Determination of the minimum anesthetic concentration of sevoflurane in thick-billed parrots (*Rhynchopsitta pachyrhyncha*)

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Objective—To determine the minimum anesthetic concentration (MAC) of sevoflurane in thick-billed parrots (*Rhynchopsitta pachyrhyncha*) and compare MAC obtained via mechanical and electrical stimulation.

Animals—15 healthy thick-billed parrots.

Procedures—Anesthesia was induced in each parrot by administration of sevoflurane in oxygen. An end-tidal sevoflurane concentration of 2.5% was established in the first bird. Fifteen minutes was allowed for equilibration. Then, 2 types of noxious stimulation (mechanical and electrical) were applied; stimuli were separated by 15 minutes. Responses to stimuli were graded as positive or negative. For a positive or negative response to a stimulus, the target end-tidal sevoflurane concentration of the subsequent bird was increased or decreased by 10%, respectively. The MAC was calculated as the mean end-tidal sevoflurane concentration during crossover events, defined as instances in which independent pairs of birds evaluated in succession had opposite responses. A quantal method was used to determine sevoflurane MAC. Physiologic variables and arterial blood gas values were also measured.

Results—Via quantal analysis, mean sevoflurane MAC in thick-billed parrots determined with mechanical stimulation was 2.35% (90% fiducial interval, 1.32% to 2.66%), which differed significantly from the mean sevoflurane MAC determined with electrical stimulation, which was 4.24% (90% fiducial interval, 3.61% to 8.71%).

Conclusions and Clinical Relevance—Sevoflurane MAC in thick-billed parrots determined by mechanical stimulation was similar to values determined in chickens and mammals. Sevoflurane MAC determined by electrical stimulation was significantly higher, which suggested that the 2 types of stimulation did not induce similar results in thick-billed parrots. (*Am J Vet Res* 2012;73:1350–1355)

Thick-billed parrots (*Rhynchopsitta pachyrhyncha*) are an endangered parrot species that has been extirpated from the United States. The species originally ranged from southern New Mexico and Arizona to central Mexico but is currently limited to small forested areas in Mexico.[1] Thick-billed parrots housed in Association of Zoos and Aquariums institutions are overseen by a Species Survival Plan, and efforts are underway to increase breeding of this species in captivity and the wild.[2] As with many bird species, anesthesia is routinely used in this species to facilitate physical examinations, data collection, diagnostic testing, and medical and surgical treatments.

Abbreviations

CI	Confidence interval
IQR	Interquartile range
MAC	Minimum anesthetic concentration
P_{ETCO_2}	End-tidal partial pressure of carbon dioxide

The potency of inhalation anesthetics in birds is commonly characterized by the MAC, which is described as the end-tidal anesthetic concentration at 1 atm at which 50% of anesthetized individuals will not move in response to a supramaximal noxious stimulus.[3–5] This is equivalent to the minimum alveolar concentration described in mammals but differs in that birds do not have alveoli; therefore, a pulmonary an-

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esthetic concentration is measured.[6] Investigators of a study[7] in captive thick-billed parrots determined that the MAC for isoflurane is lower for this species than for other birds.

Sevoflurane is another inhalation anesthetic that is commonly used in birds. In some species, sevoflurane has shorter anesthetic induction and recovery times, less ataxia on recovery, relatively mild circulatory and respiratory depressant effects, and a reduced frequency of cardiac arrhythmias, compared with effects for isoflurane.[8–11] In dogs and cats, sevoflurane has a lower blood-gas partition coefficient than does isoflurane and is therefore expected to provide more rapid anesthetic induction and recovery times and to allow quicker changes in the depth of anesthesia.[11] These qualities make sevoflurane an attractive option for anesthetic induction and maintenance in birds. However, the only avian species for which the sevoflurane MAC has been determined is domestic chickens, with a mean ± SD MAC of 2.21 ± 0.32%.[12] This value is similar to values reported in domestic mammals, where MAC ranges from 1.71% to 3.41%.[12,13] Given that thick-billed parrots have a relatively low isoflurane MAC,[7] we hypothesized that they would also have a relatively low sevoflurane MAC. Knowledge of the MAC of sevoflurane is important if this anesthetic is to be used safely and effectively in thick-billed parrots.

Determination of MAC relies on animal responses to supramaximal stimulation. Historically, mechanical clamping or pinching of a digit or the tail has been commonly used to create supramaximal stimulation in animals.[5,12] However, electrical stimulation may be a preferred alternative to mechanical stimulation because it is thought that electrical stimulation can be applied in a more consistent manner, induce similar results, and be more appropriate in certain experimental situations.[14,15] To our knowledge, there is no information comparing mechanical versus electrical stimulation for MAC determination in birds. We are aware of only 1 prior avian study[6] in which investigators used electrical stimulation as a suprmaximal stimulus. Therefore, the purpose of the study reported here was to determine the sevoflurane MAC in thick-billed parrots and to evaluate heart rate, respiratory rate, P_{ETCO_2}, and blood gas values during anesthesia with sevoflurane and to compare the use of mechanical stimulation and electrical stimulation for MAC determination.

Materials and Methods

Birds—Fifteen thick-billed parrots (5 males and 10 females) 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. It required 20 to 30 minutes to transport birds from the zoo to the laboratory. The study was approved by the Institutional Animal Care and Use Committee of the University of California-Davis.

Procedures—After arrival at the laboratory, each bird was allowed to rest for at least 30 minutes in a dark and quiet room prior to anesthetic induction. A 30-minute acclimation period was deemed sufficient to reduce the stress of transport; a longer acclimation period would have induced additional stress as a result of a longer period of food withholding.

After the acclimation period, each parrot was physically restrained with a towel, and an initial respiratory rate and heart rate were measured by observation of the bird and auscultation with a stethoscope. Anesthesia was then induced with sevoflurane[a] in 100% oxygen (1 L/min) administered via a face mask and a Bain nonrebreathing system (designated as time 0). The initial sevoflurane vaporizer[b] anesthetic concentration was set at 5% until the bird had profound muscle relaxation.

After sufficient relaxation was achieved, each bird was intubated with a noncuffed endotracheal tube[c] (internal diameter, 2.5 to 3.0 mm). Prior to intubation, a catheter[d] was inserted into the proximal end of the endotracheal tube so that the catheter tip was close to the distal end of the endotracheal tube. This catheter was used to collect samples of end-tidal gas with minimal mixing with fresh anesthetic gas. After tracheal intubation, the sevoflurane concentration was decreased to approximately 3% while the bird was instrumented for anesthetic monitoring. Mechanical ventilation[e] was initiated at a rate of 5 breaths/min, with a peak inspiratory pressure of 10 cm H_2O.[3,7] A capnograph[f] was attached to the catheter within the endotracheal tube to provide continuous measurement of P_{ETCO_2}. A temperature probe,[g] which was calibrated against a certified thermometer prior to each experiment, was placed in the esophagus at a position distal to the thoracic inlet. A Doppler ultrasonographic probe[h] was secured over the ulnar artery for continuous monitoring of the heart rate. A forced-air warming unit[i] and circulating warm water blanket[j] were used during the procedure to minimize loss of body heat and maintain body temperature between 38° and 41°C. Heart rate, respiratory rate (the sum of spontaneous respiration and 5 manually supplied ventilator breaths/min), P_{ETCO_2}, body temperature, and sevoflurane concentration were recorded approximately every 5 minutes throughout the procedure.

Immediately after instrumentation was completed, an arterial blood sample was collected from a superficial ulnar artery and analyzed for P_{O_2}, P_{CO_2}, pH, and electrolyte and glucose concentrations via a blood gas analyzer,[k] with corrections for body temperature performed on the basis of an equation used for mammalian samples. After instrumentation was completed, the end-tidal sevoflurane concentration was set to a predetermined target concentration (2.5% for the first bird). The sevoflurane concentration was maintained at this concentration for 15 minutes to allow for anesthetic equilibration. End-tidal gas samples were then collected manually via the catheter in the endotracheal tube, and the end-tidal sevoflurane concentration was measured with a calibrated infrared spectrometer.[l]

After the target end-tidal sevoflurane concentration had been maintained for 15 minutes, 2 types of noxious stimulation (mechanical and electrical) were applied to evaluate the depth of anesthesia. The stimuli were administered in stratified random order and separated by an interval of 15 minutes. Mechanical stimulation was performed by pinching a digit on the left leg with a hemostat until gross purposeful movement was observed

or until 1 minute had elapsed. For consistency, the same investigator applied all mechanical stimuli in the study. Electrical stimulation (50 V, 50 Hz, and 10 milliseconds)[6,14] was applied via needle electrodes[m] inserted SC at the medial aspect of the right thigh in proximity to the tibial nerve; electrical stimulation was applied until gross purposeful movement was observed or until 1 minute had elapsed. For either stimulus, the response was considered positive if purposeful movement was observed (moving wings or head or retracting the contralateral leg) and negative if no response was observed. The target end-tidal sevoflurane concentration for each subsequent bird was determined on the basis of the response of the preceding bird. If the preceding bird had a positive response, the target sevoflurane concentration for the subsequent bird was increased by 10%, whereas if the preceding bird had a negative response, the target sevoflurane concentration for the subsequent bird was decreased by 10%. This procedure was repeated for all remaining birds. If the response differed between mechanical and electrical stimulation in an individual bird, the target sevoflurane concentration for the subsequent bird was changed in accordance with the response of the preceding bird to the specific stimulus being applied.

Once the responses to both stimuli had been assessed, a second arterial blood sample was collected and analyzed via the blood gas analyzer. The bird then received a complete physical examination, including measurement of body weight. One milliliter of blood was obtained from the right jugular vein for a CBC and plasma biochemical analysis to assess the overall health status of the bird. Each bird was vaccinated against West Nile virus[n] (1 mL, IM). Fluids[o] (30 mL/kg, SC) were administered. Sevoflurane administration was then discontinued, and instruments were removed. Each bird was allowed to recover until it was capable of standing and was then extubated and returned to a transport kennel until it was completely recovered. All birds were allowed to rest for at least 30 minutes prior to return transport to the Sacramento Zoo.

Time to anesthetic induction, time to intubation, time to instrumentation, time to recovery, and total anesthetic time were recorded for each bird. Time to anesthetic induction was defined as the time from the start of sevoflurane administration (via face mask) to complete muscle relaxation. Time to intubation was defined as the time from the start of sevoflurane administration to successful intubation. Time to instrumentation was defined as the time from the start of sevoflurane administration to completion of instrumentation. Time to recovery was defined as the time from termination of sevoflurane administration to extubation. Total anesthetic time was defined as the time from the start of sevoflurane administration to extubation.

Data analysis—Body weight, time intervals, physiologic variables, and blood gas values were assessed for normality via the Shapiro-Wilk test. Data were reported as median and IQR. Analysis of physiologic values, blood gas values, and time intervals associated with recovery were conducted with generalized estimating equations to determine the effects of time and sevoflurane concentration. Values of $P < 0.05$ were considered significant. These analyses were performed with commercially available software.[p]

The sevoflurane MAC for mechanical stimulation and electrical stimulation were estimated via the up-and-down method[16] and were reported as mean and 95% CI. These estimates were compared via a paired *t* test. In addition, a probit model with the assumption of equal slopes was fitted to the data with commercially available software.[q] A 90% fiducial limit was used. The 2 methods of stimulation were compared via a generalized linear model, with values of $P < 0.05$ considered significant.

Results

Physiologic variables—Most variables analyzed were not normally distributed and therefore were reported as median and IQR. Median body weight of the thick-billed parrots was 320 g (IQR, 307.5 to 332.5 g). No remarkable abnormalities were detected during physical examination. Results of the CBC and plasma biochemical analyses were within reference ranges for thick-billed parrots.[17] Median time to anesthetic induction was 3 minutes (IQR, 2 to 3.5 minutes), median time to intubation was 4 minutes (IQR, 3 to 4.5 minutes), and median time to instrumentation was 6 minutes (IQR, 5 to 6.5 minutes). No effects of sevoflurane concentration were detected for time to recovery (median, 3 minutes; IQR, 2 to 3.5 minutes) or total anesthetic time (median, 64 minutes; IQR, 60.5 to 69 minutes). Heart rate was not significantly affected by sevoflurane concentration but was significantly affected by total anesthetic time. Heart rate was highest during manual restraint (median, 300 beats/min; IQR, 240 to 340 beats/min), with similar values at the time of instrumentation (median, 300 beats/min; IQR, 240 to 320 beats/min), but was significantly lower at 40 minutes after the start of anesthesia (median, 240 beats/min; IQR, 200 to 300 beats/min). Respiratory rate (the sum of spontaneous respiration and 5 manually supplied ventilator breaths/min) was highest prior to anesthetic induction during manual restraint (median, 44 breaths/min; IQR, 40 to 60 breaths/min) and lowest once the birds were anesthetized (specifically at 5% sevoflurane, which was the vaporizer setting for anesthetic induction and some of the electrical stimulations). Most birds continued to have spontaneous respiration to some degree during anesthesia, but the respirations were usually less frequent than when birds were conscious. No significant effects of time or sevoflurane concentration were detected for body temperature or P_{ETCO_2} (**Table 1**).

Paired blood gas values were obtained in 10 of 15 birds. For blood gas values, there was no effect of time or sevoflurane concentration on P_{O_2} (median, 522 mm Hg; IQR, 513 to 540 mm Hg) and potassium (median, 3.8 mmol/L; IQR, 3.4 to 4.4 mmol/L) or sodium (median, 145 mmol/L; IQR, 143 to 147 mmol/L) concentrations. Glucose and chloride concentrations and P_{CO_2} were all affected by total anesthetic time but not by sevoflurane concentration. Glucose concentrations increased significantly ($P < 0.01$) with time, starting at a median of 250 mg/dL (IQR, 236 to 267 mg/dL) and increasing to a median of 273 mg/dL (IQR, 262 to 281 mg/dL). Chloride concentrations decreased significantly (*P*

Table 1—Median (IQR) values for heart rate, respiratory rate,* esophageal temperature, and P_{ETCO_2} in 15 thick-billed parrots (*Rhynchopsitta pachyrhyncha*) anesthetized with sevoflurane in oxygen.

Variable	Before anesthetic induction	Instrumentation	40 minutes†
Heart rate (beats/min)	300 (240–340)[a]	300 (240–320)[a]	240 (200–300)[b]
Respiratory rate (breaths/min)	44 (40–60)[a]	22 (14–26)[b]	22 (14–34)[b]
Temperature (°C)	ND	40.4 (39.9–40.8)	39.9 (39.4–40.2)
P_{ETCO_2} (mm Hg)	ND	24 (20–28)	24 (19–30)

*Includes spontaneous respirations and 5 manually supplied ventilator breaths/min. †Start of administration of sevoflurane via face mask was designated as time 0.
ND = Not determined.
[a,b]Within a row, values with different superscript letters differ significantly ($P < 0.05$).

< 0.01) with time, starting at a median of 117 mmol/L (IQR, 116 to 118 mmol/L) and decreasing to a median of 114 mmol/L (IQR, 111 to 116 mmol/L). The P_{CO_2} increased significantly ($P = 0.01$) with time, starting at a median of 37.6 mm Hg (IQR, 34.1 to 39.6 mm Hg) and increasing to a median of 38.6 mm Hg (IQR, 36.6 to 48.4 mm Hg). Blood pH (median, 7.45; IQR, 7.40 to 7.50; range, 7.32 to 7.54) was significantly affected by sevoflurane concentration ($P = 0.02$) but not by time ($P = 0.12$). Blood pH typically decreased with increases in the sevoflurane concentration.

MAC—Four crossover events (defined as instances in which separate pairs of birds evaluated in succession had opposite responses to the same stimuli) were obtained for both the mechanical and electrical stimulations. Mean MAC of sevoflurane in thick-billed parrots for mechanical and electrical stimulation estimated via the up-and-down method[16] was 2.39% (95% CI, 2.13% to 2.65%) and 3.94% (95% CI, 3.46% to 4.42%), respectively. These values differed significantly ($P = 0.002$). Mean MAC estimated with the probit model was 2.35% (90% fiducial limit, 1.32% to 2.66%) and 4.24% (90% fiducial limit, 3.61% to 8.71%) for mechanical and electrical stimulation, respectively. These 2 estimates differed significantly ($P = 0.003$).

Discussion

The MAC for sevoflurane in thick-billed parrots was found to differ depending on the type of stimulation used. Determination of MAC via electrical stimulation yielded a significantly higher MAC than that determined via mechanical stimulation, independent of the method used to estimate MAC. Although the 95% CI and 90% fiducial limit were fairly large for both values, these intervals did not overlap; therefore, we are confident that the MACs for the different types of stimulation truly differ. Although the sample size of 15 birds was small, according to the up-and-down method of MAC determination that was used,[16] the 4 crossover events for each stimulus were sufficient for accurate estimates of MAC. It would have been ideal to determine the MAC for each individual bird for each stimulus and then calculate the mean of the values; however, we could not justify the duration of anesthesia this would have required, particularly when working with an endangered species and when there are other valid methods for MAC determination.

The finding that different forms of stimulation resulted in different estimates of sevoflurane MAC in the present study was contrary to results reported in the anesthetic literature for other species. Investigators in other studies[5,18] have indicated the stability of MAC for a given inhalation anesthetic, regardless of the form of supramaximal stimulation used. In 1 study[5] conducted to evaluate the MAC of halothane in dogs, placing a clamp on the tail and providing electrical stimulation both yielded extremely similar estimates of MAC. In another study,[14] the MACs of halothane and isoflurane were determined in dogs and rabbits via tail clamping and electrical stimulation. It was found that these 2 types of stimulation resulted in a similar MAC for each anesthetic within each species.[14] In contrast, the thick-billed parrots in the present study required a higher concentration of sevoflurane to prevent movement after electrical stimulation than after mechanical stimulation. Other studies[14,19,20] have revealed that the same stimulus can result in different estimates of MAC when applied to different parts of the body.

A particular concern for all evaluations of MAC is the variability in methods within and between studies.[14,18] According to 1 report,[18] estimates of MAC may vary between 10% and 20% within a species. However, the thick-billed parrots in the study reported here had an approximately 2-fold difference in the estimated sevoflurane MACs. To the authors' knowledge, there have been no other comparisons of electrical and mechanical stimulation performed in any avian species. A study[6] on use of isoflurane in red-tailed hawks (*Buteo jamaicensis*) involved similar electrical stimulation (50 V, 50 Hz, and 7.5 milliseconds) and revealed that the isoflurane MAC was slightly higher than that reported in other studies of birds in which investigators used mechanical stimulation but did not directly compare the 2 types of stimulation. The authors of that study[6] hypothesized that the difference in the type of supramaximal stimulus may have contributed to the difference in avian isoflurane MAC estimates.

Another potential source of MAC variability within the present study could have been the difference in the number of male and female birds evaluated. In the present study, we did not evaluate a large enough number of birds to draw conclusions on the effect of sex; however, in mammalian species, it is believed that sex does not influence MAC.[18,21]

Although the different values of MAC for sevoflurane in the thick-billed parrot species are interesting, the indication for this species in particular and for all avian species in general is currently unclear. There are many possible reasons for differences in the estimated MACs. It is possible birds are more sensitive to electrical stimulation than are mammals, the locations of stimulation (toe and medial thigh) may differ in sensitivity, or this particular group of thick-billed parrots was more responsive than other thick-billed parrots to electrical stimulation. The data raise questions with re-

gard to whether toe clamping is a submaximal stimulus and whether electrical stimulation is a supramaximal stimulus in thick-billed parrots.

Another study[12] conducted to evaluate sevoflurane in chickens used toe clamping as the method of stimulation, and the estimated mean ± SD MAC was 2.21 ± 0.32%. The toe-clamp technique yielded a similar MAC of 2.35% in the thick-billed parrots of the present study. This is also comparable to reported mean mammalian sevoflurane MACs of 2.31 ± 0.11% in horses,[22] 2.36 ± 0.46% in dogs,[23] 2.29 ± 0.14% in llamas,[24] 2.33 ± 0.09% in alpacas,[24] and 2.40 ± 0.05% in rats.[25] The reptilian species in which the mean ± SD MAC of sevoflurane has been reported (2.51 ± 0.46% in Dumeril monitors[26] and 3.1 ± 1.0% in green iguanas[27]) have MACs within the range of values for mammalian species. In these studies,[22,24,26,27] the MACs were determined by mechanical stimulation (usually tail clamping) or electrical stimulation. Despite the variability in stimulation techniques, the reported MACs were fairly consistent among species. This information further complicates interpretation of the markedly higher sevoflurane MAC of thick-billed parrots when electrical stimulation was used in the present study.

In 1 study,[7] thick-billed parrots were found to have a lower isoflurane MAC than that for other avian species. Although it is thought that an individual's sensitivity to an inhalation anesthetic should be consistent across a variety of anesthetic agents,[13] this did not appear to be the case for thick-billed parrots, which required a higher concentration of sevoflurane than that reported in chickens.[12] However, this comparison should be made with great care because there can be substantial variability among studies and only limited numbers of birds were evaluated in that study[12] and the study reported here. Further studies to determine the MAC of sevoflurane in other avian species would help clarify this issue.

Heart rate and respiratory rate both decreased with increasing total anesthetic time and with increases in sevoflurane concentration. Numerous studies[7,8,11,28,29] have found that inhalation anesthetics, including sevoflurane, can cause dose-dependent respiratory depression in birds and other species. The effects of time and sevoflurane concentration on avian heart rate are less consistent, with 1 study[9] indicating no change in heart rate and others[8,12,28,29] indicating increased heart rates in birds anesthetized with sevoflurane. However, in 2 studies,[28,29] heart rate did not change significantly when ventilation was controlled. The decrease in heart rate with time that was detected in the present study could have been attributable in part to an initial tachycardia due to a stress response with subsequent return of heart rate to within the reference range with time. This progression has also been reported.[8] Although there were significant changes in heart rate and respiratory rate in the present study, the changes did not appear to be clinically relevant.

The observed increase in blood glucose concentration with time was likely a result of a stress response to handling and anesthetic induction. The significant decrease in chloride concentration cannot be explained but does not appear to be clinically relevant because all values were still within the reported reference interval. The increasing P_{CO_2} and decreasing blood pH were likely a reflection of decreased ventilation in anesthetized birds, despite the application of controlled mechanical ventilation. Similar to the results for the chloride concentration, changes in P_{CO_2} and pH did not appear to be clinically important during the short duration of anesthesia. However, these patterns could certainly become clinically important under different circumstances.

Overall, the P_{CO_2} values obtained in the present study were higher at both time points than were values reported in a previous study[7] of thick-billed parrots in which investigators used isoflurane, yet they were still considered within an acceptable range. The degree of ventilation, unless extreme, is not known to affect MAC.[18,30] All other physiologic values were relatively similar between that other study[7] and the study reported here.

To our knowledge, the study reported here was the first in which the MAC of sevoflurane was determined in a parrot species and different types of supramaximal stimulation were compared in birds. This information is valuable for clinicians who use sevoflurane to anesthetize parrot species. The comparison of mechanical and electrical stimulation in the present study highlights the need for additional research on the validity of various supramaximal stimuli in birds.

a. SevoFlo, Abbott Laboratories, North Chicago, Ill.
b. Sigma vaporizer, Penlon Inc, Minnetonka, Minn.
c. Sheridan uncuffed endotracheal tube, Hudson RCI, Research Triangle Park, NC.
d. 3.5F tom cat catheter, Tyco Healthcare Group LP, Mansfield, Mass.
e. Bird Mark 7, VIASY Siasys Healthcare, Palm Springs, Calif.
f. Raman spectrometer Rascal II, Ohmeda, Salt Lake City, Utah.
g. Physiograph, Gould Instruments Systems, Valley View, Ohio.
h. Ultrasonic Doppler flow detector, Parks Medical Electronics Inc, Aloha, Ore.
i. Bair Hugger, model 505, Arizant Healthcare Inc, Eden Prairie, Minn.
j. K Aquamatic module, Hamilton Industries, Cincinnati, Ohio.
k. ABL800 Flex automated benchtop analyzer, Radiometer Medical ApS, Brønshøj, Denmark.
l. Medical gas analyzer LB1, Beckman Instruments, Schiller Park, Ill.
m. Grass S88 stimulator, Grass Instruments, Quincy, Mass.
n. Recombitek, Merial Inc, Athens, Ga.
o. Normosol-R, Hospira Inc, Lake Forest, Ill.
p. SPSS, IBM SPSS Statistics, Chicago, Ill.
q. SAS/STAT, version 9.3, SAS Institute Inc, Cary, NC.

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