

Immobilisation of impala (*Aepyceros melampus*) with a ketamine hydrochloride/medetomidine hydrochloride combination, and reversal with atipamezole hydrochloride

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ABSTRACT

A combination of medetomidine hydrochloride (medetomidine) and ketamine hydrochloride (ketamine) was evaluated in 16 boma-confined and 19 free-ranging impalas (*Aepyceros melampus*) to develop a non-opiate immobilisation protocol. In free-ranging impala a dose of $220 \pm 34 \mu\text{g}/\text{kg}$ medetomidine and $4.4 \pm 0.7 \text{ mg}/\text{kg}$ ketamine combined with 7500 IU of hyaluronidase induced recumbency within 4.5 ± 1.5 min, with good muscle relaxation, a stable heart rate and blood pH. PaCO₂ was maintained within acceptable ranges. The animals were hypoxic with reduced oxygen saturation and low PaO₂ in the presence of an elevated respiration rate, therefore methods for respiratory support are indicated. The depth of sedation was adequate for minor manipulations but additional anaesthesia is indicated for painful manipulations. Immobilisation was reversed by $467 \pm 108 \mu\text{g}/\text{kg}$ atipamezole hydrochloride (atipamezole) intramuscularly, but re-sedation was observed several hours later, possibly due to a low atipamezole:medetomidine ratio of 2:1. Therefore, this immobilisation and reversal protocol would subject impalas to possible predation or conspecific aggression following reversal if they were released into the wild. If the protocol is used on free-ranging impala, an atipamezole:medetomidine ratio of 5:1 should probably be used to prevent re-sedation.

Key words: α^2 agonist, α^2 antagonist, *Aepyceros melampus*, atipamezole hydrochloride, blood gases, impala, hyaluronidase, ketamine hydrochloride, medetomidine hydrochloride, oxygen saturation, piloerection, re-sedation.

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INTRODUCTION

The impala is a sentinel species for disease monitoring in South Africa (especially foot-and-mouth disease) and it is also important in game ranching. In a captive setting the sleek and agile impala is an attractive addition to an African hoof-stock collection, but its excitable character presents management challenges. The impala is a moderately sized antelope lending itself to physical restraint once confined, but physical restraint induces physiological and behavioural alterations indicative of stress and can induce a mortality rate of 10–40%^{10,11,12}. The induced stress of physical restraint is detri-

mental to animals, can attenuate physiological sampling results^{5,10} and raises animal welfare concerns. Historically, chemical immobilisation of impala was associated with various difficulties, including mortality and morbidity (JPR, pers. obs.). Therefore, management of this species would be facilitated by a rapid, safe, reversible and reliable chemical immobilisation protocol.

Rapid onset of immobilisation is important in free-ranging impala, since they usually inhabit fairly dense bush and once darted, will run off and can be lost prior to the onset of immobilisation. Various protocols for chemical immobilisation have been reported using opioids combined either with xylazine hydrochloride (xylazine) or a tranquilliser¹⁴. In a captive situation xylazine alone is not effective in excited or free-ranging animals due to prolonged induction times of 9–13 min⁸. Etorphine or carfentanil protocols to immobilise impala present several complications, including regurgitation,

poor muscle relaxation, respiratory depression, hyperthermia, and relatively long induction times with elevations of cortisol⁵. More rapid and smoother inductions were accomplished when xylazine was added to either etorphine⁷ or carfentanil⁵, but these combinations are potentially dangerous to humans and, in addition, opioids are subject to regulations associated with registered narcotics.

Combinations of a specific α^2 agonist, medetomidine hydrochloride (medetomidine) [Medetomidine (20 mg/ml) Wildlife Pharmaceuticals, Karino, South Africa), and the dissociative anaesthetic, ketamine hydrochloride (ketamine) [Ketamine hydrochloride (200 mg/ml) Wildlife Pharmaceuticals, Karino, South Africa], with or without hyaluronidase (hyaluronidase, Sigma Chemical Co, St Louis) have been successfully used to immobilise various species⁹, including giraffe³. Ketamine and medetomidine have an apparent synergistic action. Ketamine also enhances sedation and analgesia and minimises medetomidine-induced bradycardia. The purpose of this study was to investigate a combination of medetomidine and ketamine with hyaluronidase as a non-narcotic drug combination for the rapid and safe immobilisation of free-ranging impala, and reversal by a specific α^2 antagonist, atipamezole hydrochloride (atipamezole) [Antisedan[®], (5.0 mg/ml) Orion Corp., Orion-Farmos, Espoo, Finland].

MATERIALS AND METHODS

The study was conducted in the Kruger National Park, South Africa, and consisted of 3 related trials. Trial 1 included 16 boma-confined subadult impala males (6–18 months old). The dosage ranges and immobilisation times are listed in Table 1 and reflects a range of dosages tested to determine the effectiveness and safety of this drug combinations in a controlled boma situation prior to field use. The boma was divided into 4 sections, each measuring 15 × 20 metres, and the interconnecting gates were open, allowing access to the 4 areas. The animals were fed

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Table 1: Dosage and results from Trial 1 boma-confined, subadult male impala ($n = 16$).

Parameters	Mean \pm SD	Range
Mass (kg)	33.3 \pm 7.8	20–48
Medetomidine ($\mu\text{g}/\text{kg}$)	237 \pm 68	167–400
Ketamine (mg/kg)	6.0 \pm 2.0	1.7–8.3
Initial signs (min)	4.3 \pm 2.0	2–9
Recumbent (min)	7.8 \pm 3.5	3–14
Handle (min)	16.1 \pm 4.4	9–24
Atipamezole ($\mu\text{g}/\text{kg}$)	405 \pm 71	200–588
Standing (min)	5.2 \pm 2.7	1–10

fresh leaves, branches and lucerne hay and given continual access to water. All animals appeared clinically normal and had normal muscle mass and body condition for their size, age and time of year. The animals were darted by a CO_2 -powered remote injection device delivering a 3-ml plastic, air-pressurised dart with a 40×2 mm collared needle (Dan-Inject Skukuza, Kruger National Park, South Africa) to insure a deep intramuscular (i.m.) injection. All darts were injected in the muscle mass of the hind limb. The following times were recorded; 1) 1st signs of drug effect, 2) recumbency, 3) time to manipulation after the animal became recumbent (lag time).

In a preliminary trial with boma-confined young males, using a combination medetomidine/ketamine, when impala were approached just after they became recumbent, most responded and attempted to flee. Once up they were extremely difficult to capture. We therefore instituted a waiting period from recumbency to handling. To facilitate sedation and minimise arousal, impalas were left undisturbed for an average of 12.1 ± 3.5 min (mean \pm standard deviation) following recumbency. Once handled, each impala was blindfolded, ear-plugs inserted, intubated with a cuffed endotracheal tube² and weighed, prior to collection of physiological data. Each animal was maintained in sternal recumbency for the duration of the monitoring period with the head positioned above the rumen and the nose angled downward to facilitate fluid drainage.

Physiological parameters (respiration and heart rate, pulse oximetry (Nellcor 200, Nellcor Inc., Haywood, CA, USA) and indirect systolic blood pressure) were measured every 5 min for 30 min. Each impala's responsiveness to tactile stimulation of the ears, palpebral reflex, response to painful stimuli (needle prick at the coronary band) and muscle tone was evaluated. Rectal temperature was measured initially and at the end of the monitoring period. Indirect systolic blood pressure was measured using a blood

pressure cuff, of the appropriate diameter, placed above the carpus and a standard sphygmomanometer. The cuff was inflated to 200 mm Hg and the pressure reduced slowly to the point where the sphygmomanometer needle deflections were synchronised with the heart rate and this pressure was read as the systolic pressure. The pulse oximetry sensor was placed on the tongue or a shaved portion of the ear.

Arterial blood gas samples from the auricular artery were collected in heparinised syringes. Time 0 was the collection of the 1st set of physiological data, including the blood gas sample once the animal was handled. The samples were sealed with a rubber stopper over the needle, placed on ice and assayed within 3 h of collection (1302 pH/Blood Gas Analyzer, Instrumentation Lab. Systems, Pretoria, South Africa).

Following 30 min of data collection the medetomidine was antagonised with atipamezole administered either $\frac{1}{4}$ intravenously (i.v.) and $\frac{3}{4}$ i.m. ($n = 3$) or i.m. at 2 sites ($n = 16$). The animal's recovery was monitored up to 6 h (Table 1). At the completion of the boma trial the animals were released into the wild.

Trial 2 evaluated the effect of ketamine

or medetomidine alone on boma-confined subadult males. Four impala received ketamine (average dose = 8.1 ± 0.3 mg/kg), either by hand-injection following physical capture and restraint ($n = 2$) or plastic dart as in Trial 1 ($n = 2$). Twelve impala were divided into 3 groups of 4 and received either 200, 300 or 400 $\mu\text{g}/\text{kg}$ medetomidine. In each medetomidine group, 2 animals were given medetomidine by hand syringe and two by plastic dart. The animals were observed as in Trial 1. All but one impala immobilised with medetomidine were given atipamezole i.m. following observation, and their recovery monitored.

Trial 3 was conducted on 19 free-ranging adult and subadult male and female impala, darted from a vehicle with the same delivery system used in Trial 1. Hyaluronidase (7500 units) (Hyaluronidase, Sigma Chemical Co, St Louis) was added to all darts in this trial. The impala in Trial 3 were observed and monitored as in Trial 1. Following monitoring, the impala were moved to the bomas described in Trial 1 where the medetomidine was antagonised with atipamezole i.m. and the impala were observed as before.

RESULTS

There was no mortality or morbidity associated with the immobilisation procedures in the 3 trials. The average dose and dosage ranges for medetomidine and ketamine, the onset of drug effect, time to recumbency, and lag time to manipulating the animal in Trial 1 are listed in Table 1.

In Trial 1 the shortening of the time to 1st signs and recumbency were directly related to the increasing dose of medetomidine with dosages above 200 $\mu\text{g}/\text{kg}$ (Fig. 1).

There was no dose-dependent shorten-

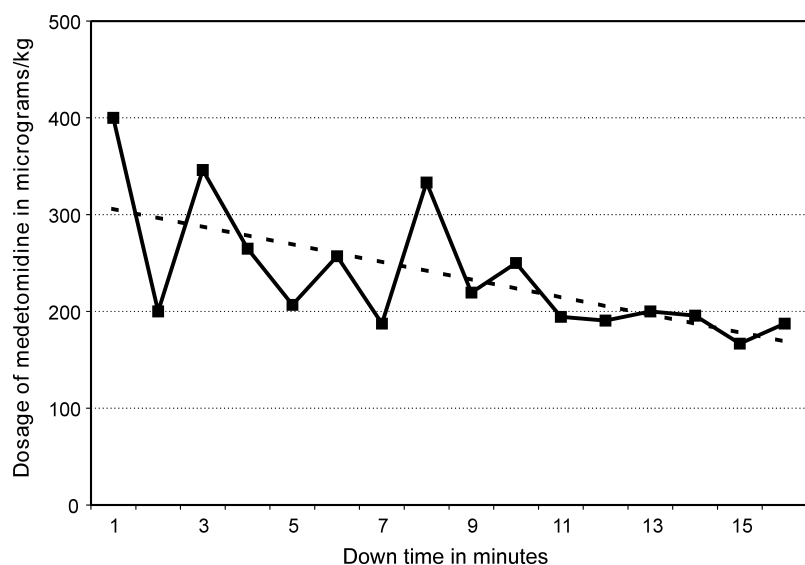


Fig. 1: Dosage of medetomidine versus down time in Trial 1 (with a dotted trendline).

Table 2: The physiological data for impalas in Trial 1 ($n = 16$).

Parameters	Time 0	5 min	10 min	15 min	20 min	25 min	30 min
Respiration rate (min)	39 ± 19	48 ± 26	43 ± 19	45 ± 17	48 ± 17	46 ± 17	46 ± 16
Heart rate (min)	47 ± 9	49 ± 7	48 ± 7	48 ± 6	49 ± 7	50 ± 8	49 ± 6
O ₂ saturation (%)	86 ± 7	86 ± 8	85 ± 14	87 ± 7	87 ± 6	89 ± 6	89 ± 6
Systolic blood pressure (mm Hg)	156 ± 15	158 ± 15	157 ± 16	161 ± 20	158 ± 18	155 ± 19	155 ± 17
Rectal temp (°C)	39.4 ± 0.5						38.6 ± 0.9

ing of time to 1st signs or recumbency seen with increasing the ketamine dosage. An area of piloerection, up to 10 cm in diameter, was observed around the dart site prior to recumbency. Tracheal intubation was possible due to adequate muscle relaxation and minimal pharyngeal response. There was minimal chewing movement once the tube was in place. Stimulation of the ears and palpebral area resulted in minor twitching responses in all animals during the monitoring period. Minimal response to painful stimulation was noted and minor intermittent paddling in one-third of the impalas was present, which was not related to stimulation and did not hinder the monitoring. Slight bloating was noted in one-third of the animals, which was resolved by slightly changing the sternal position of the animal from one side to the other. There was no regurgitation but moderate salivation was noted in most animals. The results of physiological monitoring listed in Table 2 were very consistent throughout the monitoring period.

Impalas had increased respiration rates, hypoxia and slight hypertension. The heart rate and rectal temperature were considered to be within expected values. Table 3 shows the average arterial blood gas values. A consistent hypoxia was present while the pH and pCO₂ remained within an acceptable range.

The effect of the reversal drug, atipamezole, in Trial 1 is summarised in Table 1. The initial 3 impalas received atipamezole ¼ i.v. and ¾ i.m., as previously recommended⁹, and resulted in a very rapid arousal with excessive excitement, ataxia, disorientation and a tendency to run into barriers. Subsequently the reversal protocol was changed and atipamezole was given i.m., resulting in a smoother recovery, with the animal 1st raising its head and progressing to a sternal position before rising with slight ataxia lasting up to 15 min. When these animals were observed 4–6 h later, they showed sedation by either standing with their heads down or lying down with their heads on their side. With minor stimulation, either visual or auditory, they responded normally by running off, but when left undisturbed they reverted to a sedated state. All ani-

Table 3: Arterial blood gas data from impala in Trial 1.

Blood gas parameter	Time 0	10 min	20 min	30 min
pH	7.419 ± 0.036	7.429 ± 0.044	7.437 ± 0.041	7.441 ± 0.045
PCO ₂ (mm Hg)	41.3 ± 3.5	40.7 ± 3.2	39.1 ± 3.4	39.1 ± 3.6
PO ₂ (mm Hg)	51.6 ± 11.1	51.9 ± 14.3	53.4 ± 12.3	57.0 ± 17.5

mals appeared normal and responded in a normal way to stimuli the next day.

In Trial 2, some impala became recumbent, but sedation was inadequate for physiological monitoring. Two of 4 animals given ketamine by hand-injection showed transient ataxia at 10 min, lasting about 5 min, while the other 2, which were darted, showed no clinical signs. The 8 impala that received medetomidine at 200 or 300 µg/kg became recumbent on average after 9.75 and 16 min, respectively. The darted animals in each group became recumbent on average in half the time, 8 min, compared to an average of 16 min for hand-injected animals. The down and recumbent impala became alert, responsive and struggled to escape when approached and handling was attempted. The 2 darted animals that received 400 µg/kg became recumbent after 3 and 7 min while one hand-injected impala required 11 min and the other remained mobile and avoided capture. Piloerection was noted in impalas darted with medetomidine, but not in hand-injected animals. In 11 of 12 impalas that received medetomidine it was possible to hand-catch them and administer atipamezole i.m., which produced initial arousal followed by re-sedation, as observed in Trial 1.

The data from all the impala in Trial 3 were combined, since no statistical difference related to age or sex was observed on the measured parameters. The average dose and dosage ranges for medetomidine and ketamine, the onset of initial drug effect, time to recumbency, and lag time to manipulating the animals are listed in Table 4.

The distance covered after darting varied from a few metres up to 200 m and seems to depend on several factors. Impalas that were either alone, excited or interacting with others, especially in bachelor herds, travelled the farthest, while impalas within a quiet group tended to remain near the group and travelled the shortest distance. No dose-dependent shortening of time to initial signs or recumbency associated with the medetomidine dosage was noted. An area of piloerection was observed around the dart site of up to 10 cm in diameter. Tracheal intubation was possible due to good muscle relaxation and minimal pharyngeal response. Stimulation of the ears and palpebral area resulted in a varied twitching response irrespective of the level of sedation. There was minor response to painful stimulation such as needle pricks to the area of the coronary band. There was also minor paddling in a

Table 4: Dosage and results from Trial 3 field-anaesthetised, male and female impala combined ($n = 16$).

Parameters	Mean ± SD	Range
Mass (kg)	41.7 ± 5.6	28–52
Medetomidine (µg/kg)	220 ± 34	173–321
Ketamine (mg/kg)	4.4 ± 0.7	3.5–6.4
Initial signs (min)	3.0 ± 1.2	0.5–5
Recumbent (min)	4.5 ± 1.5	2–7.4
Handle (min)	12.1 ± 3.5	6–17
Atipamezole (µg/kg)	467 ± 108	381–695
Standing (min)	6.6 ± 3.9	3–20

Table 5: Physiological data from field-anaesthetised, male and female impala in Trial 3 ($n = 17$).

Parameters	Time 0	5 min	10 min	15 min	20 min	25 min	30 min
Respiration rate (min)	35 ± 15	36 ± 17	36 ± 12	34 ± 11	34 ± 11	38 ± 10	35 ± 11
Heart rate (min)	53 ± 11	53 ± 10	53 ± 12	52 ± 10	52 ± 11	51 ± 11	52 ± 10
O ₂ saturation (%)	86 ± 3	84 ± 7	87 ± 5	85 ± 6	87 ± 3	86 ± 3	87 ± 3
Systolic blood pressure (mm Hg)	156 ± 12	149 ± 9	155 ± 15	148 ± 11	150 ± 12	148 ± 8	147 ± 13
Rectal temperature (°C)	38.8 ± 0.7						38.0 ± 0.7

1/3 of the impalas, which was not related to stimulation and did not impede monitoring. Two impala seemed to be very relaxed but became very reactive irrespective of stimulation and then relaxed and calmed down for the remaining monitoring period. Slight bloating was noted in half of the animals, which was resolved by changing the position of the animal, allowing eructation with no regurgitation.

The physiological data for Trial 3 are presented in Table 5. The parameters remained stable throughout the monitoring period, and were comparable to values obtained in Trial 1, including an increased respiration rate, hypoxia and slight hypertension. The heart rate and rectal temperature were within expected ranges. The average arterial blood gas values are shown in Table 6 and as in Trial 1 show hypoxia, with the pH and pCO₂ remaining within acceptable ranges.

The reversal drug and its effects for Trial 3 are listed in Table 4. It was noted that field-anaesthetised individuals required longer to stand and showed ataxia for 5–15 min.

DISCUSSION

Trial 2 showed that the immobilisations observed in Trials 1 and 3 were the result of an apparent synergistic action since ketamine alone produced no sedation and there was minimal sedation from using medetomidine even at the higher dosages (400 µg/kg). Impala appear to tolerate moderate to high doses of ketamine i.m. with no obvious adverse effect. It was surprising that medetomidine alone failed to produce more sedation than observed since ruminants have increased sensitivity to the sedative effects of α₂ agonists compared to other species⁹, but experience has shown that impala generally require higher doses of most immobilising drugs to achieve a desired effect.

Impalas in Trial 3 had a faster onset and recumbency than impalas in Trial 1 even though they were heavier on average and also received lower doses of the medetomidine/ketamine combination. This may be the result of hyaluronidase added to the dart in Trial 3, but in some

Table 6: Arterial blood gas data for field-anaesthetised, male and female impala in Trial 3 ($n = 17$).

Blood gas parameter	Time 0	10 min	20 min	30 min
pH	7.416 ± 0.047	7.441 ± 0.053	7.448 ± 0.030	7.459 ± 0.036
PCO ₂ (mm Hg)	41.3 ± 5.0	39.1 ± 5.2	39.6 ± 3.4	39.4 ± 3.9
PO ₂ (mm Hg)	46.1 ± 10.0	47.7 ± 10.3	48.5 ± 9.2	48.0 ± 10.0

species such as giraffes³ and chamois¹⁵ there was no observed shortening of time to observed drug effect and recumbency by adding hyaluronidase to a medetomidine/ketamine combination. This is thought to be due to the inherent rapid action of the medetomidine/ketamine combination. Another consideration for longer times to drug effect and recumbency in Trial 1 was the stimulation of other impala in the confinement of the boma. This continual interaction and stimulation during induction and initial recumbency probably led to a longer induction time.

The immobilisations in Trials 1 and 3 provided comparable physiological data, showing stable heart rates, rectal temperatures within acceptable ranges and adequate muscle relaxation, which facilitated tracheal intubation. Animals in both trials also had increased respiration rates and slightly elevated systolic blood pressure. In both trials the blood gas values for pH and PaCO₂ were stable and within acceptable ranges. Both pulse oximetry and PaO₂ indicated hypoxia in the presence of an elevated respiration rate, indicating that the tidal volume was inadequate; therefore, when using this combination, it is suggested that equipment for respiratory support be available as with any chemical immobilisation procedure⁶. Sheep anaesthetised with α₂ agonists, specifically medetomidine i.v., also showed decreased PaO₂, stable PaCO₂ and an increased respiration rate⁴.

The immobilisation produced by this combination was adequate for monitoring, capture procedures or minor manipulations, but if major manipulation such as surgery were performed, then additional anaesthetics would be required as previously recommended with this combination⁹.

The cause of the piloerection observed

is uncertain. Piloerection only occurred in impalas that received medetomidine by dart and was absent when medetomidine was hand-injected, which indicates that the phenomenon is a result of combination of drug action and delivery procedure. Piloerection was useful in the field situation, allowing identification of the darted animal even if the dart fell out.

Atipamezole is described as a selective α₂ antagonist but in this study did not completely reverse the sedative effect of medetomidine at the dose used since we observed re-sedation after initial arousal. Reports of re-sedation occur either following i.v. atipamezole or when the ratio of atipamezole:medetomidine was less than 5:1⁹. Re-sedation 2–4 h after i.m. atipamezole was reported in free-ranging cattle darted with medetomidine and reversed at a atipamezole:medetomidine ratio of 2:1^{1,15}. In the current study we used a high dose of atipamezole/kg, but since the dosage of medetomidine was also high we only obtained an average ratio of 2.3:1 in Trial 1 and 2.1:1 in Trial 3. Therefore, in field situations where the animal is released into the wild after the procedure, re-sedation would not be observed and the animal would be at risk of predation and/or conspecific aggression. Further studies are needed to assess whether increasing the atipamezole:medetomidine ratio to 5:1 would prevent re-sedation. In a captive situation the recovering animal can be isolated and observed and the re-sedation could be beneficial in acclimatising the animal to new surroundings.

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