Anesthesia with Isoflurane and Sevoflurane in the Crested Serpent Eagle (Spilornis cheela hoya): Minimum Anesthetic Concentration, Physiological Effects, Hematocrit, Plasma Chemistry and Behavioral Effects

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ABSTRACT. The initial goal of this study was to determine the minimum anesthetic concentration (MAC) for isoflurane (ISO) and sevoflurane (SEVO) for the crested serpent eagle. Next, we compared the anesthetic effects of each on the physiological effects, hematocrit, plasma chemistry values and behavior in spontaneously breathing captive adult crested serpent eagles. Sixteen eagles were randomly allocated to two groups for anesthesia with ISO (n=8) or SEVO (n=8). First, we measured the MAC values of ISO and SEVO, and four weeks later, we investigated the effect of each on the physiological effects, hematocrit (HCT) and plasma chemistry values. The MAC values of ISO and SEVO for crested serpent eagles were 1.46 ± 0.30 and 2.03 ± 0.32%, respectively. The results revealed no significant differences between the two anesthetics in induction time, while time of extubation to recovery was significantly shorter with SEVO. A time-related increase in end-tidal CO₂ and decreases in body temperature and respiratory rates were observed during anesthesia with each anesthetic. There were no significant differences between the effect of the two anesthetics on heart rate, hematocrit, plasma chemistry values or respiration, although each caused minor respiration depression. We concluded that SEVO is a more effective inhalant agent than ISO for use in eagles, showing the most rapidest induction and recovery from anesthesia.

KEYWORDS: crested serpent eagle, isoflurane, minimum anesthetic concentration, sevoflurane.

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The crested serpent eagle (Spilornis cheela hoya) is a protected species with a high morbidity rates and is often rescued by surgical procedures performed by veterinarians in Taiwan [3]. In most anesthetic modes, inhalant anesthesia is frequently used in avian species for physical examination, diagnostic investigation, surgical procedures and research and management purposes [10]. Traditionally, isoflurane (ISO) has been a common inhalant anesthetic of choice for general avian anesthesia [15, 37]. However, sevoflurane (SEVO), a new inhalant agent, may have several potential advantages over ISO. Moreover, previous investigations reported SEVO as a suitable agent for induction and quick recovery in mammals and birds [25, 26, 39].

In mammals, anesthetics are typically evaluated according to their minimum alveolar concentration [40], a measure of the potency of an anesthetic inhalant. At this concentration (vol %), also referred to as the effective dose 50 (ED 50) [7], 50% of the anesthetized animal population will not respond when exposed to a noxious stimulus. Since birds do not have an alveolar lung, anesthetic potency has been determined using the minimum anesthetic concentration (MAC), a method similar to that used for mammals [7, 28–31]. The MAC values for ISO and SEVO have been determined in a number of bird and mammal species. Although some data exist for species of eagles [29–31], specifically no information exists on the clinical use of ISO and SEVO for adult crested serpent eagles. Therefore, in this study, we aimed to first determine the MAC value of ISO and SEVO in captive adult crested serpent eagles and then to compare the physiological effects, hematocrit, plasma chemistry values and behavioral effects of ISO and SEVO at 1 MAC depth of anesthesia in spontaneously ventilating captive adult crested serpent eagles. Further, the study was performed to conclude which anesthetic can provide more satisfactory anesthesia with rapid induction and recovery for minor surgical procedures for the crested serpent eagle.

MATERIALS AND METHODS

Animals: We used sixteen captive healthy adult crested serpent eagles (Spilornis cheela hoya) (ten males, six females) ranging in body weight from 1,200 to 1,800 g. Eagles were obtained from the Wildlife First Aid Station of the Endemic Species Research Institute (ESRI) in Taiwan (120°48′5.38″E, 23°49′43.22″N). The station is at an elevation of 230 m and simulates natural breeding conditions. All birds were used in analyses due to previous injuries. Crested
serpent eagles were housed under natural temperature and light conditions, in open-air 6 x 3 x 3-m aviaries, equipped with wooden perches and enclosed by wire netting on the sides and top. Eagles were fed dead laboratory mice or one-day-old chicks every day and provided fresh water ad libitum. These diets were chosen to be representative of the birds’ natural diet [19]. Regular examinations by a veterinarian during the study period found no evidence of malnutrition or dehydration, and we did not observe malnutrition or dehydration of eagles at the clinic. The procedure used in this study was approved by the Animal Care and Use Committee of ESRI. None of the eagles developed any observable complications throughout the duration of the study.

Anesthetic procedures: Sixteen eagles were randomly allocated to two groups for anesthesia with ISO (n=8) or SEVO (n=8). First, we measured the MAC values for ISO and SEVO, and four weeks later, we investigated the effect of each on the physiologic effects, hematocrit (HCT) and plasma chemistry values. Eagles were not fasted or pre-medicated prior to or following anesthesia. The eagles were anesthetized with ISO (Forane, Abbott Laboratories, Queenborough, Kent, U.K.) or SEVO (Ultane, Abbott Laboratories), and they were intubated with a 12-F Cole endotracheal tube (Ruschelit, Willy Rusch AG, Kernen, Germany). We used an ISO vaporizer (Forawick vaporizer, Muraco Medical Co., Ltd., Tokyo, Japan), SEVO vaporizer (Vip3000; Matrix, Orchard Park, NY, U.S.A.) and ADS 1000 veterinary anesthesia delivery system (Engler, Hialeah, FL, U.S.A.), which functions as a non-rebreathing circuit and does not include a canister for chemical absorbent to eliminate carbon dioxide. It is not intended for connection to another breathing system [18].

MAC determination: Raptors were physically restrained with the aid of a falconer’s hood and a towel. Anesthesia was induced with ISO (5%) and SEVO (6%) in 100% oxygen, using an over-the-head mask attached to an ADS 1000 veterinary anesthesia delivery system. The oxygen flow rate was set at 1 l/min for induction of anesthesia. We did not perform intermittent positive pressure ventilation. When the individual lacked jaw tone, we performed intubation with a 12-F Cole endotracheal tube connected to an ADS 1000 veterinary anesthesia delivery system. Heart rate (HR), respiratory rate (RR), body temperature (BT) and end-tidal CO2 (ETCO2) and respiratory rate were monitored with a calibrated anesthetic gas monitor (Patient monitor, BP-608 Evolution; Colin Medical Technology, Tokyo, Japan), which aspirated 200 ml/min of airway gas from a port on the side of the endotracheal tube adapter. Heart rate (HR), respiratory rate (RR), body temperature (BT) and end-tidal CO2 (ETCO2) were noninvasively and continuously monitored (BP-608 Evolution) at 1 min and 10 min and every 10 min thereafter during the anesthesia event.

Intubation to induction, extubation to recovery time and smoothness of recovery were recorded for each anesthetic procedure. Time to induction was defined as the time from initial delivery of gaseous anesthesia until a medium level of anesthesia was achieved as determined by good muscle relaxation and absence of voluntary blink [1]. Time to extubation was defined as the time from ceasing anesthetic gas administration until the presence of a cough, swallow reflex or head shaking.

Hematocrit and plasma chemistry measurements: Blood samples (1.0–1.5 ml) were collected by venipuncture from either the brachial or tarsal vein into sterile syringes with 23-gauge needles. Blood samples were collected before the bird was anesthetized (baseline), at 30 min intervals throughout the anesthetic period and at one hr after discontinuing anesthesia. The blood samples were placed into a lithium heparin blood tube (Miotrainer, Becton, Dickinson and Co., Franklin Lakes, NJ, U.S.A.), and they were placed on ice following 2–3 min at room temperature [4]. In this study, samples (1.0–1.5 ml) were collected four times, and 4–6 ml blood was sampled from each eagle. The HCT was determined by centrifugation of whole blood at 10,000 g for 5 min. Plasma chemistry values were determined using an automated clinical chemistry analyzer (SPOTCHEM EZ SP-4430, Arkray, Kyoto, Japan) within 30 min and included the following seventeen parameters: uric acid (UA), plasma urea nitrogen (BUN), total protein (TP), albumin (ALB), glucose (GLU), cholesterol (CHO), triglyceride (TG), aspartate aminotransferase (AST), alanine aminotransferase...
(ALT), lactate dehydrogenase (LDH), alkaline phosphatase (ALP), total bilirubin (TBIL), creatine (CRE), creatine phosphokinase (CPK), amylase (AMY), calcium (Ca) and ionic phosphorous (IP); the concentrations of Na\(^+\), K\(^+\) and Cl\(^-\) ions were assayed with an electrolyte analyzer (SPOTCHEM EL SE-1520, Arkray).

Statistical analyses: Results are shown as mean ± SD values and were analyzed by one-way analysis of variance for repeated measures to compare time-related variables within the two anesthetic groups. Tukey’s multiple comparison test was used to identify differences between the means. The mathematical model included fixed effects due to anesthetic groups and residual error.

We used a Kolmogorov-Smirnov test to test normality. When the data had a normal distribution, a Student’s t-test was used to determine the significance. Cardiopulmonary, HCT and biochemical data were not normally distributed, and those data were analyzed using the nonparametric Mann-Whitney U test. Differences were considered to be significant at *P*<0.05.

RESULTS

MAC: The MAC value of ISO and SEVO for crested serpent eagles were 1.46 ± 0.30 and 2.03 ± 0.32%, respectively (Table 1).

Induction and recovery from anesthesia: The intubation to induction times for ISO and SEVO were 145.9 ± 16.9 and 128.4 ± 18.8 sec, respectively. Eagles did not struggle during induction with either anesthetic inhalant nor did we observe any adverse reactions during intubation. Furthermore, no significant differences were detected between ISO and SEVO for time to induction. However, the mean ± SD values for time required from extubation to recovery for ISO and SEVO, 323.3 ± 197.5 and 174.0 ± 64.9 sec, respectively, were significantly different (*P*<0.05) (Fig. 1).

Physiological effects findings: There were no significant differences in HR, RR, BT or ETCO\(_2\), between the two anesthetic groups, although these variables differed significantly from initial values (Fig. 2).

Figure 2 also shows how HR in the ISO group decreased significantly between 50 and 60 min of anesthesia and how RR in the ISO group decreased significantly throughout the evaluation period. In both groups, we observed a decrease in BT while ETCO\(_2\) increased significantly throughout the period of maintained anesthesia. Esophageal temperature was significantly (*P*<0.01) higher during SEVO anesthesia. Following anesthesia by either anesthetic, mean body temperature decreased slightly.

HCT and plasma value biochemical findings: The plasma HCT, ALB, TP, UA, CHO, ALT, ALP, TG and CRE values (Figs. 3 and 4) of both anesthetic groups decreased significantly from the baseline values during maintenance of anesthesia with no significant differences between the two. The plasma Na\(^+\), AST, Cl\(^-\), LDH, BUN, IP, AMY, Ca, TBIL and CPK values (Figs. 5 and 6) were not different from the baseline values at any time after anesthesia, with the exception of the GLU and K\(^+\) values (Fig. 6) in the ISO group, which increased significantly at one hour post anesthesia. There was no significant difference in plasma values at any time between the two groups.

DISCUSSION

To date, there have been no reported studies describing reference agents of inhalant anesthesia, especially for crested serpent eagles. On the other hand, the MAC is a well-recognized index of the potency of inhalation anesthetics in mammals [40]. However, since birds do not have an alveolar lung, the potency of inhalation anesthetics in birds has been determined by using methods similar to those used in mammals, called the MAC [30], or the ED50 [7].

We measured the MAC for ISO in crested serpent eagles (1.46 ± 0.30%) and found it to be similar to the minimal alveolar concentration MAC values reported in mammals, such as monkeys (1.28 ± 0.18%), horses (1.31 ± 0.22%), rats (1.38 ± 0.06%), dogs (1.41 ± 0.16%) [11] and goats (1.3–1.5%) [21] as well as other avian species, such as cockatoos (1.44 ± 0.07%) [7], sandhill cranes (1.34 ± 0.14%) [30], ducks (1.30 ± 0.23%) [31] and thick-billed parrots (1.07%) [32].

![Fig. 1. Induction and recovery times for ISO and SEVO anesthesia in crested serpent eagles. Each column represents the mean; a vertical bar indicates the SD; an asterisk (*) indicates a significant difference (*P*<0.05); n=8 for the two groups.](image-url)
We found similar results for MAC with SEVO in the crested serpent eagle (2.03 ± 0.32%) as found for the minimal, alveolar concentration MAC values reported in mammals, such as in children (2.49 ± 0.08%) [27], adult humans (1.7%), dogs (2.3%), cats (2.5%), pigs (1.9%), rats (2.5%) [41] and goats (2.3%) [23] as well as in other avian species, such as chickens [33]. These results suggest that different classes or even species of animals do not show a large variation in the effective concentration for inhalational anesthetics [40].

Both ISO and SEVO resulted in a smooth, rapid induction to and a relatively smooth recovery from anesthesia in captive adult crested serpent eagles, consistent with previous reports in bald eagles, chickens and psittacines [25, 34, 39]. Rapider induction and recovery is desirable following extensive anesthetic periods or in debilitated eagles [35]. The blood/gas partition coefficient of SEVO in human blood is much lower than that for halothane and ISO, but similar to that for nitrous oxide [42], indicating that induction and recovery from anesthesia with SEVO would be more rapid than with ISO. Although the time from induction to intubation was shorter with SEVO than ISO, the difference was insignificant. On the other hand, the time from recovery to extubation was significantly shorter with SEVO. These findings are in line with those obtained previously in bald eagles [26]. In this study, the lengthy duration of anesthesia did not cause morbidity or mortality. Apnea has been documented in other psittacines, cranes [30] and waterfowl [9]. In some birds, especially waterfowl, episodes of apnea and bradycardia can occur during induction of anesthesia due to a physiology response termed a dive response [9]. It is thought to be a stress response mediated by stimulation of the trigeminal receptor in the beak and nares [9]. A dive response usually occurs during the initial phase of induction gas anesthesia with a mask. We did not observe any apneas or dive response in our eagles at the 3 MAC ISO (4.3%) and SEVO (6.1%) concentrations with induction of anesthesia.

The physiological effects of inhalant agents have been studied in selected avian species [28–30]. Halothane and ISO appear to decrease ventilation through pulmonary, central and peripheral respiratory chemoreceptors. Theses receptors are acutely sensitive to carbon dioxide and insensitive to hypoxia [14]. In birds, inhalant anesthetics, such as halothane or ISO, cause marked respiratory depression and induce hypercapnia. Hypercapnia was observed in sandhill cranes anesthetized with ISO and chickens anesthetized with SEVO [30, 34]. ETCO2 could be used to predict the acute effects of altered ventilation on arterial blood pH [10]. In this study, we found that both anesthetics induced hypercapnia...
and hypoventilation during the one hour anesthesia period (at 1 MAC). The degree of respiratory depression during SEVO anesthesia did not significantly differ from that of ISO anesthesia. Our findings for crested serpent eagles were similar to previous reports in bald eagles [26], goats [23], cats [20] and sheep [21].

In a previous study, HR in bald eagles significantly decreased during anesthesia with ISO and SEVO [26], similar to our findings with crested serpent eagles. We suggest that this relative decrease may, at least partially, be a decline resulting from an initial increase caused by stress induced during manual restraining at induction. In contrast, the HRs of spontaneously ventilating chickens [33, 34] and sandhill cranes [30] anesthetized with SEVO were higher than those undergoing controlled ventilation with 2 MAC values of SEVO and ISO. We found that hypercapnia developed during maintenance of anesthesia with SEVO and ISO. Interestingly, hypercapnia may result in increased HR in mammals, although the mechanisms remain unknown [34]. There was no significant difference in HR between the two anesthetics during this study at the same MAC (1 MAC).

Normal avian body temperature ranges from 40 to 44.4°C [6, 16]. During avian anesthesia, supplement heat is recommended to counter the decrease in body temperature over time. Here, we used a circulating water warming blanket that delivered a constant temperature of 36°C during anesthesia. Despite the supplemental heat source, the body temperature of the crested serpent eagles decreased significantly during anesthesia. In this study, the mean body temperature in eagles during SEVO and ISO anesthesia was similar to previously reported body temperature ranges in bald eagles, cockatoos and crested caracaras (Caracara plancus) under ISO or SEVO anesthesia [7, 12, 26]. We found that eagles under SEVO anesthesia had a significantly higher BT compared with under ISO contrary to the results from Joyner and colleagues, who found that bald eagles under SEVO anesthesia produced a significantly lower body temperature compared with ISO treatment [26]. Previous studies indicated that hypothermia has been associated with bradycardia in birds [1]; however, we did not observe this in the crested serpent eagle during anesthesia with either SEVO or ISO.

The total blood volume in clinically normal birds is in the range of 6 to 11 ml per 100 g of body weight [43]. Using 10% as an estimate of total blood volume, a blood sample representing 1% or less of a bird’s body weight can usually be withdrawn from healthy birds without any detrimental effects [24]. In this study, an eagle weighing 1,784 g (mean for males and females) would have approximately 107 to 196 ml of blood, of which, in a clinically normal individual, up to 10% (10.7–19.6 ml) could be safely withdrawn without having any detrimental effect on the patient. In this study, blood samples were collected at baseline, 30 and 60 min and 1 hr after discontinuing anesthesia. Samples (1.0–1.5 ml) were collected four times, and no more 6 ml blood was sampled.
Decreases in HCT, ALB, BUN, UA, CRE, GTP, ALP, TG, TP and CHO values in anesthetized eagles may be consistent with relative hemodilution [8, 20, 22], as may occur in response to changes in regional blood flow associated with inhalant anesthesia. A decrease in hydrostatic pressure caused by anesthesia [40], primarily as a result of decreased vascular resistance [5], is likely responsible for the resulting vascular fluid pooling and functional sequestering of cellular elements. This decrease in hydrostatic pressure has been described in the ferret, dog and monkey as soon as 15 min after anesthetic intubation [41]. These effects were reversible, and our findings for the crested serpent eagle were consistent with previous reports on ferrets, in which the preanesthetic values returned within 45 min following anesthetic recovery [17].

This study revealed that hemodilution occurred during both SEVO and ISO anesthesia in crested serpent eagles.

Fig. 4. Effects of ISO and SEVO anesthesia on plasma chemistry UA, CHO, ALT, ALP, TG and CRE values in spontaneously breathing crested serpent eagles. Each point represents the mean; a vertical bar indicates the SD; an asterisk (*) indicates a significant difference from the first value (P<0.05); two asterisks (**) indicate a significant difference from the first value (P<0.01); n=8 for the two groups.
These findings in eagles coincide with previous results showing that a surgical depth of anesthesia with SEVO or ISO in American kestrels, cats, goats and sheep [8, 20–23] may be caused by a decrease in arterial pressure due to vasodilation and decreased cardiac output during anesthesia [20–23].

Both anesthetics in this study caused an increase in GLU during the one hour post-anesthesia period. However, the degree of hyperglycemia with SEVO was not significantly different from that with ISO. In this study, we observed increased GLU levels within one hour post anesthesia with ISO, similar to results from previous studies in the goat [22], ostrich [2], rat [13] and rabbit [44]. Tanaka et al. (2011) attributed the significant increase in GLU levels post anesthesia to the marked inhibition of glucose-induced insulin secretion by ISO [44]. On the other hand, Dressen et al. (1999) did not observe this effect on GLU levels in American kestrels following ISO anesthesia [8]. Although hyperglycemia was found with SEVO anesthesia in humans and goats [22, 36], we did not make this observation with crested serpent eagles. Anesthesia and surgery increase the plasma levels of stress hormones in humans [36]. The cause of hyperglycemia in this study is unknown. Various factors, such as decreased insulin levels, the release of stress hormones and catecholamine associated with intubation, hypercapnia and stress due
Both anesthetics in this study caused an increase in K⁺ during the one hour post anesthesia period. These findings agree with previous studies of anesthesia in the ostrich [2] and American kestrel [8] with ISO. The K⁺ levels increased significantly during the post anesthesia period, and this has been suggested to be caused by acidosis [2] and blood loss [38] that developed during the anesthesia episode. In this study, hyperkalemia in eagles in the one hour post anesthesia period may be related to acidosis resulting from blood loss during sampling and the anesthesia period.

Here, we observed no significant changes in plasma AST, ALT, ALP, LDH, CPK, T-BIL, CHO, BUN or CRE values at one hour post anesthesia in comparison with baseline values for each anesthetic. We found that the extent of renal and hepatic injury with SEVO in crested serpent eagles did not differ from that of ISO, consistent with previous reports [8, 21, 22]. In addition, no marked change in plasma Na⁺

Fig. 6. Effects of ISO and SEVO anesthesia on plasma chemistry AMY, GLU, Ca, K⁺, TBIL and CPK values in spontaneously breathing crested serpent eagles. Each point represents the mean; a vertical bar indicates the SD; an asterisk (*) indicates a significant difference from the first value (P<0.05); two asterisks (**) indicates a significant difference from the first value (P<0.01); n=8 for the two groups.
or Cl⁻ ion concentrations were observed in either anesthetic group, indicating that the electrolyte balances during and after anesthesia were similar using SEVO or ISO in crested serpent eagles.

In this study, we successfully achieved our initial aim for this study by determining the MAC values for ISO and SEVO to be 1.46 ± 0.30 and 2.03 ± 0.32%, respectively. Both inhalant anesthetics provided a smooth, rapid induction and recovery from anesthesia in eagles, although the induction time and recovery time were shorter with SEVO than with ISO. Cardiopulmonary effects of SEVO are similar to those of ISO. HCT and plasma chemistry values after SEVO administration did not differ from those of ISO. Although SEVO and ISO anesthesia resulted in minor effects on the respiratory system during spontaneous breathing, both anesthetics are considered suitable agents for anesthesia of crested serpent eagles.

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