

CARDIOVASCULAR EFFECTS OF ETORPHINE, AZAPERONE, AND BUTORPHANOL COMBINATIONS IN CHEMICALLY IMMOBILIZED CAPTIVE WHITE RHINOCEROS (*CERATOTHERIUM SIMUM*)

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Abstract: Chemical capture is an essential tool in the management and conservation of white rhinoceros (*Ceratotherium simum*); however, cardiovascular responses in immobilized megaherbivores are poorly understood. Blood pressure and heart rate responses in rhinoceros immobilized with etorphine or etorphine plus azaperone, and the effects of subsequent i.v. butorphanol administration were investigated. Six white rhinoceros were used in a randomized crossover study design with four interventions: 1) etorphine i.m.; 2) etorphine plus azaperone i.m.; 3) etorphine i.m. and butorphanol i.v.; and 4) etorphine plus azaperone i.m., and butorphanol i.v. Etorphine resulted in hypertension and tachycardia in immobilized rhinoceros on initial measurements. Over the 25-min study period, blood pressures and heart rate declined. Heart rates were slower, although the rhinoceros were still tachycardic, and blood pressures lower during the whole study period in animals immobilized with etorphine and azaperone compared with those that received only etorphine. Butorphanol administration resulted in lower arterial blood pressures and heart rates in etorphine-immobilized rhinoceros. In rhinoceros immobilized with etorphine and azaperone, heart rate slowed following administration of butorphanol i.v., although blood pressures remained unchanged. Azaperone reduced hypertension associated with etorphine immobilization, but animals remained tachycardic. Administration of butorphanol to etorphine/azaperone-immobilized rhinoceros lowered heart rate to values approaching normal resting levels without altering blood pressure.

Key words: Blood pressure, cardiovascular, *Ceratotherium simum*, heart rate, white rhinoceros.

INTRODUCTION

Chemical immobilization is an essential management tool for moving white rhinoceros (*Ceratotherium simum*) between isolated populations to maintain genetic diversity, collecting biological

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samples, attaching radio-tracking devices, facilitating dehorning procedures, and treating injured individuals.^{22,32} Etorphine plus azaperone, the preferred immobilizing drug combination, causes significant respiratory depression in immobilized white rhinoceros and butorphanol is commonly administered to counteract these effects.^{2–4,22,23} Alterations in respiratory function have been extensively studied in white rhinoceros; however, there are only a few reports on the cardiovascular effects of these drugs.^{14,22,25,32} The pressor effects of etorphine, and etorphine plus fentanyl, have been described in captive and free-ranging rhinoceros, respectively.^{13,15,18} Alterations to heart rate and blood pressure, measured noninvasively, also have been described briefly in game-ranched rhinoceros chemically captured utilizing etorphine, azaperone, and butorphanol.² Clinical observations following the administration of butorphanol in immobilized free-ranging rhinoceros suggest that it results in a substantial reduction in heart rates (Buss, pers. comm.).

The purpose of this study was to determine and compare the cardiovascular effects of etorphine or etorphine plus azaperone in boma-adapted

white rhinoceros, and to evaluate how i.v. butorphanol administration influences these effects.

MATERIALS AND METHODS

Six subadult (5–6 yr) male white rhinoceros, weighing between 1,194 and 1,420 kg, were captured in Kruger National Park (23°49'60 S, 31°30'0 E; alt. 317 m), South Africa, and habituated to captivity over a period of 4 mo. The animals were housed individually in rhinoceros-specific holding pens. Water and a 50 : 50 mix of Lucerne (*Medicago sativa*) and Tef (*Eragrostis tef*) hay were provided ad libitum. Feces were removed from the enclosures, water troughs cleaned, and food replaced daily.

A randomized crossover study design was used with a 2-wk washout period between each of four treatments: treatment 1: etorphine (Elanco, Kempton Park 1619, South Africa; 9.8 mg/ml) plus hyaluronidase (Kyron Laboratories, Benrose 2094, South Africa; 5000 i.u./vial) i.m.; treatment 2: etorphine and azaperone (Janssen Pharmaceutical Ltd., Halfway House 1685, South Africa; 40 mg/ml) plus hyaluronidase i.m.; treatment 3: etorphine plus hyaluronidase i.m. followed by butorphanol (Kyron Laboratories; 50 mg/ml) i.v.; and treatment 4: etorphine and azaperone plus hyaluronidase i.m. followed by butorphanol i.v. Doses were based on two standardized body mass categories: 1,000 to 1,250 kg received 2.5 mg etorphine, 37.5 mg azaperone, 5,000 i.u. hyaluronidase, and 25 mg butorphanol; and 1,250 to 1,500 kg received 3.0 mg etorphine, 45 mg azaperone, 5,000 i.u. hyaluronidase, and 30 mg butorphanol. Hyaluronidase (5,000 i.u.) was included in the immobilizing drug mixture as it facilitates drug absorption and reduces induction time.¹⁰

The immobilizing drugs were delivered into the muscles of the nuchal hump using a 3.0-ml plastic dart with a 60-mm uncollared needle using a compressed air rifle (DAN-INJECT International S.A., Skukuza 1350, South Africa). Once a darted animal could be safely approached, it was blindfolded and placed into lateral recumbency. Trials were conducted only if the animal was recumbent, could be safely handled within 15 min of darting, and instrumented within a further 10 min. A 22 G × 1-inch catheter (Nipro Safelet Cath, Nipro Medical Corporation, Bridgewater, New Jersey 08807, USA) was placed into a medial auricular artery. Arterial blood pressures were recorded using a transducer (TranStar 60-inch Single Monitoring Kit, Ref MX950T, Smiths Medical ASD, Inc., Dublin, Ohio 43017, USA) secured at the level of the heart and zeroed prior to

connecting to a precalibrated Cardiocap/5 physiological monitor (Datex-Ohmeda, GE Healthcare, Helsinki 00510, Finland). Heart rate was determined by chest auscultation and confirmed using the physiological monitor.

Systolic, diastolic, and mean arterial blood pressures, and heart rate were initially recorded at 10 min after the animal became recumbent ($t = 0$) and at each subsequent 5-min interval for a total of 25 min. Butorphanol (at $10 \times$ the etorphine dose in mg) was administered i.v. into an auricular vein at 2 min ($t = 2$) after the first measurement ($t = 0$) to rhinoceros in treatments 3 and 4. An equivalent volume of sterile saline was administered i.v. as a control at $t = 2$ to those animals not receiving butorphanol.

At the end of each procedure, butorphanol was administered i.v. (at $10 \times$ the etorphine dose mg) to animals in treatments 1 and 2 after the last sample at $t = 25$ min. All animals were walked into a crate of known weight and weighed by suspending the crate from a scale. Naltrexone (Kyron laboratories; 40 mg/ml) was administered i.v. (at $20 \times$ the etorphine dose in mg). Rhinoceros were kept under observation until fully recovered.

Data analysis

STATA (Stata Statistical Software: Release 14, College Station, Texas 77840, USA) was used for the statistical analysis. Descriptive statistics (means, standard deviations, medians, and 1st [Q1] and 3rd [Q3] quartile) were calculated to assess the data distribution for each treatment at different sampling points. Due to the relatively small sample size used for this study ($n = 6$), nonparametric statistical tests were used to compare median cardiovascular values at different sampling points within each treatment. Initially, the data was screened using the Kruskal-Wallis test to assess if median values for different cardiovascular parameters differed over sampling points. Subsequently, we formally tested the hypothesis that cardiovascular values changed at $t = 10$ as compared with $t = 0$. To compare differences in medians between matched pairs of cardiovascular values at $t = 0$ with $t = 10$, the Wilcoxon signed ranks test was used to account for repeated measurements over time in the same individual. The data distribution indicated that after $t = 10$, cardiovascular values tended to stabilize; thus, and to confirm that no further changes occurred after 10 min, we used linear regression (using ranks) with sampling points as fixed effects to formally assess changes on cardiovascular parameters after 10 min, using $t = 10$ as the reference value. To

Table 1. Distribution of cardiovascular parameters (median and interquartile range [IQR]) in six captive white rhinoceros ($n = 6$) for four treatments at sampling periods 0, 5, 10, 15, 20, and 25 min.

Arterial blood pressures	Treatment ^a	Time						Overall IQR
		0 min IQR ^b	5 min IQR	10 min IQR	15 min IQR	20 min IQR	25 min IQR	
Mean (mm Hg)	1	174 (155–187)	157 (143–169)	131 (129–144)	131 (121–142)	122 (112–129)	122 (112–129)	133 (124–150.5)
	2	111 (91–120)	93 (80–111)	92 (84–103)	93 (81–112)	90 (83–102)	94 (81–106)	94 (83–110.5)
	3	180 (171–188)	131 (110–136)	110 (96–116)	111 (92–122)	112 (95–128)	103 (90–114)	117 (99.5–142)
	4	131 (98–146)	81 (63–112)	77 (74–86)	80 (68–89)	83 (72–87)	88 (77–97)	87 (72.5–99)
Systolic (mm Hg)	1	192 (175–209)	173 (164–190)	154 (146–158)	155 (141–162)	146 (134–152)	144 (135–155)	158 (144–167.5)
	2	123 (110–141)	112 (92–135)	112 (101–121)	112 (91–132)	110 (101–124)	114 (90–130)	114 (97.5–130.5)
	3	211 (196–226)	155 (130–173)	138 (121–144)	141 (113–148)	140 (119–157)	135 (113–140)	146 (124.5–179.5)
	4	145 (115–169)	101 (73–120)	100 (96–114)	104 (91–111)	102 (92–114)	110 (92–119)	106 (92–120)
Diastolic (mm Hg)	1	160 (136–169)	139 (129–151)	123 (107–129)	113 (103–125)	104 (97–111)	105 (94–112)	119 (104–132.5)
	2	97 (81–106)	76 (73–94)	78 (68–88)	81 (67–90)	75 (71–79)	79 (73–85)	79 (71.5–90)
	3	161 (151–166)	116 (93–121)	93 (82–99)	96 (79–106)	98 (78–112)	85 (72–97)	101 (83.5–124)
	4	117 (84–129)	67 (53–105)	65 (62–72)	67 (58–74)	72 (63–77)	76 (73–83)	74 (60–85)
Heart rate (beats/min)	1	136 (130–144)	141 (124–144)	134 (112–136)	125 (114–128)	120 (112–128)	114 (101–124)	127 (114–139)
	2	120 (112–132)	116 (104–124)	110 (92–116)	110 (92–116)	108 (88–116)	104 (84–112)	112 (96–120)
	3	139 (122–148)	89 (82–114)	65 (58–68)	63 (52–64)	60 (48–68)	68 (48–69)	68 (60–111)
	4	134 (114–140)	68 (58–80)	54 (46–56)	52 (45–56)	49 (48–56)	58 (53–61)	56 (48–69)

^a Treatments: 1) etorphine; 2) etorphine plus azaperone; 3) etorphine and butorphanol i.v.; and 4) etorphine plus azaperone, and butorphanol i.v.

^b IQR indicates interquartile range (25th to 75th percentile).

evaluate differences on cardiovascular parameters between treatment groups, we used linear regression (using ranks) to compare median cardiovascular parameters while adjusting for the effect of time, including sampling time points, as a fixed effect in the model. Statistical significance was set at $P < 0.05$ for all statistical tests.

RESULTS

Treatment 1: etorphine

At first sampling ($t = 0$), the median values for arterial blood pressures were: mean 174 mm Hg, systolic 192 mm Hg and diastolic 160 mm Hg (Table 1). These arterial pressures decreased significantly over time to 131 mm Hg ($P =$

0.028), 154 mm Hg ($P = 0.028$), and 123 mm Hg ($P = 0.028$), respectively, at $t = 10$. Between $t = 10$ and $t = 25$, mean and diastolic arterial blood pressures decreased further to 122 mm Hg ($P = 0.017$) and 105 mm Hg ($P = 0.012$), although there was no significant change in systolic blood pressure (Table 1; Fig. 1). Heart rate decreased from 136 beats/min to 114 beats/min over the immobilization period ($t = 0$ to $t = 25$); however, changes were not statistically significant between $t = 0$ and $t = 10$ ($P = 0.059$), and $t = 10$ and $t = 25$ ($P = 0.442$) (Table 1; Fig. 2).

Treatment 2: etorphine plus azaperone

At $t = 0$, the median values for arterial blood pressures in rhinoceros immobilized with etor-

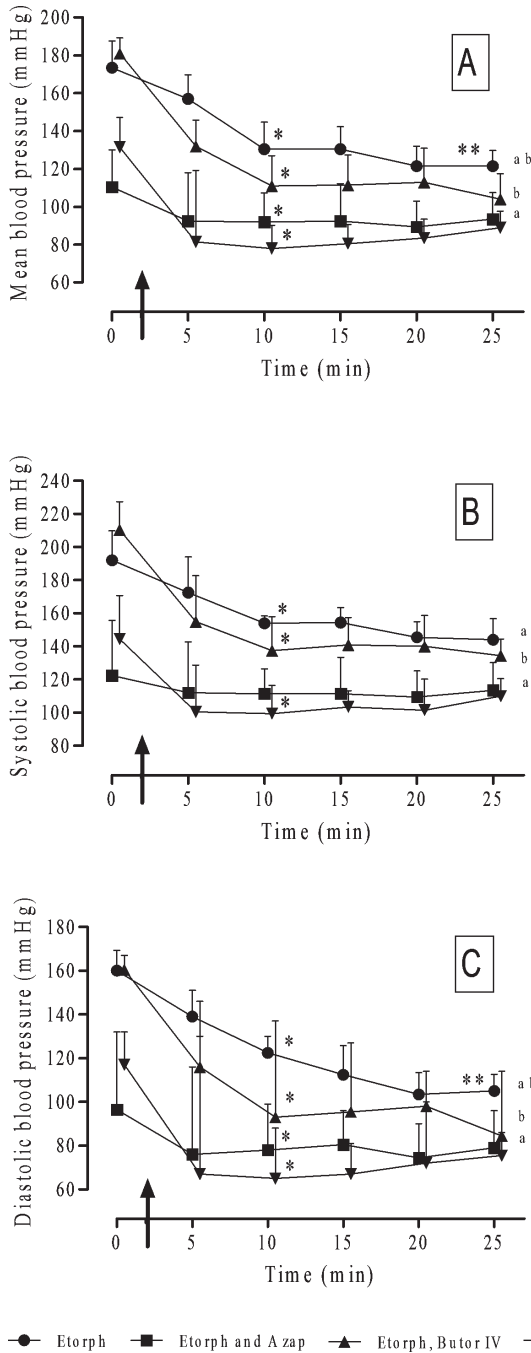


Figure 1. Arterial blood pressures: (A) Mean, (B) Systolic, (C) Diastolic. Note: Median and interquartile range of mean, systolic, and diastolic arterial blood pressures at sampling periods 0, 5, 10, 15, 20, and 25 min in six captive white rhinoceros ($n = 6$) for four treatments: 1) etorphine; 2) etorphine plus azaperone; 3) etorphine and butorphanol i.v.; 4) etorphine plus azaperone and butorphanol i.v. The black arrow indicates the time at which butorphanol was administered. *, indicates a significant ($P < 0.05$) difference within treatment between $t = 0$ and $t = 10$. **, indicates a significant ($P < 0.05$) difference within treatment between $t = 10$ and $t = 25$. ^{a-b}, the same letter indicates a significant ($P < 0.05$) difference in overall median values between treatments.

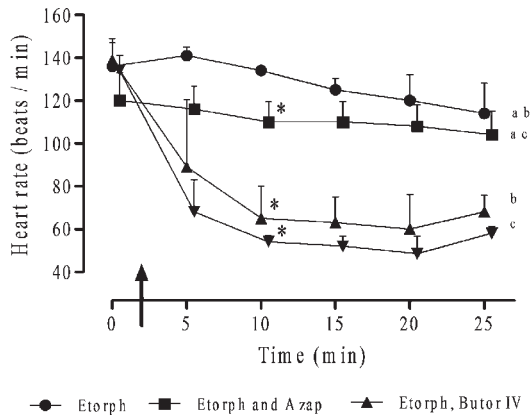


Figure 2. Heart rates. Note: Median and interquartile range of heart rates at sampling periods 0, 5, 10, 15, 20, and 25 min in six captive white rhinoceros ($n = 6$) for four treatments: 1) etorphine; 2) etorphine plus azaperone; 3) etorphine and butorphanol i.v.; 4) etorphine plus azaperone and butorphanol i.v. The black arrow indicates the time at which butorphanol was administered. *, indicates a significant ($P < 0.05$) difference within treatment between $t = 0$ and $t = 10$. **, indicates a significant ($P < 0.05$) difference within treatment between $t = 10$ and $t = 25$. ^{a-c}, the same letter indicates a significant ($P < 0.05$) difference in overall median values between treatments.

phine and azaperone were: mean 111 mm Hg, systolic 123 mm Hg, diastolic 97 mm Hg, and heart rate was 120 beats/min (Table 1). Significant decreases occurred between $t = 0$ and $t = 10$ in mean (111 mm Hg to 92 mm Hg, $P = 0.046$) and diastolic (97 mm Hg to 78 mm Hg, $P = 0.046$) arterial blood pressures; however, the decrease in systolic blood pressure (123 mm Hg to 112 mmHg) during the same period was not statistically significant ($P = 0.075$). Heart rate decreased significantly from 120 beats/min to 110 beats/min over the first 10 min of the study ($P = 0.028$) (Table 1; Figs. 1, 2). No further statistically significant changes were observed in mean, systolic, and diastolic arterial blood pressures and heart rate after 10 min.

Overall, during the 25-min immobilization period, arterial blood pressure measurements were significantly ($P < 0.001$) lower in rhinoceros immobilized with etorphine and azaperone compared with those that received only etorphine (differences in blood pressures were mean 39 mm Hg, systolic 45 mm Hg, and diastolic 40 mm Hg) (Table 1; Fig. 1). Median heart rates were also significantly different between the two treatments (15 beats/min, $P < 0.001$) (Table 1; Fig. 2).

Treatment 3: etorphine and butorphanol

After the administration of butorphanol i.v. ($t = 2$) in rhinoceros immobilized with etorphine, blood pressures decreased significantly ($P = 0.028$) between $t = 0$ and $t = 10$ (mean 180 mm Hg to 110 mm Hg, systolic 211 mm Hg to 138,

and diastolic 161 mm Hg to 93 mm Hg (Table 1; Fig. 1). Heart rate also decreased significantly ($P = 0.028$) by 74 beats/min (139 beats/min to 65 beats/min) between $t = 0$ and $t = 10$. After $t = 10$, arterial blood pressures and heart rate did not change significantly over the rest of the immobilization period (Table 1; Fig. 2).

Generally and when controlling for the effect of time, cardiovascular parameters and heart rate were significantly lower in etorphine-immobilized rhinoceros that received butorphanol compared with those given sterile saline (decreases in arterial blood pressures were mean 15 mm Hg, $P = 0.001$; systolic 12.5 mm Hg, $P = 0.019$; and diastolic 18.5 mm Hg, $P < 0.001$; and heart rate 59 beats/min, $P < 0.001$) (Table 1; Figs. 1, 2).

Treatment 4: etorphine plus azaperone, and butorphanol

The administration of butorphanol at $t = 2$ to rhinoceros immobilized with a combination of etorphine and azaperone resulted in significant changes in arterial blood pressures between $t = 0$ to $t = 10$ (mean 131 mm Hg to 77 mm Hg, $P = 0.028$; systolic 145 mm Hg to 100 mm Hg, $P = 0.046$; diastolic 117 mm Hg to 65 mm Hg, $P = 0.028$). Between $t = 10$ and $t = 25$, there were no significant changes in blood pressures (Table 1; Fig. 1). Heart rate slowed significantly between $t = 0$ and $t = 10$ (134 beats/min to 54 beats/min, $P = 0.028$) and remained unchanged for the rest of the immobilization beyond $t = 10$ ($P = 0.442$) (Table 1; Fig. 2).

Overall in rhinoceros immobilized with etorphine and azaperone, there were no significant differences in arterial blood pressures between those administered i.v. butorphanol and those that did not receive butorphanol (Table 1; Fig. 1). Heart rate was significantly ($P < 0.001$) lower in rhinoceros administered butorphanol, compared with no butorphanol (56 beats/min compared with 112 beats/min) (Table 1; Fig. 2).

DISCUSSION

Immobilization of white rhinoceros with etorphine results in hypertension and tachycardia. The inclusion of azaperone with etorphine in the immobilizing drug combination reduces blood pressure associated with etorphine administration to values lower than those reported for unrestrained zoo animals.⁶ Heart rates were also lower with this combination of opioid and tranquilizer compared with animals induced with only etorphine; however, the animals remained tachycardic with heart rates (≥ 100 beats/min) more than double those of normal values in unrestrained awake animals (32 to 42 beats/min).⁶ Intravenous administration of butorphanol to rhinoceros immobilized with only etorphine resulted in some reduction in arterial blood pressure and no significant change in animals that were induced with both etorphine and azaperone. However, in both of these treatments, butorphanol administration resulted in significant reductions in heart rate.

The rhinoceros in this study, when immobilized with etorphine, were hypertensive at $t = 0$ (10 min after becoming immobilized) compared with standing unrestrained captive white rhinoceros (mean arterial blood pressure 173.5 mm Hg vs. 124 mm Hg, systolic 192 mm Hg vs. 160 mm Hg, diastolic 160 mm Hg vs. 104 mm Hg).⁶ In addition, animals that received etorphine were tachycardic compared with standing unrestrained animals (heart rate 136 beats/min vs. 39 beats/min).⁶ Hypertension and tachycardia have been previously reported in a small number of captive white rhinoceros immobilized with etorphine.^{15,18} A mean intra-arterial blood pressure of 183 mm Hg was recorded in free-ranging animals chemically captured with an opioid combination of etorphine and fentanyl.¹³ This blood pressure is similar to values of animals in the study, but the influences of combining fentanyl with etorphine or a potential adrenergic response induced by a chase and darting from a helicopter are unknown.¹³

The underlying mechanisms for increased arterial blood pressures and tachycardia in rhinoceros immobilized with etorphine are not fully understood. The cardiovascular response observed in the study may, in part, result from hypoxia caused by drug-induced respiratory depression, a common finding in immobilized white rhinoceros.^{4,15} Hypoxia in humans resulted in increased heart rate, cardiac output, and systolic blood pressure, although mean and diastolic arterial pressures remained constant or fell slightly.²¹ Hypoxemic activation of arterial chemoreceptors increases both sympathetic vasoconstrictor outflow to vascular beds and cardiac sympathetic activity increasing heart rate.²⁷ In domestic horses administered etorphine and acepromazine, a similar cardiovascular outcome was hypothesized to result from an etorphine-induced sympathetic response through the release of catecholamines from postganglionic neurons.^{9,26} Cardiovascular pressor effects are consistent findings in opioid-immobilized perissodactyls, and have been reported in domestic and Mongolian horses, and Grevy's zebra.¹² Opioid receptors have been identified in rodent myocardial tissue preparations. If these myocardial receptors are ubiquitous in mammals, then activation of these receptors by etorphine could result in tachycardia with a potential increase in cardiac output.¹¹

The initial hypertension in rhinoceros immobilized with etorphine had resolved at $t = 25$ to values similar to those in standing unrestrained zoo animals (arterial blood pressures were mean 122 mm Hg vs. 124 mm Hg, systolic 154 mm Hg vs. 160 mm Hg, and diastolic 105 mm Hg vs. 104 mm Hg).⁶ Although the change in heart rate from 136 beats/min to 114 beats/min between $t = 0$ and $t = 25$ was not statistically significant, it may reflect a clinically relevant reduction in rate. However, rhinoceros remained tachycardic at $t = 25$ compared with unrestrained animals (heart rate 114 beats/min vs. 39 beats/min).⁶ A decrease in blood pressures with a persistent tachycardia at the end of the immobilization is an unexpected result. A decreased drug effect due to redistribution and metabolism of etorphine would account for decreasing blood pressure over time but not the elevated heart rate. A possible explanation is an increase in heart rate to maintain cardiac output and arterial blood pressures in the presence of a reduced stroke volume and/or total peripheral resistance. In a recumbent immobilized animal, stroke volume may be reduced due to limited limb skeletal muscle activity causing blood pooling and decreased venous return to the

heart. Blood pooling may also occur due to an opioid-induced venous dilation, as has been reported in humans.¹ Respiratory depression and chest wall rigidity, which commonly occurs in immobilized rhinoceros, can also potentially decrease cardiac venous return by limiting the negative intrathoracic pressure that develops in association with inspiration and usually assists blood flow through the chest.^{4,14,29} Severe hypoxia associated with opioid immobilization of rhinoceros may also be implicated in a local vasodilation in response to tissue hypoxia, a fundamental physiological response to ensure adequate oxygen supply–demand balance in metabolically active tissues.^{14,31}

A persistent tachycardia with potential increased myocardial oxygen consumption is of clinical concern due to limited anaerobic capacity of the myocardium and pronounced hypoxia commonly associated with etorphine-induced respiratory depression.^{15,30} It is also unknown how immobilizing drugs may influence the mechanisms that match coronary blood flow with myocardial oxygen requirements.³⁰ No obvious adverse effects associated with the persistent tachycardia were observed in the study animals; however, a negative outcome may occur in rhinoceros compromised due to age, disease, or poor nutrition.

Overall, arterial blood pressures in the study animals were significantly lower during the entire study period when azaperone was included with etorphine compared with etorphine only. At $t = 0$, rhinoceros had lower blood pressures compared with resting values in standing unrestrained captive animals. Heart rate decreased significantly over the first 10 min and was also clinically slower with the inclusion of azaperone; however rhinoceros were still tachycardic compared with values in standing resting animals (110 beats/min vs. 39 beats/min).⁶ By comparison, Boardman et al. (2014) reported similar heart rates (118 beats/min), but higher blood pressures (systolic 162 mm Hg and diastolic 104 mm Hg) in white rhinoceros immobilized with similar doses of etorphine and azaperone.² Those rhinoceros were free-ranging and darted from a vehicle, which may have caused a greater sympathetic response and hypertension than in the boma-acclimated rhinoceros.

Azaperone is advocated for rhinoceros immobilization to counteract etorphine-induced hypertension by antagonizing α_1 -receptors in peripheral arterioles thus limiting vasoconstriction.^{2,19,24} The hypotensive effects of azaperone have been described in domestic horses; azaperone can reduce

mean arterial pressure by approximately a third from resting levels, for up to 4 hr.²⁰

Potential mechanisms for tachycardia in etorphine/azaperone-immobilized rhinoceros could be related to a baroreceptor reflex induced by the hypotensive effects of azaperone, the sympathomimetic effects of etorphine and associated hypoxia, and possible direct opioid effects on the myocardium.^{11,19,26} However, these explanations for the cardiovascular effects may be incomplete due to a limited understanding of receptor (opioid, dopamine, and adrenergic) distributions and associated functions within rhinoceros organ systems.^{5,7}

While inclusion of azaperone reversed hypertension associated with etorphine immobilization, the lower than normal blood pressure that resulted in the rhinoceros also could be of clinical concern. A low mean arterial blood pressure can reduce blood flow through skeletal muscles, especially of the limbs, causing a buildup of metabolic waste products and a persistent hypoxia with possible myopathy and irreversible muscle damage.^{25,29} Increased muscle activity from running prior to immobilization, and compression of muscles with occlusion of blood vessels in limbs positioned underneath a recumbent animal can further increase the risk of tissue injury.²⁵ A normal pressure response in other conscious megaherbivores is an increase in blood pressure when animals move from standing to lateral recumbency.¹⁶ This compensatory response in the rhinoceros appeared to be prevented by the etorphine and azaperone combination.

Administering butorphanol to etorphine-immobilized rhinoceros resulted in rapid reductions in both arterial blood pressures and heart rate, reaching maximum changes within 10 min. Blood pressure values did not decrease to values observed in rhinoceros immobilized with etorphine and azaperone; however, heart rate was markedly slower in animals receiving butorphanol (65 beats/min compared with 110 beats/min at $t = 10$). In horses and humans, butorphanol administered alone did not significantly alter heart rate or blood pressure.^{28,33} Due to a limited understanding of the complex pharmacology of butorphanol and its interactions with potent opioids like etorphine, it is difficult to definitively explain the mechanisms that result in these cardiovascular effects.^{8,17} Etorphine is a potent pure opioid agonist at μ -, δ -, and κ -receptors, while butorphanol is a synthetic agonist–antagonist opioid that acts as a partial agonist at μ -, a pure agonist at κ -, and mainly as an antagonist at δ -receptors.^{8,34}

Pharmacological responses resulting from butorphanol activity at μ -receptors frequently overshadow those of the κ -receptors.⁸ Apart from the potential interactions of these two drugs at multiple opioid receptor types, pretreatment with etorphine can alter the binding of μ -receptors with other specific agonists.⁸ It is therefore plausible that butorphanol's partial agonist effects on μ -receptors partly antagonize some of etorphine's μ -agonist effects, like tachycardia.

Butorphanol administration in etorphine- plus azaperone-immobilized rhinoceros did not alter blood pressures, but heart rates decreased to values clinically comparable to those recorded in unrestrained zoo rhinoceros.⁶ Boardman et al. (2014) reported a decrease in both heart rate and blood pressure following the administration of butorphanol in rhinoceros immobilized with etorphine and azaperone.² The reason for the difference in blood pressure response is unknown, but may arise from the difference in the initial flight response in game-ranched animals darted from vehicles compared with boma-adapted rhinoceros darted from the ground.² Despite the difference in blood pressure responses between the two studies, rhinoceros in both groups had low blood pressures with a normal resting heart rate following butorphanol administration.

Since arterial blood pressures didn't change but heart rate slowed in etorphine- plus azaperone-immobilized rhinoceros following the administration of butorphanol, it is likely that the falling heart rate was associated with an increase in stroke volume, ejection fraction, or a combination of both to maintain cardiac output. Azaperone antagonizes peripheral vascular α_1 -receptors so it is unlikely that an increased total peripheral resistance contributed to maintaining blood pressure.^{19,29} The reduction in heart rate may result not only from a barometric response to an increase in cardiac output, but could also be related to a decrease in sympathetic response due to partial antagonism of etorphine's effects or an improvement in arterial oxygen tension. Buss et al. (2015) found that arterial oxygen partial pressures increased, from very low values, after the administration of butorphanol in boma-habituated rhinoceros immobilized with a combination of etorphine and azaperone.⁴ A reduction in heart rate subsequent to improved blood oxygen levels would be expected if the original tachycardia was caused by hypoxia.²¹

Irrespective of immobilizing drug combination used, administration of butorphanol in boma-adapted white rhinoceros reduced heart rate. As

previously mentioned, a potential benefit of decreased heart rate is a reduction in myocardial oxygen requirements in animals experiencing a marked hypoxia.

The mechanisms resulting in the cardiovascular changes recorded in this study can be elucidated further by developing a validated technique for determining cardiac output and through molecular investigation of drug-receptor interactions in rhinoceros. Cardiovascular changes associated with the use of etorphine, azaperone, and butorphanol should also be investigated in free-ranging rhinoceros as they are usually captured by darting from a helicopter, which induces a significant sympathetic response that may further alter cardiovascular function.²⁵

CONCLUSION

Hypertension and tachycardia, which occurred in rhinoceros immobilized with etorphine, were reduced by both including azaperone in the immobilizing drug combination and administering butorphanol i.v. shortly after the animal had become recumbent. Inclusion of azaperone in the dart reduced etorphine-induced hypertension but did not correct tachycardia. Intravenous administration of butorphanol reduced the heart rate to values reported for resting unrestrained animals, but did not alter blood pressure further. This reduction may be due to an improvement in hypoxemia. A decreased heart rate may have a beneficial oxygen sparing effect on the myocardium. Similarly, a reduction in peripheral resistance and blood pressure by azaperone will reduce cardiac workload and hence oxygen requirements. However, of clinical concern, is that a profound decrease in blood pressure associated with the use of azaperone could cause adverse consequences due to reduced tissue perfusion, especially in compressed skeletal muscle groups in recumbent animals. Whether a lower azaperone dose than the one used may reverse the etorphine-induced hypertension without causing hypotension requires further investigation. In summary, azaperone reduced the hypertensive effects and butorphanol reduced the tachycardic effects of etorphine in immobilized white rhinoceros, thereby reducing potential risks associated with etorphine immobilization.

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