

Reversible Immobilization of Free-ranging Red Deer (*Cervus elaphus*) with Xylazine-Tiletamine-Zolazepam and Atipamezole

Celsus Sente,^{1,2} Erling L. Meisingset,¹ Alina L. Evans,^{3,4} Sari J. Wedul,³ Barbara Zimmermann,³ and Jon M. Arnemo^{3,5,6} ¹Norwegian Institute for Agricultural and Environmental Research, Organic Food and Farming Division, NO-6630 Tingvoll, Norway; ²Department of Wildlife and Aquatic Animal Resources, School of Veterinary Medicine and Animal Resources, College of Veterinary Medicine, Animal Resources and Bio-security, Makerere University, PO Box 7062 Kampala, Uganda; ³Department of Forestry and Wildlife Management, Faculty of Applied Ecology and Agricultural Sciences, Hedmark College, Campus Evenstad, NO-2418 Elverum, Norway; ⁴Section of Arctic Veterinary Medicine, Norwegian School of Veterinary Science, NO-9292 Tromsø, Norway; ⁵Department of Wildlife, Fish and Environmental Studies, Faculty of Forest Sciences, Swedish University of Agricultural Sciences, SE-901 83, Umeå, Sweden; ⁶Corresponding author (email: jmarnemo@online.no)

ABSTRACT: Forty-eight free-ranging red deer (*Cervus elaphus*) were immobilized with xylazine (X) and tiletamine-zolazepam (TZ) by dart injection during winter 2008 in Norway. A follow-up study in five animals during winter 2010 included arterial blood samples analyzed with a portable clinical analyzer in the field. Thirty-five of 48 animals were effectively immobilized and 13 required a second dart. Mean±SD doses were 2.89±0.45 mg X/kg and 2.89±0.45 mg TZ/kg in calves and 2.97±0.66 mg X/kg and 1.91±0.43 mg TZ/kg in adults. Mean induction times for calves and adults were 8.5±5 min and 11.6±5.5 min, respectively. The main physiologic side effect during immobilization was hypoxemia (pulse oximetry, SpO₂<85%). All five animals evaluated with arterial blood gas samples were hypoxemic (PaO₂<10 kPa). Xylazine was antagonized with 0.43±0.19 mg/kg and 0.27±0.05 mg/kg of atipamezole in calves and adults, respectively. Time to standing/walking in calves and adults was 12±7 min and 12±11 min, respectively. Two capture mortalities occurred.

Key words: Atipamezole, *Cervus elaphus*, immobilization, red deer, tiletamine, xylazine, zolazepam.

Norway has a large and growing population of red deer (*Cervus elaphus*) and more than 35,000 animals are harvested annually. Although more than 1,000 free-ranging animals have been chemically immobilized for various research and management purposes during the past 30 yr, we are aware of only one scientific paper on the effects of immobilizing drugs (Arnemo et al. 1994). Here we evaluate xylazine-tiletamine-zolazepam and atipamezole for reversible immobilization of free-ranging red deer.

This study was conducted at several wintering sites of red deer in southwestern (60–63°N, 6–8°E; main study, January–April 2008) and southeastern Norway (61°25′N, 11°5′E; follow-up study, March–April 2010). The study was approved by the National Animal Research Authority, Oslo, Norway. Data from 48 of 86 red deer immobilizations were collected in the main study. Darting was carried out during dark hours from a motor vehicle or blind at supplemental feeding stations with artificial light. Drugs were administered at a distance of 10–30 m, using a CO₂-powered rifle and lightweight 2.0 mL or 3.0 mL darts with 2×30-mm barbed needles with side ports (Dan-Inject®, Børkop, Denmark). Calves (<12 mo) were darted with 1.8 mL of a solution of 500 mg xylazine (X) dry powder (Rompun®, Bayer, Leverkusen, Germany) and 500 mg tiletamine-zolazepam (TZ) dry powder (Zoletil®, Virbac, Carros, France) dissolved in 5 mL of sterile water. Adults (>18 mo) were darted with 2–3 mL of a solution of 1,000 mg X and 500 mg TZ dissolved in 5 mL sterile water based on their estimated body masses. The time to first signs of drug effects (lowering of the head, lack of coordination, and staggering) and induction time (time from darting to recumbency or manual restraint) were recorded. A recumbent animal was slowly approached, blindfolded, kept in sternal recumbency, and clinically examined by a veterinarian. Respiratory rate (observing flank movements), heart rate (cardiac auscultation), rectal temperature (using a digital

thermometer), and relative arterial hemoglobin oxygen saturation (SpO_2) using a pulse oximeter (Nellcor® N-20P, Nellcor Inc., Pleasanton, California, USA) with the sensor (VetSat®, Nellcor) attached to the tongue, were recorded 5–7 min, 12–14 min, and 19–21 min after capture. In the follow-up study, arterial blood samples were collected anaerobically from the auricular artery at 30 min and 50 min after darting, using self-filling arterial syringes with heparin (PICO™ 70, Radiometer Copenhagen, Brønshøj, Denmark) and analyzed immediately with an i-STAT®1 Portable Clinical Analyzer and i-STAT® CG4+ and EC8+ cartridges (Abbott Laboratories, Abbott Park, Illinois, USA). Measured variables included pH, partial pressure of arterial oxygen (PaO_2), partial pressure of carbon dioxide ($PaCO_2$), lactate, hematocrit, sodium, potassium, chloride, urea, and glucose. Partial pressure of arterial oxygen, $PaCO_2$, and pH were corrected based on rectal temperature. Calculated values included bicarbonate, hemoglobin, oxygen hemoglobin saturation, total carbon dioxide, base excess, and anion gap. The palpebral reflex was assessed by touching the medial canthus of the eye and the anal reflex was checked when the thermometer was inserted. Hypothermic animals (rectal temperature ≤ 35 C) were placed on an insulating blanket to avoid further cooling. All animals were tagged in both ears and selected adults were instrumented with a geographic positioning system collar (Tellus Basic 5H2D, Followit AB, Lindesberg, Sweden). Animals were weighed (± 0.5 kg) using a scale (Salter 233-10, Salter Brecknell, Smethwick, West Midlands, UK). Body condition was assessed according to Neary and Yager (2002). Each animal was given a body condition score (BCS) from 1 (very thin) to 5 (very fat). Body measurements were recorded and blood, fecal, and hair samples were collected. Atipamezole (Antisedan® 5 mg/ml, Orion Pharma Animal Health, Turku, Finland) was administered intramuscularly

at 0.15 mg/mg xylazine and the animals were left in sternal recumbency to recover. The time from darting to administration of atipamezole (time to reversal) and time from administration of atipamezole to lifting of the head (head-up time) and to standing (on-feet time) were recorded. The animals were observed for as long as possible to assess postreversal drug effects. In the main study, hypoxemia was defined as pulse oximetry reading $< 85\%$. In the follow-up study, hypoxemia was defined based on arterial blood oxygen levels as mild ($PaO_2 = 8\text{--}10$ kPa), marked ($PaO_2 = 5.5\text{--}8$ kPa), or severe ($PaO_2 < 5.5$ kPa). Acidemia was defined as a $pH < 7.35$, and was considered marked if pH was < 7.20 . Hypocapnia was defined as a $PaCO_2 < 4.5$ kPa and was defined as mild ($PaCO_2 = 6\text{--}8$ kPa) or marked ($PaCO_2 > 8$ kPa). Recorded data were analyzed using a general linear model in SPSS version 16.0.1 (SPSS Inc., Chicago, Illinois, USA). Results from the first and second arterial blood samples were compared using a two-tailed paired *t*-test (JMP 8, SAS Inc., Cary, North Carolina, USA). Differences in the models and the *t*-tests were considered significant at $P < 0.05$. Thirteen calves (six females, seven males) and 22 adults (10 females, 12 males) immobilized after one dart were included in the statistical analysis. Thirteen animals that required a second dart to achieve complete immobilization were excluded. In the follow-up study, five adult females were immobilized with one dart. Data from these animals were not included in the main study.

Summary statistics from the main study are presented in Table 1. The first signs after darting included hind limb ataxia, followed by front limb ataxia, reduced alertness, head and ear drooping, lack of coordination, and then recumbency. Induction time in adults was correlated with BCS; lean animals had a significantly shorter induction time, even after controlling for body weight and drug dose. Pulse

TABLE 1. Results of immobilization of free-ranging red deer (*Cervus elaphus*) with xylazine-tiletamine-zolazepam, administered by dart syringe, and remobilized with atipamezole intramuscularly in Norway, 2008.

Parameter ^a	Calves		Adults	
	n	Mean ± SD (range)	n	Mean ± SD (range)
Body mass (kg)	13	52 ± 8 (39–65)	22	100 ± 20 (59–138)
Body condition score	13	3 ± 1 (2–4)	22	2 ± 1 (1–3)
Xylazine (mg/kg)	13	2.89 ± 0.45 (2.31–3.85)	22	2.97 ± 0.66 (1.85–4.22)
Tiletamine-zolazepam (mg/kg)	13	2.89 ± 0.45 (2.31–3.85)	22	1.91 ± 0.43 (1.23–2.82)
Time to first sign	11	5.4 ± 4.4 (2.0–15.0)	18	4.4 ± 3.7 (1.0–17.0)
Induction time (min)	13	8.5 ± 5 (3.0–17.0)	22	11.6 ± 5.5 (4.0–23.0)
SpO ₂ (%), 5–7 min	7	72 ± 18 (48–96)	20	74 ± 20 (48–97)
SpO ₂ (%), 12–14 min	7	72 ± 17 (48–100)	20	73 ± 19 (48–95)
SpO ₂ (%), 19–21 min	7	73 ± 17 (48–99)	20	76 ± 20 (46–99)
Pulse rate (beats/min), 5–7 min	13	40 ± 14 (24–74)	22	44 ± 16 (24–80)
Pulse rate (beats/min), 12–14 min	13	44 ± 15 (24–80)	22	44 ± 13 (24–84)
Pulse rate (beats/min), 19–21 min	13	56 ± 26 (24–107)	22	44 ± 12 (20–80)
Rectal temperature (C), 5–7 min	13	37.6 ± 1.0 (35.3–38.8)	22	37.8 ± 0.7 (35.8–39.0)
Rectal temperature (C), 12–14 min	13	37.6 ± 0.9 (36.1–38.7)	22	37.8 ± 0.8 (35.5–38.8)
Rectal temperature (C), 19–21 min	13	37.3 ± 1.6 (33.0–39.0)	22	37.7 ± 0.7 (36.0–39.0)
Respiratory rate (breaths/min), 5–7 min	13	25 ± 17 (14–78)	22	24 ± 12 (10–62)
Respiratory rate (breaths/min), 12–14 min	13	27 ± 17 (14–80)	22	27 ± 12 (10–50)
Respiratory rate (breaths/min), 19–21 min	13	35 ± 19 (18–80)	22	29 ± 19 (10–88)
Time to reversal (min)	13	30 ± 9 (19–46)	22	36 ± 11 (18–59)
Atipamezole (mg/kg)	13	0.43 ± 0.19 (0.17–0.90)	22	0.27 ± 0.05 (0.17–0.37)
Head up (min)	13	8 ± 7 (0–26)	22	8 ± 10 (1–47)
On-feet time (min)	13	12 ± 7 (3–26)	22	12 ± 11 (3–48)

^a SpO₂ = relative arterial hemoglobin oxygen saturation.

oximetry levels were negatively correlated with the drug dose (lower SpO₂ in animals receiving more drugs per kilogram) and positively correlated with induction time (higher values with increasing induction times). Body condition score tended to be negatively related to SpO₂ (lower saturation in animals with high BCS). There was a significant negative association between pulse rate and induction (shorter induction time was associated with higher pulse rate) and a negative correlation between pulse and respiratory rates (the higher the respiratory rate the lower the pulse rate). Rectal temperature was inversely related to induction time and BCS (rectal temperature decreased as induction time and BCS increased and vice versa), and was directly proportional to dose, body weight, and SpO₂. Five of seven calves and 10 of 20 adults exhibited hypoxemia (SpO₂ < 85%), and one calf that died postcapture was

hypothermic (rectal temperature < 35 C). Palpebral and anal reflexes were present in lightly immobilized animals and absent in moderately to deeply immobilized animals.

Results from the arterial blood samples analyzed with the iSTAT[®] during the follow-up study are presented in Table 2. All animals had at least one arterial oxygen tension reading below 10 kPa (mild hypoxia) and one animal was below 8 kPa (moderate hypoxia). Two animals had pH values below 7.35 (mild acidemia). All animals had mild hypercapnia (PaCO₂ = 6–8 kPa), which increased significantly over time.

A female calf died 3 hr postcapture. This animal did not recover after reversal and developed bradycardia (pulse rate < 20) and hypothermia (33 C). Additionally, one yearling male was found dead 15 days postcapture, with most of its parts consumed by scavengers.

TABLE 2. Summary statistics for free-ranging red deer (*Cervus elaphus*) immobilized with xylazine-tiletamine-zolazepam and evaluated with the iStat® blood gas analyzer in Norway, 2010.

Time from darting Variable ^a	27–43 min		46–80 min	
	<i>n</i>	Mean ± SD (range)	<i>n</i>	Mean ± SD (range)
Pulse rate (beats/min)	5	31 ± 3 (28–36)	5	32 ± 3 (28–36)
Respiratory rate (breaths/min)	5	14 ± 6 (10–24)	5	13.4 ± 4.1 (9–20)
Rectal temperature (C)	5	38.4 ± 1.0 (36.6–39.1)	5	38.5 ± 0.6 (37.7–39.2)
SaO ₂ ^b (%)	5	91 ± 5 (83–95)		92.3 ± 3.3 (88–96)
PaO ₂ ^b	5		4	
(kPa)		9.6 ± 1.4 (7.7–11.2)		9.7 ± 1.1 (9.0–11.3)
(mmHg)		72 ± 10 (58–84)		73 ± 8 (67–85)
PaCO ₂ ^b	5			
(kPa)		6.5 ± 0.2 (6.2–6.7)		6.8 ± 0.4 (6.3–7.1)
(mmHg)		49 ± 2 (47–50)		51 ± 3 (47–53)
pH ^{b,c}	5	7.37 ± 0.06 (7.29–7.44)	4	7.39 ± 0.07 (7.30–7.47)
Lactate ^c (mmol/L)	3 ^d	1.3 ± 1.0 (0.7–2.5)	2	1.0 ± 0.6 (0.6–1.5)
Anion gap (mEq/L)	5	13 ± 3 (10–18)	5	11 ± 2 (10–14)
Na (mmol/L)	5	138 ± 2 (135–139)	5	137 ± 2 (135–139)
K (mmol/L)	5	4.3 ± 0.5 (3.7–4.5)	5	4.5 ± 0.4 (4.1–4.9)
Cl (mmol/L)	5	102 ± 4 (99–109)	5	101.4 ± 3.5 (97–105)
Urea (mmol/L)	5	3.1 ± 1.2 (1.2–4.5)	5	3.1 ± 1.2 (1.3–4.6)
Glucose ^c (mmol/L)	5	9.3 ± 1.7 (7.8–12.1)	5	11.0 ± 1.5 (8.9–12.7)
Hematocrit (% PCV)	5	36 ± 2 (33–39)	5	35.6 ± 2.6 (33.0–40.0)
Hemoglobin (g/L)		121 ± 8 (112–133)	5	121 ± 9 (112–136)
Base excess ^c (mmol/L)	5	1.6 ± 4.7 (–4.0–+6.0)	5	3.8 ± 4.2 (–1.0–+9.0)
HCO ₃ ^c (mmol/L)	5	26.7 ± 3.7 (22.7–30.2)	5	28.9 ± 3.2 (25.0–32.5)

^a SaO₂ = arterial oxygen saturation; PaO₂ = partial pressures of arterial oxygen; PaCO₂ = partial pressures of carbon dioxide; Na = sodium; K = potassium; Cl = chloride; PCV = packed cell volume; HCO₃ = bicarbonate.

^b Blood gas values were corrected to rectal temperature.

^c Significant difference between sample 1 and sample 2.

^d Two animals had lactate values too low for the blood-gas analyzer to read and were not included in the mean.

Due to panic reactions during aerial approach, helicopter capture is not a recommended capture technique in red deer and chemical immobilizations are usually carried out by darting from a motor vehicle or blind. Xylazine-tiletamine-zolazepam (XTZ) provided adequate immobilization and induction times were comparable to the findings of Janovsky et al. (2000), who used a 1:1 ratio of X:TZ in semicaptive red deer. Short induction times are important to reduce the risk of injury and side effects (Caulkett and Arnemo 2007). Although all immobilizing drugs can be dangerous to humans, XTZ poses a lower risk than the highly potent opioid combinations (Kreeger and Arnemo 2012).

Although XTZ successfully immobilized all animals, hypoxemia (SpO₂ < 85%) occurred in calves and adults. Read et al. (2001) evaluated the XTZ combination in elk and reported a PaO₂ of 43 ± 11.8 mmHg and an SpO₂ of 45–91%, indicating marked hypoxemia. The five animals in our study in which arterial oxygen saturation was measured had only mild hypoxemia and hypercapnia, indicating that the respiratory depression with this drug combination in red deer was less than reported in other studies. However, the oxygen status of these animals could still be improved by supplemental oxygen as has been documented in other ruminants. For Read et al. (2001), hypoxemia was corrected after only 5 min of oxygen

administration. Paterson et al. (2009) compared nasal oxygen and medical air supplementation in elk before and during anesthesia with carfentanil and xylazine and found that elk receiving 10 L/min of oxygen had a significantly faster induction and recovery, less hypoxemia, less rigidity and movement, but more apnea, hypercapnia, and acidosis. Read (2003) found that the alpha-2 agonists commonly used in combinations with other anesthetic agents may cause serious hypoxemia in wild ruminants and that in domestic sheep, they can result in changes in pulmonary parenchyma, increased venous admixture, and pulmonary edema. As with other ruminant species, anesthetized red deer should be kept in sternal recumbency to facilitate breathing and to avoid tympany and regurgitation (Caulkett and Arnemo 2007).

Recoveries were quick in both calves and adults and comparable to recovery times reported by Janicki et al. (2006), who also used atipamezole for reversal. Atipamezole is a more potent and selective alpha₂-adrenoceptor antagonist than other antagonists such as tolazoline and yohimbine used in other studies (Millspaugh et al. 1995; Janovsky et al. 2000; Walter et al. 2005; Rosatte 2007).

The death of two animals was attributed to stress as a result of immobilization. Mortality rates reported from other studies using XTZ in red deer or elk range from zero (Millspaugh et al. 1995; Walter et al. 2005; Janicki et al. 2006) to 5% (Janovsky et al. 2000). Closer monitoring of physiologic variables, including body temperature, and corrective action (warming or early reversal) may have prevented at least one of the deaths.

The XTZ combination was effective for field immobilization of free-ranging red deer. We recommend that vital signs and oxygenation be continuously monitored during immobilization and that supplemental oxygen be considered to alleviate hypoxemia.

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