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Thiafentanil-azaperone-xylazine and carfentanil-xylazine immobilizations of free-ranging caribou (*Rangifer tarandus granti*) in Alaska

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RH: LIAN ET AL.—IMMOBILIZATIONS OF FREE-RANGING CARIBOU IN ALASKA

THIAFENTANIL-AZAPERONE-XYLAZINE AND CARFENTANIL-XYLAZINE IMMOBILIZATIONS OF FREE-RANGING CARIBOU (*RANGIFER TARANDUS GRANTI*) IN ALASKA

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ABSTRACT: Carfentanil-xylazine (CX) has been the primary drug combination used for immobilizing free-ranging ungulates in Alaska, US, since 1986. We investigated the efficacy of a potential new drug of choice, thiafentanil (Investigational New Animal Drug A-3080). To determine effective doses for helicopter darting to immobilize free-ranging caribou calves (*Rangifer tarandus granti)* for radiocollaring, initial dosing trials were conducted on captive adult caribou. Captive trials indicated thiafentanil-azaperone-medetomidine could provide good levels of immobilization. However, field trials conducted in October 2013 on free-ranging caribou calves found the combination too potent, causing three respiratory arrests and one mortality. The protocol was revised to thiafentanil-azaperone-xylazine (TAX), with good results. The induction time was not significantly different between the two combinations. However, the recovery time was significantly shorter for the TAX group than the CX group. A physiological evaluation was performed on 12 animals immobilized on CX and 15 animals on TAX. Arterial blood was collected after induction and again after 10 minutes of intranasal oxygen supplements (1 L/min). Both groups had significant increases in PaO_2 after oxygen treatment. There was a concurrent significant increase in $PaCO_2$ in both groups. Rectal temperature increased significantly in both groups during the downtime, which is consistent with other studies of potent opioids in ungulates. Based on our results, we found TAX to be a potential alternative for the current CX protocol for immobilizing free-ranging caribou calves via helicopter darting.

Key words: Anesthesia, carfentanil, caribou, hypoxemia, immobilization, opioid, thiafentanil.

INTRODUCTION

Caribou (*Rangifer tarandus granti*) have been chemically immobilized for management, translocations and research purposes in Alaska, US, since 1965 (Glenn 1967). Early

immobilizations were performed with neuromuscular blocking drugs (Bergerud et al. 1964; Glenn 1967), which were neither reliable nor safe. Etorphine in combination with acepromazine replaced the neuromuscular blocking drugs. However, etorphine was not readily available in a higher concentration than 1 mg/ml (Fong 1982; Valkenburg et al. 1983). When the potent opioid carfentanil was introduced to the market in 1986 at a concentration of 3 mg/ml, it became the drug of choice for caribou immobilizations in Alaska. For the following 28 years, carfentanil in combination with the sedative xylazine has been the primary drug combination for free-ranging caribou immobilizations (Adams et al. 1988; Boertje et al.1996; Valkenburg 1997; Valkenburg et al. 1999).

Ketamine in combination with medetomidine or xylazine is the drug of choice for rangifer immobilizations in Scandinavia and Canada (Arnemo et al. 2000; Arnemo and Aanes 2009; Arnemo et al. 2011; Cattet 2011).This combination has been tested in Alaska caribou by one of the authors (K. B. B), but with longer than preferred induction and recovery times.

Wildlife Pharmaceuticals Inc. (Windsor, Colorado, USA) and the Alaska Department of Fish and Game (ADF&G) collaborated to evaluate an alternative drug protocol and determine an effective dose for the Investigational New Animal Drug A-3080 (thiafentanil), in caribou. Thiafentanil has the potential to replace carfentanil as drug of choice for ungulate immobilizations. It has been used successfully in other wild ungulate immobilizations including moose (*Alces alces*) and elk (*Cervus canadensis*) (Stanley et al. 1988; McJames et al. 1994; Wolfe et al. 2004; Kreeger et al. 2005). Thiafentanil has a significantly shorter duration of action, and is less potent than carfentanil. Like carfentanil it is fast acting and fully reversible with the opioid antagonist naltrexone (Stanley and McJames 1986; Lance and Kenny 2011). Thiafentanil has a much higher therapeutic index than carfentanil and thus potentially offers a greater safety index for the human operator (Stanley and McJames 1986). Other studies have found that thiafentanil offers faster induction times than carfentanil and

etorphine, and a wide safety range in wild ungulates (Wolfe et al. 2004; Kreeger et al. 2005; Meyer et al. 2008). One of the authors (D. J. D.) found this in caribou as well.

Azaperone is a tranquilizer with a wide safety range. Added to a drug combination including a potent opioid it is thought to improve respiration and decrease risk of hyperthermia (Meyer et al. 2008; Kreeger and Arnemo 2012). Azaperone also causes a certain degree of vasodilation, and can counteract the initial vasoconstriction caused by the alpha-2 agonist (Meyer et al. 2008). Xylazine is an alpha-2 agonist providing sedation and muscle relaxation. Combined with thiafentanil and azaperone, xylazine is expected to improve muscle relaxation and ease handling of the immobilized animal. Medetomidine is another alpha-2 agonist, more potent and selective for the alpha-2 agonist receptors than xylazine (Read 2003).

Potent opioids like carfentanil and thiafentanil are major respiratory depressants (Haigh 1990; Caulkett et al. 1994). Normally there is considerable capacity to increase the rate of oxygen supply and CO_2 removal from the body tissues. However, during general anesthesia these adaptive systems are compromised. Anesthetized animals are incapable of regulating gas-exchange, increasing ventilation or cardiac output, and the spleen is often dilated and incapable of contracting to increase hemoglobin levels (McDonell and Kerr 2007). The result is hypoxemia; where the oxygenation of the blood is insufficient to meet the metabolic requirements, resulting in damage to vital organs (Caulkett et al. 1994). Severe hypoxemia has been documented in several immobilized wild cervids (Read et al. 2001; Read 2003; Mich et al. 2008; Paterson et al. 2009; Risling et al. 2011; Evans et al. 2012; Fahlman et al. 2012; Evans et al., 2013; Lian et al. 2014). Subsequent adverse effects on behavior after chemical immobilizations have been documented in species like moose, polar bears (*Ursus maritimus*), grizzly bears (*Ursus arctos*), black bears (*Ursus americanus*) and mountain goats

(*Oreamnus americanus*) (Ramsay and Stirling 1986; Larsen and Gauthier 1989; Cotè et al.1998; Cattet et al. 2008).

In this study the current carfentanil-xylazine protocol was compared to an alternative protocol using thiafentanil. To determine the appropriate dose for free-ranging caribou, an initial dose titration trial was conducted on captive caribou. In the free-ranging study a physiological evaluation of both the current carfentanil-xylazine protocol and the alternative protocol consisting of thiafentanil in combination with azaperone and an alpha-2 agonist was conducted.

MATERIALS AND METHODS

Study area and animals

Fieldwork was conducted at four locations in Alaska during fall 2013 and spring 2014. Forty-five female caribou calves were immobilized for radiocollaring and weighing. Only animals successfully immobilized with a single dart were included in the study. In October 2013, data were collected from 22 female calves from the Fortymile herd darted from a helicopter for radiocollaring in the Ladue River drainage in eastern Interior Alaska (63°30.70′N, 141°46.20′W), altitude 549–1,219 meters. In April 2014 data were collected from eight calves from the Mulchatna herd similarly darted and handled near Tundra Lake in southwest Alaska (61°13.50′N, 155°43.00′W), altitude 593 meters, and one from the Fortymile herd near Glacier Mountain in eastern Interior Alaska (64°42.40′N, 141°47.00′W), altitude 1,300 meters. In May 2014 data were collected from 14 Fortymile herd calves in the upper Charley River drainage in eastern Interior Alaska (64°51.40′N, 143°15.70′W), altitude 1,000 meters. Calves were approximately 5 mo old in the fall captures and 11 mo old in the spring captures. The ambient temperature ranged from −7 C to 10 C.

Captive caribou trials were performed at University of Alaska Fairbanks Agricultural Research Station in Palmer Alaska in September 2013. Five yearlings were hand injected and two adults were darted with a CO_2 powered rifle (Dan-Inject, Børkop, Denmark) with different drug combinations.

All captures and handling methods were approved by ADF&G's Division of Wildlife Conservation Institutional Animal Care and Use Committee (IACUC) protocol number 2013-029F011.

Drug combinations

The carfentanil-xylazine (CX) drug combination for the fall captures (CX fall dose) was made by combining 1.5 mg carfentanil citrate (3 mg/ml, Zoopharm, Inc., Laramie, Wyoming, USA) and 20 mg xylazine (AnaSed® 100 mg/ml, Iowa, USA) in a 1 ml dart (Palmer Cap-Chur®, Powder Springs Georgia, USA). To account for heavier calves during the spring captures the carfentanil-xylazine dose was increased to 1.8 mg carfentanil and 25 mg xylazine (CX spring dose). The thiafentanil-azaperone-xylazine (TAX) drug combination was a mixture of 1.5 mg thiafentanil (10 mg/ml, Zoopharm, Inc.), 25 mg azaperone tartrate (50 mg/ml, Zoopharm, Inc.), and 20 mg xylazine in a 1 ml dart. The thiafentanil-azaperone-medetomidine drug combination was made in two concentrations: TAM_{high} and TAM_{low}. The TAM_{high} dose was made by adding 1.5 mg thiafentanil, 25 mg azaperone and 5 mg medetomidine hydrochloride (20 mg/ml, Zoopharm Inc) to a 1 ml dart. The TAM_{low} dose was made in the same way but with 2 mg medetomidine. In some instances thiafentanil had crystallized in the vial. This was solved by heating the vial with hot water or by a lighter flame under the vial. All darts had a 19 mm barbed, end-ported needle.

Female caribou calves were identified and darted from a Robinson R-44 helicopter (Robinson Helicopter Company, Torrance, California, USA) using a 22 caliber cartridge-fired

or CO_2 powered remote delivery system (Palmer Cap-Chur® pistol or Pneu-Dart® rifle, Williamsport, Pennsylvania, USA) in epaxial or gluteal muscles from a distance of 5 - 10 meters. After handling procedures were completed carfentanil was antagonized with 100 mg naltrexone hydrochloride (50 mg/ml, Zoopharm Inc) per 1 mg carfentanil. Xylazine was antagonized with 2 mg tolazoline hydrochloride (200 mg/ml Zoopharm Inc) per kg body weight. Thiafentanil was antagonized with 33 mg naltrexone/1 mg thiafentanil. Medetomidine was antagonized with atipamezole (20 mg/ml, Zoopharm Inc), in a relationship of 5 mg atipamezole for every 1 mg of medetomidine. This resulted in 150 mg naltrexone to antagonize 1.5 mg of carfentanil and 180 mg naltrexone to antagonize 1.8 mg of carfentanil. Fifty-milligram naltrexone was used to antagonize thiafentanil. Tolazoline at a dose of 100 – 200 mg was used to antagonize xylazine and 25 mg atipamezole was used to antagonize 5 mg of medetomidine, whereas 10 mg atipamezole was used to antagonize 2 mg of medetomidine. The antagonists were administered intramuscularly (IM) in brachial or femoral muscle groups. For caribou recumbent with head on the ground at approach, tolazoline was administered immediately after the first arterial sample to achieve similar level of immobilization in all animals. Three emergencies required immediate intervention including intravenous (IV) administration of antagonists into the cephalic vein. For respiratory emergencies, doxapram (Dopram Injection 20 mg/ml, New Jersey, USA) was administered at a dose of 100 mg (5 ml) IV.

Twenty-one calves from the Fortymile herd were immobilized with the CX combination. Six were immobilized with the CX fall dose and 15 were immobilized with the CX spring dose. . Nineteen calves were immobilized with the TAX combination. Out of these 11 were immobilized in the fall from the Fortymile herd and eight were immobilized in the spring from the Mulchatna herd. Two calves were immobilized with the TAM_{high} dose and

three were immobilized with the TAM_{low} dose. These five caribou were all from the fall captures in the Fortymile herd. The TAX dose was the same for both fall and spring captures.

Captive caribou were immobilized with one of the following combinations: thiafentanil-medetomidine, thiafentanil-azaperone or thiafentanil-azaperone-medetomidine. Before the immobilization, all animals were weighed on a chute scale. Five yearlings were manually restrained for hand injection with an 18-gauge, 40 mm needle. Two adults were darted with a CO_2 powered dart rifle (Dan-Inject, Børkop, Denmark). Antagonists were the same as for the free-ranging animals.

Physiological evaluation and oxygen treatment

Variables recorded for the free-ranging animals included time from darting to recumbency (induction time), time from recumbency to approach and handling the animal (capture time), time from darting until antagonists were administered (handling time) and time from administration of antagonist to standing (recovery time). These variables are summarized in Table 1. For both captive and free-ranging animals induction and recovery quality were assessed subjectively. Caribou found in lateral recumbency at approach, were placed in sternal recumbency. For those with head on the ground, snow was cleared around the nostrils. Pulse rate was measured by palpation of the auricular artery, respiration rate by counting thoracic elevations and rectal temperature with a digital thermometer. All variables were measured when the animal was first approached after recumbency, and repeated 10 minutes later.

Degree of central nervous system (CNS) depression was classified as level I (mildly affected, voluntary movement and intact reflexes), level II (no voluntary movement and intact reflexes), level III (unconsciousness, depressed reflexes, muscular relaxation) and level IV

(ceased respiration, dilated pupils). This evaluation was assessed at approach, and again 10 minutes later.

As soon as possible after approach and again 10 minutes after start of intranasal oxygen supplement an arterial blood sample was collected anaerobically from the auricular artery, using a pre-heparinized (Heparin Sodium Injection, USP 1,000 USP units/ml) 1 ml slip-tip syringe (Tuberculin Slip Tip Syringe, New Jersey, USA) and a 23-gauge needle. The blood sample was analyzed immediately using an i-STAT®1 Portable Clinical Analyzer and i-STAT® CG4+ cartridges (Abbott Laboratories, Illinois, USA). The analyzer was kept in an insulated box with warm water bottles to keep it at optimum temperature (16 - 30 C).

Measured variables included pH, partial pressure of arterial oxygen (PaO_2), partial pressure of arterial carbon dioxide ($PaCO_2$) and lactate. PaO_2, $PaCO_2$ and pH were corrected based on rectal temperature. Calculated values included bicarbonate (HCO_3^-), hemoglobin oxygen saturation (SaO_2), and base excess (BE).

All animals were administered intranasal oxygen insufflation from a portable oxygen cylinder at a flow rate of 1 L/min (all free-ranging animals and four captive animals) or 2 L/min (three of the captive animals). The oxygen nasal line was inserted 10 cm into one nostril and fixed with a clothes pin or tape. Venous blood samples for serum biochemistry were collected from the jugular or the cephalic vein.

Hypoxemia was defined as mild (60 – 79 mmHg), marked (40 – 59 mmHg), or severe (< 40 mmHg). Hypercapnia was defined as mild (45 – 59 mmHg), marked (60 -79 mmHg) or severe (> 80 mmHg) (McDonnell and Kerr 2007).

Statistical analysis

All statistical analyses were performed with linear models in statistical software R, version 3.1.1 (R Development Core Team 2014). Variables analyzed included rectal temperature, lactate, pH, PaO_2, $PaCO_2$, induction time and recovery time. All variables were tested for normal distribution using a histogram, and the assumptions for normality were fulfilled. For both drug combinations, the difference between the first and second sample was analysed using a two-tailed paired *t*-test. An ANOVA was used to compare variables (induction time, recovery time and rectal temperature change) from the two drug combinations. *P* values less than 0.05 were considered significant. Mean±SE (range) values are presented.

RESULTS

The CX combination was used to immobilize 21 calves. One calf darted with CX required an additional dart. Induction times and immobilization levels did not differ between CX fall and CX spring dose, and data for these animals were therefore pooled. The TAX combination was used to immobilize 19 calves. Two calves required one additional dart to complete induction. These two calves were excluded from further analysis. All inductions induced with a single dart were assessed as fast and smooth inductions. There were no differences in induction times between the CX and TAX combinations ($F_{1,38} = 1.46$, $P = 0.23$) (Fig. 1), however the recovery time was significantly ($F_{1,38} = 14.49$, $P = <0.001$) faster for the TAX group (Fig. 2). Induction times and recovery times for the TAX and CX groups are presented in Table 1.

Degree of CNS depression was assessed to be level I for 18 calves and level II for one calf immobilized with TAX. None of the animals in the TAX group needed early antagonism of xylazine. The same evaluation was done for nine calves immobilized on CX, where four

calves were level I, one calf was level II and four calves were level III. Seven out of 21 animals had xylazine antagonized early in the CX group.

Blood gases were collected from 12 animals in the CX group and 15 animals in the TAX group. All calves were hypoxemic in the first blood gas measurement. In the CX group two calves were mildly hypoxemic, five calves were markedly hypoxemic and four calves were severely hypoxemic. One calf did not get a PaO_2 reading. In the TAX group three calves were mildly hypoxemic, eight calves were markedly hypoxemic and four calves were severely hypoxemic before oxygen treatment. There was no difference ($F_{1,25}$ >0.001, $P = 0.99$) between the groups in arterial oxygenation in the first sample. Both groups had a significant ($T_{1,25} = -9.68$, $P < 0.001$) increase in PaO_2 after oxygen treatment.

All calves, but two, were hypercapnic in the first blood gas measurement. In the CX group four calves were mildly hypercapnic and five calves were markedly hypercapnic. One calf was not hypercapnic and one calf did not get a $PaCO_2$ reading. In the TAX group seven calves were mildly hypercapnic and six calves were markedly hypercapnic. One calf was not hypercapnic and one calf did not get a $PaCO_2$ reading. There was no difference ($F_{1,24} = 0.06$, $P = 0.85$) in $PaCO_2$ between the groups in the first arterial sample. Both the CX group ($T_{1,10} = -2{,}555$, $P = 0.029$) and the TAX group ($T_{1,13} = -2.642$, $P = 0.020$) had a significant increase in $PaCO_2$ after oxygen treatment .

Lactate data was pooled for all caribou, as chase time is a confounding variable. Chase time was not collected consistently, and therefore not used as a variable. All animals had increased lactate in the first sample, with a value of 9.5±5.0 (1.1 - 19.5). All animals had a significant ($T_{1,25} = 4.793$, $P = <0.001$) decrease in lactate between samples.

Rectal temperature is also confounded by chase time. However, both the CX group ($T_{1,20}$ = -3.875, $P < 0.001$) and the TAX group ($T_{1,18}$ = -2.7, $P = 0.004$) had a significant increase in rectal temperature during downtime (Fig. 3). The increase did not differ between the two treatments ($F_{1,38}$= 1.83, $P = 0.18$).

All physiological variables from the CX group are presented in Table 2 and from the TAX group in Table 3.

The TAM_{high} dose was used in two captures. The first calf was dead at approach. The other calf was not breathing at approach. This calf was successfully resuscitated by immediate intravenous administration of doxapram, naltrexone and atipamezole. It was also stimulated by twisting of nose and ears, and fierce rubbing of the back.

The TAM_{low} dose was used on three captures. Degree of central nervous system depression was assessed to be level III, and medetomidine was antagonized immediately at approach. One calf stopped breathing after 5 minutes of handling time, but was also successfully resuscitated. Following these five captures, both TAM combinations were discontinued. All doses and physiological variables from TAM_{high}, TAM_{low} and captive animals are presented in Table 4.

All caribou were considered healthy, based on results from serum chemistry.

Radio tracking 2 weeks post capture detected two mortalities in the CX group and two mortalities in the TAX group. One of the mortalities in the TAX group was found eaten by lynx 5 days post capture. Carcasses from these mortalities were not available for necropsy. All mortalities are considered capture related. A field necropsy was performed on the carcass from the mortality with TAM_{high}. Histopathology confirmed aspiration pneumonia as cause of death.

DISCUSSION

This study documented use of thiafentanil-azaperone-xylazine and carfentanil-xylazine in free-ranging caribou. Further, blood gas values describing severe hypoxemia, which was easily resolved with intranasal oxygen, was documented.

TAX immobilizations were characterized by caribou remaining sternal, head upright, relaxed, spontaneous palpebral reflex, and with a minor responsiveness to tactile stimulation. All calves, but one, were in the desired level I of CNS depression, and therefore did not need early antagonism of xylazine. Correct positioning and head holding behavior is important when immobilizing ruminants to prevent bloat and aspiration pneumonia (Kreeger and Arnemo, 2012). In the CX group caribou generally seemed to be in a deeper level (level II – III) of CNS depression. This assessment was only performed on nine animals, but seven out of 21 animals were antagonized early with tolazoline in this group. In contrast with findings in other studies (Kreeger et al. 2005; Meyer et al. 2008), TAX did not offer faster induction times than CX. However, the recovery time was significantly shorter for the TAX group than the CX group. This has also been documented in Shiras moose (Kreeger et al. 2005) and pronghorn antelope (*Antilocapra americana*) (Kreeger et al. 2001). These observed differences between carfentanil and thiafentanil are probably related to individual pharmacological potency.

In conscious and healthy animals, the partial pressure of arterial oxygenation (PaO_2) is expected to be between 80 and 100 mmHg (Steffey 2001; Fahlman 2014). In this study both groups were hypoxemic before oxygen supplement with a mean PaO_2 value of 46 mmHg, which borderlines severe hypoxemia. This was not unexpected, based on results from previous blood gas studies on reindeers immobilized with other drug combinations (Risling et al. 2011; Fahlman et al. 2012; Evans et al. 2013). Hypoxemia leads to hypoxia which can

have devastating results with tissue necrosis on vital tissues including brain, heart, kidneys and liver, ending in organ failure (Caulkett et al. 1994; Fahlman 2014). Morbidity from hypoxia has been a neglected area in wildlife anesthesia. However, there are a few documented effects of behavior change after chemical immobilizations (Ramsay and Stirling 1986; Cattet et al. 2008; Rode et al. 2014). Mountain goats were found to be more likely to abandon their kids after chemical immobilization than with physical immobilization (Cotè et al. 1998). Moose that had been chemically immobilized were found to have lower calf survival during a November moose census compared with moose that had not been chemically immobilized (Larsen and Gauthier 1989). Hypoxia affects all vital organs, but the cerebral cortex is especially vulnerable, and changes in cerebral function are recognized as an early sign of hypoxia in humans (Steffey 2001). The hypoxemia documented in this study was resolved with intranasal oxygen supplement.

The increase in $PaCO_2$ documented in this study was not unexpected, as it is a known side effect of both oxygen therapy and opioid induced hypoventilation (Schumacher et al. 1997; Paterson et al. 2009; Risling et al. 2011; Evans et al. 2013; Lian et al. 2014). Ventilation is mainly controlled by central chemoreceptors in the medulla sensitive to pH and $PaCO_2$ and peripheral chemoreceptors in the aortic and carotid bodies sensitive to pH and PaO_2. During chemical immobilization the chemo receptors are depressed by opioids and sedatives, resulting in hypoventilation. Prolonged hypercapnia can lead to direct depression of cardiac contractility and a depressant effect on myogenic activity in the vascular system (Steffey 2001; McDonell and Kerr, 2007). To minimize hypercapnia it is important to keep oxygen flow rates minimal, and perform oxygen titration studies for new species receiving oxygen. Other measures against hypercapnia is short handling times, or endotracheal intubation and intermittent positive-pressure ventilation for longer handling operations (Hartsfield; 2007).

Both groups had a significant decrease in lactate between samples. The excitement and helicopter pursuit leads to an increased production of lactic acid in skeletal muscles. Lactic acid acidifies the blood, with a subsequent metabolic acidosis (Haga et al. 2009). Hypoxemia and subsequent muscle hypoxia can also produce lactic acid resulting in metabolic acidosis. Even though muscle tissue is one of the more robust tissues, the severe PaO_2 values found in this study may have contributed to the high lactate levels (Steffey 2001; McDonell and Kerr 2007). Similar values have been documented in chemically immobilized wild reindeer and moose, which also had a significant decrease during handling time (Haga et al. 2009; Evans et al. 2012; Evans et al. 2013; Lian et al. 2014).

The normal body temperature in resting reindeer is 38.1-38.6 C (Blix et al. 2011). With helicopter pursuit, even though kept short, body temperatures are expected to increase due to the physical strain. Additionally, the anesthetic drugs interfere with the animal's thermoregulation (Harthoorn and Walt 1974; Evans et al. 2013; Ko and Krimins 2014). In our study hyperthermia was present in both groups with mean values >40 C. Capture related hyperthermia is not uncommon, especially in rangifer, a species adapted to an arctic climate (Blix et al. 2011; Evans et al., 2013). Both treatment groups had a significant increase in temperature during handling time. This can possibly be descriptive of a temperature that will keep increasing post recovery, as described in other ungulate species immobilized with potent opioids (Meyer et al. 2008). There was a trend of higher increase in the CX group than the TAX group, which can be related to different pharmacological potency.

The TAM combination was not an acceptable drug combination for free-ranging caribou calves at the doses tested. The combination resulted in one mortality and two respiratory arrests. Medetomidine is a potent alpha-2 agonist, with side effects including respiratory depression, aspiration pneumonia and decreased gastrointestinal motility. These

side effects are reinforced in combination with a potent opioid (Haigh 1990; Read 2003). The mortality with TAM_{high} was due to aspiration pneumonia, most likely a direct cause of the high medetomidine dose.

This study found TAX to be an efficient drug combination for free-ranging caribou calves. The doses tested in this study can be used as an alternative to the current CX combination. Thiafentanil should not be used in combination with medetomidine for immobilizing free-ranging caribou calves. The study also documented severe hypoxemia with both drug combinations. Hypoxemia was resolved with intranasal oxygen at a flow rate of 1 L/min. For chemical immobilizations, we recommend keeping handling times to a minimum and using intranasal oxygen supplement. More research is needed on long term effects from chemical immobilizations that have been conducted without oxygen supplement.

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Table 1. time from darting to recumbency (induction time), time from recumbency to approach and handling the animal (capture time), time from darting until antagonists were administered (handling time) and time from administration of antagonist to standing (recovery time) in free-ranging caribou (*Rangifer tarandus granti*) darted from helicopter with carfentanil-xylazine (CX) or thiafentanil-azaperone-xylazine (TAX) during fall 2013 and spring 2014 in Alaska, USA. Mean ± SE (range) values are presented.

Variable	***Units***	**CX**	**TAX**
Induction time	Minutes	3.2±1.4 (1.5 - 7.0)	3.6±1.6 (2.0 - 7.5)
Capture time	Minutes	1.9±2.2 (0.25 - 5.0)	2.8±1.3 (2 - 5)
Handling time	Minutes	31.0±5.6 (21 - 40)	31.6±5.1 (26 - 44)
Recovery time	Seconds	220±85 (120 - 420)	133±67 (60 - 375)

Table 2. Physiological variables from free-ranging caribou (*Rangifer tarandus granti*), during chemical immobilization with carfentanil-xylazine (CX), delivered by dart syringe from helicopter, during fall 2013 and spring 2014 in Alaska, USA. Mean ± SE (range) values are presented for all variables. *Significant difference after oxygen treatment. T1 is time from darting to collection of first sample. T2 is time from darting to second sample.

			CX		**CX**	
			Before O_2		**After 10 min of O_2**	
Variable	**Unit**	N	T1: 6 - 15 minutes	N	T2: 19 - 31 minutes	*P* value
Rectal temp	C˚	12	40.5±0.5 (39.7-41.2)	12	40.8±0.6 (40.0-41.8)	0.106
Pulse	Beat/min	9	52±8 (40-60)	9	50±15 (32-80)	0.301
Respiratory rate	Breaths/min	12	22±9 (8-40)	12	18±7 (8-28)	0.064
pH		11	7.21±0.1 (6.99-7.40)	11	7.23±0.2 (6.98-7.56)	0.254
$PaCO_2$	mmHg	11	58.0±9.2 (42.8-69.1)	11	65.6±11.2 (41.3-80.9)	0.029*
PaO_2	mmHg	11	46±13 (33-74)	11	96±17 (60-115)	<0.001*
Lactate	mmol/L	12	10.8±5.9 (1.1-19.5)	12	7.8±5.1 (1.3-16.6)	<0.001*

Table 3. Physiological variables from free-ranging caribou (*Rangifer tarandus granti*), during chemical immobilization with thiafentanil-azaperone-xylazine (TAX), delivered by dart syringe from helicopter during fall 2013 and spring 2014 in Alaska, USA. Mean ± SE (range) values are presented for all variables. *Significant difference after oxygen treatment. T1 is time from darting to collection of first sample. T2 is time from darting to second sample.

			TAX		**TAX**	
			Before O_2		**After 10 min of O_2**	
Variable	**Unit**	N	T1: 6 - 13 minutes	N	T2: 19 - 27 minutes	*P* value
Rectal temp	C˚	16	40.2±0.5 (39.0-40.8)	16	40.5±0.6 (39.4-41.4)	<0.001*
Pulse	Beat/min	12	54±10 (40-68)	12	52±11 (28-66)	0.492
Respiratory rate	Breaths/min	10	17±10 (8 - 40)	10	15±7 (6 - 24)	0.317
pH		14	7.25±0.07 (7.06-7.36)	14	7.28±0.08 (7.10-7.37)	0.024*
$PaCO_2$	mmHg	14	57.0±10.1 (35.8-73.1)	14	63.8±11.1 (52.9-83.3)	0.020*
PaO_2	mmHg	15	46±13 (24-74)	15	96±21 (61-132)	<0.001*
Lactate	mmol/L	15	8.4±3.9 (2.1-15.5)	15	5.3±3.1 (1.5-11.4)	<0.001*

Table 4. Drug doses and physiological variables collected from captive and free-ranging caribou (*Rangifer tarandus granti*), during chemical immobilization with thiafentanil-azaperone-medetomidine combinations. Caribou in a captive facility were hand injected (hi) or ground darted (d) in September 2013 and free-ranging caribou were darted from helicopter in October 2013, in Alaska, USA. Mean ± SE (range) values are presented for all variables. *Not temperature corrected values.

Age, body weight kg Captive	**Thiafentanil** mg (mg/kg)	**Azaperone** mg (mg/kg)	**Medetomidine** mg (mg/kg)	**Level immobilization** (1-5)	**Overall**	**Flow rate** L/min	**Oxygen**	**RR** brths/min	**PaO_2** mmHg	**$PaCO_2$** mmHg	**lactate** mmol/L
yearling, 91[hi]	2.73 (0.03)		5 (0.055)	4	poor	2	Before O_2	8	29	61.5	12.6
							With O_2	4	144	94.2	10.6
yearling, 89[hi]	1.46 (0.0164)		4 (0.045)	3	excellent	1	Before O_2	20	44	56	8.7
							With O_2	10	131	66.4	3.6
yearling, 95[hi]	1.6 (0.0168)		4 (0.042)	3	good	1	BeforeO_2	28	48	56.6	10.4
							With O_2	28	82	65.8	5.6
yearling, 87[hi]	1.6 (0.0183)	25 (0.29)	4 (0.046)	3	poor	1	Before O_2	40	37	57.7	14.3
							With O_2	N/R	53	92.9	7.9
yearling, 82[hi]	1.5 (0.0183)	24.5 (0.29)	3.8 (0.046)	3	excellent	1	BeforeO_2	N/R	38	60.2	10.4
							With O_2	12	115	83.5	5
adult, 109[d]	3.1 (0.028)	25 (0.23)		2	poor	1	Before O_2	28	52*	31.3*	4.2
							With O_2	N/R	90*	24.2*	2.1
adult, 100[d]	3 (0.03)	25 (0.25)		2	poor	2	BeforeO_2	16	61	53	7.7

							With O_2	24	N/R	N/R	N/R
Free-ranging											
5 months, 64[d]	1.5 (0.023)	25 (0.39)	5 (0.078)	dead	poor		N/A				
							N/A				
5 months, N/R[d]	1.5	25	5	5	poor		N/A	0			
							N/A	breathing			
5 months, N/R[d]	1.5	25	2	4	poor	1	Before O_2	2	52	46	5.5
							With O_2	8	N/R	N/R	N/R
5 months, 57[d]	1.5(0.026)	25 (0.44)	2 (0.035)	3	good	1	Before O_2	30	34	64.9	16.8
							With O_2	22	88	80.8	13.3
5 months, N/R[d]	1.5	25	2	5	poor	1	Before O_2	24			
							With O_2	0			

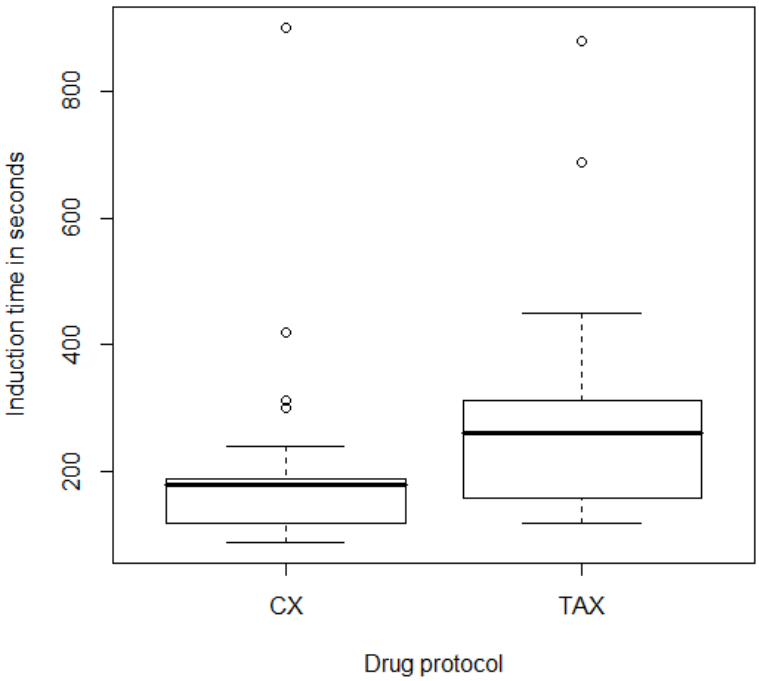

FIGURE 1. Induction times for free-ranging caribou (*Rangifer tarandus granti*) calves immobilized with carfentanil-xylazine (CX) or thiafentanil-azaperone-xylazine (TAX) during fall 2013 and spring 2014 in Alaska, USA, were not significantly different ($F_{1,38} = 1.46$, $P = 0.23$).

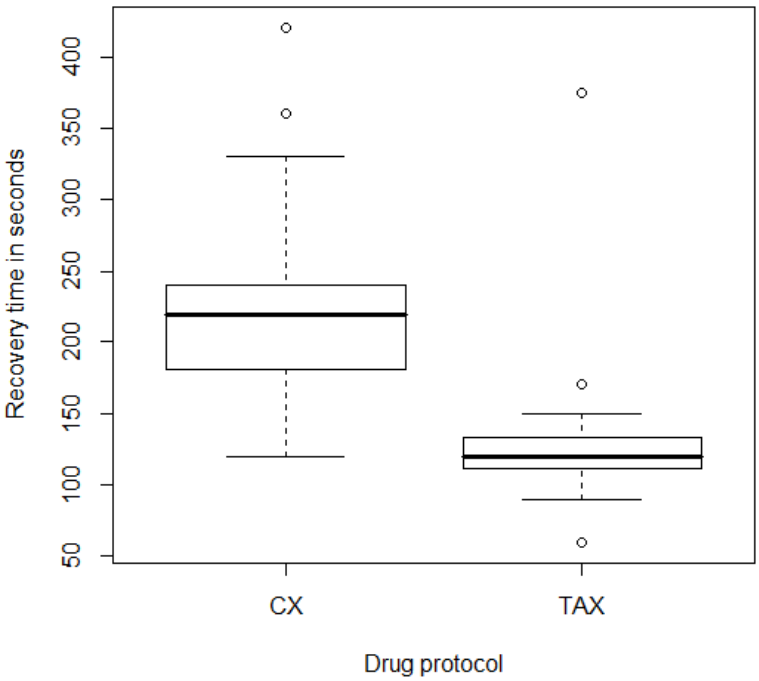

FIGURE 2. Recovery times were significantly ($F_{1,38}$ = 14.49, P = <0.001) shorter for caribou (*Rangifer tarandus granti*) calves immobilized with thiafentanil-azaperone-xylazine (TAX) than with carfentanil-xylazine (CX), during fall 2013 and spring 2014 in Alaska, USA.

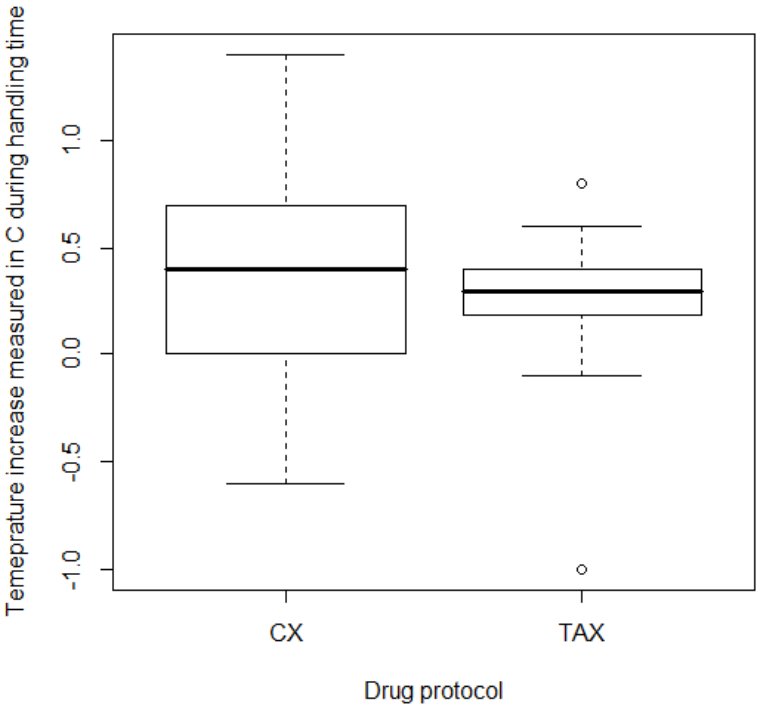

FIGURE 3. There was a significant increase in temperature during handling time for caribou (*Rangifer tarandus granti*) calves immobilized with thiafentanil-azaperone-xylazine (TAX) and carfentanil-xylazine (CX) during fall 2013 and spring 2014 in Alaska, USA.