A randomized clinical trial to compare ketamine-butorphanol-azaperone-medetomidine and detomidine-etorphine-acepromazine for anesthesia of captive Przewalski horses (*Equus przewalskii*)

Ellie L. Milnes, VetMB, DVSc, DACZM, DECZM1,2,*; Alicia M. Skelding, DVM, DVSc, DACVAA3; Cédric B. Larouche, DVM, DVSc, DACZM2; Angelica Ferro, DVM3; Pauline Delnatte, DMV, DVSc, DACZM, DECZM2; Christopher Dutton, BVSc, MSc, DACZM, DECZM2; Neil E. Anderson, BVSc, PhD2

1Toronto Zoo, Toronto, ON, Canada
2The Royal (Dick) School of Veterinary Studies and the Roslin Institute, University of Edinburgh, Midlothian, UK
3Toronto Animal Health Partners Emergency and Specialty Hospital, Toronto, ON, Canada
*Corresponding author: Dr. Milnes (emilnesdvm@yahoo.co.uk)

https://doi.org/10.2460/ajvr.21.10.0165

OBJECTIVE
To compare ketamine-butorphanol-azaperone-medetomidine (KBAM) to detomidine-etorphine-acepromazine (DEA) for field anesthesia in captive Przewalski horses (*Equus przewalskii*).

ANIMALS
10 adult Przewalski horses.

PROCEDURES
A prospective randomized crossover trial was conducted. Each horse was immobilized once with KBAM (200 mg ketamine, 109.2 mg butorphanol, 36.4 mg azaperone, and 43.6 mg medetomidine) and once with DEA (40 mg detomidine premedication, followed 20 minutes later by 3.9 to 4.4 mg etorphine and 16 to 18 mg acepromazine). Both protocols were administered by IM remote dart injection with a washout period of 6 months between treatments. Selected cardiorespiratory variables and quality of anesthesia were recorded. Antagonists were administered IM (KBAM, 215 mg atipamezole and 50 mg naltrexone; DEA, 4 mg RX821002 and 100 mg naltrexone).

RESULTS
All horses were anesthetized and recovered uneventfully. Inductions (DEA, 6.8 min; KBAM, 11.6 min; *P* = 0.04) and recoveries (DEA, 3.2 min; KBAM, 19.6 min; *P* < 0.01) were faster with DEA compared with KBAM. Quality scores for induction and recovery did not differ between protocols, but maintenance quality was poorer for DEA (*P* < 0.01). Clinical concerns during DEA immobilizations included apnea, severe hypoxemia (arterial partial pressure of oxygen < 60 mm Hg), muscle rigidity, and tremors. Horses treated with KBAM were moderately hypoxemic, but arterial partial pressures of oxygen were higher compared with DEA (*P* < 0.01).

CLINICAL RELEVANCE
Captive Przewalski horses are effectively immobilized with KBAM, and this protocol results in superior muscle relaxation and less marked hypoxemia during the maintenance phase, but slower inductions and recoveries, compared with DEA.

The ultrapotent opioid (UPO) etorphine is the drug most commonly used to immobilize nondomestic equids because of its potency, rapid onset of action, reliability, reversibility, and the historic lack of availability of efficacious alternative protocols. However, adverse effects of UPOs include respiratory depression, hypertension, muscle tremors, renarcotization, and the risk of accidental human exposure. Etorphine is usually administered in combination with a tranquilizer or sedative to reduce the etorphine dose required to achieve a working level of immobilization and to mitigate UPO-associated
adverse effects. Drug administration is usually by remote IM injection in nondomestic equids. A typical immobilization protocol for both wild free-ranging and captive Przewalski horses combines etorphine with butorphanol and detomidine in a single dart. At the Toronto Zoo, a 2-step strategy consisting of premedication with detomidine IM followed by etorphine-acepromazine IM, all by remote dart injection, has historically been the immobilization protocol of choice for Przewalski horses.

Some countries have experienced periodic shortages in etorphine supply, prompting zoo veterinarians to seek alternatives for nondomestic equid immobilization. The UPOs carfentanil and thifentanil have been trialed with variable success. Several non-UPO combinations have been reported in nondomestic equids managed in zoological institutions. Standing sedation is relatively easily achieved with non-UPO combinations such as butorphanol-detomidine, but, as in domestic horses, additional drugs (normally ketamine) are required to induce recumbency. Combinations of $\alpha_2$-adrenergic agonists (such as medetomidine, xylazine, and romifidine) and ketamine or tiletamine-zolazepam have been used to induce recumbent immobilization in captive nondomestic equids. Medetomidine-ketamine (MK) combinations reported in the zoological medicine literature have variable efficacy in Przewalski horses with some individuals showing substantial ataxia but failing to become recumbent, whereas others underwent calm induction, excellent muscle relaxation, and uneventful recovery. In boma-confined wild zebra (Equus zebra), ketamine-butorphanol-medetomidine (KBAM) was as effective as etorphine-azapente, but adverse effects of KBM included ataxic and prolonged inductions, hypertension, and hypoxemia.

The non-UPO combination butorphanol-azaperone-medetomidine (BAM) effectively immobilizes a wide range of nondomestic ungulate species, although reported adverse effects include prolonged inductions and hypoxemia. Ketamine is added to BAM for the immobilization of nondomestic equids. The aim of the study reported here was to characterize and compare the quality of induction, maintenance, and recovery and the cardiorespiratory effects of 2 drug protocols, ketamine-butorphanol-azaperone-medetomidine (KBAM) and detomidine-etorphine-acepromazine (DEA), administered by remote IM injection via dart, for the immobilization of captive Przewalski horses. It was hypothesized that KBAM would result in greater muscle relaxation and higher arterial partial pressure of oxygen ($Pao_2$) than DEA.

### Materials and Methods

Research methodology was approved by the Toronto Zoo Animal Care & Research Committee (reference No. 2018-09-19) and the Royal (Dick) School of Veterinary Studies Veterinary Ethical Review Committee (reference No. 81.89).

### Animals

Ten captive-born Przewalski horses were immobilized in April and October of 2019 for routine veterinary care at the Toronto Zoo. Seven mares, 2 geldings, and 1 stallion (estimated average body weight of 500 kg) were included in the study; all horses were healthy.

### Study design

This was a randomized, prospective, crossover clinical trial. Each horse received treatments KBAM and DEA in random order. The order in which horses received the 2 treatments was allocated using an online randomization plan generator, in which the 10 subjects were randomized into 1 block with balanced permutations. Initial treatment allocation was concealed from the investigators during the randomization process, but no further blinding was performed during the animal phase because of the human safety concerns associated with exposure to etorphine and concentrated medetomidine. Animals received the first treatment in April 2019, and the alternative treatment in October 2019. The 6-month washout period coincided with planned routine immobilizations for scheduled veterinary care.

Sample size was calculated by power based on $Pao_2$ values obtained by arterial blood gas analysis. A clinically significant effect size of 15 mm Hg and SD of 10 mm Hg were derived from prior clinical experiences of the authors. A power calculation was performed with power of 80% and a type I error of 5%, giving a sample size of 8 horses. An attrition rate of 20% was anticipated, so a final sample size of 10 horses was used.

### Immobilization

Immediately before darting, each horse was separated from the herd and moved to a 4 X 8-meter outdoor pen accessed by sliding doors. Ambient temperatures ranged from 9 °C to 16 °C. Horses were randomly assigned to a treatment protocol. For the DEA protocol, horses were premedicated with 40 mg detomidine delivered IM by remote dart, followed 20 minutes later by 1.6 to 1.8 mL of a premixed product containing etorphine HCl (2.45 mg/mL) and acepromazine maleate (10 mg/mL), compounded (Chiron Compounding Pharmacy Inc) from existing approved drug products with benzyl alcohol 0.5% preservative. For the KBAM protocol, horses received 200 mg ketamine combined with 4 mL of a premixed product containing butorphanol tartrate (27.3 mg/mL), azapente (9.1 mg/mL), and medetomidine HCl (10.9 mg/mL), compounded (Chiron Compounding Pharmacy Inc) from existing approved drug products and with inactive ingredients National Formulary (NF) methylparaben powder, NF tartaric acid powder, United States Pharmacopeia (USP) citric acid anhydrous fine granular granules, USP sodium citrate anhydrous powder, and benzethonium chloride powder. All IM injections were administered via dart injection using a gas-powered pistol. Detomidine and KBAM were administered using 5-mL plastic darts, and...
A 2-mL plastic dart, all with a 16-gauge, 38-mm, wire-barbed, side-ported, collared needle. Animals were darted from a distance of 3 to 5 meters into the gluteal muscles. The time elapsed from darting with KBAM or etorphine-acepromazine until the horse became recumbent was recorded as induction time.

Once recumbent, heart rate (HR), respiratory rate (RR), and body temperature were monitored by palpation of peripheral pulse or stethoscope auscultation, visual count of thoracic wall excursions, and digital rectal thermometer, respectively. Physiologic variables were recorded at 5, 10, and 15 minutes after recumbency (T5, T10, and T15). Arterial blood samples of 2 mL were collected anaerobically at T5 and again at T15 by aseptically puncturing the lateral metatarsal artery of the nondependent pelvic limb or the transverse facial artery with a 22-gauge needle attached to a heparinized syringe. Arterial blood was immediately analyzed for \( \text{Pa}_2 \), arterial partial pressure of carbon dioxide (\( \text{PaCO}_2 \)), arterial oxygen saturation of hemoglobin (\( \text{SaO}_2 \)), \( \text{pH} \), \( \text{HCO}_3^- \), and base excess (BE) using a hand-held analyzer (i-STAT; Abbott Laboratories) at 37°C. Supplemental oxygen was administered via a nasal cannula inserted into the ventral meatus of 1 nostril from a portable oxygen canister at a flow rate of 15 L/min, starting immediately after collection of the T5 arterial sample. During each immobilization horses underwent the following procedures: physical examination, venipuncture for blood collection, and prophylactic treatments as required (eg, vaccinations) performed by veterinarians, and hoof trimming performed by certified farriers.

Following completion of the above procedures, antagonist drugs were administered IM by hand injection and horses were left to stand unassisted. For the DEA protocol, RX821002 was used to antagonize detomidine (0.1 mg RX821002 per mg detomidine) and naltrexone was used to antagonize etorphine (20 mg naltrexone per mg etorphine). RX821002 was compounded in-house from bulk substance (RX821002 hydrochloride powder reconstituted with sterile water to 2.5 mg/mL; Sigma-Aldrich). For the KBAM protocol, medetomidine was reversed with atipamezole (5 mg per mg medetomidine) and butorphanol was reversed with naltrexone (0.5 mg per mg butorphanol).

Video recordings were made of the induction, maintenance, and recovery phases, and scored at a later date by a blinded observer using the scoring system of Matthews et al.7 Horses were kept under observation until fully recovered, at which point they were reintroduced to the herd.

**Statistical analysis**

Data were analyzed by use of statistical software (R version 3.6.2; R Foundation for Statistical Computing), and graphics were constructed.16,17 Data from horses that required additional drugs during the induction period (redarting or a ketamine bolus) were excluded from further analysis. For continuous data, distribution was assessed for normality using visual inspection of histograms, Q-Q plots, and performing the Shapiro-Wilk test. Results were presented as mean ± SD and sample range for continuous data, and as median and interquartile range for discrete data. Results were interpreted at the 95% level of confidence (\( P \leq 0.05 \)). Data were tabulated, summary statistics determined, and induction time compared between combinations using a paired 2-sample \( t \)-test. Data were not available for recovery times from October 2019 because of human error; therefore, recovery times were analyzed from 10 horses (5 DEA and 5 KBAM) in April 2019 using an unpaired 2-sample \( t \)-test. Subjective quality scores had a nonparametric distribution, so the Friedman test was used to compare scores between combinations (response: score; treatment: protocol; and block: horse).

Descriptive statistics for quantitative continuous data (HR, RR, body temperature, \( \text{SaO}_2 \), \( \text{PaCO}_2 \), \( \text{PaO}_2 \), \( \text{pH} \), \( \text{HCO}_3^- \), and BE) were tabulated at each time point and checked to identify any missing values and recording errors. Data were plotted to visually check for differences between and within protocols over time. Linear mixed-model analyses were implemented.18 The explanatory variables “time [from onset of recumbency]” and “protocol” were selected on the basis of biological plausibility and clinical relevance and included as fixed effects in the full model.19 “Horse ID” was included as a random effect to control for the similarity between repeated measures within an immobilization procedure. Separate multivariable models were created for each response variable. The fit of all models was examined by evaluating the residuals to ensure no violation of the model’s assumptions. This modeling strategy controlled for the repeated measurements within an immobilization procedure and also allowed for a direct effect of protocol on the value of the monitored physiologic variables as well as an effect of protocol on changes in monitored physiologic variables over time.20 The software package18 used for linear mixed-model analyses did not provide \( P \) values for variables, so these were generated using the Wald \( t \) values provided in the model output using the ANOVA function in a different software package.21 These \( P \) values were used to evaluate the significance of fixed effects in the models obtained.22 The standard errors of fixed-effects coefficients were used to construct approximate Wald confidence intervals.23

**Results**

**Animals**

Of the 10 horses enrolled in the study, 9 received both treatments in the randomized crossover study. One mare allocated to KBAM at the first trial was subsequently mated and was in early pregnancy at the time of the second trial; based on the preliminary finding of more severe hypoxemia in horses immobilized with DEA and concerns about fetal viability,
Table 1—Mean ± SD (range) results for physiologic variables and arterial blood gas analysis at 5, 10, and 15 minutes after recumbency in 10 individual, healthy adult captive Przewalski horses (Equus przewalskii) immobilized with either a ketamine (200 mg), butorphanol (109.2 mg), azaperone (36.4 mg), medetomidine (43.6 mg) protocol (KBAM), or a detomidine (40 mg; premedication), etorphine (3.9 to 4.4 mg), and acepromazine (16 to 18 mg) protocol (DEA) delivered IM by remote dart.

<table>
<thead>
<tr>
<th>Variable/Protocol</th>
<th>5 minutes</th>
<th>10 minutes</th>
<th>15 minutes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart rate (beats/min)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>KBAM</td>
<td>37 ± 6</td>
<td>(23 to 44)</td>
<td>40 ± 7</td>
</tr>
<tr>
<td>DEA</td>
<td>58 ± 19</td>
<td>(32 to 84)</td>
<td>53 ± 12</td>
</tr>
<tr>
<td>Respiratory rate (breaths/min)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>KBAM</td>
<td>19 ± 10</td>
<td>(6 to 36)</td>
<td>19 ± 12</td>
</tr>
<tr>
<td>DEA</td>
<td>8 ± 7</td>
<td>(4 to 24)</td>
<td>6 ± 3</td>
</tr>
<tr>
<td>Rectal temperature (° C)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>KBAM</td>
<td>38.9 ± 0.5</td>
<td>(38.2 to 39.7)</td>
<td>39.0 ± 0.7</td>
</tr>
<tr>
<td>DEA</td>
<td>39.2 ± 0.4</td>
<td>(38.6 to 39.6)</td>
<td>39.2 ± 0.4</td>
</tr>
<tr>
<td>Blood pH</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>KBAM</td>
<td>7.41 ± 0.05</td>
<td>(7.34 to 7.48)</td>
<td>–</td>
</tr>
<tr>
<td>DEA</td>
<td>7.35 ± 0.06</td>
<td>(7.26 to 7.43)</td>
<td>–</td>
</tr>
<tr>
<td>PaO₂ (mm Hg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>KBAM</td>
<td>47.2 ± 11.8</td>
<td>(26 to 62)</td>
<td>–</td>
</tr>
<tr>
<td>DEA</td>
<td>27 ± 9</td>
<td>(14 to 44)</td>
<td>–</td>
</tr>
<tr>
<td>SaO₂ (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>KBAM</td>
<td>80 ± 14</td>
<td>(47 to 93)</td>
<td>–</td>
</tr>
<tr>
<td>DEA</td>
<td>45 ± 22</td>
<td>(12 to 81)</td>
<td>–</td>
</tr>
<tr>
<td>PaCO₂ (mm Hg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>KBAM</td>
<td>41.9 ± 4.4</td>
<td>(36.3 to 48.7)</td>
<td>–</td>
</tr>
<tr>
<td>DEA</td>
<td>55.7 ± 11.4</td>
<td>(38.6 to 78.9)</td>
<td>–</td>
</tr>
<tr>
<td>BE (mmol/L)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>KBAM</td>
<td>2.1 ± 2.3</td>
<td>(−2.0 to 5.0)</td>
<td>–</td>
</tr>
<tr>
<td>DEA</td>
<td>5.1 ± 4.8</td>
<td>(−3.0 to 12)</td>
<td>–</td>
</tr>
<tr>
<td>HCO₃⁻ (mmol/L)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>KBAM</td>
<td>26.69 ± 2.0</td>
<td>(23.4 to 29.3)</td>
<td>–</td>
</tr>
<tr>
<td>DEA</td>
<td>30.8 ± 4.8</td>
<td>(23.6 to 36.7)</td>
<td>–</td>
</tr>
</tbody>
</table>

— = Not available, no sample collected. BE = Base excess. PaO₂ = Partial pressure of arterial carbon dioxide. PaCO₂ = partial pressure of arterial oxygen. SaO₂ = Arterial oxygen saturation of hemoglobin.

Table 2—Multivariable analysis using linear mixed-effects models for the effect of protocol (KBAM or DEA) and time (from onset of recumbency) on physiologic variables and arterial blood gas analysis for the Przewalski horses described in Table 1.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Estimate</th>
<th>SE</th>
<th>95% CI (Wald)</th>
<th>P value (Wald)</th>
<th>Estimate</th>
<th>SE</th>
<th>95% CI (Wald)</th>
<th>P value (Wald)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart rate (beats/min)</td>
<td>−17.5</td>
<td>2.48</td>
<td>−22.33 to −12.62</td>
<td>&lt; 0.01*</td>
<td>−0.11</td>
<td>0.27</td>
<td>−0.64 to 0.42</td>
<td>0.68</td>
</tr>
<tr>
<td>Respiratory rate (breaths/min)</td>
<td>11.3</td>
<td>1.92</td>
<td>7.54 to 15.07</td>
<td>&lt; 0.01*</td>
<td>−0.27</td>
<td>0.22</td>
<td>−0.69 to 0.16</td>
<td>0.22</td>
</tr>
<tr>
<td>Temperature (° C)</td>
<td>−0.43</td>
<td>0.14</td>
<td>−0.70 to −0.16</td>
<td>&lt; 0.01*</td>
<td>−0.006</td>
<td>0.015</td>
<td>−0.03 to 0.02</td>
<td>0.71</td>
</tr>
<tr>
<td>Blood pH</td>
<td>0.04</td>
<td>0.02</td>
<td>0.008 to 0.074</td>
<td>0.01*</td>
<td>0.0003</td>
<td>0.002</td>
<td>−0.0027 to 0.0034</td>
<td>0.84</td>
</tr>
<tr>
<td>PaO₂ (mm Hg)</td>
<td>24.4</td>
<td>4.94</td>
<td>14.74 to 34.09</td>
<td>&lt; 0.01*</td>
<td>1.09</td>
<td>0.47</td>
<td>0.17 to 2.00</td>
<td>0.02*</td>
</tr>
<tr>
<td>PaCO₂ (mm Hg)</td>
<td>−11.8</td>
<td>2.61</td>
<td>−16.87 to −6.66</td>
<td>&lt; 0.01*</td>
<td>0.12</td>
<td>0.26</td>
<td>−0.38 to 0.63</td>
<td>0.63</td>
</tr>
<tr>
<td>BE (mmol/L)</td>
<td>−3.1</td>
<td>1.15</td>
<td>−5.32 to −0.79</td>
<td>0.01*</td>
<td>0.11</td>
<td>0.11</td>
<td>−0.11 to 0.34</td>
<td>0.99</td>
</tr>
<tr>
<td>HCO₃⁻ (mmol/L)</td>
<td>−3.9</td>
<td>1.11</td>
<td>−6.10 to −1.75</td>
<td>&lt; 0.01*</td>
<td>0.10</td>
<td>0.11</td>
<td>−0.11 to 0.32</td>
<td>0.34</td>
</tr>
</tbody>
</table>

*Indicates a statistically significant (P < 0.05) effect. See Table 1 for key.

KBAM was used instead of DEA at the second trial for this individual. Therefore, data from 2 KBAM immobilizations were analyzed for this mare. The remaining 9 horses received both protocols in random order; all horses recovered uneventfully from each anesthetic event. Two pregnant mares were immobilized with KBAM at approximately 12 weeks of gestation and subsequently birthed live foals at term.

**Immobilization Induction**

All horses were agitated prior to and immediately following the initial dart. At 20 minutes after premedication with detomidine in the DEA group, all animals reached a moderate level of standing sedation and responded minimally to the etorphine-acepromazine dart. The mean ± SD induction time

Unauthenticated | Downloaded 05/29/22 03:49 AM UTC
was slower ($P = 0.04$) for horses treated with KBAM (11.6 ± 4.2 min; range, 7 to 21 min) versus DEA (6.8 ± 2.4 min; range, 4 to 11 min).

The induction quality score did not differ between the protocols ($P = 0.16$). However, there were notable differences in induction behavior between horses treated with KBAM and DEA. During KBAM inductions, animals were moderately to severely ataxic with multiple episodes of stumbling and almost falling before becoming recumbent. Penile prolapse during induction was a consistent finding in the stallion and geldings darted with KBAM but not with DEA. During DEA induction, horses tended to stand still with their head down or head pressing against the pen wall, then developed muscle tremors before falling heavily into lateral recumbency.

One animal in each group required supplemental induction drugs because they were sedated but remained standing following administration of the standard protocol; data from these immobilizations were excluded from subsequent analysis. In the DEA group, 1 horse received additional etorphine-acepromazine by IM dart injection, and in the KBAM group, 1 horse was administered additional ketamine by IV injection. Both horses became recumbent following supplementation. Overall, data from 10 KBAM and 8 DEA immobilizations were analyzed.

**Maintenance**

Most DEA horses showed severe muscle rigidity and tremors and spontaneous limb movements during the maintenance phase. In contrast, the KBAM maintenance phase was smooth and relaxed. Relaxation scores were significantly ($P < 0.01$) higher (ie, poorer muscle relaxation) for DEA than KBAM. One horse treated with DEA spontaneously stood up at T10; the horse was carefully approached, blindfolded, given an IV ketamine bolus, and became recumbent again with no further attempts to stand prior to administration of reversal agents.

**Recovery**

The mean ± SD recovery time was slower ($P < 0.01$) for horses treated with KBAM (19.6 ± 6.9 min; range, 2 to 30 min) compared with DEA (3.2 ± 0.8 min; range, 1 to 4 min). Recovery quality scores did not differ between DEA and KBAM ($P = 0.41$). All horses were unrestrained during recovery and stood at their first attempt; recoveries were quiet and uneventful. In the stallion and geldings immobilized with KBAM, penile prolapse persisted during the recovery period but resolved within 6 hours with no complications. No renarcotization was

---

**Figure 1**—Results for physiologic variables recorded at 5, 10, and 15 minutes after recumbency in 10 individual healthy adult captive Przewalski horses (*Equus przewalskii*) immobilized with either a ketamine (200 mg), butorphanol (109.2 mg), azaperone (36.4 mg), and medetomidine (43.6 mg) protocol (KBAM; black dashed lines) or a detomidine (40 mg; premedication), etorphine (3.9 to 4.4 mg), and acepromazine (16 to 18 mg) protocol (DEA; gray solid lines) delivered IM by remote dart. Results of linear mixed-model analyses indicated that protocol had a significant ($P < 0.05$) effect on all physiologic variables, whereas time from onset of recumbency only had a significant ($P = 0.02$) effect on the arterial partial pressure of oxygen ($PaO_2$; asterisk). $PaCO_2$ = Partial pressure of arterial carbon dioxide. $SaO_2$ = Arterial oxygen saturation of hemoglobin.
observed, and there was no study-associated morbidity or death.

Physiologic variables

Heart rates were faster and more variable ($P < 0.01$) and respiratory rates were slower ($P < 0.01$) in horses treated with DEA compared with KBAM (Table 1). Apnea of 30 seconds or longer occurred in 6 of the 8 horses treated with DEA but was not observed when the horses were treated with KBAM. Rectal temperatures were significantly ($P < 0.01$) higher for horses treated with DEA (Table 2) but remained within clinically acceptable limits. There was no evidence for any change in HR, RR, temperature, pH, PaCO$_2$, HCO$_3^-$, or BE as an effect of time (Figure 1).

Arterial blood gases at T5 and T15 revealed hypoxemia that was moderate (PaO$_2$ < 80 mm Hg) to severe (PaO$_2$ < 60 mm Hg) in most horses (Figure 2). Horses immobilized with DEA were more severely hypoxic than with KBAM ($P < 0.01$) (Table 2). Normocapnia and normal blood pH balance were recorded in all horses. Although horses treated with DEA had higher PaCO$_2$, HCO$_3^-$, and BE ($P < 0.01$) and lower pH values ($P = 0.01$) compared with KBAM horses, these differences were within clinically acceptable limits.

Discussion

Przewalski horses treated with DEA had significantly faster and less variable induction and recovery times than when treated with KBAM, and KBAM inductions were characterized by prolonged periods of incoordination in some individuals. This is consistent with previous work comparing etorphine-azaperone and KBM in zebras, in which KBM also resulted in a markedly slower and more ataxic induction than the etorphine combination. In domestic equids, increasing duration of the induction and recovery periods is associated with heightened risk of injury caused by ataxic stumbling. The length and character of KBAM inductions could potentially increase the risk of injury compared with DEA. One horse in each of the treatment groups failed to become recumbent following apparently correct dart placement. Possible causes for apparent drug failure include injection into fascial planes rather than deep IM injection, incomplete injection, incorrect drug compounding, errors in dart preparation, or the effects of sex and temperament. Poor muscle relaxation contributed to suboptimal immobilization quality for DEA compared with KBAM horses, as is observed in many species under the influence of UPOs. Excessive muscle rigidity and spontaneous movements when the horse is recumbent present a risk of injury to horse and handlers, increase rectal temperature, and make monitoring the depth of immobilization challenging, as
demonstrated by the spontaneous arousal of a DEA horse at T10.

Prolonged recoveries from KBAM immobilization may be attributable to incomplete reversal and the use of ketamine. In the DEA protocol, detomidine was reversed with RX821002 and etorphine with naltrexone, leaving only acepromazine to be metabolized, whereas for KBAM, butorphanol was reversed with naltrexone and medetomidine with atipamezole, leaving both ketamine and azaperone to be metabolized. In this study, RX821002 was used instead of atipamezole to reverse detomidine because of its low cost, its $\alpha_2$-to-$\alpha_1$-specificity ratio being considered closer to that of detomidine, the small volume required, and the authors' positive experience using this reversal in Przewalski horses. In equids, complete and rapid recovery from immobilization is desirable to prevent disruption of social structure, misadventure, and derangements in body temperature. Prolonged recumbency during slow recoveries additionally exacerbates hypoxemia caused by compression of the dependent lung, as well as increasing the risk of neuropathy and myopathy in the dependent limbs. Dividing antagonists equally between the IV and IM routes results in rapid (<5 min) recovery from KBAM in Przewalski horses (Milnes et al, VetMB, DVSc, DACZM, DECZM, Toronto Zoo, Toronto, ON, Canada, unpublished data, 2021). However, IV administration of $\alpha_2$-adrenoceptor antagonists can result in excitement and profound vasodilation with hypotension, and thus the IM route is generally recommended except for emergency situations.

Compounded butorphanol-azaperone-medetomidine and etorphine-acepromazine products were used because approved products were not available in Canada at the time of the study. Veterinarians should adhere to compounding regulations and be aware that pharmacokinetic properties may differ between compounded and approved products. The use of both etorphine and concentrated medetomidine bears an inherent risk of human injury from accidental exposure. Therefore, it is vital to preemptively establish emergency protocols in case of accidental human exposure for both KBAM and DEA.

Horses treated with DEA were severely hypoxemic ($PAO_2 < 60 \text{ mm Hg}$) at all time points and often apneic. Opioid-induced respiratory depression is the result of $\mu$-receptor-mediated depression of the central respiratory center. Etophine-associated skeletal muscle tremors increase cellular oxygen consumption and may therefore have further contributed to DEA-associated hypoxemia. Despite not receiving a UPO, moderate hypoxemia ($PAO_2 < 80 \text{ mm Hg}$) also occurred in horses treated with KBAM, as is consistently observed in nondomestic equids immobilized with medetomidine-ketamine combinations. Similarly, $PAO_2$ commonly decreases from 100 mm Hg to 60 to 80 mm Hg within 5 minutes of anesthetic induction in domestic horses not administered oxygen. This hypoxemia is mainly a result of ventilation-perfusion mismatch and, to a lesser extent, intrapulmonary vascular shunting because of atelectasis of the dependent lung fields that occurs during recumbency. Ketamine, like etorphine, increases sympathetic tone and cellular oxygen consumption, which could have additionally contributed to the low $PAO_2$ in KBAM-treated horses in the present study. Oxygen supplementation is therefore recommended with both KBAM and DEA protocols and, indeed, in all equid immobilizations. In horses, oxygen insufflation at 50 L/min has been demonstrated to normalize $PAO_2$, if the nasal line was advanced into the pharynx or trachea. In this study, nasal oxygen at 15 L/min only slightly increased $PAO_2$ over time. It could be that the nasal line was poorly positioned or that the flow rate was insufficient. However, if the predominant cause of hypoxemia was venous admixture, even at high flow rates, the fraction of inspired oxygen would have had very little impact on $PAO_2$. More research is required to establish optimal methods for respiratory support in immobilized nondomestic equids.

The $PAO_2$ values reported here are much lower than values reported for domestic horses that were recumbent, anesthetized, or both. This finding may be the result of interspecies variation, an effect of the anesthetic agents, or sampling error. It is possible that there was collection of venous or mixed sample because of difficulty palpating peripheral pulses in some of the horses, which would decrease oxygen content. Additionally, on colder days there was a time delay from sample collection to analysis because the analyzer needed to be warmed before the sample could be processed in the field; this could also decrease oxygen content of the sample. Despite very low $PAO_2$ on a number of occasions, all horses recovered from each anesthesia uneventfully with no reported long-term consequences such as cognitive deficits. Thoroughbred horses have much lower $PAO_2$ during exercise than when at rest and autoregulation of cerebral and cerebellar blood flow is maintained in exercising horses despite this hypoxemia and hypercapnia. It is not known to what extent the autoregulatory response of brain blood flow to hypoxemia is attenuated during anesthesia with KBAM and DEA.

In summary, KBAM may provide a useful alternative to etorphine-based combinations for captive Przewalski horses. Horses treated with KBAM had subjectively better immobilization quality during the maintenance phase, compared with apnea and severe muscle tremors and rigidity in horses treated with DEA. However, KBAM induction and recovery periods were longer and less predictable compared with DEA, which could increase the risk of injury caused by ataxic stumbling and may therefore limit the usefulness of KBAM to situations in which the horse’s environment can be controlled to prevent misadventure. Low $PAO_2$ (KBAM) to severe hypoxemia (DEA) occurred with both protocols, highlighting the critical importance of diligent monitoring and oxygen supplementation. Although the risk of UPOs to human health is well-understood and KBAM may therefore be viewed as a “safer” option than etorphine, the potential danger of accidental human exposure to potent medetomidine should not be ignored when working with KBAM.
Acknowledgments

This study was supported by grants from the American Association of Zoo Veterinarians Wild Animal Health Fund and The Morris Animal Foundation (D20EQ-812). The authors declare there were no conflicts of interest. The funding sources did not have any involvement in the study design, data analysis and interpretation, or writing and publication of the manuscript. The authors thank Dr. Margo Chase-Topping for assistance with statistical analysis.

This article represents a portion of a thesis submitted by Dr. Milnes to the University of Edinburgh as partial fulfillment of the requirements for a Master of Veterinary Science degree.

References


