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BUTORPHANOL/XYLAZINE/KETAMINE IMMOBILIZATION OF FREE-RANGING BAIRD'S TAPIRS IN COSTA RICA

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ABSTRACT: Cardiopulmonary effects and the utility of a butorphanol/xylazine/ketamine combination were evaluated during twenty immobilizations of sixteen Baird's tapirs (*Tapirus bairdii*) between March 1996 and January of 1998 in Corcovado National Park (Costa Rica). The animals were attracted to a bait site and darted from tree platforms. The tapirs were estimated to weigh between 200 to 300 kg. Actual weights of three tapirs taken at later dates fell within the estimated range. A butorphanol, 48 ± 1.84 ($\bar{x}$ ± SE) mg/animal IM, and xylazine, 101 ± 2.72 mg/animal IM, combination was used to immobilize the animals. In some instances, ketamine was used either IM or IV at 187 ± 40.86 mg/animal to prolong the immobilization period in addition to the butorphanol/xylazine combination. Naltrexone was used IM to reverse butorphanol at 257 ± 16.19 mg/animal. Either yohimbine, 34 ± 0.61 or tolazoline at 12 ± 10.27 mg/animal, was used to reverse xylazine. The mean time from dart impact to first visible effect was 4.63 ± 0.50 min ($\bar{x}$ ± SE). Mean time to sternal recumbency was 12.21 ± 1.08 min. Mean time the tapirs were immobilized was 45.63 ± 3.6 min. Mean time to return to sternal recumbency and standing in animals that received yohimbine and naltrexone was 3.16 ± 1.06 and 5.33 ± 1.45 min, respectively. Mean time to return to sternal recumbency and standing in animals that received tolazoline and naltrexone was 1.57 ± 0.39 and 3.14 ± 0.51 min, respectively. Cardiopulmonary parameters including heart rate, respiratory rate, body temperature, electrocardiogram, percent oxygen saturation, and indirect blood pressure were recorded. Arterial blood gas analysis was performed on four animals. A mild degree of hypoxemia was evidenced by low arterial oxygen saturations. Five of 14 (36%) animals measured had oxygen saturations below 90%. Bradycardia (heart rates <45 BPM) was an expected finding in 11 (55%) immobilizations. Induction, recovery and muscle relaxation of each immobilization was graded. Premature arousal, which occurred in six (30%) animals, was the only problem associated with the immobilizations. Butorphanol/xylazine is a recommended protocol for immobilization of calm, free-ranging tapirs lasting less than 30 min. Supplemental intravenous administration of ketamine is recommended for longer procedures. Nasal insufflation of oxygen is recommended.

Key words: Baird's tapir, butorphanol, cardiopulmonary effects, immobilization, ketamine, *Tapirus bairdii*, xylazine.

INTRODUCTION

In captivity, a wide range of drug combinations are being used to immobilize tapirs (Fowler and Miller, 1986; Short, 1987; Trim, 1998; Janssen et al., 1999). Due to their elusive nature and nocturnal behavior, capturing tapirs to attach radiotransmitting collars has been done infrequently. Until recently, immobilization of free-ranging tapirs have been accomplished with an etorphine/acepromazine combination (Immobilon®, Reckitt and Colman, Hull, England) (Williams, 1979, 1984). The main advantage to that protocol is a short induction, ideal for field use. Problems with this protocol include the potential for apnea and the human safety hazard present when using etorphine, especially during night captures. The primary goal of this project was to devise an alternative anesthetic protocol that would (1) be safe for tapirs, (2) be safe for the researchers, (3) provide both rapid induction and recovery to avoid predation, trauma or accidental drowning, (4) provide adequate immobilization and muscle relaxation, (5) be reversible, and (6) be relatively inexpensive.

A butorphanol/xylazine combination was used during 20 immobilizations of 16 Baird's tapirs (*Tapirus bairdii*) in Corcovado National Park (Costa Rica; 8°29′00″N, 83°35′00″ W). This combination of drugs was chosen because it fulfilled the above objectives and has been extensively applied and researched in the tapir's close phylogenetic relative, the horse (Muir and Hubble, 1991). The butorphanol/xylazine protocol afforded limited muscle relaxation; therefore, ketamine, another drug commonly used in the horse, was added to the protocol. This study allowed for the analysis of the efficacy of this protocol in a field situation and initiated an in-depth evaluation of its cardiopulmonary effects.

MATERIALS AND METHODS

Twenty immobilizations of 16 tapirs (9 males, 7 females), using either butorphanol/xylazine or a butorphanol/xylazine/ketamine (Torbugesic®, Fort Dodge, Iowa, USA, Rompun®, Bayer, Kansas, USA and Ketaset®, Fort Dodge, Iowa, USA) protocol, were performed between March 1996 and February 1998. The purpose of the immobilizations was to radiocollar the tapirs to record their habitat use, movement patterns and basic ecology. Biological samples including ectoparasites, blood, skin biopsies, rectal, vaginal and preputial swabs for bacterial cultures were collected at the time of immobilization as part of a separate health assessment study.

Tapirs were attracted to capture areas at night with ripe bananas. Tree platforms were constructed 10 to 15 m above the capture area. Immediately after a tapir entered a capture area and began eating, more bananas were thrown to maintain the tapir's interest. Bananas were continually thrown after the animal was darted until it showed visible drug effects. Animals were darted using a CO_2 powered rifle (DanInject®, Wildlife Pharmaceuticals, Fort Collins, Colorado, USA). Disposable darts ("P" type, Pneu-Dart Inc., Williamsport, Philadelphia, Pennsylvania, USA) with 1.5 inch barbed or gelatin collared needle were used. One animal was inadvertently trapped in a 1.5 × 0.75 × 1.0 m box trap being utilized for white-lipped peccary (*Tayassu pecari*) research. This tapir was approached and darted through the box trap. Once an animal became sternal, it was approached and a blindfold was applied to reduce visual stimuli. Cotton gauze squares were inserted in the ears to reduce auditory stimuli.

The animals were darted with a mixture of butorphanol and xylazine. Due to field work logistics, the doses reported in this study are based on estimated body weights. The animals in this study were estimated to be between 200 to 300 kg. Three awake animals were later weighed by coaxing them with bananas to a bar scale. The body weights of those three animals fell within the estimated body weight range. Mean (±SE) dose of butorphanol was 48 ± 1.84 mg/animal. Mean dose of xylazine used was 101 ± 2.72 mg/animal. Ketamine was utilized in 13 of the 20 immobilizations. It was delivered either intramuscularly in 6 cases or intravenously in 7 immobilizations. Mean dose of ketamine per animal was 187 ± 40.86 mg. Intravenous ketamine was administered via a 20 to 22 g catheter via hand syringe in the auricular vein. The use and dose of ketamine was judged subjectively based upon the degree of sedation desired and the length of time needed for immobilization. In general it was given in increments of 25 mg to produce the effect desired. Either yohimbine (Yobine® Lloyd laboratories, Shenandoah, Iowa, USA) or tolazoline (Tolazine®, Lloyd laboratories) was used to reverse the alpha-2 adrenergic agonistic effects of xylazine. Yohimbine was used in six immobilizations. Mean dose of yohimbine used was 34 ± 0.61 mg/animal. Tolazoline was used in seven immobilizations. Mean dose of tolazoline was 12 ± 10.27 mg/animal. Naltrexone (Trexonil®, Wildlife Laboratories, Fort Collins, Colorado, USA) was used to reverse the opioid agonistic effects of butorphanol at a mean dose of 257 ± 16.19 mg/animal. Reversal agents were administered intramuscularly by hand syringe no sooner than 15 min from the last administration of ketamine.

Time from dart impact to first visible effect (ataxia, inability of prehend bananas), time to sternal recumbency and total immobilization times were recorded. Time to return to sternal recumbency and time to standing were recorded. Each immobilization was rated with a subjective numerical score for the induction, recovery and degree of muscle relaxation. Induction was rated from 1 to 4 where a 1 was assigned to animals that walked away after having been darted or did not become sternal within 20 min. A score of 2 was given to an animal that remained in the bait area and became sternal in less than 20 min. Animals that remained in the capture area and became sternal in less than 17 min received a score of 3. The best score, 4, was given to animals that became sternal <14 min. Muscle relaxation was scored similarly from 1 to 4 where an animal with a score

of 1 meant that it had sat or stood up prior to reversal. A score of 2 was given if the animal voluntarily moved limbs or head but did not sit or stand up prior to reversal. The score of 3 was given to animals that moved its ears, mouth or vocalized but did not move its head or limbs or sat/stood up prior to reversal. The best score, 4, was given to those tapirs that never voluntarily moved prior to reversal. Recovery ratings were scored from 1 to 4 with a score of 1 denoting that after the reversal agents were given, the animal walked away from the capture area with a severe degree of ataxia. The score of 2 was given if the animal walked away with a moderate degree of ataxia. Animals that exhibited only a mild degree of ataxia when they left the capture area recieved a score of 3. Those animals that calmly walked away without any noticeable ataxia or resumed eating in the capture area received a score of 4.

Heart rate, oxygen saturation as determined by pulse oximetry, indirect oscillometric arterial blood pressure, lead II electrocardiogram, and body temperature were measured using a portable bedside monitor (NPB-4000, Nellcor Puritan Bennett Inc., Pleasanton, California, USA). These values were recorded as soon as the animal was approached, approximately fifteen min from dart impact, and continued to be recorded at five minute intervals whenever possible. Pulse oximetry was applied to labia, prepuce, nasal planum or, most often, tongue. A cuff bladder width to forelimb circumference ratio of 40% was used for indirect arterial blood pressure determination. Respiratory rate was counted by direct observation of respiratory excursions. Arterial blood was obtained from the facial artery. Four arterial blood gas analysis were obtained. Two readings from one animal and two readings from two additional animals. Additionally, one venous blood gas analysis was done. Blood gas analysis was done with the use of a portable clinical analyzer (i-STAT, Heska, Waukesha, Wisconsin, USA). Blood gas analysis also included electrolytes, hematocrit and hemoglobin measurements.

RESULTS

Mean ± SE time from dart impact to visible effect was 4.6 ± 0.5 min. Mean time to sternal recumbency was 12.2 ± 1.1 min. Mean time a tapir was immobilized was 45.6 ± 3.6 min. Mean time to return to sternal recumbency and standing in animals that received yohimbine and naltrexone was 3.2 ± 1.1 and 5.3 ± 1.5 min respectively. Mean time to sternal and standing in animals that received tolazoline and naltrexone was 1.6 ± 0.4 and 3.1 ± 0.5 min respectively.

Thirteen of 20 (65%) inductions received the highest score, 4. Two (10%) were rated with a score of 3. Five (25%) were given a score of 1. Of those, only one animal left the capture area before drugs took effect. Another animal remained in standing sedation until he was manually pushed to lateral recumbency both of the times he was immobilized. Twelve (60%) immobilizations were given a score of 4 for muscle relaxation. One was given a score of 3. One was given a score of 2. Six (30%) animals had a muscle relaxation score of 1. Four out of those six animals received ketamine. Only one of those four received ketamine intravenously. Sixteen (80%) recoveries were rated with a score of 4. Three (15%) recoveries received a score of 3. One (5%) was given a score of 2. This score was given to a tapir who, although she remained in the capture area, was ataxic for 40 min.

Heart rates ranged between 28 to 108 beats per minute. Respiratory rates measured between 8 to 21 breaths per minute. Pulse oximeter readings ranged between 54% to 100%. Indirect blood pressure measurements were between 202 to 99 systolic, 118 to 46 diastolic and 66 to 127 mm Hg mean arterial pressure. Body temperatures remained within 35.5 to 38.6 C. No arrhythmias were detected on electrocardiograms. In 5 of 14 (36%) animals measured, at least one reading from the pulse oximeter fell below 90% oxygen saturation. Table 1 summarizes the anesthetic monitoring data per animal. Arterial blood pH ranged from 7.366 to 7.416. Arterial partial pressure of carbon dioxide ranged from 46 to 50.5 mm Hg. Arterial partial pressure of oxygen ranged from 82 to 88 mm Hg. Arterial concentration of sodium ranged from 133 to 135 mmol/L. Arterial potassium concentrations ranged from 2.6 to 3.9 mmol/L. Hematocrit, as estimated by the portable clinical analyzer, ranged from 20 to 25%. Hemoglobin concentra-

TABLE 1. Anesthetic monitoring data taken during 20 immobilizations of 16 individual Baird's tapirs. Tapirs are listed in the order they were immobilized. The number of readings taken for each parameter follows the range reported in parentheses.

TAPIR	HR[b]	RR[d]	SPO2[e](%)	ECG[f]	BP[h]	T°[i]
023	28–32 (4)	12–16 (4)	N/A	N/A	N/A	36.8–37.0 (4)
018	32–40 (4)	12 (4)	N/A	N/A	N/A	36.1 (1)
010	36–40 (4)	20 (4)	N/A	N/A	N/A	37.4–37.8 (4)
021	N/A[c]	N/A	N/A	N/A	N/A	37.4–37.8 (2)
021 (II)[a]	N/A	N/A	N/A	N/A	N/A	N/A
006	45–57 (2)	18 (2)	75–93 (2)	N[g]	202/72 (2)	35.6–35.9 (2)
016	41 (2)	18 (2)	96–97 (2)	N	136/47–134/66 (2)	35.5–36.2 (2)
014	40 (1)	18 (1)	N/A	N	N/A	36.9 (1)
086	35–40 (4)	9–12 (4)	54–80 (4)	N/A	N/A	38.6 (3)
018 (II)	34–39 (28)	16–50 (15)	88–90 (28)	N	N/A	36.9–37.2 (27)
023 (II)	40–63 (14)	16–20 (14)	76–94 (14)	N	160/118–106/72 (12)	37.0–37.2 (13)
010 (II)	56–66 (8)	16–20 (6)	93–95 (8)	N	154/46–119/61 (7)	35.9–36.9 (5)
030	43–75 (9)	14–21 (11)	86–97 (8)	N	101/47 (1)	37.5–38.1 (5)
070	75–89 (3)	12–20 (4)	90–94 (3)	N	155/102 (123) (2)	37.2 (3)
014	37–45 (14)	14–19 (16)	92–100 (16)	N	156/88 (113)–118/71 (87) (8)	N/A
041	91–108 (7)	14–20 (7)	82–91 (7)	N	125/73 (90)–119/77 (93) (8)	36.0–36.4 (4)
122	43–49 (9)	8–12 (10)	89–95 (9)	N	160/104 (125)–105/107 (127) (8)	37.1–37.2 (7)
164	84–103 (3)	8–12 (8)	88–94 (7)	N/A	147/107 (123)–99/54 (67) (7)	36.3–36.6 (5)
134	39–42 (8)	12–18 (10)	90–92 (8)	N	179/55 (79)–168/60 (83) (3)	36.0–36.6 (5)
132	35–40 (8)	8–12 (7)	91–93 (8)	N	109/67 (81)–106/60 (75) (6)	36.5–36.7 (6)

[a] A roman numeral II denotes that animal's second immobilization.
[b] HR = heart rate expressed in beats per minute.
[c] N/A = not applicable.
[d] RR = respiratory rate expressed in breaths per minute.
[e] SPO2(%) = oxygen saturation as measured by a pulse oximeter expressed in percent.
[f] ECG = electrocardiogram.
[g] N = no arrhythmias detected.
[h] BP = indirect blood pressure measurements expressed in systolic/diastolic and as mean blood pressure ( ) whenever possible.
[i] T = body temperature expressed in degrees Celsius.

tions ranged from 7 to 9 g/dl. Base excess measured between 1 and 6 mmol/L. Bicarbonate concentration ranged from 27 to 31 mmol/L. Total carbon dioxide measured from 28 to 32 mmol/L. Oxygen saturation measured from 94 to 96%. Arterial calcium concentrations ranged from 1.23 to 1.3 mmol/L. One venous blood gas sample was analyzed. The venous pH was 7.366. The venous partial pressure of carbon dioxide was 47.7 mm Hg. The venous partial pressure of oxygen measured 64 mm Hg. The concentration of sodium was 134 mmol/L. Venous concentration of potassium was 3.7 mmol/L. The hematocrit was measured to be 25%. The hemoglobin level was 9 g/dl. The base excess in this sample was 2 mmol/L. Venous bicarbonate measured 27 mmol/L. The total carbon dioxide measured 29 mmol/L. The oxygen saturation was 91%.

DISCUSSION

The butorphanol/xylazine combination proved to be an effective method of immobilizing free-ranging Baird's tapirs. The rate of induction was adequate; however, the use of bananas as bait was found to be crucial to the success of the induction period. Bananas ensured that the animal was less distracted by the dart and remained in the capture area until the drugs took effect. The one animal that left the capture area prematurely, did so because leaf cutter ants (*Atta* sp.) were biting his lips, nose and tongue. These ants also were attracted to the banana bait. One tapir, who remained in standing sedation for over 20

min, was given the lowest score for both of his inductions. However, he was pushed to lateral recumbency without premature arousal in both instances and was scored with the highest scores for both recovery and muscle relaxation.

The butorphanol/xylazine combination provided sufficient sedation for the attachment of radiocollars. The induction of the last six animals received the best and most consistent scores. Those animals received 50 mg/animal of butorphanol and 100 mg/animal of xylazine. The butorphanol/xylazine protocol, however, was not of sufficient quality and duration to allow for extensive data and biological sample collection. Loud noises, movement, pain and other stimuli caused premature arousal in some instances. This is typical for low doses of an agonist/antagonist narcotic and an alpha-2 agonist combination in horses (Muir and Hubble, 1991). The analgesic effects of xylazine do not typically last more than 45 min in horses (Muir and Hubble, 1991). In our study, premature arousal occurred after 40 min if no supplemental drugs were administered.

In general, premature arousals were noted on animals on which either no ketamine was used, or the ketamine was administered at low doses IM. This is illustrated by the fact that the last six animals received the best and most consistent muscle relaxation and recovery scores. Those animals received ketamine intravenously consistently. The dose range of ketamine given IV to the last six animals ranged between 100 to 250 mg/animal. This dose was initially based on IV equine doses and subsequently on the subjective evaluation of the depth of sedation needed to manipulate the immobilized animals. The lower dose was sufficient for minor manipulations such as rolling the animal to lateral recumbency. Additional ketamine was utilized for major manipulations such as pushing or pulling the animal during measurements and sample gathering. Therefore, we feel that intravenous administration of ketamine is the optimum way to produce better muscle relaxation without reducing the quality of the recovery if given with enough time to allow redistribution prior to administering the reversal agents.

Reversal of butorphanol/xylazine was achieved with either yohimine/naltrexone or tolazoline/naltrexone. When comparing recovery times of animals reversed with tolazoline to those reversed with yohimbine, no significant difference was observed in either return to sternal recumbency ($F = 1.64$, $df = 1,18$; $P < 0.25$; ANOVA with different sample sizes, Sokal and Rolf, 1995) or time to standing ($F = 1.31$; $df = 1,18$; $P < 0.50$).

The cardiopulmonary effects of this protocol are reviewed in Table 1. Due to the use of different dosing protocols, cardiopulmonary data cannot be grouped. The only reported heart rate for a tapir is 45 bpm (Lee, 1993). The bradycardia exhibited by some individuals can be attributed to a reduction in central sympathetic tone caused by xylazine. An adult resting horse's heart rate is 28 to 44 bpm (Reed and Warwick, 1998). Perhaps further sampling of normal, non-anesthetized tapir heart rates would yield a wider range of rates and exclude some of the individuals currently thought to have been bradycardic. Subjectively, heart rates were higher in some of the animals in which ketamine was used, which might be due to ketamine's sympathomimetic effects.

To our knowledge, respiratory rates of tapirs have not been reported; however, observed respiratory rates were within the expected range for adult resting horses (Reed and Warwick, 1998). However, arterial partial pressure of carbon dioxide levels, when measured, was slightly elevated, indicating hypoventilation. Although in the four animals in which blood gas analysis was performed the arterial partial pressure of oxygen was above 60 mm Hg, five of the 14 animals on which oxygen saturation data was obtained had at least one reading below 90%. Assuming that the oxygen-hemoglobin disassociation curve is

the same in tapirs as other animals, oxygen saturation below 90% reflects an arterial pressure of oxygen of 60 mm Hg or below. Hypoxemia is defined in domestic mammals by an oxygen arterial partial pressure of 60 mm Hg. Therefore, those animals with pulse oximeter readings below 90% may have approached hypoxemic states. Oxygen-hemoglobin disassociation curves are species-specific and further work would have to be done to validate this point.

The hypercapnea and the lower oxygen saturations observed are probably due to the combined respiratory depressant effects of both xylazine and butorphanol, as well as the effect of lateral recumbency on ventilation-perfusion matching. The tapirs in this project were kept in lateral recumbency for more than half of their immobilization periods to accurately acquire morphometric measurements and have access to the medial saphenous vein. The use of supplemental oxygen administration should be considered for both extended procedures and immobilization of compromised tapirs.

Blood pressure readings indicated that the mean arterial pressure was probably adequate for organ perfusion (Grandy, 1987). A transient period of hypertension typically caused by alpha-2 agonists was not observed during this project; however, both the fact that xylazine was administered intramuscularly and that the anesthetic monitoring did not begin until at least 15 min from dart impact could have obscured this finding. Hematocrits, hemoglobin concentrations and electrolytes as measured by the portable clinical analyzer were within the normal ranges for Baird's tapirs (Fowler, 1999). Body temperatures also remained within normal values reported for tapirs (Lee, 1993).

In summary, a butorphanol/xylazine combination was a safe protocol for the immobilization of free-ranging tapirs. Since this is an ongoing study, our plan is to use the protocol used for the last six animals on all future captures. One must note that the weights of the animals in this study were estimated. The three actual weights obtained were done so at later dates while the animals were not anesthetized. This study and thus, the analysis of our data would be more meaningful if actual weights were measured at the time of immobilization. However, given the remoteness of the study area, and the inconsistent availability of a portable scale that would accommodate large animals, this was not possible. It must be taken into account that the butorphanol/xylazine protocol has the potential to produce a hypoxemic state and nasal insufflation of oxygen is recommended and planned for future captures. Due to the short sedation period afforded by this protocol, the use of butorphanol/xylazine alone should only be utilized in immobilizations lasting less than 30 min. Ketamine can be safely used to lengthen the immobilization period and aid in the depth of sedation. However, since ketamine is not reversible and may produce prolonged recoveries, one should allow enough time for it to be redistributed prior to reversing the butorphanol and xylazine. In short, the success of this protocol was attributed to (1) distracting the animals with the appropriate bait, in this case, bananas, (2) reducing the sensory stimuli by reducing unneccessary noise, applying a blindfold and inserting cotton gauze in the tapir's ears and (3) using intravenous ketamine.

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