Minimum anesthetic concentration of isoflurane in captive thick-billed parrots (Rhynchopsitta pachyrhyncha)

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Objective—To determine the minimum anesthetic concentration (MAC) of isoflurane in thick-billed parrots (Rhynchopsitta pachyrhyncha).

Animals—15 healthy thick-billed parrots.

Procedures—Anesthesia was induced and maintained with isoflurane in oxygen. In the first bird that was anesthetized, end-tidal isoflurane concentration was maintained at 1.0% for 15 minutes. After this period of anesthetic equilibration, an end-tidal gas sample was obtained for verification of isoflurane concentration. A toe was pinched to determine the bird’s response to pain, and the bird was then allowed to recover from anesthesia. To determine MAC, a so-called up-and-down approach was subsequently used in all 15 birds. Compared with the isoflurane concentration used for MAC determination in the first bird, maintenance isoflurane concentration for the second bird was increased by approximately 10% if the first bird reacted and decreased by approximately 10% if the first bird did not react to a toe pinch. These steps were then followed until all 15 birds had been anesthetized. Crossover events occurred when birds in sequence had discordant results (ie, 1 reactor and 1 nonreactor). The MAC was defined as the mean of the isoflurane concentrations measured during these crossover events.

Results—Mean MAC of isoflurane in thick-billed parrots was estimated to be 1.07% (95% confidence interval, 0.97% to 1.16%).

Conclusions and Clinical Relevance—Isoflurane MAC appears to be lower in thick-billed parrots than the MAC determined for other bird species. Determination of the species-specific requirements of thick-billed parrots should allow isoflurane anesthesia to be performed more safely in this endangered species. (Am J Vet Res 2008;69:189–194)

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soflurane is an inhalant anesthetic agent that was first introduced for use in avian species in 1985, and it is currently considered the most common anesthetic agent used in birds.1,2 In mammals, the potency of an inhalant anesthetic agent is typically defined by the MAC that prevents gross purposeful movement in response to a supramaximal noxious stimulus in 50% of the animals.3 The term MAC is used in a similar context in birds as it is in mammals. However, because birds do not have pulmonary alveoli, MAC in birds refers to the minimum concentration of pulmonary (rather than alveolar) anesthetic gas.4 The MAC of isoflurane in birds has only been determined for a few species. Values have been estimated to be 1.30 ± 0.23% in Pekin ducks (Anas platyrhynchos), 1.34 ± 0.14% in sandhill cranes (Grus canadensis), 1.25 ± 0.13% in domestic chickens (Gallus domesticus), and 1.44 ± 0.07% in cockatoos (Cacatua spp).4–7 Considering that there are approximately 9,700 known living species of birds,8 there appears to be a high potential for species-specific differences in isoflurane potency that have not been described. Although the MAC of a given inhalant anesthetic agent is fairly consistent among mammalian species, some species differences have been reported. For example, the MAC of isoflurane in cats has been estimated to be at least 20% higher than the MAC in dogs.9,10 Similar differences may exist among bird species.

Anecdotally, we have observed that thick-billed parrots (Rhynchopsitta pachyrhyncha) require relatively low vaporizer settings for maintenance of anesthesia. These parrots are an endangered species that are kept in zoos and private institutions for exhibit and propagation. They are native to the southwestern United States.
and northern Mexico, but have become endangered because of habitat loss and collection for the pet trade. In 2004, the veterinarian for the Thick-billed Parrot Species Survival Plan reported unexplained anesthesia-related deaths that were associated with the concurrent use of isoflurane and butorphanol among birds of this species. On the basis of our clinical experience, we were aware that this species was highly responsive to isoflurane, and we theorized that anesthesia-related deaths might be attributable, in part, to a lower isoflurane MAC than values determined for other species of birds. The purpose of the study reported here was to determine the MAC of isoflurane and assess heart rate, respiratory rate, and arterial blood gas variables during anesthesia at MAC in thick-billed parrots (Rhyynchopitita pachyrhyncha).

Materials and Methods

Birds—This study was approved by the Institutional Animal Care and Use Committee of the University of California, Davis. Fifteen thick-billed parrots (8 males and 7 females) that were housed at the Sacramento Zoo were included in the study. Birds were moved in covered transport kennels by car from the Sacramento Zoo to the anesthesia laboratory at the University of California, Davis. Duration of transportation to the laboratory was 20 to 30 minutes. Once in the laboratory, each bird was allowed to rest for at least 30 minutes in a dark and quiet room.

Procedures—After the period of acclimation, each parrot was physically restrained with a towel and anesthesia was induced with isoflurane in 100% oxygen via a face mask and a Jackson-Rees breathing system with a flow rate of 1 L/min and an initial isoflurane vaporizer anesthetic concentration setting of 2%. The vaporizer setting was increased by 0.5% every 30 seconds until the bird was no longer responsive and had profound muscle relaxation (maximum setting of 4% for 4.5% for all birds). During induction, respiratory rate (breaths/min) was assessed visually and heart rate (beats/min) was assessed via auscultation with a stethoscope. Once anesthesia was induced, the bird’s trachea was intubated with a noncuffed endotracheal tube (internal diameter, 2.5 or 3.0 mm). The isoflurane concentration was then reduced to 2.5%; it was maintained at this value while monitoring instrumentation was attached.

A 3.3-F catheter was inserted through the lumen of the endotracheal tube, so that the tip was level with the tip of the endotracheal tube. This catheter was used to collect samples of end-tidal gas with minimal mixing with fresh anesthetic gas. The catheter was connected to a Raman spectrometer for continuous measurement of end-tidal CO₂ concentration and isoflurane concentrations. This decreased the internal lumen of the 2.5- and 3.0-mm-diameter endotracheal tubes by 47% and 39%, respectively. However, this technique has been used previously in studies of small animals such as cats. As during induction of anesthesia, heart rate was monitored via auscultation with a stethoscope, and respiratory rate was monitored visually. A temperature probe, calibrated prior to each experiment against a certified thermometer, was placed in the esophagus at a position distal to the thoracic inlet. A forced-air temperature management unit was used during the entire procedure to minimize loss of body temperature. A lead II ECG was continuously recorded by use of 25-gauge needles placed through the skin and a physiograph. The bird was allowed to breathe with additional positive-pressure ventilation administered by use of a mechanical ventilator at a rate of 5 breaths/min and a peak inspiratory pressure of 10 cm H₂O. Heart rate, respiratory rate, esophageal temperature, and end-tidal CO₂ and isoflurane concentrations were recorded every 5 minutes. Each bird received a full physical examination during the procedure.

A blood sample (0.15 to 0.20 mL) was obtained from a superficial ulnar artery immediately after instrumentation was completed and again immediately after stimulation at the end of the procedure. Arterial blood samples were immediately analyzed for Pₐ, PₐO₂, and pH by use of a blood gas analyzer. Blood gas measurements were corrected for body temperature by use of mammalian formulae.

On completion of instrumentation, the end-tidal isoflurane concentration was reduced and adjusted to a predetermined target concentration. End-tidal isoflurane concentration was maintained for 15 minutes to allow for anesthetic equilibration. After the equilibration period, end-tidal gas samples (1 mL) were collected by hand over 10 to 20 breaths by use of a glass syringe, and end-tidal isoflurane concentration was measured by use of an infrared analyzer. This analyzer was calibrated before each experiment with room air and 3 calibration standards of known isoflurane concentrations (0.9%, 1.5%, and 2.3%). End-tidal samples were taken and measured in triplicate, and the mean end-tidal isoflurane concentration was calculated. If the mean end-tidal isoflurane concentration deviated by > 5% from the target concentration, the vaporizer setting was adjusted and an additional period of 10 minutes was allowed to elapse for equilibration. Sample collection and assessments of end-tidal gases were repeated at 10-minute intervals until the target concentration was reached.

A so-called up-and-down method was used to estimate MAC and determine the target end-tidal isoflurane concentrations. In the first bird, end-tidal isoflurane concentration was set at 1.0%. The target isoflurane concentration for all other birds was adjusted depending on the response of the previously evaluated bird. A digit was pinched by use of a hemostat until gross purposeful movement was observed or until 1 minute had elapsed, whichever occurred first. The response was considered positive if gross purposeful movement was observed; if no such movement was observed. If the first bird had a positive response to stimulation, the target concentration for the second bird was increased by approximately 10% to 1.10%. Conversely, if the first bird had a negative response to stimulation, the target concentration for the second bird was decreased by approximately 10% to 0.90%. This procedure was repeated until all 15 birds had been anesthetized. Crossover events were recorded; these were instances in which independent
pairs of birds evaluated in succession had opposite responses (ie, within a pair, the first bird had a negative response, but the second bird had a positive response, or vice versa). Individual birds could not be included in >1 crossover event.15,16

Once the bird’s response to stimulation was evaluated, 1 ml of blood was obtained from a jugular vein; a CBC and plasma biochemical analyses were performed to screen for any underlying medical condition that was not detected during physical examination. No bird had >5% of blood volume removed during the study, and maximum recommended values for routine phlebotomy in birds is ≤10% of blood volume.17 On completion of blood sample collection, the instrumentation was removed. Direct pressure was applied to the venous puncture site, and the bird was allowed to recover from anesthesia in hand until it was capable of standing. It was then placed back in its transport kennel until fully recovered. All birds were allowed to rest for at least 30 minutes, after which they were transported back to the Sacramento Zoo and released into their enclosure.

During the experiment, various intervals were also recorded for each bird. Time to induction was defined as the interval from placement of the anesthetic mask until intubation; time to instrumentiation was defined as the interval from intubation until placement of all monitoring equipment; procedure time was defined as the interval from placement of monitoring equipment until stimulation; and time to recovery was defined as the interval from stimulation until the bird had regained sufficient consciousness to be placed standing back in its transport carrier.

Data analysis—Body weights and physiologic variable data were tested for normality (Shapiro-Wilk test) by use of computer software.2 For data sets that were normally distributed, paired Student t tests were used to compare physiologic variables at the beginning and end of the study, and data are presented as mean ± SD. Wilcoxon matched-pair tests were used to analyze data sets that were not normally distributed, and those data are reported as median and range. Significance was set at a value of P < 0.05.

Minimum alveolar concentration of isoflurane was calculated by use of Dixon’s up-and-down technique and quantal analysis.15,16 For the up-and-down technique, MAC was defined as the mean of isoflurane concentrations measured during crossover events. The 95% confidence intervals for MAC were calculated by use of binomial probability. Quantal analysis was used to calculate the probability of movement (response to stimulus) as a function of anesthetic dose, where MAC (or ED50 [median effective dose]) was the dose at which that probability was 50%.16 Data underwent logistic and nonlinear regression by use of computer software.26 No difference in data fit among the 3 statistical procedures used was detected, and only the probit curve is reported. The ED50 and fiducial intervals were estimated from this curve. For all statistical analyses, a value of P < 0.05 was considered significant.

Results

Physiologic variables—Most physiologic data met criteria for normality and were reported as mean ± SD. Thick-billed parrots weighed 324 ± 24 g. No remarkable abnormalities were detected via physical examination. Results of the CBCs and plasma biochemical analyses were within reference ranges for thick-billed parrots.18 Heart rates, respiratory rates, and esophageal temperatures were significantly lower at the time that the monitors were attached than values recorded before induction of anesthesia and were significantly lower at the end of the experiment than values at the time that the monitors were attached (Table 1). Values for end-tidal CO2 concentration did not change over time.

Time to induction was 1.5 minutes for 6 birds and 2 minutes for 9 birds. For all birds, mean ± SD time to instrumentation was 6 ± 1 minutes, total procedure

<table>
<thead>
<tr>
<th>Variable</th>
<th>Before induction</th>
<th>Start</th>
<th>End</th>
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</thead>
<tbody>
<tr>
<td>Heart rate (beats/min)</td>
<td>365 ± 97</td>
<td>281 ± 50</td>
<td>221 ± 47</td>
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<tr>
<td>Respiratory rate (breaths/min)</td>
<td>53 ± 14</td>
<td>28 ± 9</td>
<td>19 ± 6</td>
</tr>
<tr>
<td>End-tidal CO2 concentration (mm Hg)</td>
<td>NA</td>
<td>26 ± 6</td>
<td>28 ± 5</td>
</tr>
<tr>
<td>Temperature (°C)</td>
<td>ND</td>
<td>38.7 ± 0.8</td>
<td>38.9 ± 1.3</td>
</tr>
</tbody>
</table>

Table 1—Mean ± SD heart rate, respiratory rate (with 5 assisted breaths/min), and esophageal temperature in 15 thick-billed parrots that were anesthetized with isoflurane in 100% oxygen. Measurements were obtained before induction of anesthesia, at the time that monitoring equipment was attached (start), and after supramaximal noxious stimulation had been applied (end).

<table>
<thead>
<tr>
<th>Variable</th>
<th>Start</th>
<th>End</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>7.49 (7.44–7.6)</td>
<td>7.47 (7.41–7.59)</td>
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<tr>
<td>Pco2 (mm Hg)</td>
<td>28.5 (20–35)</td>
<td>33.0 (24–42)</td>
</tr>
<tr>
<td>Paco2 (mm Hg)</td>
<td>557 (478–587)</td>
<td>553 (461–579)</td>
</tr>
</tbody>
</table>

Table 2—Median (range) arterial blood gas variables and pH in 8 thick-billed parrots that were anesthetized with isoflurane in 100% oxygen. Measurements were obtained at the time that monitoring equipment was attached (start) and after supramaximal noxious stimulation had been applied (end).

![Figure 1—Quantal analysis curve to estimate the minimum anesthesia concentration of isoflurane needed for a 50% probability (P) of nonresponse to supramaximal noxious stimulation among 15 thick-billed parrots that were anesthetized with isoflurane. The minimum anesthesia concentration of isoflurane was 1.01%](image_url)
time was 36 ± 8 minutes, and time to recovery was 7 ± 9 minutes. Because of technical problems, arterial blood samples were collected at both time points (beginning and end of procedure) from only 8 parrots. Blood gas values did not meet tests of normality and are reported as median and range (Table 2). There were no differences in blood gas variables and arterial blood pH values at the beginning and end of the anesthetic episode.

MAC—By use of the up-and-down method, the MAC of isoflurane in thick-billed parrots was estimated to be 1.07 ± 0.1%. Quantal analysis estimated that a concentration of 1.01% isoflurane was needed for a 50% probability of movement to supramaximal noxious stimulation (Figure 1). The fiducial interval for that probability was 0.30% to 1.61%.

Discussion

Determination of MAC in animals can be achieved by multiple means. In many investigations, MAC is determined by the bracketing method. With that technique, MAC is determined in each individual by evaluating the response to a supramaximal noxious stimulus during the administration of various concentrations of the inhalant anesthetic agent. The population MAC is then estimated from the mean of the individual MAC values. That method has traditionally been used in many veterinary studies because it requires use of a low number of animals. However, each individual must be anesthetized for long periods of time (often 2 to 6 hours) because multiple concentrations of the agent are investigated in a single anesthetic event and time to achieve equilibration after each change in anesthetic concentration is required. Such long anesthetic events may increase anesthesia-related morbidity and mortality rates, and it is commonly recommended to avoid such long anesthetic episodes in birds. Studies involving the bracketing method would not be acceptable for an endangered species such as the thick-billed parrot, particularly when anesthesia-related problems in that species have been reported in the past. The up-and-down method is an alternative approach to determination of MAC. Instead of administering various concentrations of inhalant anesthetic agent to each individual, the concentration is changed in animals in succession. This method requires a greater number of individuals, but the duration of the anesthetic event remains short for each individual. The up-and-down method has provided results that are comparable to those achieved by use of the bracketing method. By use of the up-and-down method, the anesthetic concentration is successively increased or decreased by approximately 10% in a series of animals; the adjustment is based on the response of the previously evaluated animal. A crossover event occurs when the response changes between one animal and the next in the experimental sequence. The anesthetic concentrations at crossover events are used to calculate MAC. A minimum of 4 crossover events is necessary to obtain a valid estimate of MAC. Among the thick-billed parrots used in the present study, 5 crossover events were obtained for the calculation of MAC.

To verify the accuracy of the MAC estimation, quantal analysis was used to fit probability curves to the data in the present study. Three statistical methods were used to determine the probability of movement by the birds as a function of isoflurane concentration. Moreover, each procedure was used to fit either a probit or logistic curve to the data. These procedures offer different algorithms for fitting curves to this type of data; because the sample size was relatively small, the choice of an algorithm may be important. However, no difference in fit was evident, and the selection of a particular procedure was therefore arbitrary. The probit curve fit was selected because it allowed the estimation of fiducial intervals. The MAC values determined by use of the 2 methods were similar (1.07% and 1.01%). These values were not considered different because a 10% concentration change was effected between birds in succession to determine MAC and the measurement error is therefore likely to be approximately 3% to 10%.

The estimate of MAC for isoflurane in thick-billed parrots (1.07%) differed by > 10% from that reported for cockatoos (1.44 ± 0.07%), sandhill cranes (1.34 ± 0.14%), domestic chickens (1.25% ± 0.13%), and Pekin ducks (1.30 ± 0.23%). This appears to confirm our clinical impression that anesthesia can be maintained in thick-billed parrots by administration of low concentrations of isoflurane, compared with concentrations required by other bird species. However, the method used to determine MAC in the thick-billed parrots was different from that used to determine MAC in those other bird species and may have contributed to this discrepancy. In domestic dogs, MAC determined via the bracketing method was similar to that determined by the up-and-down method; however, that finding does not guarantee that the same equivalency applies to other species.

It is also possible that MAC was artificially low in the thick-billed parrots because the stimulus used was not truly supramaximal. The amount of pressure placed on the thick-billed parrot digits was not measured. However, the pressure measurement and intensity of the stimulation should not be important as long as the stimulus is in the supramaximal range (ie, increases in intensity do not increase the response). The toe pinch is the stimulus most commonly used for MAC determinations in birds; among studies in which this technique is used in the determination of MAC, similarity of results would be expected. Other factors that may affect MAC include profound hypotension, severe hypoxemia, severe hypercapnia, and changes in body temperature. In the birds of the present study, arterial blood gas values indicated that neither hypercapnia nor hypoxemia developed. Furthermore, all birds received 100% oxygen and intermittent positive-pressure ventilation; procedures were quite short, the birds’ arteries and veins appeared well filled when blood samples were taken, and no more than 0.001% of blood volume was removed from any bird prior to MAC determination. However, blood pressure was not measured; thus, we cannot confirm whether hypotension was present. Profound hypotension was not suspected in these birds, and even moderate hypotension is not thought to have an effect on MAC.

During the study period, there was a significant decrease in the birds’ body temperature, but this change
was not clinically important because the magnitude of change was < 0.9°C and considered within acceptable limits for anesthetized birds. It is considered unlikely that this decrease in body temperature accounted for the relatively lower MAC value in these parrots because the reduction in temperature was comparable to decreases identified during studies of the effects of isoflurane in other avian species.

Although attempts were made to stabilize body temperature, fluid support was not administered to birds during the present study to avoid confounding effects associated with placement of additional needles and fluid therapy on cardiopulmonary and clinicopathologic variables. Although fluids certainly could have been administered at the end of the procedure, we concluded that such administrations were not necessary because of the short duration of the anesthetic episodes and low volume of blood that was collected from each bird. Birds were given free access to drinking water immediately after their return to the zoo.

The birds used in the present study were transported and anesthetized on the same day for logistic purposes. Birds were housed at the Sacramento Zoo, and the equipment used for monitoring could not be readily transferred to the zoo. However, the duration of transportation to and from the zoo was only 20 to 30 minutes. Birds are often transported a short distance from their home enclosure to veterinary facilities prior to anesthesia, and the protocol used in the present study was consistent with typical procedures. To minimize the effects of transportation, birds were allowed a period of 30 minutes to acclimate to their new surroundings prior to commencement of the experiment.

The relatively low MAC for isoflurane in thick-billed parrots may help explain the anesthesia-related deaths associated with concurrent isoflurane and butorphanol administration in this species. In cockatoos, the MAC of isoflurane decreased from 1.44% to 1.08% when butorphanol was administered. The effects of butorphanol were not assessed in the previous thick-billed parrots investigation. However, it is presumed that butorphanol would lower MAC of isoflurane in thick-billed parrots in a manner similar to that detected in cockatoos, which would decrease the MAC to < 1.0%. Such a low MAC of isoflurane in thick-billed parrots could explain the cardiopulmonary depression and deaths that occurred in thick-billed parrots that were administered both isoflurane and butorphanol. Maintenance settings of 1.5% to 2% isoflurane have been recommended for high-risk avian patients, and this may simply be too high for maintenance of anesthesia in thick-billed parrots, particularly if they have received butorphanol.

The technique used to measure end-tidal isoflurane concentration in the present study involved placement of a catheter into the lumen of the endotracheal tube with a resultant decrease in the diameter of the tube lumen. Theoretically, such a decrease would have caused increased resistance during ventilation, resulting in hyperventilation, acidosis, or increased respiratory effort. None of these changes were detected in the birds of our study, and the effects of the decrease in luminal diameter may have been mitigated by the intermittent positive-pressure ventilation that was provided in addition to the birds’ innate breathing.

In the present study, the birds’ heart rate, respiratory rate, and esophageal temperature decreased over time. Similar decreases are commonly detected in birds that are anesthetized with isoflurane, and none of these changes were considered clinically problematic. However, the decrease in heart rate was greater than expected. During anesthesia with isoflurane, cockatoos, galahs (Eolophus roseicapillus), and Hispaniolan Amazon parrots (Amazona ventralis), which are psittacine species similar in size to thick-billed parrots, had heart rates of 400 ± 16 beats/min, 318 ± 56 beats/min, and > 400 beats/min, respectively. After 30 minutes of isoflurane anesthesia, heart rate values in the thick-billed parrots (221 ± 47 beats/min) were substantially lower than values reported for those other psittacines and were more similar to values for larger birds such as Pekin ducks (236 to 248 beats/min). This may reflect species-specific effects of isoflurane in thick-billed parrots or may be related to other study factors.

Ventilation appeared to be adequate in the birds of the present study because they were normocapnic to slightly hypocapnic. Resting PaCO₂ values of 25 to 40 mm Hg have been reported in birds, and values were within this range for most birds in our study at most time points. Similarly, arterial blood pH was similar or slightly high (resulting in mild respiratory alkalosis for some study birds), compared with values in other anesthetized birds. The values for PaCO₂ were higher than values reported for other similarly sized birds that were anesthetized with isoflurane but did not receive assisted ventilation. Assisted ventilation was performed in our study in an attempt to obtain optimal end-tidal gas samples for determination of end-tidal isoflurane concentrations with optimal precision. The assisted ventilation induced only mild changes in PaCO₂, and those changes were not considered clinically important. Hyperventilation is not thought to have an effect on MAC.

The results of the present study have suggested that the MAC of isoflurane in thick-billed parrots is substantially lower than values reported for other avian species. Knowledge of this difference should enable anesthetic procedures involving thick-billed parrots to proceed more safely. Although general guidelines regarding MAC in birds may be useful, the data obtained in our study suggested that important differences in anesthetic potency may exist among avian species, and those differences should be investigated when possible.

   c. Uncuffed endotracheal tube, Rüsch Inc, Duluth, Ga.
   d. Tom Cat 3.5-French catheter, Tyco Healthcare Group LP, Mansfield, Mass.
   e. Raman spectrometer Rascal II, Ohmeda, Salt Lake City, Utah.
   g. ECG physiograph, Gould Instrument Systems, Valley View, Ohio.
   h. Bird Mark 7, VIASys Staisys Healthcare, Palm Springs, Calif.
   i. NPB 44 capnograph Nellcor, Pleasanton, Calif.
   j. ABL 3, Radiometer, Copenhagen, Denmark.
k. Medical gas analyzer LB1, Beckman Instruments, Schiller Park, Ill.
m. LOGISTIC, PROBIT, and GENMOD procedures in SAS/STAT, version 8.0, SAS, Cary, NC.

References