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## Response of Wild Subantarctic Fur Seal (*Arctocephalus tropicalis*) Females to Ketamine and Tiletamine-Zolazepam Anesthesia

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**ABSTRACT:** This study is the first to compare the anesthetic effects of two cyclohexamines on free-ranging subantarctic fur seal (*Arctocephalus tropicalis*) females. From April to July 1999, 107 females were immobilized for tooth extraction and blood sampling, using either ketamine (Ketalar®,  $n=58$ ) alone or tiletamine-zolazepam (Zoletil 100®,  $n=49$ ) mixture. Animals were injected intramuscularly at mean doses of 2.1 mg/kg for ketamine and 1.1 mg/kg for tiletamine-zolazepam mixture. Individual response to both drugs was highly variable. The dosage required to achieve a satisfactory level of anesthesia was smaller for subantarctic fur seals than for most other species of seals and was less for animals in better body condition. Few side effects were observed during the trials, aside from mild tremors caused by ketamine, and respiratory depression or prolonged apnea caused by tiletamine-zolazepam. We recommend use of ketamine, especially by those with little experience in anesthesia of fur seals. However, precautionary measures should be taken, such as using low doses for animals in good body condition and being prepared for anesthetic emergencies to avoid any casualties.

**Key words:** Anesthesia, *Arctocephalus tropicalis*, cyclohexamines, ketamine, subantarctic fur seal, tiletamine-zolazepam mixture.

Many aspects of scientific investigations and management of adult pinnipeds require chemical restraint or anesthesia to minimize the risks of handling for both the animal and researchers (Laws, 1993) and to reduce stress on the animal. Furthermore, anesthesia is required for surgery. During a study on age determination of live subantarctic fur seals (*Arctocephalus tropicalis*), we needed to immobilize adult females to remove the lower left post-canine 1 using the methods described in Arnbohm et al. (1992). The use of cyclohexamines such as ketamine hydrochloride and tiletamine hydrochloride associated with the sedative zolazepam hydrochloride have been widely reported for a number

of pinniped species (Briggs et al., 1975; Gales and Burton, 1987; Stirling and Sjare, 1988; David et al., 1988; Boyd et al., 1990; Woods et al., 1994), but very few for subantarctic fur seals (Gales, 1989; Ferreira and Bester, 1999). Primary purposes of this study were to: 1) gather information on the response of subantarctic fur seal females to ketamine and tiletamine-zolazepam, 2) determine which drug is the most useful for routine chemical restraint, and 3) discuss precautions to ensure the seals' welfare during chemical immobilizations.

The work was carried out during the autumn-winter 1999 breeding season of subantarctic fur seals at Amsterdam Island (35°55'S, 77°30'E; French Southern and Antarctic Lands) in the southern Indian Ocean. The study colony was located at La Mare aux Elephants, on the north-east side of the island, where one of the largest breeding colonies can be found (Guinet et al., 1994). In order to minimize the risk of drugged seals escaping to the water and drowning, female fur seals were physically restrained in a hand-held conical nylon mesh net, which was suspended by a polyvinyl chloride hoop on a 150 cm aluminum stick. Females were weighed using a spring scale (200±1 kg) and held in a restraint board (Gentry and Kooyman, 1986), with one person straddling the animal and gripping its pectoral flippers prior to anesthetic injection.

The drugs used were either ketamine hydrochloride (Ketalar®, Panpharma, Fougères, France), or tiletamine hydrochloride together with the sedative zolazepam (Zoletil 100®, Virbac, Nice, France) supplied in a crystalline mixture (250 mg of each) together with 5 ml of diluent. A

single dose of either drug was directly injected into the lumbar muscles of the animal (Parry et al., 1981) using a sterile 5 ml syringe with 21 gauge  $\times$  3.5 cm needle. The animal eyes and ears were then covered with a dark sheet to limit photo or acoustic stimuli. Each drugged animal was carefully observed, and its respiratory rate was continually monitored from injection to complete recovery in order to assess its level of immobilization throughout the procedure. These levels were classified by a method similar to that used by Briggs et al. (1975) and Boyd et al. (1990): 1) showing some signs of uncoordinated movement but otherwise normal behavior; 2) head down, unwilling to struggle but still capable of movement when stimulated and handled; 3) incapable of struggle or substantial movement when handled, but responsive to stimulation and head movement possible; 4) completely immobile with regular breathing, and little or no response to tooth extraction stimuli; 5) completely immobile with irregular breathing or short apnea, and no response to tooth extraction stimuli; 6) prolonged apnea leading to death.

The aim was to obtain level 3 to 5 (level 4 was preferred) of anesthesia, for tooth extraction, taking measurements of the animal, and blood collection when necessary. It was not possible to keep accurate records of the anesthesia maintenance time, because seals were immediately released after completion of the procedure, but regular supervision of its physical state was achieved until its complete recovery. On consecutive days, animals were approached to examine long term effect of the drugs by evaluating any behavior towards other animals or field workers. Respiration rate was continuously monitored during handling, with particular attention when level 5 of immobilization was reached. In the case of prolonged apnea with signs of anoxia, such as color changes in the gums, tongue, and conjunctiva, the animal was intubated using a cuffed veterinary endotracheal tube (9 mm diame-

ter) inserted into the trachea and was ventilated by mouth until it recovered the ability to breathe independently (Baker et al., 1990).

To account for individual body condition of female fur seals, we calculated the residual values of the linear regression between body mass and body length as used by Trites (1991). Negative residual values corresponded to individuals in poor condition while positive residual values corresponded to animal in good condition (Georges and Guinet, 2000). All statistical analyses were performed using SYSTAT 7.0 (SYSTAT, 7.0 statistics, SPSS Inc., Chicago, Illinois, USA). We used the Spearman rank correlation to test for relationship between levels of immobilization and the dose of anesthetic injected. The Mann-Whitney *U*-test was used to test for differences in the level of immobilization or the injected dose between body condition categories. Unless otherwise stated, values are reported as means  $\pm$  SD and statistical significance was considered to be  $P < 0.05$ .

From mid-April to July 1999, 107 procedures were performed (Table 1). The mean weight of captured animals was  $48 \pm 9$  kg (range = 21–68 kg).

Fifty-eight females were injected with a mean dose of  $2.1 \pm 0.2$  mg/kg (range = 1.85–2.77 mg/kg) of ketamine and were fully anesthetized within 15 min (range = 10–14 min) reaching a sufficient level of anesthesia to permit tooth extraction (Table 1). The dose injected to reach a satisfactory level of anesthesia was negatively related to body condition (linear regression,  $r = 0.523$ ;  $n = 58$ ;  $P < 0.001$ ). However no significant difference in the anesthetic level achieved in animals in better body condition was noticed ( $Z = -0.55$ ;  $n_1 = 33$ ;  $n_2 = 25$ ;  $P = 0.58$ ). Moreover, no correlation was found between the dose injected and the subsequent level of anesthesia achieved ( $r_s = 0.09$ ;  $df = 56$ ;  $P = 0.49$ ). Some undesirable side effects including mild tremors ( $n = 7$ ) and apnea ( $n = 2$ ) were observed on a few occasions with no adverse consequences for the animal (Table 1). All

TABLE 1. Characteristics of subantarctic fur seal female anesthesia using ketamine hydrochloride alone or a tiletamine-zolazepam mixture.

Anesthetic agent	Level of anesthesia	N	Weight of animal (kg)		Dosage (mg/kg)		Recovery time (min.)		Cases of	
			Mean	SD	Mean	SD	Mean	SD	Tremor	Apnea
Ketamine	3	6	46.8	9.0	2.06	0.12	18	4	3	0
	4	48	46.7	7.7	2.16	0.18	41	6	4	0
	5	4	49.3	10.7	2.08	0.20	56	8	0	2
	6	0	—	—	—	—	—	—	—	—
	Total	58	46.9	7.9	2.15	0.18	40	11	7	2
Zolazepam + Tiletamine Zoletil	3	14	53.7	5.9	1.10	0.25	40	15	1	0
	4	27	46.8	10.3	1.23	0.28	80	23	0	1
	5	6	50.8	5.3	0.99	0.09	101	21	0	3
	6	2	53.5	4.9	1.53	0.25	—	—	0	2
	Total	49	49.6	8.9	1.18	0.27	70	30	1	6

animals recovered within 40±11 min (range=10–60 min.) of being injected. They were usually resting and suckling their pup during that period.

The tiletamine-zolazepam mixture was injected into 49 females with a mean dose of 1.1±0.3 mg/kg (range=0.73–1.86 mg/kg of the mixture) that produced anesthetic induction of 10 min. (range=5–8 min). The level of anesthesia achieved did not relate to the dose injected ( $r_s=0.11$ ;  $df=47$ ;  $P=0.47$ ) and was not significantly different for individuals with better body condition than for animals in poor body condition ( $Z=1.12$ ;  $n_1=19$ ;  $n_2=30$ ;  $P=0.26$ ). However we had to inject a significantly higher dose in animals that were in poor body condition (linear regression,  $r=0.341$ ,  $n=49$ ,  $P<0.05$ ) to reach a similar level of anesthesia. In contrast to the response to ketamine, fewer cases of tremor occurred with tiletamine-zolazepam mixture ( $n=1$ ), but respiratory depression was the only significant adverse effect observed (Table 1). In six of the procedures, the animals developed apnea and were given artificial respiration until they were breathing regularly and independently. Moreover, two animals reached state 6 of anesthesia (Table 1) and died within 1 hr of being injected, although the doses (1.4 and 1.7 mg/kg) were within the range used in this study. All females injected with tiletamine-zolazepam recovered within 70±30 min (range=20–120 min), and none showed any sign of atypical behavior following anesthesia. Recovery times with tiletamine-zolazepam were significantly greater than that with ketamine alone ( $P<0.0001$ ).

Tiletamine-zolazepam mixture and ketamine alone appear to be effective for anesthesia of free-ranging subantarctic fur seal females for light surgery purposes. These two dissociative drugs produced a satisfactory degree of anesthesia, with a short induction time and no significant side effects although some deep apnea episodes occurred with tiletamine-zolazepam. Moreover, hallucinatory behavior that may be associated with the use of

these drugs (Woods et al., 1994) never led to any adverse long term effects such as pup abandonment; injected females were usually sleeping while suckling during the short period of recovery.

However, like previous studies on fur seals (Bester, 1988; Boyd et al., 1990), no relationship between dose and degree of immobilization was found, suggesting a highly variable degree of individual response to a given dose of either drug. In addition, animals in good condition required significantly lower dosage than those in poor condition to obtain a similar satisfactory degree of anesthesia permitting tooth extraction. This shows animal in good condition to be more sensitive to cyclohexamines. However, other physiologic characteristics could also explain some individual variations in the anesthesia level, such as foraging trip tiredness, stress, lactation, or pregnancy state.

Although not frequent, tremoring and apnea were observed in this study as well as in previous ones (Baker et al., 1990; Boyd et al., 1990; Griffiths et al., 1993). Ataractics such as zolazepam hydrochloride produce a good sedation, longer anesthesia, and significant muscle relaxation, and thus tend to reduce tremor during induction and the period of anesthesia. However the extent of muscle relaxation caused by zolazepam hydrochloride tends to increase occurrence of respiratory depression by promoting upper respiratory tract obstruction and prolonged apnea. Similar observations have been made in other species (Boyd et al., 1990; Mitchell and Burton, 1991; Griffiths et al., 1993) and we thus don't recommend the use of tiletamine-zolazepam to anesthetize subantarctic fur seals when level 4 or more is needed. Ketamine alone appeared to be the most satisfactory anesthetic agent, producing a shorter, safer, and more stable anesthesia with very few side effects. Furthermore, recovery from apnea was easier with ketamine because animals were much more sensitive to stimulation and artificial respiration. Particular attention should

however be taken during ketamine anesthesia recovery or under light ketamine immobilization, as a lack of pain control precludes any painful manipulation during that period.

Best anesthetic results were obtained at doses of 1.9–2.3 mg/kg for ketamine and 0.9–1.3 mg/kg for tiletamine-zolazepam. However we recommend reduced doses in individuals in good condition in order to reduce risk of apnea. Stressful conditions should also be avoided as much as possible; handling animals during periods of high air temperature must be precluded or undertaken with caution (Trillmich and Weisner, 1979) and it is preferable to cover the animal's eyes to prevent light stimulation. Two casualties occurred during our study. We thus strongly recommend a premedication injection of atropine sulfate at 0.005 mg/kg (Sweeney, 1974) to prevent reflex vagal bradycardia and to limit side effects such as salivation and upper respiratory tract secretions (Anderson, 1983). Furthermore, in cases of apnea, we recommend the use of respiratory stimulant doxapram (Dopram®, Tours, France; Parry et al., 1981) and tracheal intubation permitting artificial ventilation (Baker et al., 1990) by AMBU bag.

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