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IMMOBILIZATION OF CALIFORNIA SEA LIONS USING MEDETOMIDINE PLUS KETAMINE WITH AND WITHOUT ISOFLURANE AND REVERSAL WITH ATIPAMEZOLE

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ABSTRACT: The use of medetomidine and ketamine, alone and in combination with isoflurane, with atipamezole reversal was evaluated for immobilizing 51 California sea lions (*Zalophus californianus*) for a variety of medical procedures at a rehabilitation center in northern California (USA) between May 1997 and August 1998. Animals were given 140 μg/kg medetomidine with 2.5 mg/kg ketamine intramuscularly. Mean (±SD) time to maximal effect was 8 ± 5 min. At the end of the procedure, animals were given 200 μg/kg atipamezole intramuscularly. Immobilization and recovery times were, respectively, 25 ± 12 and 9 ± 7 min for 35 animals maintained with medetomidine and ketamine alone and 58 ± 30 and 9 ± 9 min for 16 animals intubated and maintained with isoflurane. No mortalities occurred as a result of the immobilizations. Disadvantages of the medetomidine and ketamine combination included a moderate variation in time to maximal effect and plane of sedation, a large injection volume and high cost. However, this combination offers safe and reversible immobilization that can be easily administered by the intramuscular route and that produces a plane of anesthesia that is sufficient to carry out most routine diagnostic procedures.

Key words: Anesthesia, atipamezole, California sea lion, immobilization, isoflurane, ketamine, medetomidine, *Zalophus californianus*.

INTRODUCTION

Safe and efficacious immobilization of pinnipeds continues to be an area of intensive investigation (Gales, 1989; Williams et al., 1990). Anatomic and physiologic adaptations for an aquatic existence such as thick blubber layers and a diving reflex that includes bradycardia and vascular shunting complicate many anesthetic methods that are routine in terrestrial species. Particularly challenging are large, free-ranging otariids that are not easily manually restrained and which do not have readily accessible blood vessels that allow intravenous administration of short-acting agents that can be safely titrated to effect. For these animals, administration of anesthetic agents continues to be best accomplished by the intramuscular (IM) route. Many currently used IM agents are associated with prolonged recovery times or may have narrow margins of safety that can create problems in field conditions. For example, mortality of over 20% was reported in free-ranging Steller's sea lions (*Eumetopias jubatus*) when a combination of tiletamine and zolazepam was used (Loughlin and Spraker, 1989).

The recent introduction of the α_2-agonist medetomidine to North America may provide a distinct advantage over previously used agents because of its reversibility by the α_2-antagonist atipamezole. However, the depth of sedation achievable with medetomidine alone may not be sufficient to adequately immobilize most animals, thus addition of a more potent anesthetic agent such as ketamine is usually required (Jalanka and Roeken, 1990). The combination of medetomidine and ketamine with reversal by atipamezole has been used successfully in a wide range of domestic and non-domestic mammals (Jalanka and Roeken, 1990; Cullen, 1996). Medetomidine alone and in combination with ketamine has been used in a small number of southern elephant seals (*Mirounga leonina*), where it did not appear to

improve sedation over previously used agents (Woods et al., 1996). However, its use has not been previously described in otariids.

In June of 1992, immobilization of five free-ranging, adult female Steller's sea lions was undertaken on Forrester Island, Alaska (USA) using a combination of medetomidine and ketamine. Incrementally increasing the medetomidine dose revealed that adequate sedation was first achieved with a minimum dosage of 140 μg/kg medetomidine and 2.5 mg/kg of ketamine (T. Spraker and D. Calkins, unpubl. data). The purpose of the current investigation was to evaluate the safety and efficacy of this dosage of medetomidine plus ketamine, alone and in combination with isoflurane, and reversal using atipamezole in California sea lions (*Zalophus californianus*).

MATERIALS AND METHODS

From May 1997 to August 1998, 24 male and 27 female California sea lions, ranging in weight from 13.5 to 145 kg (mean ± SD = 43.0 ± 25.5 kg) and in age from approximately 1 year to adult (estimated at over 5 years), were immobilized for a variety of medical procedures including wound debridement, radiographs, ultrasound, endoscopy, and major surgery at a rehabilitation center in northern California. Many of the animals were affected with a variety of potentially anesthetic-complicating disorders including pneumonia, nephritis and head trauma at the time of immobilization. Physical status prior to immobilization was scored as good (n = 13), fair (n = 29), or poor (n = 9). Animals in good physical status were bright, responsive, only mildly underweight if at all, and not obviously affected by any condition that would compromise anesthesia. Animals in fair physical condition may have been lethargic, moderately underweight, had an estimated dehydration of up to 5%, or had some suggestion of clinical disease such as mild to moderate dyspnea. Animals in poor condition showed clinical signs such as marked lethargy, weakness, anorexia, estimated dehydration approaching 10%, labored respiration, central nervous system deficits or renal failure. All animals were weighed within 3 days of immobilization as part of routine husbandry procedures.

Atropine sulfate (Radix Laboratories Inc., Eau Claire, Wisconsin, USA) at a dosage of 0.02 mg/kg was given IM to each animal at least 10 min prior to administration of the immobilizing agents. Each animal was given 140 μg/kg medetomidine (Domitor®, Pfizer Animal Health, Exton, Pennsylvania, USA) and 2.5 mg/kg of ketamine (Ketaset®, Fort Dodge Animal Health, Fort Dodge, Iowa, USA) intramuscularly by either hand injection (n = 47) using 3,6 or 12 ml syringes and 1.2 × 40 mm needles (Monoject®, Sherwood Medical, St. Louis, Missouri, USA) with manual restraint or blow dart (n = 4) using 3 ml darts and 1.2 × 38 mm needles (Telinject USA Inc., Saugas, California, USA). The darted animals ranged in weight from 25 to 72 kg and included two sea lions in good and two animals in poor preanesthetic condition. These animals were darted as a result of fewer available people to assist in restraining for hand injection or to minimize stress due to handling. Sites for all IM injections included muscle immediately surrounding the pelvis, femur and tibia and muscle overlying the scapula. The drugs were administered together in the same syringe. The concentration of the medetomidine was either the commercially-available 1 mg/ml solution or, if the injection volume was too high for larger animals or when using the blow dart, a lyophilized and sterile saline-reconstituted solution at 10 mg/ml was used.

Time to maximal effect as defined as the time from injection of medetomidine and ketamine to the point after which no noticeable increases in depth of anesthesia took place was recorded. Maximum plane of anesthesia was scored from Level I to III. Animals at Level I would react to noise, would not lie in place for radiographs, or required physical restraint to accomplish even minor procedures such as venipuncture or masking for gas anesthesia. Additional ketamine (1/2 of the original dose) was administered to 4 animals that reached only Level I. Animals reaching Level II would display a swallowing reflex and some jaw tone, reaction to deep pain caused by a flipper pinch or lancing of a dermal abscess, but would easily maintain sternal and lateral recumbency for radiographs or ultrasound without additional restraint. Sea lions at Level III could be intubated and showed no swallowing reflex or response to deep pain.

Heart rate and respiratory rate were monitored by stethoscopic chest auscultation and observed thoracic movements respectively. Additionally, pulse oximeter saturation (SpO_2) was monitored using a Nellcor® N-20 pulse oximeter with a Nellcor® DURA-Y multisite oxygen transducer placed into a Nellcor® VSC-S small veterinary sensor clip (Nellcor Incorporated,

Pleasanton, California, USA) with the clip probe attached to the distal one-third of the tongue.

Of the 51 animals, 35 sea lions (14 males and 21 females) were given only medetomidine and ketamine for immobilization. This subset of animals ranged in weight from 13.5 to 145 kg (mean ± SD = 48.0 ± 27.0 kg). Preanesthetic condition was scored as good in nine, fair in 21, and poor in five of these sea lions. Heart rate and respiratory rate were recorded from 19 animals approximately every 5 min during immobilization while SpO_2 was recorded approximately every 7 min for four animals. Immobilization time was defined as the time from first observed maximal effect to the time of atipamezole (Antisedan®, Pfizer Animal Health) injection at the end of the procedure.

The remaining 16 sea lions (10 males and six females) were intubated and maintained with isoflurane (AErrane®, Fort Dodge Animal Health) for procedures that were expected to be longer than 30 min or of a more invasive nature such as surgery. This group of sea lions ranged in weight from 13.5 to 63.5 kg (mean ± SD = 31.0 ± 17.0). Preanesthetic condition was good in four, fair in eight, and poor in four of the animals. If these animals could not be intubated after being given medetomidine and ketamine they were given 3 to 5% isoflurane with oxygen flow at 2 to 5 L/min through a mask (Jorgensen Laboratories Inc., Loveland, Colorado, USA) placed over the animal's muzzle until intubation could be accomplished. The sea lions were intubated by opening the mouth with 2 cm wide, 40 cm long nylon straps and visualizing the larynx with a 150 mm McIntosh laryngoscope blade (Rusch Inc., Duluth, Georgia, USA). The cuffed endotracheal tubes (Rusch Inc.) ranged from 7 to 10 mm. All 16 individuals were maintained with 1 to 3% isoflurane and a reduced oxygen flow of 2 to 3 L/min delivered through a precision Fluotec II vaporizer in a standard, semi-closed, small animal rebreathing system (VMS®, MDS Matrx, Orchard Park, New York, USA). The animals were maintained at anesthetic Level III for the remainder of the procedure. Heart rate and respiratory rate were recorded from nine animals approximately every 5 min. SpO_2 was recorded in three sea lions approximately every 12 min. If SpO_2 fell below 85%, the sea lions were manually ventilated by squeezing the rebreathing bag. Immobilization time was recorded as the time from first observed maximal effect after medetomidine and ketamine injection to injection of atipamezole. Atipamezole injection was given immediately after turning off isoflurane gas at the end of the procedure. Animals were allowed to recover on room air while still intubated and extubated once a swallowing reflex was re-established and the animals attempted to cough the endotracheal tube out of the trachea.

At the end of the procedure, 31 animals that had been given medetomidine and ketamine only and 13 that had also received isoflurane gas were given 200 μg/kg atipamezole IM in the muscle overlying the pelvis, femur, and tibia. Recovery time was recorded as the time elapsed from injection of atipamezole to the ability to stand on front flippers and begin locomotion.

Statistical analysis was performed using Instat version 2.01 (GraphPad Software, San Diego, California, USA). Differences between time and physiologic parameters were calculated using a two-tailed Student *t*-test and was considered significant at $P < 0.05$. If calculated means had significantly different standard deviations ($P < 0.05$), then a non-parametric Mann-Whitney test was used.

RESULTS

Injection of medetomidine and ketamine resulted in moderate variation in time to maximal effect with a significantly ($P < 0.01$) longer time in those animals that were blow darted in comparison to those that were hand injected (Table 1). There was also some variation in the plane of anesthesia that was reached (Table 2). Of the animals that were given a preanesthetic score of good, two (15%) reached only anesthesia Level I, nine (70%) reached Level II, and two (15%) reached Level III. Of the sea lions in fair preanesthetic condition, seven (24%) reached only Level I, 10 (35%) reached up to Level II, and 12 (41%) reached Level III. For the animals that were in poor condition, two (22%) reached only Level I, five (56%) reached up to Level II, and two (22%) reached Level III. There did not appear to be any direct relationship between preanesthetic score and plane of anesthesia. Overall, 40 (78%) of all the animals reached at least Level II, a plane of sedation that was sufficient to carry out most non-invasive diagnostic procedures.

Length of immobilization and recovery times are recorded in Table 1 for both animals maintained with medetomidine and

TABLE 1. Ranges and means ± SD of recorded time parameters describing the immobilization and reversal characteristics of medetomidine, ketamine, and atipamezole, used alone or with isoflurane, in California sea lions (*Zalophus californianus*).

Time parameter	Number of animals	Range (min)	Mean ± SD (min)
Time to maximal effect[a] (hand injection)	47	2–23	8 ± 5
Time to maximal effect[a] (blow dart)	4	10–23	17 ± 6
Immobilization time[b] (medetomidine/ketamine only)	35	9–60	25 ± 12
Recovery time[c] (medetomidine/ketamine only)	31	0–25	9 ± 7
Immobilization time[b] (maintained with isoflurane)	16	19–115	58 ± 30
Recovery time[c] (maintained with isoflurane)	13	2–33	9 ± 9

[a] Time from injection of medetomidine and ketamine to maximum observable effect.
[b] Time from maximum observable effect to injection of atipamezole.
[c] Time from injection of atipamezole to ability to stand on front flippers and begin locomotion.

ketamine alone and for those maintained with isoflurane. There was no significant difference in recovery time after injection of atipamezole between the two groups of sea lions. Prolonged recovery, over 20 min, was noted in four animals given medetomidine and ketamine only (Table 2) and one animal maintained with isoflurane whose preanesthetic condition was fair. Of the four animals given medetomidine and ketamine only, preanesthetic condition was fair in three and poor in one. Length of immobilization in these sea lions was 18 ± 7 min, and was not significantly different than the overall immobilization time for animals given medetomidine and ketamine only.

Six animals, three after being given medetomidine and ketamine only and three maintained with isoflurane, were euthanized prior to recovery based on findings during the procedure. Reasons for euthanasia included: untreatable blindness in two sea lions, multiple fractures with severe osteomyelitis and poor prognosis in two animals, end-stage septicemia in one animal, and metastatic carcinoma in one animal.

TABLE 2. Characteristics and results of immobilization of 51 California sea lions (*Zalophus californianus*) using medetomidine and ketamine and reversal with atipamezole.

Characteristic or result	Number of animals
Animals reaching only anesthetic Level I[a]	11 of 51 (22%)
Physical restraint used to complete procedure	7 of 11 (64%)
Additional ketamine (½ original dose) given	4 of 11 (36%)
Animals reaching up to anesthetic Level II[b]	24 of 51 (47%)
Animals reaching anesthetic Level III[c]	16 of 51 (31%)
Total intubated and maintained with isoflurane	16
Intubated after medetomidine and ketamine only	6 of 16 (37%)
Intubated after masking with isoflurane	10 of 16 (63%)
Prolonged recovery time (>20 min)	4 of 35 (11%)
Bradycardia (minimum heart rate <50 beats/min)	3 of 35 (9%)
Euthanized based on findings during procedure	3 of 35 (9%)
Mortalities due to anesthetic complications	0 of 35 (0%)

[a] Animals at Level I would react to noise, would not lie in place for radiographs, or required physical restraint to accomplish even minor procedures such as venipuncture or masking for gas anesthesia.
[b] Animals reaching Level II would display a swallowing reflex and some jaw tone, reaction to deep pain caused by a flipper pinch or lancing of a dermal abscess, but would easily maintain sternal and lateral recumbency for radiographs or ultrasound without additional restraint.
[c] Sea lions at Level III could be intubated and showed no response to deep pain.

TABLE 3. Ranges and means ± SD of recorded values of physiologic parameters in California sea lions (*Zalophus californianus*) after injection with medetomidine and ketamine alone and in combination with isoflurane.

Physiologic parameter	Number of animals	Number of recordings	Range	Mean ± SD
Heart rate (beats/min)[a]	19	82	40–120	70 ± 19
Respiratory rate (breaths/min)[a]	19	82	4–24	10 ± 4
Pulse oximeter saturation (%)[a]	4	14	66–86	75 ± 5
Heart rate (beats/min)[b]	9	83	37–115	74 ± 14
Respiratory rate (breaths/min)[b]	9	83	2–36	17 ± 8
Pulse oximeter saturation (%)[b]	3	14	74–100	90 ± 7

[a] Medetomidine and ketamine only.
[b] Medetomidine and ketamine in combination with isoflurane.

Ranges and means of recorded physiologic parameters are found in Table 3. SpO_2 was significantly lower in the animals maintained with medetomidine and ketamine only. Preanesthetic score was fair in three and poor in one of these sea lions. For those animals maintained with isoflurane, preanesthetic condition was good in one, fair in one, and poor in one. Preanesthetic physical condition in three animals given medetomidine and ketamine only where a bradycardia (less than 50 beats/min) was noted was good in 2 sea lions and fair in one. One sea lion in fair preanesthetic condition had a bradycardic episode when maintained with isoflurane.

Characteristics of the immobilization of 51 California sea lions using medetomidine and ketamine are summarized in Table 2. No animals died as a result of immobilization with medetomidine and ketamine, alone or with isoflurane.

DISCUSSION

The use of the medetomidine and ketamine with atipamezole reversal combination offers several advantages over previously used agents in otariids. Based on mortality rates alone, the agents in the current study appear safer than many of those previously reported (Bester, 1988; Gales, 1989; Loughlin and Spraker, 1989; Boyd et al., 1990; Work et al., 1993; Sepulveda, 1994; Heath et al., 1996). The reversibility of medetomidine and resultant rapid recovery avoids the prolonged recovery time reported with some other injectable agents such as Telazol® (Fort Dodge Animal Health), a 1:1 combination of tiletamine and zolazepam (Loughlin and Spraker, 1989). The use of isoflurane in otariids appears to result in both low mortality rates and rapid recovery (Heard and Beusse, 1993; Heath et al., 1997; Gales and Mattlin, 1998). However, the use of gas anesthesia alone may not be practical for larger animals that are not easily restrained for masking. Additional restraint equipment, personnel, increased handling time and increased animal struggling may not be practical, especially in field situations.

The variations in time to maximal effect and plane of anesthesia may be due to placement of the injection as evidenced by the increased time to peak effect in blow darted animals where appropriate anatomic sites and adequate depth of injection to muscle layers could not be easily controlled. It is possible that some animals received at least a portion of their dose into blubber which may have interfered with drug pharmacokinetics.

It is also possible that physiologic state of the animals prior to immobilization may have influenced speed and depth of sedation. Unfortunately, this could not be controlled for in a rehabilitation center where animals undergoing immobilization varied from normal health to severely compromised by disease (Gulland et al., 1996) or trauma (Goldstein et al., 1999). However, there was no obvious trend in plane of an-

esthesia reached with respect to preanesthetic condition. Prolonged recovery noted in some animals also did not appear to be a consequence of physical condition, or of immobilization time.

Significant difference in SpO_2 between animals given medetomidine and ketamine only and those maintained with gas supports previous recommendations of caution regarding the use of α_2-agonists in diving or carbon dioxide tolerant species (Sedgwick, 1999). However, small sample size and variation in recording intervals preclude any definitive conclusions. It is recommended that individuals be prepared to intubate and assist pulmonary ventilation if considering the use of medetomidine and ketamine in otariids. Widely ranging SpO_2 values may also have been a consequence of disease status in addition to possible effects of the agents used. Diseases such as verminous pneumonia as caused by *Parafilaroides decorus* are relatively common in young California sea lions (Gage et al., 1993) and may have significant effects on pulmonary perfusion and gas exchange. There were no obvious trends in the relationship between preanesthetic condition and SpO_2.

Medetomidine, a highly selective and potent α_2-agonist, produces sedation and analgesia by stimulating central receptors. Reported side-effects may include significant cardiovascular changes such as bradycardia and vasoconstriction and decreased respiratory rate (Cullen, 1996). In the current study, bradycardic episodes were recorded in 4 animals. Since bradycardia is a characteristic of the "diving response" in marine mammals (Kooyman et al., 1981), it is unknown whether the bradycardia was due to medetomidine or was a result of anesthesia inducing a diving reflex in the sea lions. Though some reports contraindicate the use of atropine with medetomidine (Cullen, 1996), atropine was administered in these animals as the risk due to stimulating the diving reflex was thought to outweigh the risks associated with medetomidine interactions. Specific effects of medetomidine on the diving reflex and the resultant physiological consequences in diving mammals is an area requiring further study.

The dosage of medetomidine used in the current study (140 μg/kg) is relatively high as compared to that used in most terrestrial mammals. The high end of dosages used in domestic dogs is usually about 80 μg/kg (Cullen, 1996). This high dose presented some difficulties especially in the larger animals in the current study. For example, for a 75 kg animal, the injection volume of medetomidine and ketamine was approximately 12.4 ml using the commercially-available solutions. This volume is poorly manageable in blow darts and would probably have resulted in the use of multiple darts, an undesirable task in a field situation. For larger animals, and especially for those that were blow darted, we lyophilized and reconstituted the medetomidine to a concentration of 10 mg/ml. Using the more concentrated solution results in an injection volume of approximately 2.9 ml for a 75 kg sea lion. The use of medetomidine also is relatively expensive in comparison to other injectable agents, and the large dose required in sea lions increases the cost even more. The immobilization of a 75 kg sea lion with medetomidine and ketamine and reversed with atipamezole costs approximately $94.50 at current market prices, whereas the use of Telazol® at a dosage of 2 mg/kg costs approximately US $5.70.

Disadvantages of medetomidine and ketamine with atipamezole reversal in sea lions thus include a moderate variation in induction time and plane of anesthesia, large injection volume and relatively high cost. However, advantages of this combination are safe and reversible immobilization that can be easily administered by the intramuscular route and production of a plane of anesthesia that is sufficient to carry out most routine diagnostic procedures. Furthermore, medetomidine and ketamine can be safely used in procedures of longer duration or of a more invasive

nature when used in conjunction with isoflurane.

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