney et al., 2005b, Keren and Olson, 2006, Hetem et al., 2011b).

Behavioural thermoregulatory responses such as orientation to solar radiation might be very important for the continuing survival of animal species, especially in the light of current rapid climate change. According to current climate change predictions (IPCC, 2013), many South African animals will have to adapt to warmer, drier environments if they are to survive in their current locations. Because behavioural responses are immediate and cost-effective, they are likely to be an animal’s first response to climate change. Therefore, it is important to study behavioural thermoregulation using accurate techniques.

Previous studies investigating orientation to solar radiation in endotherms have relied on behavioural observations (Berry et al., 1984, Vrahimis and Kok, 1992, Maloney et al., 2005b, Hetem et al., 2011b). However, the presence of a human observer can cause animals to orientate towards the human observer instead of orientating to solar radiation for thermoregulatory purposes. Therefore, it would be beneficial to develop a remote technique, whereby animal orientation to solar radiation can be studied without the presence of a human observer.

Recent technology, such as camera traps (Bashir et al., 2013, Ramsdell, 2013, Ross et al., 2013), GPS tracking (Eriksen et al., 2011, Van Beest et al., 2012, Lake et al., 2013, Lindberg, 2013, Whyte et al., 2013) and implanted data loggers (Cooke, 2008, Ropert-Coudert et al., 2009, Rutz and Hays, 2009, Bograd et al., 2010), has enabled biologists to study animals remotely. In this dissertation, I investigate the possibility of using implanted temperature-sensitive data loggers to study orientation to solar radiation by measuring the subcutaneous temperatures of wildebeest. This introductory chapter is an overview of the importance of behavioural thermoregulation for animal survival in the light of global climate change, different behavioural thermoregulatory strategies available to animals, particularly orientation to solar radiation, and the current methods that are being used to study thermoregulatory behaviour. The introductory chapter concludes with the hypotheses that form the basis of my study.

1.1 Climate change predictions and possible consequences for animals

Global predictions for climate change indicate an overall increase in ambient temperatures (IPCC, 2013). Specific predictions for southern Africa indicate ambient temperature increases between 1 ºC and 5 ºC by 2100, decreased winter rainfall in winter rainfall areas, and delayed onset of summer rainfall in summer rainfall areas (IPCC, 2013). Therefore, many animals in South Africa will have to cope with higher environmental heat loads and less drinking water. To prevent extinction, species can either shift their distributions to follow the ideal climate, or animals can remain in their current distribution and adapt to the changing environment.

By shifting their distribution patterns, animals can avoid climate change by moving to a place with more suitable conditions. Many animals have been observed to shift their distribution towards higher altitudes and poleward latitudes as a result of recent climate change (Parmesan and Yohe, 2003, Hickling et al., 2006, Parmesan, 2006, Chen et al., 2011). There is a lot of literature available on range shifts in invertebrates, especially butterflies (Parmesan et al., 1999, Warren et al., 2001) and pest species (Carroll et al., 2003). Herpetological species in the tropical montane forests of Madagascar showed an overall upwards shift in elevation of their distribution (Raxworthy et al., 2008). There are also several examples of range shifts in mammalian species. For example, northward distribution shifts have been recorded in mammals in the United Kingdom (Hickling et al., 2006). Red fox distributions have shifted northward, and, supposedly in response to inter-predatory competition from the red fox, so have the distribution of the arctic fox (MacPherson, 1964, Hersteinsson and MacDonald, 1992, Tannerfeldt et al., 2002). In Yosemite National Park (California), small mammals have shown a general upwards distribution shift in elevation (Moritz et al., 2008). A meta-analysis on biological responses to climate change indicates that, overall, animal movements are significantly related to the movements that would be expected as a response to climate change (Rosenzweig et al., 2008).

However, range shift is not an option for all animals. Shifting distribution is often difficult or impossible for animals due to species specific characteristics of the animal or due to geographic or man-made boundaries preventing movement. For example, in a study of multiple British butterfly species the mobile, generalist species were more likely to colonise new habitats than the less mobile habitat-specialist species (Warren et al., 2001). Animals that live in isolated habitats may be separated from other possibly suitable habitats by geographic barriers such as rivers or mountain ranges, which make it impossible for them to migrate. Animals that live at the top of mountains or in the Arctic are especially at risk, because they already live on the edge of their suitable habitats. If their current habitats warm, these animals would have nowhere cooler to go to follow suitable climate (Chen et al., 2011).

Man-made barriers also have made it difficult for animals to shift their distribution, because fences and cities can prevent animal mobility. Small animals can move through fences and flying animals such as birds can fly over barriers, but many large mammals are restricted by fences. For example, in southern Africa fences prevent seasonal migration of wildebeest towards water resources and forage sites and have had a negative impact on wildebeest populations (Williamson et al., 1988, Berry, 1997). If large mammals cannot move to a more appropriate climate within the fenced area, their survival is threatened. Climate envelope modelling predicts that animals that don't have the ability to shift their distribution have a greater chance of extinction than animals that can shift their distribution (-52 % compared to 9-32 %; Thomas et al., 2004). Climate envelope predictions, specifically for South African species, suggest a decline in species richness across different animal taxa in the western, arid regions of the country because of distribution shifts towards the eastern part of the country towards increasing altitude for many species (Erasmus et al., 2002). Climate envelope modelling considers the climate of an animal's current distribution, together with climate change predictions to estimate how far an animal would need to move to follow its suitable habitat in response to climate change. These models assume that an animal can only survive in climatic conditions such as those of its current distribution. Therefore, if the animal is restricted in its movement to follow suitable habitats, its range will decrease or the animal will die out completely.

However, climate envelope models predicting species extinctions do not take adaptations into account. Animals that are limited in their movements could use genetic or phenotypic adaptations in response to environmental changes in order to survive.

When the environment changes, certain individual animals within a species have particular characteristics that increase their chances to survive and carry their genes to the next generation, ensuring the survival of that species. This selection for certain traits is known as genetic adaptation or micro-evolution. Genetic adaptations in response to climate change can include shifts in timing of life history events, such as breeding and migration (Bradshaw and Holzapfel, 2008). For example, pitcher plant mosquitoes (*Wyeomyia smithii*) shifted their photoperiodic response, which controls their diapause, in response to longer growing seasons due to climate change (Bradshaw and Holzapfel, 2001).

When observing adaptive responses in animals it is often difficult to determine whether the response is due to genetic adaptation or a plastic response to environmental factors (Gienapp et al., 2008). For example, timing of reproduction can be genetically programmed, but sometimes species can adjust the timing of reproduction in response to environmental cues. In red squirrels (*Tamiasciurus hudsonicus*), it was found that breeding time shifted earlier within generations in response to increasing food availability (plastic response). The same study found that breeding time was also controlled by genetics, and that there was a strong selection towards earlier breeding time between generations (genetic adaptation; Reale et al., 2003).

Animals with short lifespans can adapt genetically faster than can long-lived animals, because animals with short lifespans have short generation times. Rate of evolution is related to generation time, therefore long-lived animals face a greater risk of extinction than do short-lived animals (Skelly and Freidenburg, 2010). Because short-lived animals can have many offspring in a short time span, there is potential for many genetic changes to occur within decades. For example, in fruit flies (*Drosophila* spp.), genetic changes associated with climate change adaptation have been observed over the course of only twenty years (Anderson et al., 2005, Umina et al., 2005, Van Heerwaarden and Hoffmann, 2007). Long-lived mammals, such as antelope, only reproduce once a year, and mostly only produce one offspring a year (Skinner and Chimimba, 2005), whereas short-lived mammals, such as house mice, can reproduce multiple times a year and produce many young at a time (Berry and Bronson, 1992). Because most large mammals are long-lived animals, they take longer to adapt genetically to environmental change (Fuller et al., 2010). Currently climate change is happening at a faster rate than previous changes in climate, therefore animals will have to adapt to changing environmental conditions at an unprecedented rate in order to survive (Quintero and Wiens, 2013).

Many long-lived mammals are already struggling to adapt to the consequences of climate change. For example, polar bears (*Ursus maritimus*) are losing condition as increasing ambient temperatures cause earlier ice-melting time and shorter feeding time on the ice (Stirling et al., 1999). Moose (*Alces alces*) are more prone to heat stress than cold stress (upper thermal limit of -5 to 0 ºC in winter and 14-20 ºC in the summer; Renecker and Hudson, 1986) so an increase in ambient temperature could threaten their survival. Because they are long-lived animals, they are unlikely to respond fast enough genetically to climate change. In addition, both polar bears and moose are unlikely to be able to shift their distribution to follow suitable habitat, because they live on the edge of their suitable habitats with no place to move northwards. Therefore, the survival of their species is threatened by current climate change.

If an animal is unable to migrate or to adapt genetically, their only other option to prevent extinction is to adapt by using phenotypic plasticity (Fuller et al., 2010). Although long-lived mammals require more time to adapt genetically to environmental change than do short-lived animals, they may have the plasticity to adapt phenotypically. Phenotypic plasticity is the flexibility of an animal's phenotype to respond to environmental factors, without any changes in the genetic make-up of an animal. Genetic adaptations happen slowly, over generations, whereas plastic responses enable animals to respond to environmental factors within their lifetime. For example, lizards can adjust their life history patterns such as growth, development, activity times and reproduction in response to environmental factors such as food availability and daily or annual environmental temperature rhythms (Adolph and Porter, 1993). In certain reptiles, sex is determined by the temperature experienced by the embryo during incubation. A skewed sex ratio due to increased ambient temperatures can be detrimental to the survival of the species. Female turtles (*Chrysemys picta*) choose nesting sites based on the amount of vegetation cover over the nesting site (Janzen and Morjan, 2001, Refsnider and Janzen, 2012). If this behaviour is plastic, and female turtles can select nesting sites with greater amount of shade in response to increased ambient temperatures, this behaviour could buffer the effect of climate change on temperature-sensitive sex determination (Refsnider and Janzen, 2012). However, if this behaviour is fixed, genetic adaptation would be required to prevent a skewed sex-ratio due to increased nest temperatures. In a different example, grey wolves (*Canis lupus*) can increase their pack sizes to increase their hunting success in times when heavy snowfall can make hunting difficult (Post et al., 1999). Increasing pack size is an example of an immediate behavioural response in wolves to climatic changes.

Because phenotypic plasticity does not require genetic changes, there is a lot of scope for adaptation in a short time. Since it seems likely that most large mammals will not be able to migrate or adapt genetically due to their long generation time, phenotypic plasticity could be their only chance for survival. The extent to which phenotypic expression could enable animals to adapt to changing climate is not well studied. To be able to predict whether an animal might be able to adapt, it is essential to understand the phenotypic plasticity of an animal in terms of its ability to respond to specific environmental changes, which may then help inform conservation and management decisions. One of the most important adaptations that animals would have to make in the light of global climate change is to adapt to a different thermal environment. There are various plastic responses that endotherms can use to thermoregulate if climatic conditions change.

1.2 Different thermoregulatory adaptations of animals

Thermoregulation is when an animal regulates its body temperature by using either autonomic or behavioural mechanisms. Every animal has an ideal range of body temperatures, within which it functions optimally. If body temperature increases above this thermoregulatory zone, consequences of hyperthermia include deactivation, and, ultimately, denaturation of enzymes, and reduced oxygenation of cells and tissues (Schmidt-Nielson, 1997). Similarly if an animal's body temperature drops below its thermoregulatory zone, they will experience reduced metabolic processes and decreased functioning of vital organs such as the heart and lungs (Collins, 1983).

Endotherms typically maintain their body temperature within a narrow range (between 36-40 °C for eutherian mammals; Schmidt-Nielson, 1997) by using autonomic thermoregulation. High body temperature enables optimal functioning of enzymes and metabolic pathways (Heinrich, 1977). Maintaining high body temperatures enables endotherms to remain active regardless of the environmental temperature (Bennett and Ruben, 1979), unlike in ectotherms, such as lizards, where activity time is largely limited by the time that environmental temperatures are within ideal range (Adolph and Porter, 1993). Higher body temperatures and sustained activity increase the ability of endotherms to provide warmth and food for their young (Farmer, 2000). Autonomic responses include blood flow redistribution, metabolic heat production and evaporative heat loss.

1.2.1 Autonomic thermoregulation

Endotherms can redistribute their blood flow to facilitate heat loss or heat uptake through either vasodilation or vasoconstriction. Circulatory redistribution is an economical form of autonomic thermoregulation for animals to respond to changes in their thermal environment. By increasing peripheral blood flow in certain body parts (vasodilation), animals can increase heat loss at the skin surface in response to increased environmental temperatures (Hales, 1983). A study done on humans shows that vasoconstriction in response to a drop in deep body temperature can reduce heat lost to the environment at the skin surface (Sessler et al., 1990).

Metabolic heat production is when endotherms use the energy they get from food or stored energy reserves to produce heat. In cold environments, producing enough heat to maintain homeothermy can be energetically costly. In very cold conditions, the energy that would otherwise go to growth, maintenance or reproduction would be used to produce heat instead, and the animal could lose body condition. For animals to produce enough heat during extreme cold periods, they would have to compensate by increasing food intake, dropping their metabolism or dropping their body temperature. Increasing the quantity or the energy content of food intake can be difficult in harsh winter conditions when there is limited food available. Hypometabolism is a combination of reducing metabolism and reducing body temperature to save energy during cold periods. Hypometabolism during harsh winter conditions (low ambient temperatures combined with limited food availability) has been inferred from reduced body temperatures in horses (*Equus ferus caballus*; Brinkmann et al., 2012), and reduced body temperature combined with reduced heart rate in ibex (*Capra ibex ibex*; Signer et al., 2010) and red deer (*Cervus elaphus*; Arnold et al., 2004). Hibernation and torpor are extreme forms of hypometabolism usually found in small mammals and have been detected as a drop in skin temperature in horseshoe bats (*Rhinolophus ferrumequinum;* Park et al., 2000) and dwarf lemurs (*Cheirogaleus spp.;* Blanco et al., 2013), and by measuring body temperature and metabolic rate in golden spiny mice (*Acomys russatus*; Grimpo et al., 2013).

In hot conditions, an animal needs to get rid of excessive heat absorbed from the environment together with the metabolic heat produced by its body to maintain homeothermy. Endotherms can lose heat passively through convection, conduction or radiation (Mount, 1979). However, if the air temperature exceeds the animal's body temperature, the only way an endotherm can lose heat to the environment is through evaporative heat loss. Evaporative heat loss is a very effective way of losing excessive body heat to the environment (Taylor et al., 1969) and includes sweating and panting. Water evaporates from the surface of the skin or airways of an animal and causes the animal to cool down (Taylor et al., 1969). However, evaporative cooling is costly in terms of the animal’s water balance. Evaporative cooling can cause an animal to dehydrate if water is not replaced by drinking or intake of water-rich food. If there is not enough water available, some animals can reduce evaporative cooling, often at the expense of increased body temperature, to prevent dehydration (Robertshaw and Dmi'el, 1983). For endotherms to survive in a hotter, drier environment, animals will need to maintain a delicate balance between thermoregulation and hydration. If animals cannot consume enough water to maintain stable internal temperatures, they may experience an increased amplitude of their daily body temperature rhythms. This pattern in endotherms is known as heterothermy and has been observed in animals such as the Arabian oryx (*Oryx leucorix;* Hetem et al., 2010), which inhabit arid environments.

Some animals do not possess the correct anatomical structures that enable them to use certain autonomic mechanisms of thermoregulation. For example, pigs (*Sus scrofa domesticus*) do not have active sweat glands (Ingram, 1967) and need to use mud bathing to cool down in hot conditions, instead of sweating (Bracke, 2011). Mud bathing is an example of a behavioural adaptation being used instead of an autonomic adaptation. Although the long generation time of large mammals means that they cannot adapt quickly genetically, behavioural adaptations allow them the flexibility to respond immediately to environmental changes. For example it may take many generations for pigs to evolve active sweat glands, but mud bathing is an immediate response to increased environmental temperatures (Bracke, 2011).

1.2.2 Behavioural thermoregulation

Behavioural adaptations are likely to be an animal’s first response to a changing environment since behavioural adaptations are more flexible and cost-effective than are autonomic adaptations (Bartholomew, 1964, Fuller et al., 2010). There are many examples of thermoregulatory behaviour in ectotherms such as butterflies (Kevan and Shorthouse, 1970), periwinkles (Munoz et al., 2005), lizards (Adolph, 1990) and tortoises (Coe, 2004). An Australian study found that ectotherms in various different temperature regions could maintain their body temperature within a favourable range by shuttling between shade and sun (Kearney et al., 2009). Such behavioural adaptations could buffer the impact of climate change on ectotherms, because the animals can increase shade-seeking behaviour in response to increased environmental temperatures to maintain their ideal body temperature.

Endotherms have also been documented to use behavioural thermoregulation (Bartholomew and Dawson, 1979, Gonyou and Stricklin, 1981, Kennedy and Day, 1989, Fortin et al., 2000, Maloney et al., 2005b, Keren and Olson, 2006, Hetem et al., 2011b). Because endotherms use autonomic thermoregulation, they are often considered to be less dependent on behavioural thermoregulatory strategies than are ectotherms. However, autonomic responses can be costly in terms of energy and water and may compromise the balance of other homeostatic systems. Behavioural adaptations could thus alleviate the energetic and water costs of autonomic responses. Strategies that reduce water loss could be particularly important in hot and dry environments, where food and water availability might already be limited. In these conditions, autonomic responses might prove to be too expensive and animals might need to rely on behavioural thermoregulation to buffer the effects of climate change. Therefore, it is important to learn how endotherms use behavioural strategies to thermoregulate in different environments to predict how animals could react to a change in climate.

Animals can use their behaviour to manipulate the amount of radiant heat they absorb from their environment. Radiation plays a significant role in animal thermoregulation (Kelly et al., 1954, Norris and Kunz, 2012). The amount of radiant heat absorbed by an animal is a combination of the quality and quantity of radiation it receives and the reflectance characteristics of the animal surface (Kelly et al., 1954). The reflectance characteristics of an animal surface, such as coat colour, texture and density, are properties that determine how much radiant heat is reflected by the animal. The greatest source of radiation is typically directly from the sun (Norris and Kunz, 2012). Animals can select microclimates, change their body postures or their body orientations to manipulate the amount of radiant heat they absorb from their environments.

In cold environments animals can take advantage of available solar radiation by basking in the sun to raise their body temperatures. For example, ibex can reduce the need for metabolic heat production during cold winter conditions when there is little food available by sun basking in the morning (Signer et al., 2010). Additionally, animals in cold environments can maximise the effect of sun basking, either by exposing a part of their body with darker (or less dense) fur to increase the amount of radiant heat they absorb, or by exposing a greater surface area of their body to the sun to increase the quantity of radiation they receive. For example, racoon dogs (*Nyctereutes procyonoides*) expose their dark chest area to the sun when they are sun basking to increase absorption of radiant heat (Harri and Korhonen, 1988).

In hot environments, endotherms can employ various strategies to avoid solar radiation to reduce the need for evaporative cooling in hot climates. By selecting cooler microclimates, animals can minimise the amount of radiant heat taken up from the environment, reducing the need for evaporative cooling, thereby conserving water and reducing the risk of death by heat stroke. For example, by seeking shade on hot days, cattle (*Bos indicus* and *B. taurus*) can reduce their radiant heat load by 45 % (Blackshaw and Blackshaw, 1994). For bighorn sheep (*Ovis canadensis mexicana*), shaded areas have been shown to be 3-7 ºC cooler than were unshaded areas (Cain et al., 2008). However, shade is not always available, and spending long periods of time seeking shade may come at a cost to other activities, such as grazing (Ben-Shahar and Fairall, 1987, Blackshaw and Blackshaw, 1994).

If animals cannot seek shade, they can reduce the amount of radiant heat they absorb by shifting the posture or orientation of their body relative to solar radiation. For example, incubating Heerman's gulls (*Larus heermann*) on a desert island (Isla Rasa) in the Gulf of California, just off Mexico, can maintain their body temperature and that of their eggs by using different body postures and shifting their body orientation to solar radiation and wind direction (Bartholomew and Dawson, 1979).

Orientation to solar radiation behaviour is especially relevant to species found in open habitats, foraging during the day, such as cattle and wild ungulates. For example, springbok (*Antidorcas marsupialis*) can reduce their need for shivering and panting during cold and hot periods by using thermoregulatory behaviour such as orientation to solar radiation, compared to an animal that was prevented from using behavioural thermoregulation (Hofmeyr and Louw, 1987). If ungulates orientate the long axis of their body parallel to solar radiation they reduce the surface area of their body exposed to solar radiation (Figure 1.1a) compared to when they are orientated perpendicular to solar radiation (Figure 1.1b). Therefore, ungulates can reduce the energetic costs of metabolic

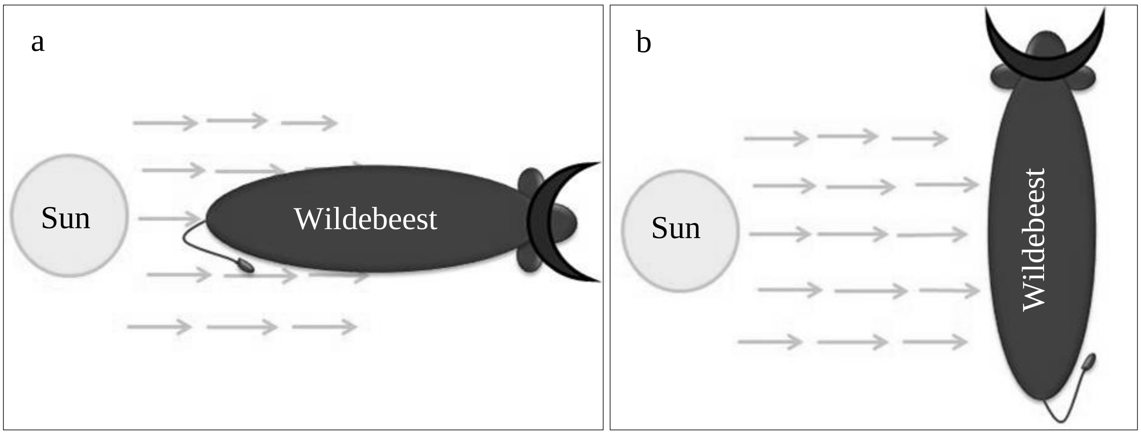


Figure 1.1 Diagram illustrating wildebeest orientation to solar radiation. A wildebeest orientated parallel to solar radiation (a) exposes a smaller surface area to solar radiation than a wildebeest orientated (b) perpendicular to solar radiation and therefore reduces radiant heat load.

heat production in cold environments and reduce evaporative water loss in hot conditions by changing their orientation to solar radiation.

The studies on orientation to solar radiation in mammals were mostly done on ungulates. Cattle prefer to orientate perpendicular to solar radiation on cold, sunny days (Gonyou and Stricklin, 1981, Kennedy and Day, 1989). By orientating perpendicular to solar radiation, cattle can reduce their metabolic requirements in winter (Keren and Olson, 2006). Wildebeest preferred to be orientated perpendicular rather than parallel to solar radiation early in the morning, especially during the cold months, to maximize the amount of heat absorbed from the sun (Berry et al., 1984, Vrahimis and Kok, 1992, Maloney et al., 2005b). Wildebeest are more likely to orientate parallel than perpendicular to solar radiation during the heat of the day (Maloney et al., 2005b), but display random orientation to solar radiation at noon (Berry et al., 1984), probably because changing orientation to solar radiation is not likely to have a great effect on the amount of surface area of the wildebeest that is exposed to when the sun shines from directly above. Similar patterns have been found in eland (*Tragelaphus oryx*), blue wildebeest and impala (*Aepyceros melampus*), which prefer to orientate parallel to solar radiation in the hot summer months and perpendicular to solar radiation in the cold winter months (Hetem et al., 2011b).

Orientation to solar radiation can reduce the costs of autonomic thermoregulation. For example, the total radiation absorbed by springbok orientated parallel was less than that of a springbok orientated perpendicular to solar radiation, especially in the winter and autumn months (Hetem et al., 2009). Black wildebeest (*Connochaetes gnou*) orientated parallel to solar radiation can decrease their radiant heat load by 30 % and reduce their water requirements by 15-30 % compared to wildebeest orientated perpendicular to solar radiation (Maloney et al., 2005b). By shifting their orientation to solar radiation, ungulates can save between 11-19 % of the energy of their basal metabolic rate (Hetem et al., 2011b).

1.3 Available techniques to study thermoregulatory behaviour

Orientation to solar radiation is a very effective method of behavioural thermoregulation. However, all previously published studies on orientation to solar radiation in free-ranging animals that I am aware of have relied on behavioural observations. Human observation of animal behaviour over long periods is arduous and potentially difficult, especially if animals live in remote areas or if they are not habituated to human presence. Additionally, human presence often influences animal behaviour (Taylor and Knight, 2003, De Boer et al., 2004). Therefore, it is difficult to know whether results of observational studies accurately reflect the natural behaviour of an animal. For example, when studying orientation to solar radiation behaviour in animals, the animals will often orientate their bodies so they can look at the human observer, which they could consider as a threat. Therefore it is difficult to determine whether the animal is changing its body orientation to thermoregulate, or to watch the human observer.

Unbiased investigation of behavioural adaptations to thermal environment requires a remote technique that excludes the effect of human presence on behaviour and enables long-term, continuous data capture of animal behaviour. Implanted data loggers have been used to measure physiological variables such as body temperature (Jessen et al., 1994, Fuller et al., 1999), activity (Mitchell et al., 1997) and heart rate (Arnold et al., 2004) remotely. Remote measurements of physiological and climatic information can be used indirectly to study behavioural thermoregulation. For example, Signer et al. (2010)

inferred sun basking behaviour in Alpine ibex by measuring activity and rumen temperature remotely. Remote measurements of physiological data can also be used to quantify the effect of thermoregulatory behaviour. For example in Cape ground squirrels, implanted miniature temperature-sensitive data loggers were used to measure the effect of behaviours such as retreating into borrows and shading of bodies with tails (Fick et al., 2009).

One example of a remote method used to study thermoregulatory behaviour is the use of miniature black globe thermometers attached to animal collars to study microclimate selection behaviour in animals. If the miniature black globe temperature on the collar of the animal is cooler than that of a similar miniature black globe thermometer set up in the sun, the animal is likely seeking shade (Hetem et al., 2007). Miniature black globe temperature has been used to study microclimate selection in wildebeest (Hetem et al., 2007), kudu (*Tragelaphus strepsiceros*; Hetem et al., 2008), angora goats (*Capra aegagrus*; Hetem et al., 2011a) and oryx (Hetem et al., 2012a). Using remote techniques to study thermoregulatory behaviour of animals could enable scientists to collect continuous long-term data, to study animals in remote habitats without the presence of a human observer and to quantify the relative advantages of different thermoregulatory behaviours. A remote method to study orientation to solar radiation behaviour would therefore be invaluable in terms of understanding how animals might react to changes in their environment.

1.4 Study species

Both black and blue wildebeest are known to use orientation to solar radiation for thermoregulatory purposes (Berry et al., 1984, Vrahimis and Kok, 1992, Maloney et al., 2005b), making them ideal species in which to validate a remote technique to study orientation to solar radiation behaviour in ungulates. Both species live in the open woodlands, grasslands and savannahs of southern Africa. Blue wildebeest prefer open grassland and woodland (Skinner and Chimimba, 2005) and spend a great amount of time seeking shade (Ben-Shahar and Fairall, 1987, Hetem et al., 2011b), whereas black wildebeest prefer open habitats and are not known to seek shade (Estes, 1966).

Black and blue wildebeest appear to have different thermoregulatory behaviour in terms of shade-seeking (Skinner and Chimimba (2005), Hetem et al. (2011b) and Ben-Shahar and Fairall (1987) compared to Estes (1966)). Therefore, it would be interesting to compare other aspects of their thermoregulatory behaviour such as the use of orientation to solar radiation for thermoregulatory purposes. Even though orientation to solar radiation has been studied in both species, the orientation to solar radiation preferences of the two species while in the same environment have not previously been compared. It is possible that black wildebeest compensate for the lack of shade-seeking by orientating their body parallel to solar radiation to reduce heat uptake during the summer months. Prior to biologging technology, comparing the two species would require observing both species at the same time. An easier way to compare orientation to solar radiation between black and blue wildebeest might be to use a remote technique to study orientation to solar radiation simultaneously in free-ranging black and blue wildebeest living in the same environment.

1.5 Aims and hypotheses of this study

I set out to develop a technique to study orientation to solar radiation behaviour in black and blue wildebeest remotely. Temperature-sensitive data loggers were implanted subcutaneously and bilaterally into nineteen wildebeest. I aimed to determine whether remote measurements of subcutaneous temperatures of the left and right flank of wildebeest could be used as a remote technique to detect and quantify orientation to solar radiation.

I hypothesised that when a wildebeest was orientated perpendicular to the sun’s rays, the subcutaneous temperatures on the side facing the sun would heat up more than the subcutaneous temperatures on the side facing away from the sun, whereas when the wildebeest was orientated parallel to the sun’s rays, subcutaneous temperatures on both sides would heat up equally. I compared the orientation to solar radiation predicted by the measured subcutaneous temperature at a given time, with visual observations of the wildebeest’s orientation to solar radiation, to validate whether subcutaneous temperatures can be used to remotely detect and quantify orientation to solar radiation.

Chapter 2

Materials and Methods

2 Materials and Methods

2.1 Animal capture

To test the hypothesis that subcutaneous temperatures can indicate orientation to solar radiation in wildebeest, free-ranging black wildebeest (*Connochaetes gnou,* Zimmermann, 1780) and blue wildebeest (*Connochaetes taurinus,* Burchell, 1823) were captured at Mokala National Park, to be implanted with temperature-sensitive data loggers. In March 2011 an experienced game capture team from the Veterinary Wildlife Services (VWS) of South African National Parks (SANParks) darted nine adult black wildebeest (five males and four females) and ten adult blue wildebeest (four males and six females) from a helicopter. The wildebeest were darted using standard SANParks procedures, using either etorphine hydrochloride (M99, Novartis, Kempton Park, South Africa (Pty) Ltd) or thiafentanil oxalate (A3080, Wildlife Pharmaceuticals South Africa (Pty) Ltd, Whiteriver, South Africa), mixed with azaperone (Stresnil, Janssen Pharmaceutical Ltd., Halfway House, South Africa). Hyalase (150 I.U., hyaluronidase, Kyron Laboratories, Benrose, South Africa) was added to all darts by dissolving it in the azaperone, to increase the absorption rate of the drugs. The black wildebeest were captured from a herd in Soutbospan (see section 2.5). The blue wildebeest were captured from different herds in different areas throughout the reserve. We chose areas where black and blue wildebeest did not appear to co-occur to minimise the chance of selecting hybridised individuals.

After they became unconscious, the wildebeest were loaded into transport trucks, where they were given the reversal drug diprenorphine hydrochloride (M5050, Novartis, Kempton Park, South Africa (Pty) Ltd) and transported to the VWS facilities in Kimberley (~80 km drive from Mokala National Park). The wildebeest were separated into five different holding pens based on species, sex, size and age, to prevent fighting between individuals. Two holding pens contained two blue wildebeest males each (the four males were separated, because there was a great size difference between them). The six blue wildebeest females were housed together in one holding pen. Another holding pen held four black wildebeest males. Two young black wildebeest males were housed with the four black wildebeest females.

Food and water was available *ad libitum*. The wildebeest were monitored and cared for by SANParks VWS staff. They were given haloperidol (0.1 mg.kg^{-1}, Kyron Laboratories, Benrose, South Africa), and perphenazine enanthate (0.3 mg.kg^{-1} Trilafon, Kyron Laboratories, Benrose, South Africa) while in the bomas to calm them down, and a multivitamin (20 ml, Phosamine Stimulans, Bayer Animal Health (Pty) Ltd, Isando, South Africa). They were kept in the holding pens for 5 weeks prior to surgery.

2.2 Surgery

For surgery (3-5 May 2011), the wildebeest were darted with etorphine hydrochloride and azaperone. After they lost consciousness, they were carried with a stretcher onto a vehicle and transported to a temporary surgery theatre set up about 50 m from the holding pens (Figure 2.1). Each wildebeest was weighed by placing them on the stretcher on a weighing scale (the weight of the stretcher was later subtracted). The wildebeest were then placed on the surgery table in a sternal position. Sand bags were used to support and maintain the animal’s position throughout surgery. Anaesthesia was maintained with 0.5 - 5 % isofluorane (Isofor, Safeline Pharmaceuticals (Pty) Ltd, South Africa) in oxygen via a facemask. A ruminal drain was inserted to prevent the wildebeest from choking on ruminal fluid.

Figure 2.1 The temporary surgery theatre set up 50 m from the holding pens where the wildebeest were housed.

The data loggers (see section 2.6.1) were implanted subcutaneously on the left and right flanks of the animals, behind the last rib, and ±100 mm below the circumference mid-line (Figure 2.2). The implant site was chosen based on a pilot study (see Appendix A) that showed that loggers placed below the mid-line would be better able to distinguish between parallel and perpendicular orientation to solar radiation than loggers placed on or above the mid-line. If the loggers were implanted above the mid-line, the loggers were more likely to be in the sun if the wildebeest orientated parallel during the middle of the day, but with the loggers implanted below the mid-line, the wildebeest's body shaded the loggers when the wildebeest was orientated parallel to solar radiation, even if the sun was directly overhead.

A patch (approximately 200 mm x 200 mm) was shaved around the implant site. Implant sites were sterilised with chlorhexidine gluconate (Hibitane; Zeneca, Johannesburg, South Africa) and injected subcutaneously with a local anaesthetic (5 ml Lignocain, 2 %, Centaur Labs, South Africa) before incisions were made. This procedure was repeated on both sides of the wildebeest.

Under sterile surgical conditions, an experienced veterinarian implanted the data loggers underneath the panniculus muscle, on both sides of each wildebeest. The loggers were tethered to the surrounding muscle with nylon (NY924, size 0, SRL, Isando, South Africa) to hold them in place. A SANParks vet assisted during the surgery.

During surgery, the wildebeest's vital signs, including blood pressure, heart rate, heamoglobin oxygen saturation, and rectal temperature were monitored with a veterinary monitor (Cardell® Multiparameter Monitor 9403, Sharn Veterinary, Inc., Tampa, FL, USA). Respiratory rate was monitored either visually or by listening to the wildebeest's

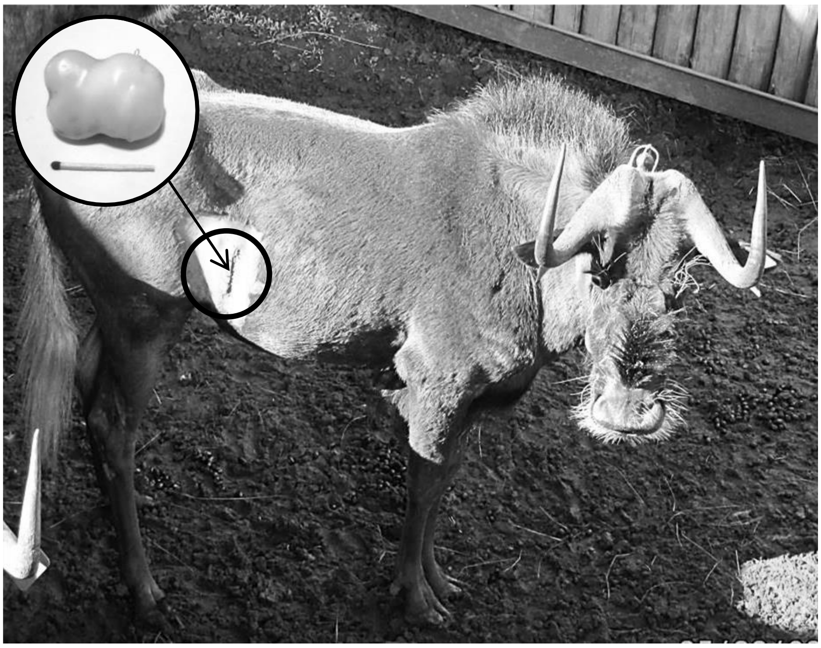


Figure 2.2 The wax-covered data loggers were implanted behind the last rib and just below the mid-line (as indicated by the red arrow) on both sides of the wildebeest.

chest with a stethoscope. If a wildebeest experienced respiratory difficulty, it was given a partial reversal drug (2.5 times the dose of etorphine hydrochloride received, butorphanol tartrate, intramuscularly, Pfizer Animal Health, Sandton, South Africa). Most wildebeest (18 out of 19) received butorphanol during surgery, and five of the blue wildebeest received two doses of butorphanol. Three blue wildebeest received 2-2.5 ml doxapram hydrochloride (Dopram, intravenously, Boehringer Ingelheim, Randburg, South Africa), in addition to the butorphanol, to stimulate breathing.

Each wildebeest received a long-acting parasiticide (0.02 $ml.kg^{-1}$, subcutaneously, Dectomax, Doramectin, 1 %, Pfizer Animal Health, Sandton, South Africa) to protect it from parasites, and a vitamin supplement (0.03 $ml.kg^{-1}$, subcutaneously, Multimin + Se, Virbac RSA Pty (Ltd), Halfway House, South Africa). The wildebeest received antibiotics (0.04 $ml.kg^{-1}$, intramuscularly, duplocilin, Intervet, Johannesburg, South Africa and 2 $mg.kg^{-1}$ intramuscularly, and Baytril, Enrofloxacin, 10 $mg.ml^{-1}$, Bayer Animal Health Division, Isando, South Africa) to prevent post-surgical infection. An analgesic (0.5 $mg.kg^{-1}$, intramuscularly, Mobic, Meloxicam, Boehringer Ingelheim, Johannesburg, South Africa) was administered to reduce pain and inflammation. Each wildebeest was fitted with an external radio collar (African Wildlife Tracking, Pretoria, South Africa) as well as a uniquely coloured ear tag for subsequent tracking and identification of individual wildebeest in the field.

Surgery lasted an average of 60 minutes per animal. After the loggers were implanted, the surgical site was sutured with dissolvable sutures (Visyn, VS108, size 2, and Viamac VM514, size 2/0, SRL, Isando, South Africa). The surgery wounds were sprayed with an antiseptic spray (Necrospray, Centaur Labs, South Africa), and smeared with tick grease (cypermethrin 0.025 %, Bayer Animal Health Pty, Isando, South Africa) to prevent infection and fly infestation. After surgery the wildebeest were returned to the holding pens and injected intravenously with the reversal drug diprenorphine hydrochloride. The wildebeest were observed until they could stand up (black wildebeest average: 3.4 minutes ±1.0 minutes, blue wildebeest average: 1.7 minutes ±1.2 minutes) to ensure they recovered successfully from anaesthesia.

The wildebeest remained in the holding pens to recover from surgery. They were monitored by myself, H. Lease and SANParks staff to ensure that no complications arose. Five weeks later (13 June 2011), the wildebeest were herded from the holding pens into the transport trucks without tranquilisers, transported back to Mokala National Park, and released.

All experimental procedures were approved by SANParks and the Wits Animal Ethics Screening Committee (clearance no. 2011/09/04). The necessary permits were obtained to transport and do surgery on the wildebeest (TOPS animal transport permit, Fauna 194/2011, and TOPS research permit, Fauna 248/2011, Northern Cape Department of Environment and Nature Conservation).

2.3 Release of wildebeest back into Mokala National Park

The blue wildebeest were released in June 2011 (winter), in the southern part of the reserve. The six blue wildebeest females formed their own herd within the first month and were joined by an untagged dominant male. These six females remained together in the same area for the majority of the study. They split up between December 2011 and February 2012, when two females moved further north. One of the tagged blue wildebeest males was solitary (mostly observed alone), while the others were usually observed within a herd of other wildebeest. The black wildebeest were released south of Soutbospan. All but one of them were observed back at Soutbospan within a month of release. The four tagged females together with one of the young tagged males formed their own herd and were joined by an untagged dominant male. The untagged dominant male was later replaced in the herd by one of the tagged males. The other black wildebeest males were seen either on their own or with one or two untagged males. The final black wildebeest male remained isolated from the rest of the black wildebeest, and was found in the southern woodland area of Mokala National Park until October 2011. There were no other black wildebeest in this area, although many other antelope, including blue wildebeest were found in this area. This male eventually relocated to a region of the reserve inhabited by other black wildebeest; in February this male was found in Soutbospan.

2.4 Study animals

Nine black wildebeest (Figure 2.3a) and ten blue wildebeest (Figure 2.3b) were used for this study. Both black and blue wildebeest are known to use orientation to solar radiation (Berry et al., 1984, Vrahimis and Kok, 1992, Maloney et al., 2005b). Furthermore, a parallel study was comparing the behaviour and physiology of black and blue wildebeest using the same study animals. Therefore, black and blue wildebeest were the ideal species in which to validate the remote technique to study orientation to solar radiation. At the time of implant surgery the black wildebeest had a body mass of 128 ± 22 kg (116 ± 2 kg for four females and 138 ± 26 kg for five males). The blue wildebeest had an average body mass of 186 ± 40 kg (158 ± 16 kg for six females and 228 ± 23 kg for four males). There was an average gain in mass for black and blue wildebeest of close to 50 kg over the course of the one-year study (Table 2.1).

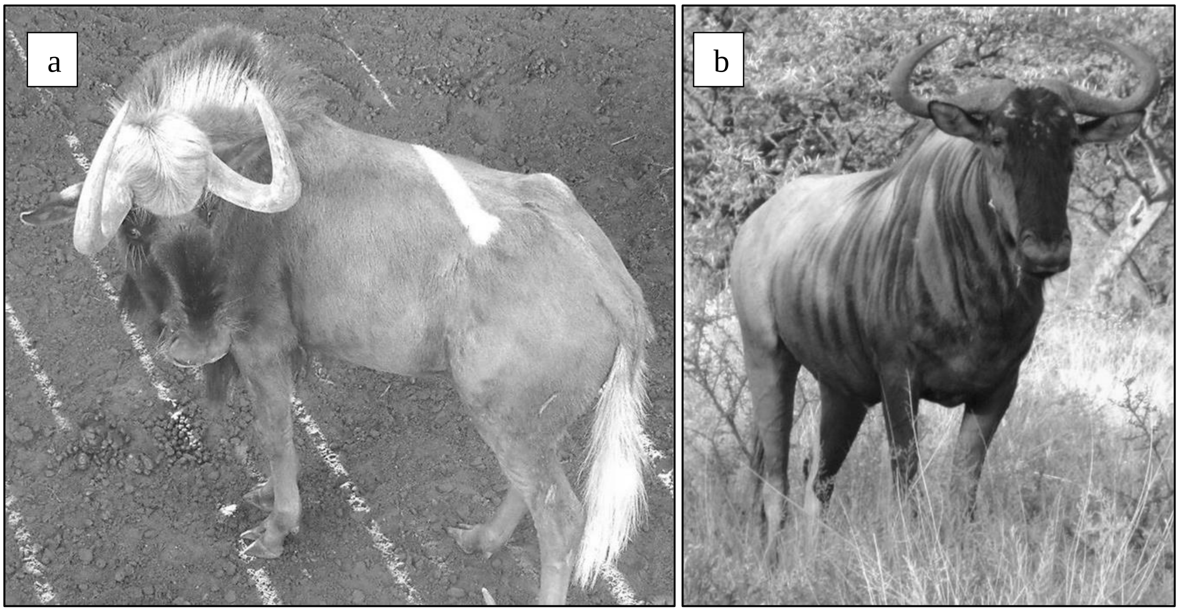


Figure 2.3 Photo of black (a) and blue wildebeest (b) taken by A. Botha (a) and P.J. van Staden (b) (permission obtained).

Table 2.1 Characteristics of study animals.

Individual	Species	Sex	Body mass at the time of implant surgery (kg)	Body mass at the time of explant surgery (kg) *
1	Black	Female	119	150
2	Black	Female	115	140
3	Black	Female	115	130
4	Black	Female	117	170
5	Black	Male	144	180
6	Black	Male	170	200
7	Black	Male	153	190
8	Black	Male	110	160
9	Black	Male	113	180
10	Blue	Female	164	220
11	Blue	Female	177	230
12	Blue	Female	159	220
13	Blue	Female	145	190
14	Blue	Female	170	240
15	Blue	Female	136	Not measured
16	Blue	Male	223	Not measured
17	Blue	Male	217	270
18	Blue	Male	210	310 **
19	Blue	Male	262	Not measured

*A different method was used to weigh animals during the explant surgery therefore the increase in temperature is not necessarily accurate.

**Probably a measurement error as the body weight falls outside the normal range for blue wildebeest (Skinner and Chimimba, 2005).

Because black and blue wildebeest can hybridise, genetic tests were done to determine whether our study animals were pure black and blue wildebeest. The genetic tests, conducted on blood samples collected during wildebeest capture, confirmed that all of the captured wildebeest were genetically pure (Desiré Lee Dalton, Appendix B). The necessary permits were obtained to transport the tissue samples (TOPS tissue transport permit, Fauna 298/2011, Northern Cape Department of Environment and Nature Conservation).

2.5 Study site

Mokala National Park (Figure 2.4) occupies 19 611 ha, and is situated 1200 m above sea level, ~80 km southwest of Kimberley, Northern Cape, South Africa (main gate coordinates: S 29° 12’ 46.5”, E 24° 19’ 34.1”). Mokala National Park was founded when Vaalbos National Park received land claims from the Sidney on Vaal claimants in 2002 (SANParks, 2012). SANParks purchased land from the Wintershoek, Plooyburg area to replace Vaalbos National Park; Mokala National Park opened officially in 2007 (SANParks, 2012).

Mokala National Park has a semi-arid climate with an average annual rainfall of 400 mm p.a. (maximum 700 mm p.a., minimum 300 mm p.a.). Rainfall primarily occurs in the summer months (November – February). However, artificial waterholes and dams supply water to animals throughout the year. Air temperatures range from -4 °C in winter to 44 °C in summer. Frost can occur between late April and end September (SANParks, 2012).

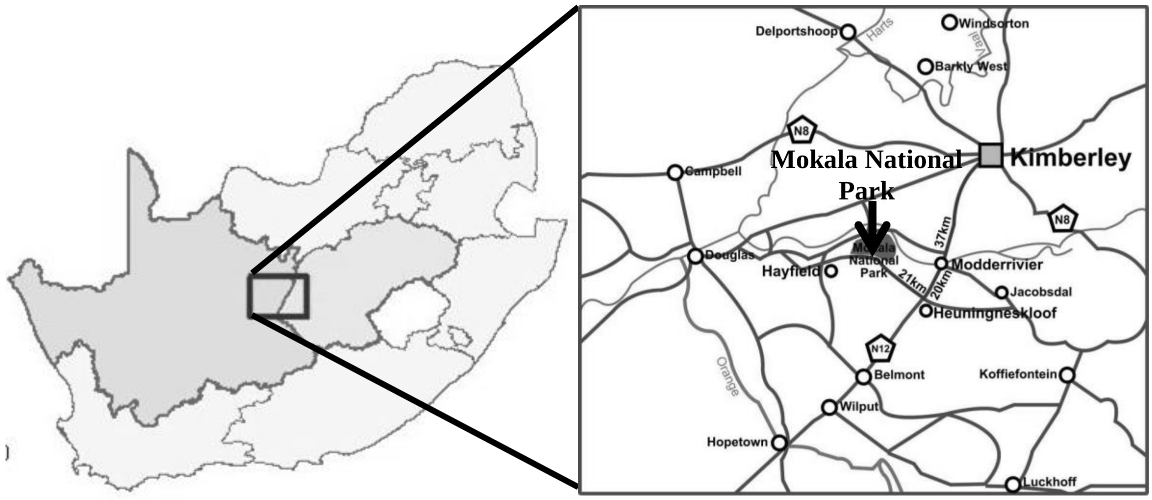


Figure 2.4 Regional map indicating the location of Mokala National Park within South Africa (SANParks, 2012).

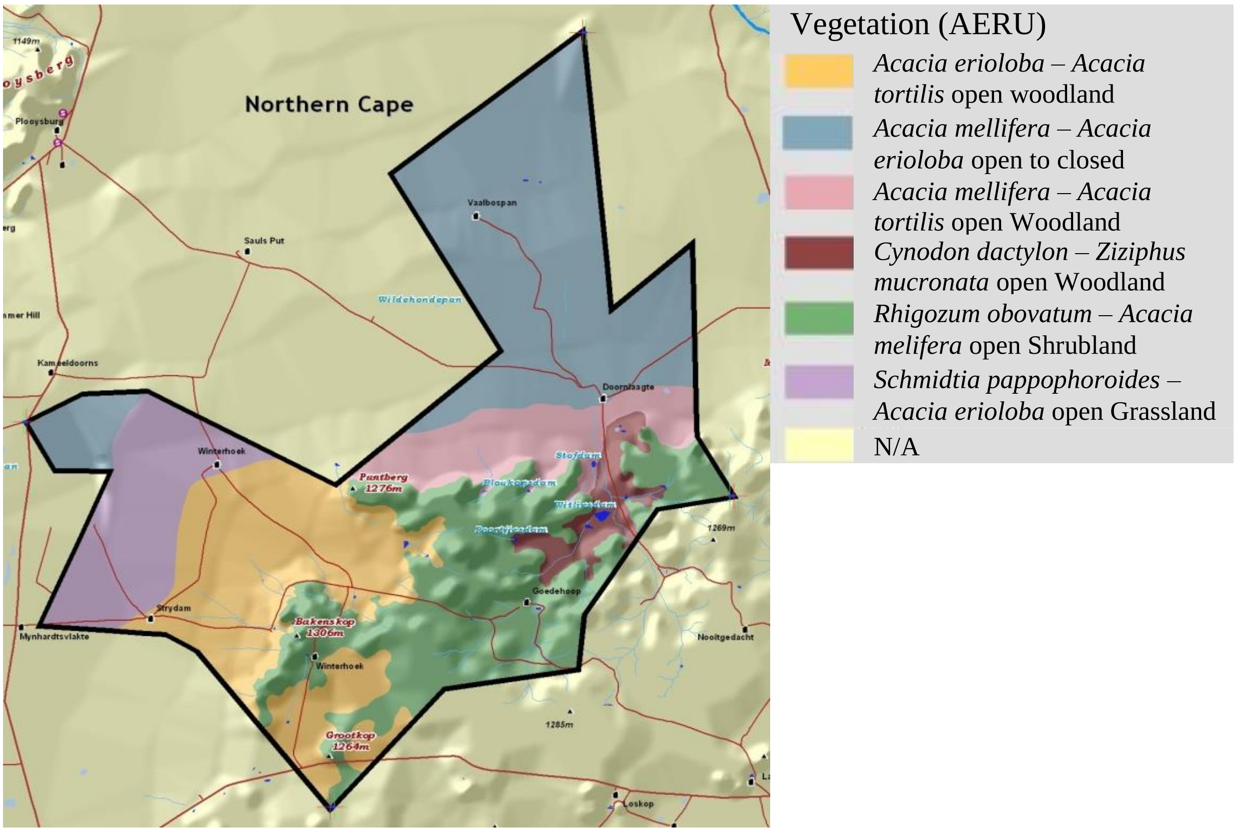


Figure 2.5 Mokala National Park Vegetation Map, 2006 (courtesy of Mokala National Park 2006).

According to Acocks' classification (Acocks, 1988), Mokala National Park falls within the Kalahari Thornveld Proper Veld Type (Veld Type 16). The camel thorn tree (*Acacia erioloba*) and grasses such as *Schmidtia pappophoroides* are characteristic of this veld type. The name "Mokala" is Setswana for camel thorn (SANParks, 2012). The reserve's variety of vegetation types (Figure 2.5) provides habitats for both black and blue wildebeest.

Mokala National Park was chosen as a field site because both black wildebeest and blue wildebeest occur in the reserve. According to the Mokala National Park census in 2011, there were 783 blue wildebeest and 278 black wildebeest. Historically, only the black wildebeest naturally occurred here (Skinner and Chimimba, 2005). The black wildebeest are found in two areas in the park: the open grassland area in the western section of the reserve known as Soutbospan, and the open woodland areas in the north known as Vaalbospan (Figure 2.6, Mokala National Park Census Map 2011). The grassland areas (Figure 2.7a) with their wide open spaces are ideal habitat for black wildebeest (Skinner and Chimimba, 2005), which seldom seek shade (Estes, 1966). The blue wildebeest are more widely distributed throughout the reserve than are the black wildebeest, and prefer the woodland areas (Figure 2.7b) that provide more shade (Berry et al., 1984, Skinner and Chimimba, 2005) than the grassland areas. The black and blue wildebeest distribution overlaps (Figure 2.6) and the two species are sometimes seen grazing together (personal observation and communication with SANParks staff), which led to the concern that the two species might interbreed.

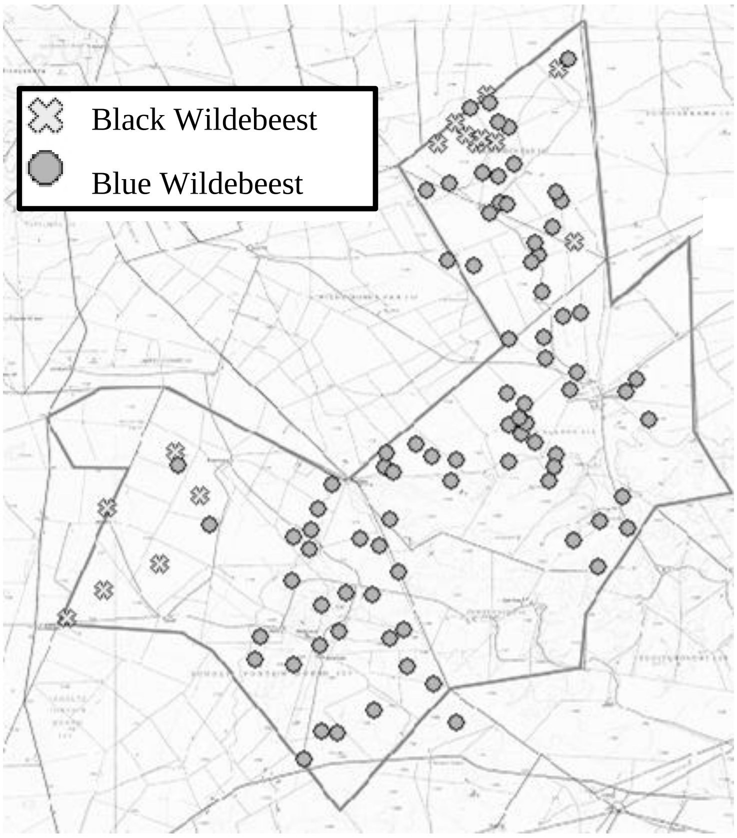


Figure 2.6 Mokala National Park Census map for 2011 showing the distribution of black wildebeest and blue wildebeest (from Mokala National Park Census 2011).

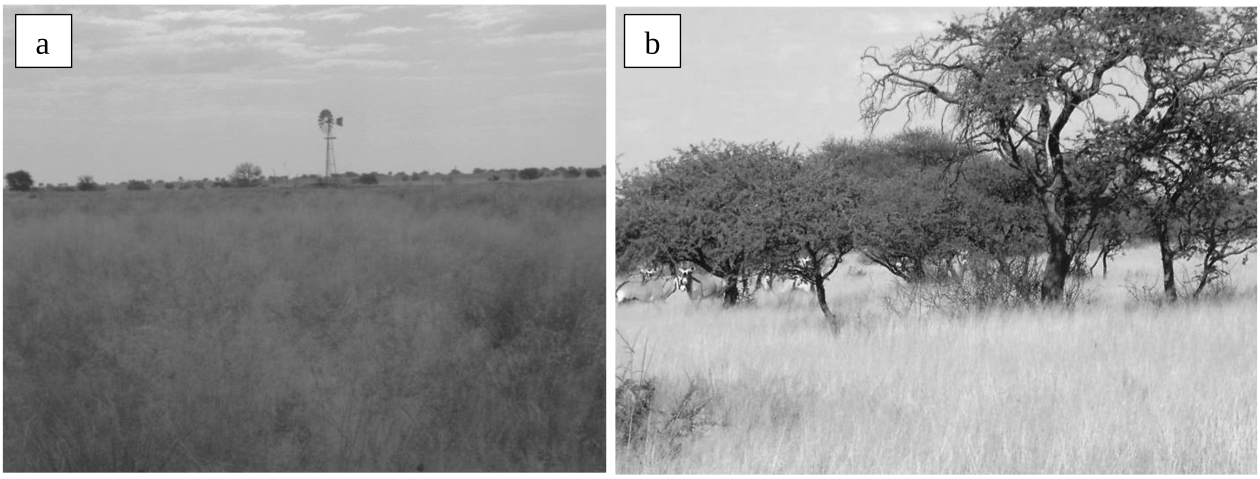


Figure 2.7 Two photographs taken by A. Botha in winter (June 2011) to illustrate the difference between the grassland (a) and woodland habitats (b) in Mokala National Park.

Aside from the black and blue wildebeest, there are many other large grazers that occur in the reserve, such as tsessebe (*Damaliscus lunatus*, Burchell, 1823), zebra (*Equus quagga*, Boddaert, 1785), red hartebeest (*Alcelaphus buselaphus*, Pallas, 1766), blesbok (*Damaliscus pygargus phillipsi*, Pallas, 1767), springbok (*Antidorcas marsupialis*, Zimmerman, 1780), eland (*Tragelaphus oryx*, Pallas, 1766), gemsbok (*Oryx gaxella*, Linaeus, 1758), white rhinoceros (*Ceratotherium simum*, Burchell, 1817), and buffalo (*Syncerus caffer*, Sparrman, 1779). There were no large predators present in the reserve that could pose a threat to our study animals.

2.6 Data collection

2.6.1 Temperature data loggers specifications

Each wildebeest received a subcutaneous implant on each lateral side of the abdominal cavity. Each implant consisted of three data loggers, one long-term Micrologger to record data for the full time period of the study (mlog T1C, Sigma Delta Technologies, Floreat, Australia) and two backup loggers to record data for small time periods within the study period (DS1922L iButtons, Maxim Integrated Products, Sunnyvale, USA) (Figure 2.8). The long-term loggers were set to record temperature data every 5 minutes for one year. They can record 500 000 data points at a resolution of 0.06 °C, and they use rechargeable lithium batteries with an average lifetime of 12 months. The backup loggers had more limited memory capacity (4 096 data points at 0.06 °C resolution) than the long-term loggers and were therefore set to record temperature every 5 minutes for two weeks during predetermined periods in the cold (July 2011) and hot (February 2012) seasons. Each logger package was covered with biologically and chemically inert wax (SASOL wax 1276, SASOL, South Africa) to make it waterproof (Figure 2.8b). Each wax-covered implant had a mass of ~43 g.

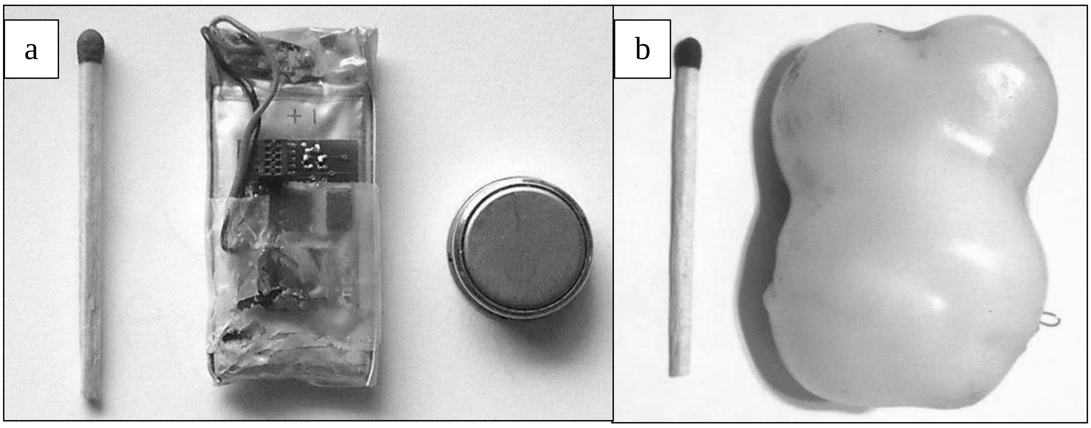


Figure 2.8 (a) Two different temperature data loggers were implanted into the wildebeest: Sigma Delta Micrologger (left) and Maxim iButton temperature loggers (right). (b) One Sigma Delta logger and two iButtons were waxed together into one package. A match is added to indicate relative size.

All data loggers were calibrated in a water bath against a certified quartz crystal thermometer (Quat 100, Heraeus, Hanau, Germany) at 5 °C increments between 25 °C and 45 °C. The loggers were left in a water bath for 1 hour at each temperature increment to ensure the logger temperature had stabilised at the raised water temperature. A calibration curve (Figure 2.9) was composed in Table Curve 2D, Version 3 (Systat Software Inc. San Jose, California). The temperatures recorded by the loggers were plotted against those recorded by the quartz thermometer and a straight line was fitted to the data. The equation of the straight line was used to calibrate the loggers. The fitted standard error of the straight line was always less than the resolution of the loggers (0.06 °C). After calibration, the accuracy of the loggers was ±0.1 °C. After the loggers were retrieved from the animals at the end of the study, they were recalibrated as described above. Calibration curves at the beginning and the end of the study were compared to each other, to detect if temperature and/or temporal drift had occurred for any loggers. Two Sigma Delta loggers drifted by 0.4 °C and 0.3 °C over one year. Their values were corrected by adding the amount of drift in temperature divided by the number of data points over which the drift occurred, multiplied by the number of the data points that had been measured.

2.6.2 Meteorological measurements

A weather station (HOBO Weather Station H21-001, Onset Computer Corporation, Massachusetts USA) was set up in a fenced-off area in the reserve, to protect it from damage by animals (Figure 2.10), and in an open area where it would not be affected by shade. The weather station measured dry-bulb temperature, black globe temperature, solar radiation, wind speed and direction, and relative humidity. Black globe temperature was measured by a thermometer inside a hollow copper ball (150 mm diameter) painted matt black. The black globe thermometer was originally designed to measure mean radiant

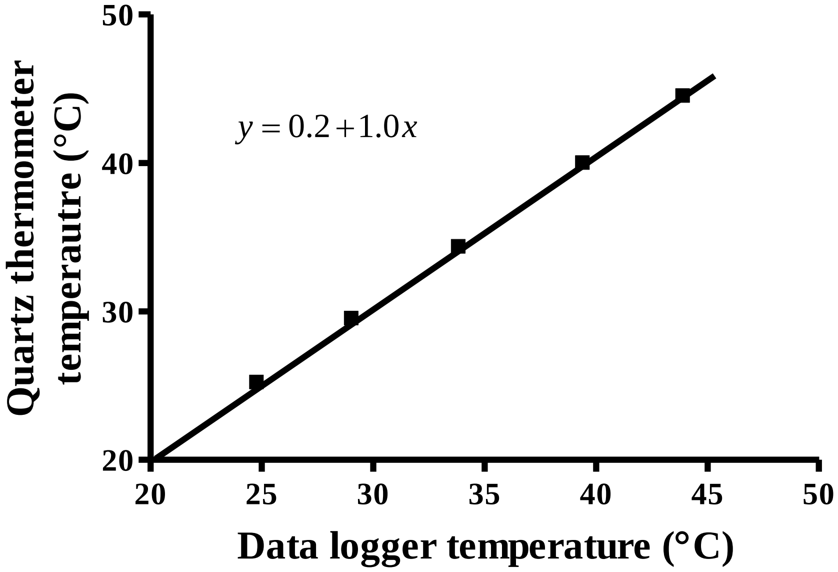


Figure 2.9 Example of a calibration curve of a Sigma Delta Micrologger calibrated against a quartz thermometer in a water bath at ~5 °C increments from 25 °C to 45 °C. Fitted standard error =0.007 °C.

Figure 2.10 The weather station set up in an open area in Mokala National Park to measure dry bulb and black globe temperature, solar radiation, wind speed and direction, and relative humidity.

temperature (Vernon, 1930). Black globe temperature incorporates the effects of radiation, wind speed and air temperature, therefore giving a better indication of the heat load experienced by an animal than would dry-bulb temperature. Black globe temperature is often used to assess environmental heat load in studies of animal thermoregulation (Mitchell et al., 1997, Fuller et al., 2000).

Miniature black globe (30 mm diameter) thermometers can be attached to an animal's collar to measure the heat load experienced by animals in their respective microclimates (Fuller et al., 1999, Hetem et al., 2007, Hetem et al., 2012a). Miniature black globe thermometers were attached to the wildebeest's collars to measure the environmental heat load experienced by the wildebeest in their immediate microclimates every ten minutes.

2.6.3 Behavioural observations

Behavioural observations were initiated in July 2011 (winter), three weeks after release. Behavioural observations of the wildebeest were conducted by A. Botha and H. Lease (occasionally with the help of one or two assistants; see acknowledgements) for one week every month from July 2011 until June 2012. At the same time, the data loggers were recording subcutaneous temperature, the weather station was recording climatic variables and the miniature black globe thermometers on the animals' collars were measuring the environmental heat load the animals were experiencing. The implanted wildebeest were tracked in the field by observers in a car or on foot using a handheld VHF antenna (RA-14K VHF Antenna, Telonics, Inc., Mesa, AZ, USA). When located, the wildebeest were observed continuously using continuous focal animal sampling methods (Altmann, 1974) until we lost sight of them. Changes in activity or orientation relative to the sun were noted as well as the time of the change. The following information was recorded: body position (standing vs. lying down), orientation relative to solar radiation and wind, cloud cover over the wildebeest and shade-seeking behaviour (see Appendix C for behavioural data sheet).

Observations on foot were attempted from a hill-top or under cover of vegetation, whenever possible, to prevent disturbing the wildebeest. Most observations were done with the help of binoculars (Bushnell Powerview 8x25 Porro Prism Binoculars 139825, Bushnell, Lenexa, KS, USA). The black wildebeest in Soutbospan were less accustomed to cars than were the blue wildebeest, because there were no tourist roads going through this area. It was difficult to get close to the black wildebeest by car, with only a few management roads available. Therefore, most observations on black wildebeest were done on foot. The open nature of the grassland (Figure 2.7a) provided only a few trees and termite mounds to conceal our presence from the black wildebeest. The black wildebeest were skittish of humans on foot, making it difficult to get close enough to the black wildebeest to be able to determine their orientation to solar radiation and to identify them by the colour and shape of their ear tags. The blue wildebeest were more often observed from the car compared to the black wildebeest. Even though the blue wildebeest were habituated to tourist cars, they were easily surprised by humans on foot. The dense vegetation of the woodland habitat (Figure 2.7b) meant that the blue wildebeest were harder to see, but at the same time the vegetation provided better cover for us than did the grassland areas. The presence of dangerous animals, such as buffalo and rhinoceros, meant that we could not always follow wildebeest on foot.

The orientation to solar radiation of the wildebeest was determined using a specially designed gnomon: a flat, round disk with a straight stick protruding at a 90 ° angle. The round disk was equally divided into 8 sections named *parallel head*, *parallel tail*, *perpendicular left*, *perpendicular right*, *oblique left* and *oblique right* (Figure 2.11). The disk had to be positioned perfectly horizontal, with the stick perfectly vertical, and the shadow of the stick had to be aligned with the dotted line. A small block of wood, which could rotate freely on the verticle stick of the gnommon, was rotated to mimic the angle of the observed wildebeest's body. The arrow on the block was pointed in the same direction as that the wildebeest was facing. The section on the disc covered by the arrow was recorded as the orientation of the wildebeest relative to the sun. When a wildebeest is orientated perpendicular (as in Figure 2.11) to the sun, the block of wood will be at a 90 ° angle to the shadow cast by the gnomon. When a wildebeest is orientated parallel to the sun, the block of wood will be parallel to the shadow cast by the gnomon.

Over the course of the study, I collected a total of ~470 hours of observations (270 for blue wildebeest and 200 for black wildebeest, 160 hours in winter, 50 hours in spring, 200 hours in summer and 60 hours in autumn). Observation periods typically lasted between 30 minutes to 3 hours, but could last up to 7 hours. On average, wildebeest did not spend more than 20 minutes in the same position, unless they were lying down.

2.7 Retrieval of loggers

After one year, the wildebeest were recaptured to remove the data loggers and radio-tracking collars. All but one of the wildebeest were recaptured between 18 and 26 June 2012. The final wildebeest (a blue wildebeest female) was culled on 8 October 2012. In the case of the nine black wildebeest, the devices were surgically removed. For management reasons, SANParks requested that the blue wildebeest be culled at the end of our study.

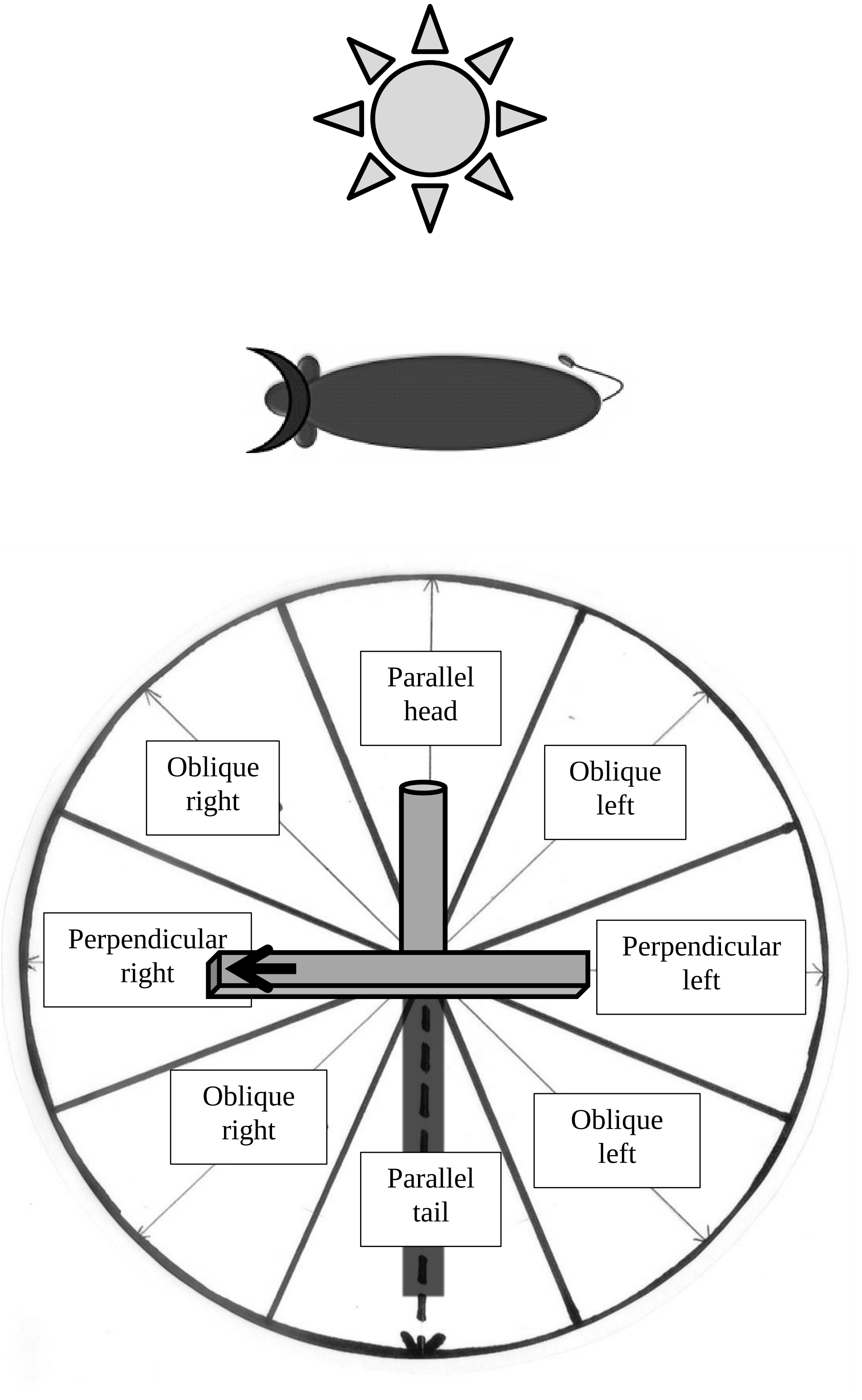


Figure 2.11 A diagram of the gnomon used to determine the wildebeest's orientation relative to solar radiation. The shadow of the stick protruding at 90 ° had to be aligned with the dashed line on the disk. The round disk was divided into 8 sections: parallel head, oblique left, perpendicular left, parallel tail, oblique right and perpendicular right. The wooden block was rotated to mimic the angle of the wildebeest's body. The arrow indicates the orientation of the wildebeest (in this case, perpendicular right).

The black wildebeest were darted from a helicopter using thiafentanil oxalate, azaperone and ketamine (Pfizer Animal Health, Sandton, South Africa) and carried with a sling attached to the helicopter to a temporary surgery theatre set up in a fenced-off area in the reserve. The load monitoring scale of the helicopter recorded the body mass of the wildebeest while they were transported. The data loggers were removed from these wildebeest using surgical procedures similar to those described earlier (section 2.2). We also removed the collars and the ear tags. After surgery, the wildebeest were given a reversal drug (naltrexone, Kyron Laboratories, Benrose, South Africa). The wildebeest stood up after an average of 3 ± 2 minutes after receiving the reversal drug.

Seven blue wildebeest were culled using standard SANParks procedures, and their data loggers were removed by myself and H. Lease. Two blue wildebeest females were not culled, because they had young calves. These females were darted and their loggers were surgically removed using procedure similar to those described for the black wildebeest.

I retrieved all of the data loggers successfully. None of the loggers had moved position, even though in ~half of my logger packages, one or both of the tethers had broken. In six of the nineteen wildebeest, the loggers were encapsulated by a thin layer of cells, on one or both sides of the animal. I observed the blue wildebeest females grazing with their calves the day after surgery.

2.8 Statistics and data analysis

2.8.1 Data sorting

The temperature data were downloaded from the retrieved data loggers and analysed. I applied the relevant calibrations (see section 2.9) to the raw downloaded data. Out of 38

Sigma Delta loggers, 26 stopped working before the end of the study period. For my study, I required data from both the left and right data loggers simultaneously. Based on these criteria, only three wildebeest had a full year of data where both data loggers were working. Five wildebeest had more than six months of data, four wildebeest had less than six months of data, six wildebeest only had data from the backup loggers for two weeks in July 2011 and/or two weeks in February 2012, and one wildebeest had no useful data.

For my analyses of orientation to solar radiation, I excluded observations when wildebeest were seeking shade or under cloud cover. Fortunately, only 17 out of 75 total days were cloudy and there were only 5 days when it rained (rain was usually in short bursts and in most cases observations could be done during the rest of the day). In the summer months, the blue wildebeest often spent several hours during the middle of the day lying down in the shade. The blue wildebeest's shade-seeking behaviour made it difficult to find the tagged blue wildebeest individuals and meant that we could not do observations on their orientation to solar radiation behaviour, since they were avoiding sun exposure.

I focused my analyses on times when I observed the wildebeest to be standing or lying perpendicular or parallel to the sun for at least ten continuous minutes. To prevent pseudo-replication, I used one value per period of observation (i.e. the time that a wildebeest remained visible and in one orientation). The threshold of ten minutes was selected because we did not expect a biologically significant change in subcutaneous temperature if the orientation relative to solar radiation was not maintained for at least ten minutes and because orientation behaviour maintained for less than ten minutes may be unrelated to thermoregulation. Using a longer time threshold (eg. twenty minutes after the beginning of the observation) greatly reduced the sample size, because most wildebeest did not remain in the same position for longer than ten minutes. My final sample size was 74 observations on 16 individual wildebeest.

This subset of calibrated temperature data was collated in Microsoft Excel (Microsoft Office Home and Student 2010, Microsoft Corporation, Redmond, WA, USA). I entered the wildebeest’s body orientation relative to solar radiation and the left and right subcutaneous temperature data into a spreadsheet. Other details included in the table are date, time, the wildebeest individual ID number, the species and sex of the wildebeest, whether the wildebeest was standing or lying down, the miniature black globe temperature from the wildebeest collar and wind speed recorded by the weather station. Basic statistics were done using GraphPad Prism (version 4, for Windows, GraphPad Software, San Diego, CA, USA). A p-value equal to or less than 0.05 was considered significant.

2.8.2 Comparison of subcutaneous temperature when wildebeest were orientated parallel and perpendicular to solar radiation

I plotted the left and right subcutaneous temperatures over time for each wildebeest. I compared these temperature profiles to the body orientation of the wildebeest as recorded during behavioural observations. I compared the temperature profiles of wildebeest observed to be orientated perpendicular to solar radiation to the temperature profiles of wildebeest observed to be orientated parallel to solar radiation. I made note of any factor that may have affected the temperature profile, such as the time a wildebeest changed position, how long it stayed in one position, the previous position the wildebeest was in, and whether the animal was lying down or standing.

To further investigate how the difference between left and right subcutaneous temperatures was affected by the orientation to solar radiation behaviour of wildebeest, I calculated the difference between left and right subcutaneous temperatures and compared that to the wildebeest's orientation to solar radiation. I calculated the difference between left and right subcutaneous temperatures ("subcutaneous temperature difference" from here on) as follows:

$$\text{Difference (°C)} = \text{Left (°C)} - \text{Right (°C)}$$

Where "Difference" is the subcutaneous temperature difference, "Left" is the left subcutaneous temperature, and "Right" is the right subcutaneous temperature. Because right subcutaneous temperatures were subtracted from left subcutaneous temperatures, a positive subcutaneous temperature difference meant that the left subcutaneous temperature was hotter than the right subcutaneous temperature, and a negative value meant that the right subcutaneous temperature was hotter than the left subcutaneous temperature.

I investigated whether there was a difference between the subcutaneous temperature difference of wildebeest orientated perpendicular and wildebeest orientated parallel to solar radiation. Because I was investigating perpendicular and parallel orientation to solar radiation and not the direction of the orientation, it did not matter which side (left or right) was warmer. Therefore, I used the absolute values of subcutaneous temperature difference. I averaged the absolute subcutaneous temperature difference for each individual orientated parallel or perpendicular to solar radiation. I compared the subcutaneous temperature difference of wildebeest orientated perpendicular and parallel to solar radiation using a paired t-test.

To determine whether subcutaneous temperature difference could distinguish the direction of perpendicular orientation of wildebeest to solar radiation (whether the left side or the right side was perpendicular to solar radiation) I used the actual values of the subcutaneous temperature difference. I used a paired t-test to determine whether the subcutaneous temperature difference of wildebeest orientated left perpendicular to solar radiation was significantly different to zero. Similarly, I tested whether subcutaneous temperature difference of wildebeest orientated right perpendicular to solar radiation was significantly different to zero

2.8.3 Subcutaneous temperature difference to predict orientation to solar radiation in wildebeest

To successfully use subcutaneous temperature difference to predict orientation to solar radiation, I had to determine the threshold subcutaneous temperature difference above which it was most likely that a wildebeest was orientated perpendicular to solar radiation. I carried out a Receiver Operating Characteristics analysis using STATA SE10 for Windows (StataCorp LP, College Station, TX, USA) to calculate the sensitivity and specificity of possible threshold values. I chose the threshold value with the highest combined sensitivity and specificity (see Chapter 3, Results, section 3.3).

I predicted the orientation of a wildebeest to solar radiation based on the subcutaneous temperature difference. If subcutaneous temperature difference was smaller than the negative threshold value, the wildebeest was predicted to be orientated right side perpendicular to solar radiation. If the subcutaneous temperature difference was larger than the positive threshold value, the wildebeest was predicted to be orientated left side perpendicular to solar radiation. If the subcutaneous temperature difference fell in-between the threshold values, the wildebeest was predicted to be orientated parallel to solar radiation.

To validate if subcutaneous temperature difference could be used to accurately predict orientation to solar radiation, I compared the orientation to solar radiation predicted by the subcutaneous temperature difference to the orientation to solar radiation we observed in the wildebeest. I calculated the percentage of times the prediction was correct for each individual wildebeest. I further calculated the mean of the individual means of the percentage of correct predictions as an indication of the accuracy of the technique.

2.8.4 Rate of change in difference between left and right subcutaneous temperatures to predict orientation to solar radiation in wildebeest

To improve the accuracy of the remote technique using subcutaneous temperatures to predict orientation to solar radiation of wildebeest, I considered using a measurement other than subcutaneous temperature difference to predict the orientation to solar radiation of wildebeest. I investigated how subcutaneous temperature difference changed over time and how temperature change was influenced by the wildebeest's orientation to solar radiation.

I calculated rate of change of difference between left and right subcutaneous temperature (rate of change (Δ) in subcutaneous temperature difference) as follows:

$$\Delta\ \text{Difference}\ (^\circ\text{C.min}^{-1}) = \frac{(\text{Left}\ ^\circ\text{C} - \text{Right}\ ^\circ\text{C})_{\text{at 10 minutes}} - (\text{Left}\ ^\circ\text{C} - \text{Right}\ ^\circ\text{C})_{\text{at 5 minutes}}}{5\ \text{minutes}}$$

Where "Δ Difference" is the rate of change in subcutaneous temperature difference, "Left" is the left subcutaneous temperature, and "Right" is the right subcutaneous temperature. Therefore, if the left subcutaneous temperature was increasing relative to the right subcutaneous temperature, there would be a greater difference between left and right subcutaneous temperatures at the 10 minute mark than there was at the 5 minute mark, and the rate of change in subcutaneous temperature difference would have a positive value, as shown in the example below (Figure 2.12), where rate of change in subcutaneous temperature difference was 0.14 °C.min^{-1}. Similarly, if the right subcutaneous temperature was increasing relative to the left subcutaneous temperature, rate of change in subcutaneous temperature would have a negative value.

I investigated whether rate of change in subcutaneous temperature difference could reflect the orientation to solar radiation of wildebeest, by comparing the rate of change in subcutaneous temperature difference of wildebeest orientated perpendicular and parallel to solar radiation using a paired t-test. To determine whether rate of change in subcutaneous temperature difference could distinguish the direction of perpendicular orientation to solar radiation, I used a paired t-test to determine whether the rate of change in subcutaneous temperature difference for wildebeest orientated left and right perpendicular to solar radiation was significantly different to zero.

To predict the orientation to solar radiation of wildebeest using rate of change in subcutaneous temperature difference, I had to determine the threshold rate of change in subcutaneous temperature difference above which a wildebeest was most likely to be orientated perpendicular to solar radiation. I carried out a Receiver Operating Characteristics analysis using STATA SE10 to calculate the sensitivity and specificity of possible threshold values. I chose the threshold value with the highest combined sensitivity and specificity (see Chapter 3, Results, section 3.4).

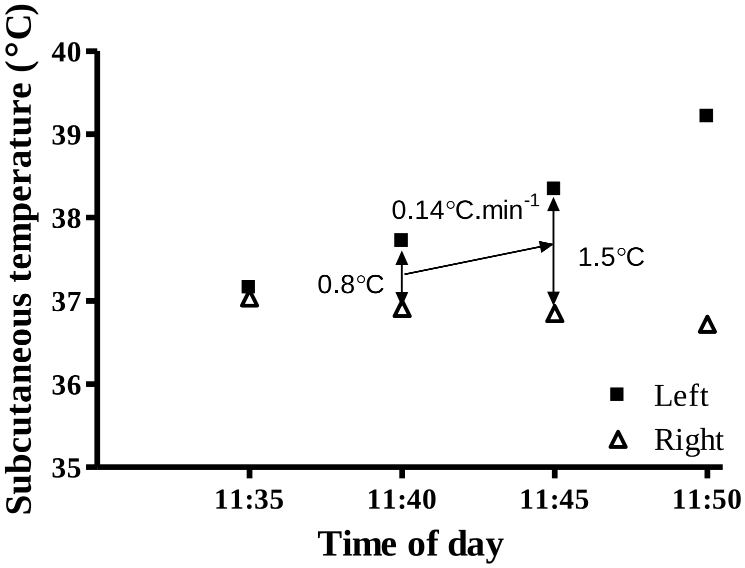


Figure 2.12 The rate of change in difference between left and right subcutaneous temperatures was calculated as the change in difference between left and right subcutaneous temperatures over time.

If the rate of change in subcutaneous temperature difference was less than the negative threshold value, the wildebeest was predicted to be orientated right perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference was greater than the positive threshold value, the wildebeest was predicted to be orientated left perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference was in-between the threshold values, the wildebeest was predicted to be orientated parallel to solar radiation.

To validate whether the rate of change in subcutaneous temperature difference could be used to predict the orientation to solar radiation of wildebeest, I compared the predicted orientation to solar radiation to the observed orientation to solar radiation of wildebeest. I calculated the percentage of correct predictions for each wildebeest individual orientated parallel, left perpendicular and right perpendicular to solar radiation. I calculated the mean of the individual means of the total percentage of correct predictions as an indication of the accuracy of the technique.

2.8.5 Prediction model incorporating both subcutaneous temperature difference and rate of change in subcutaneous temperature difference to predict orientation to solar radiation of wildebeest

For the third technique I used to predict the orientation to solar radiation of wildebeest, I designed a prediction model incorporating both subcutaneous temperature difference and rate of change in subcutaneous temperature difference (See Chapter 3, Results, section 3.5). Using the prediction model and the threshold values calculated earlier, I calculated the orientation to solar radiation of wildebeest. If the rate of change in subcutaneous temperature difference was greater than the positive threshold value, the wildebeest was predicted to be orientated left perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference was less than the negative threshold value, the wildebeest was predicted to be orientated right perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference was in-between the threshold values, but subcutaneous temperature difference was greater than the positive threshold value, the wildebeest was predicted to be orientated left perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference was in-between the threshold values, but subcutaneous temperature difference was less than the negative threshold value, the wildebeest was predicted to be orientated right perpendicular to solar radiation. If both the rate of change in subcutaneous temperature difference and the subcutaneous temperature difference was in-between the threshold values, the wildebeest was predicted to be orientated parallel to solar radiation.

I ran the prediction model in Excel using multiple if-functions and the threshold values calculated for subcutaneous temperature difference (see section 2.10.3) and rate of change in subcutaneous temperature difference (see section 2.10.4). I compared the orientation to solar radiation predicted by the model to the orientation to solar radiation we observed for the wildebeest. I calculated the percentage of times the prediction was correct for each individual wildebeest orientated parallel, left and right perpendicular to solar radiation. I calculated the mean of the individual means of the percentage of total correct predictions for all wildebeest individuals as an indication of the accuracy of the prediction model.

2.8.6 Generalised Linear Mixed Models

To determine which factors affected the prediction model, I ran three Generalised Linear Mixed Models using STATA 10. I wanted to determine which factors affected the subcutaneous temperature difference, the rate of change in subcutaneous temperature difference and the accuracy of the prediction model when all other factors were taken into account. Due to equipment failures I did not have miniature black globe temperature and wind speed for one wildebeest individual for one observation. Therefore the sample size for the Generalised Linear Mixed Models was reduced to 73 observations and 15 individuals (compared to 74 observations and 16 individuals in earlier analyses).

The first Generalised Linear Mixed Model was run to determine which factors affected the subcutaneous temperature difference of wildebeest. The absolute value of subcutaneous temperature difference was entered as the dependant variable. The second Generalised Linear Mixed Model was run to determine which factors affected the rate of change in subcutaneous temperature difference of wildebeest. The absolute value of rate of change in subcutaneous temperature difference was entered as the dependant variable. I used absolute values of subcutaneous temperature difference and rate of change in subcutaneous temperature difference to avoid the use of negative values in the models. The third Generalised Linear Mixed Model was a logistic Generalised Linear Mixed Model to determine which factors affected the accuracy of the prediction model. Whether the prediction accurately reflected the real orientation of the wildebeest (binomial: correct or incorrect) was entered as the dependent variable.

In all three models, body orientation (binomial: perpendicular or parallel), species (binomial: black wildebeest or blue wildebeest), miniature black globe temperature and wind speed were entered as independent variables to test for their relationship with the dependant variable. Because the absolute values of subcutaneous temperature difference and rate of change in subcutaneous temperature difference were used in the generalised linear mixed models, the variable “orientation” only had two categories (parallel and perpendicular), instead of three (parallel, perpendicular left and perpendicular right). Body posture (binomial: lying down or standing) was entered as a control variable because body posture can affect the way a wildebeest takes up heat from the environment (Walsberg, 1992). Animal ID was nested within date of observation as random variables to control for possible pseudo-replication, because for some individuals and on some days there were more observations than for others (Hurlbert, 1984).

2.8.7 Favourable conditions for prediction model

To support the results of the Generalised Linear Mixed Models, I compared the temperature profiles of wildebeest orientated perpendicular or parallel to solar radiation under different circumstances. I investigated the factors that were considered important according to the generalised linear mixed models to determine under which conditions the prediction model was most likely to correctly identify the orientation to solar radiation of wildebeest.

I investigated how subcutaneous temperature difference and rate of change in subcutaneous temperature difference was affected by miniature black globe temperature. The frequency plot of the miniature black globe temperatures was bimodal with a peak at 26 °C and 36 °C (Figure 2.13). Therefore, I divided miniature black globe temperature into “warm” (<30 °C) and “hot” (>30 °C) categories. I compared the subcutaneous temperature difference between wildebeest exposed to miniature black globe temperatures above and below 30 °C using a paired t-test. Similarly, I compared the rate of change in subcutaneous temperature difference when wildebeest were exposed to miniature black globe temperatures above and below 30 °C using a paired t-test. I selected temperature profile

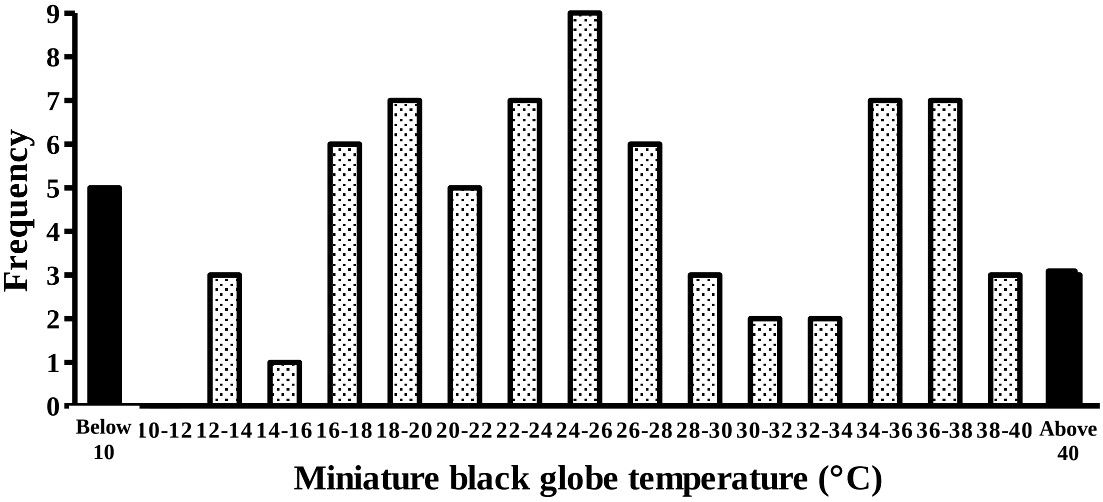


Figure 2.13 Frequency plot of miniature black globe temperatures recorded on the wildebeest collars.

examples that best illustrated the effect of miniature black globe temperature on subcutaneous temperature difference and rate of change in subcutaneous temperature difference.

I investigated the effect wind speed had on subcutaneous temperature difference and the ability of the prediction model to accurately predict the orientation of the wildebeest. I compared the wind speed when predictions were correct and incorrect using a paired t-test. I selected temperature profile examples that best illustrated the effect of wind speed on subcutaneous temperature difference and the ability of the prediction model to accurately predict the orientation of the wildebeest

I investigated whether the prediction model could predict the orientation to solar radiation of a wildebeest equally accurate when a wildebeest was orientated perpendicular and when the wildebeest was orientated parallel to solar radiation. I compared the proportion of correct predictions when a wildebeest was orientated perpendicular and parallel to solar radiation. I selected examples of temperature profiles when a wildebeest was incorrectly predicted as perpendicular when it was orientated parallel and made note of the possible reasons why the prediction was incorrect.

I investigated whether the prediction model could predict the orientation to solar radiation equally accurately for blue wildebeest and black wildebeest. I compared the proportion of correct predictions of orientation to solar radiation for black wildebeest and blue wildebeest.

I investigated whether the body posture of wildebeest affected the ability of the prediction model to accurately predict the orientation to solar radiation in wildebeest. I compared the percentage of wildebeest lying down when the prediction was correct and incorrect using bar graphs and a paired t-test. To illustrate how the body posture of a wildebeest could affect the accuracy of the prediction model, I selected a temperature profile of a wildebeest that lay down and then stood up, whilst maintaining the same orientation to solar radiation.

Chapter 3

Results

3 Results

3.1 Subcutaneous temperature patterns

For one year, the subcutaneously implanted data loggers recorded the subcutaneous temperatures of the left and right flank of free-ranging wildebeest while I conducted behavioural observations of their orientation to solar radiation in their natural environment. Throughout the study, the average daily mean subcutaneous temperature was 36.9 ± 0.6 °C, with a daily minimum of 34.1 ± 1.3 °C, and a daily maximum of 39.2 ± 0.2 °C. The range of subcutaneous temperatures recorded was between 26.3 °C and 41.9 °C, and the daily average standard deviation in subcutaneous temperature was 1.3 ± 0.3 °C. I hypothesised that subcutaneous temperatures on the left and right would differ from one another if one side of the animal had been exposed to greater solar radiation than the other side, because one side had been orientated perpendicular to solar radiation.

When I examined daily subcutaneous temperature profiles of wildebeest, I found that there were often large differences between the left and right subcutaneous temperatures (Figure 3.1). Differences in subcutaneous temperatures appeared to be more prominent in the winter months (Figure 3.1a) than in the summer months (Figure 3.1b). The maximum absolute subcutaneous temperature difference recorded during the study was 10.6 °C.

Not only were there often large differences between left and right subcutaneous temperatures, but there were also often rapid changes in subcutaneous temperatures (for example, at about 9:00 and 12:00 in Figure 3.1a). Rate of change in subcutaneous temperature difference is a measure of how the difference between left and right

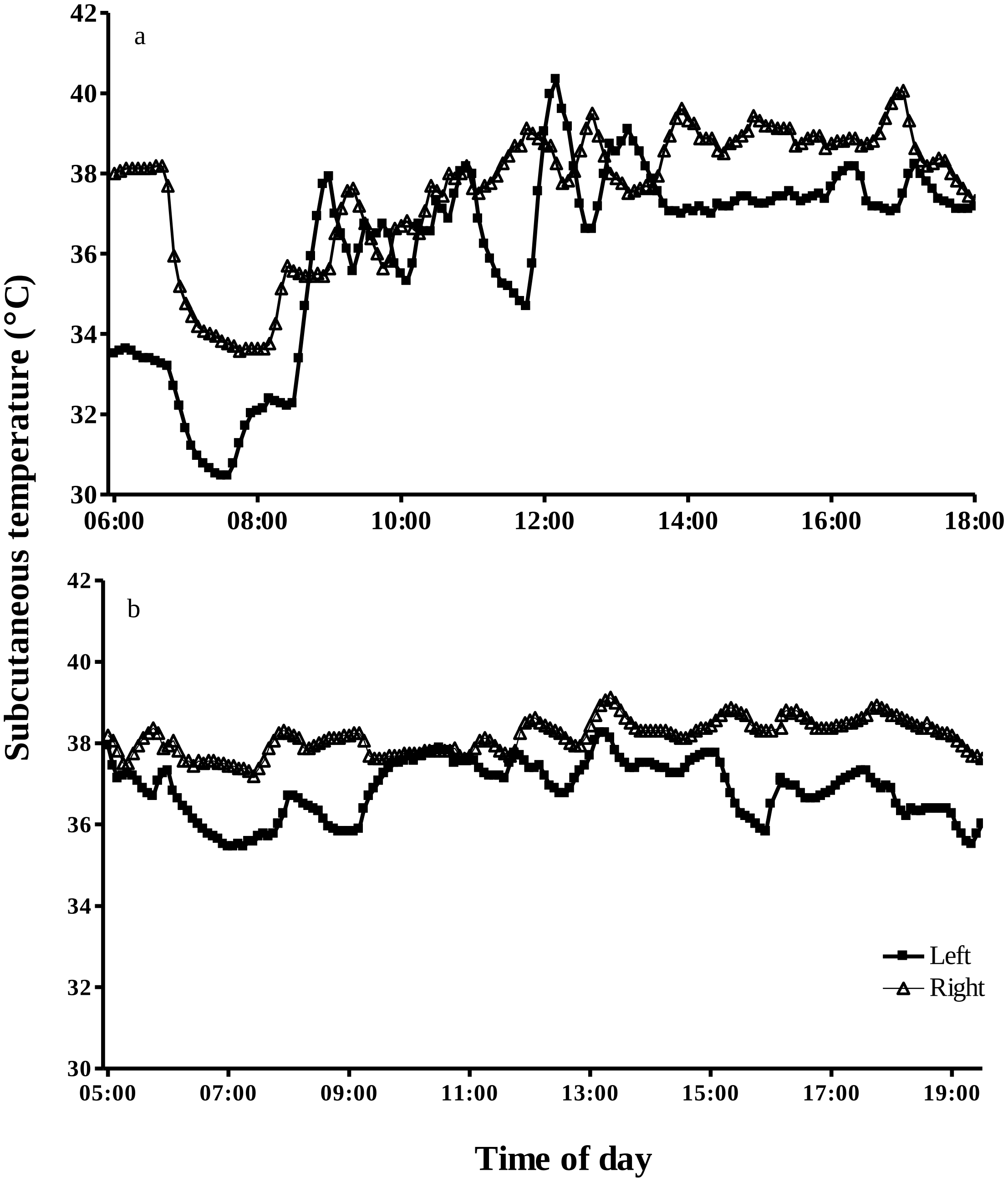


Figure 3.1 An example of the left and right subcutaneous temperatures from one blue wildebeest over the course of one day (sunrise to sunset) in winter (a) and one day (sunrise to sunset) in summer (b) to illustrate the typical daily oscillations in left and right subcutaneous temperatures.

subcutaneous temperatures changed over time. Rapid changes in subcutaneous temperatures on either the left or right side would cause a high absolute rate of change in subcutaneous temperature difference. The maximum absolute rate of change in subcutaneous temperature difference recorded during the study was 0.92 °C.min^{-1}.

3.2 Comparison of subcutaneous temperatures when wildebeest were orientated parallel and perpendicular to solar radiation.

I aimed to test whether the variation in subcutaneous temperatures, measured by the implanted data loggers, could be caused by the orientation of wildebeest relative to solar radiation. I compared the left and right subcutaneous temperatures from each wildebeest to determine whether subcutaneous temperatures displayed different patterns when wildebeest were observed orientated perpendicular to solar radiation compared to when they were observed orientated parallel to solar radiation. There were many examples of data where wildebeest were orientated perpendicular to solar radiation and the subcutaneous temperature of the side facing the sun was greater than that of the other side (Figure 3.2). In the examples when wildebeest were orientated perpendicular to solar radiation, the left and right subcutaneous temperatures could differ by almost 5 °C during the time the wildebeest were observed orientated perpendicular to solar radiation (Figure 3.2). When the wildebeest were orientated with their left side perpendicular to solar radiation (Figure 3.2, panels a, e, and g), the left subcutaneous temperature was greater than the right subcutaneous temperature. In the example where the wildebeest was orientated with its right side perpendicular to solar radiation (Figure 3.2, panel c), the right subcutaneous temperature was greater than the left subcutaneous temperature. When a wildebeest was orientated parallel to solar radiation, the subcutaneous temperatures on both sides were more similar to one another than when a wildebeest was orientated

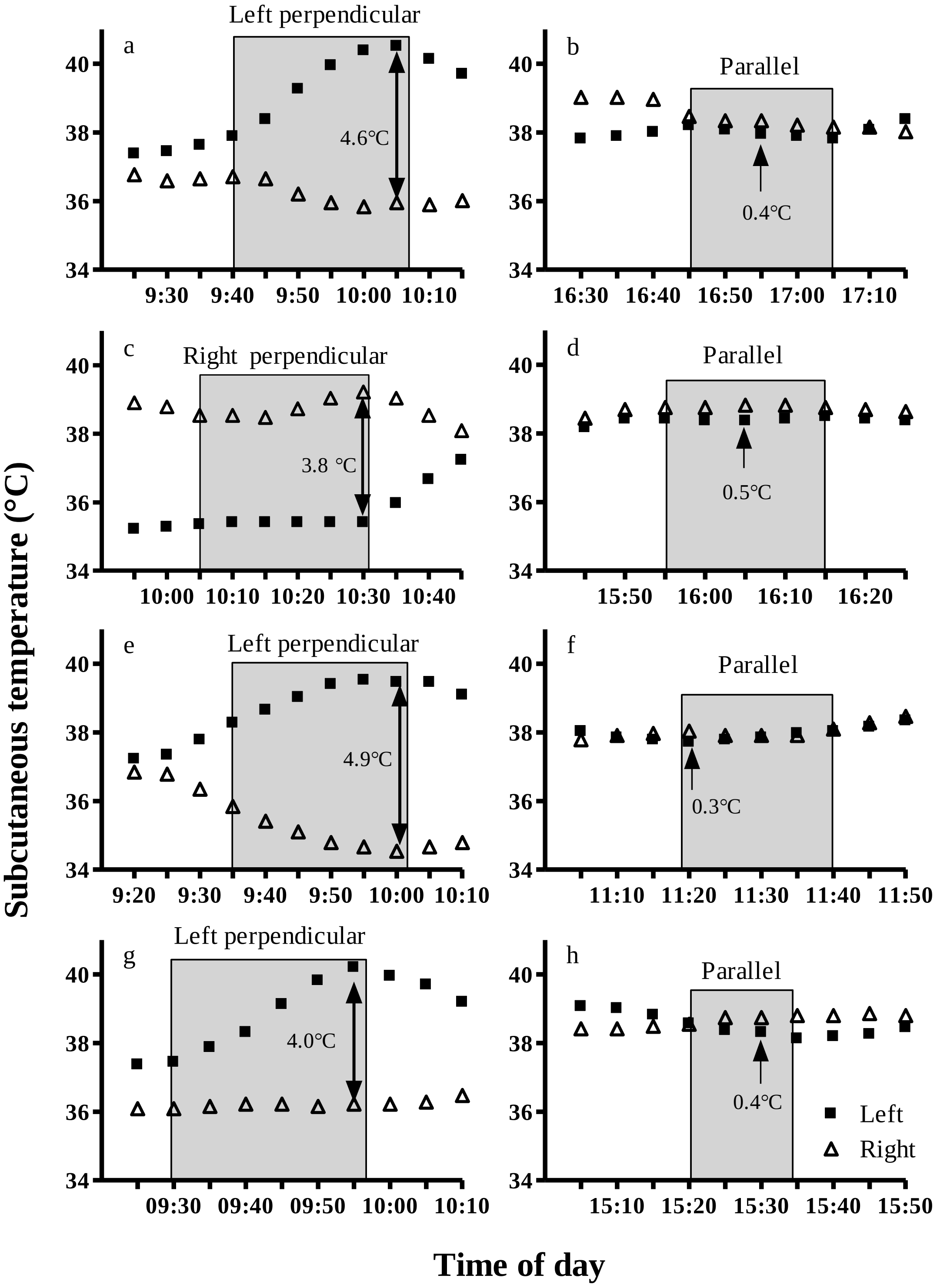


Figure 3.2 Examples of subcutaneous temperature profiles of different wildebeest individuals at times when they were observed orientating perpendicular (panels a, c, e and g) and parallel (panels b, d, f and h) to solar radiation. The graphs show left and right subcutaneous temperatures over time. The observed orientation of wildebeest relative to solar radiation is indicated on the graph. The maximum difference between left and right subcutaneous temperatures during the time the wildebeest was observed in a specific orientation to solar radiation is indicated by arrows. Miniature black globe temperatures from the wildebeest collars at the time of observation were between 17 °C and 41 °C and wind speed was between 1.3 m.s^{-1} and 1.9 m.s^{-1}.

perpendicular to solar radiation (Figure 3.2). In the shown examples of wildebeest orientated parallel to solar radiation, the maximum difference between left and right subcutaneous temperature during the time they were observed orientated parallel to solar radiation was not greater than 0.5 °C (Figure 3.2, panels b, d, f and h). Therefore, there seemed to be a relationship between the orientation of a wildebeest's body to solar radiation and the subcutaneous temperature difference.

In order to determine if the difference between left and right subcutaneous temperatures was related to the orientation to solar radiation of wildebeest, I compared the average absolute subcutaneous temperature difference for each individual wildebeest orientated parallel to solar radiation to that when they were orientated perpendicular to solar radiation (Figure 3.3). The subcutaneous temperature difference when wildebeest were orientated perpendicular to solar radiation was greater than when the animals were orientated parallel to solar radiation (paired t-test, t_9=2.59, p=0.03). On average, the subcutaneous temperature difference when wildebeest were orientating perpendicular to solar radiation was 0.7 °C greater than when they were orientating parallel to solar radiation (Figure 3.3).

To test whether the subcutaneous temperature difference could distinguish the direction of perpendicular orientation of wildebeest (left side or right side perpendicular to solar radiation), I compared the subcutaneous temperature difference when wildebeest were orientated left perpendicular and right perpendicular to solar radiation (Figure 3.4). I found that when wildebeest were orientated left perpendicular to solar radiation the subcutaneous temperature difference was not significantly different from zero (paired t-test, t_{10}=1.34, p=0.2), but when wildebeest were orientated right side perpendicular to solar radiation, the

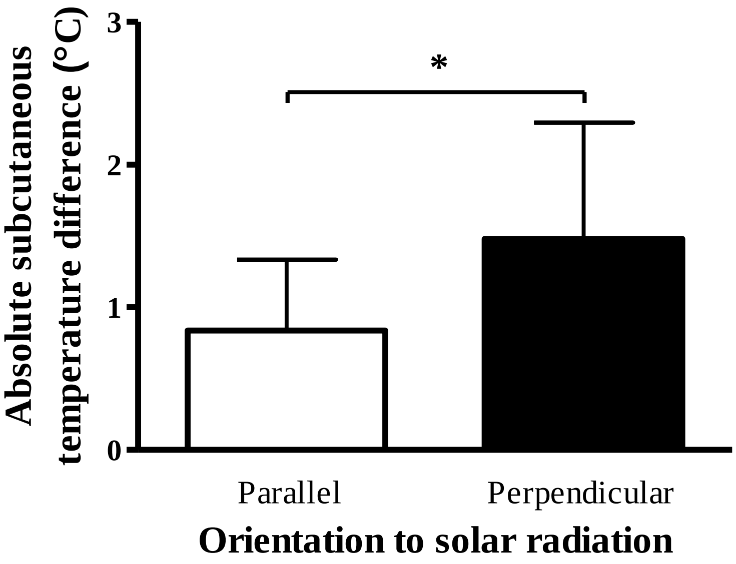


Figure 3.3 The average absolute difference between left and right subcutaneous temperatures of wildebeest orientated perpendicular to solar radiation (mean ± SD = 1.5 ± 0.8 °C) compared to when it was orientated parallel to solar radiation (mean ± SD = 0.8 ± 0.5 °C). Observations = 74, individuals =16, * $p<0.05$.

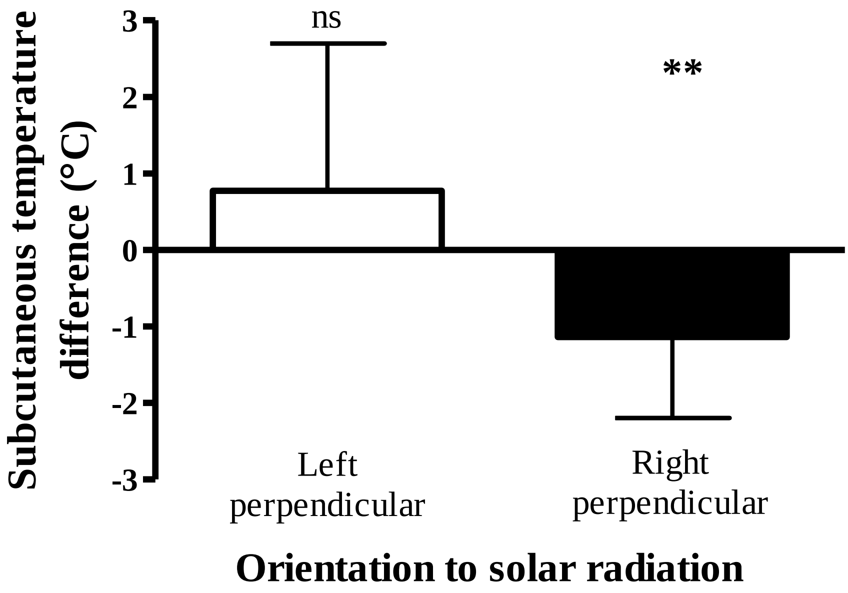


Figure 3.4 The average difference between left and right subcutaneous temperatures of wildebeest orientated left perpendicular to solar radiation, compared to zero (mean ± SD = 0.8 ± 1.9 °C) and the average difference in subcutaneous temperature of wildebeest orientated right perpendicular to solar radiation, compared to zero (mean ± SD = -1.1 ± 1.1 °C). Observations = 74, individuals =16, ns: $p>0.05$, ** $p<0.01$.

subcutaneous temperature difference was significantly different to zero (paired t-test, t_{12}=3.67, p=0.002).

3.3 Difference between left and right subcutaneous temperatures to predict orientation to solar radiation in wildebeest

To successfully predict the orientation of a wildebeest relative to solar radiation, I determined a threshold value, above which the subcutaneous temperature difference would likely indicate that the wildebeest was orientated perpendicular to solar radiation. I used a Receiver Operating Characteristic (ROC) analysis to determine the sensitivity and specificity of different threshold values. Sensitivity (the ability of the technique to make correct predictions) decreased, and specificity (the ability of the technique to exclude incorrect predictions) increased as threshold values increased (Table 3.1). For example, if 0.1 °C (the smallest subcutaneous temperature difference measured) was used as the threshold value, above which a wildebeest would be classified as orientated perpendicular to solar radiation, sensitivity was 100 %, but specificity was 0 % (Table 3.1). Such a threshold would allow me to correctly identify 100 % of all cases were wildebeest were orientated perpendicular to solar radiation. However, the 0 % specificity means that all cases where wildebeest were orientated parallel to solar radiation would have been incorrectly classified as orientated perpendicular to solar radiation. For a threshold of 4.6 °C (the largest subcutaneous temperature difference recorded), 100 % of cases where wildebeest were orientated parallel to solar radiation would be correctly identified, but 0 % of cases where wildebeest were orientated perpendicular to solar radiation would be identified.

Table 3.1 Receiver Operating Characteristics analysis to determine the threshold value with the highest sensitivity and specificity for the difference between left and right subcutaneous temperatures, to accurately predict a wildebeest's orientation to solar radiation.

Threshold subcutaneous temperature difference °C	Sensitivity	Specificity	Correctly Classified
≥ 0.1	100.0%	0.0%	62.2%
≥ 0.2	89.1%	10.7%	59.5%
≥ 0.4	78.3%	25.0%	58.1%
≥ 0.7	69.6%	46.4%	60.8%
≥ 0.8	67.4%	53.6%	62.2%
≥ 0.9	63.0%	57.1%	60.8%
≥ 1.0	**60.9%**	**60.7%**	**60.8%**
≥ 1.2	54.4%	64.3%	58.1%
≥ 1.3	52.2%	71.4%	59.5%
≥ 1.5	50.0%	85.7%	63.5%
≥ 1.7	39.1%	89.3%	58.1%
≥ 1.8	37.0%	92.9%	58.1%
≥ 2.4	26.1%	96.4%	52.7%
≥ 3.5	15.2%	100.0%	47.3%
> 4.6	0.0%	100.0%	37.8%

Observations= 74, individuals=16

Thus, a higher specificity meant that the remote technique would be better able to detect when a wildebeest was orientated parallel to solar radiation than perpendicular to solar radiation, and a higher sensitivity meant that the remote technique would be better able to detect when a wildebeest was orientating perpendicular to solar radiation than parallel to solar radiation. Because I wanted the remote technique to be equally accurate in detecting when wildebeest were orientated perpendicular and parallel to solar radiation, I selected the threshold value with the highest combined sensitivity and specificity. The threshold value I selected was 1.0 °C (Table 3.1, sensitivity: 60.9 %, specificity: 60.7 %).

I tested how often the difference between left and right subcutaneous temperature could correctly predict the orientation to solar radiation of wildebeest using the 1 °C threshold value. If the difference in subcutaneous temperature was greater than 1 °C, the wildebeest was predicted to be orientated with its left side perpendicular to solar radiation. If the subcutaneous temperature difference was smaller than -1 °C, the wildebeest was predicted to be orientated with its right side perpendicular to solar radiation. If the subcutaneous temperature difference was between -1 °C and +1 °C, the wildebeest was predicted to be orientated parallel to solar radiation. If the technique was randomly predicting wildebeest orientation to solar radiation, I expected the prediction for each orientation (parallel, perpendicular left and perpendicular right to solar radiation) to be correct 33 % of the time.

To determine whether these predictions accurately reflected orientation to solar radiation, I compared the predictions to our observations of wildebeest orientation to solar radiation. I averaged the proportion of correct predictions for each individual (Table 3.2). The predictions were more often correct when wildebeest were orientated parallel to solar

Table 3.2 The percentage of times when the difference between left and right subcutaneous temperatures could correctly predict the orientation of a wildebeest as parallel, left perpendicular or right perpendicular to solar radiation using the 1 °C threshold value. If the subcutaneous temperature difference was greater than +1 °C, the wildebeest was predicted to be orientated left side perpendicular to solar radiation. If the subcutaneous temperature difference was less than -1 °C the wildebeest was predicted to be orientated right side perpendicular to solar radiation. If the subcutaneous temperature difference fell within the threshold values (more than -1 °C, but less than +1 °C) the wildebeest was predicted to be orientated parallel to solar radiation.

Individual	Species	Total number of observations	Parallel orientations correctly predicted (%)	Left perpendicular orientations correctly predicted (%)	Right perpendicular orientations correctly predicted (%)	Average correct predictions (%)
1	Black	2			100	100
2	Black	4	0	0	100	25
3	Black	6	50		50	50
4	Black	5	67		50	60
5	Black	1		0		0
6	Black	1			0	0
7	Black	12	43	0	33	33
8	Black	4	100		50	75
9	Blue	2		100	0	50
10	Blue	11	25	50	80	55
11	Blue	5		33	50	40
12	Blue	7	100	25	100	57
13	Blue	3		100	50	67
14	Blue	5	100	0	0	20
15	Blue	3	100	0		67
16	Blue	3	100	0		67
		74	Average: 68	28	51	48

radiation (68 %, Table 3.2) than when they were orientated with their left (28 %, Table 3.2) or right (51 %, Table 3.2) side perpendicular to solar radiation. The overall average percentage of correct predictions was 48 % (Table 3.2). If I calculated the mean, weighted by the number of observations of each animal, the average percentage of correct predictions was still 48 %.

3.4 Rate of change in difference between left and right subcutaneous temperatures to predict orientation to solar radiation in wildebeest

In an effort to improve the remote technique to detect orientation to solar radiation in wildebeest, I examined the subcutaneous temperature profiles during instances when the subcutaneous temperature difference could not indicate the correct orientation to solar radiation of the wildebeest. For example, there were cases when the right subcutaneous temperature was greater than the left subcutaneous temperature, even though the wildebeest was observed to be orientated with its left side perpendicular to solar radiation. I noticed that such cases were often associated with a recent shift in wildebeest orientation to solar radiation. If a wildebeest was orientated with its right side perpendicular to solar radiation and then turned to orientate its left side perpendicular to solar radiation, the right side was still warmer than the left side from being perpendicular to solar radiation previously (Figure 3.5, at arrow a). In the example in Figure 3.5, immediately after the wildebeest turned its body orientation left perpendicular to solar radiation, the left subcutaneous temperature started increasing and the right subcutaneous temperature started decreasing until the left subcutaneous temperature was greater than the right subcutaneous temperature (at arrow b). The length of time it took for the left subcutaneous temperature to increase (and the right subcutaneous temperature to decrease) sufficiently for the left subcutaneous temperature to be greater than the right subcutaneous temperature

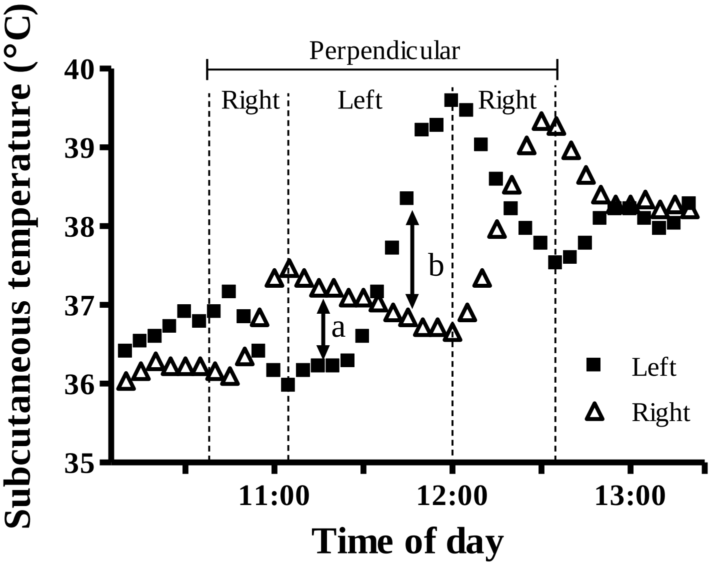


Figure 3.5 Left and right subcutaneous temperatures of a wildebeest observed to be orientated with its right side perpendicular to solar radiation, then orientated left perpendicular to solar radiation, and then orientated right perpendicular to solar radiation again. After the wildebeest orientated left perpendicular to solar radiation, the right subcutaneous temperature was still greater than the left subcutaneous temperature (a). The left subcutaneous temperature increased until it was greater than the right subcutaneous temperature (b).

depended on the difference in subcutaneous temperature before the wildebeest orientated perpendicular to solar radiation and on how quickly the subcutaneous temperatures changed after the wildebeest orientated perpendicular to solar radiation. Therefore, the previous orientation to solar radiation affected the subcutaneous temperature and may have led to inaccurate predictions of orientation to solar radiation if subcutaneous temperature difference was used, on its own, to predict the orientation of wildebeest relative to solar radiation.

In the example, during the time when left subcutaneous temperatures increased and right subcutaneous temperatures decreased, there was a rapid change in subcutaneous temperature difference. Therefore, I investigated the possibility of using rate of change in subcutaneous temperature difference to predict orientation to solar radiation instead of difference in subcutaneous temperature difference. I compared the rate of change in subcutaneous temperature difference of examples where wildebeest were orientated perpendicular and parallel to solar radiation. When wildebeest turned their bodies to be orientated perpendicular to solar radiation, the subcutaneous temperature on the side orientated perpendicular to solar radiation increased, resulting in a greater rate of change in subcutaneous temperature difference than when they were orientated parallel to solar radiation. In two examples where wildebeest were observed to be orientated perpendicular to solar radiation, the maximum rate of change in subcutaneous temperature difference during the time they were observed orientated perpendicular to solar radiation was greater than the maximum rate of change in subcutaneous temperature difference when wildebeest were orientated parallel to solar radiation (Figure 3.6). I hypothesised that a large rate of change in subcutaneous temperature difference could indicate that wildebeest were orientated perpendicular to solar radiation and a small rate of change in subcutaneous

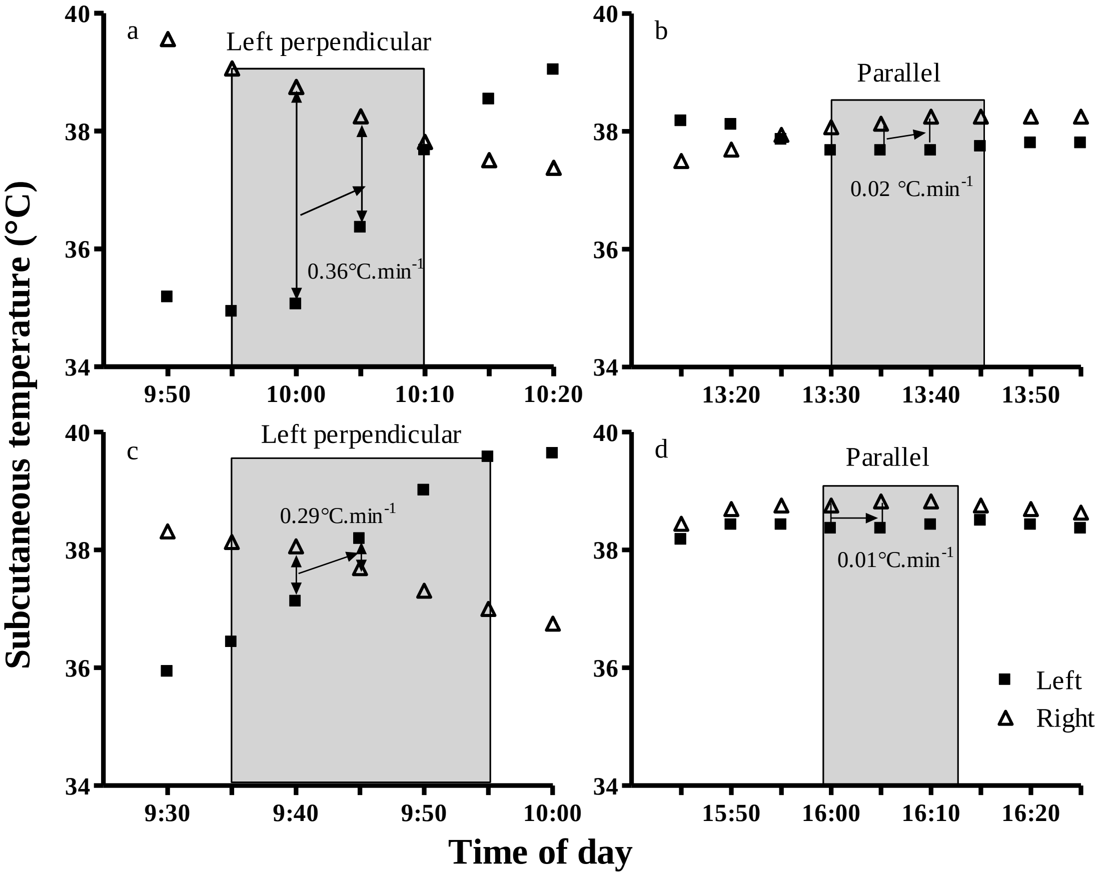


Figure 3.6 Examples of subcutaneous temperature profiles of different wildebeest individuals at times when they were observed orientating perpendicular (panels a and c) and parallel (panels b and d) to solar radiation. The graphs show left and right subcutaneous temperatures over time. The observed orientation of wildebeest relative to solar radiation is indicated on the graph. The maximum rate of change in subcutaneous temperature difference during the time the wildebeest was observed in a specific orientation to solar radiation is indicated by arrows. The miniature black globe temperatures from the wildebeest collars at the time of observation were between 17 °C and 41 °C and wind speed was between 0.7 $m.s^{-1}$ and 2.0 $m.s^{-1}$.

temperature difference could indicate that a wildebeest was orientated parallel to solar radiation.

I compared the rate of change in subcutaneous temperature difference for individuals orientated parallel and perpendicular to solar radiation (Figure 3.7). When wildebeest were orientated perpendicular to solar radiation the absolute rate of change in subcutaneous temperature difference was not significantly different from the absolute rate of change in subcutaneous temperature difference when wildebeest were orientated parallel to solar radiation (paired t-test, t_9=1.84, p=0.09). However, the rate of change in subcutaneous temperature difference when wildebeest were orientated left perpendicular to solar radiation was significantly different from zero (paired t-test, t_{10}=5.5, p=0.0003). When wildebeest were orientated right perpendicular to solar radiation, the rate of change in subcutaneous temperature difference was also significantly different from zero (paired t-test, t_{11}=3.4, p=0.006; Figure 3.8).

To use rate of change in subcutaneous temperature difference to predict orientation to solar radiation I needed a threshold value, above which the rate of change in subcutaneous temperature difference was large enough to indicate that the wildebeest was likely orientated perpendicular to solar radiation. According to the Receiver Operating Characteristic analysis, the threshold value for rate of change in subcutaneous temperature difference with the highest combined sensitivity and specificity values was 0.04 °C.min^{-1} (Table 3.3, sensitivity: 69.6 %, specificity: 46.4 %).

I therefore used rate of change in subcutaneous temperature difference to predict orientation to solar radiation using the threshold value of 0.04 °C.min^{-1}. If the rate of

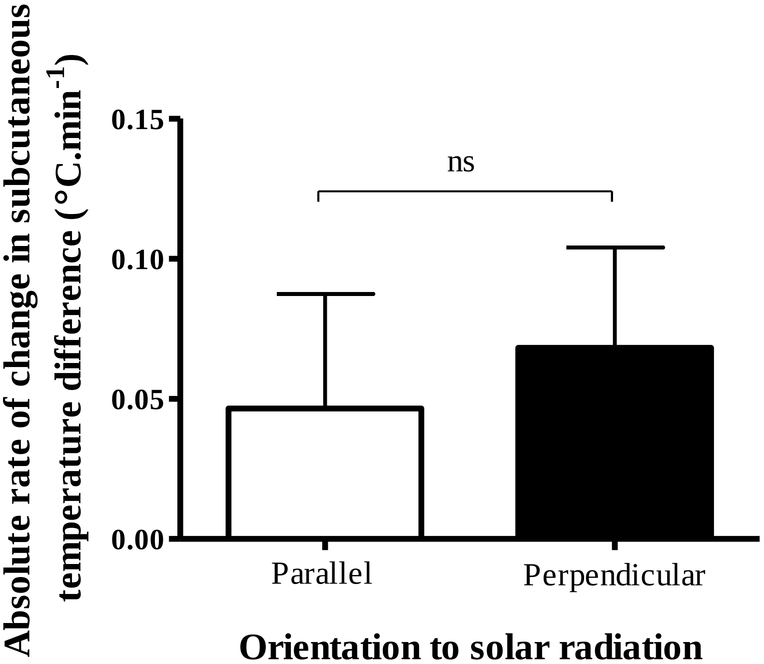


Figure 3.7 The average absolute rate of change in subcutaneous temperature difference was not significantly greater when a wildebeest was orientated parallel to solar radiation (mean ± SD = 0.05 ± 0.04 °C.min^{-1}) than when a wildebeest was orientated perpendicular to solar radiation (mean ± SD = 0.07 ± 0.04 °C.min^{-1}). Observations = 74, individuals=16, ns: $p>0.05$.

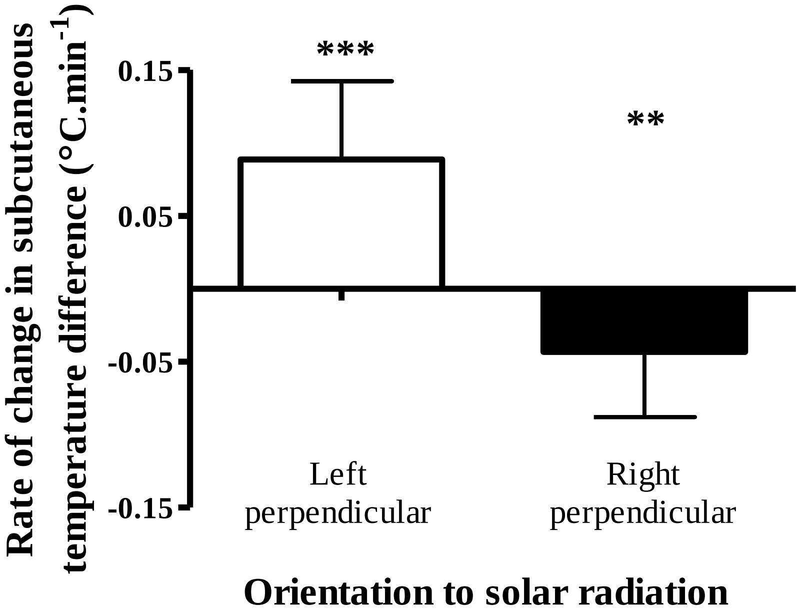


Figure 3.8 The average rate of change in subcutaneous temperature difference was significantly different from zero when a wildebeest was orientated left perpendicular to solar radiation (mean ± SD = 0.09 ± 0.05 °C.min^{-1}, SD) and when a wildebeest was orientated right perpendicular to solar radiation (mean ± SD = -0.04 ± 0.04 °C.min^{-1}, SD). Observations = 74, individuals =16, ** $p<0.01$, *** $p<0.001$.

Table 3.3 Receiver Operating Characteristic analysis to determine the threshold value with the highest sensitivity and specificity for rate of change in subcutaneous temperature difference to accurately predict the orientation of a wildebeest relative to solar radiation.

Threshold rate of change in subcutaneous temperature difference °C.min^{-1}	Sensitivity	Specificity	Correctly classified
≥ 0.00	100.0%	0.0%	62.2%
≥ 0.01	93.5%	10.7%	62.2%
≥ 0.02	78.3%	28.6%	59.5%
≥ 0.04	**69.6%**	**46.4%**	**60.8%**
≥ 0.05	56.5%	60.7%	58.1%
≥ 0.06	52.2%	67.9%	58.1%
≥ 0.07	41.3%	71.4%	52.7%
≥ 0.08	34.8%	75.0%	50.0%
≥ 0.09	32.6%	78.6%	50.0%
≥ 0.10	26.1%	82.1%	47.3%
≥ 0.12	21.7%	89.3%	47.3%
≥ 0.16	15.2%	96.4%	46.0%
≥ 0.23	8.7%	100.0%	43.2%
≥ 0.35	2.2%	100.0%	39.2%
> 0.35	0.0%	100.0%	37.8%

Observations = 74, individuals =16

change in subcutaneous temperature difference was greater than 0.04 °C.min^{-1}, the wildebeest was predicted to be orientated left side perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference was less than -0.04 °C.min^{-1}, the wildebeest was predicted to be orientated right side perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference was within the threshold values (greater than -0.04 °C.min^{-1} but less than 0.04 °C.min^{-1}), the wildebeest was predicted to be orientated parallel to solar radiation. To determine whether rate of change in subcutaneous temperature difference could accurately reflect orientation to solar radiation, the predictions were compared to the visual observations of the orientation to solar radiation of the wildebeest at that time.

By using rate of change in subcutaneous temperature difference to predict orientation to solar radiation, my remote data logging technique accurately reflected the orientation to solar radiation of wildebeest 55 % of the time (Table 3.4). The orientation of a wildebeest was more often correctly predicted when the wildebeest was orientated parallel to solar radiation (67 %, Table 3.4) than when the wildebeest was orientated with its left side perpendicular to solar radiation (56 %, Table 3.4) or when the wildebeest was orientated with its right side perpendicular to solar radiation (49 %, Table 3.4). If I calculated the mean, weighted by the number of observations of each animal, the average percentage of correct predictions was 57 %.

In the previous example of the subcutaneous temperature profile to illustrate high rate of change in subcutaneous temperature difference when a wildebeest was observed to be orientated perpendicular to solar radiation (Figure 3.6a), at the same time when the

Table 3.4 The percentage of times when rate of change in difference between left and right subcutaneous temperatures could correctly predict the orientation of a wildebeest as parallel, left perpendicular or right perpendicular to solar radiation using the 0.04 °C.min^{-1} threshold value. If the rate of change in subcutaneous temperature difference was more than +0.04 °C.min^{-1}, the wildebeest was predicted to be orientated left perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference was less than -0.04 °C.min^{-1} the wildebeest was predicted to be orientated right perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference fell within the threshold values (more than -0.04 °C.min^{-1}, but less than +0.04 °C.min^{-1}) the wildebeest was predicted to be orientated parallel to solar radiation.

Individual	Species	Total number of observations	Parallel orientations correctly predicted (%)	Left perpendicular orientations correctly predicted (%)	Right perpendicular orientations correctly predicted (%)	Average correct predictions (%)
1	Black	2			50	50
2	Black	4	100	50	0	50
3	Black	6	25		50	33
4	Black	5	0		100	40
5	Black	1		0		0
6	Black	1			0	0
7	Black	12	71	0	67	58
8	Black	4	50		50	50
9	Blue	2		100	100	100
10	Blue	11	75	50	60	64
11	Blue	5		67	0	40
12	Blue	7	50	50	0	43
13	Blue	3		100	100	100
14	Blue	5	100	100	67	80
15	Blue	3	100	0		67
16	Blue	3	100	100		100
		74	Average: 67	56	49	55

maximum rate of change in subcutaneous temperature difference was measured (0.36 °C.min^{-1}), the right subcutaneous temperature was 1.9 °C greater than the left subcutaneous temperature, even though the wildebeest was observed to be orientated left perpendicular to solar radiation. Therefore, if I had used subcutaneous temperature difference to predict orientation to solar radiation, the orientation would be incorrectly predicted as right perpendicular to solar radiation (subcutaneous temperature difference was greater than 1 °C with right subcutaneous temperature being hotter than left subcutaneous temperature). However, because there was a steep increase in left side subcutaneous temperature, the rate of change in subcutaneous temperature difference was greater than the threshold value of 0.04 °C.min^{-1} and the orientation was correctly predicted as left perpendicular to solar radiation. Therefore, in cases where the wildebeest had recently shifted its orientation to solar radiation, rate of change in subcutaneous temperature difference could, in some cases, correctly indicate the orientation to solar radiation of wildebeest regardless of the orientation to solar radiation it had maintained previously.

3.5 Comparison of the effectiveness of subcutaneous temperature difference and rate of change in subcutaneous temperature difference to predict orientation to solar radiation

Using rate of change in subcutaneous temperature difference to predict orientation to solar radiation was more often correct (55 %, Table 3.4) than was using difference in subcutaneous temperature to predict the orientation to solar radiation of wildebeest (48 %, Table 3.2). However, rate of change in subcutaneous temperature difference does not always give an accurate reflection of the orientation of a wildebeest to solar radiation. The ability to predict wildebeest orientation to solar radiation using different measurements often depended on how long the wildebeest had been orientated in a position. If a wildebeest had been orientated parallel or perpendicular to solar radiation for a long time, the subcutaneous temperatures would have reached equilibrium and subcutaneous temperature difference would be a better measurement to use to predict the orientation to solar radiation than would rate of change in subcutaneous temperature difference. For example, in Figure 3.9, a wildebeest was orientated with its right side perpendicular to solar radiation and the absolute subcutaneous temperature difference was 2.02 °C and the rate of change in subcutaneous temperature difference was 0.01 °C.min^{-1}. The subcutaneous temperature difference was greater than the threshold value of 1 °C, whereas the rate of change in subcutaneous temperature difference was less than the threshold value of 0.04 °C.min^{-1}. Therefore, if a wildebeest had been orientated in the same position for a long time and the subcutaneous temperatures had stabilised, rate of change in subcutaneous temperature difference would not be useful to distinguish between perpendicular and parallel orientation to solar radiation. In that case, using subcutaneous temperature difference would give a more accurate reflection of wildebeest orientation to solar radiation than rate of change in subcutaneous temperature difference. Subcutaneous temperatures reaching equilibrium after being orientated perpendicular to solar radiation for a long time could be the reason absolute rate of change in subcutaneous temperature difference was previously found to be not significantly greater when wildebeest were orientated perpendicular to solar radiation than when wildebeest were orientated parallel to solar radiation (Figure 3.7). Therefore, I thought to combine the two methods to improve the ability of the technique to distinguish between parallel and perpendicular orientation to solar radiation, as well as to determine the direction of the orientation to solar radiation.

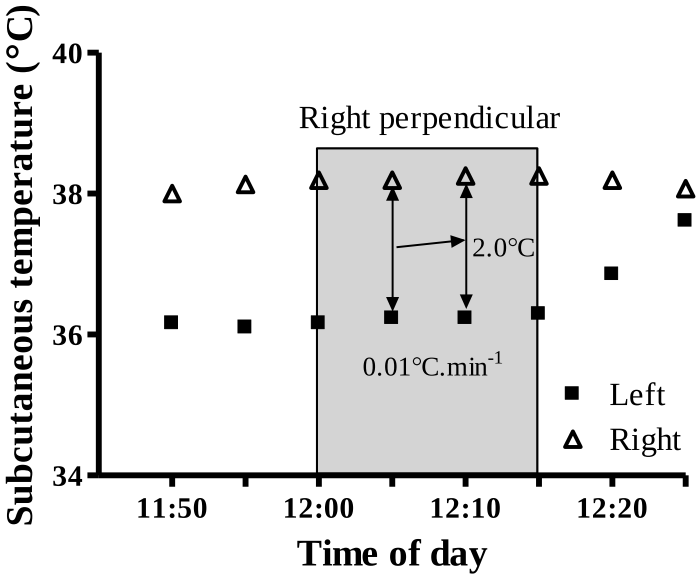


Figure 3.9 Left and right subcutaneous temperatures over time, of a wildebeest observed orientating right perpendicular to solar radiation. The wildebeest had been orientating perpendicular to solar radiation long enough for the subcutaneous temperatures to have stabilized: the subcutaneous temperature difference was 2.02 °C (greater than the threshold value of 1 °C) but the rate of change in subcutaneous temperature difference was 0.01 °C.min^{-1} (less than the threshold value of 0.04 °C.min^{-1}). At the time of observation, miniature black globe temperature from the wildebeest collar was 17 °C and wind speed was 1.3 m.s^{-1}.

I produced a hypothetical graph (Figure 3.10) to illustrate changes in subcutaneous temperature over time while a wildebeest was orientated perpendicular to solar radiation. If a wildebeest was orientated with its left side perpendicular to solar radiation long enough for the subcutaneous temperatures to stabilise, the left subcutaneous temperature would be greater than the right subcutaneous temperature (Figure 3.10, arrow a). If the wildebeest then turned its right side perpendicular to solar radiation, the left subcutaneous temperature (still greater than right subcutaneous temperature) would start to decrease and the right subcutaneous temperature would increase, causing a rapid change in subcutaneous temperature difference (Figure 3.10, arrow b), until the right side temperature was greater than the left side (Figure 3.10, arrow c). Finally, subcutaneous temperatures would stabilise again. There would be a substantial difference in subcutaneous temperatures with the right subcutaneous temperature being greater than the left subcutaneous temperature, but the rate of change in subcutaneous temperature difference would be small (Figure 3.10, arrow d). Therefore, I hypothesised that, when a wildebeest had recently changed its orientation to solar radiation, rate of change in subcutaneous temperature difference would be a better indicator of a wildebeest’s orientation to solar radiation than subcutaneous temperature difference, but when a wildebeest has maintained an orientation to solar radiation long enough for the subcutaneous temperatures to have stabilised, it would be better to use difference in subcutaneous temperature to predict the orientation to solar radiation than to use rate of change in subcutaneous temperature difference. Because I often did not know how long a wildebeest had been orientated to solar radiation in a specific way, combining subcutaneous temperature difference and rate of change in subcutaneous temperature difference to predict the orientation to solar radiation of a wildebeest could potentially improve the accuracy of the remote technique.

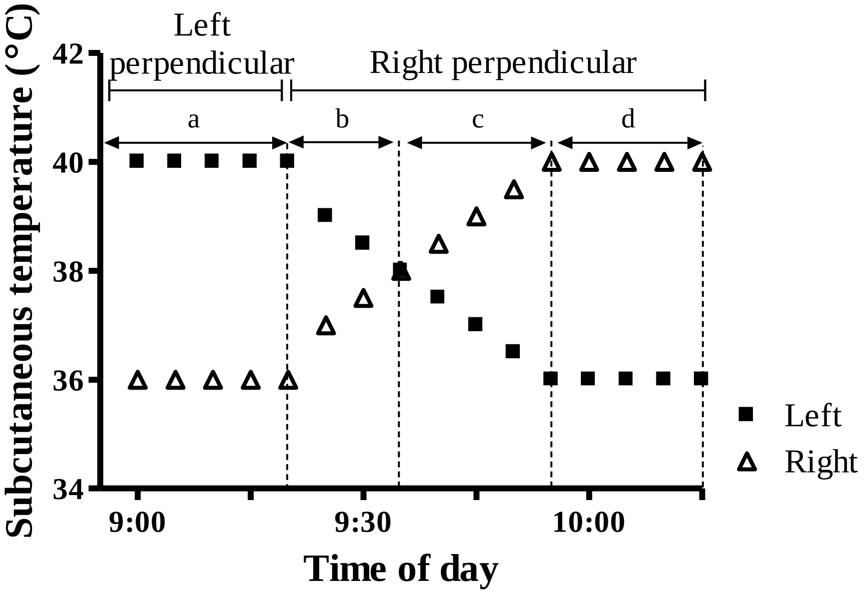


Figure 3.10 Hypothetical graph illustrating how subcutaneous temperatures theoretically change when a wildebeest was orientated with its left side perpendicular to solar radiation and then turned its body to be orientated with its right side perpendicular to solar radiation. The wildebeest had been orientated left side perpendicular to solar radiation long enough for subcutaneous temperatures to have stabilized (at arrow a). Then the wildebeest turned its body so the right side was orientated perpendicular to solar radiation, but the left subcutaneous temperature was still greater than the right subcutaneous temperature (at arrow b). Right subcutaneous temperature increased until it was greater than left subcutaneous temperature (at arrow c) and the subcutaneous temperatures stabilised again (at arrow d).

3.6 Prediction model incorporating both subcutaneous temperature difference and rate of change in subcutaneous temperature difference as predictors of orientation to solar radiation

I designed a prediction model based on the hypothetical example (Figure 3.10), incorporating both subcutaneous temperature difference and rate of change in subcutaneous temperature difference, to predict orientation to solar radiation. The prediction model used a stepwise approach to predict orientation to solar radiation (Figure 3.11).

If the absolute rate of change in subcutaneous temperature difference was greater than the threshold value (0.04 °C.min^{-1}), the wildebeest was predicted to be orientated perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference was greater than the positive threshold value ($\geq$ 0.04 °C.min^{-1}), the wildebeest was predicted to be orientated left side perpendicular to solar radiation. If the rate of change in subcutaneous temperature difference was less than the negative threshold value ($\leq$ -0.04 °C.min^{-1}), the wildebeest was predicted to be orientated right side perpendicular to solar radiation.

If the rate of change in subcutaneous temperature difference fell within the threshold values (greater than -0.04 °C.min^{-1} but less than 0.04 °C.min^{-1}), there were two possible explanations: either the wildebeest was orientated parallel to solar radiation, or the wildebeest was orientated perpendicular to solar radiation and the subcutaneous temperatures had stabilised. The subcutaneous temperature difference determined which one of these two possibilities was true. If the subcutaneous temperature difference was greater than the positive threshold value ($\geq$ 1 °C) the wildebeest was predicted to be

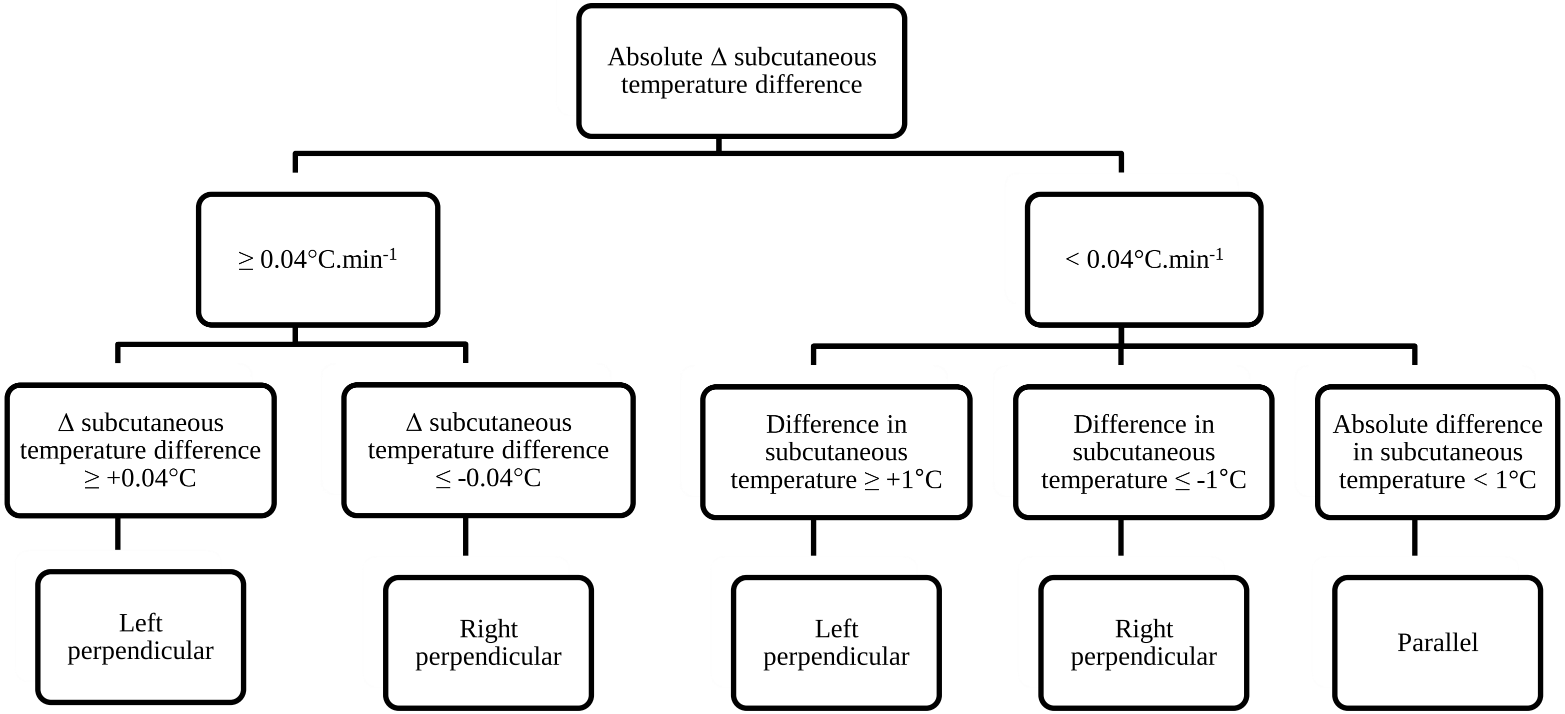


Figure 3.11 A diagram explaining the prediction model I designed to predict the orientation of a wildebeest's body relative to solar radiation using a stepwise approach incorporating both difference between left and right subcutaneous temperatures and rate of change in subcutaneous temperature difference. Δ = rate of change in.

orientated left side perpendicular to solar radiation. If the subcutaneous temperature difference was less than the negative threshold value (≤ -1 °C) the wildebeest was predicted to be orientated right side perpendicular to solar radiation. If the subcutaneous temperature difference was within these threshold values (greater than -1 °C but less than +1 °C) the wildebeest was predicted to be orientated parallel to solar radiation.

To determine how accurately the prediction model, which incorporated both difference in subcutaneous temperature and rate of change in subcutaneous temperature difference, could predict orientation to solar radiation, I calculated the percentage of times each observed orientation to solar radiation was correctly predicted by the model for each individual and averaged the percentage of correct predictions for all the individuals. The prediction model was more often correct at predicting orientation to solar radiation when wildebeest were orientated right (82 %, Table 3.5) or left (56 %, Table 3.5) perpendicular to solar radiation than when the wildebeest were orientated parallel to solar radiation (49 %, Table 3.5). In total, the prediction model correctly reflected the orientation to solar radiation 60 % of the time (Table 3.5). If I calculated the mean, weighted by the number of observations of each animal, the average percentage of correct predictions would be 59 %. If the prediction model was randomly predicting orientation to solar radiation, I would expect the prediction model to be correct 33 % of the time. Therefore the prediction model was more often correct than would be expected by chance.

The prediction model using both subcutaneous temperature difference and rate of change in subcutaneous temperature difference was more accurate (60 %, Table 3.5) than were the models using either difference in subcutaneous temperature (48 %, Table 3.2) or rate of change in subcutaneous temperature difference (55 %, Table 3.4) on their own to predict

Table 3.5 The percentage of times when the prediction model could correctly predict the orientation of black and blue wildebeest as parallel, left perpendicular or right perpendicular to solar radiation using the 1 °C threshold value for subcutaneous temperature difference and the 0.04 °C.min^{-1} threshold value for rate of change in subcutaneous temperature difference. If either subcutaneous temperature difference or rate of change in subcutaneous temperature difference was above the positive threshold value, the wildebeest was predicted to be orientated left perpendicular to solar radiation. If either subcutaneous temperature difference or rate of change in subcutaneous temperature difference was below the negative threshold value, the wildebeest was predicted to be orientated right perpendicular to solar radiation. If both subcutaneous temperature difference and rate of change in subcutaneous temperature difference fell within the threshold values, the wildebeest was predicted to be orientated parallel to solar radiation.

Individual	Species	Number of observations	Parallel orientations correctly predicted (%)	Left perpendicular orientations correctly predicted (%)	Right perpendicular orientations correctly predicted (%)	Total correct predictions (%)
1	Black	2			100	100
2	Black	4	0	50	100	50
3	Black	6	25		100	50
4	Black	5	0		100	40
5	Black	1		0		0
6	Black	1			0	0
7	Black	12	43	0	100	50
8	Black	4	50		50	50
9	Blue	2		100	100	100
10	Blue	11	25	50	80	55
11	Blue	5		67	50	60
12	Blue	7	50	50	100	57
13	Blue	3		100	100	100
14	Blue	5	100	100	67	80
15	Blue	3	100	0		67
16	Blue	3	100	100		100
		74	Average: 49	56	81	60

orientation to solar radiation. However, the prediction model incorrectly classified wildebeest orientation to solar radiation 40 % of the time. To determine why the percentage of incorrect predictions was so high, I investigated specific examples of when the technique did not work.

3.7 Border-line cases

One drawback of the prediction model is that there will always be border-line cases that fall just inside or outside of the threshold values. Because there was a lot of variance in both the subcutaneous temperature difference (Figure 3.4) and rate of change in subcutaneous temperature difference (Figure 3.7) when wildebeest were orientated perpendicular and parallel to solar radiation, there were many examples where subcutaneous temperature difference and/or rate of change in subcutaneous temperature difference fell just inside or outside of the threshold value. For example, large differences in subcutaneous temperature allowed for a high degree of certainty that the prediction model would correctly predict orientation to solar radiation, but when the subcutaneous temperature difference was closer to the threshold value, the orientation to solar radiation could easily be incorrectly classified (Figure 3.12). Similarly, there were borderline cases where rate of change in subcutaneous temperature difference was close to the threshold value.

3.8 Factors that influenced the accuracy of the prediction model

Not all instances where the prediction model incorporating subcutaneous temperature difference and rate of change in subcutaneous temperature difference incorrectly reflected the orientation to solar radiation of wildebeest were border-line cases. Therefore, I investigated which factors could influence the subcutaneous temperature difference and

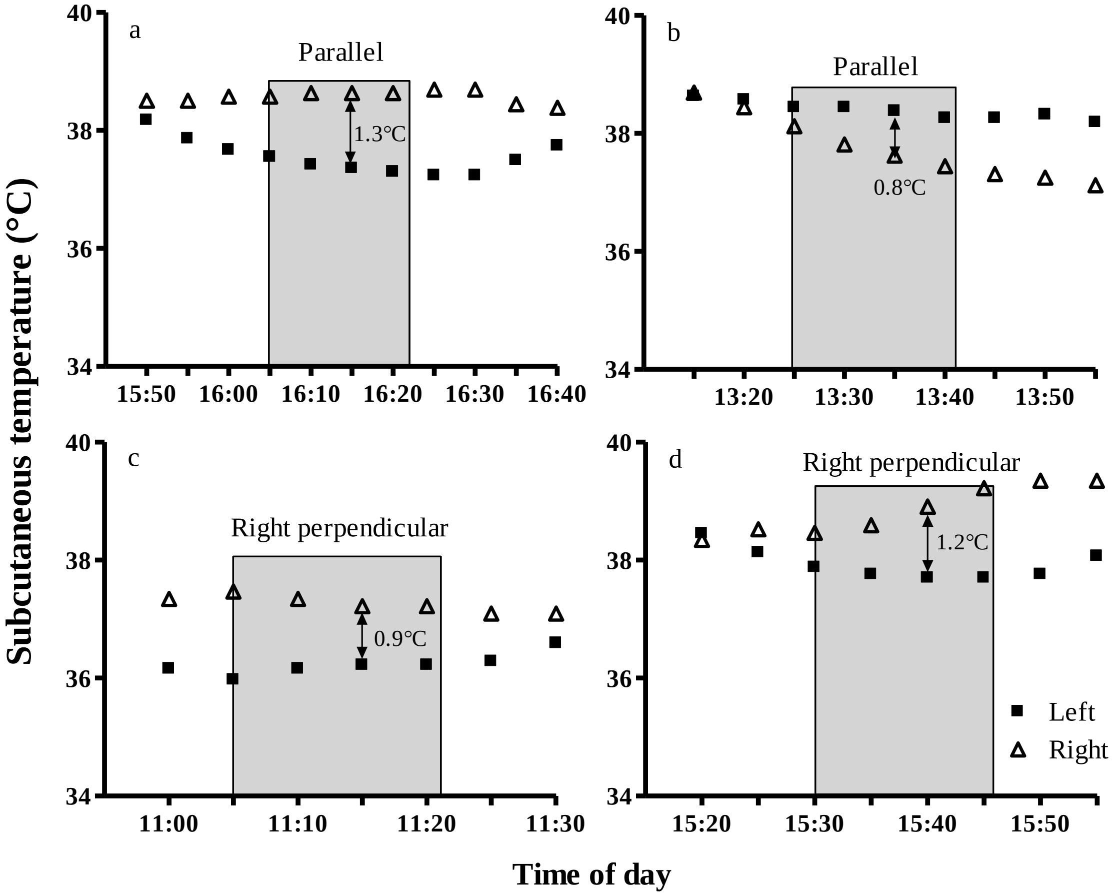


Figure 3.12 Examples of subcutaneous temperature profiles of different wildebeest individuals observed orientated parallel and perpendicular to solar radiation. In all cases the subcutaneous temperature difference was close to the threshold value (1 °C). In two of the cases (panels a and c) the orientation to solar radiation was incorrectly identified. In the other two cases (panels b and d) the orientation to solar radiation was correctly identified.

rate of change in subcutaneous temperature difference and how these factors negatively affected the accuracy of the prediction model. The factors I investigated were orientation to solar radiation (parallel vs. perpendicular), species (black wildebeest or blue wildebeest), miniature black globe temperature (measured as °C on the collar attached around the wildebeest's neck), wind speed ($m.s^{-1}$) and wildebeest body posture (standing vs. lying down). Random factors that were included were wildebeest individual and date of observation to prevent pseudo-replication (see Chapter 2, Materials and Methods, section 2.8.6 for a full description).

3.8.1 Factors that influenced the difference between left and right side subcutaneous temperatures

According to the Generalised Linear Mixed Model, orientation to solar radiation, miniature black globe temperature (°C) and wind speed ($m.s^{-1}$) had a significant ($p \leq 0.05$) effect on the difference between left and right subcutaneous temperatures when the effects of all other factors were taken into account (Table 3.6). Species and posture did not have a significant effect on the difference between left and right subcutaneous temperatures (Table 3.6, $p > 0.05$). Therefore the difference in subcutaneous temperatures did not vary between black and blue wildebeest (Table 3.6, $p = 0.36$). Whether an animal was lying down or standing did not affect the subcutaneous temperature difference (Table 3.6, $p = 0.71$).

A greater subcutaneous temperature difference was associated with wildebeest orientated perpendicular to solar radiation than with wildebeest orientated parallel to solar radiation (Table 3.6, $p < 0.05$). The significant relationship between subcutaneous temperature difference and orientation to solar radiation confirms the earlier findings that the difference

Table 3.6 Generalised Linear Mixed Model results to test which factors had a significant effect on the absolute difference between the left and right side subcutaneous temperature. The table shows the coefficient, standard error (ß ± SE), z-value, p-value and 95 % confidence interval for orientation to solar radiation, species, miniature black globe temperature (°C) measured on the wildebeest collar, wind speed ($m.s^{-1}$) and posture. Random factors that were included were animal individual and date of observation.

	ß ± SE	Z	P	95% Confidence interval
Orientation	0.47 ± 0.23	2.00	<0.05	0.01 to 0.92
Species	-0.23 ± 0.26	-0.91	0.36	-0.73 to 0.27
Miniature black globe	-0.08 ± 0.02	-5.29	<0.01	-0.11 to -0.05
Wind speed	-0.33 ±0.15	-2.20	0.03	-0.62 to -0.04
Posture	-0.09 ± 0.25	-0.37	0.71	-0.59 to 0.41
Constant	3.80 ± 0.59	6.41	<0.01	2.64 to 4.96

Random factors

Animal ID estimated variance ± SE: <0.01 ± 0.56

Date of observation estimated variance ± SE: 0.32 ± 0.24

Observations =73, individuals =15

between left and right subcutaneous temperatures was greater when wildebeest were orientated perpendicular to solar radiation than when they were orientated parallel to solar radiation (Figure 3.3) and validates the hypothesis on which I had based the prediction model (Figure 3.11).

The absolute subcutaneous temperature difference was inversely associated with miniature black globe temperature (°C) on the collar of individual wildebeest (Table 3.6, $p<0.01$). Therefore, the higher the miniature black globe temperature on the collar, the smaller the subcutaneous temperature difference. To illustrate how miniature black globe temperature on the collar of a wildebeest could influence the subcutaneous temperature difference, I compared the absolute difference in subcutaneous temperature for wildebeest exposed to "warm" (<30 °C) and "hot" (>30 °C) miniature black globe temperatures (Figure 3.13). The subcutaneous temperature difference was significantly greater when wildebeest were exposed to "warm" miniature black globe temperatures than when they were exposed to "hot" miniature black globe temperatures (paired t-test, $t_7=3$, $p=0.01$). In one example, a blue wildebeest individual was orientated perpendicular to solar radiation early in the morning when the miniature black globe temperature was 17 °C (Figure 3.14a). Later that day, when miniature black globe temperature reached 30 °C, the same individual was again orientated perpendicular to solar radiation (Figure 3.14b) and the difference in subcutaneous temperature was smaller than when the individual was exposed to a lower miniature black globe temperature.

High wind speed ($m.s^{-1}$) was associated with smaller differences between left and right subcutaneous temperatures than when wind speed was low (Table 3.6, $p=0.03$). If the wind cooled down the subcutaneous temperature on the side exposed to solar radiation, it would

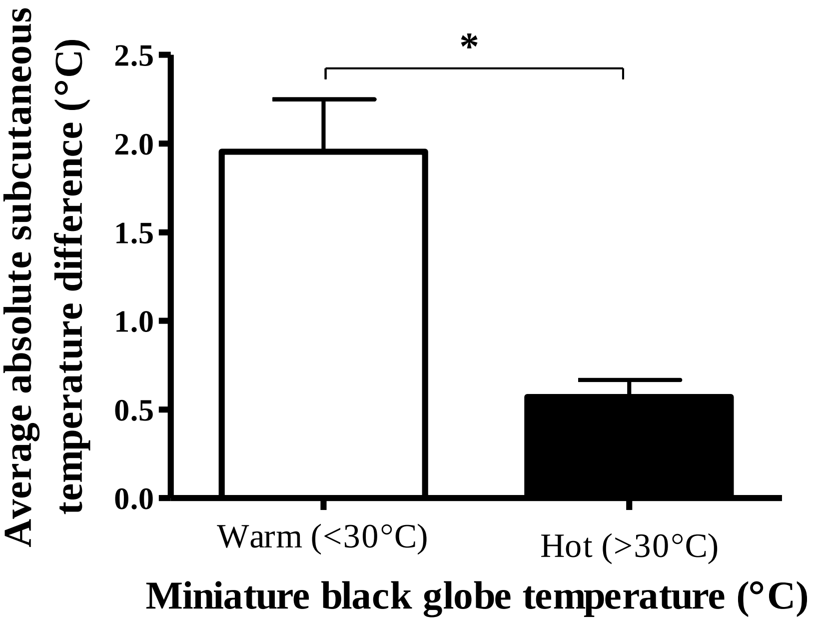


Figure 3.13 The average absolute difference between left and right subcutaneous temperatures of wildebeest exposed to miniature black globe temperatures below 30 °C (mean ± SD = 2.0 ± 1.1 °C) and miniature black globe temperatures above 30 °C (mean ± SD = 0.6 ± 0.3 °C). Observations= 73, individuals=15, * p<0.05.

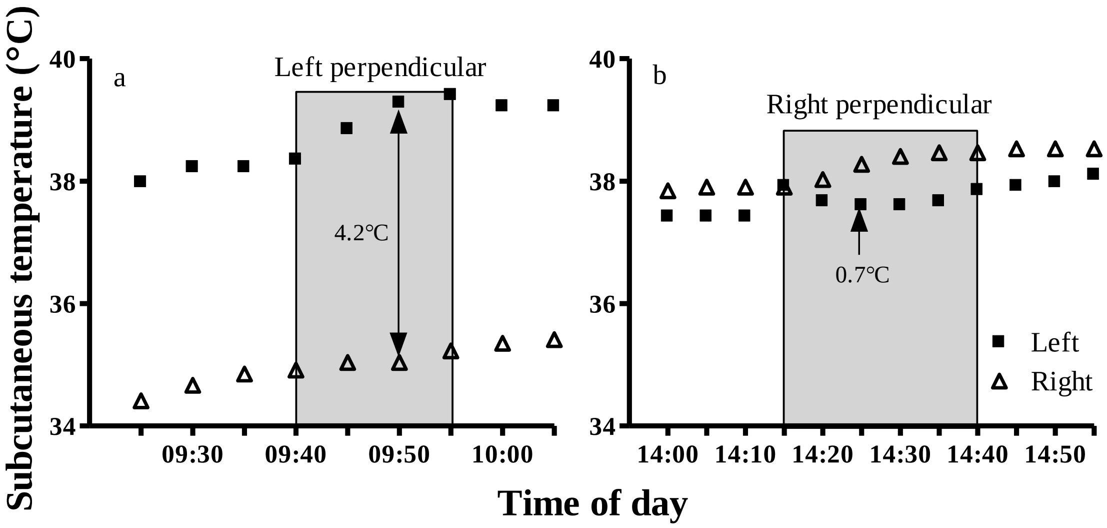


Figure 3.14 Examples of subcutaneous temperature profiles from the same blue wildebeest observed to be orientated perpendicular to solar radiation a) early in the morning when miniature black globe temperature from wildebeest collar was low (17 °C), and b) later that same day when miniature black globe temperature from wildebeest collar was high (30 °C). The maximum difference in subcutaneous temperature during the observation is indicated by the arrows. Wind speed was a) 1.3 m.s^{-1} and b) 0.7 m.s^{-1}.

reduce the subcutaneous temperature difference. For example, in Figure 3.15a the wind speed was low and the prediction model correctly predicted the orientation as right perpendicular to solar radiation. However, in Figure 3.15b, the wind speed was high and the prediction model incorrectly predicted the orientation as parallel to solar radiation. Both the sun and the wind came from the West. Even though the wildebeest was orientated left perpendicular to solar radiation, the left subcutaneous temperature did not increase, because it was being cooled down by the wind.

3.8.2 Factors that influenced the rate of change in subcutaneous temperature difference

Miniature black globe temperature (°C) had a significant effect on the absolute rate of change in subcutaneous temperature difference (Table 3.7, $p<0.01$). Orientation to solar radiation, species, wind speed ($m.s^{-1}$) and posture did not have any significant effect on the rate of change in subcutaneous temperature difference (Table 3.7, $p>0.05$).

Orientation to solar radiation did not have a significant effect on rate of change in subcutaneous temperature difference (Table 3.7, $p=0.36$), even though, previously, orientation to solar radiation had a significant effect on the subcutaneous temperature difference (Table 3.6, $p<0.05$). The lack of significance confirms the previous results found in the paired t-test done in section 3.4 (Figure 3.7), indicating that rate of change in subcutaneous temperature difference was not greater when wildebeest were orientated perpendicular to solar radiation than when they were orientated parallel to solar radiation. As shown earlier, rate of change in subcutaneous temperature difference did not always reflect orientation to solar radiation, especially if the wildebeest had been orientated in a specific position to solar radiation for a long time (Figure 3.9).

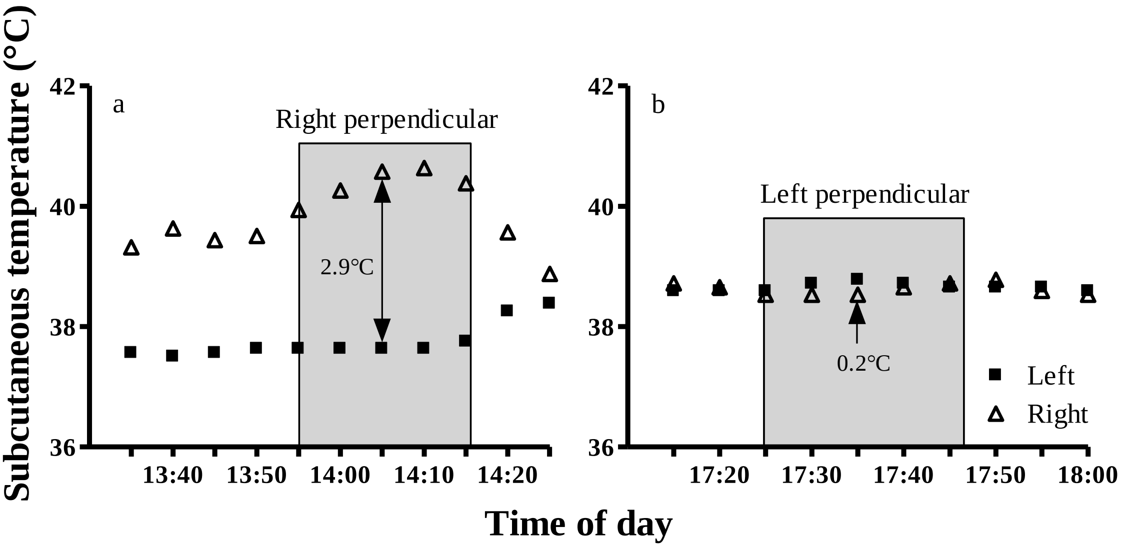


Figure 3.15 Two subcutaneous temperature profiles of two different wildebeest individuals observed orientated perpendicular to solar radiation. The first wildebeest (panel a) was exposed to low wind speed (1.5 m.s^{-1}). The second individual (panel b) was exposed to high wind speed (2.2 m.s^{-1}). In the example where the individual was exposed to high wind speed (b), the sun and the wind was coming from the same direction (West). Miniature black globe temperatures from wildebeest collars were a) 35 °C and b) 29 °C.

Table 3.7 Generalised Linear Mixed Model results to test which factors have a significant effect on the rate of change of the difference between left and right subcutaneous temperature. The table shows the coefficient and standard error (ß ± SE), z-value, p-value and 95 % confidence interval for orientation to solar radiation, species, miniature black globe temperature (°C) measured on the wildebeest collar, wind speed (m.s^{-1}) and posture. Random factors that were included were animal individual and date of observation.

	ß ± SE	Z	p	95% Confidence interval
Orientation	0.01 ± 0.02	0.92	0.36	-0.02 to 0.05
Species	-0.01 ± 0.02	-0.34	0.73	-0.04 to 0.03
Miniature black globe	>-0.01 ± <0.01	-3.70	<0.01	-0.01 to <0.01
Wind speed	-0.01 ± 0.01	-1.28	0.20	-0.03 to 0.01
Posture	-0.02 ± 0.02	-1.05	0.30	-0.05 to 0.02
Constant	0.19 ± 0.04	4.63	<0.01	0.11 to 0.26

Random factors

Animal individual estimated variation ± SE: <0.01 ± <0.01

Date of observation estimated variation ± SE: <0.01 ± <0.01

Observations =73, individuals =15

As was found with subcutaneous temperature difference, the rate of change in subcutaneous temperature difference did not vary between black and blue wildebeest (Table 3.7, p=0.73) or between wildebeest standing or lying down (Table 3.7, p=0.3). Wind speed (m.s^{-1}) did not affect rate of change in subcutaneous temperature difference significantly (Table 3.7, p=0.2) even though, previously, wind speed had a significant effect on subcutaneous temperature difference (Table 3.6, p=0.03).

High miniature black globe temperatures (°C) were associated with a smaller rate of change in subcutaneous temperature difference than low miniature black globe temperature. To illustrate how miniature black globe temperature affected rate of change in subcutaneous temperature difference, I compared the rate of change in subcutaneous temperature difference of wildebeest exposed to warm (<30 °C) and hot (>30 °C) miniature black globe temperatures (Figure 3.16). The rate of change in subcutaneous temperature difference was significantly greater for warm than for hot miniature black globe temperatures (paired t-test, t_7=3.6, p=0.009). In an example of a subcutaneous temperature profile of a wildebeest observed to be orientated perpendicular to solar radiation when miniature black globe temperature was 17 °C (Figure 3.17a), the rate of change in subcutaneous temperature difference was 0.15 °C.min^{-1} greater than the rate of change in subcutaneous temperature difference of a different wildebeest individual, which was observed to be orientated perpendicular to solar radiation when miniature black globe temperature was 33 °C (Figure 3.17b).

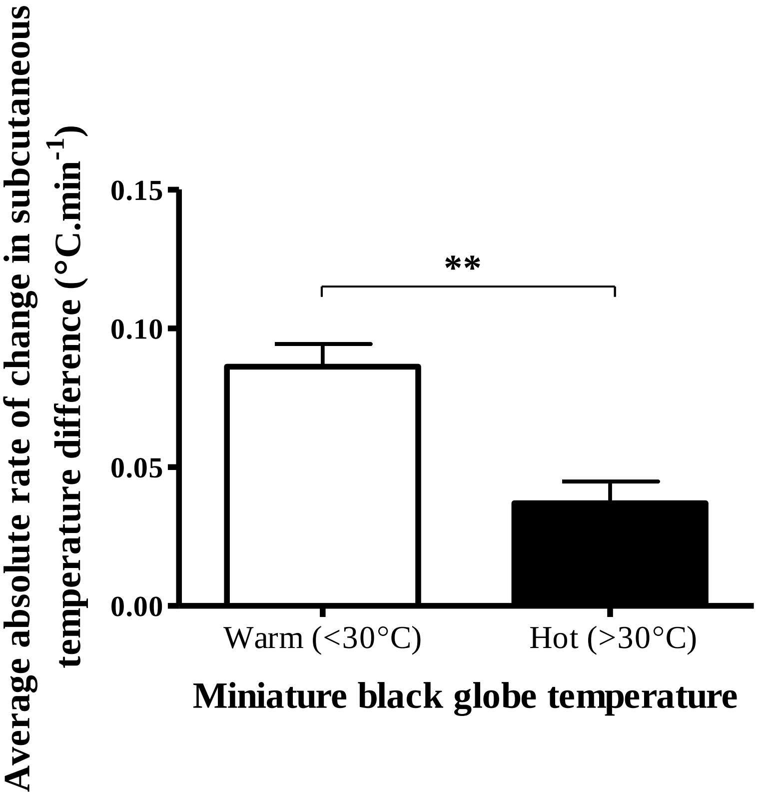


Figure 3.16 The average absolute rate of change in subcutaneous temperature difference in wildebeest exposed to miniature black globe temperatures below 30 °C (mean ± SD = 0.08 ± 0.03 °C.min^{-1}) and miniature black globe temperatures above 30 °C (mean ± SD = 0.04 ± 0.02 °C.min^{-1}). Observations= 73, individuals=15, ** p<0.01.

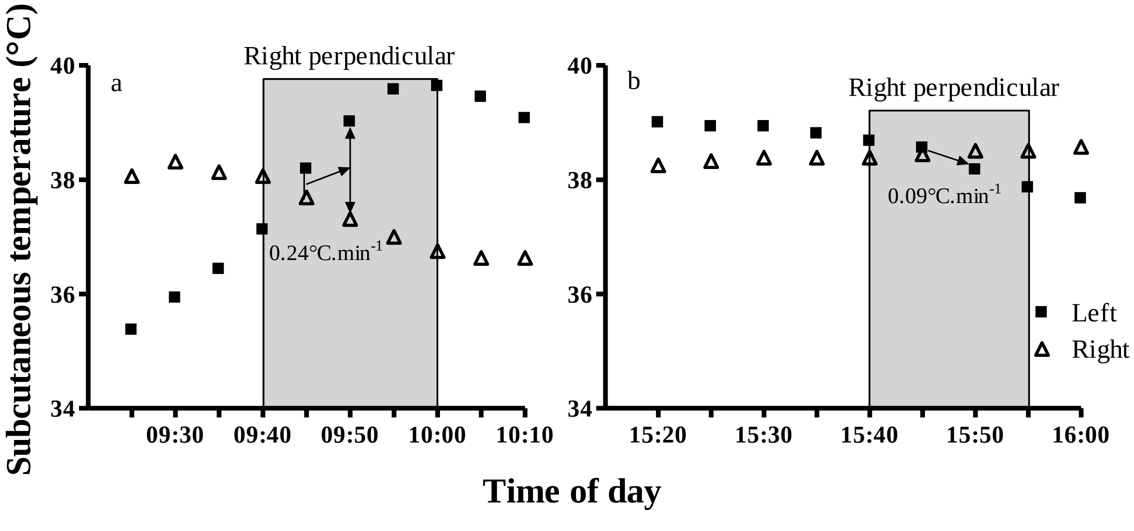


Figure 3.17 Two subcutaneous temperature profiles comparing the rate of change in subcutaneous temperature difference of two different wildebeest individuals observed orientating perpendicular to solar radiation exposed to a) low miniature black globe temperature (17 °C) and b) high miniature black globe temperature (33 °C). The rate of change in subcutaneous temperature difference is indicated by the arrows. Wind speeds recorded were a) 0.7 m.s^{-1} and b) 0.4 m.s^{-1}.

3.8.3 Factors that influenced the ability of the prediction model to accurately reflect orientation to solar radiation

Species (black or blue wildebeest) and posture (lying down or standing) had a significant ($p<0.05$) effect on whether the prediction model could accurately predict wildebeest orientation to solar radiation (Table 3.8), even though species and posture did not have a significant effect on the previous variables, subcutaneous temperature difference (Table 3.6) and rate of change in subcutaneous temperature difference (Table 3.7). Other factors that were found to have a significant effect on whether or not the prediction model could accurately predict wildebeest orientation to solar radiation were orientation to solar radiation and wind speed, (Table 3.8, $p<0.05$).

Miniature black globe temperature (°C) did not have a negative effect on the ability of the prediction model to accurately predict the orientation of a wildebeest to solar radiation (Table 3.8, $p=0.12$), even though the previous Generalised Linear Mixed Models showed that miniature black globe temperature had a significant effect on both subcutaneous temperature difference (Table 3.6, $p<0.01$) and rate of change in subcutaneous temperature difference (Table 3.7, $p<0.01$). However, in the earlier example, where high miniature black globe temperature resulted in a small subcutaneous temperature difference (Figure 3.14b), the prediction model could still correctly predict orientation to solar radiation because the rate of change in subcutaneous temperature difference was greater than the threshold value (0.06 $°C.min^{-1}$), despite the subcutaneous temperature difference being smaller than the threshold value. Therefore, even though high miniature black globe temperatures reduced the subcutaneous temperature difference, they did not negatively affect the ability of the prediction model to accurately predict the orientation of wildebeest to solar radiation.

Table 3.8 Generalised Linear Mixed Model to determine which factors affect how often a prediction is true or false. The table shows the coefficient and standard error (ß ± SE), z-value, p-value and 95 % confidence interval for orientation to solar radiation, species, miniature black globe temperature (°C) measured on the wildebeest collar, wind speed (m.s^{-1}) and posture. Random factors that were included were animal individual and date of observation.

Prediction	ß ± SE	Z	P	95% Confidence interval
Orientation	1.27 ± 0.61	2.09	0.04	0.08 to 2.47
Species	1.79 ± 0.73	2.47	0.01	0.37 to 3.22
Miniature black globe	0.06 ± 0.04	1.54	0.12	-0.02 to 0.14
Wind speed	-0.92 ± 0.42	-2.18	0.03	-1.75 to -0.09
Posture	-1.99 ± 0.72	-2.78	0.01	-3.39 to -0.59
Constant	-0.70 ± 1.48	-0.47	0.64	-3.61 to 2.20

Random factors

Animal individual estimated variance ± SD: <0.01 ± 0.48

Date of observation estimated variance ± SD: <0.01 ± 0.81

Observations =73, individuals =15

Orientation to solar radiation had a significant effect on the ability of the prediction model to accurately predict orientation to solar radiation (Table 3.8, p=0.04). The prediction model was more likely to correctly predict orientation to solar radiation if the wildebeest was orientated perpendicular to solar radiation than when it was orientated parallel to solar radiation, confirming previous results presented in Table 3.5 (56 % for left perpendicular, 81 % for right perpendicular and 49 % for parallel to solar radiation).

The way a wildebeest was orientated to solar radiation before it turned its body parallel to solar radiation could affect the subcutaneous temperature profile of a wildebeest. If the wildebeest was orientated perpendicular to solar radiation prior to orientating parallel to solar radiation, and the subcutaneous temperatures could not converge quickly enough, the difference in subcutaneous temperature would be greater than the threshold value and the wildebeest would be predicted as orientated perpendicular to solar radiation. For example, in Figure 3.18 the subcutaneous temperature difference was 1.8 °C when the wildebeest turned its body to be orientated parallel to solar radiation. The left and right subcutaneous temperatures took too long to converge. Ten minutes after the start of the observation, the subcutaneous temperature difference was 1.7 °C and the wildebeest was incorrectly predicted as right perpendicular to solar radiation. If the wildebeest was orientated perpendicular to solar radiation, then turned its body to orientate parallel to solar radiation and the left and right subcutaneous temperatures converged too quickly, the rate of change in subcutaneous temperature difference would be greater than the threshold value, and the prediction model would incorrectly predict the wildebeest as orientated perpendicular to solar radiation. For example, in Figure 3.19 the wildebeest was orientated right perpendicular to solar radiation and the right subcutaneous temperature difference was 3.1 °C greater than the left subcutaneous temperature when the wildebeest turned to

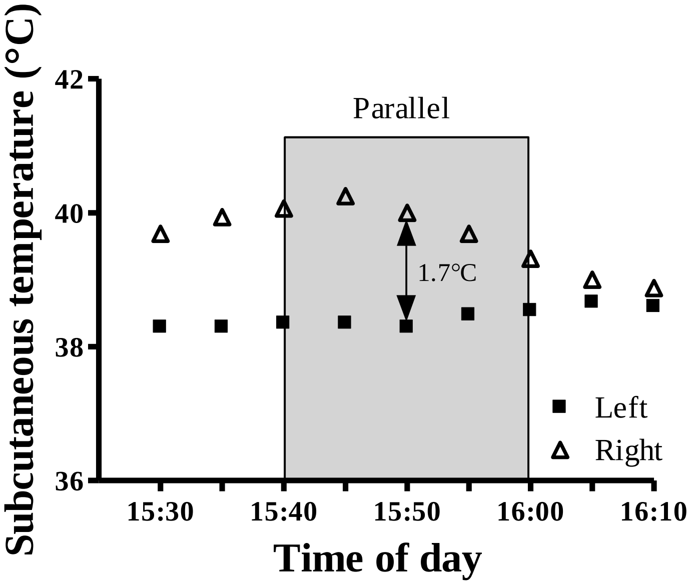


Figure 3.18 Subcutaneous temperature profile of a wildebeest incorrectly predicted to be orientated perpendicular to solar radiation while it was observed to be orientated parallel to solar radiation. The subcutaneous temperature difference was 1.8 °C prior to when the wildebeest turned parallel to solar radiation. Ten minutes after the start of the observation (indicated by the arrow), the subcutaneous temperature difference was still greater than the threshold value of 1 °C, leading to the incorrect prediction. Miniature black globe temperature from the wildebeest collar was 25 °C and wind speed was 0.9 m.s^{-1}.

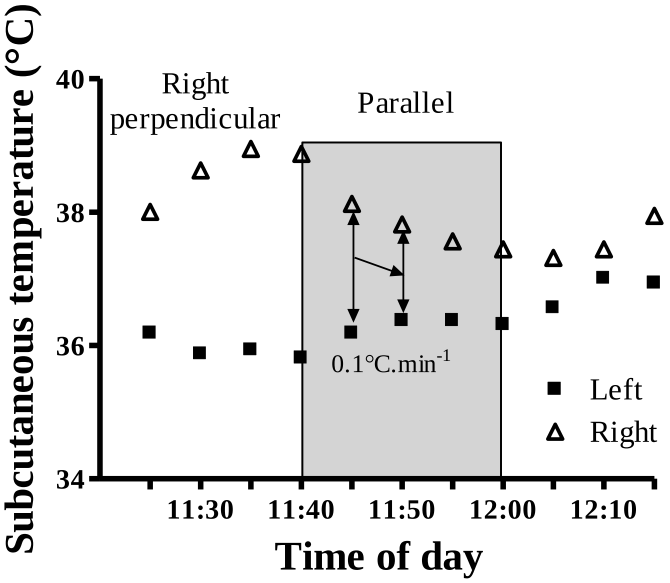


Figure 3.19 Subcutaneous temperature profile of a wildebeest observed to be orientated right side perpendicular to solar radiation before it turned its orientation parallel to solar radiation. When the subcutaneous temperatures converged, the rate of change in subcutaneous temperature difference ten minutes after the start of the observation (indicated by the arrows) was greater than the threshold value of 0.04 °C.min^{-1} and the wildebeest was incorrectly predicted to be orientated left side perpendicular to solar radiation. Miniature black globe temperature from the wildebeest collar was 20 °C and wind speed was 2.4 m.s^{-1}.

orientate parallel to solar radiation. After the wildebeest turned parallel to solar radiation, the right subcutaneous temperature decreased so quickly that the rate of change in subcutaneous temperature difference was 0.1 °C.min^{-1} and the wildebeest orientation to solar radiation was incorrectly predicted as left perpendicular to solar radiation instead of parallel to solar radiation.

After accounting for the influence of orientation to solar radiation, miniature black globe temperature, wind speed and body posture, species of wildebeest had a significant effect on the accuracy of the prediction model, with the model more likely to be correct for blue wildebeest than for black wildebeest (Table 3.8, p=0.01). When I divided the results of the prediction model according to species (Table 3.9), the prediction model was more often correct when predicting the orientation to solar radiation for blue wildebeest (77 %) than for black wildebeest (43 %), confirming the results of the Generalised Linear Mixed Model.

Wind speed (m.s^{-1}) had a negative effect on the ability of the prediction model to accurately predict a wildebeest's orientation to solar radiation (Table 3.8, p=0.03). Therefore, the greater the wind speed, the less likely it was that the prediction model would accurately reflect the orientation to solar radiation preferences of wildebeest. To illustrate how the wind speed affected the accuracy of the prediction model, I compared the average wind speed for each individual when the prediction of orientation to solar radiation was correct or incorrect (Figure 3.20). The average wind speed was greater when the prediction was incorrect than when the prediction was correct (paired t-test, t_9=4.3, p=0.002). As illustrated earlier, if the sun and the wind came from the same direction, high

Table 3.9 The percentage of times when the prediction model could correctly predict the orientation to solar radiation of black and blue wildebeest as parallel, left perpendicular or right perpendicular to solar radiation using the 1 °C threshold value for difference in subcutaneous temperature and the 0.04 °C.min^{-1} threshold value for rate of change in subcutaneous temperature difference.

Individual	Species	Number of observations	Parallel orientations correctly predicted (%)	Left perpendicular orientations correctly predicted (%)	Right perpendicular orientations correctly predicted (%)	Total correct predictions (%)
1	Black	2			100	100
2	Black	4	0	50	100	50
3	Black	6	25		100	50
4	Black	5	0		100	40
5	Black	1		0		0
6	Black	1			0	0
7	Black	12	43	0	100	50
8	Black	4	50		50	50
Average for black wildebeest			24	17	79	43
9	Blue	2		100	100	100
10	Blue	11	25	50	80	55
11	Blue	5		67	50	60
12	Blue	7	50	50	100	57
13	Blue	3		100	100	100
14	Blue	5	100	100	67	80
15	Blue	3	100	0		67
16	Blue	3	100	100		100
Average for blue wildebeest			75	71	83	77
Average for both species			49	56	81	60

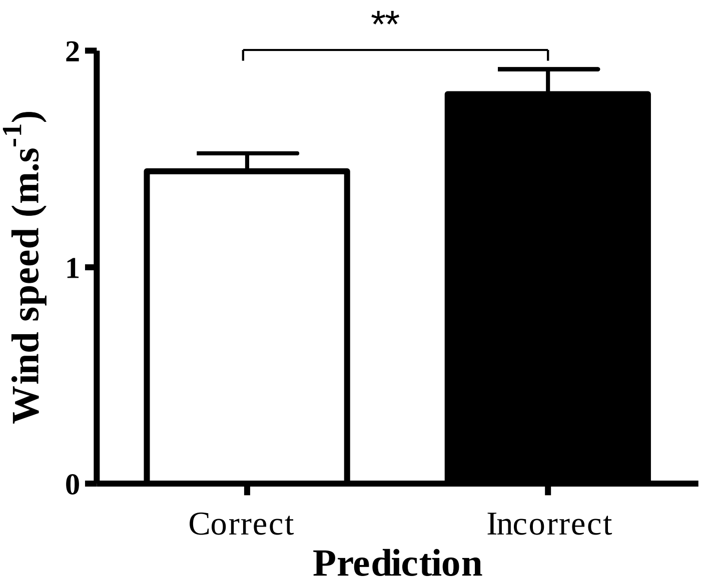


Figure 3.20 The average wind speed when the prediction model correctly reflected orientation to solar radiation (mean ± SD = 1.4 ± 0.3 m.s^{-1}) compared to when the prediction model was incorrect (mean ± SD = 1.8 ± 0.4 m.s^{-1}). Observations= 73, individuals=15, ** $p<0.01$.

wind speeds can cool down the side exposed to the sun (Figure 3.15) reducing the subcutaneous temperature difference and resulting in incorrect classification of orientation relative to solar radiation.

The body posture of a wildebeest (whether it was standing or lying down) had a significant effect on whether the prediction model could accurately reflect orientation to solar radiation of a wildebeest (Table 3.8, p=0.01). The prediction model was less likely to correctly predict orientation to solar radiation if a wildebeest was lying down than if it was standing. A paired t-test showed no difference in the percentage of wildebeest lying down when orientation to solar radiation was correctly or incorrectly predicted (paired t-test, t_9=1.6, p=0.14) (Figure 3.21). However, the lack of significance could be due to small sample size or because other factors such as orientation to solar radiation, species, miniature black globe temperature and wind speed were not taken into account, as they were in the Generalised Linear Mixed Model. In one example (Figure 3.22) a wildebeest was lying down parallel to solar radiation, but the right subcutaneous temperature was 1.1 °C greater than the left side leading to the incorrect prediction that the wildebeest was orientated left perpendicular to solar radiation. When the wildebeest stood up, still orientated parallel to solar radiation, the subcutaneous temperature difference decreased to 0.4 °C and the orientation was correctly predicted as parallel to solar radiation.

However, there were cases where the wildebeest was lying down and the prediction model correctly predicted the orientation to solar radiation. Wildebeest often maintained the same orientation to solar radiation for longer when they were lying down compared to when they were standing, which could increase the accuracy of the prediction model.

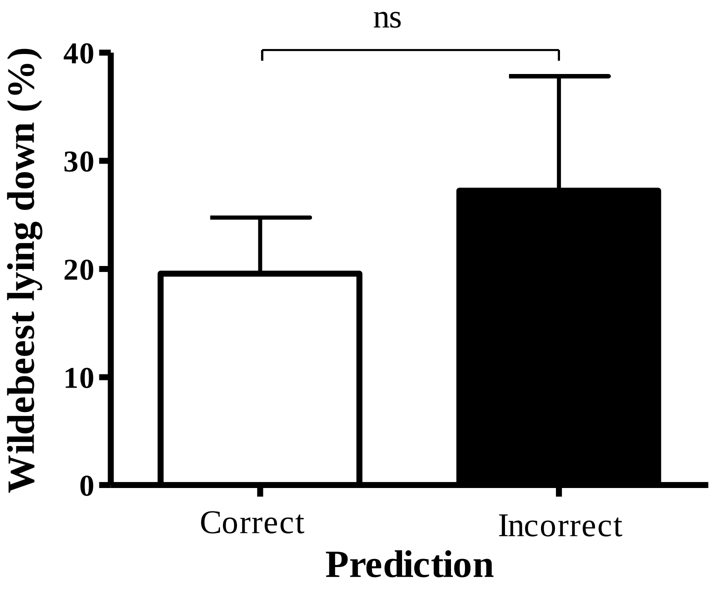


Figure 3.21 The average percentage of wildebeest lying down when the prediction model correctly reflected orientation to solar radiation (mean ± SD = 19.6 ± 19.4%) compared to when the prediction model was incorrect (mean ± SD = 27.3 ± 36.6%). Observations= 73, individuals=15, ns: p>0.05.

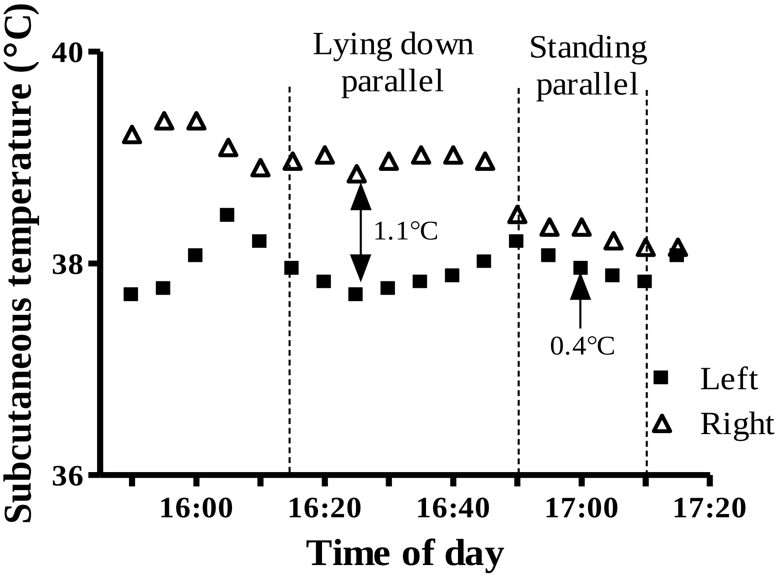


Figure 3.22 Subcutaneous temperature profile of a blue wildebeest, which was observed lying down with its body orientated parallel to solar radiation. Ten minutes after the start of the observation (indicated by the arrow), the subcutaneous temperature difference was greater than the threshold value (<-1 °C) and the model incorrectly classified the orientation of the wildebeest as perpendicular to solar radiation. After the wildebeest had been lying down for 35 minutes, the wildebeest stood up, still orientated parallel to solar radiation. Ten minutes later (indicated by the arrow), the subcutaneous temperature difference was below the threshold value of 1 °C and the wildebeest was correctly predicted to be orientated parallel to solar radiation. Miniature black globe temperature from wildebeest collar was 23 °C and the wind speed was 1.9 m.s^{-1}.

In one example (Figure 3.23) the wildebeest was lying down left perpendicular to solar radiation for 35 minutes. At the end of 35 minutes there was an almost 3 °C difference between left and right subcutaneous temperatures. Because the wildebeest was orientated left perpendicular to solar radiation for so long, there was a great subcutaneous temperature difference and the orientation to solar radiation was correctly predicted as left perpendicular to solar radiation.

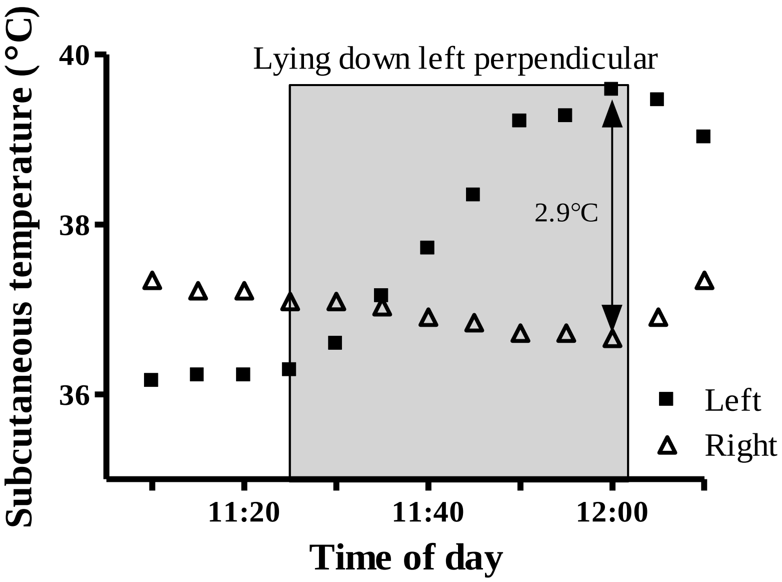


Figure 3.23 Subcutaneous temperature profile of a wildebeest, which was observed to lie down orientated left side perpendicular to solar radiation. After 35 minutes of lying down orientated left side perpendicular to solar radiation, the difference between the left and right subcutaneous temperature was greater than the threshold value of 1 °C and the orientation was correctly predicted as left side perpendicular to solar radiation. Miniature black globe temperature from wildebeest collar was 24 °C and wind speed was 2.0 m.s^{-1}.

.

Chapter 4

Discussion

4 Discussion

I show, for the first time, the potential of subcutaneous temperatures to be used in a technique to study orientation to solar radiation remotely in ungulates. Subcutaneous temperatures of wildebeest were highly variable during the course of a day (sunrise to sunset). The subcutaneous temperatures, measured by the implanted temperature-sensitive data loggers over the course of one year, ranged between 26.3 ºC and 41.9 ºC. The differences between left and right subcutaneous temperatures could be as great as 10.6 ºC (at a time when I was unable to visually confirm the orientation of the wildebeest). My hypothesis was that radiant heat may increase subcutaneous temperature on the side of the animal facing the sun, relative to the subcutaneous temperature on the opposite side of the animal's abdomen.

There was indeed a greater absolute subcutaneous temperature difference when wildebeest were observed to be orientated perpendicular to solar radiation than when they were observed to be orientated parallel to solar radiation (see Chapter 3, Results, Figure 3.3), confirming my hypothesis. The maximum subcutaneous temperature difference found when a wildebeest was observed to be orientated perpendicular to solar radiation was 4.9 ºC. However, although subcutaneous temperature difference tended to be greater when wildebeest were orientated perpendicular to solar radiation than when they were orientated parallel to solar radiation, the difference between subcutaneous temperatures accurately predicted orientation to solar radiation in only 48 % of observations.

The subcutaneous temperature difference appeared to be related not only to the current orientation, but also to the length of time a wildebeest had been orientated in a single direction. For example if a wildebeest had been orientated right perpendicular to solar radiation and then turned to orientate left perpendicular to solar radiation, the right side would have been hotter than the left side immediately after the wildebeest orientated left perpendicular to solar radiation, leading to incorrect predictions of the wildebeest's orientation to solar radiation (see Chapter 3, Results, Figure 3.5). I therefore investigated if rate of change in subcutaneous temperature difference might be a better predictor of orientation to solar radiation than would difference in subcutaneous temperature. However, because the length of time a wildebeest had been orientated in a particular position varied, rate of change in subcutaneous temperature difference accurately predicted only 57 % of observations. Therefore, using rate of change in subcutaneous temperature to predict orientation to solar radiation was more often correct than using subcutaneous temperature difference, but not sufficiently so for the technique to be used successfully.

To improve the accuracy of the technique to predict orientation to solar radiation, I developed a prediction model that incorporated both the subcutaneous temperature difference and rate of change in subcutaneous temperature difference (see Chapter 3, Results, Figure 3.11). Although the prediction model improved the accuracy of the technique to remotely predict orientation to solar radiation (60 %), more than a third of observations were still incorrectly classified. Therefore, I investigated which factors affected the accuracy of the prediction model.

The subcutaneous temperature on the left and right side of a wildebeest can be influenced by many environmental and biological factors. Any factor that influences the subcutaneous temperature independently of heat load from solar radiation can have an effect on the accuracy of the prediction model. The factors that were found to affect the accuracy of the prediction model included orientation to solar radiation, species, wind speed and posture. The remote technique worked best for blue wildebeest that were standing rather than lying down, and when wind speed and miniature black globe temperature were low.

I may have been able to improve the accuracy of the prediction model had I been able to observe the animals continuously. Since continuous observation was not feasible in free-living wildebeest, I often did not know how long a wildebeest had been orientated in a particular direction or what its previous orientation to solar radiation was. Instead I focussed my analyses on periods when a wildebeest had been observed in a single orientation for a minimum of 10 minutes, which resulted in a small subset of observations. From 470 hours of observations, I focussed on 74 observations in 16 individuals, following the exclusion of cloudy days, periods when wildebeest sought shade and times when the data loggers failed (see Chapter 2, Materials and Methods, section 2.8.1). My small sample size was a limiting factor when doing statistical analysis.

4.1 Limitations of prediction model

I assessed the remote technique to detect orientation to solar radiation by focussing on observations when wildebeest were orientated either parallel or perpendicular to solar radiation. However, there were many instances where I observed wildebeest orientated in between these two positions (oblique to solar radiation). Because of the design of the prediction model, a wildebeest that was orientated oblique to solar radiation would incorrectly be predicted to be orientated either parallel or perpendicular to solar radiation. I would expect that, in cold, sunny conditions, oblique orientation would reduce the need for metabolic heat production more than when a wildebeest was orientated parallel to solar radiation, but less than when a wildebeest was orientated perpendicular to solar radiation. Therefore, when studying thermoregulatory behaviour of wildebeest, it is also important to determine when a wildebeest is orientated oblique to solar radiation, so that the wildebeest is not incorrectly classified as orientated perpendicular or parallel to solar radiation in these times.

Theoretically, if a wildebeest was orientated oblique to solar radiation, the subcutaneous temperature difference would be greater than when the wildebeest was orientated parallel to solar radiation, but smaller than when the wildebeest was orientated perpendicular to solar radiation. For example, if the subcutaneous temperature difference was always greater than 4 °C if a wildebeest was orientated perpendicular to solar radiation and always smaller than 2 °C if a wildebeest was orientated parallel to solar radiation, and the subcutaneous temperature always stabilised within ten minutes of a wildebeest changing position, it would be possible to classify oblique orientation as the times when the subcutaneous temperature difference was between 2 °C and 4 °C. However, the subcutaneous temperature differences when wildebeest were orientated parallel compared to perpendicular to solar radiation were not as distinct as in this hypothetical example. Instead, there was substantial variance in subcutaneous temperature difference of wildebeest orientated parallel and perpendicular to solar radiation; hence, there were many instances where parallel and perpendicular orientation to solar radiation were incorrectly predicted.

The prediction model could more accurately predict the orientation to solar radiation of wildebeest orientated perpendicular to solar radiation than of wildebeest orientated parallel to solar radiation. Therefore, there were many instances when wildebeest were orientated parallel to solar radiation, but incorrectly predicted to be orientated perpendicular to solar radiation. If a wildebeest was orientated perpendicular to solar radiation, and either subcutaneous temperature difference or rate of change in subcutaneous temperature difference was above the threshold values, the orientation to solar radiation would be correctly predicted.

However, for a wildebeest to be correctly identified as orientated parallel to solar radiation, both subcutaneous temperature difference and rate of change in subcutaneous temperature difference had to be below the threshold value. If either one of these variables was greater than the threshold value the wildebeest would be incorrectly predicted as perpendicular to solar radiation. Environmental factors could have influenced subcutaneous temperatures, however, the Generalised Linear Mixed Model accounted for the effect of wind speed and miniature black globe temperature and still found a significant relationship between orientation to solar radiation and the accuracy of the prediction model.

Another explanation for the incorrect prediction is that the previous orientation to solar radiation of a wildebeest could have influenced the subcutaneous temperatures of wildebeest orientated parallel to solar radiation. I attempted to account for the effect of the previous orientation of a wildebeest by using rate of change in subcutaneous temperature difference to predict the orientation to solar radiation. However, using rate of change in subcutaneous temperature difference only accounted for the previous orientation if the wildebeest was orientated perpendicular to solar radiation. For example, if a wildebeest was orientated right perpendicular to solar radiation and then turned left perpendicular to solar radiation, the rate of change in subcutaneous temperature difference would have been greater than the threshold value and the orientation would correctly be predicted as left perpendicular to solar radiation (see Chapter 3, Results, Figure 3.5). However, if the wildebeest was orientated right perpendicular to solar radiation and then turned parallel to solar radiation, the rate of change in subcutaneous temperature differences was sometimes greater than the threshold value and the orientation was incorrectly predicted as left perpendicular to solar radiation (see Chapter 3, Results, Figure 3.19). Therefore, using rate of change in subcutaneous temperature difference did not sufficiently account for the previous orientation of a wildebeest if the wildebeest changed from being orientated perpendicular to solar radiation to being orientated parallel to solar radiation. My remote technique could possibly be improved by incorporating an appropriate time lag to allow subcutaneous temperatures to stabilise after a wildebeest changed its orientation from perpendicular to parallel to solar radiation.

Orientating parallel to solar radiation reduces the radiant heat load on wildebeest. Similarly, animals that seek shade reduce their radiant heat load. In both cases, both sides of the animal are exposed to the same amount of radiant heat. Therefore, it is understandable that the subcutaneous temperature profile of a wildebeest standing in the shade (Figure 4.1a) was similar to that of a wildebeest orientated parallel to solar radiation (Figure 4.1b). In both cases, the subcutaneous temperature difference was less than 1 °C. Therefore, I could not use subcutaneous temperature difference to distinguish between a wildebeest orientated parallel to solar radiation and a wildebeest in the shade. There is probably no thermal advantage for an animal to shift its body orientation relative to solar radiation if the animal is standing in the shade or if there is cloud cover, since orientation to solar radiation during these times is often random (Maloney et al., 2005b, Hetem et al., 2011b). When studying behavioural thermoregulation in animals, it is important to determine which behavioural responses animals are most likely to use in response to changes in environmental heat loads. Therefore, it is important to be able to distinguish between parallel orientation to solar radiation and shade-seeking behaviour. In this study, shade-seeking behaviour and cloud cover over the study animals was determined visually, but to study orientation to solar radiation remotely shade cover would need to be determined remotely. A weather station set up in close proximity to the study animals would be able to determine cloud cover by measuring radiation. Shade-seeking behaviour in animals can be quantified by attaching a miniature black globe thermometer to a wildebeest collar (Hetem et al., 2007). If the miniature black

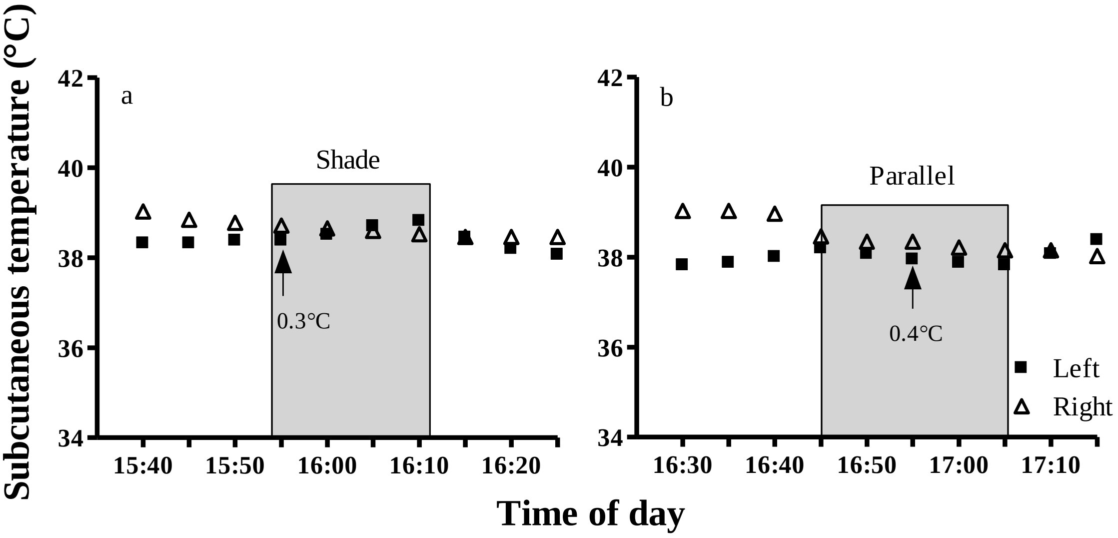


Figure 4.1 Subcutaneous temperature profiles illustrating a wildebeest standing a) in the shade and b) parallel to the sun. In both cases, the maximum difference between left and right subcutaneous temperatures (indicated by the arrows) was less than 1 °C.

globe temperature on the wildebeest collar is lower than the miniature black globe temperature measured in the sun, the wildebeest is likely seeking shade (Hetem et al., 2007).

4.2 Factors that influenced subcutaneous temperature and the accuracy of the prediction model

Subcutaneous temperatures were measured below the skin of the animal. The coat and skin of an animal is the interface where heat transfer takes place between the animal and its environment. Therefore, subcutaneous temperature can respond to changes in environmental conditions quicker that would core body temperature. Core body temperature of wildebeest varies less than does subcutaneous temperature. For example, in a study conducted on black wildebeest in Kimberley (the same area where my study was conducted), blood and brain temperatures of black wildebeest showed little variation (Jessen et al., 1994) even when exposed to intense radiation and air temperatures close to 40 °C with no shelter available. The wildebeest in this study were exposed to similar environmental conditions during the summer months. The subcutaneous temperatures of wildebeest measured during this study showed fluctuations between the average daily minimum and average daily maximum of 5 °C. The observed subcutaneous temperature fluctuations are probably due to environmental factors such as radiant heat and wind speed, and active vasodilation and vasoconstriction of peripheral blood vessels by the animal.

4.2.1 Miniature black globe temperature

Radiant heat absorbed by the coat and skin surface of the wildebeest will increase subcutaneous temperature, and wind blowing over the fur at air temperatures lower than skin temperature will decrease subcutaneous temperatures. Black globe temperature incorporates radiation, air temperature and wind speed and has been designed to study thermal comfort in humans (Vernon, 1933). In studies of wildlife physiology, smaller versions of the black globe thermometer have been attached to animal collars to measure the environmental heat load an individual animal experienced in its microhabitat (Fuller et al., 1999, Hetem et al., 2007, Hetem et al., 2008, Hetem et al., 2012a).

I found that subcutaneous temperature differences tended to be greater at lower miniature black globe temperatures than at higher miniature black globe temperatures. The small subcutaneous temperature difference during hot periods could have resulted because wildebeest preferred to reduce their body surface area exposed to solar radiation when the miniature black globe temperature was high by orientating parallel to solar radiation. Previous studies have shown that wildebeest prefer orientating parallel to solar radiation in the middle of the day and perpendicular to solar radiation early in the morning and late afternoon (Berry et al., 1984, Vrahimis and Kok, 1992, Maloney et al., 2005b, Hetem et al., 2011b). Therefore, the higher the miniature black globe temperature, the more likely the wildebeest was to be orientated parallel to solar radiation as reflected by a small subcutaneous temperature difference. However, the Generalised Linear Mixed Model found that miniature black globe temperature had a significant effect on subcutaneous temperature difference, even when the effect of all other variables, including orientation to solar radiation had been taken into account. Therefore, miniature black globe temperature had a significant effect on subcutaneous temperature difference independent of orientation to solar radiation. For example, when wildebeest were orientated perpendicular to solar radiation when miniature black globe temperature was hot, the subcutaneous temperature difference was smaller than when the wildebeest was orientated perpendicular to solar radiation when the miniature black globe temperature was cooler (see Chapter 3, Results, Figure 3.14). Therefore, orientation to solar radiation preferences cannot account for the greater subcutaneous temperature difference when miniature black globe temperatures were low.

A possible explanation for the smaller subcutaneous temperature differences when miniature black globe temperatures were high could be that wildebeest peripheral blood vessels were vasodilated when miniature black globe temperatures were high. Animals vasodilate their peripheral blood vessels in hot conditions to increase heat loss to the environment (Mount, 1979). Vasodilation increases blood flow close to the skin surface, to facilitate heat loss to the environment. Increased blood flow close to the skin surface could lead to an increase in subcutaneous temperature. If vasodilation occurred on both sides of the wildebeest, as expected, both sides of the wildebeest would increase in temperature and not just on the side facing perpendicular to the sun. Therefore vasodilation would reduce the influence of radiant heat on the subcutaneous temperature difference when miniature black globe temperatures were high.

In cool conditions, animals vasoconstrict their peripheral blood vessels to reduce heat loss to the environment (Mount, 1979). A decrease in peripheral blood flow would cause a decrease in subcutaneous temperature. The side facing perpendicular to solar radiation would be able to absorb radiant heat quicker than if it was vasodilated because of the increased temperature gradient between the skin and fur surface of the animal. The side facing away from the sun would be cold due to the decreased blood flow close to the skin surface. Therefore, on a cool but sunny morning, the orientation to solar radiation would have a greater effect on the difference between the left and right subcutaneous temperatures than when miniature black globe temperatures were high.

Alternatively, the greater difference in subcutaneous temperature when miniature black globe temperatures are cool could be caused by the angle of the sun's rays early in the morning compared to late afternoon. During the early morning and late afternoon, when miniature black globe temperatures were cooler, the sun's rays were more horizontal and shone more directly onto the flank of the animal facing perpendicular to the sun, compared to at midday, usually the hottest time of day, when the sun shone more directly from above. Therefore, I expect that, early in the mornings, when the angle of incident solar radiation was more horizontal, perpendicular orientation to solar radiation would cause a greater subcutaneous temperature difference than at midday, when the angle of solar radiation was closer to 90 °.

Even though high miniature black globe temperature was associated with a reduced subcutaneous temperature difference and rate of change in subcutaneous temperature difference when wildebeest were orientated perpendicular to solar radiation, high miniature black globe temperature did not have a negative effect on the accuracy of the technique. If the wildebeest was orientated perpendicular to solar radiation when miniature black globe temperatures were high, the high miniature black globe temperature could cause the difference between left and right subcutaneous temperatures to be less than the threshold value, leading to the incorrect prediction that the wildebeest was orientated parallel to solar radiation. However, there were examples where the wildebeest was orientated perpendicular to solar radiation when miniature black globe temperatures were high and, even though the subcutaneous temperature difference and rate of change in subcutaneous temperature difference were lower than when miniature black globe temperatures were low, either subcutaneous temperature difference or rate of change in subcutaneous temperature difference was still above the threshold value to be correctly predicted as orientated perpendicular to solar radiation (See Chapter 3, Results, Figure 3.14b). Reduced subcutaneous temperature difference and rate of change in subcutaneous temperature difference potentially reduced the ability of the prediction model to correctly predict orientation to solar radiation only if the wildebeest was orientated perpendicular to solar radiation. If the wildebeest was orientated parallel to solar radiation and high miniature black globe temperature caused a reduction in subcutaneous temperature difference, the subcutaneous temperature difference would still be below the threshold value and the wildebeest would still be correctly predicted as orientated parallel to solar radiation. Therefore, even though miniature black globe temperature had a significant effect on the subcutaneous temperature of wildebeest, the effect was not great enough to reduce the accuracy of the prediction model.

4.2.2 Wind speed

One component contributing to miniature black globe temperature is wind speed. At high wind speeds the prediction model was less accurate in reflecting the orientation of wildebeest relative to solar radiation than when the wind speed was low. High wind speed has a negative effect on the insulative properties of the coat (Dawson and Brown, 1970, Marder, 1973, Campbell et al., 1980, Walsberg, 1990). Wind displaces still air that is trapped within the coat between hairs, decreasing the boundary layer where convection takes place between the body of the animal and the surrounding air (Mount, 1979). If the coat and skin of the wildebeest happened to be wet from water or sweat, the cooling effect of wind on subcutaneous temperatures would have been increased (Gebremedhin and Wu, 2001).

Because wind can have such a profound effect on subcutaneous temperature, it could override the effect of orientation to solar radiation on the subcutaneous temperature. If the wildebeest was orientated parallel to the wind, wind would have blown over both sides of the body. Reduced subcutaneous temperatures on both sides of the wildebeest could lead to the incorrect prediction that the wildebeest was orientated parallel to solar radiation, even though it might be orientated perpendicular to solar radiation. If the wildebeest orientated its body perpendicular to the wind, subcutaneous temperature would be cooled down only on one side of the body. If the side facing perpendicular to the wind was facing perpendicular to the sun the cooling effect of the wind would mask the effect of the solar radiation on the subcutaneous temperature on that side, leading to incorrect predictions of the wildebeest's orientation to solar radiation (see Chapter 3, Results, Figure 3.15b).

Wind speed not only affects the subcutaneous temperature of wildebeest, but also their behaviour. The higher the wind speed, the more likely a wildebeest is to orientate according to the wind and not according to the sun (Berry et al., 1984, Vrahimis and Kok, 1992, Maloney et al., 2005b). In hot conditions, a wildebeest might use strong winds to cool down by orientating its body perpendicular to the wind (Berry et al., 1984). In cool weather, wildebeest can limit the cooling effect of strong winds by orientating their bodies parallel to the direction from which the wind is coming (Gebremedhin, 1987, Vrahimis and Kok, 1992), by seeking cover behind vegetation, or by lying down (Hetem et al., 2011b).

4.2.3 The effect of body posture on subcutaneous temperature

Animals can change their body posture to manipulate the amount of wind and radiant heat they are exposed to. Various animal species are known to change their body posture as a behavioural response to environmental temperatures (Hutchinson et al., 1975, Harri and Korhonen, 1988, Dasilva, 1993). If an animal lies down, it affects the surface area of the animal's body that is exposed to the sun (Walsberg, 1992). When ambient temperatures are high, black wildebeest prefer to lie down during the heat of the day to reduce their heat uptake (Vrahimis and Kok, 1993, Maloney et al., 2005a). However, in one example on blue wildebeest (Hetem et al., 2011b), the hottest time of the day coincided with the time of day when wind speed was highest. Therefore it is not certain whether the wildebeest were lying down to avoid the wind or to avoid radiant heat. Wildebeest can also lie down to reduce the surface area where heat loss takes place to the environment. During the cold season, black wildebeest preferred to lie down when it was cold, especially at night (Maloney et al., 2005a).

Wildebeest may lie down for reasons other than thermoregulation. Animals can lie down for rumination and digestive purposes or to save energy (Jarman and Jarman, 1973, Mitchell, 1977, Berry et al., 1982). Therefore, in many cases it is difficult to determine why animals are lying down and whether lying down has any thermoregulatory advantages.

In this study I found that when wildebeest were lying down, the prediction model was less likely to correctly predict the orientation of a wildebeest relative to solar radiation. How the wildebeest lies down would affect which parts of the wildebeest’s body were shaded as well as the amount of radiation, reflected from the ground and nearby objects, that was absorbed by the wildebeest. Wildebeest seldom lay down perfectly sternal. They usually lay down leaning to one side. The amount of surface area exposed to solar radiation while a wildebeest is lying down will have an effect on its subcutaneous temperature. The subcutaneous temperature on the side the wildebeest is lying on will be affected by conductive heat transfer with the ground. Depending on the temperature of the ground, the subcutaneous temperature would either increase or decrease on the side lying against the ground. Therefore the patterns of left and right subcutaneous temperatures of a wildebeest will be determined by how the wildebeest is lying down and not only by the orientation of the wildebeest relative to solar radiation, and could lead to incorrect predictions of orientation to solar radiation using the prediction model.

4.2.4 The effect of evaporative cooling on subcutaneous temperature

Subcutaneous temperature is determined, not only by environmental factors, but also by biological factors. If the animal was exposed to high environmental temperatures, or high amounts of radiation, thermoregulatory responses of endotherms such as sweating would decrease the subcutaneous temperature. Blue wildebeest can sweat at high ambient temperatures, but rely more on panting than sweating for evaporative heat loss (Taylor et al., 1969). Although there are intra-individual differences, the amount of sweat produced by a blue wildebeest can be less than half that of other antelope such as eland (*Taurotragus oryx*) and buffalo (*Syncerus caffer*; Robertshaw and Taylor, 1969). The preference of panting over sweating is probably an adaptation to survive in arid conditions, since panting caused a lower amount of evaporative water loss in black beduoin goats (*Capra hircus*) compared to when they were sweating (Robertshaw and Dmi'el, 1983). During times of dehydration, black beduoin goats increased panting and decreased the amount of sweating compared to when they were hydrated to reduce evaporative water loss (Robertshaw and Dmi'el, 1983).

I am not aware of any literature on sweating in black wildebeest. It is likely that they have active sweat glands, because other alcelaphines like red hartebeest (*Alcelaphus buselaphus cokii*; Finch, 1972) and blue wildebeest (Taylor et al., 1969) do have active sweat glands. However, the phylogenetic relationship of antelope had no influence on their sweat gland activity (Robertshaw and Taylor, 1969), since various sweating patterns were found amongst antelope within the same subfamilies and even within individuals of the same species (Robertshaw and Taylor, 1969). Therefore, we cannot assume that the sweating patterns of black wildebeest would be similar to that of the blue wildebeest. I do not know whether sweating affected the subcutaneous temperatures of the wildebeest in our study. I would expect that sweat would be produced evenly on both sides on the wildebeest. Therefore sweating should not affect the subcutaneous temperature difference or the rate of change in subcutaneous temperature difference.

Evaporative heat loss is known to be an effective way of losing heat to the environment (Taylor et al., 1969), however it can be costly in terms of the water balance of the animal. When dehydrated, animals will decrease sweating and panting to reduce water lost to the environment (Taylor, 1970). By using mud or water to wet the skin, animals can cool their bodies using evaporative heat loss, without the risk of dehydration associated with sweating and panting. Mud and water bathing are known to be used by various animals such as elephants, rhino, suids and buffalo (Estes, 1997, Skinner and Chimimba, 2005, Bracke, 2011). However, the purpose of these behaviours is sometimes unclear (Emmons et al., 2004). Bathing has many other advantages, such as removal of parasites, social bonding, sunburn protection, and forms part of sickness behaviour and sexual behaviour (Bracke, 2011). Measuring the effect of bathing on subcutaneous temperature can help to determine whether these activities are used for thermoregulatory purposes or not. If these behaviours have no effect on the subcutaneous temperature, it is more likely that bathing has other purposes such as skin care, hygiene or social cohesion, instead of thermoregulatory purpose.

We observed mud bathing in the collared blue wildebeest as well as in untagged blue wildebeest individuals in the park. One (artificial) water hole, which the blue wildebeest often frequented, was a landscape depression filled with water. The water hole often had lots of mud due to animals trampling the ground when they came to drink water. The blue wildebeest were often observed to mud bathe at this water hole. When blue wildebeest were observed grazing in the area close to the water hole, they sometimes had a layer of mud somewhere on their bodies (the mud on their coats was especially obvious because of the red colour of the earth), suggesting that they had been mud bathing earlier. Unfortunately we did not have enough observations on blue wildebeest mud bathing to quantify its effect on subcutaneous temperature in this study.

We did not observe mud bathing in black wildebeest, but we never observed them close to muddy places. The black wildebeest were often observed from further distances away than were the blue wildebeest, so mud on the coats of the black wildebeest would not necessarily be as visible as was the mud on the blue wildebeest coats. Therefore, it is possible that black wildebeest employ mud bathing just like blue wildebeest do, although we didn’t witness it in this study. A layer of mud on the coat of a wildebeest would affect the thermal properties of the coat. Wet mud would facilitate heat loss by increasing evaporative heat loss, and dry mud could act as an extra insulation layer. Therefore, mud would likely affect the subcutaneous temperatures and could lead to inaccurate predictions of wildebeest orientation to solar radiation using the prediction model, especially if mud was located only on one side.

In this study, we could not quantify the effect of evaporative heat loss on the prediction model. Further research into the sweat gland activity of black wildebeest and into the effect of mud bathing on subcutaneous temperature would be necessary to determine the effect of evaporative cooling on the prediction model. It is also uncertain how differences in sweat excretion and mud bathing behaviour between the two species could affect the accuracy of the prediction model.

4.2.5 Differences between black and blue wildebeest that could affect subcutaneous temperature

The prediction model was more accurate in reflecting the orientation to solar radiation of blue wildebeest than that of black wildebeest. The two species lived on the same reserve and were therefore exposed to similar climatic conditions. It is possible that biological differences between black and blue wildebeest could be the reason the remote technique could more accurately reflect the orientation behaviour of blue wildebeest than black wildebeest. There are biological differences between black and blue wildebeest that could affect their thermoregulation. For example, black wildebeest have a smaller body size and thicker fur than do blue wildebeest.

The black wildebeest is about 30 % smaller than the blue wildebeest. Size is known to have a correlation with basal metabolic rate and body temperature (White and Seymour, 2003, Clarke and Rothery, 2008). Smaller antelope cope better with heat stress than do larger antelope. For example, it has been found that, during spring, the activity patterns of impala (*Aepyceros melampus, 50 kg*) was less influenced by increased ambient temperatures than that of blue wildebeest (180 kg) and eland (420 kg; Shrestha et al., 2013). In a study comparing two species of desert-adapted antelope in Saudi Arabia, the Arabian oryx (*Oryx leucoryx*, 70 kg) had a lower mean body temperature than did the Arabian sand gazelle (*Gazella subgutturosa marica*, 15 kg; Hetem et al., 2012b).

Body size is correlated, not only with core temperature, but also with surface temperature. Surface temperature has a positive correlation with body size across species within a wide range of body weight (0.02 kg – 4 000 kg; Phillips and Heath, 1995). Because the blue wildebeest has a greater body size than the black wildebeest, we might expect the blue wildebeest to have a greater ability to regulate its surface temperature than the black wildebeest. According to the equation VMI = 0.27717 + 0.27929 log(weight) (Phillips and Heath, 1995) the vasomotor index (VMI) of black wildebeest would be 0.87 compared to the vasomotor index of blue wildebeest 0.91. The vasomotor index is an indication of the ability of an animal to control their surface temperature. The vasomotor index of the blue wildebeest was only 0.04 greater than that of black wildebeest. The size difference between black and blue wildebeest was probably too small to influence their subcutaneous temperatures.

Since difference in body mass was unlikely to account for the difference in our prediction model detecting orientation to solar radiation behaviour, I investigated other morphological characteristics that differed between the two wildebeest species. The black wildebeest are often exposed to high amounts of solar radiation with no shelter available (Estes, 1966), therefore we can expect to find that black wildebeest have developed adaptations to protect themselves against solar radiation. One way in which an animal can reduce the amount of solar radiation absorbed from its environment when standing out in the open is by increasing their insulation. The black wildebeest’s coat is darker and thicker than that of the blue wildebeest (Estes, 1997). The amount of insulation a wildebeest’s coat provides would affect the accuracy of the prediction model to detect orientation to solar radiation behaviour of wildebeest.

It is generally assumed that a light coloured coat is more advantageous in hot conditions than a dark coat, because the dark colour absorbs more radiant heat from the sun than light coloured coats (Finch et al., 1980, Hetem et al., 2009). However, the amount of radiant heat absorbed by the dark coat is not necessarily the same as the amount of radiant heat that gets absorbed by the animal’s body. For example, in a study conducted on emu’s and kangaroos living in the Australian desert, the emu's feathers, although darker in colour, provided better protection from solar radiation than the light fur of kangaroos (Dawson and Maloney, 2004). Even though the plumage of an emu has a high absorptivity because of its dark colour, most of the radiant heat gets absorbed by the outer layer of the plumage and dissipated into the environment, causing a low penetrance of solar radiation to the skin of the animal (Maloney and Dawson, 1995). Another study has shown that the dark coloured feathers of ravens in Israel provide protection against solar radiation, because the heat gets absorbed by the tip of the feather and dissipated into the air, without getting absorbed by the skin of the animal (Marder, 1973). A study in squirrels has shown that a thick, dark coloured coat provides just as much insulation from solar radiation as a thick, light coloured coat (Walsberg, 1990).

Coat thickness plays an important role in the amount of radiant heat that reaches the animal's body. A thick coat can either reduce heat loss, by trapping heat inside the fur, retarding heat loss to the environment, or it can decrease heat gain, by preventing penetration of radiation through the coat and being absorbed by the skin (Walsberg, 1992). Koala's and polar bears live in extreme opposite thermal environments and have different coloured coats. However, both their coats were of similar thickness and provided the same amount of insulation against solar radiation at low wind speeds (Dawson et al., 2014). Thick wool is thought to protect sheep from solar radiation. When sheep were exposed to the sun in hot conditions, shorn sheep showed higher respiration rates (indicating thermal panting in response to heat stress) than unshorn sheep (Macfarlane et al., 1958). Therefore, other insulative properties of the coat (such as the texture, density and depth) also affect the amount of insulation the coat offers against solar radiation and could play as significant a role in insulation as the colour of the coat (Walsberg, 1992, Dawson and Brown, 1970).

The temperature-sensitive data loggers were implanted subcutaneously. Therefore, the loggers measured the amount of radiant heat absorbed by the skin, not the amount of radiant heat the wildebeest were exposed to. If the thick, dark coat of the black wildebeest provided insulation from solar radiation, this insulation would decrease the amount of solar radiation absorbed by the side facing perpendicular to solar radiation, decreasing the subcutaneous temperature difference. Therefore, it would be more difficult to detect orientation to solar radiation using subcutaneous temperatures in black wildebeest than in blue wildebeest. However, I am not aware of any published data quantifying the insulation value of the black wildebeest coat. To study orientation to solar radiation behaviour in black and blue wildebeest using subcutaneous temperature, it is important to understand the thermal properties of the coats of the two species. The structure, density, reflectance, absorbance and optical properties of the winter and summer coat would have to be quantified and compared between the two species (similar to studies done by Walsberg, 1990, Dawson and Brown, 1970, Walsberg and Schmidt, 1989) to understand how their subcutaneous temperature is affected by solar radiation. Knowing the thermal properties of their coats will assist in determining why the prediction model worked better for blue wildebeest than for black wildebeest. Biological differences between species such as size and insulation levels could complicate the use of the prediction model for comparison of orientation to solar radiation between different species. The prediction model would have to be validated for each species and for different seasons (to account for seasonal variation in coat properties) before it could be used to study orientation to solar radiation in a different species.

4.2.6 Orientation to solar radiation in black and blue wildebeest

Previous studies have shown that wildebeest prefer to orientate perpendicular to the sun early in the morning and late afternoon, when ambient temperatures are cool, to increase the uptake of radiant heat, and prefer to orientate parallel to the sun in the heat of the day to expose the smallest possible surface area to the sun (Berry et al., 1984, Vrahimis and Kok, 1992, Maloney et al., 2005b, Hetem et al., 2011b). Blue wildebeest were more likely to orientate to the sun in the early morning and late afternoon and less likely to orientate towards the sun in the middle of the day (Berry et al., 1984), probably due to the angle of the sun at midday. Because the sun was shining from directly above, wildebeest could not avoid the sun by changing their orientation.

However, wildebeest body orientation is not always determined by the direction of the sun. There are times when wildebeest orientation is determined by other factors. When they are seeking shade or when there is cloud cover, their body orientation relative to solar radiation is more likely to be random (Maloney et al., 2005b, Hetem et al., 2011b). If wildebeest are walking towards a water hole or a grazing area (Berry et al., 1984), their orientation will be based on the direction they are walking in and not necessarily be related to the direction of solar radiation. If wind speeds are high animals might prefer to orientate relative to the wind direction (Berry et al., 1984, Vrahimis and Kok, 1992) rather than to solar radiation. Wildebeest can orientate towards a threat, for example, by turning to look at a human observer (personal observation). One study argued that cattle and deer preferred to orientate to magnetic north at times when they were unlikely to use orientation for thermal advantages (Begall et al., 2008).

The times when wildebeest body orientation relative to the sun is best reflected by the subcutaneous temperature difference are probably the times when orientation to solar radiation is most effective and when wildebeest are most likely to rely on orientation to solar radiation for thermoregulatory purposes. For example, if, on a cold day, the wind speed was so high that it cooled down the subcutaneous temperature on the side facing perpendicular to solar radiation, perpendicular orientation to solar radiation would no longer be an effective method to increase heat gain from solar radiation. In that case, the wildebeest might be more likely to orientate to the wind than to solar radiation. Similarly, if, on a hot day, the angle of the sun during midday was so that the wildebeest could not reduce heat gain by orientating parallel to solar radiation, they were more likely to use other thermoregulatory strategies, such as shade-seeking, instead of orientation to solar radiation.

Blue wildebeest often prefer to seek shade in the middle of the day, especially in the summer (Ben-Shahar and Fairall (1987), Hetem et al (2011b) and personal observation), while the black wildebeest are mostly in the sun all day (Estes, 1966). It could be that blue wildebeest only orientate parallel to solar radiation up to a certain black globe temperature and seek shade to avoid the sun when it gets too hot. Shade-seeking is a very effective thermoregulatory strategy and many animals in hot environments might be dependent on shade-seeking for survival (Blackshaw and Blackshaw, 1994, Cain et al., 2008, Hetem et al., 2012a). However, the disadvantage of shade-seeking behaviour is that it decreases time spent grazing (Bennett et al., 1985, Ben-Shahar and Fairall, 1987). The advantage of orientation to solar radiation behaviour is that, in hot conditions, it enables an animal to be out in the sun grazing for a greater proportion of the day, than it would have been had it been in shade.

4.3 Perspective and significance

This study is the first attempt to study orientation to solar radiation behaviour in any species using a remote technique. The prediction model correctly reflected the actual orientation to solar radiation of wildebeest more often than would be expected by chance. However, factors other than solar radiation also influenced the subcutaneous temperatures of wildebeest, leading to incorrect predictions of orientation relative to solar radiation, and reduced accuracy of the technique. Even though the prediction model described in this study is not accurate enough to be used on its own to study orientation to solar radiation behaviour of ungulates remotely, it is a good first step towards that goal. This study was also able to identify which environmental conditions and biological factors affected the accuracy of the remote technique. Therefore future studies can focus on compensating for the effect of these factors on the accuracy of the remote technique. If this remote technique could be used to study orientation to solar radiation behaviour successfully in wildebeest, it would be a significant advancement in the field of behavioural thermoregulatory studies of free-living ungulates.

4.4 Possible ways to improve the remote technique to quantify orientation to solar radiation behaviour

Before orientation to solar radiation can be studied remotely using subcutaneous temperatures, further investigation will be necessary. One possible way of improving the method is to measure surface temperature instead of subcutaneous temperature. Because the loggers were implanted below the skin and panniculus muscle of the wildebeest, it measured the amount of solar radiation that was absorbed through the pelt and skin of the wildebeest and not the amount of solar radiation the wildebeest was exposed to. To measure the radiant heat the wildebeest was exposed to, it would be necessary to place a temperature, light or radiation sensor on the outer coat surface. However, having a data logger attached on the outside of the animal holds the risk that the data logger can easily get damaged or dislodged. Alternatively, the data loggers can be implanted subcutaneously with an external thermistor measuring surface temperature. However, having an external thermistor can increase the risk of infection for the animal and water damage to the data logger. The other option is to attach temperature, light or radiation sensors to a collar. There is a risk that a data logger attached to a collar could get covered by mud and dust, which could interfere with light and radiation measurements. A further possible limitation is that a wildebeest’s neck and head is not always orientated the same way as the wildebeest’s body. However, despite these limitations, attaching a temperature light or radiation sensor to a collar may provide a promising technique to determine orientation to solar radiation, since it excludes the effects of core temperature, coat insulation and peripheral blood flow.

The advantage of using subcutaneous temperature is that subcutaneous temperatures can quantify radiant heat absorbed by the skin (not just what the animal is exposed to) and is therefore a better indication of the thermal load the animal is exposed to than surface temperature would be. Therefore, I suggest future studies should still focus on using subcutaneous temperatures to indicate orientation to solar radiation. Studying subcutaneous temperatures of animals employing different orientations to solar radiation can answer questions about orientation preferences, such as “Are the wildebeest orientating according to the sun, the wind, or magnetic north?” An improved remote technique using subcutaneous temperatures has the potential to accurately quantify the orientation to solar radiation of wildebeest and simultaneously measure the effect of the orientation behaviour on the thermal balance of the wildebeest. Measuring the effect of orientation to solar radiation on the subcutaneous temperatures under different environmental conditions enables us to improve our understanding of how animals utilise orientation to solar radiation in their natural environment and how they could employ orientation to solar radiation to adapt to changes in climate. Therefore, instead of using a different measurement to study orientation to solar radiation, I suggest that other measurements can be made in addition to subcutaneous temperature to improve the accuracy of the technique, such as measuring animals’ orientation relative to magnetic North.

The animal’s orientation to magnetic North can be measured using either a mechanical or a solid state compass (Wilson et al., 2007). The mechanical compass is a ship compass consisting of a bar magnet inside two fluid-filled spheres with two Hall sensors placed on the outside of the outer sphere at 90° angles to each other, which are connected to a data recording unit (Hochscheid and Wilson, 1999). The newer solid state compass is a tri-axial magnetometer with 3 axes at 90° angles to one another (Wilson et al., 2008). By comparing the wildebeest orientation relative to magnetic north to the direction of solar radiation, one could infer the orientation of the wildebeest relative to solar radiation. The angle of the sun can be calculated for a specific place at a specific date and time (Walsberg, 1992). Using compass orientation to determine orientation to solar radiation means that the orientation of the wildebeest can be accurately determined, not only for perpendicular and parallel orientation to solar radiation but also for orientations that are oblique to solar radiation. Previous orientation positions, environmental factors and body posture of the animal will not affect the compass measurements. The combination of the compass direction and the subcutaneous temperatures would allow us to quantify, not only the orientation behaviour of the wildebeest, but also the physiological effects of orientation to solar radiation of the wildebeest.

However, there are limitations to using a compass to determine the orientation of the animal. Body orientation of an animal relative to solar radiation will not necessarily have the same thermal effect when wildebeest are lying down compared to when they are standing. Therefore, if the compass module predicts the wildebeest to be orientated perpendicular to solar radiation when it is lying down, the thermal load on that wildebeest will not necessarily be the same as when the wildebeest is standing orientated perpendicular to solar radiation.

Therefore it would be essential to use a remote method to determine if a wildebeest was lying down or standing. A pedometer attached to the wildebeest's leg to determine whether the leg of the wildebeest is vertical or horizontal can be used to detect the body posture of the wildebeest (similar to the pedometer used in horses Brinkmann et al., 2012).

A compass module also cannot detect shade cover. If the compass module predicts that the wildebeest is orientated perpendicular to the sun, but the wildebeest is either seeking shade or under cloud cover, its orientation to solar radiation will not have any thermoregulatory advantages. A miniature black globe thermometer attached to the collar of the animal can detect shade-seeking behaviour (Hetem et al., 2007). The miniature black globe temperature would further enable us to study microclimate selection in animals together with orientation to solar radiation. Studying both orientation to solar radiation and microclimate selection simultaneously will increase our understanding of how animals employ behavioural strategies for thermoregulation, and which environmental factors drive behavioural thermoregulation. Climatic data from a weather station set up close to the animals will enable us to exclude times when the wildebeest are less likely to use orientation to solar radiation for thermoregulatory behaviours, such as when wind speed is high or when it is overcast. In addition, climatic data would enable us to study which environmental factors drive orientation to solar radiation behaviour in ungulates.

Vasodilation of peripheral blood vessels in hot conditions could affect subcutaneous temperatures. Measuring core temperature in addition to the subcutaneous temperature would give an indication of vasomotor state in wildebeest. Since peripheral vasodilation and vasoconstriction could affect the subcutaneous temperature, measuring core temperature is important to exclude the effect of vasodilation and vasoconstriction on the accuracy of the prediction model. Additionally, measuring abdominal temperature will indicate how the animal's core temperature is affected by the heat load it is exposed to.

Because there were so many factors influencing subcutaneous temperatures, a greater sample size of observations would be advantageous to study the effect of multiple factors on subcutaneous temperature and to study the accuracy of the remote technique under different conditions. In previous studies where remote techniques were used to study animal behaviour remotely, the remote technique was sometimes validated in captive animals before the technique was used to study free-living animals (Hochscheid and Wilson, 1999, Charrassin et al., 2001). For future studies I would suggest that implanted individuals should be kept in a pen where they can be observed easily to validate the remote technique before using it to study orientation to solar radiation in free-living animals. Validating the technique in captive animals could involve extra costs and time, but I believe that it would improve the effectiveness of the study as it would increase the amount of observations that can be used and would reduce difficulties associated with studying free-living animals.

I recommend that future studies to develop a remote technique to study orientation to solar radiation use a combination of subcutaneous temperatures, core temperature, compass direction of the animal's body orientation relative to magnetic north, a pedometer attached to the animal's leg, miniature black globe temperature, and a weather station, to study orientation to solar radiation remotely. The advantage of measuring all of these variables simultaneously, as mentioned above, is that it will enable us to quantify the value of orientation to solar radiation in relation to the thermal balance of the wildebeest and enable us to study which behavioural thermoregulatory strategies animals employ in different environmental conditions.

Chapter 5

Conclusion

5 Conclusion

Developing a remote technique to accurately quantify the orientation to solar radiation of wild animals would be invaluable to future research and would improve both the quality and quantity of data that can be collected on animal thermoregulatory behaviour. My study showed, for the first time, the potential of using biologging of subcutaneous temperatures to remotely study orientation to solar radiation in black and blue wildebeest. The difference between left and right subcutaneous temperatures together with the rate of change of difference between left and right subcutaneous temperatures accurately reflected the orientation of wildebeest to solar radiation more often than would be expected by chance. Therefore the orientation of a wildebeest relative to solar radiation has an influence on the subcutaneous temperatures of a wildebeest. However, the technique was correct only 60 % of the time. Therefore, before this prediction model can be used to study orientation to solar radiation remotely in free-living wildebeest, further studies need to be done to refine the technique.

The remote technique to predict orientation to solar radiation can be improved by attaching a compass to the animal collar to measure the direction of an animal's body relative to magnetic north. Further measurements would be necessary to exclude conditions in which the remote technique would not be effective, such as times when the wildebeest are lying down and times when the wildebeest are in the shade. A pedometer can be attached to the animal's leg to determine when the animal is lying down. A weather station set up close to the study animals and a miniature black globe thermometer attached to the wildebeest collar can indicate when an animal is seeking shade or under cloud cover.

When studying thermoregulatory behaviour in animals, it would be beneficial to study all the thermoregulatory responses an animal could employ. For example, microclimate selection can be studied together with orientation to solar radiation. To better understand why animals select certain microclimates in their environment, the temperature experienced by animals in the different microclimates that are available to them can be measured by placing temperature data loggers and miniature black globes in different microclimates. For example, temperature data loggers attached to an animal collar, together with GPS coordinates, can be used to measure the air temperature in various microclimates and study habitat use and habitat selection (Van Beest et al., 2012).

To quantify the effect of certain behavioural strategies on the thermoregulation of the animal, various physiological variables of the animal (such as subcutaneous and core temperature) can be measured whilst studying the different behavioural strategies animals employ under different environmental conditions. For example, by measuring the subcutaneous temperature of a wildebeest while quantifying its orientation to solar radiation, we can quantify the amount of radiant heat it absorbs compared to when it is orientated perpendicular to solar radiation. Vasomotor state can be studied by measuring abdominal temperature combined with subcutaneous temperatures. It is also important to study the environmental factors that drive these behavioural responses. Climatic variables measured by a weather station close by to the study animals can be used to study which environmental variables drive orientation to solar radiation preferences in animals.

Many remote techniques have been used to study animal behaviour. GPS tracking devices are often used to study home range characteristics (Whyte et al., 2013), activity patterns (Eriksen et al., 2011) and movement (Van Beest et al., 2012, Lindberg, 2013, Tremblay et al., 2014) or to track animals during behavioural studies (Lake et al., 2013). Camera traps placed in strategic places have been used in animal conservation to determine distribution (Ross et al., 2013) and population densities (Silver et al., 2004, Bashir et al., 2013). In animal behaviour studies, camera traps have been used, for example, to study oviposition timing in the Oregon Spotted Frog (*Rana pretiosa*; Ramsdell, 2013) and small mammal activity in suburban backyards (Kays and Parsons, 2014). Alternatively, a small camera can be attached to the animal's head to record what the animal sees, to study animal movement and foraging behaviour over a short term (Davis et al., 1999, Tremblay et al., 2014).

Implanted biologgers have been used to study different behavioural and physiological aspects of animals. Biologgers are especially popular in studies of marine animals (Wilson et al., 2002, Wilson et al., 2007, Ropert-Coudert et al., 2009, Bograd et al., 2010), probably because it is difficult to use visual observations to study marine animals when they are out at sea (Bograd et al., 2010). For example, in free-ranging penguins, temperature data loggers were implanted in each animal's oesophagus to measure feeding behaviour while at sea (Charassin et al., 2001).

Biologging has also been used in terrestrial animal studies to measure both physiological and behavioural patterns. In ostriches, brain activity has been measured during sleep using implanted electrodes (EEG, EOG and EMG) and a thermistor in the brain (Lesku et al., 2011). Implanted activity loggers have been used to detect activity patterns in relation to environmental conditions (Mitchell et al., 1997, Hetem et al., 2012b). Implanted ECG electrodes have been used to measure heart rate as an indication of metabolic rate in red deer (Giacometti et al., 2001, Arnold et al., 2004). Thermistors implanted in the brain and carotid artery have been used to study selective brain cooling in ungulates (Mitchell et al., 1997, Fuller et al., 1999). Microclimate selection can be quantified using miniature black globe temperatures (Hetem et al., 2007). Body temperature can be measured with abdominally implanted temperature data loggers and has been used to study daily body temperature rhythms (Hetem et al., 2012b), heterothermy (Hetem et al., 2010) fever responses (Hetem et al., 2008), metabolic rate (Signer et al., 2010) and torpor (Grimpo et al., 2013). The above examples illustrate some of the vast possibilities of using remote techniques to study animal behaviour and physiology. A wide range of behavioural and physiological variables can be measured using appropriate technology, opening up many possibilities for studying animal behaviour and physiology remotely.

However, there are certain risks and limitations related to using remote techniques. Some of the limitations are related to the technology that is currently available. The battery life and memory space of data loggers that are currently available can be a limiting factor when doing long-term studies on animals. Implanted biologgers that store data, but do not transmit the data, can only be accessed after termination of the study, because it is necessary to retrieve the data logger to access the data. There are risks involved in retrieving the data loggers, such as failure to recapture study animals, data loggers lost due to predation of animals or scavenging of carcasses of animals that died during the study. Recapture of animals may be more difficult than the initial capture, depending on the learning and memory capacity of the animals and the capture method used. There is the risk that data loggers can fail before the end of the study. Because data can only be accessed at the end of the study, it is not known beforehand how much data is being recorded during the time period of the study until the data loggers are retrieved, so failed equipment cannot be repaired or replaced throughout the study. A substantial amount of time and money is often invested into the use of remote techniques, therefore, it would be a great loss if loggers failed to record data during a study.

To avoid the risks associated with logging data on a device that needs to be retrieved, many researchers are opting for devices with a remote download capacity. When using biologgers that send data via radio or satellite, the data can be accessed and analysed throughout the study and devices that stop recording during the study can be fixed or replaced. Ethically, animals should still be recaptured to remove all devices once they stop functioning. Data loggers that send data via radio, cellphone or satellite need to be either external data loggers, or implanted data loggers that transmit recorded data to an external collar (Arnold et al., 2004, Signer et al., 2010). When using VHF transmitters, animals have to be close enough to the receiver for data to be transmitted. For cellphone transmitters, animals need to be in an area with cellphone reception. There are data loggers available that can both store data and send data to a receiver (Cooke, 2008), but these are often expensive and large in size, because they require more battery power than data loggers that either only store or only transmit data, and therefore the battery needs to be greater in size to have the same battery life (Cooke et al., 2004, Cooke, 2008).

The equipment needed for remote monitoring requires a certain amount of expertise for use, since great care needs to be taken when deciding which equipment would be most appropriate to use to answer the specific study question (Cooke, 2008). Managing and analysing the large quantities of data recorded by these loggers can also be challenging (Rutz and Hays, 2009). Managing large amounts of data could slow down the process of analysing, writing up and publishing data that has been collected. Due to costs and logistic limitations of using biologgers (e.g. how many animals you manage to capture and implant) the sample size of animal subjects used in a study can sometimes be small, causing difficulties during statistical analysis of data (Cooke et al., 2004).

Biologging involves certain risks regarding animal welfare (Hawkins, 2004). External marker tags can decrease survival rates in penguins (Gauthier-Clerc et al., 2004, Dugger et al., 2006) and external data loggers can restrict movement or foraging behaviour of animals (Wilson et al., 1986, Bannasch et al., 1994). Implanting data loggers surgically is invasive and there is a morbidity and mortality risk involved for the animal, both during and after the surgery (Hawkins, 2004). The capture, handling and surgery of animals should be done by experienced professionals. Surgery should be performed under conditions as sterile as possible and should be conducted by a veterinarian. After surgery the animals should be monitored in case any complications arise. Many studies are being done to reduce the risks associated with the use of biotelemetry, by improving the attachment or surgical implantation of biologgers to reduce possible negative effects on animal welfare and natural behaviour (Bannasch et al., 1994, Giacometti et al., 2001). An important aspect of animal welfare, which should be considered, especially in small animals, is the total mass of equipment that is implanted or attached to the animal (Hawkins, 2004). Therefore, the size of the animal limits the amount and type of loggers that can be used.

There have been many advances in biologging over the past few years. Not only have there been improvements in technology, such as a decrease in device size and increase in memory capacity and battery life span, but also in the use of available technology (Rutz and Hays, 2009, Bograd et al., 2010). For example, a “backpack”, which could be attached to a penguin’s back to record various physiological and environmental measurements, was designed to provide a “diary”, recording the daily interactions of an animal with its environment (Wilson et al., 2008). This “Daily Diary” measured animal location and movement (using dead-reckoning), behaviour (by using tri-axial magnetometer and tri-axial accelerometer), energy expenditure and environmental conditions. The device could record data for 35 days using a memory card of 1 GB. Today, such memory cards are available with much greater capacity. For example, a 32 GB SD memory card can be easily acquired in South Africa for less than R300 (Price Check, South Africa, www.pricecheck.co.za, search: SD memory card, 7 March 2014) and technology is increasing memory capacity of these memory cards all the time. Therefore, in terms of memory capacity, it would be easy to collect data for longer than a year. Battery life is more likely to be a constraint than is memory capacity. The Daily Diary used a lithium battery (3.6 V), which could last 7 days. A larger battery would last longer, but would increase the size of the package.

Measuring a range of environmental, physiological and behavioural variables could be advantageous when studying how animals respond to changes in their environment. As biologgers are becoming smaller and battery life and memory capacity of loggers is increasing, it will be easier to study smaller animals for longer periods of time. The advantage of studying large mammals (greater than 100 kg) is that it is possible to measure a range of variables (for example body temperature, activity and microclimate) recorded every 10 minutes for more than a year.

Measuring multiple variables continuously for one year or longer, will enable us to study thermoregulatory strategies animals employ during all seasons. We can determine which strategies are most essential to animal survival, and what are the threshold conditions where these strategies are either no longer effective or become too costly for the animal. For example, there are trade-offs between foraging and seeking shade (Bennett et al., 1985, Ben-Shahar and Fairall, 1987). Therefore, if animals have access to water and can use evaporative cooling they may prioritize foraging. However, if water becomes limited, as predicted with climate change in many regions, evaporative cooling will become too costly to their water balance and the animals might prefer behavioural responses such as seeking shade or shifting their orientation to solar radiation.

Climate change predictions state that many parts of South Africa will become hotter and drier (IPCC, 2013). There have been predictions of how animals will respond to climate change, especially in terms of distribution shifts (Erasmus et al., 2002, Thomas et al., 2004, Rosenzweig et al., 2008, Chen et al., 2011) and genetic adaptation (Bradshaw and Holzapfel, 2008, Skelly and Freidenburg, 2010, Quintero and Wiens, 2013). However, to really understand how animals will respond to climate change, phenotypic plasticity needs to be taken into account (Fuller et al., 2010). Because behavioural responses are immediate and cost-effective, they are likely to be an animal's first response to environmental change. Therefore studying animal behaviour is essential to understanding how animals might respond to climate change. Currently, we don't know enough of the behaviour, ecology, physiology or genetics of animals to accurately predict responses to climate change (Gienapp et al., 2008). The development of remote techniques has increased the possibilities to study behavioural and physiological ecology of animals. Studying behavioural thermoregulation in animals remotely can help us to better understand how animals might utilise behavioural strategies to adapt to changes in their environment.

Information on the behavioural and physiological adaptations that are vital to animal survival is important when making management decisions affecting animal welfare and conservation (Cooke, 2008, Heller and Zavaleta, 2009, Bograd et al., 2010). Biologging could enable scientists to study mortality and survival rates, reproduction biology, distribution, abundance, and movement of animals (Cooke, 2008). Studying animal physiology and behaviour could indicate how animals could adapt to changes in their current habitats. For example, detailed information on the interactions between lizard physiology and the environment enabled the calculation of the basic niche in which a lizard could survive and reproduce (Kearney and Porter, 2004, Kearney and Porter, 2009, Kearney et al., 2012). The same could be done for large mammals. Measuring core, subcutaneous and miniature black globe temperature could give an indication of the heat load an animal is exposed to and the microclimates available in its current habitat and how animals use these microclimates (Van Beest et al., 2012). The effect of other biological factors, such as coat colour and thickness, on the thermoregulation of an animal also needs to be studied and quantified. Climate change predictions can then be used to predict animal distribution shifts (Kearney and Porter, 2004, Kearney and Porter, 2009, Kearney et al., 2012).

Using biologging to study animals can enable us to predict how animals might respond to climate change and identify which species might be most vulnerable to changes in temperature and aridity predicted for climate change. We can then focus conservation strategies on those sensitive species to ensure their survival in the future. For example, in cases where species are unable to shift their distribution to follow climate change, “corridors” can be made between isolated habitats (Heller and Zavaleta, 2009) or species can be physically removed from their original habitat and moved to habitats with more suitable climate (Hoegh-Guldberg et al., 2008).

However, further research is necessary to guide management and conservation decisions (Heller and Zavaleta, 2009). For example, there are many different opinions regarding assisted colonization (McLachlan et al., 2007, Hoegh-Guldberg et al., 2008, Ricciardi and Simberloff, 2009), since it could have unforeseen ecological impacts associated with species introduced into new habitats (Ricciardi and Simberloff, 2009). Biologging provides the necessary tools to increase our knowledge of animal ecology, physiology and behaviour. Increased knowledge of ecology, behaviour and physiology of animals might enable us to determine which conservation strategies would be best in specific situations.

References

ACOCKS, J. P. H. (1988) *Veld types of South Africa*, Botanical Research Institute, Department of Agriculture and Water Supply, South Africa.

ADOLPH, S. C. (1990) Influence of behavioral thermoregulation on microhabitat use by two sceloporus lizards. *Ecology,* 71**,** 315-327.

ADOLPH, S. C. & PORTER, W. P. (1993) Temperature, activity and lizard life histories. *The American Naturalist*, 142**,** 273-295.

ALTMANN, J. (1974) Observational study of behavior: sampling methods. *Behaviour,* 49**,** 227-267.

ANDERSON, A. R., HOFFMANN, A. A., MCKECHNIE, S. W., UMINA, P. A. & WEEKS, A. R. (2005) The latitudinal cline in the *IN(3R)Payne* inversion polymorphism has shifted in the last 20 years in Australian *Drosophila melanogaster* populations. *Molecular Ecology,* 14**,** 851-858.

ARNOLD, W., RUF, T., REIMOSER, S., TATARUCH, F., ONDERSCHEKA, K. & SCHOBER, F. (2004) Nocturnal hypometabolism as an overwintering strategy of red deer (*Cervus elaphus*). *American Journal of Physiology - Regulatory, Integrative and Comparative Physiology*, 286**,** 174-181.

BANNASCH, R., WILSON, R. P. & CULIK, B. (1994) Hydrodynamic aspects of design and attachment of a back-mounted device in penguins. *Journal of Experimental Biology,* 194**,** 83-96.

BARTHOLOMEW, G. A. (1964) The roles of physiology and behaviour in the maintenance of the homeostasis in the desert environment. IN HUGHES, G. M. (Ed.) *Society of experimental biology symposium.* Cambridge University Press.

BARTHOLOMEW, G. A. & DAWSON, W. R. (1979) Thermoregulatory behavior during incubation in Heermann's gulls. *Physiological Zoology,* 52**,** 422-437.

BASHIR, T., BHATTACHARYA, T., POUDYAL, K., SATHYAKUMAR, S. & QURESHI, Q. (2013) Estimating leopard cat *Prionailurus bengalensis* densities using photographic captures and recaptures. *Wildlife Biology,* 19**,** 462-472.

BEGALL, S., CERVENY, J., NEEF, J., VOJTECH, O. & BURDA, H. (2008) Magnetic alignment in grazing and resting cattle and deer. *Proceedings of the National Academy of Sciences,* 105**,** 13451-13455.

BEN-SHAHAR, R. & FAIRALL, N. (1987) Comparison of the diurnal activity patterns of blue wildebeest and red hartebeest. *South African Journal of Wildlife Research,* 17**,** 49-54.

BENNETT, A. F. & RUBEN, J. A. (1979) Endothermy and activity in vertebrates. *Science,* 206**,** 649-654.

BENNETT, I. L., FINCH, V. A. & HOLMES, C. R. (1985) Time spent in shade and its relationship with pysiological factors of thermoregulation in three breeds of cattle. *Applied Animal Behaviour Science,* 13**,** 227-236.

BERRY, H. H. (1997) Aspects of wildebeest *Connochaetes taurinus* ecology in the Etosha National Park - a synthesis for future management. *Madoqua,* 20**,** 137-148.

BERRY, H. H., SIEGFRIED, W. R. & CROWE, T. M. (1982) Activity patterns in a population of free-ranging wildebeest *Connochaetes taurinus* at Etosha National Park. *Zeitschrift fur Tierpsychologie,* 59**,** 229-246.

BERRY, H. H., SIEGFRIED, W. R. & CROWE, T. M. (1984) Orientation of wildebeest in relation to sun angle and wind direction. *Madoqua,* 13**,** 297-301.

BERRY, R. J. & BRONSON, F., H, (1992) Life history and bioeconomy of the house mouse. *Biological Reviews,* 67**,** 519-550.

BLACKSHAW, J. K. & BLACKSHAW, A. W. (1994) Heat stress in cattle and the effect of shade on production and behaviour: a review. *Australian Journal of Experimental Agriculture,* 34**,** 285-295.

BLANCO, M. B., DAUSMANN, K. H., RANAIVOARISOA, J. F. & YODER, A. D. (2013) Underground hibernation in a primate. *Scientific Reports,* 1768**,** 1-4.

BOGRAD, S. J., BLOCK, B. A., COSTA, D. P. & GODLEY, B. J. (2010) Biologging technologies: new tools for conservation. Introduction. *Endangered Species Research,* 10**,** 1-7.

BRACKE, M. B. M. (2011) Review of wallowing in pigs: Description of the behaviour and its motivational basis. *Applied Animal Behaviour Science,* 132**,** 1-13.

BRADSHAW, W. E. & HOLZAPFEL, C. M. (2001) Genetic shift in photoperiodic response correlated with global warming. *Proceedings of the National Academy of Sciences of the United States of America,* 98**,** 14509-14511.

BRADSHAW, W. E. & HOLZAPFEL, C. M. (2008) Genetic response to rapid climate change: it's seasonal timing that matters. *Molecular Ecology,* 17**,** 157-166.

BRINKMANN, L., GERKEN, M. & RIEK, A. (2012) Adaptation strategies to seasonal changes in environmental conditions of a domesticated horse breed, the Shetland pony (*Equus ferus caballus*). *Journal of Experimental Biology,* 215**,** 1061-1068.

CAIN, J. W., JANSEN, B. D., WILSON, R. R. & KRAUSMAN, P. R. (2008) Potential thermoregulatory advantages of shade use by desert bighorm sheep. *Journal of Arid Environments,* 72**,** 1518-1525.

CAMPBELL, G. S., MCARTHUR, A. J. & MONTEITH, J. L. (1980) Windspeed dependence of heat and mass transfer through coats and clothing. *Boundary-Layer Meteorology,* 18**,** 485-493.

CARROLL, A. L., TAYLOR, S. W., REGNIERE, J. & SAFRANYIK, L. (2003) Effect of climate change on range expansion by the mountain pine beetle in British Columbia. *The Bark Beetles, Fuels, and Fire Bibliography,* 195.

CHARASSIN, J., KATO, A., HANDRICH, Y., SATO, K., NAITO, Y., ANCEL, A., BOST, C., GAUTHIER-CLERC, M., ROPERT-COUDERT, Y. & LE MAHO, Y. (2001) Feeding behaviour of free-ranging penguins determined by oesophageal temperature. *Proceedings of the Royal Society of Londond B,* 268**,** 151-157.

CHARRASSIN, J., KATO, A., HANDRICH, Y., SATO, K., NAITO, Y., ANCEL, A., BOST, C. A., GAUTHIER-CLERC, M., ROPERT-COUDERT, Y. & LE MAHO, Y. (2001) Feeding behaviour of free-ranging penguins determined by oesophageal temperature. *Proceedings of the Royal Society of Londond B,* 268**,** 151-157.

CHEN, I. C., HILL, J. K., OHLEMULLER, R., ROY, D. B. & THOMAS, C. D. (2011) Rapid range shifts of species associated with high levels of climate warming. *Science,* 333**,** 1024-1026.

CLARKE, A. & ROTHERY, P. (2008) Scaling of body temperature in mammals and birds. *Functional Ecology,* 22**,** 58-67.

COE, M. (2004) Orientation, movement and thermoregulation in the giant tortoises (*Testudo (Geochelone) gigantea*) of Aldabra Atoll, Seychelles. *Transactions of the Royal Society of South Africa,* 59**,** 73-77.

COLLINS, K. J. (1983) *Hypothermia - the facts*, Oxford University Press, Oxford, UK.

COOKE, S. J. (2008) Biotelemetry and biologging in endangered species research and animal conservation: relevance to regional, national, and IUCN Red List threat assessments. *Endangered Species Research,* 4**,** 165-185.

COOKE, S. J., HINCH, S. G., WIKELSKI, M., ANDREWS, R. D., KUCHEL, L. J., WOLCOTT, T. G. & BUTLER, P. J. (2004) Biotelemetry: a mechanistic approach to ecology. *Trends in Ecology and Evolution,* 19**,** 334-343.

DASILVA, G. L. (1993) Postural changes and behavioural thermoregulation in *Colobus polykomos*: the effect of climate and diet. *African Journal of Ecology,* 31**,** 226-241.

DAVIS, R. W., FUIMAN, L. A., WILLIAMS, T. M., COLLIER, S. O., HAGEY, W. P., KANATOUS, S. B., KOHIN, S. & HORNING, M. (1999) Hunting behavior of a marine mammal beneath the antarctic fast ice. *Science,* 283**,** 993-996.

DAWSON, T. J. & BROWN, G. D. (1970) A comparison of the insulative and reflective properties of the fur of desert kangaroos. *Comparative Biochemistry and Physiology,* 37**,** 23-38.

DAWSON, T. J. & MALONEY, S. K. (2004) Fur versus feathers: the different roles of red kangaroo fur and emu feathers in thermoregulation in the Australian arid zone. *Australian Mammalogy,* 26**,** 145-151.

DAWSON, T. J., WEBSTER, K. N. & MALONEY, S. K. (2014) The fur of mammals in exposed environments; do crypsis and thermal needs necessarily conflict? The polar bear and marsupial koala compared. *Journal of Comparative Physiology B,* 184**,** 273-284.

DE BOER, H. Y., VAN BREUKELEN, L., HOOTSMANS, M. J. M. & VAN WIEREN, S. E. (2004) Flight distance in roe deer *Capreolus capreolus* and fallow deer *Dama dama* as related to hunting and other factors. *Wildlife Biology,* 10**,** 35-41.

DUGGER, K. M., BALLARD, G., AINLEY, D. G. & BARTON, K. J. (2006) Effects of flipper bands on foraging behaviour and survival of adelie penguins (*Pygoscelis adeliae*). *The Auk,* 123**,** 858-869.

EMMONS, L. H., FLORES, R. P., ALPIRRE, S. A. & SWARNER, M. J. (2004) Bathing behaviour of Giant Anteaters (*Myrmecophaga tridactyla*). *Edentata,* 6**,** 41-43.

ERASMUS, B. F. N., VAN JAARSVELD, A. S., CHOWN, S. L., KSHATRIYA, M. & WESSELS, K. J. (2002) Vulnerability of South African animal taxa to climate change. *Global Change Biology,* 8**,** 679-693.

ERIKSEN, A., WABAKKEN, P., ZIMMERMANN, B., ANDREASSEN, H. P., ARNEMO, J. M., GUNDERSEN, H., LIBERG, O., LINNELL, J., MILNER, J. M., PEDERSEN, H. C., SAND, H., SOLBERG, E. J. & STORAAS, T. (2011) Activity patterns of predator and prey: a simultaneous study of GPS-collared wolves and moose. *Animal Behaviour,* 81**,** 423-431.

ESTES, R. D. (1966) Behaviour and life history of the wildebeest (*Connochaetes taurinus* Burchell). *Nature,* 212**,** 999-1000.

ESTES, R. D. (1997) *The behaviour guide to African mammals,* Russel Friedman Books, Kyalami, South Africa

FARMER, C. G. (2000) The key to understanding endothermy and other convergent features in birds and mammals. *The American Naturalist,* 155**,** 326-334.

FICK, L. G., KUCIO, T. A., FULLER, A., MATTHEE, A. & MITCHELL, D. (2009) The relative roles of the parasol-like tail and burrow shuttling in the thermoregulation of free-ranging Cape ground squirrels, *Xerus inauris*. *Comparative Biochemistry and Physiology A,* 152**,** 334-340.

FINCH, V. A. (1972) Thermoregulation and heat balance of the East African eland and hartebeest. *American Journal of Physiology,* 222**,** 1374-1379.

FINCH, V. A., DMI'EL, R., BOXMAN, R., SHKOLNIK, A. & TAYLOR, C. R. (1980) Why black goats in hot deserts? Effects of coat color on heat exchanges of wild and domestic goats. *Physiological Zoology,* 53**,** 19-25.

FORTIN, D., LAROCHELLE, J. & GAUTHIER, G. (2000) The effect of wind, radiation and body orientation on the thermal environment of Greater Snow goose goslings. *Journal of Thermal Biology,* 25**,** 227-238.

FULLER, A., DAWSON, T., HELMUTH, B., HETEM, R. S., MITCHELL, D. & MALONEY, S. K. (2010) Physiological mechanisms in coping with climate change. *Physiological and Biochemical Zoology,* 83**,** 713-720.

FULLER, A., MALONEY, S. K., KAMERMAN, P. R., MITCHELL, G. & MITCHELL, D. (2000) Absence of selective brain cooling in free-ranging zebras in their natural habitat. *Experimental Physiology,* 85**,** 209-217.

FULLER, A., MOSS, D. G., SKINNER, J. D., JESSEN, P. T., MITCHELL, G. & MITCHELL, D. (1999) Brain, abdominal and arterial blood temperatures of free-ranging eland in their natural habitat. *European Journal of Physiology,* 438**,** 671-680.

GAUTHIER-CLERC, M., GENDNER, J.-P., RIBIC, C. A., FRASER, W. R., WOEHLER, E. J., DESCAMPS, S., GILLY, C., LE BOHEC, C. & LE MAHO, Y. (2004) Long-term effects of flipper bands on penguins. *Proceedings of the Royal Society of London B,* 271**,** 423-426.

GEBREMEDHIN, K. G. (1987) Effect of animal orientation with respect to wind direction on convective heat loss. *Agricultural and Forest Meteorology,* 40**,** 199-206.

GEBREMEDHIN, K. G. & WU, B. (2001) A model of evaporative cooling of wet skin surface and fur layer. *Journal of Thermal Biology,* 26**,** 537-545.

GIACOMETTI, M., JANOVSKY, M., FLUCH, G., ARNOLD, W. & SCHOBER, F. (2001) A technique to implant heart-rate transmitters in red deer. *Wildlife Society Bulletin,* 29**,** 586-593.

GIENAPP, P., TEPLITSKY, C., ALHO, J. S., MILLS, J. A. & MERILA, J. (2008) Climate change and evolution: disentangling environmental and genetic responses. *Molecular Ecology,* 17**,** 167-178.

GONYOU, H. G. & STRICKLIN, W. R. (1981) Orientation of feedlot bulls with respect to the sun during periods of high solar radiation in winter. *Canadian Journal of Animal Science,* 61**,** 809-816.

GRIMPO, K., LEGLER, K., HELDMAIER, G. & EXNER, C. (2013) That's hot: golden spiny mice display torpor even at high ambient temperatures. *Journal of Comparative Physiology B,* 183**,** 567-581.

GROBLER, J. P., HARTL, G. B., GROBLER, N., KOTZE, A., BOTHA, K. & TIEDEMANN, R. (2005) The genetic status of an isolated black wildebeest (*Connochaetes gnou*) population from the Abe Bailey Nature Reserve, South Africa: Microsatellite data on a putative past hybridization with blue wildebeest (*C. taurinus*). *Mammalian Biology,* 70**,** 35-45.

GROBLER, J. P. & VAN DER BANK, F. H. (1993) Genetic variability in South African blue wildebeest (*Connochaetes taurinus*). *Comparative Biochemistry and Physiology,* 106B**,** 755-762.

GROBLER, J. P. & VAN DER BANK, F. H. (1995) Allozyme divergence among four representatives of the subfamily Alcelaphinae (family: Bovidae). *Comparative Biochemistry and Physiology,* 112B**,** 303-308.

HALES, J. R. S. (1983) Thermoregulatory requirements for circulatory adjustments to promote heat loss in animals. *Journal of Thermal Biology,* 8**,** 219-224.

HARRI, M. & KORHONEN, H. (1988) Thermoregulatory significance of basking behaviour in the raccoon dog (*Nyctereutes procyonoides*). *Journal of Thermal Biology,* 13**,** 169-174.

HAWKINS, P. (2004) Bio-logging and animal welfare: practical refinements. *Memoirs of National Institute of Polar Research Series Special Edition,* 58**,** 58-68.

HEINRICH, B. (1977) Why have some animals evolved to regulate a high body temperature? *The American Naturalist,* 111**,** 623-640.

HELLER, N. E. & ZAVALETA, E. S. (2009) Biodiversity management in the face of climate change: A review of 22 years of recommendations. *Biological Conservation,* 142**,** 14-32.

HERSTEINSSON, P. & MACDONALD, D. W. (1992) Interspecific competition and the geographical distribution of red and arctic foxes *Vulpes vulpes* and *Alopex lagopus*. *Oikos,* 64**,** 505-515.

HETEM, R. S., DE WIT, B. A., FICK, L. G., FULLER, A., KERLEY, G. I. H., MEYER, L. C. R., MITCHELL, D. & MALONEY, S. K. (2009) Body temperature, thermoregulatory behaviour and pelt characteristics of three colour morphs of springbok (*Anticorcas marsupialis*). *Comparative Biochemistry and Physiology A,* 152**,** 379-388.

HETEM, R. S., DE WIT, B. A., FICK, L. G., FULLER, A., MALONEY, S. K., MEYER, L. C. R., MITCHELL, D. & KERLEY, G. I. H. (2011a) Effects of desertification on the body temperature, activity and water turnover of Angora goats. *Journal of Arid Environments,* 75**,** 20-28.

HETEM, R. S., MALONEY, S. K., FULLER, A., MEYER, L. C. R. & MITCHELL, D. (2007) Validation of a biotelemetric technique, using ambulatory miniature black globe thermometers to quantify thermoregulatory behaviour in ungulates. *Journal of Experimental Zoology,* 307A**,** 342-356.

HETEM, R. S., MITCHELL, D., MALONEY, S. K., MEYER, L. C. R., FICK, L. G., KERLEY, G. I. H. & FULLER, A. (2008) Fever and sickness behavior during an opportunistic infection in a free-living antelope, the greater kudu (*Tragelaphus strepsiceros*). *American Journal of Physiology - Regulatory, Integrative and Comparative Physiology,* 294**,** 246-254.

HETEM, R. S., STRAUSS, W. M., FICK, L. G., MALONEY, S. K., MEYER, L. C. R., SHOBRAK, M., FULLER, A. & MITCHELL, D. (2010) Variation in the daily rhythm of body temperature of free-living Arabian oryx (*Oryx leucoryx*): does water limitation drive heterothermy? *Journal of Comparative Physiology B,* 180**,** 1111-1119.

HETEM, R. S., STRAUSS, W. M., FICK, L. G., MALONEY, S. K., MEYER, L. C. R., SHOBRAK, M., FULLER, A. & MITCHELL, D. (2012a) Activity re-assignment and microclimate selection of free-living Arabian oryx: responses that could minimise the effects of climate change on homeostasis? *Zoology,* 115**,** 411-416.

HETEM, R. S., STRAUSS, W. M., FICK, L. G., MALONEY, S. K., MEYER, L. C. R., SHOBRAK, M., FULLER, A. & MITCHELL, D. (2012b) Does size matter? Comparison of body temperature and activity of free-living Arabian oryx (*Oryx leucoryx*) and the smaller Arabian sand gazelle (*Gazella subgutturosa marica*) in the Saudi desert. *Journal of Comparative Physiology B,* 182**,** 437-449.

HETEM, R. S., STRAUSS, W. M., HEUSINKVELD, B. G., DE BIE, S., PRINS, H. H. T. & VAN WIEREN, S. E. (2011b) Energy advantages of orientation to solar radiation in three African ruminants. *Journal of Thermal Biology,* 36**,** 452-460.

HICKLING, R., ROY, D. B., HILL, J. K., FOX, R. & THOMAS, C. D. (2006) The distributions of a wide range of taxonomic groups are expanding polewards. *Global Change Biology,* 12**,** 450-455.

HOCHSCHEID, S. & WILSON, R. P. (1999) A new method for the determination of at-sea activity in sea turtles. *Marine ecology progress series,* 185**,** 293-296.

HOEGH-GULDBERG, O., HUGHES, L., MCINTYRE, S., LINDENMAYER, D. B., PARMESAN, C., POSSINGHAM, H. P. & THOMAS, C. D. (2008) Assisted colonization and rapid climate change. *Science,* 321**,** 345-346.

HOFMEYR, M. D. & LOUW, G. N. (1987) Thermoregulation, pelage conductance and renal function in the desert-adapted springbok, *Antidorcas marsupialis*. *Journal of Arid Environments,* 13**,** 137-151.

HURLBERT, S. H. (1984) Pseudoreplication and the design of ecological field experiments. *Ecological Monographs,* 54**,** 187-211.

HUTCHINSON, J. C. D., ALLEN, T. E. & SPENCE, F. B. (1975) Measurement of the reflectances for solar radiation of the coats of live animals. *Comparative Biochemistry and Physiology,* 52**,** 343-349.

INGRAM, D. L. (1967) Stimulation of cutaneous glands in the pig. *Journal of Comparative Pathology,* 77**,** 93-99.

IPCC (2013) Climate change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. IN STOCKER, T. F., QIN, D., PLATNER, G. K., TIGNOR, M., ALLEN, S. K., BOSCHUNG, J., NAUELS, A., XIA, Y., BEX, V. & MIDGLEY, P. M. (Eds.), Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA

JANZEN, F. J. & MORJAN, C. L. (2001) Repeatability of microenvironment-specific nesting behaviour in a turtle with environmental sex determination. *Animal Behaviour,* 62**,** 73-82.

JARMAN, M. V. & JARMAN, P. J. (1973) Daily activity of impala. *East African Wildlife Journal,* 11**,** 75-92.

JESSEN, C., LABURN, H. P., KNIGHT, M. H., KUHNEN, G., GOELST, K. & MITCHELL, D. (1994) Blood and brain temperatures of free-ranging black wildebeest in their natural environment. *American Journal of Physiology,* 267**,** 1528-1536.

KAYS, R. & PARSONS, A. W. (2014) Mammals in and around suburban yards, and the attraction of chicken coops. *Urban Ecosystems***,** 1-15.

KEARNEY, M., MATZELLE, A. & HELMUTH, B. (2012) Biomechanics meets the ecological niche: the importance of temporal data resolution. *Journal of Experimental Biology,* 215**,** 922-933.

KEARNEY, M. & PORTER, W. P. (2004) Mapping the fundamental niche: physiology, climate, and the distribution of a nocturnal lizard. *Ecology,* 85**,** 3119-3131.

KEARNEY, M. & PORTER, W. P. (2009) Mechanistic niche modelling: combining physiological and spatial data to predict species' ranges. *Ecology Letters,* 12**,** 334-350.

KEARNEY, M., SHINE, R. & PORTER, W. P. (2009) The potential for behavioral thermoregulation to buffer "cold-blooded" animals against climate warming. *Proceedings of the National Academy of Sciences,* 106**,** 3835-3840.

KELLY, C. F., BOND, T. E. & HEITMAN, H. J. (1954) The role of thermal radiation in animal ecology. *Ecology,* 35**,** 562-569.

KENNEDY, A. D. & DAY, A. C. (1989) Orientation to the sun of British and exotic breed cross cows during the winter. *Applied Animal Behaviour Science,* 24**,** 115-125.

KEREN, E. N. & OLSON, B. E. (2006) Thermal balance of cattle grazing winter range: Model application. *Journal of Animal Science,* 84**,** 1238-1247.

KEVAN, P. G. & SHORTHOUSE, J. D. (1970) Behavioural thermoregulation by high arctic butterflies. *Arctic,* 23**,** 268-279.

LAKE, B. C., BERTRAM, M. R., GULDAGER, N., CAIKOSKI, J. R. & STEPHENSON, R. O. (2013) Wolf kill rates across winter in a low-density moose system in Alaska. *The Journal of Wildlife Management,* 77**,** 1512-1522.

LESKU, J. A., MEYER, L. C. R., FULLER, A., MALONEY, S. K., DELL'OMO, G., VYSSOTSKI, A. L. & RATTENBORG, N. C. (2011) Ostriches sleep like platypuses. *Plos One,* 6**,** 1-7.

LINDBERG, J. (2013) Selection of habitat and resources during migration by a large mammal. *Dissertation.* Swedish University of Agricultural Sciences, Umea, Sweden.

MACFARLANE, W. V., MORRIS, R. J. H. & HOWARD, B. (1958) Heat and water in tropical Merino sheep. *Crop and Pasture Science* 9**,** 217-228.

MACPHERSON, A. H. (1964) A Northward range extension of the red fox in the eastern Canadian arctic. *Journal of Mammalogy,* 45**,** 138-140.

MALONEY, S. K. & DAWSON, T. J. (1995) The heat load from solar radiation on a large, diurnally active bird, the emu (*Dromaius novaehollandiae*). *Journal of Thermal Biology,* 20**,** 381-387.

MALONEY, S. K., MOSS, G., CARTNELL, T. & MITCHELL, D. (2005a) Alteration in diel activity patterns as a thermoregulatory strategy in black wildebeest (*Connochaetes gnou*). *Journal of Comparative Physiology A,* 191**,** 1055-1064.

MALONEY, S. K., MOSS, G. & MITCHELL, D. (2005b) Orientation to solar radiation in black wildebeest (*Connochaetes gnou*). *Journal of Comparative Physiology A,* 191**,** 1065-1077.

MARDER, J. (1973) Body temperature regulation in the brown-necked raven (*Corvus corax ruficollis*)- II. Thermal changes in the plumage of ravens exposed to solar radiation. *Comparative Biochemistry and Physiology A,* 45**,** 431-440.

MCLACHLAN, J. S., HELLMANN, J. J. & SCHWARTZ, M. W. (2007) A framework for debate of assisted migration in an era of climate change. *Conservation Biology,* 21**,** 297-302.

MITCHELL, A. W. (1977) Preliminary observations on the daytime activity patterns of lesser kudu in Tsavo National Park, Kenya. *East African Wildlife Journal,* 15**,** 199-206.

MITCHELL, D., MALONEY, S. K., LABURN, H. P., KNIGHT, M. H., KUHNEN, G. & JESSEN, C. (1997) Activity, blood temperature and brain temperature of free-ranging springbok. *Journal of Comparative Physiology B,* 167**,** 335-343.

MORITZ, C., PATTON, J. L., CONROY, C. J., PARRA, J. L., WHITE, G. C. & BEISSINGER, S. R. (2008) Impact of a century of climate change on small-mammal communities in Yosemite National Park, USA. *Science,* 322**,** 261-264.

MOUNT, L. E. (1979) *Adaptation to thermal environment,* Edward Arnold Publishers Limited, London, UK.

MUNOZ, J. L. P., FINKE, G. R., CAMUS, P. A. & BOZINOVIC, F. (2005) Thermoregulatory behavior, heat gain and thermal tolerance in the periwinkle *Echinolittorina peruviana* in central Chile. *Comparative Biochemistry and Physiology, Part A*, 142**,** 92-98.

MUTH, A. (1977) Thermoregulatory postures and orientation to the sun: A mechanistic evaluation for the zebra-tailed lizard, *Callisaurus draconoides*. *Copeia,* 1977**,** 710-720.

NEI, M. (1973) Analysis of gene diversity in subdivided populations. *Proceedings of the National Academy of Sciences of the United States of America*, 70**,** 3321-3323.

NORRIS, A. L. & KUNZ, T. H. (2012) Effects of solar radiation on animal thermoregulation. IN BABATUNDE, E. B. (Ed.) *Solar radiation.* 195-220, InTech (www.intechopen.com).

PARK, K. J., JONES, G. & RANSOME, R. D. (2000) Torpor, arousal and activity of hibernating Greater Horseshoe Bats (*Rhinolophus ferrumequinum*). *Functional Ecology,* 14**,** 580-588.

PARK, S. D. E. (2001) Trypanotolerance in West African cattle and the population genetic effects of selection. *Doctoral dissertation.* University of Dublin, Dublin, UK.

PARMESAN, C. (2006) Ecological and evolutionary responses to recent climate change. *Annual Review of Ecology, Evolution, and Systematics,* 37**,** 637-669.

PARMESAN, C., RYRHOLM, N., STEFANESCU, C., HILL, J. K., THOMAS, C. D., DESCIMON, H., HUNTLEY, B., KAILA, L., KULLBERG, J., TAMMARU, T., TENNENT, W. J., THOMAS, J. A. & WARREN, M. (1999) Poleward shifts in geographical ranges of butterfly species associated with regional warming. *Nature,* 399**,** 579-583.

PARMESAN, C. & YOHE, G. (2003) A globally coherent fingerprint of climate change impacts across natural systems. *Nature,* 421.

PHILLIPS, P. K. & HEATH, J. E. (1995) Dependancy of surface temperature regulation on body size in terrestrial mammals. *Journal of Thermal Biology,* 20**,** 281-289.

POST, E., PETERSON, R. O., STENSETH, N. C. & MCLAREN, B. E. (1999) Ecosystem consequences of wolf behavioural response to climate. *Nature,* 401**,** 905-907.

QUINTERO, I. & WIENS, J. J. (2013) Rates of projected climate change dramatically exceed past rates of climatic niche evolution among vertebrate species. *Ecology Letters,* 16**,** 1095-1103.

RAMSDELL, K. D. (2013) Diel breeding patterns of the Oregon Spotted Frog (*Rana pretiosa*). *Master's dissertation.* The Evergreen State College, Olympia, WA, USA.

RAXWORTHY, C. J., PEARSON, R. G., RABIBISOA, N., RAKOTONDRAZAFY, A. M., RAMANAMANJATO, J. B., RASELIMANANA, A. P., WU, S., NUSSBAUM, R. A.

& STONE, D. A. (2008) Extinction vulnerability of tropical montane endemism from warming and upslope displacement: a preliminary appraisal for the highest massif in Madagascar. *Global Change Biology,* 14**,** 1703-1720.

REALE, D., MCADAM, A. G., BOUTIN, S. & BERTEAUX, D. (2003) Genetic and plastic responses of a northern mammal to climate change. *Proceedings of the Royal Society of London B,* 270**,** 591-596.

REFSNIDER, J. M. & JANZEN, F. J. (2012) Behavioiural plasticity may compensate for climate change in a long-lived reptile with temperature-dependent sex determination. *Biological Conservation,* 152**,** 90-95.

RENECKER, L. A. & HUDSON, R. J. (1986) Seasonal energy expenditures and thermoregulatory responses of moose. *Canadian Journal of Zoology,* 64**,** 322-327.

RICCIARDI, A. & SIMBERLOFF, D. (2009) Assisted colonization is not a viable conservation strategy. *Trends in Ecology and Evolution,* 24**,** 248-253.

ROBERTSHAW, D. & DMI'EL, R. (1983) The effect of dehydration on the control of panting and sweating in the black bedouin goat. *Physiological Zoology,* 56**,** 412-418.

ROBERTSHAW, D. & TAYLOR, C. R. (1969) A comparison of sweat gland activity in eight species of east african bovids. *Journal of Physiology* 203**,** 135-143.

ROPERT-COUDERT, Y., BEAULIEU, M., HANUISE, N. & KATO, A. (2009) Diving into the world of biologging. *Endangered Species Research,* 10**,** 21-27.

ROSENZWEIG, C., KAROLY, D., VICARELLI, M., NEOFOTIS, P., WU, Q., CASASSA, G., MENZEL, A., ROOT, T. L., ESTRELLA, N., SEGUIN, B., TRYJANOWSKI, P., LIU, C., RAWLINS, S. & IMESON, A. (2008) Attributing physical and biological impacts to anthropogenic climate change. *Nature,* 453**,** 353-358.

ROSS, J., HEARN, A. J. & MACDONALD, D. W. (2013) Recent camera-trap records of Malay Weasel *Mustela nudipes* in Sabah, Malaysian Borneo. *Small Carnivore Conservation,* 49**,** 20-24.

RUTZ, C. & HAYS, G. C. (2009) New frontiers in biologging science. *Biology Letters,* 5**,** 289-292.

SANPARKS (2012) Mokala National Park offical website. *http://sanparks.org.za/parks/mokala/all.php*, 6 March 2012.

SCHMIDT-NIELSON, K. (1997) *Animal physiology: Adaptation and environment (5th ed.),* Cambridge University Press, New York, NY, USA.

SESSLER, D. I., MOAYERI, A., STOEN, R., GLOSTEN, B., HYNSON, J. & MCGUIRE, J. (1990) Thermoregulatory vasoconstriction decreases cutaneous heat loss. *Anesthesiology,* 73**,** 656-660.

SHRESTHA, A. K., WIEREN, S. E., VAN LANGEVELDE, F., FULLER, A., HETEM, R. S., MEYER, L. C. R., DE BIE, S. & PRINS, H. H. T. (2013) Larger antelopes are sensitive to heat stress throughout all seasons but smaller antelopes only during summer in an African semi-arid environment. *International Journal of Biometeorology,* 58**,** 41-49.

SIGNER, C., RUF, T. & ARNOLD, W. (2010) Hypometabolism and basking: the strategies of Alpine ibex to endure harsh over-wintering conditions. *Functional Ecology***,** 1-11.

SILVER, S. C., OSTRO, L. E. T., MARSH, L. K., MAFFEI, L., NOSS, A. J., KELLY, M. J., WALLACE, R. B., GOMEZ, H. & AYALA, G. (2004) The use of camera traps for estimating jaguar *Panthera onca* abundance and density using capture/recapture analysis. *Oryx,* 38**,** 148-154.

SKELLY, D. K. & FREIDENBURG, L. K. (2010) Evolutionary responses to climate change. *Encyclopedia of Life Sciences.* John Wiley & Sons, Ltd, Chichester (www.els.net).

SKINNER, J. D. & CHIMIMBA, C. T. (2005) *The mammals of the southern African subregion*, Cambridge University Press, Cape Town, South Africa.

SPENCER, C. C., NEIGEL, J. E. & LEBERG, P. L. (2000) Experimental evaluation of the usefulness of microsatellite DNA for detecting demographic bottlenecks. *Molecular Ecology,* 9**,** 1517-1528.

STIRLING, I., LUNN, N. J. & IACOZZA, J. (1999) Long-term trends in the population ecology of polar bears in western Hudson Bay in relation to climatic change. *Arctic,* 52**,** 294-306.

TANNERFELDT, M., ELMHAGEN, B. & ANGERBJORN, A. (2002) Exclusion by interference competition? The relationship between red and arctic foxes. *Oecologia,* 132**,** 213-220.

TAYLOR, A. R. & KNIGHT, R. L. (2003) Wildlife responses to recreation and associated visitor perceptions. *Ecological Society of America,* 13**,** 951-963.

TAYLOR, C. R. (1970) Dehydration and heat: effects on temperature regulation of East African ungulates. *American Journal of Physiology,* 219**,** 1136-1139.

TAYLOR, C. R., ROBERTSHAW, D. & HOFMANN, R. (1969) Thermal panting: a comparison of wildebeest and zebu cattle. *American Journal of Physiology,* 217**,** 907-910.

THOMAS, C. D., CAMERON, A., GREEN, R. E., BAKKENES, M., BEAUMONT, L. J., COLLINGHAM, Y. C., ERASMUS, B. F. N., DE SIQUEIRA, M. F., GRAINGER, A., HANNAH, L., HUGHES, L., HUNTLEY, B., VAN JAARSVELD, A. S., MIDGELEY, G. F., MILES, L., ORTEGA-HUERTA, M. A., PETERSON, A. T., PHILLIPS, O. L. & WILLIAMS, S. E. (2004) Extinction risk from climate change. *Nature,* 427**,** 145-148.

TREMBLAY, Y., THIEBAULT, A., MULLERS, R. & PISTORIUS, P. (2014) Bird-borne video-cameras show that seabird movement patterns relate to previously unrevealed proximate environment, not prey. *Plos One,* 9**,** 1-6.

UMINA, P. A., WEEKS, A. R., KEARNEY, M. R., MCKECHNIE, S. W. & HOFFMANN, A. A. (2005) A rapid shift in a classic clinal pattern in drosophila reflecting climate change. *Science,* 308**,** 691-693.

VAN BEEST, F. M., VAN MOORTER, B. & MILNER, J. M. (2012) Temperature-mediated habitat use and selection by a heat-sensitive northern ungulate. *Animal Behaviour,* 84**,** 723-735.

VAN HEERWAARDEN, B. & HOFFMANN, A. A. (2007) Global warming: fly populations are responding rapidly to climate change. *Current Biology,* 17**,** 16-18.

VERNON, H. M. (1930) The measurement of radiant heat in relation to human comfort. *Journal of Physiology (London),* 70**,** 15-17.

VERNON, H. M. (1933) The estimation of solar radiation in relation to its warming effect on the human body. *Quarterly Journal of the Royal Meteorological Society,* 56**,** 239-252.

VRAHIMIS, S. & KOK, O. B. (1992) Body orientation of black wildebeest in a semi-arid environment. *African Journal of Ecology,* 30**,** 169-175.

VRAHIMIS, S. & KOK, O. B. (1993) Daily activity of black wildebeest in a semi-arid environment. *African Journal of Ecology,* 31**,** 328-336.

WALSBERG, G. E. (1990) Convergence of solar heat gain in two squirrel species with contrasting coat colors. *Physiological Zoology,* 63**,** 1025-1042.

WALSBERG, G. E. (1992) Quantifying radiative heat gain in animals. *American Zoology,* 32**,** 217-224.

WALSBERG, G. E. & SCHMIDT, C. A. (1989) Seasonal adjustment of solar heat gain in a desert mammal by altering coat properties independently of surface coloration. *Journal of Experimental Biology,* 142**,** 387-400.

WARREN, M. S., HILL, J. K., THOMAS, J. A., ASHER, J., FOX, R., HUNTLEY, B., ROY, D. B., TELFER, M. G., JEFFCOATE, S., HARDING, P., JEFFCOATE, G., WILLIS, S. G., GREATOREX-DAVIES, J. N., MOSS, D. & THOMAS, C. D. (2001) Rapid responses of British butterflies to opposing forces of climate and habitat change. *Nature,* 414**,** 65-69.

WHITE, C. R. & SEYMOUR, R. S. (2003) Mammalian basal metabolic rate is proportional to body mass. *Proceedings of the National Academy of Sciences of the United States of America,* 100**,** 4046-4049.

WHYTE, B. I., ROSS, J. G. & BLACKIE, H. M. (2013) Differences in brushtail possum home-range characteristics among sites of varying habitat and population density. *Wildlife Research,* 40**,** 537-544.

WILLIAMSON, D., WILLIAMSON, J. & NGWAMOTSOKO, K. T. (1988) Wildebeest migration in the Kalahari. *African Journal of Ecology,* 26**,** 269-280.

WILSON, R. P., GRANT, W. S. & DUFFY, D. C. (1986) Recording devices on free-ranging marine animals: does measurement affect foraging performance? *Ecology,* 67**,** 1091-1093.

WILSON, R. P., GREMILLET, D., SYDER, J., KIERSPEL, M. A. M., GARTHE, S., WEIMERSKIRCH, H., SCHAFER-NETH, C., SCOLARO, J. A., BOST, C. A., PLOTZ, J. & NEL, D. (2002) Remote-sensing systems and seabirds: their use, abuse and potential for measuring marine environmental variables. *Marine ecology progress series,* 228**,** 241-261.

WILSON, R. P., LIEBSCH, N., DAVIES, I. M., QUINTANA, F., WEIMERSKIRCH, H., STORCH, S., LUCKE, K., SIEBERT, U., ZANKL, S., MULLER, G., ZIMMER, I., SCOLARO, A., CAMPAGNA, C., PLOTZ, J., BORNEMANN, H., TEILMANN, J. & MCMAHON, C. R. (2007) All at sea with animal tracks; methodological and analytical solutions for the resolution of movement. *Deep-Sea Research II,* 54**,** 193-210.

WILSON, R. P., SHEPARD, E. L. C. & LIEBSCH, N. (2008) Prying into the intimate details of animal lives: use of a daily diary on animals. *Endangered Species Research,* 4**,** 123-137.

Appendix A - Pilot study

Methods

A pilot study was conducted during 4 days in March and April 2011 to test the possibility of using a remote technique to quantify orientation to solar radiation in animals. A barrel was used to represent the body of a wildebeest. Miniature temperature data loggers were attached bilaterally to the sides of the barrel at three positions (below mid-line, mid-line and above mid-line) and the barrel was covered with a blue wildebeest pelt (Figure 1). This model animal was rotated at regular time intervals (15, 30, 60 and 120 minutes) throughout a ten hour day and orientated either perpendicular or parallel to solar radiation.

Results

An example of the results obtained from one of these days is shown in Figure 2. In the shown example, when the model was orientated perpendicular to solar radiation, the temperature increased on the side facing the sun, while the temperature dropped on the side facing away from the sun. When the model was orientated parallel to solar radiation the temperatures merged until both sides had a similar temperature reading.

The position of the loggers on the animal's body had an effect on the temperature patterns (Figure 3). Figure 3.a depicts the temperature readings of a logger attached to the barrel above the mid-line, whereas Figure 3.b shows temperature readings from a logger attached below the mid-line of the barrel. In Figure 3.a the difference between left and right temperatures when the animal was standing parallel to solar radiation was greater compared to in Figure 3.b.

Discussion and conclusion

When the loggers were located below the mid-line it was shaded by the barrel when the sun was directly overhead during the middle of the day, whereas the logger attached above the mid-line was always in the sun during the middle of the day irrespective of how the animal's body was orientated. Because the data loggers attached below the mid-line showed a smaller difference in temperature when the model animal was orientated parallel to solar radiation than the loggers attached above the mid-line, the temperature patterns from the data logger attached below the mid-line showed a clearer difference between perpendicular orientation to solar radiation and parallel orientation to solar radiation. Based on these results, we decided to implant the loggers into the live wildebeest just below the mid-line.

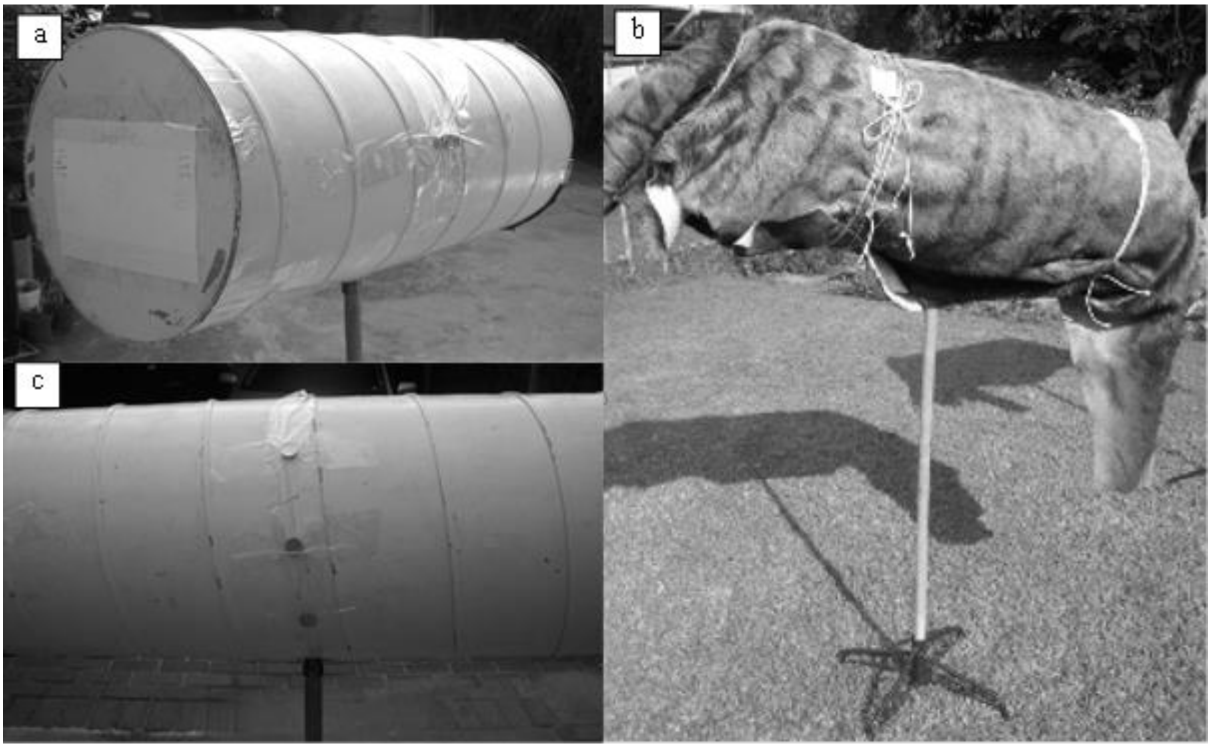


Figure 1 The set-up for the pilot study. A barrel (a) mounted on a stick and stand was covered with a blue wildebeest pelt to represent a wildebeest (b). Temperature-sensitive data loggers were attached in three positions (above, on and below the mid-line) to the barrel underneath the blue wildebeest pelt (c). The model animal was placed out in the sun and rotated at 15, 30, 60 and 120 minute intervals to face different orientations relative to the sun.

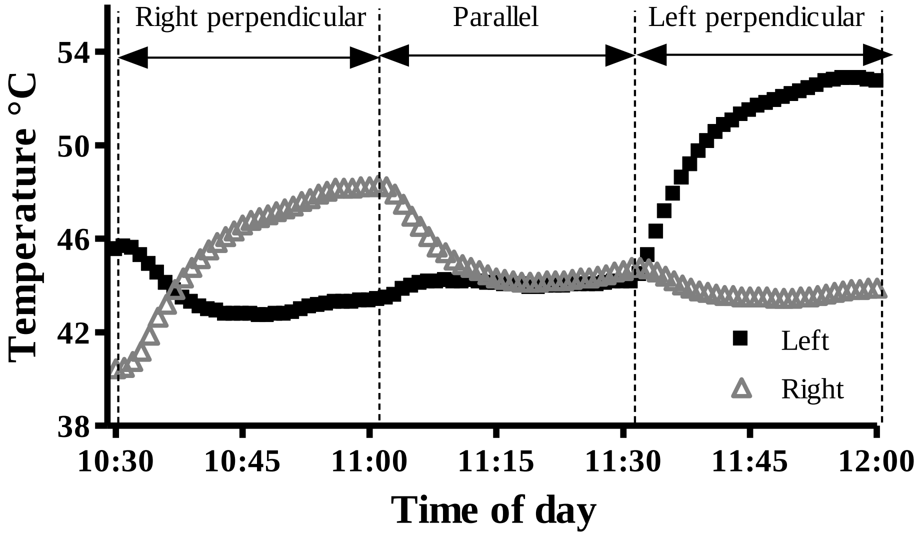


Figure 2 Left and right temperatures of model wildebeest orientated right perpendicular, parallel and left perpendicular to solar radiation.

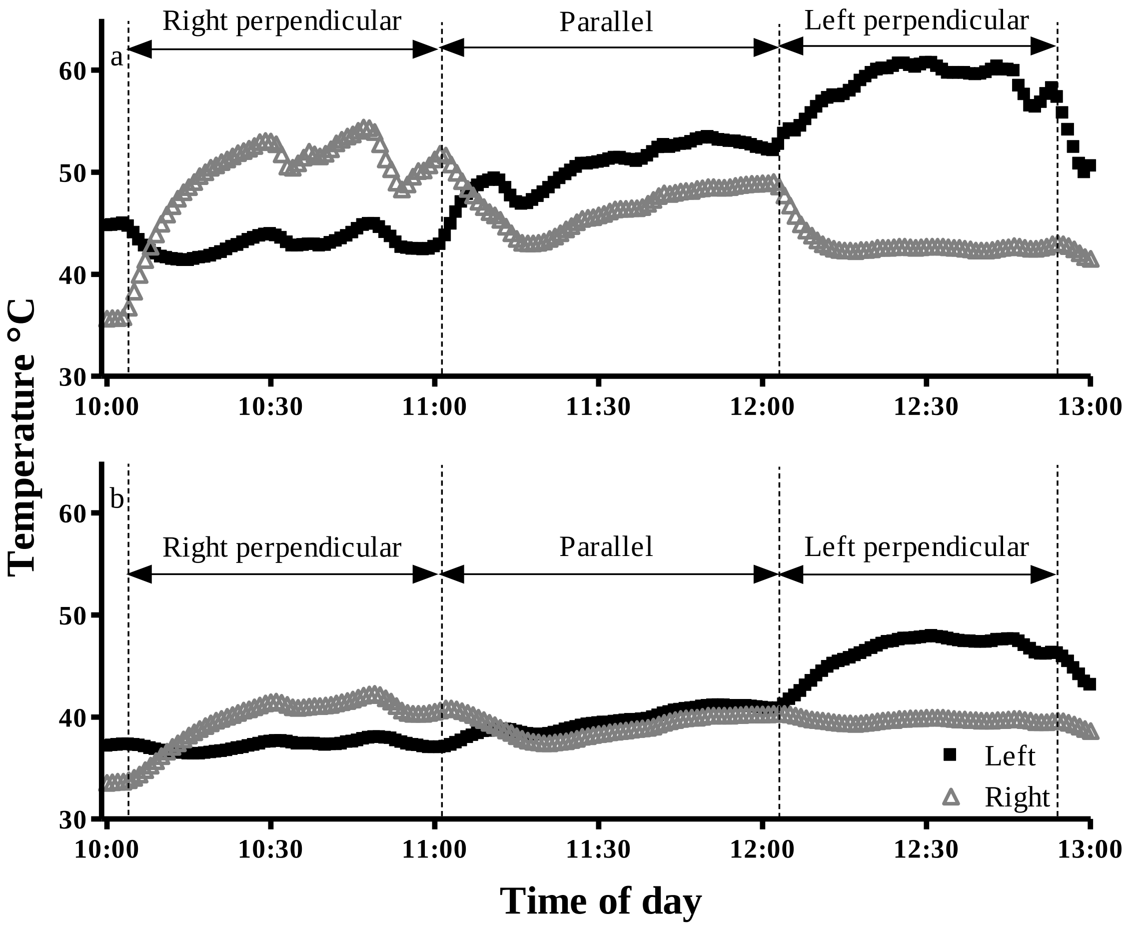


Figure 3 Left and right temperatures of the model wildebeest orientated right perpendicular, parallel and left perpendicular to solar radiation with the temperature logger attached above (a) and below (b) the mid-line of the barrel.

Appendix B - Wildebeest DNA typing results

This document was compiled by Dr Desiré lee Dalton from the National Zoological Gardens and edited by myself to complement the rest of the dissertation. The genetic tests were conducted to determine whether the black and blue wildebeest of Mokala National Park were hybridising. Similar tests have been conducted in previous studies to determine the genetic purity of wildebeest populations (Grobler and Van der Bank, 1993, Grobler and Van der Bank, 1995, Grobler et al., 2005).

Results

Wildebeest typing

Enquiries: Dr Desiré lee Dalton
Email: desire@nzg.ac.za
Tel: +27 12 328 3265

Date: 04 May 2011

For Attention: Dr. Hilary Lease

Results: Wildebeest DNA typing:

The National Zoological Gardens of South Africa (NZG) is currently conducting projects on the genetic diversity, inbreeding and relatedness in a variety of wildlife species. In order to conduct this research, different genetic markers are being used. Microsatellite genotyping of the wildebeest population was conducted using 6 cross species markers (loci). Each parent transmits one allele (copy) to his offspring; therefore each individual presents 2 alleles per locus. Within the same species one locus could present several alleles. In this way, microsatellites are ideal for determining paternity, population genetics, hybridisation and genetic variability.

Herewith are the results of the samples received in April 2011.

According to the DNA marker set typed (Table 1.), as compiled through research for the detection of possible hybrid animals the following result was obtained from an assignment test for blue and black wildebeest.

The marker test indicates the following samples are NOT possible hybrids as they were assigned only to Black Wildebeest: BW284-292.

Statistical analysis

This section presents a motivation for the analyses, describes the analytical methods and software employed, presents and explains results, and discusses the implications of results.

Rational for measuring genetic diversity

Genetic diversity provides the basis for adaptability (through natural selection) as environmental conditions change. If a population lacks genetic diversity, it is in great danger of lacking the resources to survive environmental change. Low levels of genetic diversity may also lead to the expression of deleterious recessive alleles. In practice, loss of genetic diversity (or "inbreeding") may result in reduced survival, reproductive abnormalities, juvenile mortalities, physical deformities and reduced growth in populations. It is therefore important to conserve representative levels of genetic diversity in artificially managed populations.

Genetic diversity may be lost through events such as (i) genetic bottlenecks, which occur when a population is reduced to a few reproducing individuals which offspring then increase in numbers to re-establish the population; (ii) founder events, when a population is started (or founded) using a small number of individuals which may not contain the full range of

Table 1 Results of DNA marker test for detection of black and blue wildebeest hybridization.

Lab Nr.	Sample Nr.	Results of Marker 1		Results of Marker 2		Results of Marker 3		Results of Marker 4		Results of Marker 5		Results of Marker 6		Assignment
BW284	7	129	129	117	117	198	200	202	202	106	135	246	246	Black wildebeest alleles only
BW285	8	129	129	117	117	198	200	202	202	106	139	230	230	Black wildebeest alleles only
BW286	9	129	129	117	117	198	200	202	204	106	139	228	230	Black wildebeest alleles only
BW287	10	129	129	117	117	194	200	202	204	106	135	230	246	Black wildebeest alleles only
BW288	12	129	129	117	117	198	200	202	204	106	129	230	246	Black wildebeest alleles only
BW289	13	129	129	117	117	198	200	202	202	139	145	228	230	Black wildebeest alleles only
BW290	14	129	129	117	117	196	200	202	202	135	139	230	230	Black wildebeest alleles only
BW291	15	129	129	117	117	198	198	204	204	106	129	228	246	Black wildebeest alleles only
BW292	16	129	129	117	117	200	200	202	202	129	135	230	230	Black wildebeest alleles only
BW293	17													To repeat

variation that occurs in the species; (iii) isolation and lack of migration; and (iv) small population size.

Calculation of genetic diversity values:

Genetic diversity in populations is commonly quantified using three measures:

- Average heterozygosity: this measure was formulated by (Nei, 1973) and is probably the most widely used measure to quantify genetic diversity across loci in populations. Possible values of heterozygosity range from zero (no variation) to just below one.
- Average number of alleles per locus: this refers to the average number of alleles per locus across all loci screened in a population. The number of alleles per locus is thus a mathematical average of the number of alleles at each locus, in each population. The lowest possible value for number of alleles is zero, typically increasing to 10-15 or more in highly polymorphic populations.

During the current project, these coefficients of genetic diversity within populations were calculated using MSToolkit (Park, 2001), an add-in for Excel software. (MSToolkit was also used to prepare or initiate input files for all other software used).

Results and discussion for genetic diversity

A summary of results regarding the genetic diversity for the wildebeest is presented in Table 2.

Table 2 Genetic diversity of wildebeest population.

Population	Heterozygosity ± SD	Number of alleles ± SD
Wildebeest	0.432 ± 0.1411	2.83 ±1.60

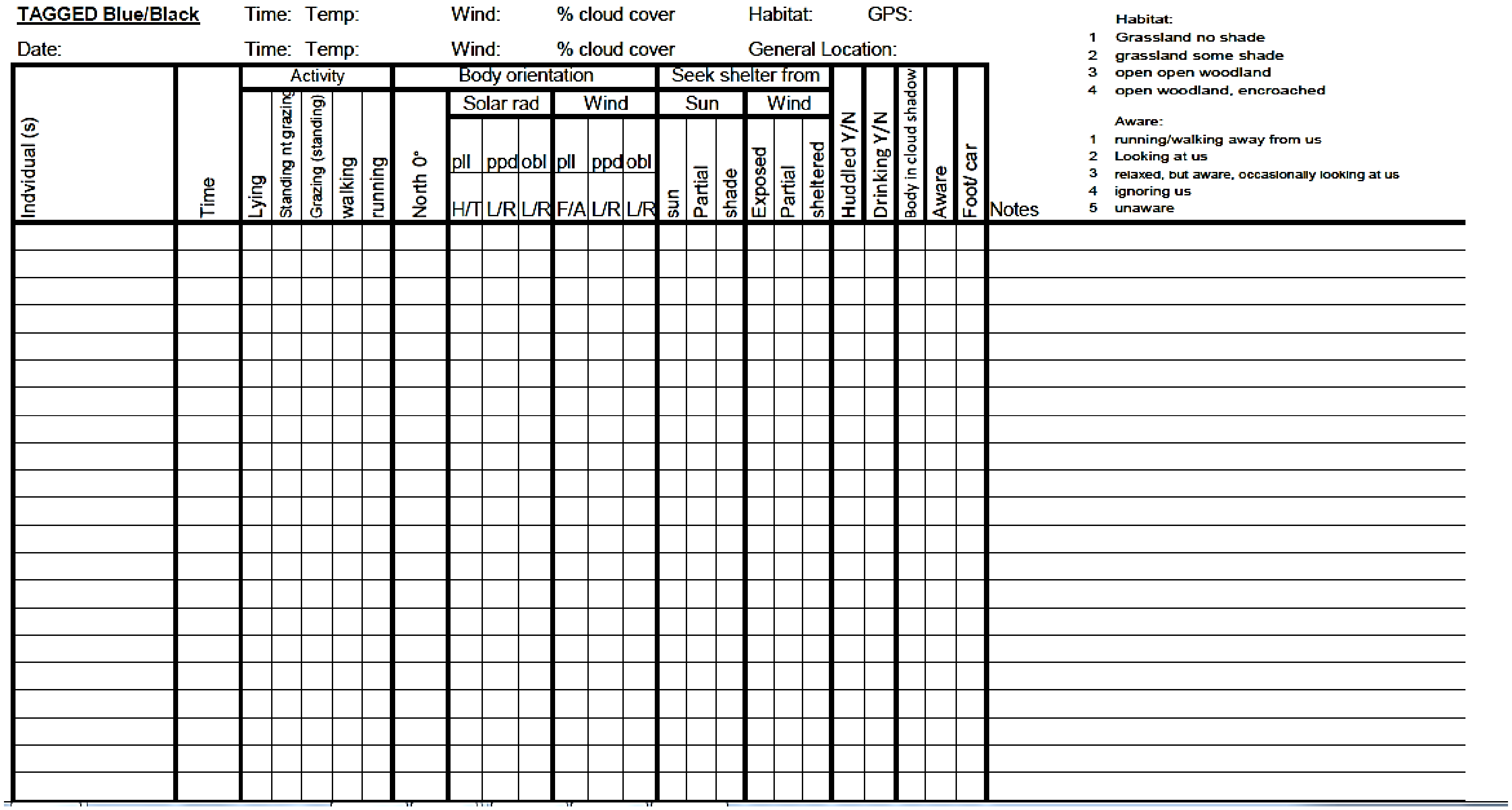
The level of average heterozygosity for the wildebeest is 0.4732. This is lower than results obtained for other wildebeest (0.37-0.50) from the Free State, Groote Schuur Estate, Benfontein and Bloem dam. The average number of alleles per locus is, according to some authors (Spencer et al., 2000) a more powerful indicator of bottlenecks and loss of diversity compared to average heterozygosity. The number of alleles per locus (2.83) is similar to results obtained from other wildebeest studies (2.5 - 3.33), which suggests that this population has sufficient genetic diversity and is currently not at risk for inbreeding.

We thank you for your support and hope to be of service in the future.

Yours sincerely,
Dr Desiré Lee Dalton

Appendix C - Behavioural sheet

The behaviour sheet used during the behavioural observations to determine orientation to solar radiation of free-ranging wildebeest for this study was also used in a parallel study on the same species. Therefore, some of the variables mentioned in the table were not discussed in this dissertation.



TAGGED Blue/Black Time: Temp: Wind: % cloud cover Habitat: GPS:

Date: Time: Temp: Wind: % cloud cover General Location:

		Activity					Body orientation							Seek shelter from											
								Solar rad			Wind			Sun			Wind								
								pll	ppd	obl	pll	ppd	obl												
Indvidual (s)	Time	Lying	Standing nt grazing	Grazing (standing)	walking	running	North 0°	H/T	L/R	L/R	F/A	L/R	L/R	sun	Partial	shade	Exposed	Partial	sheltered	Huddled Y/N	Drinking Y/N	Body in cloud shadow	Aware	Foot/ car	Notes

Habitat:

1 Grassland no shade
2 grassland some shade
3 open open woodland
4 open woodland, encroached

Aware:

1 running/walking away from us
2 Looking at us
3 relaxed, but aware, occasionally looking at us
4 ignoring us
5 unaware