

CHAPTER 1:

INTRODUCTION

1. INTRODUCTION

1.1 Capture of game animals

Capture is a necessary practice in the management of wildlife populations, as the need often arises to translocate, handle or treat wild animals in the field. Wild animals may also be captured for scientific purposes, for example, animals may be radio-collared for monitoring of seasonal movements (Jacques et al., 2009). Although, there have been advancements and refinements in the techniques of capture and the drugs used for darting since the 1960's (where only physical restraint with a rope was used), there still is a significant risk of morbidity and mortality associated with capture of wild animals (Kock and Meltzer, 2006).

The process of capture, over the last 30 years, has undergone several changes and improvements, as techniques and knowledge have improved. Today there is a variety of capture techniques to suit the species captured, terrain, number of animals caught and subsequent transport of captured animals. Despite the availability of different capture methods, capture is a stressful procedure and is associated with animal distress and frequently morbidity and mortality (Ganhao et al., 1988).

A common practice in capture is to immobilise animals with the use of darting drugs. The drugs immobilise the animal by disrupting signalling between synapses in the central nervous system. Immobilising drugs can be, and often are, combined with a tranquiliser or sedative such as azaperone, xylazine, medetomidine, diazepam and midazolam (Kock and

Meltzer, 2006). Different drugs, drug combinations and drug doses are used for different species. Common immobilising drug groups are the opioids, neuromuscular blockers and cyclohexylamines. Of the opioids, etorphine is one of the most widely used drugs and is commonly used in ungulates, elephants and rhinoceroses (Swan, 1993; Meltzer et al., 2006b). Thiofentanil, also an opioid, has shown to be effective in immobilising specific ungulate species such as kudu, nyala and waterbuck (Meltzer et al., 2006b). The advantage of thiofentanil is that it has a shorter induction time (the time that the animal takes to be immobilised after it has been darted) than does etorphine (Meltzer et al., 2006b). Carnivores, reptiles, birds and primates are not immobilised with opioids (as this can cause excitement, convulsions and respiratory depression). Instead, the cyclohexylamine group of drugs are used for immobilisation. Of the cyclohexylamines, ketamine and tiletamine are most commonly used for immobilisation (Swan, 1993; Burroughs et al., 2006). Neuromuscular blocking agents, such as gallamine, are not commonly used for immobilisation because they do not cause anaesthesia or analgesia, but rather muscle paralysis. This paralysis may include the paralysis of respiratory muscles, which can cause respiratory depression. However, gallamine is used for the immobilisation of crocodiles because this drug is well tolerated (has a wide therapeutic index) in this species (Swan, 1993; Meltzer et al., 2006b).

Capture does not necessarily require chemical immobilisation, but can also be done by using physical restraint, confinement or mass capture techniques. Physical restraint could include physical force to hold on an animal, but is only used if an animal is small enough to do so safely (Atkinson et al., 2006). Large carnivores, such as leopards and cheetahs, are usually first baited into cage traps and then chemically immobilised either by pole syringes or darts

(Burroughs et al., 2006). Animals can also be chased into nets or baited into an area where a net will be dropped on them. Small carnivores, such as African wild cat, are first caught in nets and then injected with an immobilising agent by hand (Burroughs et al., 2006). Lastly, physical barriers are often used to direct animals into a temporary enclosure or vehicle after they have been driven, in that direction, using a helicopter, motor vehicle or motorcycle (Atkinson et al., 2006). Once animals have been captured or confined, they can be placed into bomas for housing, or in crates for transport, but it is important to note that any type of confinement will cause chronic stress in a wild animal (Atkinson et al., 2006). Capture of an animal also causes stress, but this stress is intense and usually of an acute nature. The most appropriate method of capture will differ for different species and terrain but the ultimate aim of capture is to do so as quickly as possible to minimise stress and the adverse physiological changes associated with stress (Foster, 2005).

The stress associated with capture causes an increase in energy metabolism (Ganhao et al., 1988). This increase in energy metabolism helps an animal escape, but may cause a homeostatic imbalance in the animal, which may alter normal physiological functions, such as temperature regulation, acid-base balance, reproduction, tissue growth and maintenance and immunity (Kock et al., 1987a, Cattet et al., 2003).

Because of the disturbance of normal physiological function during capture, animals face an increased risk of morbidity and mortality. Animals can also physically injure themselves in their attempts to escape being captured. Injuries, usually from by running into objects, can result in severe bone fractures and open wounds (Kock and Meltzer, 2006). Respiratory function of animals can also be negatively altered by capture-drugs. Opioids can cause

respiratory depression resulting in hypercapnia, hypoxia and an acidosis (Swan, 1993; Kock and Meltzer, 2006). Bloat is also a possible consequence when ungulates are captured using chemical means. Some drugs decrease rumen movement and eructation and thus fermenting gas in a ruminant's stomach is unable to escape. The increased pressure from the fermenting gas expands the stomach and decreases the space available for lung expansion, which can inhibit breathing and cause cardiovascular collapse (Kock and Meltzer et al., 2006). In animals, such as ruminants, carnivores, primates and pigs, chemical immobilisation can also cause vomiting or regurgitation, which may result in aspiration pneumonia (Kock and Meltzer et al., 2006). An animal's normal body temperature also is likely to be altered because of capture. The increased body temperature is believed to be caused by ambient temperature, stress, exertion and the disruption of thermoregulation through drugs, which can cause hyperthermia and an acidosis. However, if ambient temperatures are too low (especially in winter) animals can also experience hypothermia, particularly after immobilisation (Kock et al., 1987; Kock and Meltzer, 2006).

Capture myopathy is a pathology that is often observed after capture, which also contributes to the morbidity and mortality risk of a captured animal. Capture myopathy is a syndrome of homeostatic imbalance and is thus exacerbated and encompassed by physiological changes in the acid-base balance, tissue injury, body temperature and changes in enzyme levels (Montane et al., 2003). Prior to the 1960s (and before the onset of capture drug use) mortality rates from capture myopathy were as high as 50% (Kock and Meltzer, 2006). The primary cause of capture myopathy is the damage to the muscle tissue as a consequence of overexertion and the subsequent biochemical changes (Montane et al., 2003; Kock and Meltzer, 2006). One of the biochemical changes that occurs after

overexertion is the elevation of enzyme levels. Elevated levels of enzymes such as creatine phosphokinase, aspartate aminotransferase and lactate dehydrogenase are commonly seen in animals affected by capture myopathy (Kock et al., 1987). Skeletal and cardiac muscle damage results in an increase in myoglobin and lactic acid in the blood, which leads to a decrease in pH and kidneys and liver damage. Animals usually die acutely from capture myopathy but animals may die up to two months after the capture event (Meltzer et al., 2006b).

1.2 Capture-induced hyperthermia

Capture-induced hyperthermia is dangerous side-effect of capture that may be a major cause of capture myopathy. Hyperthermia is “the condition of a temperature regulator when core temperature is above its range specified for the normal active state of the species” (The Commission for Thermal Physiology of the International Union of Physiological Sciences, 2001). Capture-induced hyperthermia, specifically, is the body temperature increase, above a normal regulated range for a species, during the capture process.

1.2.1 Pathophysiology of hyperthermia

Hyperthermia is distinctly different from fever because in fever the set-point for a normal body temperature is increased in the hypothalamus and thermoregulatory mechanisms are stimulated to increase body temperature, whereas in hyperthermia the body's thermoregulatory mechanisms are unable to maintain the body temperature at the normal set-point (Harvey, 1993). Depending on the cause, the magnitude of the temperature

increase, and the severity of adverse effects, the type of hyperthermia can manifest as various differing heat stress syndromes, such as, malignant hyperthermia (a metabolic disorder that is induced by specific anaesthetic agents), heat exhaustion (a body temperature disorder associated with the depletion of water and electrolytes) and heat stroke (Knochel, 1989; Harvey, 1993).

Of the heat illnesses, in humans, heat stroke is the most dangerous and is considered a medical emergency (Harvey, 1993). Heat stroke, a pathological syndrome, is characterised in humans by a core body temperature increase above 40°C with central nervous system dysfunction, such as delirium or coma (Harvey, 1993), and occurs when normal physiological mechanisms fail to dissipate sufficient heat to overcome the heat gained either from the environment or from intense exercise (Bouchama and Knochel, 2002). Heat stroke is more likely to develop if underlying diseases (heart disease or neurological disorders) or predisposing factors (dehydration or impaired consciousness) are present that will compromise cardiac output or thermoregulatory mechanisms, such as sweating and cutaneous vasodilation (Bouchama and Knochel, 2002). Clinical features of heat stroke include hypotension, hyperventilation, altered electrolytes and changes in the pH of the blood, which may progress to multi-organ failure if heat stroke is inadequately treated (Harvey, 1993).

In heat stroke, multi-organ damage results from the complex interplay of systemic inflammation, coagulation, blood circulatory changes, oxidative damage and cytotoxicity (direct cell damaging effect) caused by a severely increased body temperature (Bouchama and Knochel, 2002; Yan et al., 2006). Cells are directly damaged by periods of heat stress. A

body temperature of greater than 40°C can alter cell membrane permeability, increase the degradation of intracellular proteins, inhibit DNA synthesis and disrupt the cytoskeleton (Yan et al., 2006). Organs, such as the kidneys, liver and intestines, are prone to be damaged during periods of heat stress because the blood supply to these organs is shunted to the periphery to increase heat loss (Knochel, 1989; Bouchama and Knochel, 2002). That is, to increase the rate of sweating, radiation and evaporative heat loss, blood is shunted from the viscera to the skin. To increase the amount of blood flow to the periphery heart rate also increases and thus tachycardia is a common clinical feature in heat stroke (Knochel, 1989; Harvey, 1993). With an increase in body temperature there is also an increase in the oxygen consumption and metabolic rate of tissues, but because of the decreased blood flow to organs a subsequent hypoxia may result in the organs (Harvey, 1993; Bouchama and Knochel, 2002). The hypoxia may also then result in the production of radicals that further injure tissues by oxidation (Bouchama and Knochel, 2002). If hypoxia and tissue injury occur in the brain then sleepiness, confusion, delirium and coma may develop as the body temperature increases (Harvey, 1993). During heat stress the intestines become more permeable because of local tissue injury and increased pro-inflammatory cytokines may be produced. In many species, including rats and primates, it has been shown that endotoxins and bacteria are able to move from within the gut into the general circulation because of the increased gut permeability during heat stress (Bouchama and Knochel, 2002). The endotoxins may further aggravate hypotension resulting in circulatory shock (Yan et al., 2006). During periods of heat stress coagulation is activated because of decreased circulating anti-clotting factors and thus heat stroke can also be characterised by diffuse microvascular blood clots (Bouchama and Knochel, 2002). Changes in pH and electrolytes may also result in muscle tissue damage (rhabdomyolysis); this is more common in

hyperthermia that is caused by extreme exertion. Rhabdomyolysis may then result in the release of myoglobin into the blood (Harvey, 1993). The circulating myoglobin then may contribute to worsening kidneys dysfunction and may contribute to causing acute renal failure in heat stroke (Bouchama and Knochel, 2002; Kosaka et al., 2004).

Capture-induced hyperthermia is not identical to heat stroke because there is also a psychological stress component that contributes to the capture-induced hyperthermia. Also, heat stroke, that is caused by high environment temperatures, may develop over two or more days of exposure to the high environmental temperatures (Knochel, 1989), whereas capture-induced hyperthermia develops acutely during the process of capture. However both these conditions may share something in common. Heat stroke can also be caused by intense exercise in hot and humid conditions (Knochel, 1989), and capture-induced hyperthermia can be compounded by intense exercise during a chase. This is because in both situations there is an increased heat gain to the animal through the increased metabolic activity of the muscles during extreme exertion. This added heat would normally be dissipated by heat loss mechanisms (cutaneous vasodilation and sweating) but in hot humid conditions these mechanisms may be insufficient and a rise in core body temperature occurs (Harvey, 1993; Kock and Meltzer, 2006). Moreover, it is likely that the pathophysiological effects on the body from heat stroke and capture-induced hyperthermia may be similar because of the effect of the increased body temperature that occurs in both events.

1.2.2 Consequences of hyperthermia for animal welfare

The magnitude and the duration of the capture-induced hyperthermia are both detrimental to the animal and can produce organ damage, rhabdomyolysis, alterations in electrolyte balance, increased oxidative stress and may result in death (Kosaka et al., 2004; Kock and Meltzer, 2006). Capture-induced hyperthermia "is one of the primary indications for the development of capture myopathy" (Drew, 1996). Even if animals do not die outright from the hyperthermia or capture myopathy, they may die from predation because of the morbidity associated with hyperthermia; animals are less alert and weakened and less able to defend themselves against predators (Abbott et al, 2005).

1.2.3 Incidence and magnitude

The incidence of hyperthermia during capture is not well reported, because not all studies report the number of animals afflicted with hyperthermia or very few animals' temperatures are recorded during capture. When deaths occur, the cause is often assumed to be capture myopathy with little investigation into the pathology.

Mortality rates in wild species have been rarely documented for different capture methods.. What has been documented is that polar bears are susceptible to overheating especially when chased by helicopters in summer (Stirling et al., 1989). In black bears that were chemically immobilised after they were trapped in snares, it was found that their rectal temperatures ranged from 35.5°C-43°C. Two bears (from a total sample size of 84 bears) died from hyperthermia, as concluded through necropsy (Hellgren and Vaughan, 1989). Few

studies have properly investigated whether hyperthermia can be implicated as a cause of capture-related death. However, many other studies have documented hyperthermia as a common occurrence that occurs post-capture. Table 1.1 is an overview of some of the literature to illustrate the incidence of hyperthermia reported. Table 1.2 is an overview of reported species that are susceptible to hyperthermia.

1.2.4 Causes of capture-induced hyperthermia

The exact mechanisms that cause the increase in body temperature during a capture-induced hyperthermia are not fully understood, but the cause of capture-induced hyperthermia has been elucidated to be stress (Meyer et al., 2008a). However, game capture professions still believe that the primary causes are the ambient conditions and animal exertion (The South African Bureau of Standards, 2000; Ebedes, 2003). Ambient conditions may confound a capture-induced hyperthermia, especially after capture, and therefore the guidelines state that animals should not be captured if the environmental temperature is greater than 25°C, because the temperature difference between the animal's body and the environment may not be great enough for sufficient heat loss (The South African Bureau of Standards, 2000; Atkinson et al., 2006), which may lead to heat storage and hyperthermia.

However, the 25°C ambient temperature guideline has been questioned by capture professionals in Southern Africa who argue for capture outside the demarcated capture season (from 1 March to 31 October) of certain species (Masterson et al. 2009). They

Table 1.1 An overview of hyperthermia and the mortalities associated with capture.

Species	Number of animals captured	Peak (P) and mean (M) body temperature during or after capture (°C)	Ambient temperature (°C)	Site of temperature measurement	Mortality	Comments	Capture type	Capture drugs	Reference
African elephant (<i>Loxodonta africana</i>)	20	P: 37.8°C M: 36.4 ± 0.72°C	21°C to 35°C	Rectal	Not reported		Chemical immobilisation	9.5 ± 0.5 mg etorphine; 2000 IU hyaluronidase	Osofsky, 1997
African lion (<i>Panthera leo</i>)	5	P: 40.2°C M: 38 ± 1.4°C (at 30 minutes after capture)	up to 46°C	Rectal	No deaths; 1 lion had a decreased respiratory rate while immobilised	Rectal temperature remained stable; animals were sprayed with water	chemical immobilisation - darted from car	0.07 mg.kg ⁻¹ medetomidine; 1.8 mg.kg ⁻¹ tiletamine-zolazepam (Zoletil)	Jacquier et al., 2006
American river otter (<i>Lutra canadensis</i>)	10	P: ~ 39°C M: ~38°C (after ~ 50 minutes of immobilization)	15°C to 22°C	Rectal	2 deaths - cause not reported	Body temperature decreased with time	Initial capture prior to start of study not reported	After initial capture: 16.8 mg.kg ⁻¹ ketamine solution (10 ml ketamine, 0.5ml xylazine and 0.5ml acepromazine); 1.25% isoflurane	Hoover and Jones, 1986
Amur tiger (<i>Panthera tigris altaica</i>)	19	M: 40°C ± 1°C (for tigers caught from helicopter)	Average summer: 15°C; Average winter: -14°C	Not reported	Helicopter capture: No deaths - but potential existed from heat stress. Snare captures: 2 (4.5%) severe injuries - fractured metatarsals	In summer animals were cooled with water	Aldrich leg-hold snares; darting by helicopter	After initial capture: 10.8mg.kg ⁻¹ ketamine; 3.4mg.kg ⁻¹ xylazine	Goodrich et al., 2001
Baird's tapir	16	P: 38.6°C Body temperature ranged from 35.5 to 38.6°C	Not reported	Not reported	Not reported	Body temperature was within the reported normal range for tapirs	baited and then darted from tree platform	48 mg butorphanol + 101 mg Xylazine + 187 mg ketamine if immobilisation needed to be extended	Foerster et al., 2000
Bighorn sheep (<i>Ovis canadensis</i>)	634 - 158 by drop-net; 90 by chemical immobilisation; 137 by net-gun; 247 by drive-net	P: 43.8°C M: 42.8°C (in stressed sheep)	Not reported	Rectal	12 - died from capture myopathy; 11 - died from accidents; 95 - showed evidence of capture stress which may have included hyperthermia;	young sheep had higher temperatures than in older sheep; net-gun capture caused lower rectal temperatures than drive-net and chemical immobilisation	chemical immobilisation, drive-net, net-gun and drop-net	Not reported	Kock et al., 1987a; Kock et al., 1987b
	45	P: 41.5°C M: 40.9 ± 0.7°C (after capture)	About 21°C	Rectal and tympanic	1 (2.22%) - died probably from hyperthermia; 11 - had signs of capture stress and hyperthermia; 3 - rectal temperature > 41°C	All sheep were water-doused after capture; tympanic temperature was lower than rectal temperature	Helicopter net-gun	No chemical immobilization	Drew, 1996
Black bear (<i>Ursus americanus</i>)	84	P: 43°C M: 40.1 ± 0.4°C (for male bears in late summer)	Average summer: 26°C; average winter: 5.1°C	Rectal	2 died - necropsy revealed cause of death to be hyperthermia; 5 - showed signs of heat stress	Water was used to cool heat stressed animals with a rectal temperature greater than 42°C	Cable snares	After initial capture: 4.4 mg.kg ⁻¹ ketamine; 2.2 mg.kg ⁻¹ xylazine	Hellgren and Vaughan, 1989

Species	Number of animals captured	Peak (P) and mean (M) body temperature during or after capture (°C) (°C)	Ambient temperature (°C)	Site of temperature measurement	Mortality	Comments	Capture type	Capture drugs	Reference
Black wildebeest (<i>Connochaetes gnou</i>)	4	P: 41°C M: 38.62 ± 0.02°C	Range in summer: 16-31°C; range in winter: 6-21°C	Brain by a thermistor in the brain stem; blood by a thermistor in the carotid artery	Not reported - assume none	No correlation between the intensity of solar radiation and body temperature; increased brain and blood temperature to 41°C after recapture with a helicopter chase	Chemical immobilisation; for recapture animals were darted from a helicopter	3-4 mg per individual of etorphine; 25 mg per individual of xylazine or 50 mg per individual azaperon	Jessen et al., 1994
Boar (<i>Sus scrofa</i>)	47	Not reported	Mean year temperature: 12-13°C	Not reported	5 (10.63%) died: post mortem revealed no gross pathology; 3 animals within 2-3 hours of immobilisation; 2 animals died within 3 days of immobilisation	Animals were placed in a cool, shady area to recover from immobilisation	cage traps; medium sized pens; large sized pens	After initial capture: 2.5 mg per individual of xylazine; 1.25 mg per individual of Zoletil	Fenati et al., 2008
Bobcat (<i>Felis rufus</i>)	21	P: 42.7°C	Average summer: 26.4°C; Average winter: 18°C	Rectal	Not reported	Traps were positioned in the shade to decrease hyperthermia; higher initial temperatures in winter may have indicated stress-induced hyperthermia	wire box traps	After initial capture: 13.3 mg.kg ⁻¹ ketamine; 1.2 mg.kg ⁻¹ xylazine	Beltran and Tewes, 1995
Collared peccaries (<i>Tayassu tajacu</i>)	19	Not reported	Not reported	Not reported	15 % died - probably heat stress in summer		baited clover deer traps	After initial capture: 18.4 mg.kg ⁻¹ mg ketamine	Gallagher et al., 1985
	207	P: 40.1°C M: 37.6 ± 2°C	2°C to 37°C	Rectal	1 death from drug overdose	59 feral hogs were also immobilised in this study; one hog was hyperthermic with a body temperature greater than 41°C	baited box traps	After initial capture: 4.73 mg.kg ⁻¹ Telazol	Gabor et al., 1997
Coyote (<i>Canis latrans</i>)	10	P: not reported M: 40.4°C ± 0.8°C	Average: 5.9°C ± 4.6°C (ranged from -3.9°C to 18°C)	Rectal	Not reported	3 animals reported as disoriented and hyperthermic	Soft-catch leg-hold traps and mesh-wire live traps	After initial capture: 0.12 ± 0.02 mg.kg ⁻¹ medetomidine	Baldwin et al., 2008
Crested porcupine (<i>Hystrix cristata</i>)	17	Not reported	20°C to 28°C	Not reported	2 (11.76%) - 1 "probably" from hyperthermia		live-trapped	After initial capture: 20mg xylazine; 11mg.kg ⁻¹ ketamine	Pigozzi, 1987
	31	P: 38.6°C M: 36.92 ± 0.18°C (after first handling)	Average: 15°C	Rectal	1 (2%) - after immobilisation; no cause reported	1 female aborted two fetuses because of stress; females had a increased rectal temperature in comparison to males; no correlation between rectal temperatures and drug dose	Baited box traps	After initial capture: 30-80mg zoletil	Massolo et al., 2003

Species	Number of animals captured	Peak (P) and mean (M) body temperature during or after capture (°C)	Ambient temperature (°C)	Site of temperature measurement	Mortality	Comments	Capture type	Capture drugs	Reference
Elk (<i>Cervus canadensis</i> and <i>Cervus manitobensis</i>)	18	P: 45°C M: 43 ± 1°C (in both groups after being stressed)	Greater than 33°C	Rectal	3 (16.66%) - died one week after experiments from hyperthermia; temperatures from dead animals were 45°C the previous week; post mortem revealed: muscular oedema and haemorrhage, cardiac haemorrhage and pulmonary oedema; 2 other animals - had signs transient signs of capture myopathy	Mean temperature after stress was 43°C	Baited corral	After initial capture: 0.1 mg.kg ⁻¹ ketanserin; saline	Stanley et al., 1986
European badger (<i>Meles meles</i>)	43	Not reported	Not reported	Not reported	2 (4.65%) died - post mortem revealed predation as the cause	Xylazine administered intra-nasally	Helicopter with net-gun	After initial capture: 1.5-2 mg.kg ⁻¹ xylazine; saline	Cattet et al., 2004
	93	P and Me: 38.3°C ± 0.2°C (for animals that spontaneously recovered from anaesthesia)	Not reported	Rectal	Not reported	High initial rectal temperatures were correlated with spontaneous recovery from anaesthesia	Baited cage traps; chemical immobilisation	A combination of medetomidine, ketamine and butorphanol administered in a ratio of: 20:40:80 µg.kg ⁻¹ ; 20:40:60 µg.kg ⁻¹ and 20:60:40 µg.kg ⁻¹ ; 0.2 mg.kg ⁻¹ ketamine alone	McLaren et al., 2005
	4	P: ~ 38.5°C M: ~37.4 (after 15 min of disturbed normal activity)	Not reported	By radio-telemetry in the abdomen	Not reported	Animals were semi-tame	Animals raised in captivity; restrained in cage traps	No chemical immobilisation	Schutz et al., 2006
European mink (<i>Mustela lutreola</i>)	24	P: 38.3°C M: ~38°C	Not reported	Rectal	Not reported	After capture animals were placed in a shaded room; moderate hyperthermia 5 min post immobilization; rectal temperature decreased over time	Wire mesh traps	After initial capture: 10 mg.kg ⁻¹ ketamine; 0.2 mg.kg ⁻¹ medetomidine	Fournier-Chambrillon et al., 2003
Gray wolf (<i>Canis lupus</i>)	16	P: ~ 38.8°C (at time zero) M: ~38.5 (after 30 minutes)	Not reported	Rectal	No mortality, one animal post immobilisation displayed thrashing and self-mutilation	Rectal temperatures were stable	Not reported	5 or 10 mg.kg ⁻¹ teletamine and zolazepam (Telazol)	Kreeger et al., 1990
Grizzly bear (<i>Ursus arctos</i>)	46	P: not reported M: 38.9°C ± 0.22°C (in free ranging bears)	Not reported	Rectal	Not reported	Rectal temperatures correlated with induction times	Chemical immobilisation - helicopter darting; leg hold snare	8-10 mg.kg ⁻¹ Telazol	Cattet et al., 2003

Species	Number of animals captured	Peak (P) and mean (M) body temperature during or after capture (°C)	Ambient temperature (°C)	Site of temperature measurement	Mortality	Comments	Capture type	Capture drugs	Reference
Impala (<i>Aepyceros melampus</i>)	530	P: 41°C M: 41 ± 0.6°C (during individual restraint); 40.6 ± 0.9°C (after 18.5 min, for animals restrained in the shade)	20°C to 30°C	Rectal	45 had a temperature greater than 41°C; 13 died during individual restraint - post mortem revealed terminal circulatory failure; 2 died in boma - post mortem revealed severe pulmonary odema; 3 died in boma - died due to predation	Rectal temperatures did not return to normal when restrained	Herded by helicopter into a boma; groups of 15 animals caught by nets; during processing animals were restrained with shoulder and flank straps	After initial capture: 0.1-0.2 mg.kg ⁻¹ acetylpromazine	Murray et al., 1981
	15	P: 43.5°C M: 3.2 ± 0.6°C (mean body (increase in naïve animals)	Average dry bulb: 30.1°C; average black globe: 37.4°C ± 5.6°C	Abdominal temperature measured by miniature data loggers	3 deaths – body temperatures of dead animals exceeded 43°C, post mortem revealed lesions characteristic of capture myopathy; initial respiratory depression: 33% with thiafentil and azaperone, 13% with thiafentil and medetomidine, 7% with etorphine and medetomidine and 0% with etorphine and azaperone	Body temperature increase not correlated to drug combinations or ambient temperature but induction times	Chemical immobilisation - animals darted in boma	1.5 mg with etorphine and 40 mg azaperone; 1.5 mg etorphine and 2 mg medetomidine; 1.2 mg thiafentil and 40 mg azaperone; 1.3 mg thiafentil and 2 mg medetomidine	Meyer et al., 2008a; Meyer et al., 2008b
North American porcupine (<i>Erethizon dorsatum</i>)	150	Not reported	Not reported	Not reported	3 deaths (0.87%) - from immobilisation		Night capture with dip net; vertical trap; noose pole; gloved hands and chemical immobilisation	10 mg.kg ⁻¹ ketamine; 4 mg.kg ⁻¹ xylazine (starting doses, doses were lowered over time)	Morin and Berteaux, 2003
Polar bear (<i>Ursis maritimus</i>)	213	P: 40.7°C M: 38.9°C (for bears caught at the Beaufort Sea)	Western Coast and autumn: -2°C to 15°C; Eastern Coast spring: -27°C to -2°C	Rectal	No animals died; 29 had a body temperature greater than 40°C in		Chemical immobilisation - helicopter darting	8-9 mg.kg ⁻¹ Telazol	Stirling et al., 1989
Pronghorn (<i>Antilocapra americana</i>)	281	M and P: 40.8°C (for post-release dead animals)	-21°C to 13°C	Rectal	25 (8.9%) deaths - 12 from direct injuries during capture; 13 post-release	Mortalities within 26 days were considered related to capture; correlation between ambient temperature and rectal temperature not significant; mean rectal temperature was higher for post-release animals that died	Helicopter net-gun	Not reported; assume no chemical immobilisation	Jacques et al., 2009

Species	Number of animals captured	Peak (P) and mean (M) body temperature during or after capture (°C)	Ambient temperature (°C)	Site of temperature measurement	Mortality	Comments	Capture type	Capture drugs	Reference
Raccoon (<i>Procyon lotor</i>)	41	P: 41.1°C M: 38.2 ± 1.2°C	Not reported	Rectal	No mortalities; two animals had transient hind leg paralysis; 1 animal had excessive salivation when immobilised	Small increases in rectal temperature were associated with an increase in ambient temperature; heart rate and respiration rate increased with an increase in rectal temperature	Mesh-wire live traps; chemical immobilisation	15 or 20 mg per individual of Zoletil	Pitt et al., 2006
	31	P: not reported M: 39.6°C ± 1.0°C	Average: 5.9°C ± 4.6°C (ranged from -3.9°C to 18°C)	Rectal	Not reported		Soft-catch leg-hold traps and mesh-wire live traps	After initial capture: 0.21 ± 0.05 mg.kg ⁻¹ medetomidine	Baldwin et al., 2008
Red fox (<i>Vulpes vulpes</i>)	30	P: not reported M: 40.5°C ± 1.1°C	Average: 5.9°C ± 4.6°C (ranged from -3.9°C to 18°C)	Rectal	Not reported		Soft-catch leg-hold traps and mesh-wire live traps	After initial capture: 0.14 ± 0.04 mg.kg ⁻¹ medetomidine	Baldwin et al., 2008
Southern chamois (<i>Rupicapra pyrenaica</i>)	43	P: ~ 41°C (at time zero) M: -40.5°C (at ~40 minutes)	Not reported	Rectal	Not reported	Body temperature decreased with time; body temperature was higher in control animals; initial temperature increase likely due to stress-induced hyperthermia	Drive-nets	After initial capture: 0.11 ± 0.02 mg.kg ⁻¹ acepromazine; control animals received saline	Lopez-Olvera et al., 2007
Springbok (<i>Antidorcas marsupialis</i>)	8	P: 43°C M: 39.22 ± 0.07°C (during winter)	Ranged from -6°C to 34°C	Abdominal temperature recorded with miniature data loggers	No deaths reported	Animals showed short durations of exercise induced hyperthermia of about 41°C; at the end of the study capture resulted in significantly increased temperature than any other time of recording	pop-up corral; capture nets	10-15 mg per individual of haloperidol; 50-100 mg per individual of perphenazine	Fuller et al., 2005.
White-tailed deer (<i>Odocoileus virginianus</i>)	984 overall; 171 reported rectal temperatures	P: 41.9°C M: 38.9°C and 39.5°C (for the initial temperature from capture with clover traps and by net-gun respectively)	Ranged from -28°C to 5.4°C	Rectal	Of the overall 984 animals: 2.8% - accidental deaths and deaths associated with chemical and physical restraint; 2.5% - post-release deaths: died from predation or from complications during handling including hyperthermia and emaciation. Of the 171 reported temperatures: 66.1% of the initial temperatures were or were close to being hyperthermic; only 33.9% of final recorded temperatures were still considered high; only one deer with a rectal temperature between 41.2-41.9°C died - post mortem revealed death was because of predation	Cooling of deer with snow to the abdomen at body temperatures greater than 39.4°C; deer with a body temperature greater than 41°C had chemical immobilisation reversed; body temperature decreased over time; body temperature was not correlated with capture mortality	Collapsible clover traps; helicopter with net-gun; rocket-netting	After initial capture: 75-100 mg per individual with xylazine; 300-400 mg per individual with ketamine	DelGiudice et al., 2001; DelGiudice et al., 2005

Species	Number of animals captured	Peak (P) and mean (M) body temperature during or after capture (°C)	Ambient temperature (°C)	Site of temperature measurement	Mortality	Comments	Capture type	Capture drugs	Reference
White-tailed deer (<i>Odocoileus virginianus</i>)	208	M and P: 40.8°C (for post-release dead animals)	Ranged from -27°C to 13°C	Rectal	3 (1.4%) deaths- from direct injuries during capture	considered mortalities within 26 days related to capture; correlation between ambient temperature and rectal temperature not significant; mean rectal temperature was higher for post-release dead animals than survivors	Helicopter net-gun	Not reported; assume no chemical immobilisation	Jacques et al., 2009
Wolverine (<i>Gulo Gulo</i>)	24: 12 juveniles, 12 adults	P: 42.2°C M: 40.1 ± 0.8°C (after 15-30 min of immobilisation)	-5°C to 25°C	Rectal	No deaths reported; 1 juvenile and 11 adults were hyperthermic : 6 adults had a rectal temperature greater than 41°C	Animals with a rectal temperature greater than 41°C were cooled with snow applied to the tongue, axilla, groin and footpads	Hand capture with snare pole, chemical immobilisation - helicopter darting and ground darting	juveniles - 0.14 mg.kg ⁻¹ medetomidine, 7.5 mg.kg ⁻¹ ketamine; adults - 0.37 mg.kg ⁻¹ medetomidine, 9.4 mg.kg ⁻¹ ketamine	Fahlman et al., 2008

Table 1.2 An overview of species that are reported to be susceptible to hyperthermia, capture myopathy or stress

Species	Account	Reference
Bighorn sheep (<i>Ovis canadensis</i>)	Susceptible to hyperthermia	Foster, 2005
Black bear (<i>Ursus americanus</i>)	Susceptible to hyperthermia	Hellgren and vaughan, 1989
Blue wildebeest (<i>Connochaetes taurinus</i>)	Susceptible to capture myopathy	Burroughs et al., 2006
Buffalo (<i>Syncerus c. caffer</i> and <i>Syncerus c. nanus</i>)	Susceptible to hyperthermia	Burroughs et al., 2006
Bushpig (<i>Potamochoerus porcus</i>) and warthog (<i>Phacochoerus aethiopicus</i>)	Susceptible to stress and hyperthermia	Burroughs et al., 2006
Chinchilla (<i>Chinchilla laniger</i>)	Susceptible to hyperthermia	Orcutt, 2005
Deer	Susceptible to hyperthermia	Caulkett and Haigh, 2004
Eland (<i>Taurotragus oryx</i>)	Susceptible to hyperthermia and capture myopathy	Burroughs et al., 2006
Guinea pig (<i>Cavia porcellus</i>)	Susceptible to hyperthermia	Orcutt, 2005
Impala (<i>Aepyceros melampus</i>)	Susceptible to stress	Murray et al., 1981; Knox and Hattingh, 1990
Impala (<i>Aepyceros melampus</i>)	Excitable nature	Meyer et al., 2008a and 2008b
Klipspringer (<i>Oreotragus oreotragus</i>)	Susceptible to capture myopathy	Burroughs et al., 2006
Nyala (<i>Tragelaphus angasi</i>)	Susceptible to stress	Burroughs et al., 2006
Polar bear (<i>Ursus maritimus</i>)	Susceptible to overheating	Stirling et al., 1989; Bourne [Accessed 11 Jan 2010]
Rabbit	Susceptible to overheating	Bourne [Accessed 11 Jan 2010]
Sable (<i>Hippotragus niger</i>)	Susceptible to hyperthermia	Burroughs et al., 2006
Tsessebe (<i>Damaliscus lunatic</i>)	Susceptible to capture myopathy	Burroughs et al., 2006
Waterbuck (<i>Kobus ellipsiprymnus</i>)	Susceptible to heat stress	Burroughs et al., 2006

propose that the duration an animal is chased poses a greater adverse effect and mortality risk than does high ambient temperature (Masterson et al. 2009). Capture outside of the cool capture season months is plausible because it has been shown that the stress of capture is the primary cause of capture-induced hyperthermia, and the magnitude of capture-induced hyperthermia is independent of ambient temperature (Meyer et al., 2008a; Meyer et al., 2008b). Meyer et al. (2008a) correlated the amount of stress in captured animals to the magnitude of the capture-induced hyperthermia, using miniature data loggers to continuously record body temperature before, during and after capture. The technology had been used before in other ungulate studies and capture-induced hyperthermia had been seen in the data (Fuller et al., 2005) but the cause of capture-induced hyperthermia had not been investigated. Rectal temperatures, usually documented in capture studies, usually provide only a single reading and are only taken during the time the animal is immobilised and therefore have not been useful in describing the cause of capture-induced hyperthermia. Meyer and his colleagues (Meyer et al., 2008a) varied capture times, human exposure, animal microclimate and animal activity to examine the cause of capture-induced hyperthermia. The study animals showed an increase in body temperature with net-capture, chemical immobilisation and even exposure to the presence of humans (without capture). The results showed that the magnitude of the capture-induced hyperthermia was not related to the intensity of exercise, activity levels and environmental temperatures during capture. However, the level of stress in the impala was correlated to the body temperature increase in capture. That is, that impala that were more habituated had smaller increases in body temperature during capture. Therefore, at least in the initial body temperature increase seen in captured animals, the rise in body temperature may be caused predominantly by stress.

Meyer et al., (2008a) propose that capture-induced hyperthermia may be a form of stress-induced hyperthermia, the mechanisms of which are not fully elucidated but are likely mediated in the central nervous system (Oka et al., 2001). What is known about stress-induced hyperthermia, which has been studied mainly in small laboratory animals, is that stress-induced hyperthermia activates the hypothalamic-pituitary-adrenal axis when a stressor is perceived (Vinkers et al., 2009). This activation then mediates a rise in body temperature that will occur in either a hot or cool environment. Therefore, the increase in body temperature will rise irrespective of the ambient temperature. The activation of the autonomic nervous system lasts as long as the exposure to the stressor and therefore the rise in body temperature will last the same duration (Vinkers et al., 2009). This stressor can be physical and psychological, such as handling stress, restraint stress or a novel stimulus in laboratory animals or suppressed emotional expression in humans (Moe and Bakken, 1997; Oka et al., 2001). Stress-induced hyperthermia occurs in many species and has been found in man, mice, lizards and animals such as silver foxes and deer (Moe and Bakken, 1997; Montane et al., 2003). The psychological stress experienced by animals during capture occurs in the initial engagement of capture and is responsible for the likely initial stress-induced body temperature increase of a captured animal. Anxiolytic drugs, such as benzodiazepines and certain serotonergic agonists, have been shown to attenuate stress-induced hyperthermia when the drugs were administered prior to the exposure of a stressor (Lecci et al., 1990). However, if these drugs are administered to an immobilised animal sometime after the initial capture engagement has elapsed they may not be as effective (Montane et al., 2003).

That is not to say that various other factors that include high ambient temperature, immobilising drugs and the animal's exertional state will not influence capture hyperthermia (Hattingh et al., 1990; Kock and Meltzer, 2006). These factors may not be the underlying cause of the initial body temperature increase during capture but will exacerbate the already existing body temperature increase and confound a capture-induced hyperthermia. During capture the sympathetic response increases the metabolic rate of the animal, which increases its metabolic heat production. However, this increase in sympathetic drive is also stress-related as animals increase energy metabolism in their attempt to escape a stressor (Ganhao et al., 1988). Exertion also increases muscle metabolism thus increasing metabolic heat production (Montane et al., 2003). Normally, when an animal is not in a capture situation, ambient air temperature does not correlate to the normal body temperature of that animal. In antelope, such as impala, their normal body temperature is the highest in the late afternoon when air temperatures are decreasing (Meyer et al., 2008a). However, in hot and humid conditions (when ambient temperature exceeds body temperature) the increased ambient temperature will exacerbate the hyperthermia by limiting heat loss to the environment (Art and Lekeux, 2005). Tranquilising drugs and anaesthetics used for capture can also contribute to the hyperthermia because they can hinder the physiological mechanisms responsible for countering the increased heat load (Kock and Meltzer, 2006). The combination of high ambient temperatures and capture drugs may only become important when animals are immobilised for a prolonged period of time with drugs that induce thermal lability (Meyer et al., 2008a).

The immobilising and sedative drugs used in capture have various pharmacological properties and therefore have different side-effects on thermoregulation. The opioid drugs

are potent analgesics but can have severe side-effects such as respiratory depression (the animal stops breathing), hypertension, and paradoxical excitability in some species. An important side-effect is that the suppression of thermoregulatory mechanisms may occur (Swan, 1993). This suppression may make the animal thermally labile (meaning that the animal's body temperature will approach the environmental temperature). The cyclohexylamines, especially ketamine, may also cause hyperthermia because of the side-effects of increased muscle tone and convulsions (Swan, 1993; Meltzer et al., 2006b). Sedative drugs (such as xylazine and medetomidine) can also disrupt thermoregulation by suppressing central nervous system function (xylazine and medetomidine are alpha-2 receptor agonists), decreasing heart rate and suppressing peripheral blood flow through alpha-2 receptor activation (Swan, 1993). Tranquillising drugs such as azaperone inhibit alpha-1 receptors (Swan, 1993; Meltzer et al., 2006b) and therefore can cause peripheral vasodilation, which may alleviate a capture-induced hyperthermia by increasing heat loss from the periphery. However, if ambient temperature is high then tranquillising drugs may aggravate a hyperthermia because the peripheral vasodilation would allow for increased heat transfer from the environment to the animal. Meyer et al. (2008b) conducted a study of the thermal effects in impala of a number of drug combinations made up of thiafentanyl, etorphine, azaperone and medetomidine. The results showed that the choice of drug was not the overriding factor in causing an increase in magnitude of capture-induced hyperthermia. Once again the amount of stress an animal experienced during capture correlated with the magnitude of the capture-induced hyperthermia. The longer an animal was conscious during capture the greater the amount of stress was experienced by the animal and subsequently there was a greater body temperature increase. No matter the drug combination, some animals had shorter induction times which correlated with less

cortisol secretion and a smaller capture-induced hyperthermia than animals that took longer to become recumbent. Other studies have reported a similar correlation between limiting stress and induction times and the rectal temperatures of wild animals (Kock et al., 1987a; Beltran and Tewes, 1995; Cattet et al., 2003). That is, the shorter the period of stress for an animal, the smaller the magnitude of the body temperature increase.

Long-acting tranquilising drugs such as haloperidol, perphenazine enanthate and zuclopenthixol, which are long-acting neuroleptics, have been thought to also inhibit thermoregulation, and have been investigated in scientific studies. These drugs were tested in both goats and wildebeest and found to have no significant effect on thermoregulation (Fick et al., 2006; Fick et al., 2007). Wildebeest were housed in a boma and had no significant changes in normal body temperature when dosed with these tranquilisers (Fick et al., 2006).

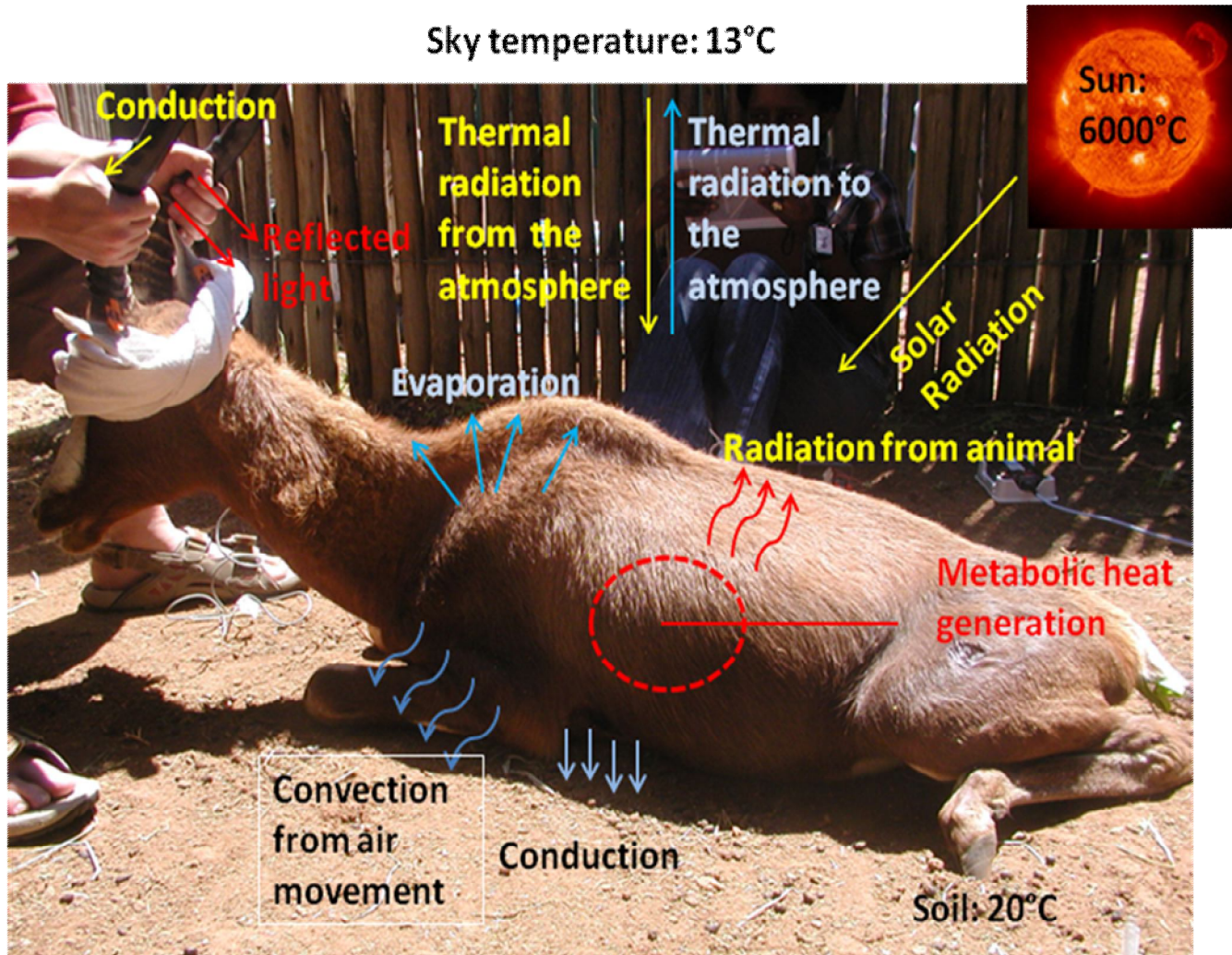
More studies investigating the mechanisms of the causes of capture-induced hyperthermia are needed. Because stress seems to be the major cause of the initial body temperature increase in capture, its occurrence can be limited but most likely not eliminated. Therefore, during capture the stress and the hyperthermia need to be sufficiently controlled and treated to ensure optimal animal wellbeing.

1.3 Cooling of hyperthermic animals

1.3.1 Principles of heat loss

A possible solution to reduce the complications caused by capture hyperthermia is to physically cool captured animals. Physical cooling methods use the principles of conduction, radiation, evaporation and convection. Figure 1.1 summarises the main avenues of how animals lose and gain heat to and from the environment by radiation, evaporation and conduction. Conduction is the heat exchange across a surface through direct contact and can be increased by increasing the temperature gradient and area of contact (Willmer et al., 2005). A large ice-pack will have greater conductive cooling than will a smaller pack, while frozen water cools more by conduction than does warmer water. Convection is heat exchange due to gas or liquid movement over a surface, for example fanning, which increases heat loss through increased convection (Weller, 2005). Convection can be increased by increasing the air velocity. Radiation is the emission of heat through infrared rays. A hot body will lose heat to its cooler surroundings through radiation (Weller, 2005; Willmer et al., 2005).

Lastly, evaporation is the use of thermal energy to vaporise water, which causes a subsequent loss of heat on the evaporating surface (Weller, 2005; Bouchama et al., 2007). Evaporation cools the surfaces by using energy to transform water into a gas from a liquid. This energy is in the form of heat removed from the evaporating surface, which is thereby cooled (Weller, 2005). The amount of energy needed is called the latent heat of vaporisation and is 2400 J.g^{-1} at an air temperature of 25°C (Willmer et al., 2005; Haeussermann et al., 2007). The rate of evaporative cooling depends on a combination of variables, which are the water-vapour pressure gradient, the surface temperature and the amount of water on the surface, which depends on the water permeability of that surface (Willmer et al., 2005). That is, an environment with high humidity will allow less evaporation than will a low



Core body temperature: 38.8°C

Air temperature: 28°C

Figure 1.1 An overview of how animals lose and gain heat to and from the environment. Adapted from Willmer P, Stone G, Johnston I. 2005. Temperature and its effects. In: *Environmental Physiology of Animals*. Blackwell Publishing. Victoria. p. 197

humidity environment, at the same ambient temperature. The greater the temperature difference between the surface and environment the quicker evaporation will occur, and surfaces covered in feathers and hairs will increase the resistance to evaporation and water loss and have less evaporation than will a hairless surface. That is why in humans, who are relatively hairless mammals, evaporative cooling is such a powerful cooling tool that can cause 80% of all heat loss during exercise (Weller, 2005; Willmer et al., 2005).

In animals evaporation is also one of the most important cooling avenues. Increased evaporative cooling can be achieved through sweating or increasing the respiratory evaporative heat loss through the mouth by panting, or through the respiratory system by increasing airflow. However, increased evaporative cooling removes water, and the water balance of the animal may be affected with prolonged sweating or panting (Willmer et al., 2005; Cain et al., 2006). Therefore there is a limit to the amount of evaporative heat loss an animal is capable of, and when that limit is reached the animal then becomes dehydrated, thus decreasing or ceasing evaporative heat loss. The efficacy of sweating depends on wind speed, the rate of sweating and the water-vapour pressure gradient (Cain et al., 2006). Heat loss by panting occurs because increased air flows over the evaporative surfaces of the upper respiratory tract and therefore the amount of heat lost depends primarily on the volume of respired breaths and the frequency of the breaths, but the amount of evaporative heat loss due to panting will also depend on the water-vapour pressure and the salivation rate (Cain et al., 2006).

Heat loss by sweating and panting in animals is a part of thermal homeostasis, which is maintained by the balance of heat gain and heat loss (Weller, 2005). In mammals and birds this balance is maintained within a narrow temperature range that is the optimal temperature for biochemical reactions in the body, as the activation energies of enzymes are affected by temperature changes (Willmer et al., 2005). Animals can gain heat by radiation from the sun and surroundings and also lose heat by radiation to their surrounding environment. Primarily, endothermic animals, such as ungulates, will produce heat through metabolic activity, muscle activity and assimilation of food (Drew, 1996). Therefore, exercise and increased food intake will increase their metabolic heat production (Drew, 1996). However, animals also gain or lose heat from their environment (Figure 1.1) but can alter their behaviour to avoid undesirable environmental temperatures, for example by moving into the shade or by basking in the sun (Willmer et al., 2005; Cain et al., 2006).

In hot environments animals must employ techniques to dissipate heat to maintain their normal body temperature. By changing their body position and behaviour animals can alter the conductive cooling of a substrate by altering how much of their body is in contact with that substrate – for example, standing versus lying down (Willmer et al., 2005).

Animals also transfer heat to the environment through convection which removes warm air that surrounds the body (Willmer et al., 2005). To increase convective cooling to the environment, animals can align more of their body surface towards wind or can alter the insulative properties of their hair, fur, or feathers (Willmer et al., 2005). In summer, animals may shed thicker winter coats and grow lighter coats (both in colour and thickness) to decrease insulation and improve convective heat loss (Willmer et al., 2005). Animals also

vasodilate blood vessels in the skin so that more warm blood flows through the periphery from the core, which increases convective cooling away from the core, increases radiation from the skin surface and increases convective and conductive cooling at the skin surface (Weller, 2005; Willmer et al., 2005; Cain et al., 2006).

Usually the signal to activate heat loss mechanisms is activated in the central nervous system. Thermoreceptors sense changes in body (and skin) temperature and then transmit this afferent signal to the central nervous system where the afferent signal is compared to the normal body temperature set point (Weller, 2005). If a difference from the normal body temperature set point is perceived then efferent nerves are stimulated to activate heat loss mechanisms such as vasodilation and sweating (Weller, 2005).

When animals are immobilised during capture the aforementioned heat loss avenues may be compromised (such as inhibition of the thermoregulatory centre in the central nervous system by immobilising drugs). Animals may also be captured on hot, still, sunny days where an increased heat load on the animals can occur from the environment (through increased radiation and conduction and diminished convection). After capture animals may be placed into vehicles for transport where animals may also face a similar problem of decreased efficacy of heat loss mechanisms because truck compartments and crates are confined spaces where air movement is lessened (causing decreased convective cooling) and the apparent specific humidity of the air may increase over time (causing decreased evaporative cooling). Therefore, after the process of capture the need to cool animals artificially is often necessary.

1.3.2 Cooling methods used for humans

In regards to this dissertation, cooling refers to techniques used with the purpose of decreasing core body temperature and not for creating thermal comfort only. Thermal comfort in the heat relates to the level of fatigue and perception of feeling cool. It is possible to increase thermal comfort, for example by activating cold-sensitive thermoreceptors on the face (by splashing with cold water), without significantly decreasing the core body temperature.

Various means of reducing body temperature have been investigated in humans. These range from increasing airflow of intubated patients to using cooling plates on the body (Andrews et al., 2005; Bayegan et al., 2008). Other methods include the application of ice-packs to areas where large arteries pass near the body surface such as the neck, axilla and groin (Clements et al., 2002). However, only placing ice-packs over major arteries alone has been proven to be ineffective, or only effective if combined with another cooling technique (Smith, 2005). Table 1.3 provides an overview of cooling techniques and cooling experiments done in humans and in animals (that were used to model human heat loss).

Most human studies have focused on cooling athletes with exercise-induced hyperthermia, cooling heat stroke victims and inducing hypothermia in a clinical setting to prevent further tissue damage from stroke and cardiac arrest (Clements et al., 2002, Moore et al., 2008). One technique used to induce hypothermia is infusion of 4°C ice-cold intravenous (IV) fluid, which in humans had been infused at 30ml.kg⁻¹ for 30 minutes or 4l over an hour (Schneider et al., 2006). A concern in using cold or icy water (1-3°C) for dousing and 4°C IV fluid is that

Table 1.3 An overview of cooling techniques that have been tested for the purpose of cooling humans

Cooling type	Reason for cooling	Subjects	Ambient temperature	Site of temperature measurement	Lowest temperature or cooling rate reported	Comments	Associated adverse effects	Reference
Increased airflow through the upper respiratory tract	To decrease the temperature increase (pyrexia) of the brain after brain injury	15 brain injured, intubated and mechanically ventilated patients	Not reported	Brain and oesophageal	Brain temperature: little difference of 0.13°C between airflow and no airflow state.	Air flow was at room temperature at 115 ml.kg ⁻¹ .min ⁻¹ into the nostrils; no decrease in oesophageal temperature recorded; Concluded that increased airflow into the airways did not significantly lower brain temperature but reckon that the air conditioning increased convective cooling of the skull and decreased brain temperature	Not reported	Andrews et al., 2005
Water immersion (1-3°C; 10-11°C; 15-16°C; 18-20°C and 25°C)	To cool heatstroke victims	40 dogs; collapsed in a heat chamber and then cooled	45°C (for climatic chamber; not reported during cooling)	Rectal	1-3°C water: 9.9 ± 0.6 kcal.m ⁻² .min ⁻¹ 10-11°C water: 9.9 ± 2.1 kcal.m ⁻² .min ⁻¹ 15-16°C water: 11 ± 2.8 kcal.m ⁻² .min ⁻¹ 18-20°C water: 5.4 ± 0.8 kcal.m ⁻² .min ⁻¹ 25°C water: 5.2 ± 0.6 kcal.m ⁻² .min ⁻¹	Dogs had heat stroke induced in a climatic chamber and were then cooled. 18 dogs were comatose from heatstroke. It was found that the 15-16°C water had a similar cooling rate to the 1-3°C water. Water that was warmer than 18°C still cooled significantly but cooled not as quickly as the 1-3°C and 10-11°C water.	2 dogs died after immersion	Magazanik et al., 1980
Water immersion at: 2°C, 8°C, 14°C and 20°C	To cool exercise-induced hyperthermia	7 healthy humans; exercised until rectal temperature reached 40°C	39°C	Oesophageal, aural and rectal	2°C water immersion: 34.5 ± 1.2°C (oesophageal temperature) 8°C water immersion: 35.4 ± 0.9°C 14°C water immersion: 36.3 ± 1.1°C 20°C water immersion: 36.2 ± 1.1°C	Volunteers were cooled in a circulating water bath until rectal temperature was 37.5°C. Found that the fastest core cooling rate (calculated from rectal temperature) was from the 2°C water immersion but experienced continued core-cooling after the immersion had stopped in all the water temperatures	No adverse effects	Proulx et al., 2003; Proulx et al., 2006
Water immersion (13°C-15°C and 23°C-24°C); cold air (0°C and 5°C)	A review of methods for precooling athletes before events to increase exercise performance	No subjects; review article	5°C - 40°C	Rectal and oesophageal	Water immersion (17°C): A decrease of 4.2°C over 30 minutes Cold air (0°): reduced core body temperature by ~1°C	For a range of ambient temperature, water immersion and forced air movement improved exercise performance when subjects were precooled	If body core temperature was lowered to hypothermic levels (less than 35°C) then exercise performance was compromised	Marino, 2002

Cooling type	Reason for cooling	Subjects	Ambient temperature	Site of temperature measurement	Lowest temperature or cooling rate reported	Comments	Associated adverse effects	Reference
Ice-water immersion; circulating water; forced air movement; bladder lavage	To actively cool patients undergoing surgery	6 Normothermic healthy human volunteers	23°C	Tympanic	Ice-water immersion: $9.7 \pm 1.1^{\circ}\text{C}\cdot\text{hr}^{-1}$ Circulating water: $1.6 \pm 1.1^{\circ}\text{C}\cdot\text{hr}^{-1}$ Forced air movement: $1.7 \pm 0.5^{\circ}\text{C}\cdot\text{hr}^{-1}$ Bladder lavage: $0.8 \pm 0.3^{\circ}\text{C}\cdot\text{hr}^{-1}$	Forced air technique: $1000\text{L}\cdot\text{min}^{-1}$ of air at 10°C ; circulating water technique: through the use of a circulating water blanket and mattress at 5°C ; Bladder lavage: a 10 min cycle of 300 ml of fluid; ice-water immersion: 50l of crushed ice and water. Recommend ice-water immersion when non-invasive cooling methods are inadequate in treating hyperthermia or inducing hypothermia.	Ice-water immersion: terminated after 20 minutes because temperature approached 34°C , afterdrop of 2°C reported	Plattner et al., 1997
Ice-water immersion ($1-3^{\circ}\text{C}$); whole body cooling by wet towels and air exposure	To cool exercise-induced hyperthermia	21 highly physically fit humans; had been exercised to a rectal temperature of 41.2°C	24.4°C	Rectal	Ice-water immersion $0.20^{\circ}\text{C}\cdot\text{min}^{-1} \pm 0.02\text{C}\cdot\text{min}^{-1}$ Air exposure while wrapped in wet towels: $0.11 \pm 0.02\text{C}\cdot\text{min}^{-1}$	Volunteers ran a 11.5km race and were then cooled in the field. Ice-water immersion was compared to air exposure with wet towels wrapped around the body. It was found that ice-water cooled significantly faster than the air exposure cooling method	No adverse effects	Armstrong et al., 1996
Ice-water immersion (5.2°C); cold-water (14°C) immersion and control	To cool exertional heat stroke victims	17 highly physically fit humans (distance runners); exercised by running 19km	$27^{\circ}\text{C} \pm 1^{\circ}\text{C}$	Rectal	Ice-water immersion $-0.13 \pm 0.01^{\circ}\text{C}$ (from start of immersion to 3 minutes after immersion had ended) Cold-water immersion: $0.13 \pm 0.01^{\circ}\text{C}$	Volunteers after exercise were immersed for 12 minutes in tubs. Found that ice-water immersion and cold-water immersion had similar rectal temperatures and cooling rates; both cooling methods cooled significantly greater than the control of no cooling (only passive cooling)	No adverse effects	Clements et al., 2002
Water immersion; evaporative cooling; conductive cooling; invasive cooling	A review of cooling methods for treating heatstroke victims	No subjects; review article	Not reported	Not reported	Ice-water immersion: cooled heat stroke victims to 38.3°C Rest of cooling techniques actual body temperatures reached not reported	Conductive cooling included: cooling blankets and ice-packs; invasive cooling included: intravenous solution and lavage. Concluded that there was not enough evidence to declare one cooling method the best cooling method for heatstroke victims.	Ice-water immersion mortality rate: 14.3%; Conductive cooling: 15-33%;	Bouchama et al., 2007

Cooling type	Reason for cooling	Subjects	Ambient temperature	Site of temperature measurement	Lowest temperature or cooling rate reported	Comments	Associated adverse effects	Reference
Cold-water immersion	A review on cold-water immersion	No subjects; review article			0.15-0.35 °C.min ⁻¹ for cold-water (14°C) to ice-water immersion (2°C)	This review aims to elucidate cooling misconceptions, especially the misconception that vasoconstriction and shivering associated with cold water immersion impede cooling. The article concludes that strong evidence supports cold-water immersion as the first line treatment for heatstroke	Some discomfort reported but not a primary concern in the acute treatment stage	Casa et al., 2007
Cold-water immersion; evaporative cooling; invasive cooling; Dantrolene as a pharmacological adjunct in heatstroke	A review of methods for treating exertional heatstroke	No subjects; review article	Not reported	Not reported	Ice-water immersion: 0.15-0.2°C.min ⁻¹ Evaporative cooling: 0.05-0.310.2°C.min ⁻¹ for a body cooling unit. Invasive cooling: 0.11 °C.min ⁻¹ with peritoneal lavage. Dantrolene adjunct: cooled as quickly as cooling without dantrolene	Ice-water immersion is the most effective cooling method; further work recommended to specifically compare water immersion and evaporative cooling techniques	Some patients reported to find immersion uncomfortable	Smith et al., 2005
Cold-water immersion (8°C)	To determine if body fat effected cooling rate	17 healthy humans; low body fat group had 8 volunteers and high body fat group had 9 volunteers; exercised until rectal temperature reached 40°C	40°C	Rectal, ear canal, skin and oesophageal	Higher fat group: 0.20 ± 0.09°C.min ⁻¹ Lower fat group: 0.23 ± 0.09°C.min ⁻¹	Volunteers were exercised and then placed into a water bath. The body fat percentage was significantly different (a difference of about 10%) between the two groups, however cooling rates were similar for the two groups. Therefore, it was concluded that a 10% body fat difference does not affect the cooling rate of water immersion in exercise-induced hyperthermia.	Not reported - assume none	Lemire et al., 2008

Cooling type	Reason for cooling	Subjects	Ambient temperature	Site of temperature measurement	Lowest temperature or cooling rate reported	Comments	Associated adverse effects	Reference
Hand immersion; whole body fanning; air cooled garment; liquid cooled garment and phase change garment	To cool exercise-induced hyperthermia	9 healthy humans; exercised until body temperature reached 38.5°C	31.2°C	Rectal and skin	Whole body fanning: 0.92°C (change in rectal temperature after 30 minutes of exercise) Hand immersion: 0.8°C Air cooled garment: 0.76°C Liquid cooled garment: 0.77°C Phase change garment: 0.69°C	Volunteers were exercised to a rectal temperature of 38.5°C, then cooled for 15 minutes and then exercised again. Hands were immersed up to wrists in 17°C water; phase change garments cool the chest by conduction; air cooled garments cool the chest by fanning; liquid cooled garments cool the chest by cooling circulating saline in the vest to 12.3°C; in the whole body fanning treatment subjects were cooled by being seated in front of a fan with an airspeed of 3.5-3.8 m.s ⁻¹ . Concluded that whole body fanning was the most effective technique for cooling athletes undergoing repeated bouts of exercise in the heat.	No adverse effects	Barwood et al., 2009
Cold (-20°C) metal plates applied to body surface which were compared to ice-packs, fans and alcohol rubs	To induce a therapeutic hypothermia	12 human-sized swine	Not reported	Brain, bladder, pulmonary artery tympanic and oesophageal	Cold metal plates: 9.3 ± 1.4°C.hr ⁻¹ Control of ice-packs, alcohol rub, and fans: 6.7 ± 1.4°C.hr ⁻¹	Control animals were cooled with ice-packs distributed over the animal and by an alcohol spray to the abdomen and neck of the swine and evaporated by two electric fans. Concluded that the use of cold metal plates was effective in inducing therapeutic hypothermia in swine.	No adverse reaction	Bayegan et al., 2008
Ice-jacket	To increase exercise performance	7 highly physically fit humans; exercised for 80 minutes	30°C	Rectal and skin	A 1.2°C decrease in rectal temperature when wearing the ice-jacket	Ice-jacket was of a design that allowed numerous ice-packs to be placed in pockets to cool the chest. Found no significant difference between exercise performance with and without the ice-jacket	Not reported - assume none	Duffield et al., 2003

Cooling type	Reason for cooling	Subjects	Ambient temperature	Site of temperature measurement	Lowest temperature or cooling rate reported	Comments	Associated adverse effects	Reference
Hand and head wash with 2l of 23°C water	To cool workers working in high ambient temperature environments to prevent heat stroke and heat exhaustion	11 healthy and physically fit humans	35°C	Rectal, oesophageal, skin and ear canal	Rectal: ~ 37.8°C; oesophageal: ~37.5°C	Water was applied to the head and hands for one minute after 20 minutes of exercise; subjects were only allowed an increase of core body temperature to 38.5°C, otherwise the experiment was terminated therefore subjects only had a maximum of 1°C body temperature increase. Skin and ear canal temperatures decreased but rectal and oesophageal temperatures were unchanged by cooling. Concluded that this method is an effective and economical method for reducing heat stress.	1 individual reached a core body temperature of 38.5°C and experimentation ceased	Fuji et al., 2008
Extracorporeal cooling; endovascular cooling	To induce a therapeutic hypothermia	6 human-sized swine	Not reported	Brain, bladder, pulmonary artery and tympanic	Extracorporeal cooling: $8.2 \pm 2.8^{\circ}\text{C}\cdot\text{hr}^{-1}$ (for decreasing brain temperature) Endovascular cooling: $2.6 \pm 0.8^{\circ}\text{C}\cdot\text{hr}^{-1}$	Extracorporeal venovenous cooling: venovenous flow rate was $300 \text{ ml}\cdot\text{min}^{-1}$ at 4°C; endovascular cooling: saline flow rate $100 \text{ ml}\cdot\text{min}^{-1}$ at 4°C. Found that extracorporeal cooling was significantly faster than endovascular cooling and that extracorporeal cooling was a safe and effective method for inducing hypothermia in swine	No adverse effects	Holtzer et al., 2005
Central venous infusion (4°C and 20°C)	To induce a therapeutic hypothermia	9 healthy human volunteers		Tympanic, muscular, skin	4°C intravenous infusion : $2.5 \pm 0.4^{\circ}\text{C}$ (decrease in body temperature) 20°C intravenous infusion: $1.4 \pm 0.2^{\circ}\text{C}$	$40 \text{ ml}\cdot\text{kg}^{-1}$ of saline was infused over 30 minutes. It was found that infusing centrally with cold fluid decreased core body temperature more than they had expected. They suggest 4°C intravenous infusion for inducing hypothermia.	Not reported	Rajek et al., 2000

Cooling type	Reason for cooling	Subjects	Ambient temperature	Site of temperature measurement	Lowest temperature or cooling rate reported	Comments	Associated adverse effects	Reference
Intravenous infusion (fridge temperature, exact temperature not reported; ice-packs to areas of large arteries (axilla, groin and neck) and fanning (with an industrial fan) with an intermittent water spray	To treat exercise-induced hyperthermia and to examine the effect of cooling on heart rate	11 healthy humans; exercised until core temperature reached about 40°C	34.2°C ± 0.5°C	Core (via an ingestible thermometer pill) and skin	Fanning: 37.7 ± 0.3°C (after 40 minutes of cooling) Cold saline IV infusion: 37.8 ± 0.3°C Ice-packs: 37.7 ± 0.3°C	After an exercise-induced hyperthermia was elicited cooling techniques were applied with the aim to investigate the effects on heart rate. Ice-packs and fanning significantly lowered heart rate in comparison to the intravenous infusion treatment.	Not reported - assume none	Leicht et al., 2009
Intravenous infusion (4°C and 23°C)	To induce a therapeutic hypothermia	16 normothermic humans	23.9°C	Core (via an ingestible thermometer pill) and skin	4°C intravenous infusion: 1 ± 0.4°C (decrease in core temperature) 23 °C intravenous infusion: 0.5 ± 0.1°C	30 ml.kg ⁻¹ of saline was infused over 30 minutes. The 4°C IV infusion resulted in core cooling but did not induce a therapeutic hypothermia. The 23°C IV infusion only mildly cooled the core.	Visible shivering noted	Moore et al., 2008
Iced gastric lavage	To cool heatstroke victims	11 dogs; externally heated to a body temperature of 43°C	20°C	Brain, pulmonary artery, rectum and chest wall subcutaneous tissue	Iced gastric lavage: 0.200-0.216°C.min ⁻¹ .m ⁻² cooling rate	Control dogs cooled passively; dogs in the experimental group were treated with boluses of 300 ml of iced water (1-3°C) into their stomach. Concluded that iced gastric lavage, at least in dogs, was a safe and effective adjunctive technique for treating heatstroke.	2 control animals died; post mortem in all animals revealed tissue damage from heatstroke	Syverud et al., 1985

Cooling type	Reason for cooling	Subjects	Ambient temperature	Site of temperature measurement	Lowest temperature or cooling rate reported	Comments	Associated adverse effects	Reference
Placement of icepacks to large arteries of the axilla, neck and groin; covering the body with ice-packs with and without evaporative cooling; evaporative cooling	To cool heatstroke victims	Five physically fit humans; subjects were exercised until rectal temperature increased by 2°C	40°C	Rectal	Combined - ice-packs to the body surface and evaporative cooling: $-0.036^{\circ}\text{C}\cdot\text{min}^{-1}$ Selected ice-pack placement: $-0.028^{\circ}\text{C}\cdot\text{min}^{-1}$ Ice-packs over the body surface without evaporative cooling: $-0.034^{\circ}\text{C}\cdot\text{min}^{-1}$ Evaporative cooling alone: $-0.034^{\circ}\text{C}\cdot\text{min}^{-1}$	Evaporative cooling was achieved by blowing compressed air over subjects that had been splashed with water; ice-packs to only the major arteries was insignificantly more effective than not applying any external cooling. Found that the highest cooling rate was achieved by combining ice-packs over the body surface with evaporative cooling.	Not reported	Kielblock et al., 1986
Ice-packs to body surface	To increase exercise performance	Five dogs; exercised on a treadmill	$22^{\circ}\text{C} \pm 1^{\circ}\text{C}$	Hypothalamic, rectal and muscle	Rectal: -41°C at the end of an hour after exercise with cooling; $\sim 43^{\circ}\text{C}$ without cooling	Dogs were exercised until exhaustion; exercise with cooling was compared to exercise without cooling. It was found that cooling with ice-packs significantly increased exercise performance	Not reported	Kruk et al., 1985

peripheral vasoconstriction and the induction of shivering may occur, decreasing the cooling rate (Proulx et al., 2003). However, it has been shown that cold-induced vasodilation, rather than vasoconstriction, occurs if the temperature of doused water is below a critical temperature of 16°C (Daanen, 2003). In addition, the slowing of cooling by vasoconstriction has not been proven experimentally (Smith, 2005).

Although there are many cooling techniques in humans, cold-water immersion is considered the gold standard technique for whole body cooling (Casa et al., 2007a), but there has been controversy surrounding this technique because of the view that cutaneous vasoconstriction would decrease heat transfer between the patient and the water (Clements et al., 2002). There was also a concern that water immersion would increase metabolic heat production by inducing shivering, if skin temperature dropped below 28°C (Clements et al., 2002). However, studies (Costrini, 1990; Armstrong et al., 1996; Smith, 2005; Casa et al., 2007a) have shown water immersion to be the most effective method for body cooling and that its cooling power overcomes any cutaneous vasoconstriction that may be present. It has now been established that cold water immersion is the best method for alleviating hyperthermic illness (Casa et al., 2007a). A study using an ice-water slurry, in which volunteers were immersed in 50l of icy water, found that it cooled humans at a rate of $9.7^{\circ}\text{C}\cdot\text{h}^{-1} \pm 4.4^{\circ}\text{C}\cdot\text{h}^{-1}$, but these values were obtained from normothermic individuals and not hyperthermic ones (Plattner et al., 1997). This technique is also associated with an “afterdrop” phenomenon, which is the continued cooling of the core after the intervention has ended, which may then lead to a hypothermic state (Plattner et al., 1997).

Although cold-water immersion may have the largest cooling rate, cooling techniques need

to be tailored to the situation in which they will be used. For example in outdoor endurance events it may not always be feasible to use ice-water. So in one study (Clements et al., 2002) the efficacy of ice-water immersion versus cold-water immersion was investigated; runners were cooled with ice-water immersion (average of 5.2°C water temperature) and cold-water immersion (average of 14.0°C water temperature) to compare the rate of the different water temperatures in cooling athletes with exercise-induced hyperthermia. Both the water temperatures cooled the patients significantly faster than did no cooling but neither technique cooled significantly faster compared to the other (Clements et al., 2002). Another study (Barwood et al. 2009) investigated alternatives to water immersion because water immersion is not practical for large groups of athletes, with short breaks between events. In this study, hand immersion, whole body fanning, air-cooled garments, liquid-cooled garments and phase-change garments were compared to determine their capacity to extract heat in exercising participants in hot humid conditions. Whole body fanning was found to be the most effective and cost-effective technique for these participants. This technique is ideal for athletes because they wear little clothing and sweat profusely, and this technique may be used on more than one of athlete at a time (Barwood et al., 2009).

1.3.3 Cooling methods used for domesticated animals

Cooling studies in animals have focused on domesticated and farm animals, such as pigs, dogs, cattle and horses, and no studies have been done on wild animals. In all these studies the animals have been able to practise normal autonomic thermoregulation, a scenario unlike that in captured wild animals in which the animals are immobilised and the animals may be unable to thermoregulate as they would if free-living.

Animals have been used as models to study malignant hyperthermia and methods of inducing therapeutic hypothermia. Pigs, especially, have been used because they are susceptible to hyperthermia (Holtzer et al., 2005; Rosenberg et al., 2007). The difference between malignant hyperthermia and capture-induced hyperthermia is that malignant hyperthermia is caused by a genetic disorder with a metabolic dysfunction that is triggered by drugs (by anaesthetics such as halothane) or rarely with exercise and excitement (Rosenberg et al., 2007), whereas capture-induced hyperthermia is caused by stress (Meyer et al., 2008). Malignant hyperthermia can be alleviated with the administration of dantrolene but the administration of dantrolene does not alleviate other forms of hyperthermia (Smith, 2005; Rosenberg et al., 2007) and has not been tested as a method to alleviate capture-induced hyperthermia.

Animal models have also been used to investigate methods in treating heatstroke in man (Table 1.3). Techniques that have been investigated include the use of iced gastric lavage at a water temperature of 0 to 1°C and peritoneal lavage at a water temperature of 6°C in dogs in which heatstroke had been induced. Both techniques cooled the dogs significantly quicker than the controls in which no cooling was applied (Bynum et al., 1978; Syverud et al., 1985). Another study, also using dogs, investigated water immersion and compared the effect of different water temperatures on the rate of cooling. The water temperatures investigated were ice water (1 to 3°C), cold water (10 to 11°C) and tap water (15 to 16°C) on dogs that had collapsed from heat stroke in a climatic chamber. The findings were that all the water temperatures led to equal cooling rates (Magazanik et al., 1980).

Animals have also been cooled after intense exercise to investigate the best technique to alleviate exercise-induced hyperthermia. Cooling in exercised dogs with ice-packs has been compared to passive cooling (Kruk et al., 1985). The ice-packs were placed around the dog's trunk and were successful in decreasing the rectal temperature and increasing the exercise performance of the dogs. Substantial work also has been done examining cooling in exercising horses. Horses, especially race horses, exercise outdoors in hot, humid conditions and undergo intense exercise during races, which may cause a hyperthermia. Race horses can produce high metabolic heat loads because of their large metabolic capacity. Because of their smaller surface area to volume ratio, compared to humans, it can be difficult for horses to dissipate the heat generated after exercise. Therefore, horses, like humans, can succumb to heat stress and heat stroke (Hodgson et al., 1993; Kohn et al., 1999). The use of dousing with cold water (9°C to 15.6°C) on hyperthermic horses has been investigated and is the accepted treatment for hyperthermic horses; the cold water-dousing significantly decreased rectal and pulmonary artery temperature (Williamson et al., 1995; Kohn et al., 1999). The hydration status, muscle health and serum electrolyte levels of the horses were unaffected by the cold water-dousing and so it was concluded that this is a safe and effective cooling technique (Kohn et al., 1999).

In cooling of farm animals, most studies have focused on environmental management of animal housing. Environmental management is especially important in pigs and poultry, which are not capable of sweating, but environmental management is important in any housed species as it influences yields and reproductive performance (Aksit et al., 2006; Haeussermann et al., 2007). For example, lactating cows have decreased milk yields when they are exposed to high ambient temperatures (Chaiyabutr et al., 2008). Stress (both

emotional and physical) from inappropriate environmental management in housed animals causes an increase in body temperature, acidaemia and an increase in muscle metabolism (Wariss, 1990; Sigholt et al., 1997). Also, Wariss (1990) found that emotional and physical stress in cattle before slaughter caused an increase in glycolysis (the utilization of stored glycogen for energy), which resulted in a decrease in liver yield, overall carcass yield and a decrease in meat quality. Poorer quality meat has an altered colour, texture and is less palatable, which decreases the economic value of the meat in both domesticated animals and captured wild animals (Wariss, 1990; Sigholt et al., 1997). It is therefore important to keep farmed animals comfortable and cool to gain optimal economic value from their carcasses.

For effective environmental management, combining cooling methods, such as wetting animals and increasing air movement over them, is recommended. Animals may be wet by the use of intermittent sprinkling; the recommended water droplet size for intermittent sprinkling is to be as large as (or larger than) than 150 microns to better saturate hair (Gaughan et al., 2004). Intermittent sprinkling in the environmental management of animals can be combined with a method that lowers the metabolic heat production of farm animals, such as decreasing their feed intake (Gaughan et al., 2004; Mader and Davis, 2004). The air movement over farm animals can also be altered to increase convective and evaporative cooling. Where increased air flow is used in the environmental management of farm animals the recommended air speed over cattle is 2 to 4ms⁻¹ (Gaughan et al., 2004). To further increase the evaporative cooling effect of the airflow over farm animals the air in the animals' housing can be forced through cooling pads by fans. One study (Chaiyabutr et al., 2008) investigated the effect of increasing evaporative cooling in the animal house by

forcing air through cooling pads first and found that the cooled animals had significantly lower rectal temperatures and an increased milk yield in comparison to non-cooled cows (Chaiyabutr et al., 2008). However, often environmental management only increased the thermal comfort of housed animals, or tested the cooling preferences of housed animals given the choice of different pens that had either sprinkler systems, increased air flow or floor cooling (Bull et al., 1997; Huynh et al., 2006; Silva et al., 2006; Barbari and Conti, 2009). The emphasis in these studies is to increase productivity and reproductive performance. However, these studies increased the thermal comfort of the animals rather than necessarily lowering of core body temperature of the housed animals. However, in a study in pigs (Bull et al. 1997), in which pigs had the choice of selecting different pens with different cooling treatments, there was a significant decrease in the pigs' rectal temperatures in pigs that preferred a conductive cooling pad on the floor.

1.3.4. Cooling methods used for game animals

The cooling methods in wild animals in Africa follow capture guidelines, which recommend placing animals in the shade (but with heavy animals, such as rhino, moving an immobilised animal is not always possible) and to have plenty of water available for dousing the animal (Meltzer et al., 2006b). Other recommendations to treat capture-induced hyperthermia include the use of a portable mist sprayer and intravenous shock therapy to cool animals (Meltzer et al., 2006a). In North America specific guidelines exist for the capture of wild sheep. The use of water rubbed into the hair is recommended, especially rubbing the water into the neck, belly and groin. Also recommended is to use a mixture of part water, part alcohol for rubbing to increase evaporative cooling and the use of a cold water enema by

applying a human enema bag into the anus of wild sheep that have a body temperature greater than 41°C (Foster, 2005). The guidelines for brown bears, gray wolves, wolverines and lynxes, in Europe, define an animal as hyperthermic if their body temperature exceeds 40°C and recommend treating the hyperthermia by applying snow or water (depending on the season) to the axilla, groin and tongue. If the body temperature continues to increase to greater than 41°C then an intravenous infusion of 10-15ml.kg⁻¹.hr⁻¹ of Ringers lactate is then recommended (Arnemo and Fahlman, 2008). Different capture guidelines and cooling protocols therefore exist for different species in different climates.

Yet, dousing with water is the cooling method employed most routinely during capture (Kock and Meltzer, 2006; Meltzer et al., 2006b). Wild animals are usually doused with water that is at an ambient temperature. That is, that the water storage containers are often left in the sun and the water becomes warmed (the water may even exceed air temperature if the water container is exposed to a sufficiently high radiant heat load). Even if the water is warmed, but has a temperature still lower than body temperature, there is a question if this method is effective enough to rapidly cool hyperthermic animals. This is because the methods used to cool animals in the field are based largely on anecdotal evidence that they appear to cool an animal. As far as I am aware, there has been no scientifically sound study to validate any of the techniques used to cool captured animals.

In trying to determine the most effective cooling methods for wild animals, unfortunately, it must be recognised that the most effective methods in humans are not always feasible for use on animals in the wild. For example, extracorporeal blood cooling is a method of rapid, direct core cooling in a clinical setting, but is impractical for wild animals in the field;

extracorporeal blood cooling requires specialised apparatus such as cardiopulmonary bypass machines (Schneider et al., 2006). Using an ice immersion in an ice-water slurry would be impractical in the field, as the amount of ice that is used to cover a human patient could amount to 100kg (Plattner et al., 1997), and therefore to cool large animals in this way is not feasible. Indeed the use of any water immersion technique would be difficult because studies in humans utilize large water baths (Clements et al., 2002) and it would be impossible to take a large enough tub into the field, yet alone enough water to fill such a tub. Therefore, other methods which mimic water immersion, like dousing the body surface of captured animals with water, are more feasible. Water-dousing also has been shown to be an effective cooling method in horses with exercise-induced hyperthermia (Kohn et al., 1999).

In domesticated animals manipulating the environmental conditions is the most common method of cooling animals. However, it is not feasible to alter the environment for wild animals, as wild animals are caught in field conditions and, if they are housed, they are housed typically in open pens, such as in bomas. However, environmental management may be possible in certain circumstances, such as in trucks in which animals are transported.

Other methods are more practical and show promise in cooling wild animals, such as cooling by the application of ice-packs to areas where large blood vessels run (Clements et al., 2002). An added possible advantage of this technique in animals is that the axilla and groin regions, which have large superficial blood vessels, have less fur, which would increase skin contact with the cooling substance and therefore possibly increase body cooling. Smith (2005) has suggested that in humans for this technique to be effective it must be combined

with another cooling technique. However, ice-packs placed over the dorsal body surface of hyperthermic dogs effectively lowered the body temperature of the dogs (Kruk et al., 1985). Another technique that could be transposed from humans to wild animals is the infusion of 4°C ice-cold IV fluid as it was shown to be effective in lowering body temperature over an hour in humans (Schneider et al., 2006).

1.4 Dissertation aims

My study therefore investigated cooling methods on hyperthermic animals with the aim of determining the most effective, practical and economical means of reducing body temperature during capture. We doused hyperthermic blesbok after capture with different water temperatures of 4°C, 17°C and 28°C and determined the effect that each water temperature had on decreasing the body temperature. We investigated the effect of fanning in combination with 28°C water dousing on lowering body temperature and compared it to dousing with 28°C water alone. We also investigated alternative methods to water dousing that could be utilised on captured animals. Hyperthermic captured blesbok were covered in ice-packs on their dorsal body surface and also had ice-packs placed under their axilla, ventral abdomen and groin. The hyperthermic blesbok also were sprayed with a fine mist spray at a water temperature of 28°C and were infused with a 1l intravenous saline infusion at 4°C. The ice-packs, mist spray, cold IV infusion were compared against each other and to a 28°C water-dousing intervention to determine the efficacy of each technique in lowering body temperature. It is important to note that the detrimental effects of hyperthermia not only depend on the magnitude of the hyperthermia but also on the duration of the hyperthermia (Kosaka et al., 2004). It is therefore important that an

effective cooling technique be rapid and produce a significant (at least 2°C) decrease in body temperature. The efficacy of a technique was therefore determined through the rate and magnitude of cooling.

In this chapter the problem of how animals become hyperthermic during capture is outlined and it is shown that although there has been much investigation into cooling humans and domesticated animals, there has been no published research on cooling hyperthermic African wildlife after capture. In chapter 2 the experimental protocol including surgery protocol, equipment used and study animals is described. I describe how we were able to record body temperature continuously through surgically implanted data loggers and how we elicited a hyperthermia prior to the start of our cooling interventions. The specific details of each cooling intervention (water volumes and cooling procedure) are also described in chapter 2. Chapter 3 illustrates the results of our key findings through the analysis of the body temperature measurements taken in our study animals. The normal body temperature of the blesbok, peak body temperatures, how quickly animals were cooled and how effectively the blesbok were cooled by each intervention is shown. Chapter 4 discusses the importance of these results in relation to the outcome for best cooling methods. In this chapter we show how effective the placement of ice-packs and dousing with water were in decreasing body temperature and how ineffective, overall, the IV infusion and mist spray were in alleviating capture-induced hyperthermia. Lastly chapter 5 contains my concluding remarks and suggestions for future research solutions.

CHAPTER 2:

MATERIALS AND METHODS

2. MATERIALS AND METHODS

2.1 Subjects and study site

Nineteen adult blesbok (*Damaliscus dorcas phillipsi*; eight males and eleven females) were implanted with miniature temperature-sensitive data loggers during surgery. Data collection for this study occurred in January and February 2009, in the southern hemisphere summer. The data loggers were implanted in December 2008 and complete data sets were retrieved from 12 animals in March 2009. Seven animals were not included in the results of this study as we did not obtain sufficient data due to logger failure in those animals.

The study was carried out at the National Zoological Garden's Biodiversity Conservation Centre in Lichtenburg (26°06'55" S, 26°09'50" E) (project number P08/20). The Centre is a 4500ha nature reserve situated in the North-West Province, South Africa. Animals were captured from the reserve by using a passive boma-capture technique. After the capture the animals were housed in boma units (two adjoining bomas, each of 5m x 5m), in groups of two to three animals per boma unit. The animals were identified by coloured and numbered ear-tags. The staff members at the Centre were responsible for animal husbandry, which included water *ad libitum*, a diet of Lucerne supplemented by antelope pellets and regular boma cleaning. All experimental and surgical procedures were approved by the Animal Ethics Screening Committee of the University of the Witwatersrand (clearance number 2008/34/04).

2.2 Surgery

2.2.1 Anaesthesia

A temporary surgical theatre was set-up in a roofed area adjacent to the bomas. A combination of etorphine hydrochloride (3-3.5mg intramuscularly - IM, M99, Novartis, Johannesburg, South Africa) and azaperone (40-60mg IM, Stresnil, Kyron Laboratories, Johannesburg, South Africa) was used to dart and immobilize the blesbok for surgery. These doses were later repeated in our experimental procedures (Section 2.2.2). The sites for data logger implantation were shaved and sterilized with chlorhexidine gluconate (Hibitane, Astra Zeneca, Johannesburg, South Africa) and the incision line was injected subcutaneously (SC) with 1ml of a local anaesthetic (Lignocaine Injection 2%, Centaur Labs, Johannesburg, South Africa). Once the animals were immobilised, anaesthesia was maintained with isoflurane (Isofor, Safe Line Pharmaceuticals, Johannesburg, South Africa), 1-4% in 100% oxygen, via a facemask.

2.2.2 Implantation and removal of data loggers

A miniature Stowaway temperature-sensitive data logger (see Section 2.3 for logger details), covered in inert wax (Sasol wax EXP987, Sasol, Johannesburg, South Africa), was surgically implanted into the abdomen through an incision in the right para-lumbar fossa (Figure 2.1). This logger was not tethered but could move freely in the abdominal cavity to record core body temperature. To measure subcutaneous temperature four pairs of iButton (see Section 2.3 for logger details) temperature-sensitive data loggers were implanted subcutaneously (Figure 2.2) at



Figure 2.1 The typical size of the Stowaway miniature data logger, waxed (left) and unwaxed (right), which measured core body temperature intra-abdominally.



Figure 2.2. The typical size of the iButton miniature data logger unit, waxed (left) and unwaxed (right), which measured subcutaneous body temperature.

four sites. A pair of these data loggers was covered in inert wax to make one logger unit. Two of these logger units were implanted in the neck (in the region of the upper neck and lower neck), one logger unit was implanted in the flank (behind the last rib and before the hind limb) and the last logger unit was implanted in the groin (Figure 2.3).

During surgery each animal received an injection of a long-acting penicillin (5-6ml IM Peni La Phenix, penicillin, Virbac Animal Health, Centurion, South Africa), an analgesic anti-inflammatory (1ml IM Phenylarthrite injectable solution, phenylbutazone, Bayer Animal Health, Centurion, South Africa or 3-5ml IM Dexamethasone, Centaur Labs, Johannesburg, South Africa), a Vitamin B injection (4ml IM Rucenta Vitamin B Complex, Rucenta Medical Supplies, Johannesburg, South Africa) and a parasiticide (0.5ml SC Dectomax, Pfizer Laboratories, Sandton, South Africa). The surgical procedure lasted about 30 minutes and the respiratory rate, peripheral haemoglobin oxygen saturation, heart rate and rectal temperature were monitored throughout the surgical procedure. To prevent fly worry, myiasis and infection post-surgery the surgical wounds were sprayed with a topical antiseptic spray (Necrospray, Centaur Labs, Johannesburg, South Africa) and covered with an anti-tick grease (Tick grease, cypermethrin 0.025% m/m, Bayer Animal Health, Centurion, South Africa).

After surgery the pharmacological action of etorphine was reversed with diprenorphine hydrochloride (M5050 0.7ml IV, Novartis, Johannesburg, South Africa) and the animals were released back into the bomas and their health was monitored closely for three days. The blebsok were allowed one month to recover from the surgery before the commencement of our cooling experiments.

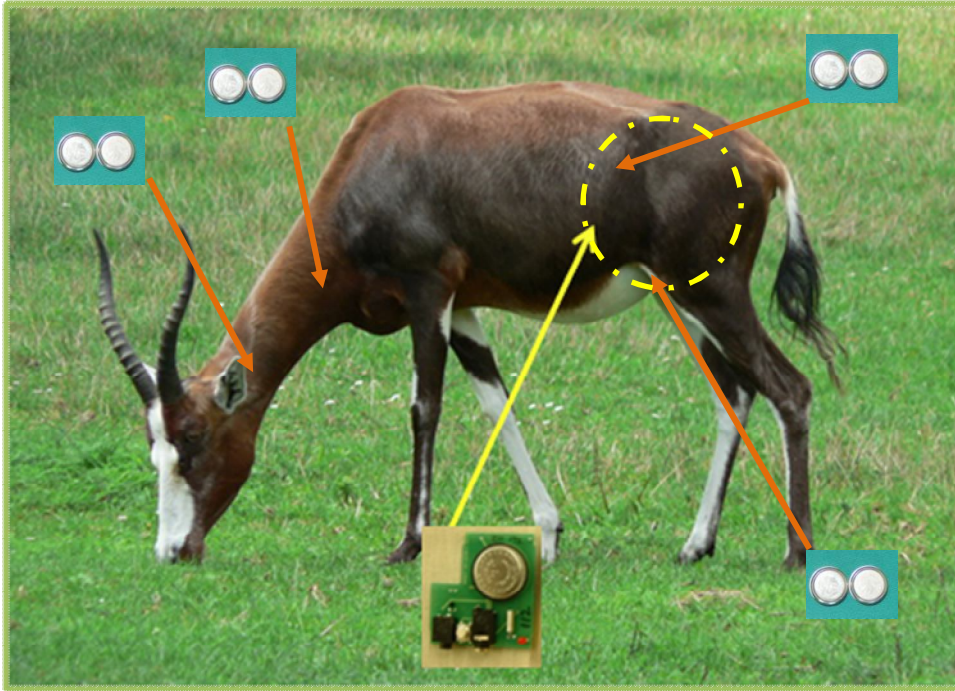


Figure 2.3 The data logger implantation sites in the blesbok. The iButton logger units remained fixed at their subcutaneous implantation sites, whereas the Stowaway loggers moved freely in the abdomen, but ultimately settled in (and were removed from) the pelvic canal.

A similar surgical and anaesthetic procedure was used to remove the loggers at the end of the study in March 2009. The blesbok were once again tranquilised and anaesthetised for the removal of the loggers. The loggers used to measure core body temperature were found and recovered from the pelvic canal area of the abdomen. Except for one of these loggers, the loggers were not encapsulated in connective tissue. The loggers to measure subcutaneous temperature were found at the implantation sites. For post-surgical recovery the blesbok were then released into a 75ha camp in the reserve.

2.3 Body temperature measurements

2.3.1 Core body temperature

The Stowaway miniature temperature-sensitive data logger, size 40 x 40 x 20mm, (StowAway XTI, Onset Computer Corporation, Pocasset, Massachusetts, United States of America) that was implanted into the abdomen, and recorded core body temperature continuously throughout the study period, had a resolution of 0.04°C and a range of +32°C to +46°C. Prior to surgery the logger was set at a recording interval of 6 minutes and covered in inert wax. The loggers were calibrated against a high accuracy thermometer (Quat 100, Heraeus, Hanau, Germany), in an insulated water-bath, to an accuracy of 0.05°C. Prior to implant surgery the logger was sterilized with formaldehyde vapour (Paraformaldehyde, Kyron Laboratories, Johannesburg, South Africa) in a sealed drum.

2.3.2 Subcutaneous temperature

The iButton miniature temperature-sensitive data loggers, size 20 x 20 x 10mm, (iButton DS1922T, Maxim, Dallas Semiconductor, Dallas, Texas, United States of America) that were implanted to measure subcutaneous temperatures had a resolution of 0.0625°C and range of 0°C to 125°C. These loggers were set to record at an interval of every 6 minutes. Due to their small memory (4 kb) two loggers were sequentially programmed (one was on a delayed start) to continuously record subcutaneous temperature over the entire study period at each site. These two loggers were waxed as one unit and were implanted at each site. Similarly to the abdominal loggers these loggers were also calibrated prior to surgery to an accuracy of 0.05°C and sterilised in formaldehyde vapour. Both the subcutaneous and the abdominal loggers were calibrated three times before implantation surgery to determine if there was any temperature drift (the change in accuracy of the temperature recordings over time). It was determined that there was no change in the accuracy of the temperature recordings over time.

2.4 Experimental procedure

After the post-surgical recovery period the experimental trials commenced. Blesbok were darted at a weekly interval from January 2009 to February 2009 on five separate occasions. Some animals were darted on a sixth occasion to repeat cooling interventions that had been confounded by rainy weather. The time of day at which capture, darting and the cooling interventions took place was standardised, so as to minimise the variability in each group of animals that may have occurred as a result of the animal's circadian rhythm and the ambient environmental conditions. The first group to be darted and to receive their cooling

intervention was always darted between 9:29 and 9:59. The next group was then be darted an hour later after the first group and so on for all the groups. This ensured that darting always occurred within the same time interval for all the groups throughout the investigation.

To induce hyperthermia before the start of the cooling interventions, the blesbok from one housing boma unit, which housed two to three blesbok, were chased through a boma corridor into a larger boma (10 x 30m) where they were encouraged to keep running by investigators clapping hands, as required, for ten minutes. After ten minutes they were herded back to their housing boma (Figure 2.4). Following this exercise, they were immediately darted (with 3-3.5mg etorphine and 40-60mg azaperone) and immobilised for the cooling intervention. An assessment of the depth of immobilisation was performed by team members scoring how much movement was present in the blesbok's limbs and body and also scoring how much shivering occurred during immobilisation.

After darting, each blesbok was kept immobilised for 30 minutes, during which the animal was orientated facing the sun so that the sun's rays fell on the blesbok's back evenly. During each trial care was taken to ensure animals were not shaded by the researchers and animals lay on the same sandy substrate of the boma housing floor. The blesbok were also placed in sternal recumbency with the head above the thorax, the nose pointing downwards, and the head aligned with the spinal column, so as to prevent obstruction of the airways and to allow ruminal gas to escape. Cotton wool was placed into the ears of the blesbok and the blesbok were blindfolded, to reduce the stress of external auditory and visual stimuli.

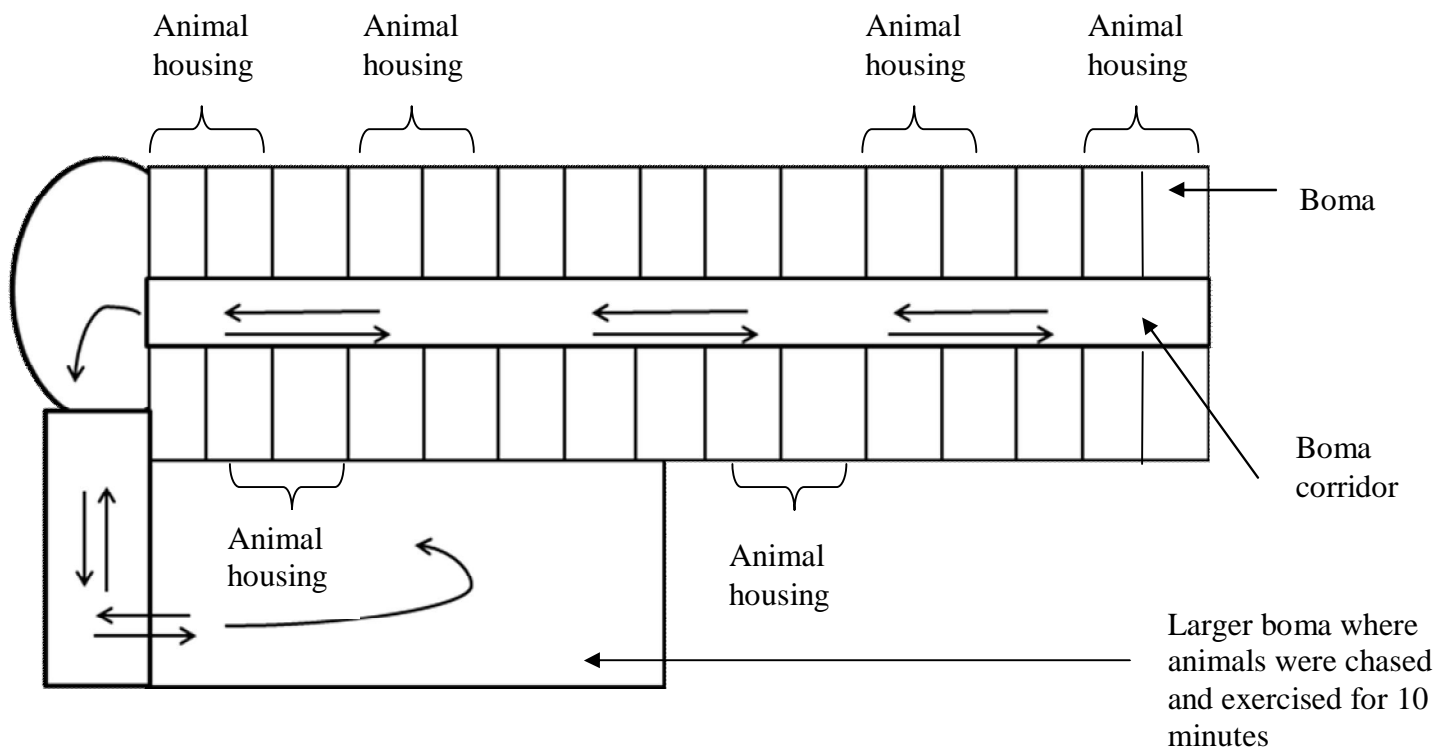


Figure 2.4. Schematic representation of the bomas and chase sequence.

Equipment was placed by the investigators onto the blesbok five minutes after they were darted to measure their vital signs and skin and rectal temperature. Skin temperature was measured by placing a thermistor (bead diameter 1.5mm, 27-10K4A801, Onset Computer Corporation, Pocasset, Massachusetts, United States of America). The thermistor bead had been attached to a washing line peg that was then positioned over the skin of the inside of the ear of the blesbok. The ear temperature was logged at a two-minute interval by attaching the thermistor probe to a temperature-sensitive data logger (StowAway XTI, Onset Computer Corporation, Pocasset, Massachusetts, United States of America). Rectal temperature was measured by either, or both, a thermocouple (copper constantan insulated thermocouple wire, NN-T-24, Omega Engineering, Connecticut, United States of America) connected to a digital reader (BAT-12 Microprobe Thermometer, Phsyitemp Instrumments, New Jersey, United States of America) and a veterinary mercury rectal thermometer. However, we were only able to obtain sporadic readings and a future study would have to further elucidate the real time rectal temperatures of immobilised blesbok during cooling. The animal's heart rate and peripheral haemoglobin oxygen saturation were measured with a veterinary pulse oximeter (Nonin 9847V, Nonin Medical, North Plymouth, USA) and respiratory rate was assessed by counting the breaths per minute.

Cooling interventions took place five minutes after the equipment was placed on the blesbok. The blesbok were divided into two groups, a water-dousing group (n = 5) and an alternative methods group (n = 7), to investigate different cooling interventions. Both groups had a control (animals that received no cooling intervention, and were merely chemically immobilised for 30 minutes) and a 28°C water-dousing intervention to compare against common practice. The water temperature of 28°C was selected because the water

used in common practice in South African game capture can equilibrate to environmental temperatures while being transported to capture sites. Prior to this study, we tested how warm water, in a typical plastic drum used in the field, becomes when left in the sun on typical summer days by measuring the temperature of the water within the drum and the ambient air temperature. The water, within the drum, when left in the sun warmed to about 28°C, which was approximate to the ambient temperature.

Within the water-dousing group we investigated the cooling efficacy of different water temperatures, namely a cold temperature of 4°C, a cool temperature of 17°C and a tepid temperature of 28°C (Table 2.1). In this group we also examined if the addition of fanning air over an animal doused with the 28°C water-dousing (28°C+fanning) improved the efficacy of cooling with 28°C water-dousing. Animals were doused with a total of 10l of water over the dorsal body surface. The first 5l was poured slowly over the animal and rubbed into the skin, five minutes after the animal had been immobilised. The remaining 5l was poured 15 minutes after immobilisation. At each pouring, it took about five minutes to pour the total volume of 5l over the animal. In the 28°C+fanning intervention, the fanning was done by manual means using a rigid plastic tea tray, a size of 531mm x 381mm (Rec1, Blue Swallow, Kraaifontein Industria, South Africa), which was waved up and down to produce a wind of $1.2 \pm 0.6\text{ms}^{-1}$.

Within the second group - the alternative methods group - we investigated alternatives to water-dousing cooling methods (Table 2.1). One intervention was to soak the animal with a fine mist spray of 28°C water, which was generated by a manual garden sprayer ($0.435\text{l}\cdot\text{min}^{-1}$, Premium 5l Pressurised Sprayer, Model G-2317, Shih Kuo, Taiwan). The fine mist spray

Table 2.1 A summary of animal body masses and cooling interventions for both the water-dousing group and the alternative methods group.

Group	Mean \pm SD body mass (kg) at start of study		Cooling interventions
	(December)	(March)	
Water-dousing group (n = 5)	42.2 \pm 10.3	44.4 \pm 8.3	Control 28°C water dousing 4°C water dousing 17°C water dousing 28°C water dousing with the addition of fanning
Alternative methods group (n = 7)	51.8 \pm 2.6	47.6 \pm 4.0	Control 28°C water dousing Fine mist spray at 28°C One litre of sterile isotonic saline at 4°C (IV) Ice-packs

was applied to the dorsal body surface of the animal at five minutes and 15 minutes after the animal was immobilised. The mist spray was sprayed up and down the body surface (not wetting the upper neck) for a five-minute interval. Another intervention in this group was infusing 1l of sterile isotonic saline (Adcock Ingram Critical Care, Johannesburg, South Africa) at 4°C via a Jelco and 18 gauge catheter (BD Insyte IV catheter, 1.3 x 30mm, 105ml.min⁻¹, BD, New Jersey, United States of America) into the cephalic vein in the blesbok's forelimb. The solution was infused as fast as possible (an average time of 12.75 ± 1.47 min) until it was finished. Prior to the start of the study I had calculated the theoretical cooling potential of the IV infusion on the blesbok. The cooling was calculated from the proportions of body mass and temperature using the following equations:

$$(\text{mass of animal} \times \text{initial body temperature}) + (\text{mass of fluid} \times \text{temperature of fluid}) = (\text{mass of animal} + \text{mass of fluid}) \times X$$

Initial body temperature – X = change in body temperature

X in the above equations is the final body temperature after the fluid infusion. For a body mass of 45kg, infused fluid of 1kg at a temperature of 4°C, and an initial body temperature of 41°C, then X equals 40.2°C. Therefore the theoretical change in body temperature would be 0.8°C.

The other intervention tested conductive cooling by us placing ice-packs (Techni-Ice, Reusable Dry Ice Packs, HDR Model, Victoria, Australia) over the dorsal body surface of the blesbok and onto areas of less fur and good superficial blood supply (the groin, lower neck and axilla) for 25 minutes. This group also contained the 28°C water-dousing (same pouring procedure of 10l as in the water-dousing group) and a control. The interventions were

randomised within the groups between the animals and each animal in the group received all the interventions of that group.

To achieve the desired water temperatures, I had cooled the 5l water bottles in a fridge (to achieve the 17°C water temperatures) or heated the 5l water bottles in a water bath to the 28°C temperature. The ice-packs were kept in the freezer compartment of the fridge to remain frozen until the start of the cooling intervention. Five-litre water bottles and isotonic saline drip bags were also kept temporarily in the freezer to achieve the 4°C water temperature. Prior to the start of the intervention I had tested the water temperature of the 5l water bottles by using a thermometer (SAMA Partial, 76mm Immersion Thermometer, CP15, Brannan, England) to confirm the correct temperature. The water bottles, drip bags and ice-packs were transported and stored in cooler boxes for a short period (no more than 10 min) prior to the start of the interventions.

After 30 minutes of immobilisation, the immobilising effects of etorphine were reversed with 0.6-0.7ml of diprenorphine hydrochloride. Once a set of two to three animals had received an intervention, the process was repeated with the next set of animals. Post-intervention, I and the investigative team monitored the animals for thermoregulatory behaviours such as shivering, huddling, panting, lying or standing. An overview of the experimental procedure can be seen in Figure 2.5.

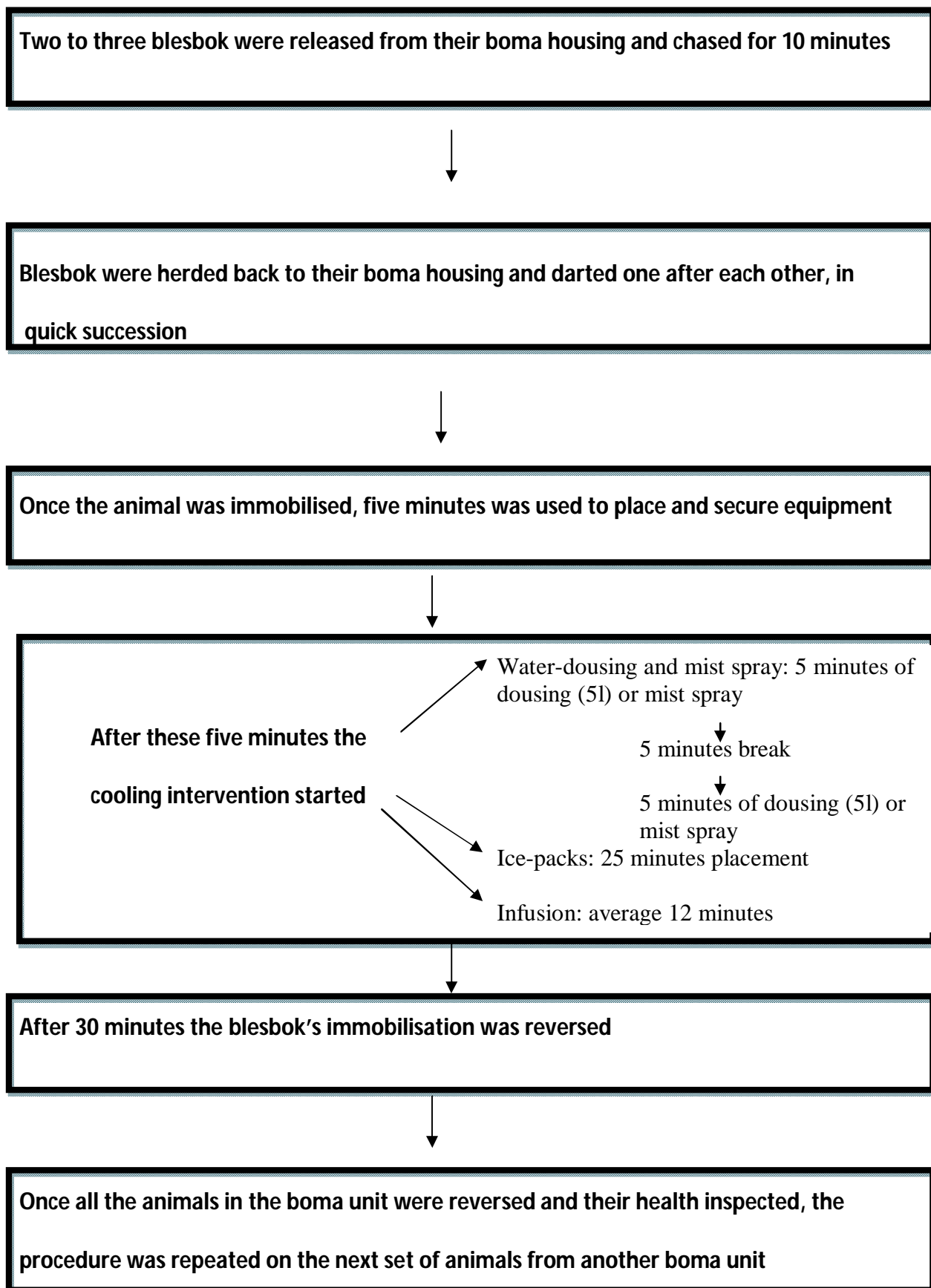


Figure 2.5 A flow diagram showing the order of the experimental procedures.

2.5 Microclimate measurements

Microclimate measurements were taken to determine the ambient environmental conditions. A sling psychrometer (Whirling Hygrometer, M112022, Casella Measurement, Bedford, England) was used to determine wet and dry bulb temperature. Vapour pressure was then measured from a psychrometric chart of 85 kPa (Barenbrug, 1974) using the dry and wet bulb temperatures. An anemometer (Thermo-Anemometer, GGA-65, Alnor, Finland) was also used to determine wind speed by placing the sensor 100 mm from the animal and also by standing in the sun and holding the sensor 1.5m above the ground in the air. The black globe temperature, which integrates the effects of air temperature, wind speed and solar radiation and therefore provides an index of the environmental heat load on an animal (Yaglou, 1968) was also measured.

2.6 Data analysis

Results are reported as mean \pm standard deviation (SD) and $P < 0.05$ was considered statistically significant. I used a paired Student's t-test for pair-wise comparisons, while a repeated measures analysis of variance (RM-ANOVA) was used for multiple comparisons. Statistical values were calculated from Graphpad Prism 5 (GraphPad Prism version 5.00 for Windows, GraphPad Software, California, USA). The results reported are body temperature, which was measured intra-abdominally, unless otherwise stated as subcutaneous temperature.

The thermo-sensitive logging equipment recorded data at an interval of 6 minutes for a month prior to the start of the interventions, allowing us to establish the normal body temperature pattern of the blesbok ($n = 12$). This normal pattern was determined from ten days after surgery up until two days before the start of interventions (a total period of 23 days) by averaging each six-minute recording of body temperature to determine the 24 hour rhythm of the blesbok.

For analyses, the time of the start of the cooling intervention was taken as time zero. The peak temperature elicited after the chase procedure was the absolute peak temperature within 30 minutes after time zero (as there was never a peak temperature before time zero during the experimental procedure). The minimum body temperature was calculated as the absolute minimum reached within an hour from time zero; a one-hour period was analysed because the body temperature decreased up to an hour after the start of the cooling intervention. The change in body temperature after cooling (the amplitude of cooling) was calculated as the difference between the body temperature at time zero and the body temperature at the time of 30 minutes and one hour. The time of 30 minutes was analysed for the amplitude of cooling because the cooling interventions were applied during that interval of 30 minutes. In addition, an hour was analysed because the effect of the cooling interventions was seen typically up to an hour after the onset of the cooling (time zero). Thermal response indices (TRI) are the areas under the temperature curves and are used as a measure of the extent of cooling, integrated over time. The TRI was calculated from the time integral of the change in body temperature from time zero. The greater a negative TRI value, the larger the area under the temperature curve, and therefore the greater the integrated temperature decrease over time. The rate of cooling is a measure of how quickly

the interventions decreased body temperature. It was calculated from the slope of the body temperature curve over 30 minutes, that is, the minimum temperature within 30 minutes was subtracted from the peak temperature, then this value was divided by the time difference between the peak and minimum body temperature (this calculation assumed a mostly linear change in abdominal temperature, this linear change can be seen later in Section 3.3).

The temperature difference between the body core and the subcutaneous layer was calculated as the difference between core temperature and subcutaneous temperature. Subcutaneous temperature was calculated as the average of three subcutaneous measurement sites over which cooling interventions were applied (the lower neck, the flank and the groin). To compare the core – subcutaneous temperature difference curves between interventions, areas under the curve and peak differences for the period of time zero to time one-hour were calculated. The core - skin temperature difference also was determined by subtracting the ear skin temperature from the core temperature between the interval of time zero and 24 minutes. The purpose of this calculation was to estimate if vasoconstriction was occurring in the peripheral vasculature. This interval was analysed to compare the differences when the cooling intervention had started to when it had finished. The interval of 24 minutes and not 30 minutes (like the other analyses) was used (and was analysed with a paired Student's t-test) because the cooling intervention would have just been completed at time 24 minutes (at 30 minutes the animals may have had a sufficient length of recovery time from the cooling intervention to return any changes in ear skin vasculature back to normal). A TRI was also analysed for the interval of time zero to 24 minutes to also estimate if there had been vasoconstriction present during the 24-minute

interval. In this instance a one-way ANOVA was used because of two missing data sets in the ear skin temperature data.

CHAPTER 3:

RESULTS

3. RESULTS

3.1 Normal body temperature rhythm

In blesbok, normal body temperature fluctuates cyclically during the day; the temperature follows a regular pattern every day and a clear 24 hour body rhythm is exhibited (Fig. 3.1). Figure 3.1 shows that body temperatures fluctuates daily by about 1-2°C from a minimum about 38.5°C to a maximum of about 40°C. The figure depicts the body temperature of one blesbok but this cyclic fluctuation was typical for all the study animals.

The average 24 hour body temperature on days when capture did not occur (23 days) is illustrated in Figure 3.2. The mean 24 hour body temperature of all the blesbok (12 animals over 23 days) was $38.78^{\circ}\text{C} \pm 0.37^{\circ}\text{C}$. Daylight hours occurred from about 05:00 to about 19:00. The mean blesboks' body temperature revealed a minimum of $37.88^{\circ}\text{C} \pm 0.09^{\circ}\text{C}$ at about 08:30 each day. During the course of the day the body temperature increased to reach a maximum of $39.37^{\circ}\text{C} \pm 0.11^{\circ}\text{C}$ in the evening at about 18:00. On average, the body temperature during the night was $38.83^{\circ}\text{C} \pm 0.26^{\circ}\text{C}$ and during the day was $38.73^{\circ}\text{C} \pm 0.54^{\circ}\text{C}$. These values did not differ significantly (Student's t-test, $P = 0.092$, $t = 1.69$).

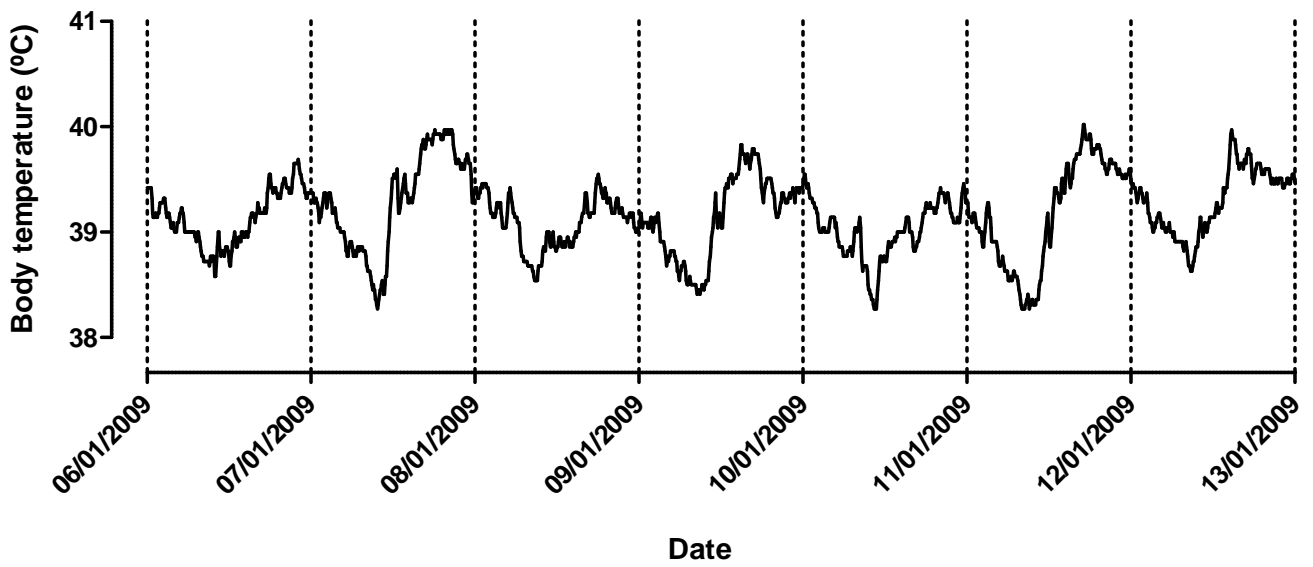


Figure 3.1 The body temperature of one blesbok over seven days.

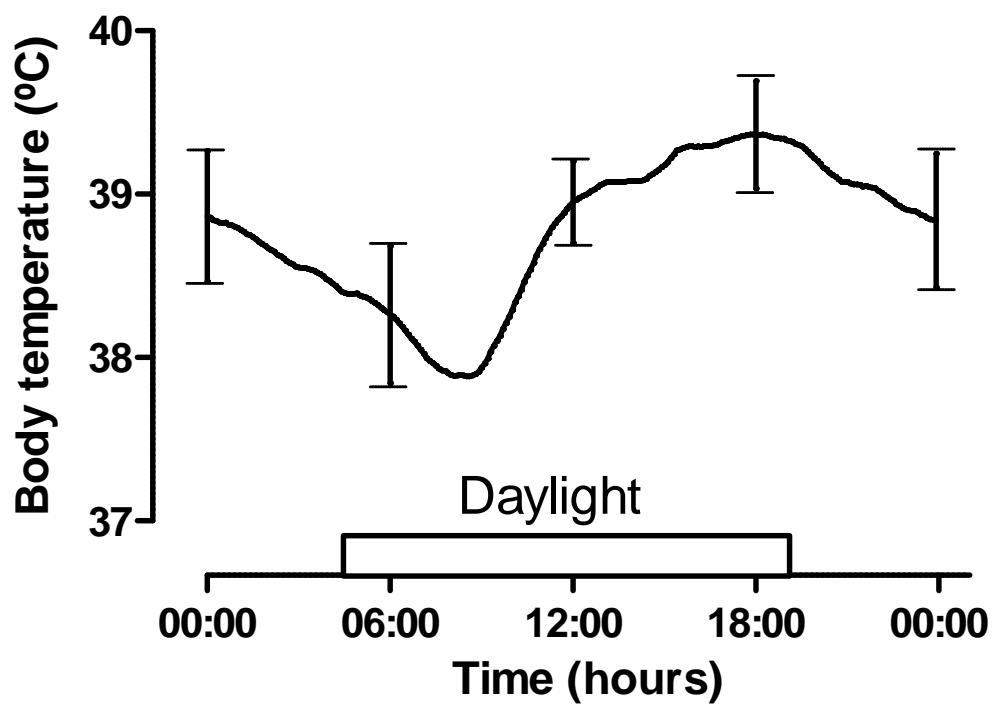


Figure 3.2 The typical mean body temperature rhythm of all the blesbok shown as temperature over 24 hours. The solid bar indicates daylight hours. The minimum occurs in the morning and the peak temperature occurs before sunset. Vertical bars (I) indicate the standard deviation from mean body temperature at that hour.

3.2 Body temperature changes during capture

Our exercise protocol and darting mimicked a capture procedure and induced a precipitous rise in body temperature (Fig. 3.3) that was significantly different from the normal body temperature before capture (the peak temperature was compared to the normal temperature for all capture events, Student's t-test, $P = 0.0001$, $t = 10.79$). This sudden peak in body temperature depicted in Figure 3.3 was about 42°C and 41°C for that animal. Figure 3.3 depicts one animal's body temperature change during capture, but this precipitous rise in body temperature was typical for all the blesbok when captured.

In Figure 3.4 the different stages of the capture protocol that was used to elicit a capture-induced hyperthermia are depicted. The figure depicts the experimental procedure times for when the cooling intervention was started (time zero), when the cooling intervention was completed, when the animal was given a reversing agent for the action of the opioid and when the animal was up and awake. The figure also shows the normal body temperature that would have occurred at that time, without capture and cooling, which was calculated from the mean normal body temperature of that blesbok at that time (the natural history of the animal) on days without capture (23 days). Figure 3.4 reveals that the temperature increased during chasing and darting of the animals. Once an animal was fully immobilized and recumbent, the cooling intervention was applied (time zero) and there was a decrease in body temperature over time. The control and ice-pack interventions (Fig. 3.4) are shown as an example of typical experimental procedure times and of typical body temperature responses obtained from the experimental procedures. In both panels it can be

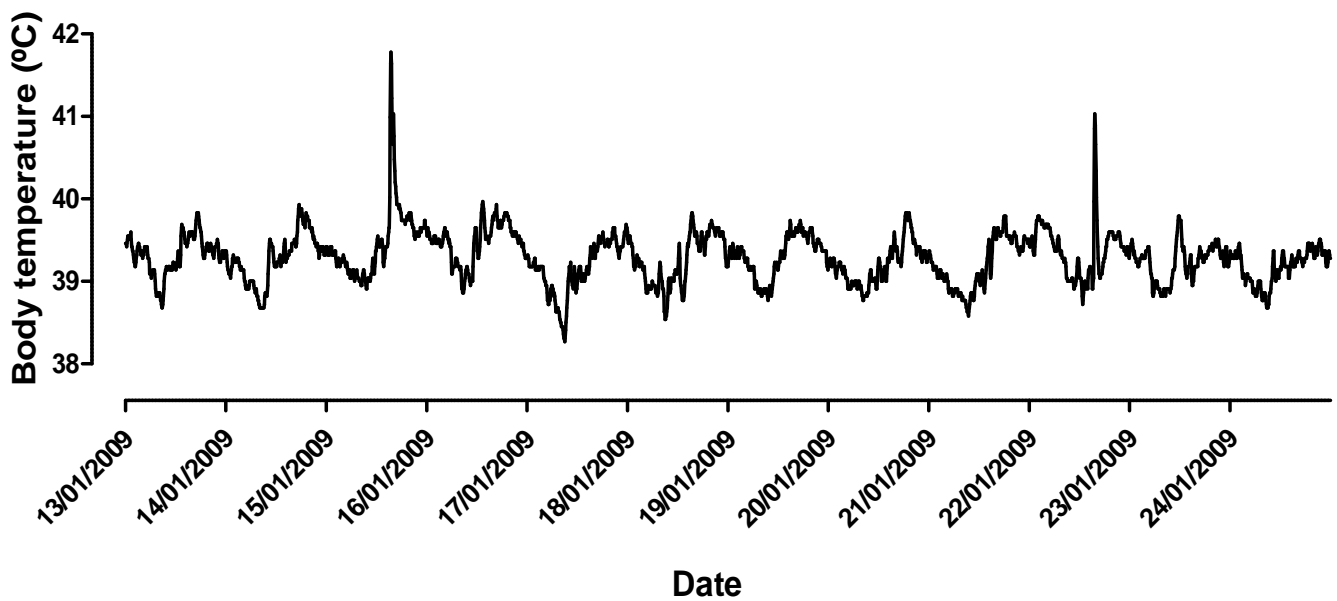


Figure 3.3 The body temperature of one blesbok over eleven days that include two capture days. The 15/01/2009 and 22/01/2009 were trial days where a capture-induced hyperthermia was elicited. The sudden increase in body temperature, of about 3°C, is clearly visible as two spikes in the temperature curve.

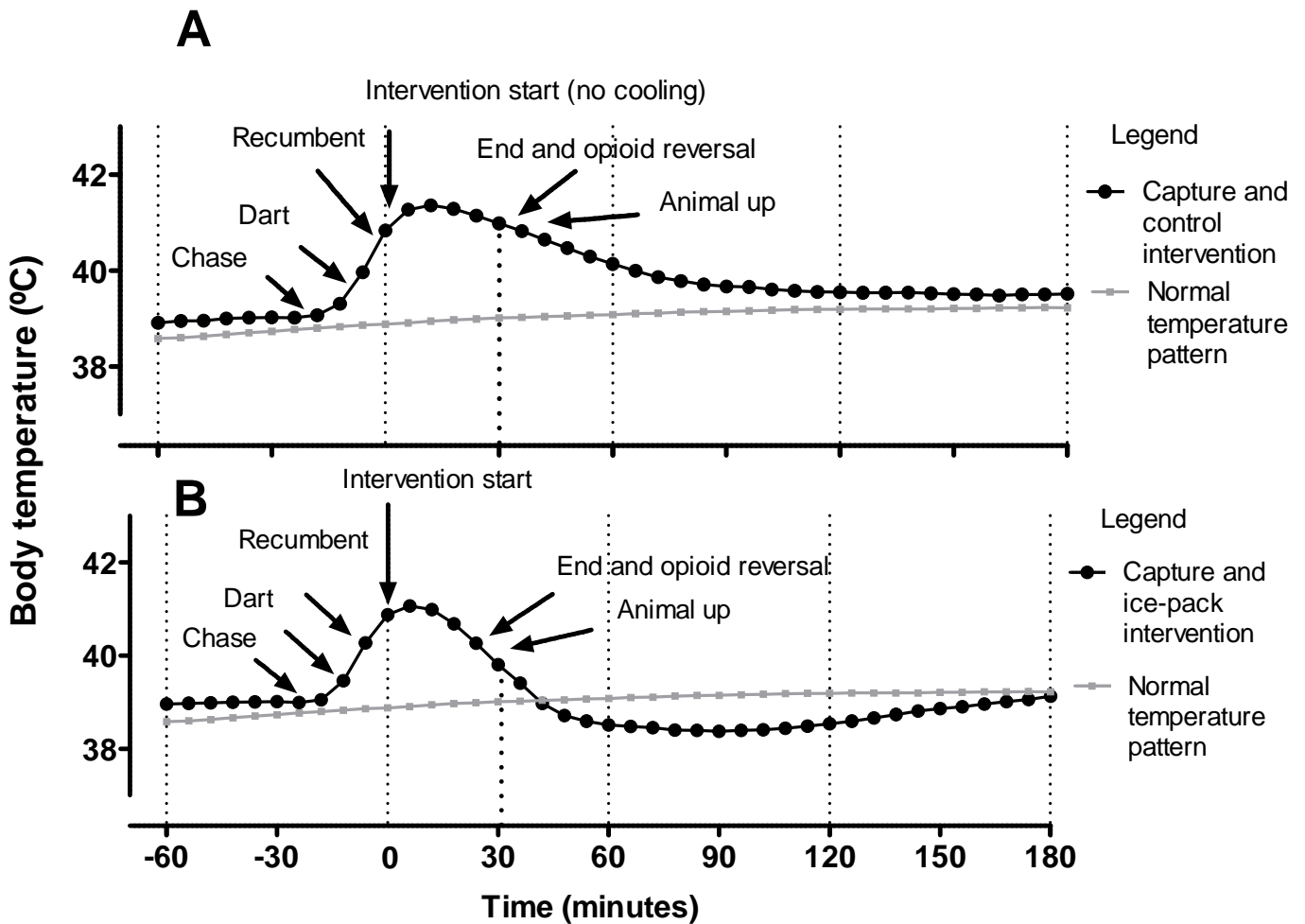


Figure 3.4 The body temperature of one blesbok before capture, during capture, during experimental interventions and after experimental interventions. Panel A depicts a temperature curve (bold black curve) when no intervention occurred (control) after capture and also depicts the normal body temperature pattern (grey curve) for that blesbok at the same time of day on days when no capture occurred (23 days). Panel B shows the increase in body temperature during the capture procedure and how the ice-pack interventions decreased the body temperature (bold black curve) and how the body temperature during capture differed from the normal body temperature pattern (grey curve) for that blesbok at the same time of day on days when no capture occurred.

seen that capture increased the body temperature above what the normal temperature would have been at that time without capture.

3.3 Mean body temperatures

The mean and SD of body temperature during each intervention are shown in Figure 3.5 for both the water-dousing group and the alternative methods group. The rise in temperature from the capture procedure is apparent before time zero. At time zero the start of cooling began. At 30 minutes the cooling intervention ended. Thus, a decrease in body temperature is shown between time zero and time 30 minutes in the cooling interventions (Fig. 3.5). The body temperature would typically continue to decrease until 60 minutes had elapsed, then the body temperature would plateau, and finally the body temperature would return to normal after time 60 minutes. The return to normal body temperature was considered a return to $38.78 \pm 0.37^{\circ}\text{C}$, which was the mean body temperature of all the blesbok over 24 hours over 23 days (Section 3.1). In the water-dousing group, for all the cooling interventions (4°C water-dousing, 17°C water-dousing, 28°C water-dousing and 28°C water-dousing with fanning), the mean body temperature within one hour is significantly more decreased than the mean body temperature of the control (RM-ANOVA, $P < 0.0001$, $F = 16.33$). In the alternative methods group, for all the cooling interventions (ice-packs, IV infusion, mist spray and the 28°C dousing), this decrease in mean body temperature within one hour is significantly decreased more than the mean body temperature of the control (RM-ANOVA, $P < 0.0001$, $F = 21.63$). Also, the ice-packs and the 28°C water-dousing had a greater decrease in the mean body temperature in one hour than the IV infusion and mist spray cooling interventions (RM-ANOVA, $P < 0.05$).

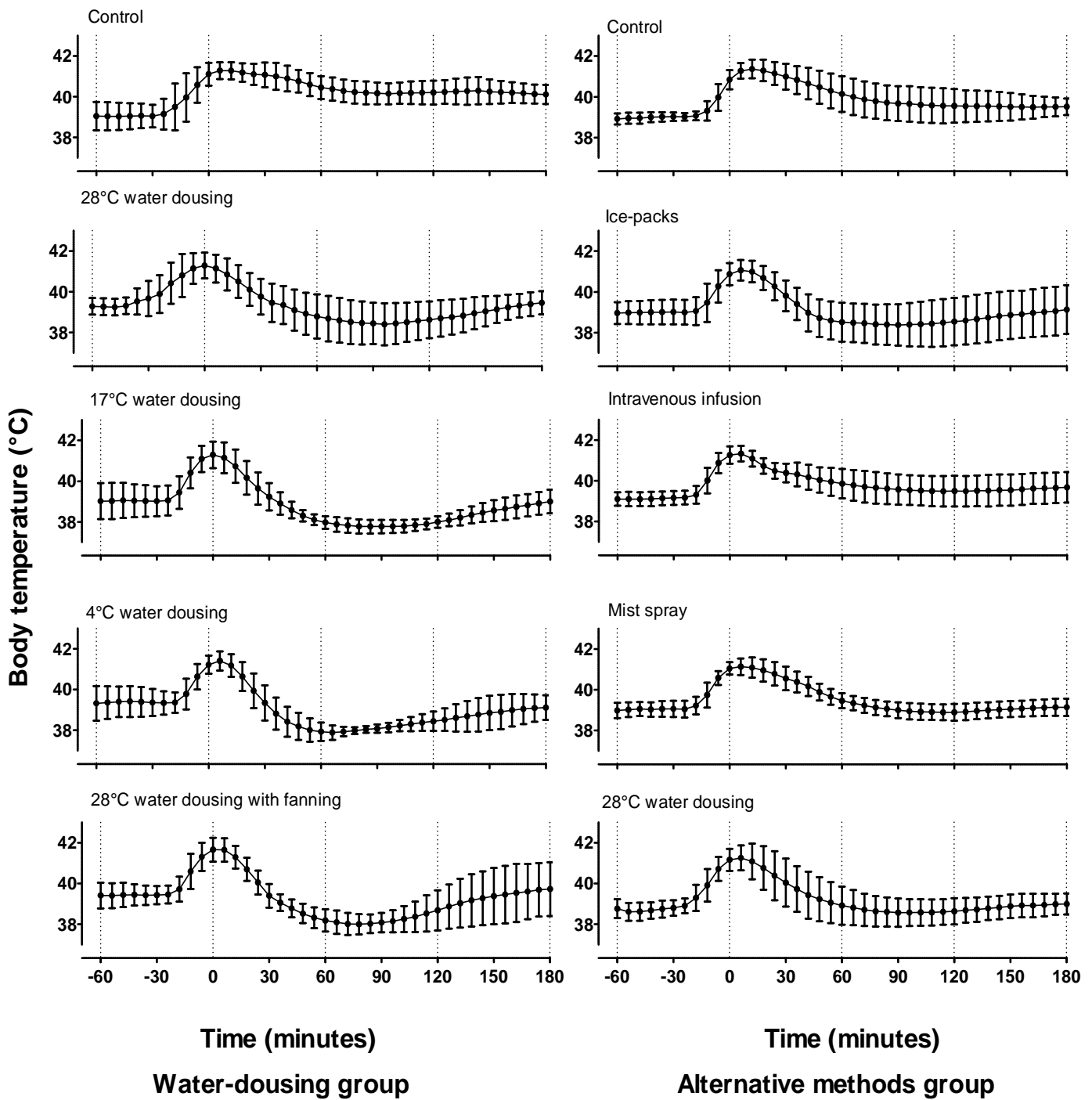


Figure 3.5 The mean body temperature of the blesbok for each cooling intervention over time. The panels on the left show the mean body temperature and standard deviation of blesbok in the water-dousing group. The panels on the right depict the mean and standard deviation of the body temperature for blesbok in the alternative methods group. Vertical bars (I) indicate the standard deviation from mean body temperature at that hour.

3.4 Peak body temperatures

The peak body temperatures were compared to confirm that blesbok in each group had a similar body temperature before cooling started. Peak body temperatures were not different for both the water-dousing group and alternative methods group and were in the range of 41 -42°C (RM-ANOVA, $P = 0.106$, $F = 2.280$; $P = 0.831$, $F = 0.365$; Fig. 3.6).

Peak body temperature occurred close to the start of the cooling intervention (denoted as time zero) in most animals but the peak temperature could occur up to and including 12 minutes after the start of the cooling intervention. The mean time for the occurrence of the peak in body temperature in the water-dousing group was 4.3 ± 3.9 minutes after the start of cooling and in the alternative methods group the mean time that the peak body temperature occurred was 6.1 ± 0.9 minutes after the start of cooling.

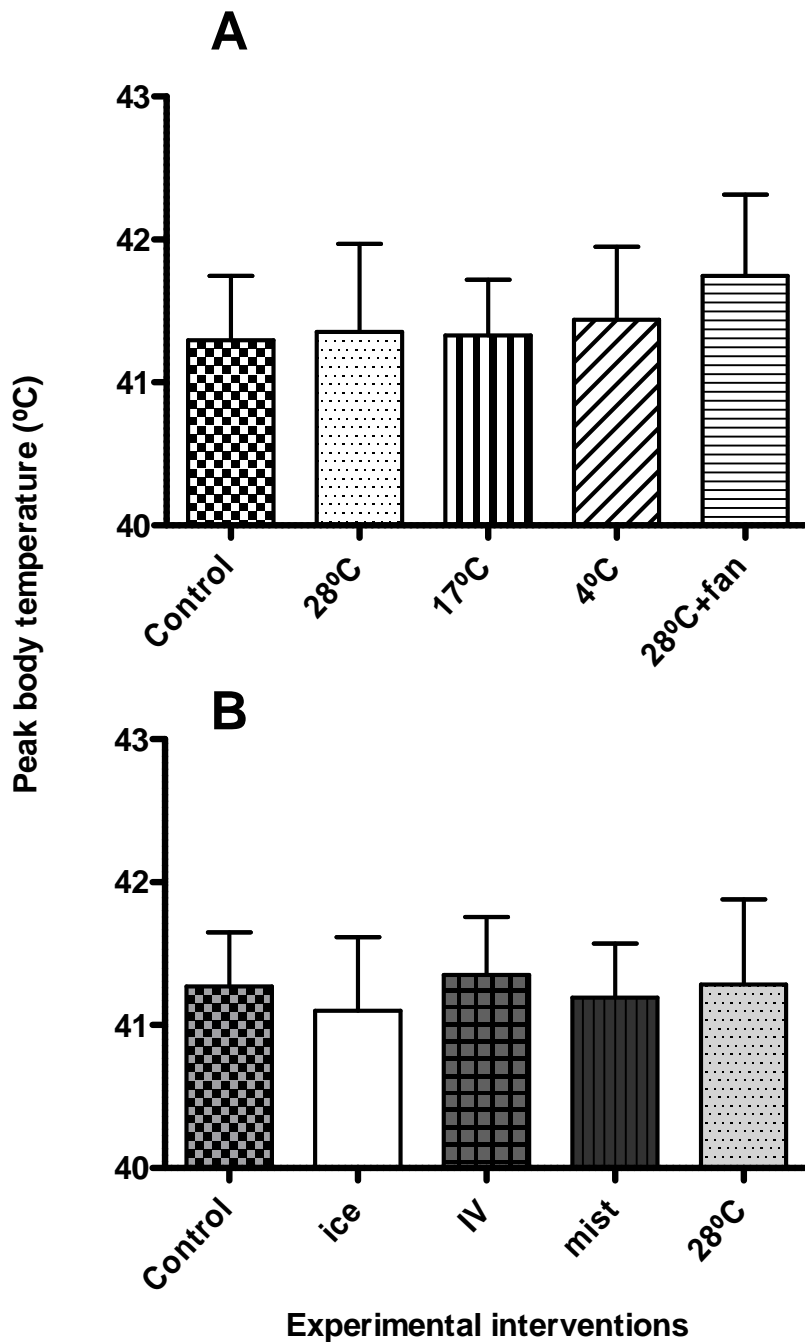


Figure 3.6 The peak body temperatures for each experimental intervention. The top panel (A) illustrates no significant difference in peak temperatures in the water-dousing group whereas the bottom panel (B) illustrates no significant difference in peak temperatures in the alternative group. The bars show the mean and SD of the peak body temperature of the blesbok and the term IV refers to intravenous infusion.

3.5 Minimum body temperatures

The absolute minimum body temperatures of the blesbok after cooling in each intervention were compared to those in the other interventions, and to the control within the groups. The water-dousing interventions significantly lowered body temperature to about 38°C in the blesbok compared to the temperature reached of about 40.5°C in the blesbok of the control intervention (RM-ANOVA, $P < 0.0001$, $F = 14.87$) as seen in Figure 3.7. The minimum body temperature of the blesbok between each cooling intervention (4°C water-dousing, 17°C water-dousing, 28°C water-dousing and 28°C water-dousing with fanning) did not differ (RM-ANOVA, $P > 0.05$, $F = 14.87$). There was also no significant difference in the minimum body temperature of the blesbok between the 28°C water-dousing and the 28°C water-dousing with fanning intervention (RM-ANOVA, $P > 0.05$).

In the alternative methods group, only the ice-packs and the 28°C water-dousing significantly lowered body temperature in the blesbok comparison to the control (RM-ANOVA, $P = 0.0005$, $F = 7.431$; Fig. 3.7). The minimum body temperature in the blesbok after ice-packs placement and 28°C water-dousing did not significantly differ (RM-ANOVA, $P > 0.05$). The IV infusion and mist spray treatments did not significantly lower body temperature in the blesbok in comparison to the control (RM-ANOVA $P > 0.05$). Thus, ice-packs and the 28°C water-dousing, which led to a significantly lower minimum body temperature than the control, also had a significantly lower minimum body temperature than did the IV infusion and mist spray (RM-ANOVA, $P < 0.05$).

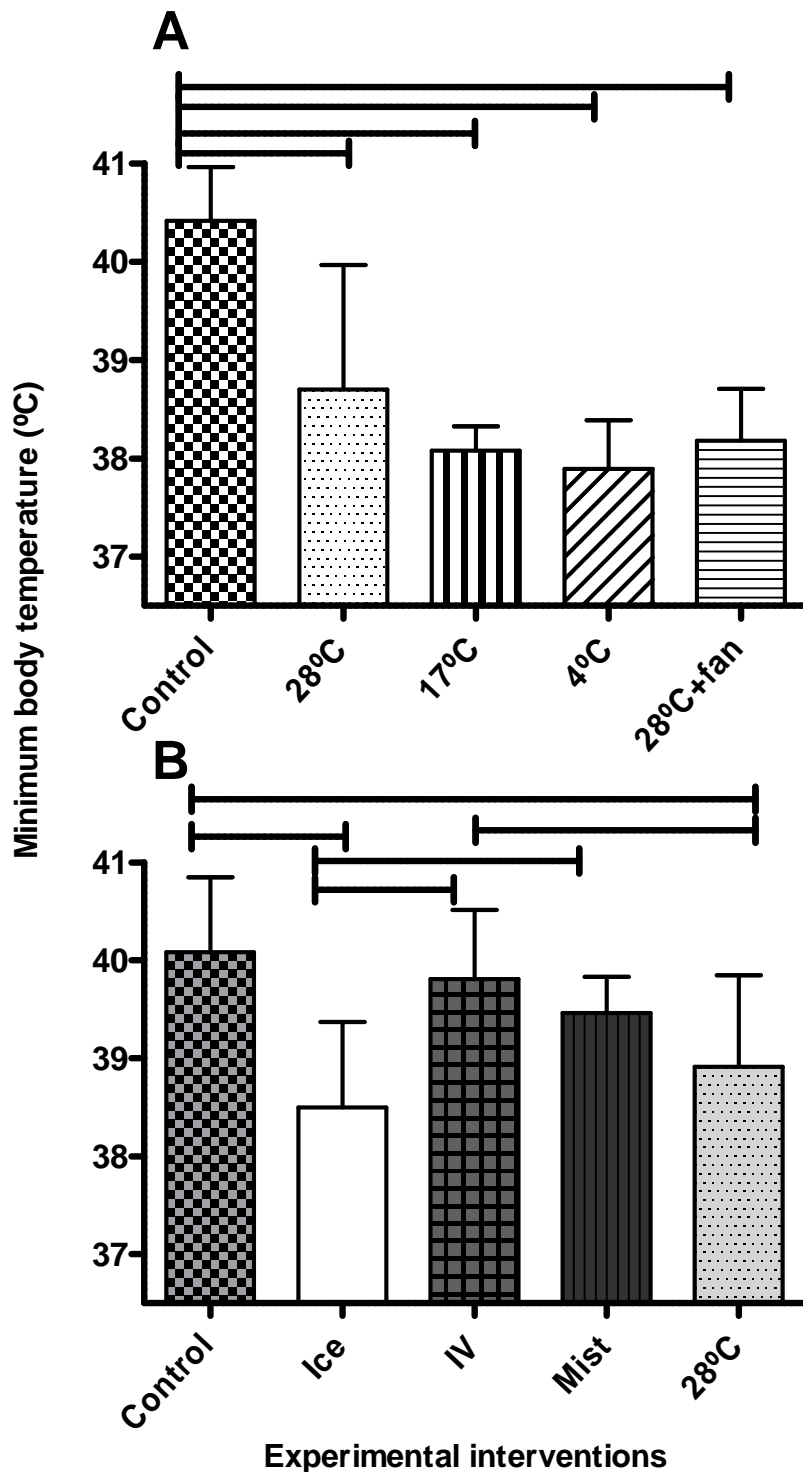


Figure 3.7 The minimum body temperatures reached within an hour from the start of cooling. The top panel (A) illustrates the significance of the water-dousing interventions in comparison to the control in lowering body temperature in one hour. The bottom panel (B) illustrates the differences in minimum body temperature in the alternative methods group. The solid bar above the columns (—) indicates a significant difference ($P < 0.05$). The bars show the mean and SD of the minimum body temperature reached during the experimental interventions and the term IV refers to intravenous infusion.

3.6 Amplitude of cooling

The amplitude of cooling, which was the absolute change in body temperature from time zero, was calculated after 30 minutes and one hour. Figure 3.8 illustrates this change in body temperature of the blesbok in the water-dousing and alternative methods groups. The negative values in Figure 3.8 indicate a fall in body temperature of the blesbok. After 30 minutes, all the water-dousing interventions had led to a significant decrease (RM-ANOVA, $P = 0.0002$, $F = 10.45$) in body temperature, of about 2°C, in comparison to the control. There was no significant decrease (RM-ANOVA, $P > 0.05$) in fall of body temperature between the water-dousing interventions. After an hour, all the interventions were significantly more effective (RM-ANOVA $P = 0.0001$, $F = 12.12$) than the control in decreasing temperature in the blesbok, but no intervention was more effective (RM-ANOVA $P > 0.05$) than another in decreasing the body temperature; the decrease in temperature was about 3°C for all the water-dousing interventions (Fig. 3.8). A Student's t-test confirmed that there was no significant difference in the decrease in the body temperature of the blesbok between 28°C water-dousing and 28°C water-dousing with the addition of fanning, at either 30 minutes or one hour ($P = 0.176$ and 0.062 respectively).

In the alternative methods group after 30 minutes, ice-packs, IV infusion and the 28°C water-dousing significantly decreased the body temperature in comparison to the control (RM-ANOVA, $P < 0.05$, $F = 2.906$), by about 1°C (Fig. 3.8). The fall in the body temperature after the mist spray did not differ from that of the control (RM-ANOVA, $P > 0.05$), with the mean decrease in body temperature of the blesbok being less than 0.5°C. The mist spray also did not differ significantly (RM-ANOVA, $P > 0.05$) from the IV infusion in decreasing the

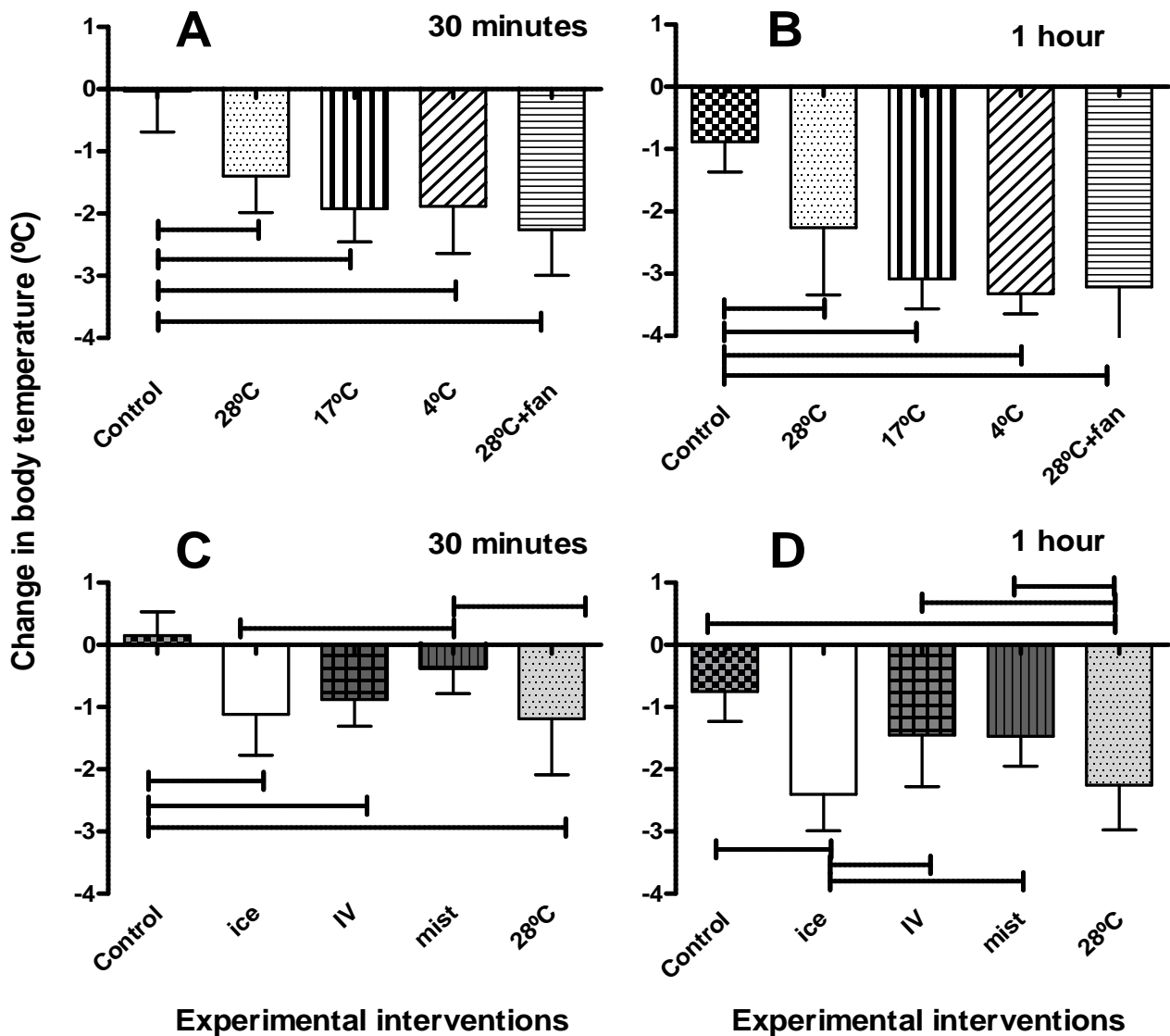


Figure 3.8 The magnitude of the decrease in temperature within 30 minutes (panels A and C) and 1 hour (panels B and D). The graphs on the left (panels A and C) illustrate the magnitude of the decrease in 30 minutes and the graphs on the right (panel B and D) illustrate the magnitude of the decrease in one hour. In the water-dousing group in both 30 minutes and one hour (Panel A and B respectively) all the water-dousing interventions significantly decreased temperature in comparison to the control. In the alternative methods group, ice-packs, IV infusion and 28°C water-dousing decreased the temperature significantly in 30 minutes (panel C) but after an hour (panel D) only the ice-packs and 28°C water dousing lowered the temperature significantly. The solid bar above the columns (—) indicates a significant difference ($P < 0.05$). The bars show the mean and SD of the magnitude of the body temperature decrease for each intervention and the term IV refers to intravenous infusion.

temperature, and ice-packs and 28°C water-dousing were significantly (RM-ANOVA, $P < 0.05$) more effective than mist spray in decreasing the temperature. After an hour, only the ice-packs and the 28°C water-dousing had significantly (RM-ANOVA, $P < 0.05$, $F = 10.36$) decreased the temperature in comparison to the control. These interventions also significantly (RM-ANOVA, $P < 0.05$) decreased body temperature of the blesbok in comparison to the IV infusion and mist spray. After an hour, the change in body temperature of the hyperthermic blesbok (without any cooling method applied to the animals) in the control intervention was by about 1°C.

3.7 Thermal response indices

The thermal response index (TRI, °C.min) was calculated for each intervention over two time periods, 30 minutes and one hour. In the water-dousing group, within 30 minutes (RM-ANOVA, $P = 0.0052$, $F = 5.593$) and one hour (RM-ANOVA, $P < 0.0001$, $F = 18.80$), all the water-dousing interventions (4°C water-dousing, 17°C water-dousing, 28°C water-dousing and 28°C water-dousing with fanning) led to a significantly larger TRI value (which is a negative value) than the control in the blesbok (Fig. 3.9). The water-dousing interventions did not differ (RM-ANOVA, $P > 0.05$) in their TRI values. A Student's t-test confirmed that there was no difference in the TRI

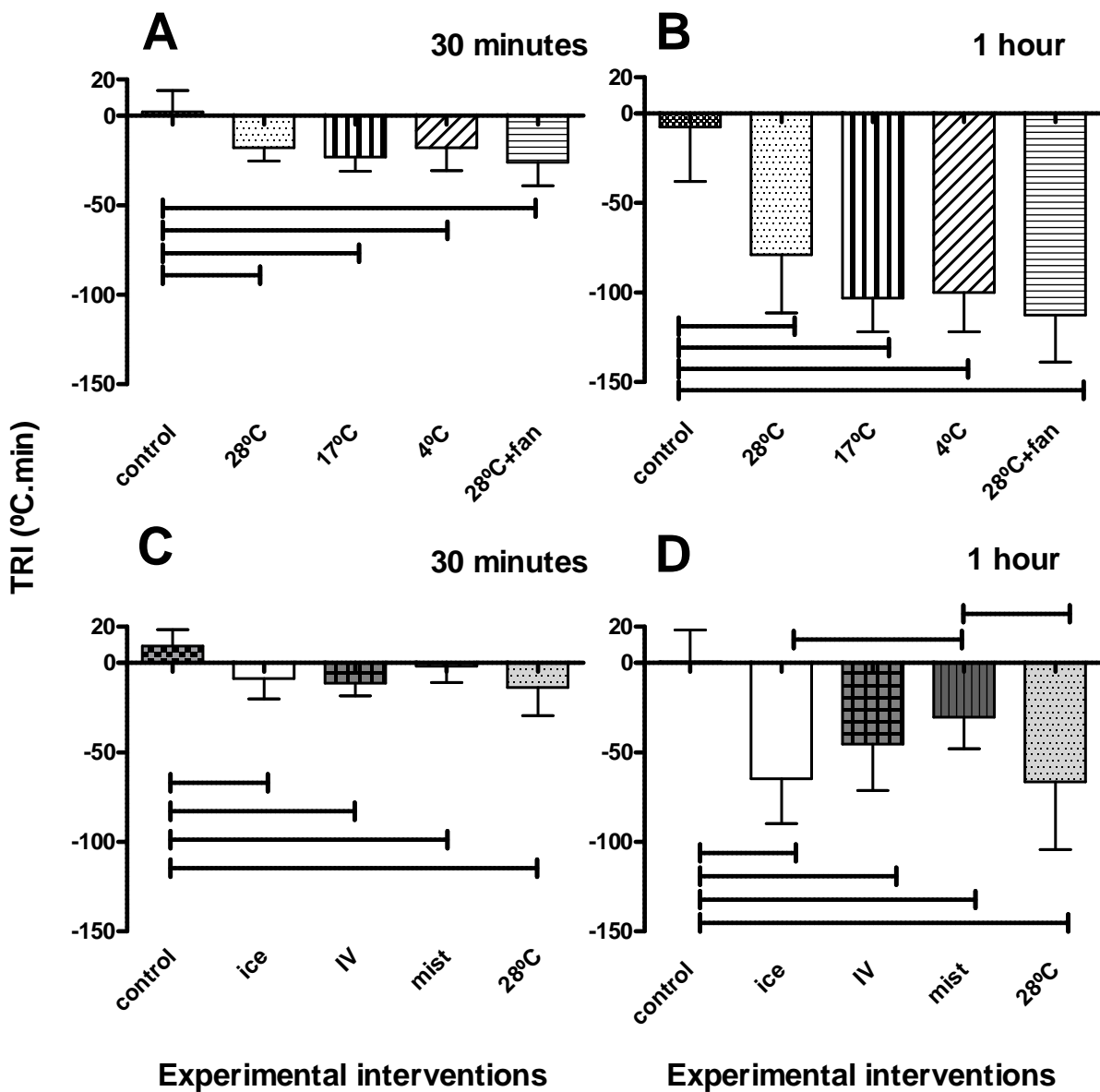


Figure 3.9. The TRI values within 30 minutes (panels A and C) and within one hour (panels B and D) from the start of cooling for the water-dousing and alternative methods groups. The top graphs (A and B) are the water dousing group. All the water-dousing interventions had significantly larger TRI values than the control. The bottom graphs (C and D) are the alternative methods group. Within one hour the ice-packs and 28°C water dousing have significantly larger TRI value than the mist spray. The solid bar above the columns (—) indicates a significant difference ($P < 0.05$). The bars show the mean and SD of the TRIs for each experimental intervention and the term IV refers to intravenous infusion.

values between the 28°C water-dousing and the 28°C water-dousing with fanning over 30 minutes and one hour ($P = 0.4036$ and 0.1819 respectively). In the alternative methods group all the body temperature curves (28°C water dousing, ice-packs, IV infusion and mist spray) of the blesbok had a significantly larger TRI value than did the control (Fig. 3.9). This was true for both the 30 minute (RM-ANOVA, $P = 0.0015$, $F = 6.158$) and one hour interval (RM-ANOVA, $P < 0.0001$, $F = 11.92$). In the one hour interval the ice-packs and the 28°C water-dousing were more effective in negatively increasing the TRI value than were the mist spray, as these two interventions also had a significantly (RM-ANOVA, $P < 0.05$) larger TRI value than the mist spray. However, the control had a TRI value with a positive value which means that even though there was some decrease in body temperature, overall, the blesbok body temperature increased over one hour. Similarly, the mist spray had a positive TRI even though it caused slight cooling and this TRI value was significantly smaller than the control.

3.8 Rate of cooling

The rate of cooling ($^{\circ}\text{C}\cdot\text{min}^{-1}$) is indicative of how quickly an animal was cooled. In the water-dousing group the 4°C, 17°C and 28°C with fanning cooled the blesbok significantly faster than did the control (RM-ANOVA, $P = 0.014$, $F = 7.477$; Fig. 3.10). Dousing the animals with the cold temperature water of 4°C cooled the animals at a rate of about $0.10^{\circ}\text{C}\cdot\text{min}^{-1}$, significantly (Student's t-test, $P < 0.0428$) faster than the 28°C water dousing, which cooled the blesbok at a rate of $0.05^{\circ}\text{C}\cdot\text{min}^{-1}$.

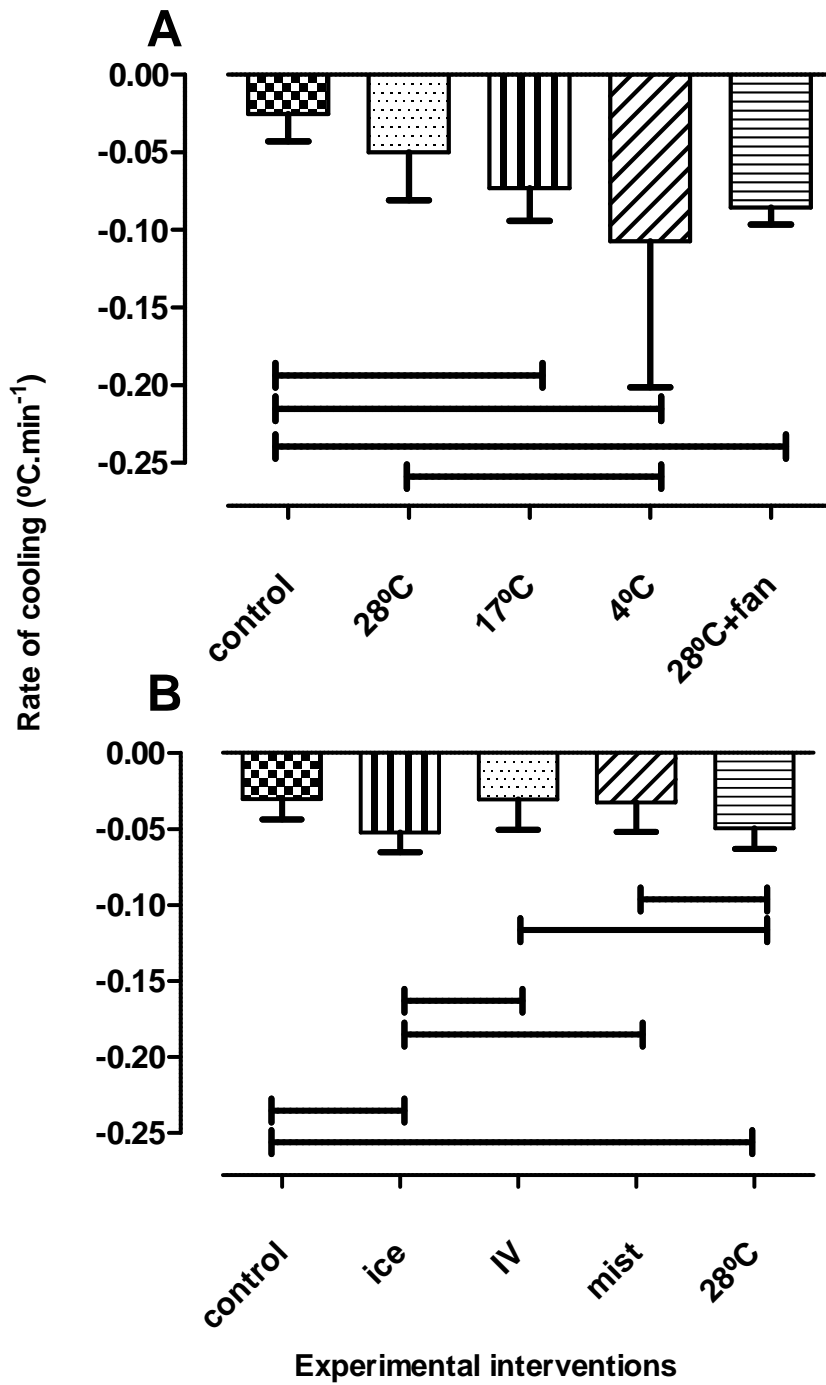


Figure 3.10 The rate of cooling over 30 minutes by each experimental intervention. The top panel (A) illustrates the water-dousing group and the bottom panel (B) illustrates the alternative methods group. The solid bar above the columns (—) indicates a significant difference ($P < 0.05$). The bars show the mean and SD of cooling rate and the term IV refers to intravenous infusion.

In the alternative methods group the ice-packs and the 28°C water-dousing cooled the animals quicker than did the control (RM-ANOVA, $P < 0.05$, $F = 8.386$; Fig. 3.10). These two interventions also cooled the animals significantly faster than did the mist spray and the IV infusion (RM-ANOVA, $P < 0.05$), while the cooling induced by the mist spray and IV infusion did not differ from that of the control (RM-ANOVA, $P > 0.05$), at less than $0.05^{\circ}\text{C}\cdot\text{min}^{-1}$. A Student's t-test confirmed that the 28°C water-dousing cooled the animals faster than did the mist spray ($P = 0.0268$).

3.9 The temperature difference between core and subcutaneous temperature

In Figure 3.11 the temperature difference between the abdominal body temperature (core temperature) and subcutaneous temperature is shown. The temperature difference between the core and the periphery reflect conductance to the core and the periphery and if the conductances are similar then there is no temperature difference. In the control curves, for both groups, the temperature difference between the core and the subcutaneous layers did not change over time.

In the water-dousing group a distinct rise, with a maximum amplitude of about 2°C , was seen in all the water-dousing interventions (Fig. 3.11). The peak difference between the core and subcutaneous temperature for 28°C, 17°C, 4°C and 28°C with fanning was significantly larger than for the control (RM-ANOVA, $P < 0.0001$, $F = 22.72$). The largest amplitude of about 3.5°C is seen in the curve of the 4°C water-dousing intervention; this amplitude is significantly larger than those of the other water-dousing interventions (28°C water-dousing, 17°C water-dousing and 28°C water dousing with fanning) and the control

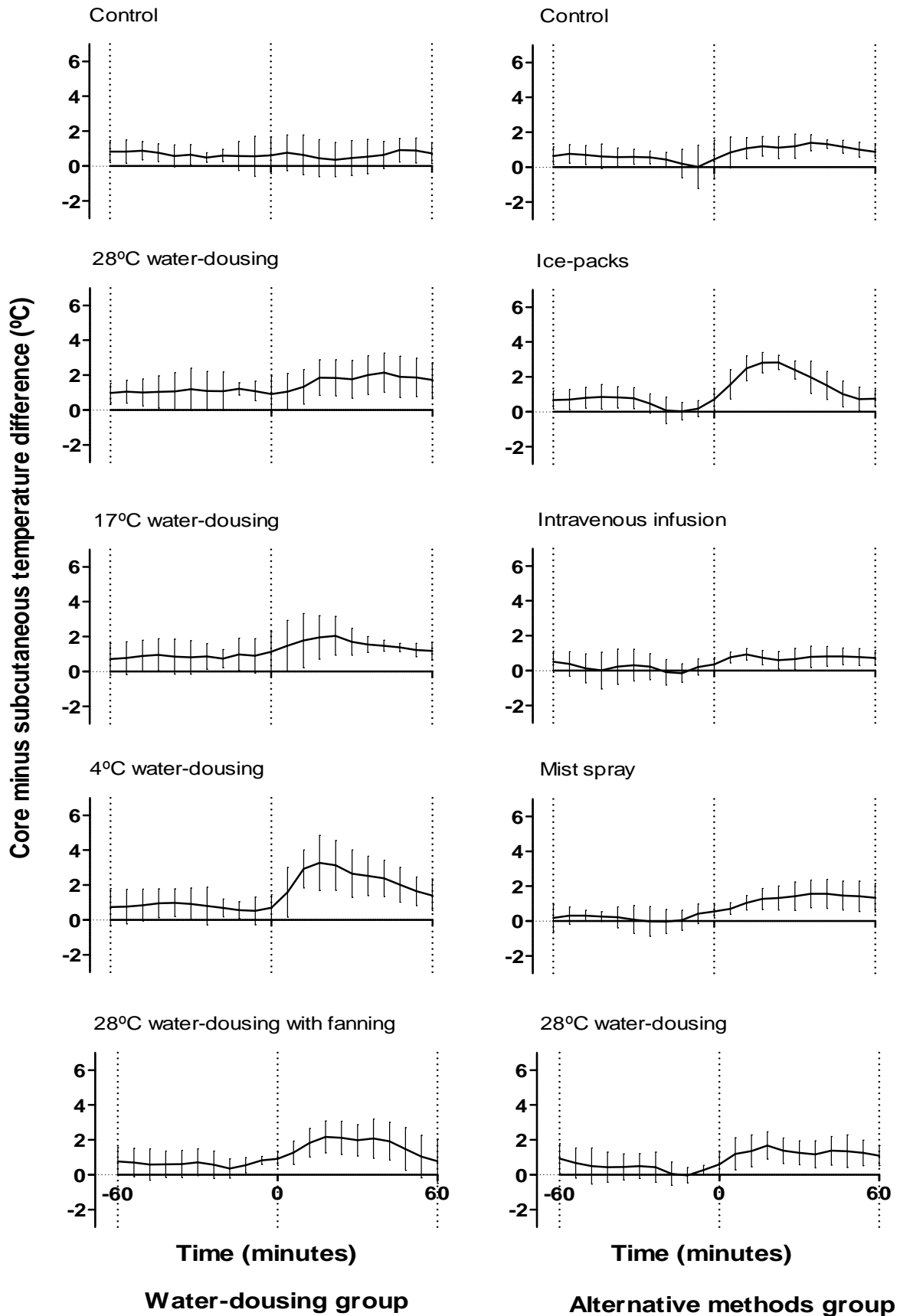


Figure 3.11 The temperature difference between the core and the periphery over time. The graphs on the left illustrate the water-dousing group and the graphs on the right illustrate the alternative methods group. The highest amplitude curve, in the water dousing group, was in the 4°C intervention. In the alternative group, the ice-packs had the highest amplitude in the temperature difference curve. Vertical bars (I) indicate the standard deviation from temperature difference at that hour.

(RM-ANOVA, $P < 0.05$, $F = 22.72$). For the area under the curve values calculated from the curves from time zero to 30 minutes, the 4°C water-dousing was significantly greater than the control, 28°C water-dousing, 17°C water-dousing and the 28°C water-dousing with fanning (RM-ANOVA, $P = 0.014$, $F = 7.468$). Over a one-hour period the 4°C water dousing had a significantly larger area under the curve value than only the control and the 17°C water-dousing (RM-ANOVA, $P = 0.032$, $F = 3.467$).

In the alternative methods group the ice-packs, the IV infusion and the 28°C water-dousing led to a significantly larger peak temperature difference between the core and the periphery than did the control (RM-ANOVA, $P < 0.0001$, $F = 10.10$). The mist spray elicited a similar peak core minus subcutaneous temperature difference compared to the control in the blesbok (RM-ANOVA, $P > 0.05$). The highest amplitude, of about 3°C, was seen in blesbok receiving the ice-pack intervention (Fig. 3.11), which led to a significantly larger peak difference between the core and subcutaneous temperature than the control, IV infusion, mist spray and 28°C water-dousing (RM-ANOVA, $P > 0.05$). The 28°C water-dousing had a higher peak core minus subcutaneous temperature difference than did the IV infusion. For the area under the curve over 30 minutes, only the ice-packs had a significantly larger area under the curve than did the other alternative methods interventions and the control (RM-ANOVA, $P = 0.0072$, $F = 4.538$). However, over one hour there were no differences in the area under the curve (RM-ANOVA, $P = 0.474$, $F = 0.910$).

3.10 The temperature difference between core and ear skin temperature

Figure 3.12 depicts the temperature difference between the core and the ear skin. The temperature difference at 24 minutes (when cooling had just been recently stopped) was compared to time zero (when cooling was started). There was no significant temperature difference (Student's t-test, $P > 0.05$) between these two times for any of the interventions in either group.

The area under the curve for the temperature difference between the core and the ear skin was also calculated for the interval of time zero to 24 minutes to determine if there had been any vasoconstriction during the 24 minute interval of cooling. In the water-dousing group there was no difference in any of the water-dousing interventions (4°C water-dousing, 17°C water-dousing, 28°C water-dousing and 28°C water-dousing with fanning) in the temperature difference with time in comparison to the control (one-way ANOVA, $P = 0.09609$, $F = 0.149$).

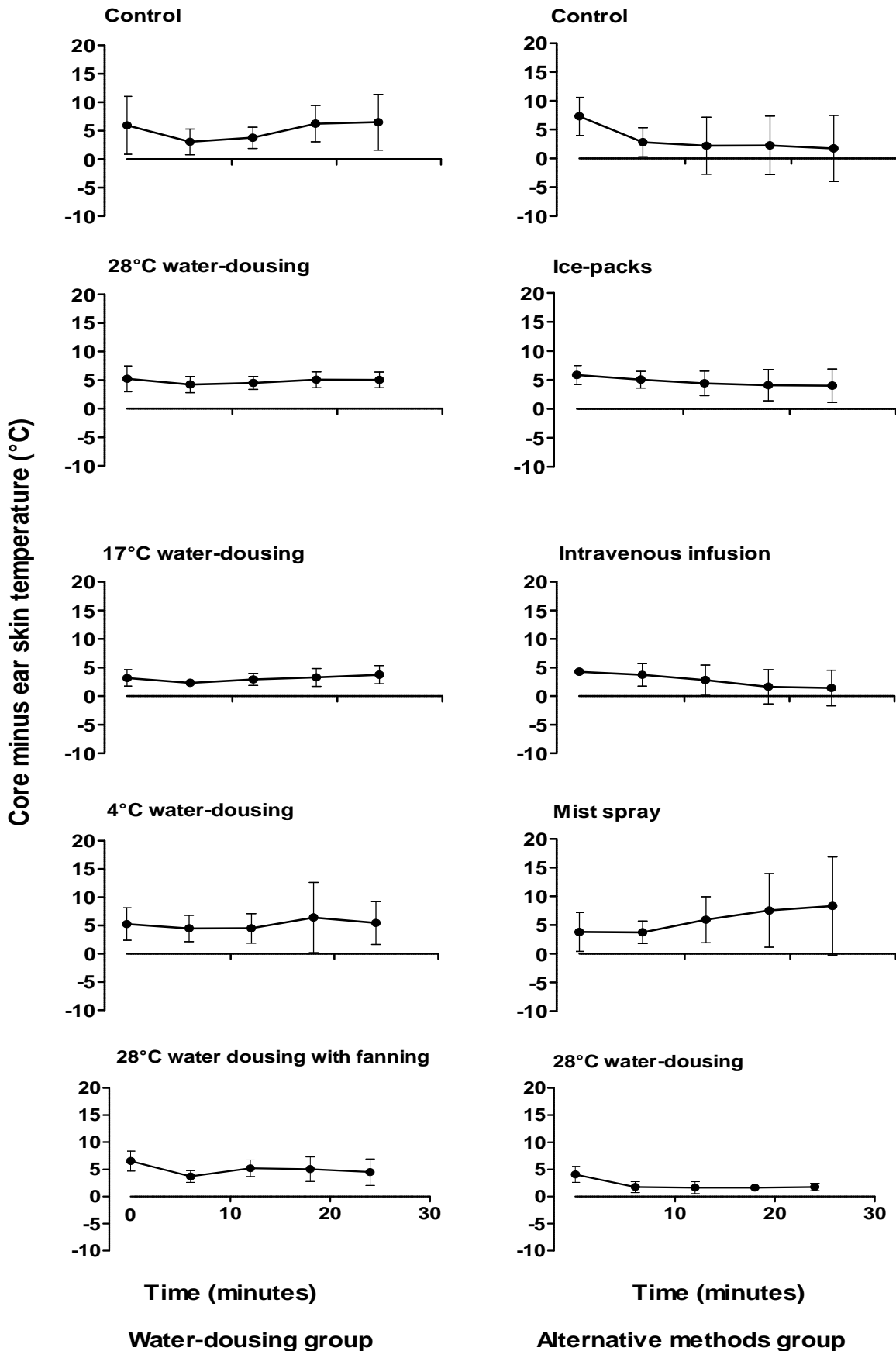


Figure 3.12 The temperature difference between the core and ear skin over time. The graphs on the left illustrate the water-dousing group and the graphs on the right illustrate the alternative methods group. Vertical bars (I) indicate the standard deviation from temperature difference at that hour.

In the alternative methods group there was also no significant temperature difference with time in either the ice-packs, IV infusion, mist spray or 28°C water-dousing interventions in comparison to the control intervention (one-way ANOVA, $P = 0.2495$, $F = 1.446$). Therefore, considering the difference between time zero and 24 minutes and the area under the curve values for the 24 minute interval, it did not appear that any generalized vasoconstriction was present in the blesbok during the cooling interventions.

3.11 Core abdominal temperature versus rectal temperature

Real-time rectal temperatures were monitored using a combination of digital readers and rectal thermometers. In some instances rectal temperatures fell faster than did the core temperature (reported by the abdominal loggers) for the same recording period time. This anomaly was seen only in the cooling interventions which significantly lowered the body temperature of blesbok (the water-dousing and ice-packs), but when no significant cooling (the IV infusion, mist spray and control trials) had occurred then rectal and abdominal loggers' temperatures were similar (Fig. 3.13) The discrepancy between the abdominal and rectal temperature is depicted in Figure 3.13, with examples typical for all the blesbok.

3.12 Environmental conditions

Overall there was no significant difference (ANOVA, $P > 0.05$) in any environmental conditions between interventions in either group. The summary of environmental conditions can be seen in Table 3.1. Dry-bulb temperature, wet-bulb temperature,

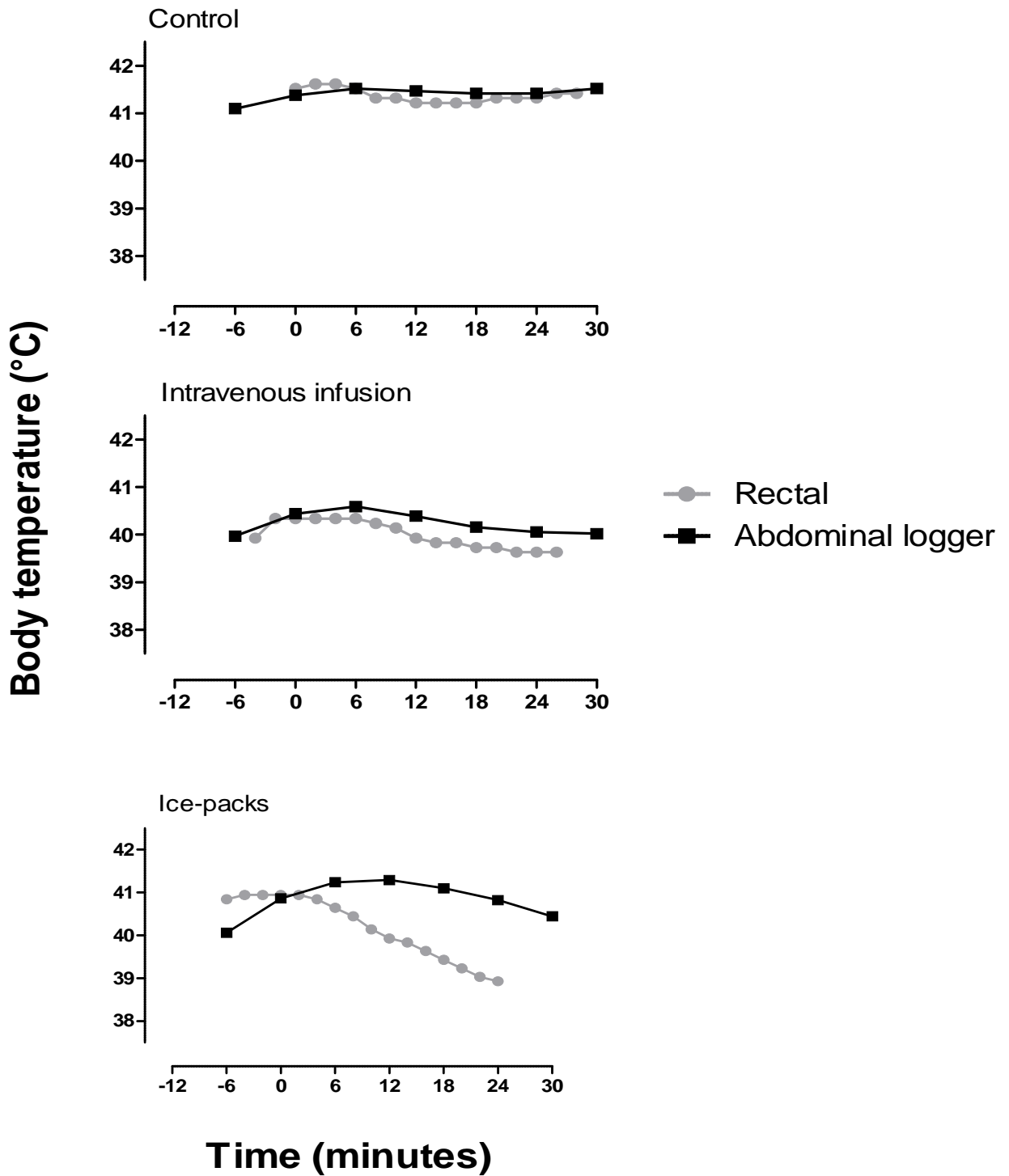


Figure 3.13 The body temperature discrepancy between abdominal temperature and rectal temperature. When no cooling interventions (top panel) and when IV fluids (middle panel) were administered then the rectal and abdominal core temperatures were similar. However, when ice-packs (bottom panel) were placed the rectal temperatures decreased faster than the abdominal core temperatures.

Table 3.1 The environmental conditions for both groups.

Group	Interventions	Environmental condition				
		Dry-bulb temperature (°C)	Wet-bulb temperature (°C)	Wind speed (m.s ⁻¹)		Vapour Pressure (kPa)
				Boma	Animal	
Water-dousing group	Control	27.7 ± 2.3	22.7 ± 2.3	0.3 ± 0.1	0.4 ± 0.1	2.5 ± 0.4
	28°C water-dousing	27.6 ± 2.5	22.5 ± 2.1	0.3 ± 0.1	0.3 ± 0.2	2.4 ± 0.3
	17°C water-dousing	27.2 ± 1.4	22.1 ± 0.7	0.3 ± 0.1	0.9 ± 1.2	2.4 ± 0.1
	4°C water-dousing	26.4 ± 3.5	22.6 ± 1.3	0.4 ± 0.3	0.3 ± 0.2	2.5 ± 0.1
	28°C water-dousing with fanning	28.3 ± 1.6	24.1 ± 2.1	0.3 ± 0.1	1.2 ± 0.6 ^a	2.8 ± 0.4
Alternative methods group	Control	28.5 ± 3.8	22.9 ± 2.6	0.3 ± 0.1	0.8 ± 1.0	2.5 ± 0.4
	Ice-packs	27.4 ± 1.9	22.7 ± 1.7	0.3 ± 0.1	0.3 ± 0.2	2.5 ± 0.3
	IV infusion	29.3 ± 3.0	23.8 ± 1.4	0.4 ± 0.4	0.3 ± 0.1	2.7 ± 0.2
	Mist spray	28.4 ± 2.5	22.9 ± 2.0	0.3 ± 0.2	0.3 ± 0.2	2.5 ± 0.3
	28°C water-dousing	28.7 ± 2.4	23.2 ± 1.9	0.3 ± 0.2	0.3 ± 0.1	2.6 ± 0.4

^a: This is our fanning speed that we generated over the animal

wind speed, black globe temperature and vapour pressure were similar for all interventions within a group.

CHAPTER 4:

DISCUSSION

4. DISCUSSION

To my knowledge, this is the first study comparing different cooling techniques for captured wild antelope. All the different temperatures (4°C, 17°C and 28°C) of the water-dousing interventions and ice-packs decreased the body temperature successfully over an hour by about 2.5-3°C more than did the control (in which no cooling intervention occurred). The cooling was sufficient to return the body temperature of the blesbok close to their normal body temperature over 24 hours during the day ($38.78^{\circ}\text{C} \pm 0.37^{\circ}\text{C}$) after an hour period. Cooling with the IV fluids and mist spray did not return the blesboks' body temperature back to normal within an hour of cooling.

By chasing the blesbok, we elicited an increased body temperature of 41-42°C, which was about 3°C more than the mean daily average body temperature of the animals ($38.78^{\circ}\text{C} \pm 0.37^{\circ}\text{C}$). In one group of blesbok we tested the effect of pouring water of different temperatures (4°C, 17°C and 28°C) over the animals, and the effect of including fanning when 28°C water was doused over the blesbok, on body temperature. In this group the decrease in body temperature after the water-dousing interventions was about 3°C (Figure 3.8), which returned body temperature to about 38°C (Figure 3.7). Therefore, all the water-dousing temperatures in this group can be considered effective in significantly lowering body temperature. In the other group we tested alternative methods (ice-packs, IV infusion and a mist spray) to water-dousing for cooling hyperthermic animals and compared these alternatives to a control (of no external cooling applied) and a water-dousing method (dousing with 28°C water). The ice-packs were the only alternative method to significantly lower body temperature to a minimum of about 38.5°C; the control (Figure 3.7) caused only

a lowered body temperature of about 40°C. However, the ice-packs when compared to the water-dousing method (28°C water-dousing) both had a similar effect on lowering body temperature and thus can be considered equally effective (Figure 3.8). The ice-packs and 28°C water-dousing were more effective than the IV infusion and the mist spray in lowering body temperature in the blesboks; the IV infusion and mist spray caused similar changes in body temperatures (Figure 3.7) as the control after an hour.

Initially, in the first 30 minutes after the start of cooling, there was a greater change in the blesboks' body temperature with the IV infusion than in the control (Fig 3.8) but this was only about a 1°C change in body temperature. After an hour the change in body temperature from the IV infusion in the animals was similar to the control and this is why the IV infusion (and mist spray) resulted in a similar minimum body temperature (within an hour) to the control, which was about 40°C (Figure 3.7). The IV infusion and mist spray cooled the animals gradually, but no differently to that of the control (Figure 3.10). Instead it was the ice-packs that cooled the blesbok significantly quicker than the control over 30 minutes (Figure 3.10). However, the ice-packs cooled as quickly as using water-dousing with 28°C water. There was also a distinction in how quickly the animals were cooled using different water temperatures in the water-dousing group. The 4°C water-dousing cooled the animals quicker than did the 28°C water-dousing. Therefore, in order to cool animals more quickly a cold water temperature (4°C) is more effective than a warmer water temperature (28°C).

The only instance when the IV infusion and mist spray were significantly different to the control was in the decrease in body temperature with time over 30 minutes and one hour

(thermal response indices; Figure 3.9). Figure 3.9 shows that the control generally has an increase in body temperature over one hour (giving a positive TRI value), in the alternative methods group. Figure 3.5 shows that during that hour mean body temperature was elevated above the initial temperature for a longer period than when the temperature was lower. Therefore, an overall positive change in body temperature over time was recorded for the control group in the alternative methods group. In contrast Figure 3.10 shows a negative rate of cooling for the control because the cooling rate is calculated by determining how quickly the body temperature changed from the peak body temperature to the minimum body temperature during that hour (that is, the slope of the temperature curve was analysed when temperatures were decreasing). So the body temperature of the blesbok in the control group increased in one hour more than the body temperature decreased which resulted in a positive TRI (Figure 3.9). However, because there still was a change in the absolute values from the peak body temperature to the minimum body temperature within the first hour there was a negative rate of cooling (Figure 3.10).

Again in Figure 3.9 it can be seen that the water-dousing interventions also caused a lower body temperature to be maintained for longer (up to an hour) than did the control. There were similar TRI values (at 30 minutes and one hour) in the blesbok when either 4°C, 17°C or 28°C water-dousing (Figure 3.9) was used. Therefore, the 4°C water-dousing, 17°C water-dousing, 28°C water-dousing and the 28°C water-dousing with fanning all similarly decreased the body temperature with time (up to an hour) in the water-dousing group.

We also specifically compared the 28°C water-dousing to the 28°C water-dousing with fanning to investigate what effect the addition of fanning had on body temperature of the

blesbok. The 28°C water-dousing with fanning did not change the body temperature significantly more than did just the 28°C water-dousing. Both these interventions lowered the body temperature to a similar minimum body temperature in the blesbok (Figure 3.7) and both the 28°C water-dousing with fanning and just the 28°C water-dousing led to similar decreases in body temperature with time (thermal response indices; Figure 3.9). Lastly, the addition of fanning to the 28°C water-dousing did not cool the animals any quicker than just the 28°C water-dousing. Therefore the addition of fanning to the 28°C water-dousing did not significantly increase heat loss.

A potential problem when attempting to lower body temperature rapidly in mammals is that the cooling intervention may elicit vasoconstriction, leading to an attenuated fall, or even a rise, in body temperature. Therefore we measured ear skin temperature, as an indicator of vasomotor tone, throughout the animals' immobilisation. We subtracted this skin temperature from core abdominal temperature, to estimate if vasoconstriction occurred in response to cooling. An increase in the difference between the two temperatures, from the start of cooling to the end of cooling, would have indicated a change in the vasomotor tone and therefore would have revealed if cutaneous vasoconstriction was present. It could be argued that with a constant temperature difference of 5°C between the body temperature and ear skin temperature that the blesbok were vasoconstricted for the entire time from the start of cooling to the end of cooling. Even if vasoconstriction was present for the entire time it is important, that we found no evidence of significant changes in the temperature differences, and therefore in cutaneous vasomotor tone, in any of our cooling interventions (Figure 3.12). Therefore changes in blood flow to the skin did not reduce the efficacy of the different cooling interventions.

It is possible that there was no vasoconstriction because we had used azaperone (in our drug mixture in the darts), which has α_1 antagonist properties and is therefore a vasodilator (Meltzer et al., 2006b). Shivering during immobilisation also would have affected the efficacy of our cooling techniques. Shivering is stimulated by surface cooling and would have been counter-productive to our cooling intentions as it increases metabolic heat production (Holtzer, 2005). However, we observed no shivering in the blesbok during the immobilisation and cooling period.

Although possible cutaneous vasoconstriction was not a problem in the study, what became problematic were our rectal temperature measurements. We experienced problems measuring rectal temperature measurements in the blesbok because of some equipment failures. Rectal temperatures were recorded during immobilisation initially with digital readers, but some of the devices gave anomalous readings during the study period. These anomalous readings could have been caused by broken thermocouples, a variable depth of insertion and a fault in the electronic reference point within the unit (the ice-point). When the digital readers failed, they were replaced with mercury rectal thermometers, however these mercury rectal thermometers were not calibrated and many different ones were used as quite a few broke in the field. Therefore, I was unable to perform statistical analyses on the rectal temperature results. However, not all of our rectal temperatures measurements were compromised and we were able to obtain some complete data sets with functioning equipment. In these instances, we observed a discrepancy between the rectal temperatures and abdominal temperatures, which were measured by the implanted miniature loggers. Abdominal temperature is closely related to arterial temperature, with a difference of 0.2°C-0.3°C being recorded between the mean abdominal and arterial blood temperatures in a

previous study in eland (Fuller et al., 1999), whereas rectal temperatures can vary during the day due to ambient temperature fluctuations (Fuller et al., 1999). Therefore rectal temperatures are not as a good measure of central blood temperature measurements and thus abdominal temperatures (Fuller et al., 1999). In our study, it seemed that during cooling with water-dousing or ice-packs, the rectal temperature decreased faster than did the abdominal temperature recorded for the animal (Figure 3.13). However, for interventions such as the mist spray, IV infusion and the control, the rectal temperatures and abdominal temperatures were similar (Figure 3.13). The quicker decrease in rectal temperatures during interventions where the body temperature decreased significantly (the water-dousing and ice-packs) may be explained by rapid local cooling in the hindquarter muscles and skin with the venous drainage from the area passing close to the rectum. However, because of the equipment failure these differences between rectal and abdominal temperatures during cooling need to be replicated before we can investigate the reasons for this apparent disparity. If this result is replicated it may cast doubt on the accuracy of rectal temperature readings used in the field while animals are being cooled with water or ice-packs, because in these cases rectal temperature may be a poor reflection of core temperature because it may over-exaggerate cooling of the body.

Besides the equipment problems we experienced there had also been the potential for adverse effects on the blesbok themselves. One concern of cooling hyperthermic animals is the risk of "afterdrop". That is, body temperature continues to fall after the cooling intervention has ended, which may then cause hypothermia (when the body temperature is decreased below the normal temperature range), which may become dangerous for animals when they are to be transported in a cold environment such as at night. Or perhaps a

continued lowered temperature may even be beneficial for an animal after capture (perhaps if they are being transported after capture in a hot humid environment). A novel aspect of this study was that we recorded body temperature of the blesbok continuously before, during and after capture and cooling and therefore we were able to determine how the body temperature changed after cooling had stopped. Other studies that have also recorded body temperature of captured animals have used rectal temperatures (Kock et al., 1987a and 1987b; DelGuidice et al., 2005; Jacques et al., 2009) at the time of immobilisation, thus not providing insight into the body temperatures changes that occur once cooling has ended or when the immobilising agent has been reversed and the animal released. It is important to take measurements during and after capture as after capture rectal measurements become impossible to do once the animal is awake and free. An animal's body temperature may still change after waking up and unless body temperature is recorded after capture we cannot know if the animal will become hypothermic, hyperthermic or return to their normal body temperature. Meyer et al. (2008a and 2008b) recorded body temperature continuously of impala to better elucidate the causes of capture stress, but our study is the first to have obtained continuous body temperature measurements for hyperthermic antelope that were cooled. It is clear that capture increases body temperature dramatically from the normal body temperature rhythm of blesbok (Figure 3.3). It was also clear that some cooling did occur after the cooling intervention had ended. We found that cooling could continue up to 30 minutes after the cooling intervention had ended (an hour after time zero; Fig 3.4). We found that although the cold IV fluids initially (in the first 30 minutes) decreased the body temperature of the blesbok by 1°C, but after an hour the minimum body temperature was similar in comparison to the control (Figure 3.8). Most importantly, we found that in the cooling interventions where

body temperature was significantly decreased up to an hour after time zero (ice-packs and water-dousing interventions; Figure 3.8), in comparison to the control, the body temperature was returned close to the normal body temperature of about 38°C. That is, the animals that received the water-dousing and ice-pack interventions did not have a body temperature decrease at any stage below their daily average body temperature 24 hours after their initial capture. Therefore, the cooling interventions in our study did not cause hypothermia during or after cooling (from the hour after time zero to the next day).

Other potential adverse effects to the blesbok may have arisen from surgery and the invasive nature of implanting the loggers. Even though our team is well experienced and adept at the nature of this surgery, any surgery has inherent risks of infection and can affect an animal's well-being. Therefore, we kept our sample size small for ethical reasons and made use of a repeated measures study design. In our study it was worth having a smaller sample size to gain the continuous data logging during the study period that we could only accomplish through the invasive procedures during surgery. Our sample size ($n = 5$ and $n = 7$ for the two groups) was suitable and sufficient for adequate statistical power, as we found significant differences between the cooling interventions and the control in both groups in all our statistical analyses of body temperature. Furthermore, the responses in both groups to the 28°C water-dousing and the control were similar (unpaired Student's *t*-test, $P > 0.05$). Therefore, the responses between the two groups were similar and hence it was valid to compare across the groups, and combine the two groups to form a larger sample size of 12 animals; when comparing body temperatures and body temperature changes we found that the 28°C water-dousing and the control interventions were also significantly different (paired Student's *t*-test, $P < 0.05$) in this larger group. In addition, this finding reveals that

our smaller sample size in each group reflects the effects that would occur in a larger group of animals. Kock et al's (1987a and 1987b) study is one of the few that have investigated the best capture method for wild sheep and the physiological responses of the animals to capture in North America, which had used a sample size of 634 animals. Our sample size may seem small in comparison to studies such as Kock et al. However, in Kock et al's study rectal temperatures were measured and thus Kock et al. were not able to record the body temperature of animals prior, during and after capture. Also, not only would it be costly and logistically difficult to capture so many animals in South Africa, it is not ethically justifiable to expose so many animals to surgery and for logger implantation (as surgery does bear some risk to the animal itself), especially if a smaller sample size would suffice in a repeated measures design study like ours.

The cooling of wild animals presents unique challenges for capture teams and conservationists, because amenities such as running water and electricity are not available, and events often are unpredictable. Therefore, practicality has to be balanced with efficacy; the best cooling techniques used on humans, or even other domesticated animals, are not always feasible for wild animals. Guidelines do exist for practically and effectively cooling wild animals for Africa, Europe and America (Foster, 2005; Meltzer et al., 2006a; Arnemo and Fahlman, 2008). However, the practice of how animals are cooled has come from years of field experience and has not been validated through scientific studies. So, although current cooling techniques may seem effective, validation is required, particularly to identify the most effective method for a particular capture.

One common cooling practice in field conditions is to pour water over captured animals and to rub the water into the hair. This cooling technique is recommended by capture guidelines in Africa, Europe and America (Foster, 2005; Meltzer et al., 2006a; Arnemo and Fahlman, 2008) but the temperature of the water used may be highly variable because the water may equilibrate to ambient temperature over time. Water stored in a container is likely to be warmer in hot African countries than in European countries with milder ambient temperatures. However, we have shown that even water that has been warmed to 28°C (although, in field conditions in Africa it may be possible that the available water even be hotter than 28°C) will effectively cool an animal when doused over its body surface, because, from our cooling interventions, we have demonstrated that the 28°C water significantly decreased body temperature to less than 39°C, which was a similar body temperature as that which resulted after the 4°C water-dousing. Therefore we have validated the common field practice of dousing with water at a temperature of 28°C or less to cool captured blesbok.

Pouring water over animals can be considered a modified form of water immersion for use in the field. Pouring water over animals and water immersion have a similar purpose to soak the animals in water. Cold water immersion (with water less than 10°C) is the gold standard in human whole body cooling for hyperthermic patients (Casa et al., 2007a). However, there is no added cooling through evaporation during water immersion, but evaporation may be present when water is poured over the animal's surface instead. The blesbok of the water-dousing group were not immersed in water but their fur was thoroughly soaked by pouring water (of 4°C, 17°C and 28°C) over the dorsal body surface and rubbing the water into the fur. Previously, non-anaesthetised hyperthermic horses have been effectively and safely

cooled with 6°C and 15°C water in two different studies with water poured over the horses' body surface (Marlin et al, 1998; Kohn et al., 1999). We found that the 4°C water cooled the animals the quickest, but ultimately there was no significant difference in the minimum body temperature reached between the different water temperatures. All the water temperatures effectively cooled the animals.

We expected that the cooler water (4°C) would decrease the body temperature quicker because of an increased conductive cooling power resulting from a greater temperature difference between the water and the animal (Jessen, 2001; Willmer, 2005). Magazanik et al. (1980) also investigated water immersion with different water temperatures (1-3°C, 10-11°C, 15-16°C, 18-20°C and 25°C) in hyperthermic dogs, which were immersed in water baths. They showed that cooling occurred at all temperatures; the cooling rates of icy water (1-3°C), cold (10-11°C) and cool water (15-16°C) were similar. Clements et al. (2002) also found that ice-water (5°C) and cold-water (14°C) immersion induced similar cooling rates over 12 minutes in humans with exercise-induced hyperthermia. Cold water immersion is considered the gold standard of treatment for heat illnesses in humans; a water temperature range 1-15°C has also been shown experimentally to be highly effective for the treatment of hyperthermia during immersion (Smith, 2005; Casa et al., 2007a). Magazanik et al. (1980) found that cooling with 1-16°C water was quicker than with 18-25°C water. That is, the dogs still had a significantly decreased body temperature with 18-25°C water but cooling was faster with cooler water. We had a similar result where the 28°C water-dousing effectively lowered the body temperature (to a minimum similar to the 4°C water) in the blesbok but not as quickly as when the 4°C water was used.

Other heat loss principles used in cooling techniques are evaporative and convective heat loss using air movement. Runners have been cooled successfully with fanning air over their body surface (Barwood et al., 2009). The use of electric fans in sporting events was found to be practical, cheap and effective in decreasing body temperature (Barwood et al., 2009). In cooling human heat stroke victims, evaporative cooling is not the first line choice of treatment (water immersion is), but evaporative techniques (such as fanning and whole body cooling and wetting units) have also been shown to be an effective method to cool heatstroke victims, particularly when combined with other cooling techniques (Smith, 2005; Bouchama et al., 2007). It is because of the increased efficacy of fanning when combined with another cooling technique that we decided not to investigate fanning alone but to investigate fanning combined with water-dousing. I compared the addition of fanning to the 28°C water-dousing to dousing with 28°C water alone, but I found no significant differences between the two cooling methods in lowering the body temperature of blesbok. The vapour pressure of the air in the bomas was, on average, 2.5kPa and was therefore low enough (as the kPa of wet skin at a temperature range of 34°C - 43°C is 4.9kPa to a value greater than 6.3kPa, which forms a vapour pressure gradient with water being vaporised and moving from the higher skin vapour pressure to the lower ambient vapour pressure) to allow for evaporative heat loss. Our expectation was that fanning when combined with the 28°C water-dousing, which increased the wind speed over the animal, would have increased heat loss by increasing the air convection over the animal and the evaporation of water off the animal (Jessen, 2001). However, we fanned our animals with a rigid sheet of plastic and did not use electric fans, as there is no electricity in field conditions. Even battery-operated fans are too inconvenient to carry into a field situation, and so we simulated what is currently performed in the field to cool hyperthermic animals. The lack of a significant additional fall

in body temperature with the addition of fanning may be because our fanning wind speed was too low. The wind speed we generated was about $1.2\text{m}\cdot\text{s}^{-1}$, but the recommendation for alleviating heat stress in cattle is a wind speed of 2 to $4\text{m}\cdot\text{s}^{-1}$ (Gaughan et al., 2004). Also, when athletes were exercised in hot, humid conditions and were then cooled by fanning alone over their body surface, it was a wind speed of 3.5 to $3.84\text{m}\cdot\text{s}^{-1}$ that was successful in decreasing body temperature back to the pre-exercise normal (Barwood et al., 2009). However, to maintain such a wind speed would be difficult without the use of electronic or battery-operated fans, as we fanned for periods of five minutes, a tiring effort for volunteers. It is also likely that the reason that fanning did not significantly improve the 28°C efficacy is because it may be that whole body cooling by immersion is in itself much more effective than evaporative cooling techniques, and so the addition of fanning to our water-dousing would only have a minimal additional benefit (Marlin, 1998).

Capture guidelines also recommend the use of cold IV infusions, mist sprays and packing ice under the axilla, groin and tongue (Foster, 2005; Meltzer et al., 2006a; Arnemo and Fahlman, 2008). We investigated some of these methods as alternatives to water-dousing. Because the placement of ice-packs to just the axilla and groin has proven ineffective in humans in significantly decreasing core body temperature (Smith, 2005), we placed ice-packs over the body surface as well as ice-packs in the axilla and groin. In dogs exercised on a treadmill, ice-jackets around their trunks have been shown to decrease rectal temperature in comparison to non-cooled dogs (Kruk et al., 1985). In our study we also found that ice-packs cooled effectively by decreasing body temperature in comparison to the control intervention.

In humans, both ice-jackets (Quod et al., 2006) and water immersion, in baths or showers, (Marino, 2001) of 28°C have been used to successfully pre-cool athletes before sporting events and after sporting events from an exercise-induced hyperthermia (Armstrong et al., 1996), but no direct comparison, to my knowledge, has been conducted between these two methods. In our study the ice-packs had similar effects to the 28°C water-dousing in that their effects on minimum body temperature reached, change in body temperature, the decrease in body temperature with time and how quickly the animal was cooled were no different. The two methods may have had similar outcomes, despite the much lower temperature of the ice-packs, because cooling the core from the surface by ice-packs is a slow process (Rajek et al., 2000). This pattern is demonstrated for ice packs in Figure 3.8 and Figure 3.9, which show how the change in body temperature and the decrease in body temperature with time are much greater after an hour than over 30 minutes. The ice-packs also would have warmed over the 30 minutes of application, both from the animal's body heat and from the ambient temperature, whereas the 28°C water was maintained at 28°C throughout dousing. It is worthwhile to note that our ice-packs may have warmed but did not melt during the 25 minutes of placement over the blesboks' body surface, and therefore all the cooling was conductive. Also, the 28°C water was rubbed into the fur and therefore there was direct contact between water and the skin (which would have increased heat loss by conduction and convection), but the ice-packs over the body surface were placed on the fur, therefore the trapped air between the fur and skin, that acts as an insulative layer, would have been undisturbed (Jessen, 2001). Fur is also less thermally conductive than skin so heat would have been transferred slower between the ice-packs, fur and skin than between water and the skin (Jessen, 2001). The ice-packs may have also prevented

convective heat loss by forming a barrier over the fur and preventing air movement between the fur and over the skin (Jessen, 2001).

The use of cold IV fluids and mist sprays has also been recommended in the field to cool wild animals (Foster, 2005; Meltzer et al., 2006a; Arnemo and Fahlman, 2008). In our study the IV infusion led to a significant decrease in body temperature with time (thermal response indices, Figure 3.9) in the blesbok over 30 minutes and over one hour. At 30 minutes, the IV infusion rapidly decreased the temperature by 1°C (Figure 3.8), which was a large enough decrease to affect the magnitude of the decrease in body temperature over time (thermal response indices at 30 minutes and one hour) but was not an adequate decrease to return hyperthermic animals' body temperatures to normal (Figure 3.7). At 30 minutes, the IV infusion decreased the body temperature over time similarly to that of the ice-packs (Figure 3.9). This similarity in the decrease in body temperature over time between the IV infusion and ice-packs is also because of the rapid 1°C decrease in body temperature that occurred within the first 30 minutes of the IV infusion. The ice-packs decreased the temperature by about 2.5°C, but slowly over a period of one hour. The ice-pack's larger, but relatively slower change, in body temperature and the IV infusion's rapid change in body temperature had a similar effect on altering the magnitude of the decrease in body temperature over the first 30 minutes, which would have led to similar decreased temperature values with time within those 30 minutes. Even though there may have been statistical differences in the temperature decrease with time (at the end of 30 minutes and one hour) between the control and the IV infusion and mist spray, these were not physiologically important differences as there was no significant change in the minimum body temperature in comparison to the control (within one hour), nor was there a

difference in how quickly both these cooling interventions cooled in comparison to the control. Therefore, the IV infusion and mist spray, when considered overall, were clinically ineffective in causing a substantial decrease in body temperature to alleviate a capture-induced hyperthermia.

Cold IV infusions directly reduce the temperature of the blood and mist sprays are believed, anecdotally, to better soak an animal than does water-dousing. Some people also believe, anecdotally again, that the water sprayed becomes cooler from the effect of evaporation on the water droplets as it travels through the air and therefore cooler water lands on the animal. Mist sprays, I believe, are used in the field because those who use them have had a previous experience of the thermal comfort they produce (largely by lowering facial skin temperature). However, our study has shown that a fine mist spray was ineffective in decreasing the body temperature of animals. The IV infusion did decrease the animals' temperature by about 1°C during the 30 minutes immobilisation, but this decrease was not enough to return the animals' body temperature to normal body temperature for that time of day. Studies in humans using 4°C isotonic saline have achieved varying success. One study reported a mean decrease of 2.5°C over 30 minutes of 4°C isotonic saline by a central infusion (via the use of a central venous catheter) of 40ml.kg⁻¹ of saline in healthy volunteers (Rajek et al., 2000), whereas another study reported a decrease in body temperature of only 1°C by infusing 30ml.kg⁻¹ of 4°C saline peripherally in healthy volunteers for 30 minutes (Moore et al., 2008). Rajek et al. (2000) argued that infusing peripherally is not as effective as infusing centrally because heat is transferred mainly to peripheral tissues rather than core tissues. However, the primary cause for the inefficacy of the IV infusion is the volume that was infused. Prior to the start of the study we calculated the cold load of the IV infusion

on the animal. The change in body temperature we calculated for 1L of saline (0.8°C) was similar to the actual change in body temperature we recorded during the study (about 1°C). Therefore, from this calculation it was clear that the body temperature decrease would only be about 1°C, which was independent of the infusion site (peripherally or centrally) but dependent on the volume infused. Doubling the volume of the infusion should double the fall in body temperature. However, for practical reasons it would be difficult to infuse larger volumes in the field as the use of a too large a volume of an IV infusion may lead to pulmonary oedema and would take a prohibitively long time to infuse (Bouchama et al., 2007). We took, on average, about 12 minutes to infuse one litre of saline.

The mist spray was ineffective because not enough water was able to saturate the fur of the animals. The animals were sprayed with 28°C water; 5L at the same five-minute intervals as the 28°C water-dousing, but at the end of the five-minutes not all the water had been sprayed on the animals (about 1-2L were left). Indeed, it was impossible to spray all of the 5L onto the animal in five-minutes. In the 28°C water-dousing intervention the water was also rubbed into the fur; this procedure was not done in the mist spray intervention as the rationale of using the spray in the field is that it soaks the fur and rubbing is not necessary. Fine mist sprays are used in humans to increase thermal comfort because small water droplets evaporate quickly off bare skin, but again fur hinders the evaporation of droplets off the skin and thus hinders evaporative cooling.

I also believe the reason why an IV infusion and mist spray technique hold such appeal for capture teams is because of their ease of use in the field. An IV bag is small, easy to transport and to keep chilled in a cooler box with ice. A mist spray is practical because it is

also light and easier to carry than a water drum of the same volume because of the shoulder straps on the sprayer. However, as these techniques have no advantage in cooling an animal their practical appeal should be disregarded in favour of a cooling technique that is effective and practical. Ice-packs may be as effective as a 28°C water-dousing but they are not that practical because they are bulky and many sheets are required to cover an animal's body surface, which makes ice-packs expensive and difficult to transport if a large enough cooler box is available. Also, ice-packs may only be effective on one animal in a day due to melting. Water-dousing is an effective and practical cooling method. Firstly, water is the most inexpensive of all the cooling materials. Also, a water source is may be available in the field for refilling water drums (if they run empty); water drums are hardy and easily transported, even in larger volumes. Smaller volumes of water can also be kept cool in cooler boxes.

We have demonstrated which techniques are effective in cooling animals in our protocol. The mechanism by which cooling may have occurred in the water-dousing and ice-packs interventions was through the convective heat transfer via blood flow through the body compartments and from conductive heat transfer (conduction through the tissues), from the body core to the muscle and through the muscle to the skin (Ducharme and Tikuisis, 1991). Although, subcutaneous fat is three to four times less thermally conductive than muscle, thermal conductivity experiments in the human forearm immersed in cold water and warmer water (of 15 to 30°C) have shown that it is muscle and not subcutaneous fat that accounts for 92% of the total thermal insulation of the forearm during cold water immersion (Ducharme and Tikuisis, 1991). Muscle has been said to insulate 70-80% of the body in humans and therefore is a considerable factor in overcoming insulation for core cooling (Lemire et al., 2008). The thermal conductivity of a tissue is dependent on the

tissue's composition and the blood flow through that tissue (Ducharme and Tikuisis, 1991). As blood flow decreases through a tissue the greater the tissue insulates (Ducharme and Tikuisis, 1991). I have previously mentioned that there were no changes in the vasomotor tone (Figure 3.12) and that with the possibility of constant cutaneous vasoconstriction present in the blesbok (and therefore decreased blood flow to the periphery). This vasoconstriction could have also occurred in the muscle and may have also resulted in a lower thermal conductivity through the muscle. Muscle blood vessel vasoconstriction may have occurred in the blesbok because it is well known that the stress of capture causes an increase in sympathetic tone and this increase in tone causes vasoconstriction in muscles (Thomas and Segal, 2004).

However, if the blood vessels were mostly vasodilated after the capture, as during exercise the contraction of skeletal muscles increases blood flow through the muscle (Thomas and Segal, 2004) the conductive (Marlin et al., 1998) and the convective heat transfer between the muscle and the blood (Weller, 2005) would have increased. This heat transfer through the different compartments would have overcome the insulative properties of the muscle and fat. Heat would also be exchanged through conduction between the cooling intervention on the skin surface and the warmer muscle. Also because of the increased blood flow and volume in the limbs after exercise, more hot blood would be cooled at the skin surface and would be returned as cooler blood from skin to the muscle and into the core (Lemire et al., 2008). Marlin et al. (1998) used hyperthermic horses to investigate changes in compartmental body temperature after cooling by water-dousing the horses with 5l of cold water (6°C) at 30 second intervals. They also observed no initial vasoconstriction (a similar finding to the lack of cutaneous vasoconstriction that we

measured through our ear-skin temperatures) and through thermography and muscle temperatures demonstrated that heat exchange occurs primarily between the muscle and the cooling intervention. Skin itself is also an important avenue of heat loss. As in muscle, after exercise blood vessels in the skin are vasodilated and increased blood pressure (because of the increased cardiac output after exercise) shunts blood to the periphery and the skin (Willmer et al., 2005). The increased blood flow to the skin increases the heat radiated and convected from the skin to the environment; sweating and evaporation (in animals that are capable of sweating) and the conductive heat loss to the environment from the physical contact between the skin and the environment, for example the ground or a cooling intervention (Jessen, 2001). Subsequently, cooled blood from the skin is transported back to the core.

We measured the subcutaneous temperature of the animals and compared it to core temperature to determine if heat was being transferred between the core and the periphery, that is, if there was any subcutaneous vasoconstriction. The core temperature minus subcutaneous temperature was compared between the animals by using the different water temperatures and a control intervention. The 4°C water-dousing led to the largest peak difference (a difference of 3.5°C) in the core minus subcutaneous temperature in the blesbok (Figure 3.11). That is, there was some vasoconstriction occurring in the blesbok, however this vasoconstriction was not sufficient to prevent the 4°C water from decreasing the body temperature by about 3°C (Figure 3.8). The mechanism by which the 4°C cooled quicker than the control and the 28°C water is most likely through increased conductive and convective cooling.

In the ice-packs intervention there also was an increased temperature difference between the core minus subcutaneous temperature in the blesbok. The peak difference was 3°C (Figure 3.11) between the core temperature and subcutaneous temperature in the ice-pack intervention. This peak difference, caused by the ice-packs, between the core and the periphery was significantly larger than in the IV infusion, mist spray and 28°C water-dousing interventions. So again, it is likely that there was some subcutaneous vasoconstriction but the ice-packs were still an effective intervention and decreased the blesbok body temperature by about 2.5°C after an hour (Figure 3.8). The ice-packs' efficacy is due to a high conductive cooling power because of the prolonged contact between the ice-packs and animal (which was over 30 minutes whereas water was poured for only five-minute intervals). However, even though there was an increased temperature difference between the core and the periphery, which may have been due to subcutaneous vasoconstriction, there was ultimately no difference between the ice-packs and the 28°C water-dousing in how quickly the animals were cooled. A possible reason why the ice-packs and 28°C water dousing had similar cooling rates was because the ice-packs were placed over the surface of the blesbok and would have impaired air and water movement over the blesbok's skin surface and thus only cooled via conduction. Whereas, the 28°C water dousing would have cooled by conduction and convection and these two combined avenues of heat loss may have been equal in cooling power to the increased conductive cooling of the ice-packs.

I now believe we have sufficient evidence to recommend the most effective cooling techniques for blesbok (and possibly other medium body sized antelope). It is clear that irrespective of the water temperature (at least up to 28°C) water-dousing is the most effective method for cooling hyperthermic blesbok in the field. The water-dousing was

effective because all the water temperatures (4°C, 17°C, 28°C) decreased the body temperature significantly in comparison to the control. No water temperature decreased the body temperature more than did another. Common practice in the field may be to use whatever water is available and we have validated a water temperature range (4 to 28°C) which will cool hyperthermic blesbok effectively. However, the 4°C water-dousing cooled the animals the quickest. Therefore, if an animal is hyperthermic it is important to douse it with water, and water with a temperature between 4°C and 28°C will be effective. However, if cold water is available and can be kept chilled in the field then this water will cool an animal the quickest. Ice-packs were also effective in lowering the body temperature, and were as effective as the 28°C water-dousing. However, the ice-packs were quite bulky and heavy, and may be cumbersome to use in the field, difficult to transport and to keep cold, so, using 28°C water in plastic drums may be more practical. The mist spray was ineffective in decreasing body temperature, and therefore I cannot recommend its use for hyperthermic animals. The cold IV infusion had a limited effect on decreasing the blesboks' body temperature (about 1°C decrease over 30 minutes and an hour) because not enough cold water could be infused without risking adverse effects to the animal. Therefore I cannot recommend its use for cooling hyperthermic animals if other cooling methods are available.

CHAPTER 5

CONCLUSION

5. CONCLUSION

Our study has been the first to systematically investigate the cooling methods used to alleviate capture-induced hyperthermia in wildlife. Previously, the cooling techniques used in the field have been selected based on anecdotal evidence that they are effective. The most common method that is used in the field is the dousing of animals with water but, in recent years, techniques such as using mist sprays also have been employed (Meltzer et al., 2006a).

It is clear that irrespective of the water-temperature (between 4°C and 28°C), water-dousing is an effective method for cooling hyperthermic animals in the field. These recommendations probably apply only to antelope of a medium body size, as the amount of heat exchange between an animal and its environment will change in different body sizes, shapes, pelages, and other morphological and physiological characteristics specific to a particular species. A change in heat transfer with body size results mainly because of a change in the surface area to volume ratio. That is, smaller animals may require less cooling because of their increased surface area to volume ratio, which means these animals lose heat more rapidly to the environment than does a larger animal (Willmer et al., 2005). Conversely, large animals will have a larger thermal inertia (that is, because of their larger body size it takes larger animals much longer to lose any gained heat because of their smaller body surface area to volume ratio) than a smaller animal's body and would require greater cooling (Willmer et al., 2005). Excessive water-dousing in smaller animals may be associated with the risk of overcooling (decreasing the body temperature below the target normal range), potentially leading to hypothermia. Therefore it is important that further

studies be conducted in different species to determine the most effective cooling method for that species or groups of species of similar sizes.

Differing size, may be the most important, but is not the only variable between species that will affect the efficacy of cooling. The animals' pelage will affect the amount of heat transferred between the skin of the animal and the environment. Fur has a lower thermal conductivity than does skin (Jessen, 2001), and so heat is transferred more slowly across fur than it would be with direct skin contact between the cooling intervention and an animal. Different species also have a varying denseness of fur, and fur density may vary seasonally. Colder climate species, such as polar bears and reindeer, have a thicker pelage than do blesbok. Thicker dorsal fur may also be found in desert sheep and goats, which inhabit environments where the air temperature often exceeds the animals' body temperature (Willmer et al., 2005). In these conditions thicker fur protects against heat gain by decreasing radiation, convection and conduction from the environment to the animal (Willmer et al., 2005). However, thicker fur may impede heat transfer between a cooled substance and an animal and therefore a more intense cooling method may be necessary (for example, with greater water volumes or lower water temperatures). Animals can also develop thicker fur as part of their long-term strategy to conserve heat in cold ambient temperatures; hair may be lighter and thinner in summer when ambient temperatures are warmer and hair may become denser during winter for better insulation (Willmer et al., 2005). Therefore the efficacy of a cooling technique may change with the seasonal variation of a species' fur.

Other factors will also affect the ability of animals to lose heat, for example, health status (the body condition), age and state of hydration (Jessen, 2001). These factors also may contribute to how much an animal's body temperature rises in response to capture. Animals that are in poorer physical fitness are more susceptible to the adverse effects of hyperthermia, such as decreased cardiac output and increased central nervous system dysfunction, and therefore are more susceptible to morbidity and mortality (Jessen, 2001). In these less physically fit animals it becomes important to cool them as quickly as possible to decrease the risk of adverse effects developing. However, a thin, small animal in poor condition may become hypothermic after cooling is applied after capture compared to a larger, healthier animal that is treated with the same cooling intervention. Therefore, effective and quick cooling methods, that do not cause adverse effects such as hypothermia, should also be investigated in less physically fit animals, or care should be taken to monitor the fall in body temperature during cooling efforts.

Especially as there are certain species that are more susceptible to developing hyperthermia than others. Eland tend to run long distances during capture and are susceptible to hyperthermia (because intense exercise may exacerbate a capture-induced hyperthermia) and capture myopathy (Burroughs et al., 2006) and impala, which have an excitable nature, also tend to be highly stressed and thus susceptible to hyperthermia during capture (Meyer et al., 2008a). We elicited a hyperthermia of 41-42°C in our blesbok but the hyperthermia experienced in the field in blesbok and other species may be as great as, or even greater, than 43°C (Meyer et al., 2008a). We would need to investigate if our cooling techniques are still effective if body temperature is increased to, or greater than, 43°C. In animals that are susceptible to hyperthermia it may be necessary to cool the animals as quickly as possible;

this rapid cooling may be achieved by dousing with larger volumes (greater than 5l) of water. In addition, using small volumes (5l) of cold water (4°C) may be as useful as large volumes of warmer water because we have shown that 4°C water induces rapid heat loss and provides sustained cooling.

Besides interspecies variables that may warrant further investigation on how they affect cooling efficacy, there are also still a variety of other cooling techniques in the field, which need investigating. However, firstly it would also be worthwhile to investigate if our techniques are effective in other antelope species and if dousing with water that is warmer than 28°C is effective. Other techniques that have been anecdotally reported to be used in the field are the placement of leaves or hessian sacks over animals, wetting the ground under the animal, enemas with cold water, and alcohol and water mixtures poured over the animal (Fowler, 1995; Foster, 2005; Meltzer et al., 2006a). However, there is no published data on how effective these cooling methods are. We have demonstrated that mist spray, a recommended technique (Meltzer et al., 2006a), is an ineffective cooling method. Therefore, future studies should also evaluate how effective other recommended cooling techniques actually are.

The capture method used is also important when determining the cooling intervention for an animal. Animals that are darted in woody areas can be placed under the shade of trees and larger species (that are difficult to move) can be covered in leaves (to decrease radiant heat gain from the sun) (Meltzer et al., 2006a), but this may compromise heat loss from evaporation and convection. Animals that are chemically restrained can be doused with water and can be given IV fluids (Meltzer et al., 2006a), but as we've shown, IV fluids should

not be the first choice for cooling animals. For animals that are physically restrained it would be more difficult to administer IV fluids or to douse them with water, especially if these animals are caught with mass capture techniques. It would also be difficult to individually hold and douse 40 animals that have been caught in nets, but this technique still may be the most practical method for animals in nets in comparison to any of the other techniques that I investigated. Therefore, other cooling techniques would warrant investigation under these circumstances, such as hosing animals with water, or transporting them in a well ventilated vehicle to increase convective cooling.

Along with the consideration of using different cooling techniques for different capture methods, these considerations may also apply to the variety of capture drugs used. Future studies could focus specifically on the effects of capture drugs on heat loss and temperature regulation. Some studies have already examined the relationship between thermoregulation and capture drugs (Meyer et al., 2008b). Fick et al (2006) studied the effects of zuclopenthixol, haloperidol and perphenazine (tranquilising drugs used after capture) on boma-housed wildebeest and found that these drugs did not adversely affect thermoregulation. However, more studies specifically examining how specific capture drugs affect heat loss are needed because different classes of capture drugs affect thermoregulation in different ways. The opioid class of drugs inhibit central thermoregulation (in the pre-optic anterior hypothalamus) and therefore induce thermal lability, which means that the effector pathways to correct increased heat gain are inhibited and temperature regulation becomes poikilothermic (Clark, 1979; Swan, 1993). That is, that the body temperature will tend towards ambient temperature. Cyclohexamines, specifically ketamine, can cause hyperthermia by increased metabolic activity, muscle convulsions and

increased muscle tone (Swan, 1993). Sedative drugs, such as xylazine, also disrupt central thermoregulation, decrease cardiac output and decrease peripheral blood flow (Swan, 1993). One drug that may increase heat loss to the environment is azaperone, as it increases peripheral blood flow by increasing peripheral vasodilation (Swan, 1993; Meltzer et al., 2006b). The different ways these drugs effect thermoregulation may influence the efficacy of a cooling technique. Therefore, different capture drugs combined with different cooling interventions should be investigated to optimise the choice of cooling methods for specific drug combinations.

All of the above mentioned capture factors can be combined to reduce heat stress. That is, capture technique should be refined so that the ideal capture drug and capture method is used in each situation. Capturing animals quickly, reducing human interference, reversing drugs timeously and releasing animals quickly will decrease the amount of heat stress that an animal is subjected to and would therefore attenuate the initial hyperthermia present in the animal.

It is important to improve heat transfer by optimising cooling and drug combinations but it is also important to research and better understand the mechanisms of heat loss available during immobilisation. A technique that could be used is to examine the anatomy of captured animals through the use of ultrasound. A portable ultrasound could examine the thickness of the subcutaneous layer of the captured animal (DelGiudice et al., 2005) to correlate the amount of subcutaneous insulation with effective heat transfer. The amount of fat would also provide a quantitative index for the physical condition of an animal to determine if animals in poorer physical condition are susceptible to overcooling. The

ultrasound may also be used to examine organs that are prone to damage after capture and during hyperthermia, to examine if swelling has occurred or there is any cardiac dysfunction during capture. Then it can be investigated if cooling methods have any visible protective effect on these organs.

The use of remote monitoring equipment, such as implanted loggers, may also reveal further physiological changes during and especially after capture, because the morbidity from capture-induced hyperthermia may make animals more prone to predation because of decreased activity (Abbot et al., 2005). It is possible that because cooling alleviates hyperthermia that the animals may recover faster and not experience decreased activity after capture. It may be possible that continued cooling of the body after a cooling intervention may make an animal sluggish and therefore also prone to predation. However, during our cooling interventions the body core temperature after cooling typically still decreased 30 minutes after the cessation of cooling, but this decrease returned body temperature to normal and did not cause hypothermia. Therefore, it should be investigated if cooling aggravates a decrease in an animal's activity after capture or if cooling promotes a return to normal activity levels and may enable captured animals to be less susceptible to predation. Implanted or collared activity loggers could be used to remotely monitor activity and assess if a change in activity is one of the aspects of the long-term consequences of applying cooling interventions. Also, night-time hypothermia that has been observed post-capture and it would be worthwhile to establish if inactivity and a change in thermoregulatory behaviour in the late afternoon influence this hypothermia. Furthermore, it would be worth investigating whether cooling in the late afternoon would predispose an animal to developing hyperthermia at night.

Further elucidation of site-specific temperature changes in the body of captured animals is also possible with further temperature measurements. One method of recording surface temperature is thermography, which uses infrared wavelengths to image the temperature changes occurring on the skin surface. Marlin et al. (1998) used thermography in combination with the recording of muscle and rectal temperatures to determine the dynamics of compartmental changes in horses after they had been doused with water. The use of thermography on wild animals could reveal sites on the animal's body surface that act as thermal windows (where body heat is actively exchanged with the environment) or could reveal sites where animal's body surface is the hottest. Knowing where these sites are would reveal body areas that future cooling interventions efforts could be focused upon or reveal how effectively heat is being transferred from the cooling intervention to the animal's body surface.

Lastly, the recording and reporting of body temperature in the field needs to be improved. The most common method of measuring body temperature in the field is with rectal thermometers and it is recommended that body temperature be monitored regularly as part of the standard practice during capture (Meltzer et al., 2006a; Arnemo and Fahlman, 2008). I believe that body temperature measurements should still be recorded rectally, even though we observed a discrepancy between rectal temperature and core temperature during cooling in our blesbok (Section 3.11). At the moment rectal temperature measurements are the only practical and economic method of obtaining temperature measurements that usually are considered to be near core temperature in captured animals in a usual field setting. In North America, digital thermometers are recommended because

they are easier to read than glass thermometers (Foster, 2005). However, digital thermometers that have been designed and tested in and for laboratory settings may be affected by solar radiation, wind and ambient temperature, which could affect the integrity of body temperature measurements (Casa et al., 2007b). Also rectal thermometers (digital or mercury) usually are not calibrated, which can also lead to inaccurate measurements of body temperature in captured animals. Inaccurate equipment in the field may be problematic because inaccurate thermometers may lead to the delay in initiating the cooling of hyperthermic animals and therefore decrease the efficacy of cooling techniques in alleviating hyperthermia. Ideally, field workers and researchers should calibrate and report the accuracy of any rectal thermometers used.

However, few published studies have recorded or reported rectal temperatures, or performed autopsies on animals that died post-capture to determine the precise cause of death, which makes it difficult to determine the incidence of hyperthermia and the relationship between hyperthermia and mortality (Gallagher et al., 1985, Pigozzi, 1987; Cattet et al., 2003; Viljoen et al., 2008). The papers cited in this dissertation may only be few but it is important to remember that capture operations are routine for both conservation and game management operations, and the findings from there may not be published in the form of scientific studies. Therefore the knowledge is not easily accessed or assessed and it becomes easy for anecdotal evidence to become part of the capture routine without sound scientific basis. That is, field workers may record rectal temperature while an animal is being translocated but that temperature record is for their own purposes and is therefore not generally available. If high body temperatures (perhaps 3°C above the animal's normal body temperature) during a capture operation were reported regularly and collated into a

database by field workers (that other capture professionals, veterinarians and scientists had access too) then it would be possible to determine the incidence of hyperthermia per species, mortalities and the cause of death and the type of cooling procedures used. Therefore, better reporting of body temperature during capture will lead to a better understanding of how prevalent capture-induced hyperthermia is and what effect it has on morbidity and mortality and whether cooling techniques are protective or not. Once these aspects of capture and cooling are known (through further scientific studies and better reporting of body temperature during capture), then perhaps new guidelines may be drawn up for the capture of animals.

There is still much research that needs pursuing in regards to capture-induced hyperthermia itself and how cooling methods can be optimised to reduce animal morbidity and mortality. However, I do feel that we have taken the first steps in scientifically investigating and researching methods that could be used in South Africa and perhaps internationally to alleviate capture-induced hyperthermia, at least in antelope. We now know that water-dousing and ice-packs are effective methods but that cold IV infusions and mist sprays are ineffective in substantially reducing the body temperature increase from capture. Therefore, I believe that dousing animals with water should continue as the standard practice, as it a practical, cheap and effective method, at least until other cooling techniques are investigated and prove to be more effective. I hope that these results will help wildlife professionals make an informed choice as to which cooling technique to use in cooling hyperthermic animals.

CHAPTER 6:

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6. REFERENCES

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