

## Thermal, cardiorespiratory and cortisol responses of impala (*Aepyceros melampus*) to chemical immobilisation with 4 different drug combinations

L C R Meyer<sup>a\*</sup>, R S Hetem<sup>a</sup>, L G Fick<sup>a</sup>, A Matthee<sup>b</sup>, D Mitchell<sup>a</sup> and A Fuller<sup>a</sup>

### ABSTRACT

Thermometric data loggers were surgically implanted in 15 impala (*Aepyceros melampus*) to investigate the consequences of chemical capture. Impala were darted and chemically immobilised for 30 min with each of the following drug combinations: etorphine and azaperone; etorphine and medetomidine; thiafentanil and azaperone, and a thiafentanil medetomidine combination. During immobilisation, pulse oximeter readings, respiratory rhythm, the plane of immobilisation and plasma cortisol concentrations were measured and recorded. The impala developed an extremely high rise in body temperature, which peaked 20–30 min after reversal of the immobilisation. The magnitude of the rise in body temperature was similar for all the drug combinations ( $F = 0.8$ ,  $P = 0.5$ ), but the duration of the hyperthermia was shorter when the thiafentanil and azaperone combination was used ( $F = 3.35$ ,  $P < 0.05$ ). Changes in body temperature were related to the time that it took for an animal to become recumbent after darting ( $r^2 = 0.45$ ,  $P = 0.006$ ) and not to the effect of the drug combination on time to recumbency ( $r^2 = 0.29$ ,  $P = 0.46$ ). The relationship between time to recumbency and body temperature change, and also to plasma cortisol concentration ( $r^2 = 0.67$ ,  $P = 0.008$ ), indicated that physiological consequences of capture were related to the duration of exposure to a stressor, and not to the pharmacology of the capture drugs. Although shorter time to recumbency in individuals resulted in the benefit of smaller stress responses and body temperature changes, those individuals were predisposed to developing hypoxia and possibly induction apnoea. When animals are chemically immobilised, reducing the thermal consequences of capture requires limiting the exposure of the animal to a psychological 'fright stress'.

**Key words:** body temperature, stress, etorphine, thiafentanil, medetomidine, azaperone, stress-induced hyperthermia, impala (*Aepyceros melampus*).

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ungulates. Cardiorespiratory and plasma cortisol responses to capture were also measured. The drug combinations used consisted of a narcotic opiate drug, used to induce catatonic immobilisation and sedation, and a tranquiliser or sedative, used as an adjuvant to further calm the animals. No studies have investigated the differences between the thermoregulatory effects of etorphine and thiafentanil in ungulates. Thiafentanil induces immobilisation more rapidly and has a shorter duration of action than does etorphine<sup>15,20</sup>. Medetomidine and azaperone are supposed to disrupt central thermoregulatory control and cause thermal lability<sup>8,14</sup>; they affect peripheral blood flow differently, and, hence, heat flow to and from the environment. Azaperone causes vasodilation through its  $\alpha_1$ -antagonistic effects, and therefore increases peripheral blood flow<sup>8,14</sup>; whereas medetomidine causes vasoconstriction and a fall in blood pressure and cardiac output, thus decreasing peripheral blood flow<sup>9,20</sup>. Impala (*Aepyceros melampus*) were chosen as experimental animals, because they are known to be highly excitable and reactive to capture<sup>18</sup>, and because they are known to develop capture-induced hyperthermia<sup>22</sup>.

### MATERIALS AND METHODS

The procedures were approved by the University of the Witwatersrand's Animal Ethics Screening Committee (clearance number 2004/11/05). The study took place between December 2003 and December 2005 at the Lichtenburg Game Breeding Centre (26°07'S, 26°07'E) of the National Zoological Gardens, 220 km west of Johannesburg, South Africa.

#### Animals and surgery

Fifteen adult (mean mass of  $37.5 \pm 3.5$  kg) female impala were caught from the wild and transported to bomas (5 m × 10 m holding pens with solid 3 m-high wooden pole walls) no less than 2 weeks before surgery. On the day of surgery, the impala were herded into a game-transport vehicle where they received a

### INTRODUCTION

When wild ungulates are immobilised chemically, using capture drugs in a dart, they typically develop an extremely high rise in body temperature<sup>2,4,6,11,13,17,22</sup>. This rise in body temperature may result in lethal hyperthermia, but it also increases cellular oxygen consumption<sup>12,16</sup> in animals that may already have a negative oxygen balance from drug-induced hypoxia and a high metabolic demand from intense activity during escape attempts. If cellular energy production cannot keep pace with normal cellular needs, normal

cellular function and integrity are disrupted, which ultimately could lead to organ failure<sup>12</sup> and capture myopathy<sup>21</sup>. Understanding why body temperature rises is crucial to survival during chemical capture, but very little actually is known about its cause. Meltzer and Kock<sup>21</sup> suggested that the rise in body temperature during chemical capture may be related partly to the effects of capture drugs on the animal's thermoregulatory processes.

The potential effects of capture drugs on an animal's thermoregulation may vary depending on the pharmacological effects of the drugs, and so the use of different drugs could result in different body temperature responses during capture. The aim therefore was to determine the thermal response of impala to chemical capture using different drug combinations used to immobilise wild

<sup>a</sup>Brain Function Research Group, School of Physiology, University of the Witwatersrand, 7 York Road, Parktown, 2193 South Africa.

<sup>b</sup>National Zoological Gardens Lichtenburg Game Breeding Centre, PO Box 716, Lichtenburg, 2740 South Africa.

\*Author for correspondence.  
E-mail: leith.meyer@wits.ac.za

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tranquilliser azaperone 40 mg (Stresnil, Janssen Pharmaceutica, Johannesburg, South Africa) or haloperidol 15 mg (Kyron Laboratories, Johannesburg, South Africa) intra-muscularly (i.m.) *via* a pole syringe. Once tranquil, impala were captured individually by hand and anaesthetised using halothane (Fluothane, Astra Zeneca, Johannesburg, South Africa) in 100 % oxygen delivered *via* a face mask. A 50 mm incision was made through the midline in the ventral abdominal wall and a miniature temperature-sensitive data logger (described below) was placed, without tethering, into the abdominal cavity. Post-surgery, each impala received a long-acting penicillin-based antibiotic (4–5 ml, i.m., Peni LA Phenix, Virbac Animal Health, Johannesburg, South Africa), an analgesic and anti-inflammatory containing 140 mg/ml ramiphenazone, 70 mg/ml sodium phenylbutazone and 0.5 mg/ml dexamethasone (4–5 ml, i.m., Dexametanol, Centaur Labs, Johannesburg, South Africa), and a long-acting parasiticide, doramectin (5 mg, subcutaneously (s.c.), Dectomax, Pfizer Laboratories, Johannesburg, South Africa). The impala were marked with different coloured plastic ear tags for identification. Once the impala recovered from anaesthesia they were returned to the bomas where they were housed for the duration of the study.

At the end of the study, the impala were re-caught, and the data loggers were removed under an anaesthetic and surgical procedure similar to that used for implantation. The animals were then released back into the main reserve.

#### Body temperature measurements

Core temperature was measured at 10-min intervals in the abdominal cavity of the impala with miniature temperature-sensitive data loggers (StowAway XTI, Onset Computer Corporation, Pocasset, MA, USA). The loggers had a measurement range of +34 °C to +46 °C, a resolution of 0.04 °C and a mass of ~40 g (50 × 45 × 20 mm) when covered in an inert wax (Wax EXP986, SASOL, Johannesburg, South Africa). Before implantation the loggers were calibrated individually, in an insulated water bath, against a high-accuracy thermometer (Quat 100, Heraeus, Hanau, Germany); each logger had a calibrated accuracy of greater than 0.05 °C.

#### Experimental procedure

Experimental trials began 2–5 months after surgery. Trials were conducted using 3 groups of impala. For the 1st group ( $n = 4$ ) trials took place between February and

April (late southern hemisphere summer to early autumn) 2004, for the 2nd group ( $n = 6$ ) trials took place in February and March 2005 and for the 3rd group ( $n = 5$ ) trials took place between June and September (mid-winter to early spring) 2005. Throughout the trials, the impala were housed in 5 × 10 m bomas, with a maximum of 3 impala per boma; these bomas limited the amount of escape activity possible during and after darting. The impala received lucerne and water *ad libitum* and the bomas were cleaned regularly, so the impala were accustomed to occasional human presence. During the trials, each impala was darted by dart gun (Sabi 500, SABI Werkswinkel t/a Magnum Arms, Nelspruit, South Africa; Pneu-Dart dart type P, 3 ml volume, 25 mm long, wire-barbed needle, Pneu-Dart, Williamsport, United States of America) between 8:00 and 13:00 and each impala was darted on 4 occasions, fortnightly, in a random order. Each time a different cocktail was administered, namely: 1.5 mg etorphine hydrochloride (M99, Novartis, Johannesburg, South Africa) and 40 mg azaperone; 1.5 mg etorphine hydrochloride and 2 mg medetomidine hydrochloride (Domitor, Novartis, Johannesburg, South Africa); 1.2 mg thiafentanil oxalate (A3080, Wildlife Pharmaceuticals, Karino, South Africa) and 40 mg azaperone; and 1.2 mg thiafentanil oxalate and 2 mg medetomidine hydrochloride. The darts were fired into the gluteus muscles. The doses of the drugs, per unit body mass, were  $40.1 \pm 3.5 \mu\text{g/kg}$  (mean  $\pm$  SD) etorphine,  $32.1 \pm 2.8 \mu\text{g/kg}$  thiafentanil,  $1.07 \pm 0.09 \text{ mg/kg}$  azaperone and  $53.4 \pm 4.7 \mu\text{g/kg}$  medetomidine. Drug dosages were based on dosages recommended by experienced veterinarians working in the wild-life field<sup>1,3</sup>.

All impala in a boma were darted within 3 minutes. Time from dart impact to that at which the impala no longer could stand on its own was designated 'time to recumbency'. Once recumbent, each impala was moved to a non-shaded area in the boma, where it was held in a sternal position, with the head positioned so that the neck was aligned with the spinal column and the head was elevated above the thorax with the nose pointing downwards. This positioning allowed for unobstructed eructation of ruminal gas and maintained open upper airways. The impala were blindfolded and cotton wool was placed in their ears to reduce external sensory stimuli. Respiratory rate was measured by counting the number of breaths in a minute. To measure arterial haemoglobin oxygen saturation and heart rate, a veterinary pulse oximeter (Nonin 9847V with lingual sensor 2000SL,

Nonin Medical, North Plymouth, USA) was clipped to the wall of the vulva. According to the manufacturer's specifications, saturation was measured to an accuracy of 3 % and heart rate to an accuracy of 2 beats/min. The level of immobilisation was assessed clinically by observing movement and muscle tone.

Twenty-eight minutes after darting, blood samples were drawn from either a cephalic or a jugular vein, into a lithium heparin tube (BD Vacutainer Systems, Plymouth, UK). The samples were kept on ice until they could be centrifuged to separate the plasma. The plasma was stored at –70 °C until thawed for cortisol concentrations to be measured by radioimmunoassay (Coat-A-Count Cortisol Kit, Diagnostic Products, Los Angeles, USA). Thirty minutes after darting, the action of the immobilising drugs was reversed with 3 mg diprenorphine hydrochloride (M5050, intravenously (i.v.), Novartis, Johannesburg, South Africa) for etorphine, 12 mg naltrexone hydrochloride (Trexonil, i.v., Wildlife Pharmaceuticals, Karino, South Africa) for thiafentanil, and 10 mg atipamezole hydrochloride (Antisedan, i.m., Novartis, Johannesburg, South Africa) for medetomidine.

#### Climatic data

The microclimate in the bomas throughout darting and immobilisation was assessed by measuring dry-bulb (ambient air) and black globe temperatures 1 m above the ground at 2 min intervals by means of a Hobo data logger (H08-007-02, Onset Computer Corporation, Pocasset, MA, USA). Black globe temperature integrates the effects of ambient air temperature, solar radiation and wind speed, and provides the best single index of dry environmental heat load. Water vapour pressure was not measured, but it typically was very low (~1.8 kPa), at the site.

#### Data analysis

Results are reported as mean  $\pm$  SD, and  $P < 0.05$  was considered statistically significant. Changes in body temperature were calculated as the difference between maximum body temperature after darting and body temperature immediately before darting. The thermal response index was calculated as the time integral of the elevation of body temperature from body temperature before darting. Pearson product-moment correlations were used to compare relationships between all other relevant pairs of variables. Where appropriate, we correlated the responses of the 15 impala averaged over the 4 different drug combinations ( $n = 15$ ), or the average response of the 15 impala to each drug combination ( $n = 4$ ).

Repeated measures 1-way analysis of variance (ANOVA) followed by a Student Newman Keuls (SNK) *post hoc* test was used to test for differences between the different drug combinations in terms of times to recumbency, body temperature changes, thermal response indices, arterial haemoglobin oxygen saturations, respiratory rates and heart rates. A Kruskal-Wallis test was used to compare cortisol concentrations in response to each of the different drug combinations. A Chi-square test was used to test for an association between drug use and induction apnoea. Because of the length of time that it took for some of the impala to become recumbent with some drug combinations (up to 14 min), and because the immobilising drugs were reversed 30 min after darting, we analysed arterial haemoglobin oxygen saturation, respiratory rate and heart rate only over the 1st 16 min of the immobilisation from the time the animals became recumbent, divided into 0–8 min and 8–16 min. To determine the immediate effects of the drugs on arterial haemoglobin oxygen saturation post-darting, the data in the 1st 4 min of immobilisation (0–4 min) were analysed.

## RESULTS

### Body temperature and time to recumbency

Body temperature of the impala while they were housed in bomas was  $38.93 \pm 0.25$  °C ( $n = 15$ ), with a fluctuation of about 1.2 °C over 24 h. Body temperature followed a nycthemeral rhythm, with minimum temperatures soon after dawn and peak temperatures in the late afternoon to early evening. Whenever the impala were immobilised body temperature increased rapidly, continued to rise during immobilisation and reached a peak 20–30 min after the effects of the immobilising agents had been reversed (Fig. 1). After this peak, body temperature decreased slowly, but had not stabilised as long as 3 h after reversal of the immobilisation. The magnitude of the body temperature increase was not influenced significantly by the drug combination used to immobilise the impala. There were no significant differences between the mean body temperature changes of the impala following immobilisation with the drug combinations ( $F_{3,14} = 0.79$ ,  $P = 0.51$ ,  $n = 15$ ; Fig. 2A), nor in the thermal response indices calculated over the period of immobilisation ( $F_{3,14} = 0.62$ ,  $P = 0.60$ ,  $n = 15$ , data not shown). However, the thermal response indices calculated over 2, 3 and 4 h after darting were significantly lower when thiafentanil/azaperone was used as the immobilising agent,

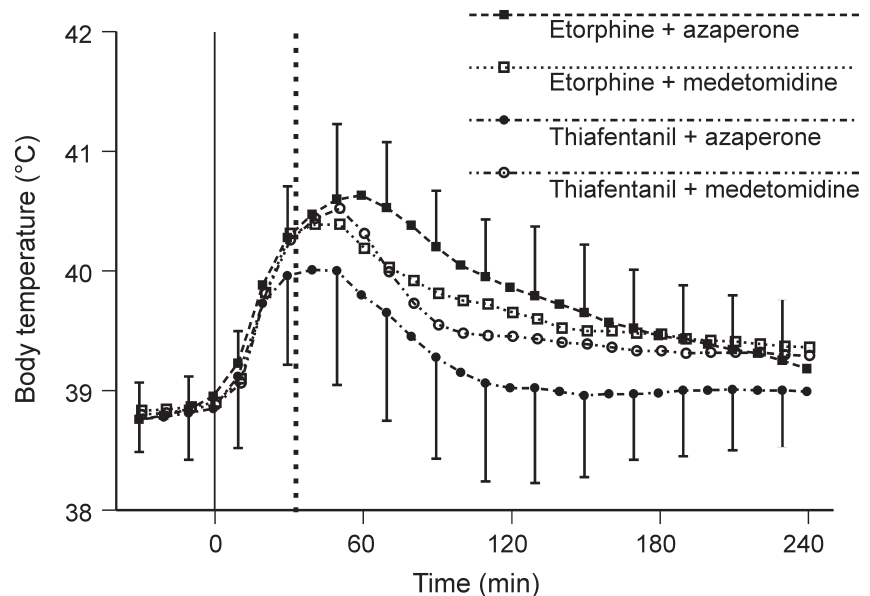


Fig. 1: **Body temperatures** (mean  $\pm$  SD,  $n = 15$ ), measured at 10-minute intervals, of impala before, during and after immobilisation with etorphine and azaperone, etorphine and medetomidine, thiafentanil and azaperone, and thiafentanil and medetomidine. The solid line at time 0 indicates the time at which the impala were darted, and the dashed line at 30 min the time at which the immobilising effects of the drugs were reversed, with diprenorphine used for etorphine, naltrexone for thiafentanil, and atipamezole for medetomidine.

than when etorphine/azaperone was used (2 h  $F_{3,14} = 3.11$ ,  $P = 0.03$ ; 3 h  $F_{3,14} = 3.62$ ,  $P = 0.02$ ; 4 h  $F_{3,14} = 3.35$ ,  $P = 0.03$ ; Fig. 2B, all  $n = 15$ ). The reduction in thermal response index associated with immobilisation with thiafentanil and azaperone occurred mainly because body temperatures recovered sooner, rather than because peak temperatures were significantly lower (Fig. 1).

The time for the impala to become recumbent also differed in response to each of the drug combinations ( $F_{3,14} = 10.29$ ,  $P < 0.0001$ ,  $n = 15$ ; Fig. 2C). When impala received thiafentanil they became recumbent more quickly than when they received etorphine, in both combinations (Fig. 2C). There was no significant correlation between time to recumbency and the magnitude of the body temperature changes ( $r^2 = 0.29$ ,  $P = 0.46$ ,  $n = 4$ ; also compare Fig. 2A to 2C), or between time to recumbency and the thermal response indices (30 min  $r^2 = 0.49$ ,  $P = 0.30$ , to 4 h  $r^2 = 0.63$ ,  $P = 0.21$ ,  $n = 4$ ; also compare Fig. 2B with Fig. 2C), across the different drug combinations. However, there was a positive linear relationship between the mean time to recumbency and mean body temperature change when the responses to the 4 drug combinations were averaged for the individual impala ( $r^2 = 0.45$ ,  $P = 0.006$ ,  $n = 15$ ; Fig. 3A). The mean time to recumbency for the 15 impala was also correlated with the mean thermal response indices over the 30 min ( $r^2 = 0.61$ ,  $P = 0.0006$ ,  $n = 15$ ) and the 4 h period ( $r^2 = 0.38$ ,  $P = 0.01$ ,  $n = 15$ ) after darting.

Because peak body temperatures occurred after the immobilising effects of the drugs had been reversed and because in the field it is impractical to measure body temperatures after the animals had recovered motility, body temperatures at 20 and 30 min after darting were investigated to predict peak body temperature. The body temperature at 30 min after darting ( $r^2 = 0.90$ ,  $P < 0.0001$ ; Fig. 4B) was a better predictor of peak body temperature than was the body temperature at 20 min ( $r^2 = 0.87$ ,  $P < 0.0001$ ; Fig. 4A). Body temperature at 30 min also weakly predicted the 4 h thermal response index ( $r = 0.56$ ,  $P = 0.03$ ), but body temperature at 20 min did not do so ( $P = 0.10$ ). Peak body temperature ( $y$ ) could be calculated from body temperatures ( $x$ ) measured during immobilisation using the following equations:

$$y = 1.06x - 1.6 \text{ (} Sy.x = 0.33 \text{ °C, body temperature at 20 min),}$$

and

$$y = 0.94x + 2.63 \text{ (} Sy.x = 0.19 \text{ °C, body temperature at 30 min).}$$

Because the slope of these equations did not differ significantly from 1 ( $F_{1,13} = 0.1$ ,  $P = 0.76$  body temperature at 20 min;  $F_{1,13} = 0.41$ ,  $P = 0.53$  body temperature at 30 min) these equations could be simplified by fitting a constrained regression line with a slope equal to 1:

$$y = x + 0.79 \text{ (} Sy.x = 0.32 \text{ °C, body temperature at 20 min),}$$

and

$$y = x + 0.38 \text{ (} Sy.x = 0.19 \text{ °C, body temperature at 30 min).}$$

Therefore, peak body temperature



could be estimated, by extrapolation, by adding 0.79 °C to body temperature as measured at 20 min, or, better, by adding 0.38 °C to that measured at 30 min.

Although trials were carried out over a range of environmental conditions (dry bulb: minimum = 18.2 °C, mean = 30.1 ± 5.1 °C, maximum = 40.3 °C; black globe: minimum = 24.5 °C, mean = 37.4 ± 5.6 °C, maximum = 47.2 °C), these conditions did not influence the magnitude of the hyperthermia that the impala developed. There was no correlation between the change in the body temperature or the thermal response index and the environmental conditions that prevailed in the bomas at the time of darting and immobilisation, irrespective of which drug combination was used (change in body temperature *vs* dry bulb temperature  $r^2 = 0.03$ ,  $P = 0.58$ ,  $n = 15$ , Fig. 5A; 4 h thermal response index *vs* dry bulb temperature  $r^2 = 0.01$ ,  $P = 0.71$ ,  $n = 15$ , Fig. 5B; change in body temperature *vs* black globe temperature  $r^2 = 0.02$ ,  $P = 0.65$ ,  $n = 15$ , data not shown; 4 h thermal response index *vs* black globe temperature  $r^2 = 0.004$ ,  $P = 0.82$ ,  $n = 15$ , data not shown).

#### Plasma cortisol

The mean time to recumbency for the 15 impala was correlated to mean plasma cortisol concentration ( $r^2 = 0.29$ ,  $P = 0.008$ ,  $n = 15$ ; Fig. 3B). The mean plasma cortisol concentration also was correlated to mean body temperature changes ( $r^2 = 0.31$ ,  $P = 0.008$ ,  $n = 15$ ; data not shown) in individual animals. The plasma cortisol concentration, however, did not differ significantly in the impala when they received each of the drug combinations ( $P = 0.92$ ,  $n = 4$ ).

#### Respiration

There was no significant effect of the different drugs on respiratory rates of the impala over the entire immobilisation period (Fig. 6A). However, induction apnoea (cessation of breathing at the beginning of the immobilisation) occurred in 33 % of the impala that were immobilised with thiafentanil and azaperone, 13 % that were immobilised with thiafentanil and medetomidine, and 7 % that were immobilised with etorphine and medetomidine. It did not occur with etorphine and azaperone. The risk of induction apnoea was significantly different ( $\chi^2 = 8.07$ ,  $P = 0.04$ ) between the 4 combinations of immobilising drugs.

When impala were immobilised with thiafentanil and azaperone, their haemoglobin oxygen saturation was significantly lower in the 1st 8 min of immobilisation than when the other drug combinations

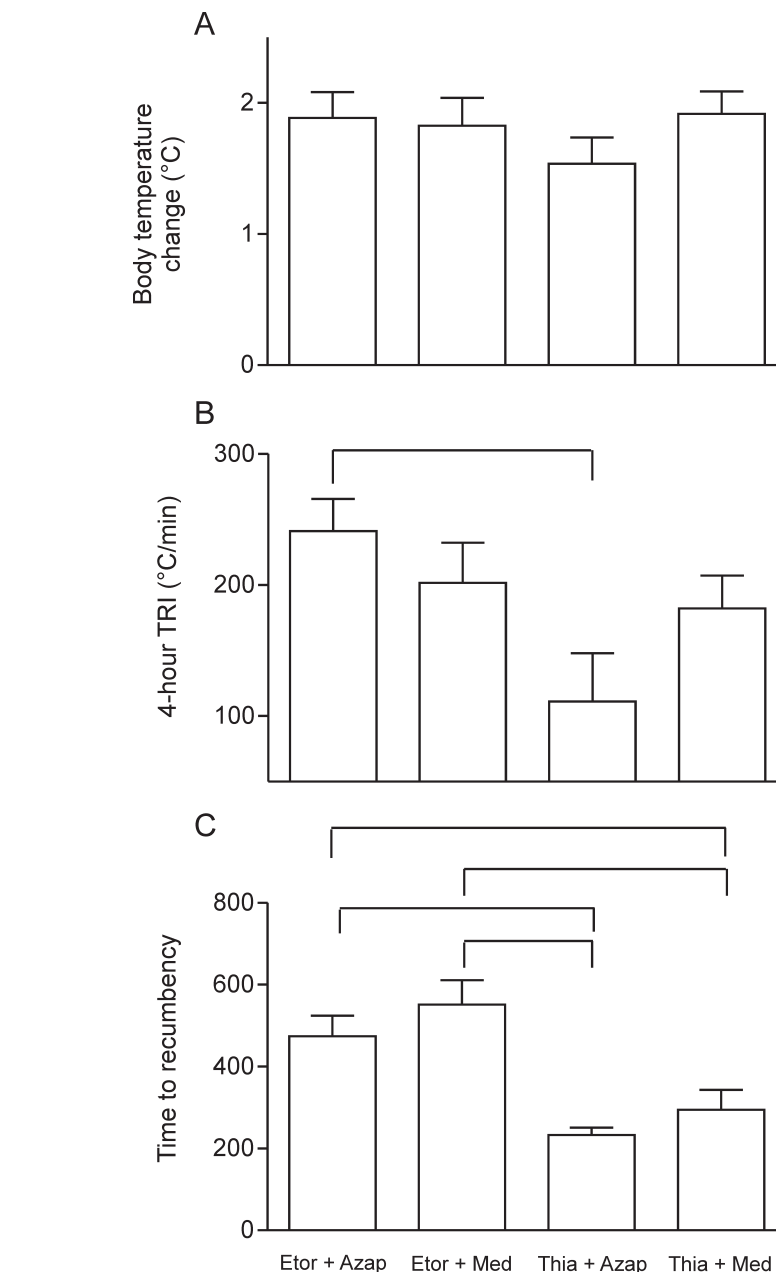


Fig. 2: **Change in body temperature (A), 4-hour thermal response index (TRI – the time integral of the elevation of body temperature from body temperature before darting) (B), and time to recumbency after darting (C) in impala (mean ± SD,  $n = 15$ ) that were immobilised with etorphine and azaperone (Etor+Azap), etorphine and medetomidine (Etor+Med), thiafentanil and azaperone (Thia+Azap), and thiafentanil and medetomidine (Thia+Med). Bars above the graphs indicate significant differences between the drug combinations (1-way ANOVA with *post hoc* SNK test,  $P < 0.05$ ).**

were used ( $F_{3,14} = 3.74$ ,  $P < 0.05$ ; Fig. 6B), but it then increased, and became significantly greater, over a period of 8–16 min than when the impala were immobilised with etorphine and azaperone ( $F_{3,14} = 2.90$ ,  $P < 0.05$ ; Fig. 6B). There was no significant difference in the haemoglobin oxygen saturations of the impala given the other drug combinations; the haemoglobin oxygen saturations remained fairly constant throughout the immobilisation period. When the responses of individual impala were analysed, haemoglobin oxygen saturation was lower in impala that had shorter times to recumbency; there was a significant linear correlation

between mean haemoglobin oxygen saturation and mean time to recumbency both for the 1st 4 min of immobilisation ( $r^2 = 0.49$ ,  $P = 0.004$ ,  $n = 15$ ; Fig. 3C) and throughout immobilisation ( $r^2 = 0.58$ ,  $P < 0.001$ ,  $n = 15$ ; data not shown). Thus impala that had shorter times to recumbency had smaller increases in body temperature and plasma cortisol concentrations, but lower haemoglobin oxygen saturations, than those with longer times to recumbency (Fig. 3).

#### Heart rate

Irrespective of which drug combination was used to immobilise the impala, their

heart rates were highest at the beginning of the immobilisation and then decreased over the period of the immobilisation (Fig. 6C). When medetomidine was combined with the opiates, the impala had lower heart rates in the 1st 8min of immobilisation than when azaperone was combined with the opiates ( $F_{3,14} = 8.81, P < 0.001$ ; Fig. 6C). For the duration of the immobilisation the impala's heart rates were significantly lower when etorphine and medetomidine, and thiafentanil and medetomidine, were used than with etorphine and azaperone ( $F_{3,14} = 11.53, P < 0.05$ ; Fig. 6C).

#### Clinical assessment of immobilisation

After becoming recumbent, most of the impala remained immobile, with infrequent and minor movements like chewing (bruxism), ear twitching and tail flicking. However, the impala that received thiafentanil and azaperone moved their legs and bodies from 8 min after becoming recumbent, and some attempted to stand. A few of the impala that received the other drug combinations also tried to move after 8 min but their movements were smaller and attempts to stand were weak.

#### DISCUSSION

When impala were darted and chemically immobilised for 30 min with 4 combinations of an opiate and a tranquiliser or sedative, they developed a high rise in body temperature that peaked almost an hour after darting (20–30 min after reversal of immobilisation) and gradually declined, in some cases, over more than 3 h after reversal of immobilisation. The rise in body temperature, which could be as much as 3 °C, varied greatly between individual animals and was not correlated to the prevailing environmental heat load, even though air temperature sometimes exceeded 40 °C. The magnitude of the rise also was not determined by the choice of darting drugs. The main factor contributing to the rise in body temperature and body temperature changes over time appeared to be the psychological 'fright stress' that each individual animal experienced during capture, which was directly related to how long the animal was conscious (*i.e.* by the time from darting to recumbency). Irrespective of the drug combination used, some animals became recumbent in a shorter time than did other animals, and those animals had smaller changes in body temperature and plasma cortisol concentrations than did the animals that took longer to become recumbent (Fig. 3A and Fig. 3B). The thiafentanil drug combinations tended to induce recumbency in shorter times than

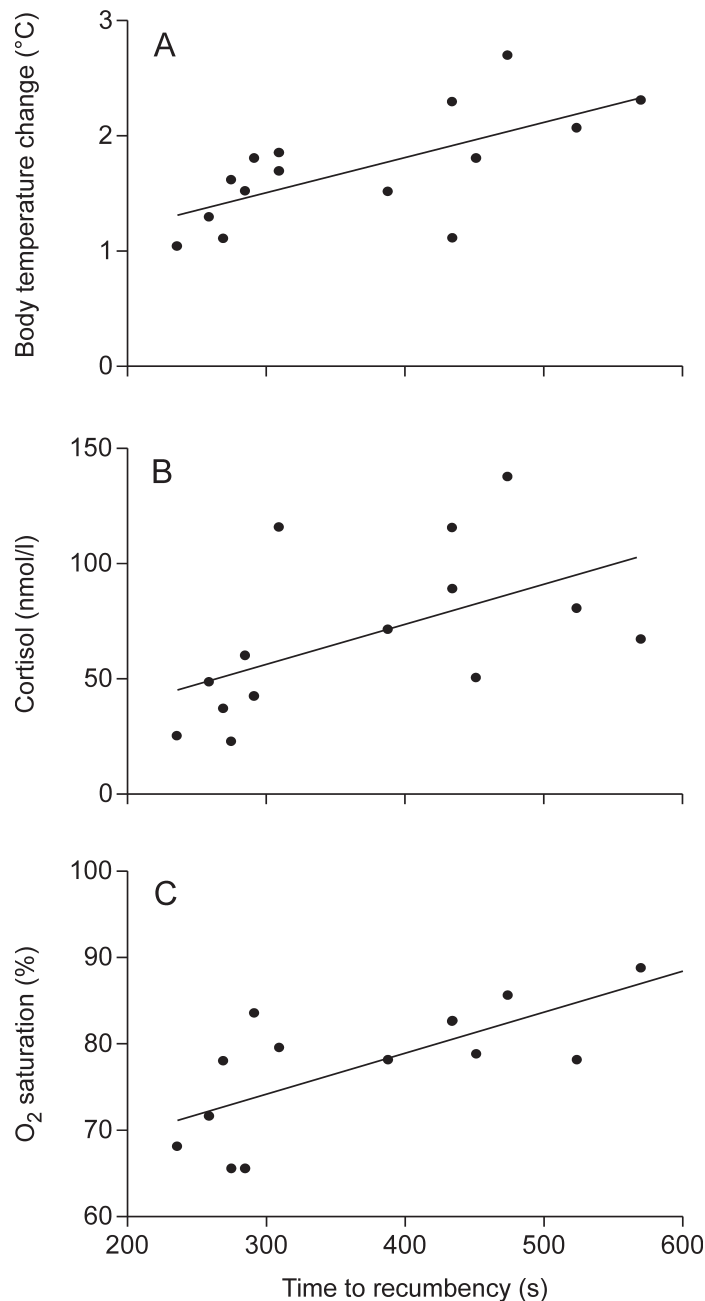


Fig. 3: Change in body temperature vs time to recumbency (A) ( $r^2 = 0.45, P = 0.006, n = 15$ ), plasma cortisol concentration vs time to recumbency (B) ( $r^2 = 0.29, P = 0.008, n = 15$ ) and haemoglobin oxygen saturation over the 1st 4 minutes of immobilisation vs time to recumbency (C) ( $r^2 = 0.49, P = 0.004, n = 15$ ), of impala in response to darting and chemical immobilisation with etorphine and azaperone, etorphine and medetomidine, thiafentanil and azaperone, and thiafentanil and medetomidine. Responses to the 4 drug combinations were averaged for each impala.

did the etorphine drug combinations (Fig. 2C), but there was a large variability in the times to recumbency between individual animals (Fig. 3). Some impala always became recumbent in about 5 min, regardless of the darting drugs used, while others took twice as long. Because of that variability, there was no statistically smaller mean rise in body temperature in the impala when they received the thiafentanil combinations (Fig. 2A).

Changes in body temperature therefore appear to have been influenced mainly by an individual animal's stress response to

capture, rather than by the pharmacological effects of the immobilising drugs themselves (see Fig. 2A). The shorter the time to recumbency, the less was the hyperthermia, irrespective of how that short time to recumbency was achieved (Fig. 3A). The variable susceptibility of individual impala to the immobilising drugs was demonstrated further by the respiratory depressant effects of the drugs. Those animals that appeared to be more sensitive to the immobilising effects of the drugs, in that they became recumbent in a shorter time and developed smaller rises in body temperature

(Fig. 3A) than those that were less sensitive, also had lower percentage oxygen haemoglobin saturations (Fig. 3C). These animals therefore were more sensitive to the effects of immobilising drugs on the respiratory system. Consequently, a risk of decreasing the time to recumbency is that the animals may develop severe respiratory depression and hypoxia.

Deciding which drug combination causes the least disturbance to the overall physiological function of impala therefore requires balancing the thermal benefits of short time to recumbency with its respiratory risks. Impala that received thiafentanil and azaperone had the smallest duration and extent of hyperthermia. However, the immobilising effects of thiafentanil and azaperone were short-lived and the impala experienced initial severe hypoxia (Fig. 6B), with a high incidence of induction apnoea (33 %). The other drug combinations also caused moderate hypoxia, but this hypoxia was not as severe as that caused by thiafentanil and azaperone. The etorphine drug combinations were less likely to cause induction apnoea than were combinations with thiafentanil, but with these combinations the animals took longer to become immobile (Fig. 2C), and etorphine and azaperone caused prolonged hyperthermia (Figs 1 and 2B). Although thiafentanil and medetomidine did not significantly reduce the magnitude or the duration of the capture-induced hyperthermia below that seen with etorphine drugs combinations, that combination immobilised the impala more quickly and induced only moderate hypoxia (Fig. 6B). Therefore, based on our choice of drugs, doses and capture method, we advocate thiafentanil and medetomidine as the most suitable combination with which to immobilise impala chemically, but caution that the risk of capture-induced hyperthermia, hypoxia and, in some animals, induction apnoea will not be eliminated.

It is possible that different doses of the drugs may have improved the clinical outcome of individual animals, but because the impala reacted to the drugs in a highly variable manner, a drug dose that would improve the clinical outcome in some individuals may well have worsened the outcome in others. It also might have been useful to test the effects of drugs on thermoregulation under controlled conditions, for example in domesticated animals in a laboratory. However, the primary aim of the study was to test the thermal consequences of using different drugs under capture conditions, rather than to systematically investigate the effects of the drugs on thermoregulation.

Opioids and adrenergic ligands have the potential to cause thermal lability and these effects may have become more apparent with longer immobilisation. Although there was no association between environmental conditions and body temperature change, the experimental animals were not exposed to prolonged immobilisation or to severe heat or cold. Impala are known to have a fractious nature and therefore their responses to stress also may be greater than those of other wild ungulate species. However, it is possible that the primary cause of hyperthermia in other wild ungulate species would be the same as that which we found in impala, namely fright stress.

There have been suggestions that capture-induced hyperthermia results partly from the effects of the capture drugs on thermoregulatory processes. Meltzer and Kock<sup>21</sup> proposed that the adrenergic effects of opiate drugs increase metabolic rate and therefore heat production during chemical capture. Although adrenergic effects of opiate drugs have been demonstrated in rats given etorphine<sup>30</sup>, these effects have not been linked to body temperature changes.

Geller and colleagues<sup>10</sup> showed that etorphine caused a dose-related dual temperature response in rats; low doses caused hyperthermia, medium doses had no effect and high doses caused hypothermia. Rosow and colleagues<sup>31</sup> showed a similar response in mice. They also showed that this response could be altered by changing environmental temperatures<sup>31</sup>, implying that the underlying malfunction is thermal lability. The varying effects of opiates on body temperatures are not limited to rats and mice but also occur in other mammalian species, including ungulates<sup>5</sup>, and appear to be related to the complex interaction of opiates on opioid receptors both in the central and peripheral nervous system<sup>10,31</sup>. While these effects are not disputed, they are probably not significant during capture with short-term immobilisation.

Like the effects of opiates on body temperatures during chemical capture, the effects of tranquilisers and sedatives on body temperature are not clear. Tranquilisers may affect thermoregulation through several mechanisms including anti-adrenergic effects, inhibition of sweating, and central dopamine antagonism

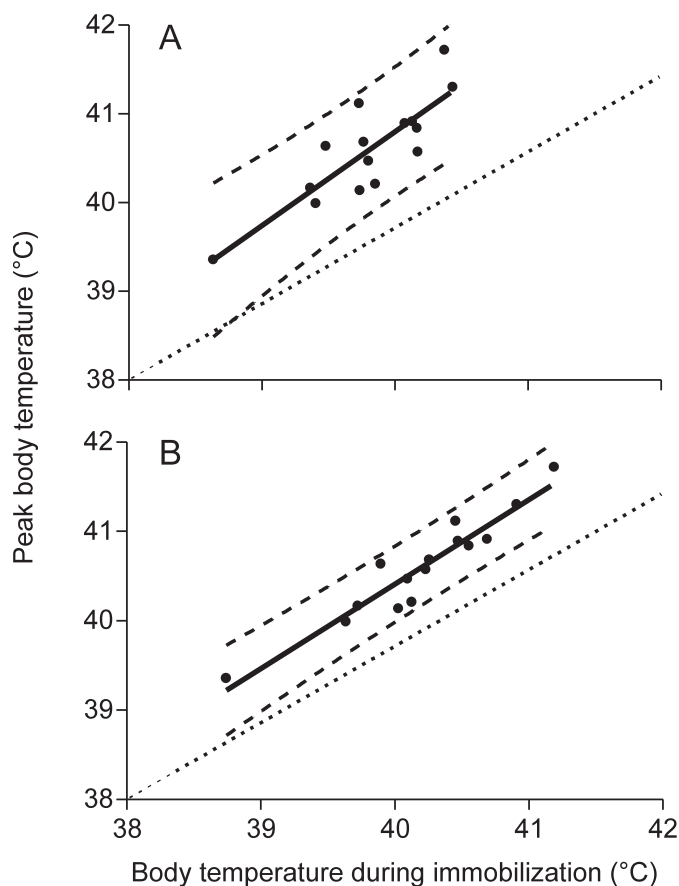


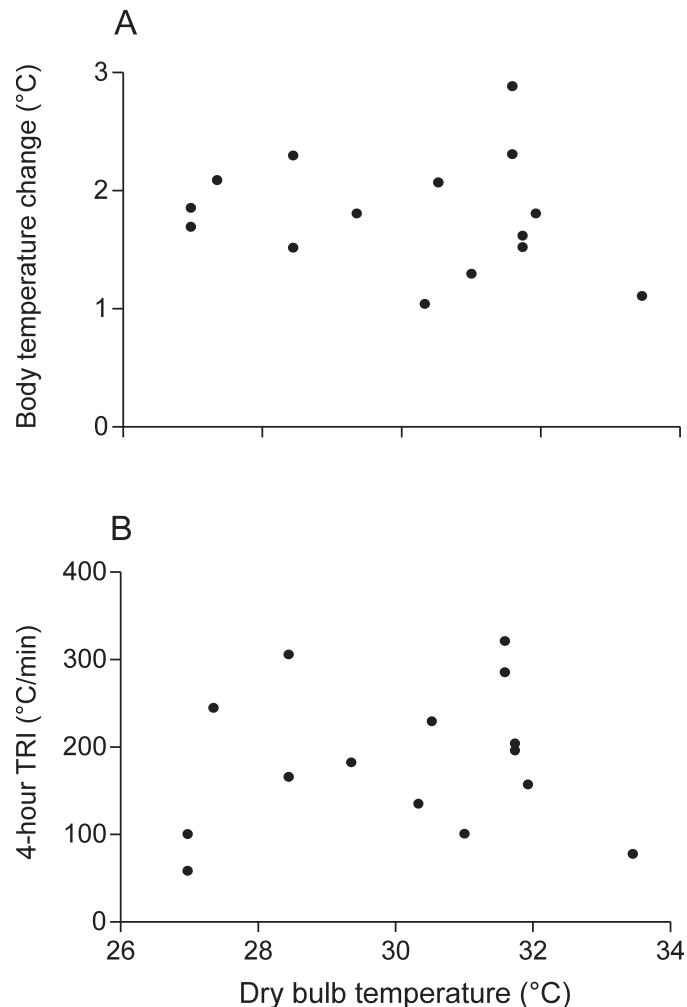
Fig. 4: Peak body temperature after darting vs body temperature at 20 minutes after darting (A) and 30 minutes after darting (B), in 15 impala in response to chemical immobilisation with etorphine and azaperone, etorphine and medetomidine, thiafentanil and azaperone, and thiafentanil and medetomidine. Responses to the 4 drug combinations were averaged for each impala. The dotted line is the line of identity ( $y = x$ ), the dashed lines are the 95 % confidence band and the solid lines are the regression lines (A,  $r^2 = 0.90$ ,  $P < 0.0001$ ; B,  $r^2 = 0.87$ ,  $P < 0.0001$ ).

that disrupts the thermoregulatory set-point causing thermal lability<sup>8</sup>. The tranquiliser we used, azaperone, has potent anti-dopaminergic effects and causes vasodilation through minor  $\alpha_1$ -antagonistic effects<sup>20,32</sup>. It would have been expected that both of these effects would have resulted in the impala becoming thermally labile and that their body temperatures, during and after immobilisation, could have been influenced strongly by environmental heat load. Similarly, environmental heat load may have been expected to influence the impalas' body temperatures when medetomidine was used, because central noradrenergic mechanisms are involved in the control of body temperature<sup>7</sup> and  $\alpha_2$ -agonists induce thermal lability<sup>14,19</sup>. Even if azaperone and medetomidine did not cause the animals to become thermally labile, some effect on the change in body temperatures would be expected because these drugs have opposite effects on peripheral blood vessel diameters<sup>9,20</sup>, and hence blood flows and heat exchange to and from the environment. However, in the impala, the effects of environmental heat load on body temperature were not statistically significant, even over a wide range of environmental conditions. Therefore any effects of environment on body temperature during capture were insignificant, compared with the effects of fright stress.

The psychological processes in fright stress that leads to hyperthermia are not well defined. One factor could be the effects of muscular activity of the animals from darting to immobilisation. To exclude the influence of activity on the body temperatures in this study, the impala were darted in small (5 × 10 m) bomas and they displayed only low-level activity before becoming recumbent. The factor most likely to have induced excessive body temperature rises in the impala was the stress response to capture. Psychological stress is known to induce hyperthermia which causes an acute rise in body temperature similar to that seen in the impala<sup>24,25,28,29</sup>. The precise mechanisms underlying stress-induced hyperthermia are not known, but the hyperthermia does not arise solely as the result of the metabolic and vascular effects of catecholamine release<sup>23,27,28</sup> and rather appears to be centrally regulated in response to a psychological stimulus<sup>28</sup>.

It is well known that the behaviour of animals in a group is not uniform and there are distinct differences in stress responses between individual animals when they are placed in captivity<sup>18</sup>. The magnitude of the rise in body temperature that occurred in our impala was

**Fig. 5: Change in body temperature vs dry bulb temperature (A) ( $r^2 = 0.03$ ,  $P = 0.58$ ,  $n = 15$ ) and 4-hour thermal response index (TRI – the time integral of the elevation of body temperature from body temperature before darting) vs dry bulb temperature (B) ( $r^2 = 0.01$ ,  $P = 0.71$ ,  $n = 15$ ) of impala in response to darting and chemical immobilisation with etorphine and azaperone, etorphine and medetomidine, thiafentanil and azaperone, and thiafentanil and medetomidine. Responses to the 4 drug combinations were averaged for each impala.**



related strongly to an individual animal's response to capture. Even though all the animals received similar drug doses per kilogram body mass, some individuals were affected more by the effects of the drugs than were others; some individuals always became recumbent quickly and typically experienced hypoxia. Therefore the variable thermal responses in our animals can most likely be explained by the inter-individual differences in behaviour and stress responses to chemical capture. An animal's susceptibility to the drugs may be related to how 'tame' (not afraid of humans) or how 'wild' (afraid of humans) an animal is. 'Tame' animals generally become recumbent sooner, are more affected by the side-effects of immobilising drugs<sup>20</sup>, show a smaller response to stress<sup>26</sup> and have smaller changes in body temperature compared to 'wild' animals<sup>22</sup>.

In summary, the acute body temperature elevation that occurs during chemical capture of impala does not appear to be determined primarily by the pharmaco-

logical effects of the capture drugs, but rather is related to the animal's psychological stress response to capture. This hyperthermia peaks only after the reversal of the immobilisation and also resolves only hours later. We suggest that limiting the magnitude and duration of this hyperthermia requires minimising the time from first encounter until the animal becomes recumbent. When animals are not confined to a small boma before capture, steps also should be taken to limit additional heat production by decreasing the length, duration and intensity of pre-capture exercise. Restricting capture to cool months of the year, or cool times of the day, will not protect animals from capture-induced hyperthermia; its magnitude is independent of environmental conditions. If animals are severely hyperthermic and especially if they are hyperthermic and hypoxic, steps should be taken to decrease the hyperthermia and ensure that adequate tissue oxygenation takes place. Although our study was not designed to produce the supportive



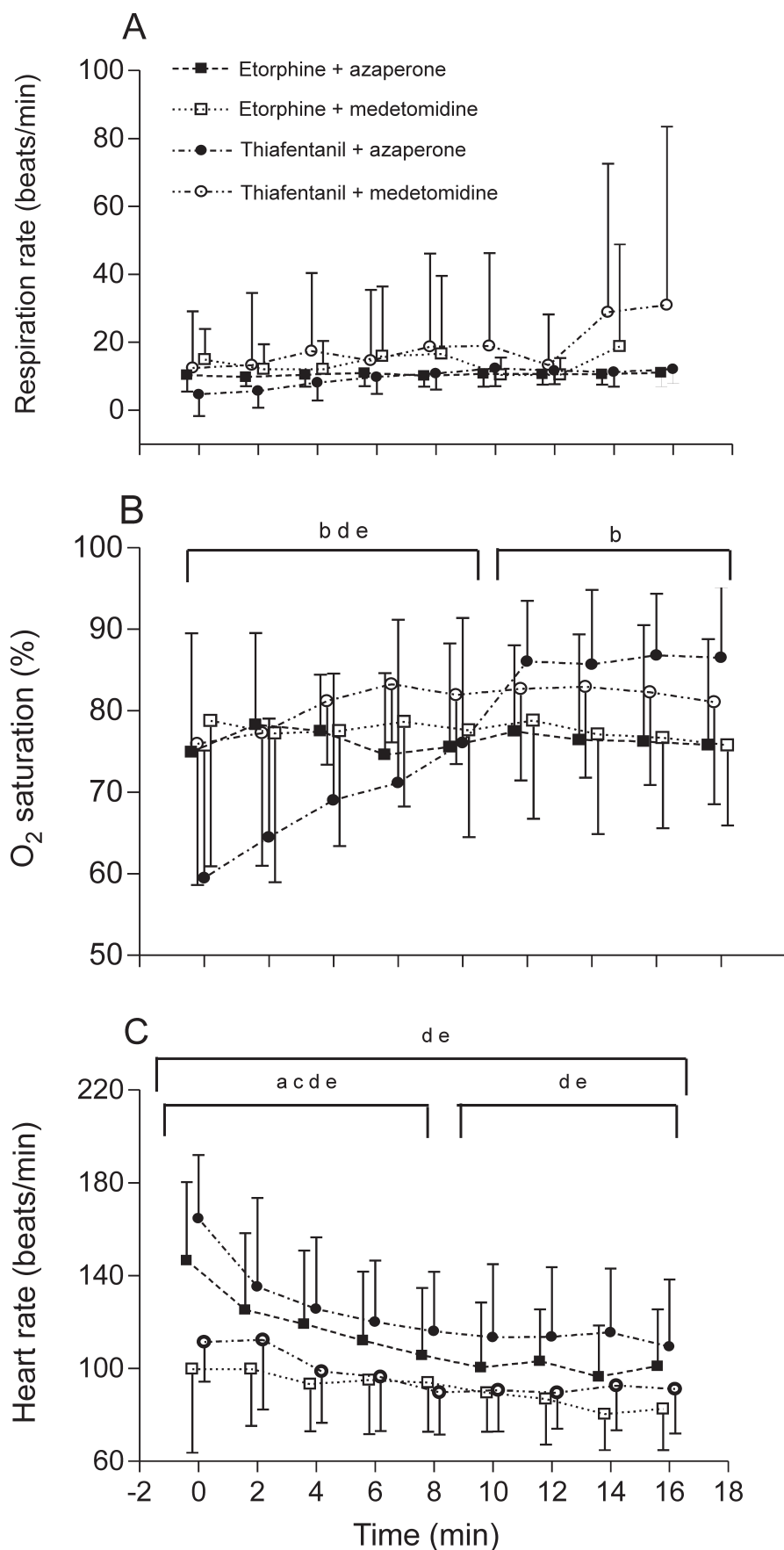


Fig. 6: Respiratory rate (A), haemoglobin oxygen saturation (B) and heart rate (C) in impala (mean  $\pm$  SD,  $n = 15$ ) from time to recumbency (time 0). a,  $P < 0.05$  etorphine and azaperone vs etorphine and medetomidine; b,  $P < 0.05$  etorphine and azaperone vs thiafentanil and azaperone; c,  $P < 0.05$  etorphine and azaperone vs thiafentanil and medetomidine; d,  $P < 0.05$  etorphine and medetomidine vs thiafentanil and azaperone; e,  $P < 0.05$  thiafentanil and medetomidine vs thiafentanil and azaperone, 1-way ANOVA with *post hoc* SNK test on means over the periods 0–8 min, 8–16 min and 0–16 min.

evidence, we believe that, once animals are immobilised, they should be protected from exposure to high or low temperatures. In conclusion, limiting the thermal consequences of capture requires limiting the exposure of the animal to fright stress, and selecting chemical agents that cause rapid recumbency in all individuals without inducing unmanageable respiratory depression and hypoxia.

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