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Authors: Vogel, Ingrun, de Thoisy, Benoît, and Vié, Jean-Christophe

Source: Journal of Wildlife Diseases, 34(3) : 555-566

Published By: Wildlife Disease Association

URL: <https://doi.org/10.7589/0090-3558-34.3.555>

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## COMPARISON OF INJECTABLE ANESTHETIC COMBINATIONS IN FREE-RANGING TWO-TOED SLOTHS IN FRENCH GUIANA

Ingrun Vogel, Benoît de Thoisy, Jean-Christophe Vié

Programme Faune Sauvage, EDF/CNEH, Savoie Technolac, 73373 Le Bourget-du-Lac cedex, France

**ABSTRACT:** Immobilization was studied in 202 free-ranging two-toed sloths (*Choloepus didactylus*). All the sloths were in good condition with a body weight  $>2$  kg, and were anesthetized for a variety of minor clinical procedures. Intramuscular anesthetic combinations included 0.1 mg/kg acepromazine + 10 mg/kg ketamine (A/K,  $n = 30$ ), 1 mg/kg xylazine + 10 mg/kg ketamine (X/K,  $n = 89$ ), 10 mg/kg tiletamine/zolazepam (T/Z,  $n = 37$ ), and 0.04 mg/kg medetomidine + 3 mg/kg ketamine (M/K,  $n = 46$ ) antagonized by 0.2 mg/kg atipamezole. The animals were quiet during the induction stage and complete recumbency was reached in (mean  $\pm$  SD)  $2.5 \pm 2.0$  min with A/K,  $2.7 \pm 1.7$  min with X/K,  $1.8 \pm 0.6$  min with T/Z, and  $2.5 \pm 5$  with M/K. Utilization of A/K was not satisfactory because of poor anesthetic level and lack of muscle relaxation. T/Z induced immobilization was characterized by deep anesthesia and good myorelaxation, but often was associated with irregular respiration and low relative oxyhemoglobin saturation values ( $SpO_2$ ). Ketamine in combination with  $\alpha_2$ -agonists, xylazine or medetomidine, provided suitable anesthesia, with good to excellent muscular relaxation, good analgesia, high  $SpO_2$  values, moderate bradycardia, but strong bradypnea with medetomidine. Anesthesia with M/K was reversed after 41.6 min of immobilization with atipamezole. Calm recoveries were obtained and the animals were able to hang up after  $10.0 \pm 7.9$  min. The first signs of arousal were observed within an average of 43 to 51 min after the injection of the three other combinations. Recoveries from X/K immobilization were quiet; sloths held on after 34 min. With T/Z, recovery duration was long and very irregular at  $76.7 \pm 31.3$  min, some animals required 3 hr before being able to hang up. Finally, ketamine in association with an  $\alpha_2$ -agonist appeared to give the best chemical immobilization in wild two-toed sloths for 40 min procedures including minor surgery.

**Key words:** Acepromazine, *Choloepus didactylus*, immobilization, ketamine, medetomidine, tiletamine-zolazepam, two-toed sloth, xylazine.

### INTRODUCTION

Two-toed sloths (*Choloepus* spp.) are arboreal Xenarthra inhabiting Neotropical rainforests. Sloths have physiological particularities including a low basal metabolic rate (McNab, 1985), poikilothermism (Britton and Atkinson, 1938), a ruminant-like stomach (Goffart, 1971) and the possibility to support long periods of apnea (Irving et al., 1942). There are some reports of anesthesia of captive two-toed sloths with a limited number of samples and without study of anesthetic effects. Chemical immobilizations with pentobarbital alone or in combination with promazine were the first reported (Meritt, 1972, 1974; Toole, 1972; Wallach and Boever, 1983). Ketamine has been used alone or in association with diazepam, acepromazine, xylazine or gas anesthesia to achieve satisfactory immobilization (Wallach and Boever, 1983; Dedet et al., 1988; Rappa-

port and Hochman, 1988; Gillespie, 1993; Wallace and Oppenheim, 1996). Difficulties may occur in controlling volatile anesthetics because of the ability of the sloths to cease breathing for up to 20 min (Irving et al., 1942). One study reported the utilization of a mixture of tiletamine and zolazepam (Bush and Gilroy, 1979).

During the course of a wildlife rescue and research project, we had a unique opportunity to capture and study a large sample of southern two-toed sloths (*Choloepus didactylus*). A 30 to 40 min-long immobilization was necessary to conduct a variety of clinical procedures. Four anesthetic combinations were used, including acepromazine/ketamine, xylazine/ketamine, tiletamine/zolazepam and medetomidine/ketamine. Medetomidine was antagonized with atipamezole. Acepromazine, a phenothiazine derivative, is a potent neuroleptic agent with low toxicity. This drug has been given to a wide variety of species

TABLE 1. Number of two-toed sloths, sex ratio (males : females) and total dose for acepromazine/ketamine (A/K), xylazine/ketamine (X/K), tiletamine/zolazepam (T/Z), and medetomidine/ketamine (M/K) anesthetic drug combinations used for field immobilization in French Guiana.

	Group A/K		Group X/K		Group T/Z		Group M/K	
Number of sloths	30		89		37		46	
Sex ratio	12:18		37:52		16:21		20:26	
Total dose (mean $\pm$ SD), in mg/kg	0.1 $\pm$ 0.0	10.1 $\pm$ 0.05	1.0 $\pm$ 0.15	9.9 $\pm$ 1.5	10.1 $\pm$ 0.5	0.039	0.005	3.0 0.3

(Lumb and Jones, 1984) and has been rarely used alone in wild animals (Fowler, 1986). Its use to achieve neuroleptanalgesia is possible by combination with a powerful analgesic such as ketamine (Löscher et al., 1991). Xylazine is the most frequently used immobilizing drug in zoo and wild animals (Gölthenboth, 1995). It acts synergistically with most anesthetics by reducing the total dose needed to achieve immobilization (Greene and Thurmon, 1988). The combination of xylazine and ketamine is widely used because it produces relatively safe and reliable short-term anesthesia (Hatlapa and Wiesner, 1982; Lumb and Jones, 1984). Tiletamine, a dissociate cyclohexamine anesthetic combined in a 1:1 ratio with the benzodiazepine zolazepam has many advantages such as the small volume required, ease of administration, wide safety margin, and dose-related effects, thus making this combination popular as an immobilizing drug for use in many wild species (Lin et al., 1993; Gölthenboth, 1995). Medetomidine, a potent and selective  $\alpha_2$ -adrenoceptor agonist, potentiates ketamine to a greater extent than xylazine (Arnemo et al., 1993). It allows to reduce the effective ketamine dose by as much as 75% (Jalanka, 1991). Medetomidine has been used both alone and in combination with ketamine for rapid induction of reversible immobilization in various zoo and wild species (Jalanka and Röken, 1990). Rapid reversal is achieved by using the potent and selective  $\alpha_2$ -adrenoceptor antagonist atipamezole (Jalanka and Röken, 1990). The primary objective of this study is to compare these injectable anesthetic combinations in

order to develop a safe and effective short-time anesthesia procedure for wild two-toed sloths.

## MATERIALS AND METHODS

### Animals

A large number of mammal and snake species, and some birds and amphibians, were captured at the Petit Saut dam on the Sinnamary river (4°45'–5°04'N, 52°55'–53°15'W, French Guiana) during the flooding of the forest between January 1994 and July 1995. Two-toed sloths were caught by climbing in the trees either with a lasso or by cutting branches down and were picked up in the water from boat with lasso or net. After capture, they were put immediately into individual cages and transferred to the veterinary facility after a pirogue trip lasting no more than 2 hr. At their arrival, they were held in a quiet place for a minimum of 1 hr.

All mammals were anesthetized for a variety of minor clinical procedures including blood sampling, skin biopsy, measurements and tattooing. Anesthetic data were recorded precisely in 202 healthy two-toed sloths. Body weight averaged 6.92 kg (SD = 2.05 kg) and ranged from 2.22 to 11.80 kg. The number of sloths and sex-ratio for each group are summarized in Table 1. The unbalanced sex-ratio reflects the demographic structure of wild *Choloepus* sp. populations (Meritt and Meritt, 1976). Pregnancy diagnostic was done by physical (rectal and abdominal) palpation during anesthesia. Fourteen females were pregnant.

### Anesthetic drugs

Four drug combinations were used. Group A/K consisted of 0.1 mg/kg acepromazine maleate (Calmivet®, Vétroquinol S.A., B.P. 189, 70204 Lure cedex, France) plus 10 mg/kg ketamine hydrochloride (Ketamine 500 U.V.A.®, Laboratoires U.V.A., 94200 Ivry-sur-Seine, France). Group X/K was composed of 1 mg/kg xylazine (Rompun 2%®, Bayer Pharma, 49–51, quai de Dion-Bouton, 92815 Puteaux cedex,

France) plus 10 mg/kg ketamine hydrochloride. Group T/Z was 10 mg/kg tiletamine/zolazepam (Zoletil 50®, Reading, B.P. 27, 06511 Carros cedex, France). Group M/K was a formulation of 0.04 mg/kg medetomidine hydrochloride (Domitor®, Pfizer Corporation, 91407 Orsay, France) plus 3 mg/kg ketamine hydrochloride. When the procedures were completed, 0.2 mg/kg atipamezole hydrochloride (Antisedan®, Pfizer Corporation, 91407 Orsay, France) were used for reversal.

### General procedures

A 30 to 45 min-long immobilization was expected. The animals were not fasted before anesthesia. They were weighed with the cage prior to drug administration. The exact body weight ( $\pm 20$ g) was determined after anesthesia with a digital balance (Table 1). These aggressive animals were physically restrained with a lasso for a single anesthetic injection given intramuscularly (i.m.) in the hind leg. Supplement doses of immobilization agents were administered i.m. when needed.

The duration of anesthesia was evaluated by recording the following time intervals after administration of the anesthetic mixture: first signs of ataxia, time of recumbency, complete immobilization (no reaction to stimulation), appearance of first signs of recovery (first movements of the claws) and ability to hang on with at least two limbs. Total immobilization was defined as the time between lack of reaction to stimulation and appearance of first signs of recovery. Recovery period was the time between appearance of first signs of recovery and ability to grasp with at least two extremities. It was impossible to assess return to normal locomotion, i.e., complete recovery, because the animals remained motionless in the cage even after complete recovery.

The following events were monitored during anesthesia. A 0 to 6 degree scale was used to estimate the anesthetic level in which there was no effect (0); moderate sedation (1); heavy sedation (2); light anesthesia (3); complete anesthesia (4) required for our procedures; deep anesthesia (5); and death due to drug overdose (6). The degree of muscle relaxation was evaluated on the basis of ease of opening the mouth and relaxation of leg muscles and expressed as excellent, good, moderate, or poor. Heart and respiration rates, relative oxyhemoglobin saturation values ( $SpO_2$ ) and rectal temperature were recorded 5, 15, 30 and eventually 45 min after injection of the drugs. Heart rate was measured by cardiac auscultation, respiratory rate by direct observation and body temperature by rectal digital thermometer. Ox-

yhemoglobin saturation was monitored using a portable pulse oximeter (model N20 P, Nellcor Inc., Hayward, California, USA), with the sensor located on the tongue. After manipulation, the animal was placed back into its cage where the recovery process was monitored. All visible side effects like seizures, salivation and cyanosis were recorded throughout the experiment. Our own observations on the whole population led us to define the following events as anesthetic related complications: tachycardia with heart rate  $>170$  beats/min, bradycardia with heart rate  $<40$  beats/min, tachypnea with respiratory rate  $>70$  breaths/min, bradypnea with respiratory rate  $<7$  breaths/min, hypoxemia with the relative oxyhemoglobin saturation values  $<80\%$ . The animals were released within 24 hr according to the manipulation time. A post-release survey using visual collars and radiotracking was then conducted for 1 yr.

### Data analysis

Quantitative data are given as mean  $\pm$  standard deviation (SD) and qualitative data are presented as percentages. All statistical analysis were performed with Statistical Analysis Program (SAS Institute, Cary, North Carolina, USA). Calculated  $P$  values  $\leq 0.05$  were considered as statistically significant. In each protocol, serial recordings of clinical parameters (at  $T_5$ ,  $T_{15}$ ,  $T_{30}$  and  $T_{45}$ ) were performed with the Friedman test for the quantitative parameters. The comparisons between data obtained in the different procedures were compared for the quantitative parameters using the General Linear Mean procedures to calculate Least Square Means and were performed with the chi square test for qualitative data.

## RESULTS

### Induction time

All trials produced a smooth and uneventful induction (Table 2). Mean induction times were globally similar for sloths immobilized with A/K, X/K and M/K but T/Z resulted in significantly shorter induction periods. Supplemental doses were needed to achieve induction in one sloth in the A/K and M/K groups, in five of the 89 sloths in the X/K group and in one animal in T/Z group. These animals were excluded from the statistical analysis.

### Anesthetic level and muscle relaxation

No significant differences were recorded in anesthetic levels of X/K, M/K and

TABLE 2. Comparative anesthesia intervals (mean  $\pm$  SD (range)) for two-toed sloths immobilized with acepromazine/ketamine (A/K), xylazine/ketamine (X/K), tiletamine/zolazepam (T/Z) and medetomidine/ketamine (M/K).

Anesthesia times (min)	A/K	X/K	T/Z	M/K
<b>Induction</b>				
Ataxia	1.49 $\pm$ 0.54 <sup>1,4a</sup> (0.52–3.17)	1.27 $\pm$ 0.45 <sup>1,5</sup> (0.50–3.08)	1.01 $\pm$ 0.38 <sup>4,5,6</sup> (0.50–2.50)	1.34 $\pm$ 0.40 <sup>6</sup> (0.50–2.28)
Recumbency	2.82 $\pm$ 2.03 <sup>2</sup> (0.53–11.0)	2.73 $\pm$ 1.73 <sup>7</sup> (1.00–5.00)	1.75 $\pm$ 0.59 <sup>2,3,7</sup> (1.03–3.50)	2.51 $\pm$ 0.54 <sup>3</sup> (1.42–3.83)
Complete immobilization	6.33 $\pm$ 2.25 (4.5–10.17) <sup>b</sup>	3.99 $\pm$ 1.57 <sup>8</sup> (1.82–8.00)	2.8 $\pm$ 1.15 <sup>8,9</sup> (1.33–6.00)	4.13 $\pm$ 1.35 <sup>9</sup> (2.50–8.50)
<b>Spontaneous recovery</b>				
First signs	43 $\pm$ 12 <sup>c</sup> (22.5–70)	50 $\pm$ 17.5 <sup>1</sup> (18–100)	51 $\pm$ 13.5 <sup>2</sup> (32–86)	45.5 $\pm$ 10.5 <sup>d</sup> (35–66)
Hanging up	72 $\pm$ 20 (46–125)	89 $\pm$ 34 <sup>3,5</sup> (38–180)	127 $\pm$ 31 <sup>4,5</sup> (75–200)	63 $\pm$ 15 <sup>e</sup> (45–83)
Total immobilization time	44.4 $\pm$ 10.7 <sup>c</sup> (28.3–50)	46.5 $\pm$ 16.5 (18.5–90.0)	48.7 $\pm$ 13.8 (27.0–83.7)	43.2 $\pm$ 12.1 (30.0–62.2)
Recovery time	29 $\pm$ 13 <sup>c</sup> (11–68)	40.2 $\pm$ 25.4 <sup>1</sup> (2–138)	76.7 $\pm$ 31.3 <sup>1</sup> (27–159)	9.3 $\pm$ 8.5 (1–21) <sup>e</sup>

<sup>a</sup> Mean values with same superscripts (1,2,3, etc.) are significantly different ( $P < 0.05$ ).

<sup>b</sup> Reached only by 1/3 of sample.

<sup>c</sup> Only 1/3 of sample is considered.

<sup>d</sup>  $n = 9$ .

<sup>e</sup>  $n = 4$ .

T/Z. Sixty percent of the animals reached degree 4 or 5 on our scale. X/K, M/K and T/Z respectively induced 4, 12 and 19% of degree 5 on the scale. Utilization of A/K induced poor anesthetic levels: only 36% of the animals in degree 4, 57% in degree 3 and 7% in degree 2.

Regarding myorelaxation, significant differences were observed between all four trials except T/Z and X/K. Group A/K showed a lack of muscle relaxation. At the beginning of the anesthesia the sloths remained tense and rolled up into a ball. After approximately 5 min, a low muscle relaxation appeared, which made the manipulation of the animals possible. A poor, moderate or good myorelaxation were observed respectively in 15, 12, and only one of the 28 sloths. T/Z and X/K resulted in a satisfactory myorelaxation (with 70% good or excellent myorelaxation). M/K induced the highest degree of relaxation. No animal showed poor myorelaxation and only one was recorded as moderate.

#### Clinical data

Figure 1 shows the comparison between the effects of A/K, X/K, M/K, and T/Z on rectal temperature, heart rate, respiratory rate, and relative arterial oxygen saturation.

Initial rectal temperature was highly variable: 32 C to 38 C. In all groups, there was a significant decrease (0.5–1.3 C) in the mean rectal temperature during the immobilization period. There was no significant difference between A/K, X/K, M/K and T/Z with respect to mean rectal temperature recorded 5, 15, 30, 45 min after injection.

With A/K, no significant alteration of heart rate was recorded during anesthesia period. With ketamine in association with an  $\alpha_2$ -agonist, there was a significant decrease in heart rate: 61  $\pm$  15 to 46  $\pm$  13 bpm in X/K group, 61  $\pm$  17 to 47  $\pm$  10 bpm in M/K group. Bradycardia was recorded with X/K combination in six animals and in two individuals after M/K ad-

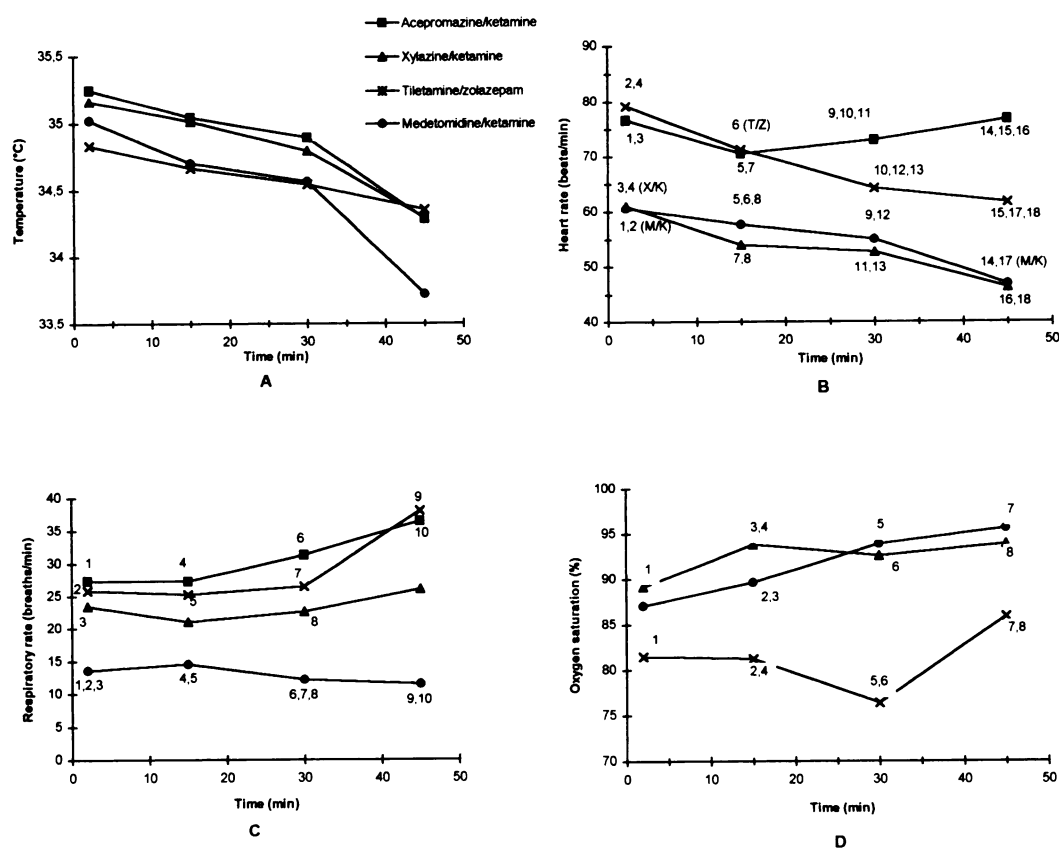


FIGURE 1. Mean values for rectal temperature (A), heart rate (B), respiratory rate (C) and oxyhemoglobin saturation (D) at 15 min intervals for wild two-toed sloths anesthetized with acepromazine/ketamine, xylazine/ketamine, tiletamine/zolazepam, and medetomidine/ketamine. Values with same superscript numbers are significantly different ( $P < 0.05$ ).

ministration. Administration of T/Z also was associated with a significant, persistent decrease in heart rate, but less pronounced. The mean heart rate dropped gradually from  $79 \pm 21$  bpm to  $62 \pm 16$  bpm, down to 34 bpm in one animal.

Respiratory rates were highly variable during anesthesia from one sloth to another, mainly with T/Z. Nevertheless, in all groups, mean values remained stable throughout anesthesia. M/K induced significantly lower mean rates than did T/Z, A/K and X/K. The initial mean respiratory rate was  $14 \pm 8$  bpm and decrease non-significantly to  $11.5 \pm 5$  bpm. Slight anesthetic related complications were observed with this last combination: brief periods of apnea observed during the first

few minutes in one animal and bradypnea in four additional individuals. The highest mean respiratory rates were observed with A/K. Tachypnea was recorded on 13 occasions either at the beginning or at the end of anesthesia. Mean values observed with A/K were similar to those with T/Z and X/K. Tachypnea also was observed in four and three animals belonging to the X/K and T/Z groups respectively. Two individuals in group X/K and group T/Z exhibited bradypnea. One animal had an irregular respiratory rate with the X/K combination.

Relative arterial oxygen saturation ( $SpO_2$ ) was not recorded during A/K immobilization due to technical problems. The animals anesthetized with T/Z showed

TABLE 3. Reversal of M/K anesthesia with atipamezole given intramuscularly (min) in 23 two-toed sloths.

	Mean $\pm$ SD	Range
Time of atipamezole injection (after anesthetic injection)	41.6 $\pm$ 8.9	31.0–68.0
First signs of recovery (after Atipamezole injection)	4.5 $\pm$ 1.8	0.5–7.4
Hanging up (after Atipamezole injection)	10.0 $\pm$ 7.9	3.7–29.5

the lowest mean values for the duration of immobilization. The initial value was  $81 \pm 12\%$ ; it reached its lowest level after 30 min ( $76 \pm 11\%$ ), and increased during the last 15 min to  $86 \pm 8\%$ . Animals immobilized with X/K and M/K combinations presented similar values increasing from 89 to 94% and 87 to 96% respectively throughout anesthesia. Eight animals (1 in X/K group; 3 in M/K group and 5 in T/Z group) showed hypoxemia; extreme  $\text{SpO}_2$  levels of 50 to 52% were observed in one animal anesthetized with X/K during the immobilization period. However, it was not associated with clinical signs, such as tachycardia, cyanosis, or any other complication.

#### Anesthesia intervals

When the procedures were completed, M/K anesthesia was reversed by atipamezole at a i.m. dose of five times the medetomidine dose (Table 3). Nine sloths did not receive atipamezole in order to observe spontaneous recovery. Eleven sloths showed first signs of spontaneous recovery before the injection of atipamezole; they are not included in the calculation of arousal time. Atipamezole was injected before the first signs of recovery in 23 animals  $42 \pm 9$  min after anesthetic injection. Recovery was quiet; animals were able to hang up after a mean period of 10 min, ranging from 3.7 min to 29.5 min. Three sloths received atipamezole intravenously (i.v.). The recovery was uneventful and shorter; first signs were observed after 0.5 to 1 min, and animals were able to hang up after 1.5 to 2 min. No signs of re-sedation or abnormal behavior like over-alertness were observed within 3 hr after i.m. or i.v. reversal.

In all four groups, mean total immobilization with spontaneous recovery varied between 40 and 50 min (Table 2). With A/K, complete immobilization was successful in only one third of the animals although this result was not representative of the results for the sample as a whole. Thus, this time is not statistically compared to those obtained with other drugs combinations. Mean duration of M/K anesthesia was the shortest ( $43.2 \pm 12.1$  min) followed by X/K ( $46.5 \pm 16.5$  min) and T/Z ( $48.7 \pm 13.8$  min). Some individuals anesthetized with T/Z required over 3 hr to hang up. For all groups, no abnormal behavior was noted within 3 hr after mobility was regained.

#### Side effects

No side effects such as regurgitation, bloating, excessive salivation, cyanosis, were observed during anesthesia. Excitation during recovery was observed on one occasion after A/K injection. However, one animal anesthetized with A/K combination had a prolonged recovery time (2 hr) and was found dead the following day at the release area. Two additional animals anesthetized with M/K and T/Z were found dead a few days after release, although no problem had been observed during anesthesia. The animals were in good body condition and no abnormal findings were recorded at necropsy. Stress induced by capture and translocation to a new habitat was probably important, exact role of anesthesia in the three deaths is difficult to assess. Two pregnant females were anesthetized with A/K, eight with X/K and four with T/Z. They were observed 12 hr after reversal and none showed spontaneous abortion.

## DISCUSSION

A large number of captured sloths was expected to be captured given of their density (Montgomery and Sunquist, 1975). This translocation program provided unique opportunity to use and compare the effects of several anesthetic combinations on a large sample, with some drugs previously used in this species or others that had not been tested.

Two major factors could influence the anesthetic effects and increase individual variations. First, physical condition has a major influence. Because of the deterioration of the environment (defoliation of the trees), animals before capture were facing considerable variation in their habitat. However, to estimate the nutritional status is difficult in two-toed sloths because of absence of a subcutaneous layer of adipose tissue between skin and musculature and low muscle mass (Wislocki, 1928). Therefore, it was only possible to exclude obviously weak animals from the study. Second, stress is responsible for an increase of liberation of  $\text{Ca}^{2+}$  inducing a decrease in the effectiveness of anesthetics on the excitable membranes (Löscher et al., 1991). Stress also increases the activity of sympathetic nervous system and thus increases catecholamines liberation, inhibiting  $\alpha_2$ -agonists (Lance, 1991); so, the induction could be prolonged and stress could result in an increase of body temperature and heart and respiratory rates (Fowler, 1986). The magnitude of the effects of stress is difficult to assess and was probably reduced throughout the study with an increasing knowledge of handling these animals. We assume that these factors could be considered as greatly minimized because of the large sample size.

Combinations A/K and X/K were used at doses recommended by Wallach and Boever (1983). To our knowledge one study used T/Z anesthesia, at a dosage of 3 mg/kg (Bush and Gilroy, 1979). These authors reported long induction time and too short immobilization time for our pro-

cedures and we increased the dose to 10 mg/kg. Numerous studies have shown that medetomidine is effective for the chemical restraint of non-domestic or domestic animals, especially when combined with ketamine (Jalanka and Röken, 1990; Jalanka, 1991) and we used it in other species from French Guiana, including howler monkeys (*Alouatta seniculus*) (Vié et al., 1998), golden-handed tamarins (*Saguinus midas*), and white-faced sakis (*Pithecia pithecia*) (Vié et al., unpub. data), three-toed sloths *Bradypus tridactylus* (P. Chabaud, unpub. data) and kinkajous *Potos flavus* (Fournier et al., 1998). To our knowledge there is no published report on its use in sloths. An initial trial with 2 mg/kg ketamine and 50  $\mu\text{g/kg}$  medetomidine gave poor myorelaxation and a marked bradypnea:  $5.0 \pm 3.2$  bpm after 15 min of immobilization. Effects on the respiratory rate was less pronounced with 3 mg/kg and 40  $\mu\text{g/kg}$ . Two-toed sloths are aggressive animals that can cause severe injuries to handlers or their own carried infants. A short and smooth induction period was required to reduce stress and to decrease the risk of injury both for humans and animals. In the present study, induction periods were calm and satisfactory for all the animals. The i.m. T/Z combination quickly induced complete immobilization in the sloths, as reported in other species (Gray et al., 1974; Eads, 1976). By comparison, with similar dosages, the induction period was 30% longer with M/K, A/K and X/K, and 20% longer with T/Z in three-toed sloths *Bradypus tridactylus* under the same conditions.

Two-toed sloths are incomplete homeotherm animals, with a mean body temperature of 34.5 C (Goffart, 1971). Body temperature varies with outside temperature, especially in periods of inactivity (Britton and Atkinson, 1938). Throughout anesthesia, ambient temperature was around 30 C. At this temperature, the risk of hypothermia in two-toed sloths is very low, because physiological values can reach 24 C, but sunshine (35–40 C) usually is sufficient to increase body temperature up to a le-



thal level (Britton and Atkinson, 1938). Individuals showed very different initial body temperature ranging from 31 to 38.2 C and a slight variation of temperature throughout anesthesia. Thus, we consider that temperature is unsuitable for the study of anesthetic effects of these drugs.

Heart rates range between 70 and 130 bpm (Goffart, 1971). A/K produced minimal influence on heart rate as also shown in three-toed sloths. In contrast, T/Z combination induced a significant decrease in cardiac rates ( $61 \pm 16$  bpm) throughout the experimental period. We recorded similar results in other species including red howler monkeys and three-toed sloths. Nevertheless, effects of TZ on cardiovascular system appear to be greatly variable according to studies and species (Lin et al., 1993). Xylazine and medetomidine exhibit the typical cardiovascular effects of  $\alpha_2$ -agonists compounds when used in combination; they induced centrally mediated bradycardia and peripheral vasoconstriction (Löscher et al., 1991). The stimulating effects of ketamine on the central nervous system (Wright, 1982) should balance the depressive effects of  $\alpha_2$ -agonist compounds. In the two sloth species, ketamine effects could not balance  $\alpha_2$ -agonist effects, as observed in other studies (Jalanka and Röken, 1990; Jalanka, 1991; Fournier-Chambrillon et al., 1997).

The respiratory rate is a very variable parameter in two-toed sloths ranging from 10 to 78 bpm in normal conditions, according to activity (Meritt, 1985). Mean values for X/K, A/K, T/Z remained within physiological interval all along anesthesia, but M/K was associated with a marked depression of respiratory rate due to medetomidine effect (Jalanka, 1991). Bradypnea was observed in 9% of the animals and apnea in one case only. We observed such side effects in other Xenarthra; apnea was occasionally recorded in three-toed sloths and one nine-banded armadillo (*Dasypus novemcinctus*) had evidence of cyanosis. Monitoring of oxyhemoglobin saturation with pulse oximetry has been reported in

many wild species but to our knowledge never in two-toed sloths. This monitoring became routine in zoological medicine but some technical problems occur to adapt the equipment to a large set of species. The precision can be influenced by various parameters such as hypothermia and hypotension (Saint John, 1992). Measurement of the systolic pressure is not possible below 50 mm Hg (Erhardt et al., 1989). Unfortunately, there is no specific data on blood pressure in two-toed sloths, although it is supposed to be low (Goffart, 1971). The pigmentation of tissue of the sensor localization is also important (Saint John, 1992). It has been suggested that the tongue is the best localization of the sensor (Jacobson et al., 1994). Frequently we failed to record measures in two-toed sloths but even more difficulties occurred with three-toed sloths. This could be due to their low body temperature, deep pigmentation of the tongue or low blood pressure, or a combination of all these factors. The decrease of SpO<sub>2</sub> during T/Z anesthesia has been previously reported (Helleyer et al., 1988). In our study, lowest values were observed at T<sub>30</sub>, with values below 70% in seven animals. As in other studies (Helleyer et al., 1988), an increase was observed later. Lowest values (57% for T/Z, 50% for M/K) were not related to clinical signs or any complications. The low basal metabolism rate of sloths compared to other mammals (McNab, 1985) may explain the absence of clinical symptoms despite relative hypoxemia and low values of oxygen blood saturation. A specificity of sloths is to survive a phase of apnea of up to 20 min (Irving et al., 1942). Physiological events during the period of apnea is not well known, however, in non anesthetized animals, there is a strong decrease in heart and metabolic rates. The produced CO<sub>2</sub> is replaced in the blood by lactate and dispersed. Thus, the CO<sub>2</sub> blood concentration remains below the threshold of sensitivity and peripheral and central CO<sub>2</sub> receptors are not stimulated (Irving et al., 1942). Moreover, the high content of myo-

globin in the muscle represents an additional O<sub>2</sub> reserve (Goffart, 1971). In most mammal species saturation values lower than 70% can be dangerous (Mihm and Halperin, 1985), but it does not seem to be a risk factor in two-toed sloths. In one animal with an initial SpO<sub>2</sub> value of 61% and short apnea episodes in the first 8 min, an increase to 95% was observed in 15 min without supplemental oxygen flow. However, low values during a long period (>20 min) could have harmful effects.

As in three-toed sloths, 10 mg/kg ketamine associated with 1 mg/kg acepromazine failed to induce myorelaxation and anesthesia of level 4 in most of two-toed sloths. Similar results were obtained in armadillos (*Dasypus kappleri* and *D. novemcinctus*). Because of the anticonvulsive properties of acepromazine (Lumb and Jones, 1984), another ratio might induce a more satisfactory relaxation and provide the good anesthesia recommended by others (Fowler, 1986). Nineteen percent of T/Z injections induced a level 5 anesthesia. Deep anesthesia of level 5 may induce a useless strain to the organism, that can be avoided in only classical and non-invasive clinical procedures. A less stressful immobilization could be obtained by reducing the dose as observed with three-toed sloths successfully immobilized with 5 mg/kg. As in other species (Lin et al., 1993), myorelaxation with T/Z was good in the two sloth species and armadillos. Despite a low xylazine dose, myorelaxation effects of xylazine (Lumb and Jones, 1984) also were satisfactory in the four species and in another Xenarthra, the collared anteater (Fournier-Chambrillon et al., 1997). Nevertheless, the best results were obtained with M/K and this confirms the excellent myorelaxation properties of this association (Jalanka, 1989, 1990; Jalanka and Rönken, 1991; Arnemo and Soli, 1992; Arnemo et al., 1993; Spelman et al., 1994).

T/Z combination induced a prolonged period of recovery, which was documented in different species (Eads, 1976; Payton and Pick, 1989) and also observed during

our field investigations on howler monkeys and three-toed sloths. Four two-toed sloths needed more than 2 hr for complete recovery. Such a long recovery period is undesirable for field immobilizations, but Bush and Gilroy (1979) using a lower dosage (2–6 mg/kg) in the same species failed to obtain significantly shorter recovery times.

Antagonization of M/K anesthesia with atipamezole was rapid and complete within  $10.0 \pm 7.9$  min in the 23 sloths reversed by i.m. route. Intravenous administration of 0.2 mg/kg atipamezole shortened the recovery after M/K induced immobilization, however the intramuscular route is preferable mainly given the sloths particular limb vascular system, *retia mirabile* (Goffart, 1971), makes intravenous injections difficult under field condition. Atipamezole i.v. has been reported to induce overalterness in carnivores (Jalanka, 1989) or resedation in ungulates (Jalanka and Rönken, 1990). We did not observe such phenomena but this is probably less obvious to diagnose in sloths than in other species. Very few side effects were observed between the immobilization in the veterinary facilities and the release in the nearby forest. One animal was excited during the recovery process and one had a prolonged recovery. It was not possible to observe long term side effects on the majority of the animals.

## CONCLUSIONS

The A/K combination induced a short and calm induction time and resulted in little influence on the cardiovascular system. However, the irregular respiratory rate, the low degree of anesthesia (only sedation) and the poor myorelaxation make it not suitable for *C. didactylus* at the doses we used. The T/Z combination produced the shortest induction time and a deep anesthesia with good muscle relaxation and minimal influence on the cardiovascular functions. But irregular respiratory rate, low values of hemoglobin saturation, and especially, a long recovery were

recorded. Additionally, the perishable solution can be a disadvantage during field work. The use of an  $\alpha_2$ -agonist in combination with ketamine provides the most suitable immobilization for wild two-toed sloths. M/K anesthesia was characterized by a short induction time, a good level of anesthesia and an excellent myorelaxation. Cardiorespiratory depressive effects were recorded, but no clinical complications occurred. Moreover, oxyhemoglobin saturation remained at a high level during the length of immobilization period. Immobilization could be reversed with the antagonist atipamezole resulting in a reduction of recovery period. Bradycardia and irregular respiration were noted with X/K but in contrast to M/K depression of respiration was not recorded. The induction period was short and calm. A satisfactory anesthetic level with good muscle relaxation and excellent values of hemoglobin saturation were recorded. The recovery period was smooth and relatively short. No attempt to antagonize xylazine was made. Thus, we recommend medetomidine 40  $\mu$ g/kg with ketamine 3 mg/kg, or xylazine 1 mg/kg with ketamine 10 mg/kg for immobilization of two-toed sloths, for 30 to 40 min-long handling, including classical clinical procedures, and even minor surgeries.

#### ACKNOWLEDGMENTS

The first author is grateful to H. Wiesner (Münchener Tierpark Hellabrunn Director) for advice during the composition of her thesis on the two-toed sloth. The three authors thank the whole Faune Sauvage staff for capture and manipulation of animals, and especially P. Chabaud for data on three-toed sloths. They also thank Bayer Pharma for supplying Rompun® and Pfizer Corporation for supplying a part of Domitor® and Antisedan®.

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*Received for publication 18 August 1997.*