Journal of the American Association for Laboratory Animal Science
Vol 64, No 3
May 2025
Pages 446–456

Premedication Strategies in Turkeys (*Meleagris gallopavo*) Utilizing Either Intramuscular Midazolam or Midazolam/ Butorphanol Compared to Saline Control

Ryan P Buhrer, DVM,[1,*] Loni A Taylor, DVM, MPH, PhD,[2] Robert E Rose, DVM, MS, DACLAM,[3] Jonathan F Bova, DVM, MS, DACLAM,[3] and Tracy H Vemulapalli, DVM, MS, DACLAM[4]

Avian anesthesia in translational medicine must account for various species, yet there is limited research specifically on turkeys used in scientific studies. Premedication is commonly used in veterinary medicine to produce smoother anesthetic events for patients and staff. This study assessed premedication of turkeys using intramuscular midazolam (MDZ) or midazolam/butorphanol (MDZ-B) compared with saline control to improve handling/patient behaviors. Female turkeys (*n* = 35) undergoing a surgical procedure for an orthopedic study were randomly divided into 3 groups (midazolam: 2 mg/kg IM; midazolam: 2 mg/kg IM and butorphanol: 1 mg/kg IM; or saline control: 0.45 mL/kg). Blinded raters qualitatively scored perianesthetic criteria at approximately 10 min post premedication (body position, restraint required, and muscle relaxation), following induction (intubation attempts/ease and apnea), and at recovery (restraint required, tremors, and wing flapping). Relevant time points tracked quantitative data including time of premedication, induction, intubation, inhalant anesthesia end time, and recovery. Premedication with either MDZ or MDZ-B improved perianesthetic parameters such as achieving sternal recumbency, lessening the restraint required, and improving muscle relaxation when compared with saline control. Use of MDZ or MDZ-B significantly decreased the time from premedication to intubation compared with control. The saline control group had significantly faster recoveries than the premedication groups. Choosing an appropriate anesthetic premedication protocol involves considering many factors. Turkeys premedicated with either MDZ or MDZ-B had easier induction leading to improved animal handling and restraint by staff. Due to significantly longer recovery times, MDZ or MDZ-B may increase the periprocedural labor required yet offer a smoother, less traumatic recovery.

Abbreviations and Acronyms: IPPV, intermittent positive pressure ventilation; MDZ, midazolam; MDZ-B, midazolam/butorphanol combination; PMED, birds receiving either MDZ or MDZ-B, analyzed as a single "premedication group"; PS-Int, time range for injection with premedication/saline to intubation; PS-Iso, time range for injection with premedication/saline to isoflurane administration

DOI: 10.30802/AALAS-JAALAS-24-129

Introduction

Anesthesia plays a crucial role in biomedical research involving surgical procedures. Most common laboratory animal species (e.g., mice, dogs, and nonhuman primates) have a variety of validated anesthetic protocols to choose from. While substantial research exists on various drug combinations in avian species, there is a specific lack of studies on premedication strategies for domestic poultry and, in particular, turkeys.

Domestic turkeys (*Meleagris gallopavo*) and chickens (*Gallus domesticus*) are excellent models for studying the tendons of the human hand due to their anatomic similarities.[1] Model development in less commonly used species necessitates the development of premedication strategies to facilitate handling and lower potential stress during induction. A frequent practice in avian medicine is to induce anesthesia via delivery of the anesthetic inhalant by face mask (i.e., "mask induction").[2] Mask induction, without premedication, has the potential to cause significant stress and risk of injury to both the patient and the staff due to the large size and strength of turkeys.[3–5] Periprocedural sedation provided by premedication drugs benefits the patient by alleviating handling- and restraint-induced stress.[6]

Route of administration is one factor to consider in alleviating the handling-associated stress of premedication. Drug delivery options include intranasal, oral, intramuscular, and intravenous. Due to the large size of the pectoral musculature in poultry species, these muscles are a commonly used site for premedication drug administration; thus, the choice of intramuscular administration was used in the present study.[2,6]

Premedication with drugs such as midazolam or butorphanol, used alone or in various combinations, is commonly

Submitted: 09 Nov 2024. Revision requested: 10 Dec 2024. Accepted: 06 Mar 2025.
[1]Department of Veterinary Pathobiology, College of Veterinary Medicine and Biomedical Sciences, Texas A&M University, College Station, Texas; [2]Department of Epidemiology and Biostatistics. School of Medicine, Texas A&M University, College Station, Texas; [3]Comparative Medicine Program, Department of Research, Texas A&M University, College Station, Texas; and [4]Departments of Small Animal Clinical Sciences and Veterinary Pathobiology (joint appointment), College of Veterinary Medicine and Biomedical Sciences, Texas A&M University, College Station, Texas
**Corresponding author. Email: rpbuhrer@tamu.edu*
This article contains supplemental materials online.

administered to avian patients to reduce handling stress and promote smooth anesthetic induction.[6–8] Currently, pharmacokinetic data for midazolam and butorphanol are limited within our common poultry species, such that dosing must be extrapolated from other avian species' pharmacokinetic data. While a recent review article[6] compiled numerous published studies evaluating the effect of midazolam or butorphanol on birds (primarily psittacine species and some poultry), none of the studies included in the review investigated their effects on turkeys.

Midazolam (MDZ) is classified as a benzodiazepine class drug. MDZ acts in the central nervous system on the γ-aminobutyric acid type A ($GABA_A$) receptors, resulting in rapid sedation, muscle relaxation, and anxiolysis.[2,6,9] Benzodiazepine drugs do not typically produce significant negative effects on the cardiovascular and respiratory systems. In addition to its beneficial sedative effects, midazolam provides a reduction in the minimum alveolar concentration.[2,10] Another benefit of MDZ is that it is water soluble, allowing it to be readily absorbed by the body when administered intramuscularly.[11] In a recent study[12] examining MDZ as a potential marker for cytochrome P450 3A activity, researchers determined the half-life of elimination for MDZ when administered intravenously in turkeys ($t_{1/2}$ = 25.2 min) and found this to be comparable to that reported in mammalian species.

Butorphanol, another premedication drug commonly used in avian patients, is a partial κ agonist and μ antagonist opioid. Opioid agents have potential anesthetic and analgesic benefits in avian species.[8] A study[13] in pigeons (*Columba* spp.) found that the predominant opioid receptor type in the forebrain was composed of approximately 76% κ receptors. This finding suggests that μ opioid agonists may not provide adequate pain control in many bird species.[14–16] Studies[15,17,18] in poultry have assessed the analgesic effects of butorphanol; the authors were unable to find studies that looked directly at the sedative effects of butorphanol as a single agent in poultry, specifically in turkeys. Studies[16,19] investigating the pharmacokinetics of butorphanol found that the terminal half-life when given intravenously in broiler chickens was 71.3 min, and when given intramuscularly in red-tailed hawks (*Buteo jamaicensis*) and great horned owls (*Bubo virginianus*) was 56.4 and 110.4 min, respectively. In a pharmacokinetic study involving Hispaniolan Amazon parrots (*Amazona ventralis*), dosing butorphanol every 2 to 3 h was required to maintain therapeutic levels.[20] Like midazolam, butorphanol has been shown to reduce minimum alveolar concentration levels in cockatoos (*Cacatua* spp.) and African gray parrots (*Psittacus erithacus*).[20,21] Butorphanol, when combined with midazolam (MDZ-B), was shown to produce moderate sedation in pigeons.[22] MDZ-B has also been shown to produce analgesia and more profound sedation with limited adverse effects in cockatiels and other psittacines.[2]

In the present study, we aimed to assess the effect of intramuscular premedication of turkeys with either MDZ or MDZ-B to improve periprocedural handling and patient behaviors at time points including postpremedication, induction/intubation, and during recovery when compared with birds receiving no premedication drugs (i.e., saline controls). We hypothesized that the use of premedication drugs would result in significant differences in the time to successful intubation as well as recovery times when compared with our control group. A secondary aim was to create and evaluate a scoring system used in assessing perianesthetic behaviors. We predicted that birds premedicated with either MDZ or MDZ-B would exhibit improved perianesthetic scores compared with control birds, resulting in smoother anesthetic induction and recovery from the surgical procedure. We also hypothesized that the scoring system would perform comparably well despite the experience level of the raters.

Methods

Animals. Thirty-six female broad-breasted white turkeys (*M. gallopavo*) were acquired from the Texas A&M University (TAMU) Poultry Science Center (College Station, TX) for a study evaluating flexor tendon repair and were used in this premedication study. For practical reasons, 18 surgeries were performed each week (6 surgeries per day for 3 d) for 2 consecutive weeks, when the turkeys were 12 and 13 wk old, respectively. The turkeys arrived from the center in 2 groups of 18 approximately 1 week before their scheduled surgery for simultaneous quarantine and acclimation. The study was approved by the TAMU IACUC, and the turkeys were housed in an AAALAC-accredited facility. The turkeys were group-housed in 6 ft × 8 ft (1.8 m × 2.4 m) kennel runs in groups of 6, divided into 3 groups per room, occupying 2 different rooms. They were housed on wood shavings (Producers Cooperative Association; Tractor Supply Premium Pine Shavings) and hay (Producers Cooperative Association) with free access to feed (Producers Cooperative Show Broilers Finish) and city water. The room set point for temperature was 71 °F and remained within the recommended range for poultry (61 to 81 °F) as outlined in the *Guide for the Care and Use of Laboratory Animals.* Humidity stayed within the 30% to 70% accepted range. The room had a 12:12 light-dark cycle. Six kennels were present per room so that groups could be moved to a clean run every 2 wk. Spot cleaning was performed daily with fresh hay and wood shavings added as needed. All groups were housed on the same side of the room, with barriers between adjacent groups to help prevent disruptions in hierarchical dominance between groups. Turkeys were assessed for wounds or other obvious issues at the initiation of the quarantine/acclimation period. No screening for pathogens was performed before the study. As they were placed in the kennels, those birds undergoing surgery during week 1 were assigned identification nos. 1 to 18; those undergoing surgery during week 2 were assigned nos. 19 to 36. Birds 1 to 18 were then randomly divided into kennels 1 to 3. Birds 19 to 36 were randomly divided into kennels 4 to 6. Kennel numbers corresponded to the surgical day, with turkeys in kennels 1 to 3 being assigned to week 1 and kennels 4 to 6 being week 2 surgeries. Turkey no. 22 was removed from the study on the day of surgery due to a swollen hock and, thus, was not an ideal candidate for the flexor tendon repair study. Turkey no. 30 was removed after a piece of hay was found partially obstructing the trachea. For the animal's clinical welfare, the veterinarians removed the hay, which caused a 4-min delay unrelated to aspects of this current study. Due to this delay, time points were affected, and there is potential that due to the time spent off isoflurane, other variables of interest could have been affected, which led to the author's election to remove the animal from the study analysis.

Perianesthetic scoring. To assess the perianesthetic scoring metric between raters of different experience levels, 2 blinded raters were used who both independently scored every bird used in this experiment. Rater A is a licensed veterinarian completing specialty training in laboratory animal medicine, with experience in avian species. Rater B is a laboratory animal surgical technician with decades of anesthetic experience in multiple species but limited avian experience. Due to the limited number of animals in the study, no formal live animal training was performed on the scoring rubrics before the start of the study. Rater A was responsible for modifying previously used rubrics for use in this study.[4,23] Rater B discussed the rubrics with rater A before the start of the study and was able to ask for clarification about what the score should be if they found the rubric unclear before rating any birds.

To assess the difference associated with premedication administration, turkeys were assessed at 3 different time points during the experiment with different criteria scored during those events by only rater A (Tables 1 and 2). Turkeys were assessed 10 min after premedication (or control) intramuscular injection, with body position/sedation quality scored. Immediately after scoring body position, the birds were removed from the transport crate by rater A, and the remaining premedication criteria, restraint required and muscle relaxation, were scored based on behaviors at the time of removal from the transport crate (Table 1, Premedication). Turkeys were scored immediately after intubation, and the criteria evaluated were as follows: intubation ease and presence of apnea requiring intermittent positive pressure ventilation (IPPV; Table 2). The following recovery criteria were scored for behaviors spanning the entire recovery period spanning from the end of isoflurane administration to recovery completion (ability to maintain sternal recumbency unassisted): restraint required, muscle tremors, and wing flapping (Table 1, Recovery). Standing was not used as a 'recovery complete' criterion due to the potential for casting of the surgical limb to affect coordination. In cases where birds' recovery skipped a period of sternal recumbency and they were able to stand almost immediately, recovery was marked complete at the time of standing.

Premedication. Animals were randomly assigned to 1 of 3 experimental groups. The control group ($n = 10$), received 0.45 mL/kg saline IM (Saline Solution 0.9%; Clipper Distributing Company). Experimental group one ($n = 12$) received 2 mg/kg midazolam IM (Midazolam Injection, USP; Akorn). The second experimental group ($n = 12$) received 2 mg/kg Midazolam IM mixed with 1 mg/kg butorphanol IM (Torbugesic; Zoetis). Turkeys were weighed 24 to 48 h. presurgery for premedication drug calculations. Before receiving their assigned premedication protocol, birds were separated from their pen mates and wheeled to the prep room in a transport kennel. Turkeys were held and restrained by rater A for all birds. They were restrained using a forward-facing modified "football hold," keeping wings tucked into each side with additional restraint of the feet by gently grasping the lower limbs in the opposite hand. Rater B performed the intramuscular injection with the premedication or saline. All intramuscular injections were given in the right pectoral muscles, divided evenly between 2 sites that were at least 1 in. apart. Personnel administering premedication drugs or saline control were blinded to the contents of the syringe and each of the birds' weights. The syringe was covered with tape to prevent observing the volume in the syringe; it could be argued that the syringe plunger position would indicate premedication volume and could have implied a particular treatment group (i.e., MDZ-B, saline, MDZ: least to greatest total volume). However, the variable weights of the turkeys offset this effect by similarly causing variation in syringe position; thus, the person administering the drug was effectively kept blinded to treatment. Following intramuscular injection, the birds were returned to the transport crate, and premedication was scored as described in perianesthetic scoring.

Induction/intubation. Turkeys were restrained in the same manner as during premedication/saline control injection, with the birds resting on a table for support as they lost righting reflex during induction. Rater B gently grasped the turkey's neck to maintain appropriate face mask positioning during induction. The time inhalant administration began was recorded. Turkeys were masked down using 2.5% isoflurane (Isospire; Dechra) in 100% O_2 until there was a lack of movement, sternal body

Table 1. Perianesthetic scoring metrics used during premedication and recovery

	Criteria for individual scores				
	1—Very poor	2—Poor	3—Good	4—Very good	5—Excellent
Premedication					
Body position/ sedation	Standing Fully awake (no sedation)	Standing Mild sedation	Standing Moderate sedation	Sternal Moderate sedation (able to right self from lateral recumbency)	Lateral recumbence (unable to right self)
Restraint required	Significant	Moderate	Minimal	No restraint required	No restraint required (plus lateral recumbence)
Muscle relaxation	Poor muscle relaxation Violent wing flapping	Minimal muscle relaxation Mild wing flapping	Minimal muscle relaxation No wing flapping	Muscle relaxation No wing flapping	Good muscle relaxation No wing flapping
Recovery					
Restraint required	Significant manual restraint required	Manual restraint required	No manual restraint required (but not smooth and calm)	No manual restraint needed (smooth and calm recovery)	No manual restraint required (exceptionally smooth and calm)
Muscle tremors	Prolonged tremoring (tremors persist throughout recovery)	Moderate tremors (persist for more than half of recovery)	Mild tremoring (tremors lasted from 1/4 to 1/2 of the recovery)	Slight short-lasting tremors (last less than 1/4 of recovery)	No muscle tremors
Wing flapping	Violent wing flapping Rolling	Moderate wing flapping	Minimal wing flapping	No wing flapping	No wing flapping

Variables were scored at the end of premedication and recovery (see Methods). Scores of 1 represented poor perianesthetic outcomes while scores of 5 represented excellent outcomes.

Table 2. Perianesthetic scoring metrics used during induction

	Score		
	1—Poor	2—Good	3—Excellent
Intubation	Difficult	Minimal difficulties	Easy
	Multiple attempts required	One or more attempts required	Anesthetic depth sufficient for successful intubation in one attempt
	Further masking required	Anesthetic depth complicates but does not prevent intubation	
		Additional masking not required	
Apnea	Prolonged apnea (>30 s) or <2 breaths per min	Mild to moderate apnea (15–30 s) or <4 breaths per minute	Minimal to no apnea (<15 s) or >4 breaths per minute

A score of 1 represents a poor induction outcome, while a score of 3 represents an excellent outcome.

positioning, loss of righting reflex was achieved (when larger muscle groups of body, legs, and wings were relaxed with no leg or wing movement to correct to sternal), and/or birds lost jaw tone. Jaw tone was assessed by removing the head from the face mask when the patient was deemed at a suitable anesthetic depth (based on body position and muscle relaxation). Intubation was attempted if the mouth could be opened adequately to view the tracheal opening. If jaw tone prevented adequate visualization, the birds were returned to mask induction. Mask induction was overseen by rater A, a veterinarian who assessed when the turkey's anesthetic depth was adequate to attempt intubation. Turkeys were intubated in sternal recumbency. Both raters performed intubations and are experienced in intubation of various species. Time of successful intubation and the number of attempts to achieve it were recorded. If the turkey resisted intubation and an attempt was unsuccessful, further masking with inhalant was performed until a deeper state of anesthesia was reached. In the event of 2 unsuccessful intubation attempts, isoflurane was increased to 3%. Induction scoring was performed as described in perianesthetic scoring after successful intubation.

Surgical prep/surgery/postoperation procedures. Once animals were intubated, the appropriate leg was prepped for tendon surgery by performing an alcohol/chlorhexidine scrub. While prepping, oxygen saturation and heart rate levels were monitored via pulse oximetry. Respiratory rate was counted manually by observing chest excursions. IPPV was administered at a rate of one breath per minute, regardless of respiratory rate.[24] A local block of the surgical limb was provided with 0.5 mL bupivacaine (0.25% Bupivacaine Hydrochloride Injection, USP; Hospira). After the preparation of the leg was complete, turkeys were transported into the operating room for the completion of a flexor tendon repair procedure. All turkeys received identical surgical manipulation with the only variation being one of 3 topical treatments (saline, alternate hydrogel sheet, or collagen hydrogel) applied to the tendon repair. The topical treatments have shown no systemic distribution and are only locally active. During surgical preparation and surgery, isoflurane was maintained at 2.5% for all animals unless clinical signs (i.e., decreased respiration rate, decreased heart rate, reacting to stimuli, etc) necessitated changes in the dose of isoflurane administered. Upon surgical completion, the turkeys were wheeled back into the prep room, where they received 1.0 mL meloxicam (Metacam; Boehringer Ingleheim) in the left pectoral muscles. The surgical foot was epoxy resin cast using 1-in. casting tape (3M Scotchcast Plus Casting Tape) as part of the primary investigator's experimental design. Once casting was complete, inhalant anesthesia was discontinued, and the recovery period began.

Recovery. Inhalant anesthesia end time was recorded. On surgery days 1 and 2, 100% O_2 was continued until extubating. Turkeys with procedures on days 3 to 6 were left intubated but returned to room air 5 min after the inhalant was discontinued. Turkeys were monitored during recovery for their eyes opening/blinking, head movement, jaw tone, and leg movement. Extubation was performed once the turkey was able to hold its head up or regain enough jaw tone that the animal could maintain its own airway. Once extubated, birds were moved back to the transport crates and monitored until they could maintain a sternal position and elevate their head unassisted. Once sternal and able to maintain head position, recovery was deemed complete for this study. At that point, the birds were checked on periodically throughout the rest of the day but not continuously monitored. Recovery was scored as described in perianesthetic scoring.

Time points and periods. Time points recorded included the time of premedication/saline injection, inhalant start time, intubation time, inhalant end time, and recovery complete time (Figure 1). From these time points, time periods were calculated. PS-Iso was the time from the premedication/saline injection to the start of isoflurane inhalant anesthesia. Induction time was the period from the start of inhalant anesthesia to the time of successful intubation. PS-Int is the time from injection of premedication/saline to successful intubation. Inhalant time was the time from the start of isoflurane inhalant to the time isoflurane was discontinued. Recovery time was the time from discontinuation of the isoflurane inhalant to recovery complete. Procedural time was the total procedure time from the injection to complete recovery.

Heart rate, respiratory rate, and CO_2. Subsequent to the study, heart rate and respiratory rate were collected from the anesthesia records. Times for respiratory rate and heart rate are given from time of injection with premedication/saline control. CO_2 is presented as millimeters of mercury at the first CO_2 reading and minutes from the time of injection. Values were recorded approximately every 5 min from inhalant start until the turkey was moved into the operating room (~35 min from injection). Raw data time points were adjusted to the nearest 5-min interval with times ending in digits 3 to 7 rounded to the nearest 5 and times ending 0 to 2 or 8 to 9 digits rounded to the nearest 10 (e.g., 13 min were rounded to 15 min and 28 min rounded to 30 min). Time points 0, 5, and 10 min did not have any values since patients were left undisturbed for 10 min following premedication/saline injection. Data from 40 min on were not consistently recorded at 5-min intervals and was thus not present consistently for time point comparisons. CO_2 was only monitored in the operating room and not in the prep/casting room so data for that were limited; thus we selected to

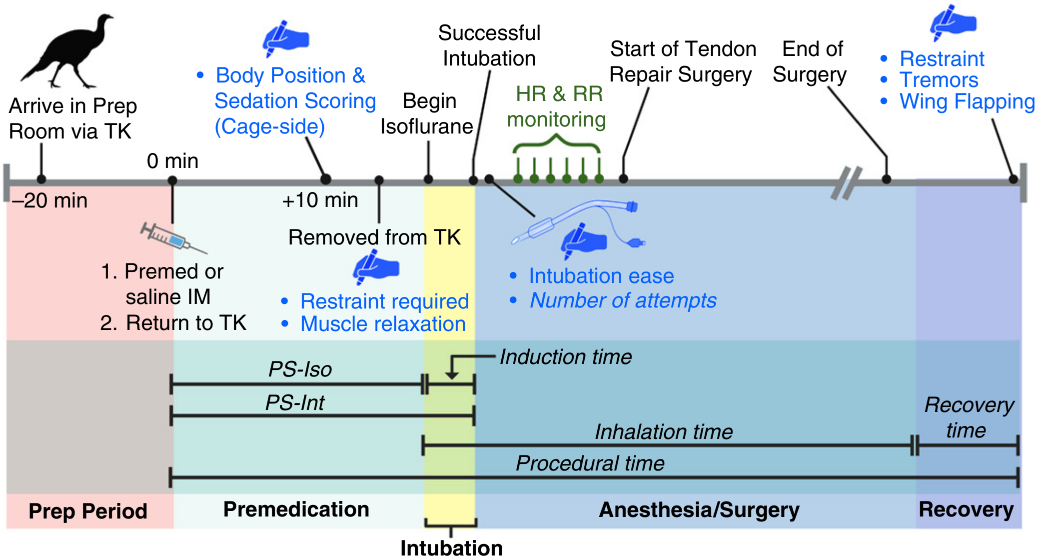

Figure 1. Study timeline for each turkey, from arrival to the prep room and premedication through complete recovery. Approximately 10 to 20 min after arrival, turkeys received a single intramuscular injection of MDZ, MDZ-B, or saline control. They were returned to and left undisturbed in their travel kennels for at least 10 min before initiating the first assessment. Assessment of the turkeys on each of 8 qualitative measures (handwriting icon and blue font) occurred at specified points during the perianesthetic periods (i.e., premedication, induction, and recovery); all quantitative data (italics) was obtained during these same periods. The 6 calculated time periods (black bars) are shown relative to their associated perianesthetic periods (color-shaded vertical columns). HR, heart rate; RR, respiratory rate; TK, travel kennel. This figure was created in BioRender. Vemulapalli, T. (2025) https://BioRender.com/m95n189.

only evaluate the initial CO_2 measured in surgery and the time of that reading. One PMED turkey (no. 10) had no recorded vitals due to a sensor malfunction.

Statistical analysis. Statistical analysis was completed using STATA SE 18.0. The 2 raters' results were compared using the Cohen's κ using all animals. To keep consistency in the analysis, rater A's results were used as the sole outcome variable throughout the premedication analysis.

All ordinal and time variables in the data were first tested for normality using the Shapiro-Wilk test. Of the 8 ordinal variables tested, 4 exhibited a normal distribution: premedication body position, premedication restraint, induction ease, and apnea at induction. The remaining 4 variables were not normally distributed: premedication muscle relaxation, recovery restraint required, recovery tremors, and recovery wing flapping. Of the 6 time variables tested, 3 exhibited a normal distribution: PS-Iso, inhalant time, and procedural time. The remaining 3 were not normally distributed; these were induction, PS-Int, and recovery time. To maintain consistency in reported points of central tendency, all ordinal and time range variables in the data were reported as medians, with the first and third quartiles reported in brackets, and analyzed using the more conservative nonparametric test, Wilcoxon (Mann-Whitney) rank sum test. This approach also helped maintain consistency in analyzing the differences between the MDZ and MDZ-B premedication treatment groups. Due to no statistically significant difference between the 2 premedication treatment groups (MDZ and MDZ-B), the groups were collapsed into a single, overarching premedication group (PMED). PMED was then compared with the saline control group using the same method as described above. A P value of less than 0.05 was considered statistically significant.

Heart rate, respiratory rate, and CO_2 were tested for normality using the Shapiro-Wilk test. Data were normally distributed for respiratory and heart rates at all time intervals. There were multiple missing data points in the heart rate and respiratory rate data, so a 1-way ANOVA with the Bonferroni correction for multiple comparisons was performed for heart rate and respiratory rate at each time interval. The data for the time of the first CO_2 measurement was normal. The CO_2 millimeters of mercury were not normally distributed. The Kruskal-Wallis equality of population statistic was performed, followed by the Dunn pairwise comparison with the Bonferroni correction for both CO_2 measurements to maintain consistency.

Results

Perianesthetic scoring system rater agreement. Agreement between raters was greater than 70% for all perianesthetic score variables. The Cohen's κ values for all scoring variables can be found in Supplementary Table S1.

Comparison of MDZ compared with MDZ-B groups. Turkey weights between the MDZ (median = 9.39 kg) and MDZ-B (median = 9.32 kg) groups showed no statistically significant differences ($P = 0.9892$). A comparison of the MDZ premedication and the MDZ-B revealed no significant differences ($P > 0.05$) for any of the qualitative parameters scored (Table 3) or the

Table 3. Comparison of perianesthetic-related scores for MDZ compared with MDZ-B at specified procedural time points

	MDZ	MDZ-B	P value
Premedication			
Body position	4 [4, 4]	4 [4, 4]	0.4783
Restraint required	4 [2.5, 4]	4 [3.5, 4]	0.4426
Muscle relaxation	4 [2, 4]	4 [3.5, 4]	0.4606
Induction			
Intubation ease	3 [3, 3]	3 [3, 3]	1.0000
Apnea at induction	3 [3, 3]	3 [3, 3]	1.0000
Recovery			
Restraint required	5 [4.5, 5]	5 [5, 5]	0.3416
Tremors	5 [5, 5]	5 [5, 5]	1.0000
Wing flapping	5 [4.5, 5]	5 [5, 5]	0.3416

Data are presented as median [quartile 1, quartile 3]. For Premedication and Recovery, scores ranged 1 to 5, with 1 indicating poor scoring outcomes and 5 indicating excellent scoring outcomes. For Inductions, scores ranged from 1 (poor) to 3 (excellent).

Table 4. Comparison of quantitative data for MDZ compared with MDZ-B at specified procedural time points

Variable	MDZ	MDZ-B	*P* value
PS-Iso (min)	12 [11, 12]	12.5 [11.5, 13]	0.0701
Induction (min)	5.5 [5, 7]	5.5 [3.5, 7]	0.6757
PS-Int (min)	17 [16.5, 19]	17.5 [16, 20.5]	0.6356
Intubation number of attempts	1 [1, 1]	1 [1, 1]	1.0000
Inhalant time (min)	69 [64, 72.5]	71.5 [68, 75.5]	0.3689
Recovery (min)	20 [15, 45]	21.5 [16.5, 29.5]	0.9890
Procedural time (min)	108 [92, 121]	107 [99, 118.5]	0.8317

Data are presented as median [quartile 1, quartile 3].

time ranges recorded (Table 4). No turkey in either treatment group reached lateral recumbency at 10 min postinjection of premedication. Heart rate and respiratory rate showed no significant differences ($P > 0.05$) between MDZ and MDZ-B at all time points evaluated. CO_2 data also had no significant differences (Table 5). Based on the lack of statistically significant differences between the 2 individual premedication groups (MDZ and MDZ-B), these data were collapsed into a single premedication group (PMED) that was then compared with the saline control group.

Comparison of premedication compared with saline. The results discussed in this section and subsequent sections unless otherwise specified are for the comparison between PMED and saline control. Turkey weights between PMED (median = 9.39 kg) and saline control (median = 9.48 kg) groups showed no statistically significant differences ($P = 0.9492$).

Comparison of premedication scores. The results in this, and all subsequent, sections are presented as (group = median [quartile 1, quartile 3] and group = median [quartile 1, quartile 3], *P* value) unless otherwise stated (Table 6). PMED birds scored significantly better than saline control birds for the qualitative parameters of premedication body position (PMED = 4 [4, 4]; saline = 1 [1, 1]; $P < 0.0001$), premedication required restraint (PMED = 4 [3, 4]; saline = 2 [1, 2]; $P < 0.0001$), and premedication muscle relaxation (PMED = 3 [3, 4]; saline = 1.5 [1,2]; $P < 0.0001$). No turkey in either treatment group reached lateral recumbency at 10 min postinjection of premedication.

Comparison of anesthetic induction/intubation. PMED turkeys had significantly shorter median inductions (PMED = 5.5 min [4.5, 7]; saline = 7.5 min [6, 17]; $P = 0.0388$) and PS-Int (PMED = 17 min [16.5, 19]; saline = 20 min [18, 28]; $P = 0.0139$) (Tables 6 and 7; Figure 2). However, there was no significant difference found in the number of intubation attempts. When assessing qualitative perianesthetic parameters, there was no statistical significance with respect to intubation ease or presence of apnea between PMED and saline control. Two of the 10 saline control birds required 3 and 4 intubation attempts, respectively, and Isoflurane was increased to 3% as described in Methods. One of the 24 PMED birds required 2 intubation attempts. One bird receiving PMED (MDZ-B) had an apnea score of 1 (i.e., >30 s) during the induction period. Isoflurane was reduced to 1.5% and IPPV was administered following intubation. Based on anesthesia records, it required IPPV at 4 bpm for approximately 10 min until spontaneous breathing returned to a rate >4 bpm. Heart rates recorded during this period of IPPV were 145 and 141 bpm. CO_2 was not recorded during this time as it was only recorded in the operating room and the apnea occurred in the prep room.

Comparison of anesthetic recovery. PMED had longer recovery times (PMED = 21.5 min [16, 38], saline = 7 min [6, 8]; $P < 0.0001$) compared to control birds. Procedural time was longer for premedicated birds (PMED = 107 min [94.5, 120.5]; saline = 90 min [86, 96]; $P = 0.0015$) (Tables 6 and 7; Figure 3). Inhalant time was not statistically different between groups (PMED = 70 min [67, 74.5]; saline = 70 min [68, 77]; $P = 0.9036$). Recovery scores for required restraint and wing flapping were qualitatively better in premedicated birds than for control birds (restraint: PMED = 5 [5, 5], saline = 3 [2, 5]; $P = 0.0031$; wing flapping: PMED = 5 [5, 5]; saline = 3 [2,5]; $P = 0.0081$).

Comparison of vital parameters (heart rate, respiratory rate, and CO_2). Respiratory rate and heart rate are presented as median ± SD. CO_2 data are presented as median [IQR] (Table 8). The respiratory rate was not significantly different between PMED and saline control groups for any time point analyzed. Heart rate was significantly higher in the PMED group at the 20- and 25-min time points. Heart rates at the 20-min time point

Table 5. Measure vital parameters during anesthesia at specific time points for MDZ compared with MDZ-B

	Midazolam	Midazolam-butorphanol	*P* value
Respiratory rate (breaths/min)			
15 min	14.29 ± 5.09 (*n* = 7)	16.50 ± 5.74 (*n* = 4)	0.5231
20 min	11.29 ± 4.42 (*n* = 7)	13.00 ± 5.02 (*n* = 6)	0.5259
25 min	11.55 ± 3.57 (*n* = 9)	10.50 ± 3.66 (*n* = 10)	0.5341
30 min	10.89 ± 4.11 (*n* = 9)	11.40 ± 3.13 (*n* = 5)	0.8139
35 min	10.43 ± 2.82 (*n* = 7)	10.14 ± 3.48 (*n* = 7)	0.8689
Heart rate (beats/min)			
15 min	162.75 ± 39.55 (*n* = 8)	185.5 ± 25.52 (*n* = 6)	0.2443
20 min	195.40 ± 21.77 (*n* = 10)	191.20 ± 26.64 (*n* = 10)	0.7040
25 min	197.00 ± 30.06 (*n* = 9)	196.00 ± 25.25 (*n* = 11)	0.9364
30 min	196.60 ± 30.16 (*n* = 10)	194.00 ± 30.26(*n* = 8)	0.8583
35 min	179.17 ± 33.49 (*n* = 6)	184.75 ± 25.45 (*n* = 8)	0.7283
CO_2			
Minutes from injection	37 [32.5, 38.5] (*n* = 12)	39 [38, 45] (*n* = 11)	0.0603
Millimeters of mercury	75.5 [68, 82] (*n* = 12)	69 [67, 78] (*n* = 11)	0.4570

Times for respiratory rate and heart rate are given from injection with premedication. Heart rate and respiratory rate are presented as mean ± SD (sample size). CO_2 is presented as the median millimeters of mercury at the first CO_2 reading and median minutes from premedication of the first reading. CO_2 data are presented as median [quartile 1, quartile 3] (sample size at that time point).

Table 6. Comparison of scores for saline control compared with PMED at specified procedural time points

	Saline control	PMED group	*P* value
Premedication			
Body position	1 [1, 1]	4 [4, 4]	<0.0001‡
Restraint required	2 [1, 2]	4 [3, 4]	<0.0001‡
Muscle relaxation	1.5 [1,2]	4 [3, 4]	<0.0001‡
Intubation			
Intubation ease	3 [3, 3]	3 [3, 3]	0.1604
Apnea during induction	3 [3, 3]	3 [3, 3]	1.0000
Recovery			
Restraint required	3 [2, 5]	5 [5, 5]	0.0031†
Tremors	5 [5, 5]	5 [5, 5]	0.6765
Wing flapping	3 [2, 5]	5 [5, 5]	0.0081†

Data are presented as median [quartile 1, quartile 3]. For Premedication and Recovery, scores ranged from 1 to 5, with 1 indicating poor scoring outcomes and 5 indicating excellent scoring outcomes. For inductions, scores ranged from 1 (poor) to 3 (excellent).
†, $P \leq 0.01$; ‡, $P \leq 0.0001$.

for PMED and saline were 193.3 ± 23.78 bpm and 158.75 ± 18.16 bpm, respectively, with a *P* value of 0.0011. Heart rates at the 25-min time point for PMED and saline were 196.45 ± 26.77 and 172.63 ± 27.13 bpm, respectively, with a *P* value of 0.0437. The median time of the first recorded CO_2 and the CO_2 millimeters of mercury at the first recorded value were not significantly different between groups.

Discussion

Domestic turkeys, while not commonly used in research, are still an important animal model and therefore require the exploration and refinement of premedication and anesthetic protocols to improve handling and promote smooth recovery following surgery and other potentially painful procedures. The current study demonstrated that MDZ premedication, with or without butorphanol, led to improved qualitative scores during the premedication and recovery periods for almost all parameters measured. The only parameter showing no improvement was muscle tremors where median scores in both groups corresponded to excellent, which clinically represents no muscle tremors. The higher qualitative scores in PMED birds indicate that premedication significantly decreased several behaviors associated with alert and potentially stressed birds (i.e., struggling as examined via the amount of restraint required, muscle relaxation, and wing flapping) and thus likely aided in the smoother induction and recovery periods seen in these birds as compared with those receiving no premedication (saline control). Similar results were found in a study of restrained cockatiels (*Nymphicus hollandicus*), which showed that MDZ or MDZ-B led to less struggling and lowered respiratory rates as compared with saline controls.[25]

The comparison of MDZ and MDZ-B found no statistically significant differences. Given the lack of statistically significant differences, the dose of butorphanol may not have been adequate to produce noticeable sedative effects in the turkeys in this study. Further studies could investigate a higher dose of butorphanol to determine if that could improve the quality of sedation in a multimodal anesthetic approach. Due to the lack of difference in premedication groups, the authors considered combining MDZ and MDZ-B into 1 group (PMED), given the potential that butorphanol had a limited sedative effect based on the analysis of MDZ compared with MDZ-B.

Periprocedural use of sedation often decreases the need for heavy restraint as well as the likelihood of stress-induced injurious behaviors by the animal.[2,5,6,18,26] Following premedication, birds in the PMED group had identical median scores across 3 qualitative measures that corresponded to a clinical picture of sternal recumbency, required no restraint, and no wing flapping observed. Conversely, saline control birds had median scores that varied among those same 3 measures, which clinically presented as standing, fully awake birds that required restraint and were wing flapping. Due to the anxiolytic and sedative effects of midazolam and sedative effects of butorphanol, this result for the PMED group is expected when compared with birds receiving no premedication.[2,6,8,9] The improved scores seen during premedication resulted in easier animal handling of the turkeys by ensuring that birds were more tractable and thereby facilitated smoother mask induction compared with the control turkeys. A similar study[27] involving intramuscular MDZ-B premedication in hospitalized great white pelicans (*Pelecanus onocrotalus*) found that these patients were less aggressive, permitted handling, and enabled physical examination without the need for restraint. One notable finding is that, in the current study, no PMED turkeys achieved lateral recumbency by the 10-min post-premedication time point. Achieving lateral recumbency in premedicated turkeys was not a necessary outcome as all PMED birds received good/excellent scores without reaching lateral recumbency. A higher dose or a more extended waiting period may be necessary for procedures requiring deeper sedation where a loss of consciousness is desired.

Mask induction with isoflurane (used in this study) and other fluorinated hydrocarbon gas anesthetics is known to be highly aversive to many animal species, including people, most likely due to their pungent odors.[3,4,28–33] The aversive effect of these

Table 7. Comparison of quantitative data for saline control compared with PMED at specified procedural ranges

Variable	Saline control	Premedication group	*P* value
PS-Iso (min)	12.5 [11, 14]	12 [11, 13]	0.3706
Induction (min)	7.5 [6, 17]	5.5 [4.5, 7]	0.0388*
PS-Int (min)	20 [18, 28]	17 [16.5, 19]	0.0139*
Intubation number of attempts	1 [1, 1]	1 [1, 1]	0.1604
Inhalant time (min)	70 [68, 77]	70 [67, 74.5]	0.9036
Recovery time (min)	7 [6, 8]	21.5 [16, 38]	<0.0001‡
Procedural time (min)	90 [86, 96]	107 [94.5, 120.5]	0.0015†

Data are presented as median [quartile 1, quartile 3].
*, $P \leq 0.05$; †, $P \leq 0.01$; ‡, $P \leq 0.0001$.

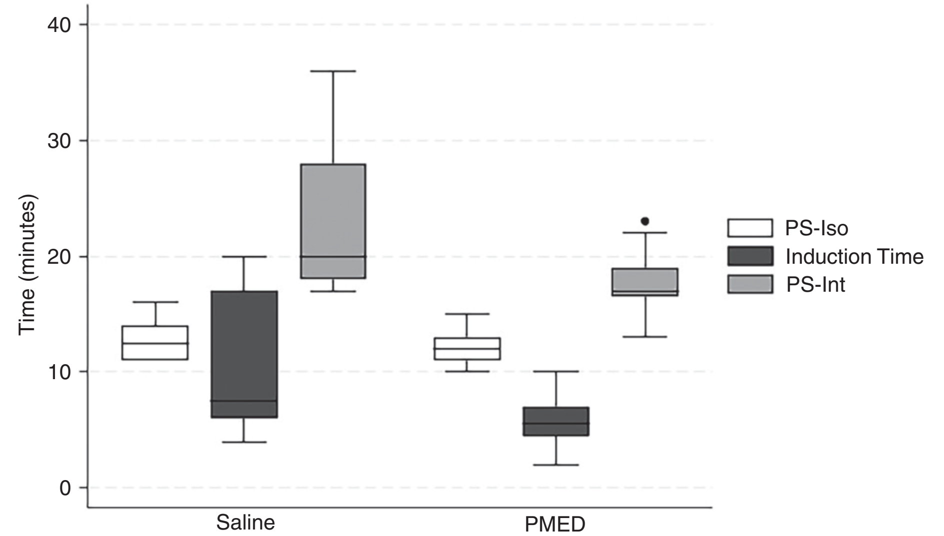

Figure 2. Premedication and induction inclusive time ranges (PS-Inh, Induction, and PS-Int) comparing the PMED group (receiving premedication) to the saline control group. The dot above PMED PS-Int represents an outlier in the data.

gases can result in significant resistance to mask induction often resulting in a longer time for induction and possibly necessitating the use of a higher percentage of the anesthetic mixed with O_2.[3,29–32] In the present study, the PMED group saw reduced times for induction as well as time from premedication to intubation (PS-Int) when compared with the saline control group. This would be expected since, when using multimodal anesthesia, the premedication would provide additional sedation, which, in most individuals, should lower the inhalant required to induce anesthetic depth sufficient for intubation. The median number of intubation attempts and the median scores for intubation ease and apnea during induction were identical for both PMED and saline control groups. Comparable intubation attempts and intubation ease scores between the PMED and control birds are to be expected, given that anesthetic depth was assessed before attempting intubation and the same parameters were checked before attempting intubation. Of note, 1 bird (no. 20) receiving midazolam/butorphanol premedication experienced brief apnea during induction, but the cause could not be determined. While the exact cause of the apnea in our study could not be determined, chickens experience respiratory depression from inhalant anesthetics comparably to their mammalian counterparts.[34] For example, a study[35] in New Zealand White rabbits found that mask induction with either isoflurane or sevoflurane anesthetics led to periods of apnea during induction. For turkey no. 20, we also cannot rule out a potential synergistic interaction between MDZ-B and isoflurane resulting in oversedation and subsequent apnea.

Median inhalant time was not significantly different between the PMED and control groups, which implies that isoflurane had a negligible effect on the differences observed during recovery between the 2 groups and that these differences could be attributed to the presence or absence of premedication. Recovery time, as well as procedural time, was significantly longer in the PMED group than the saline control group. Although the recovery time was lengthened, in the authors' opinion, this extended recovery period was not excessively long and, when matched with the improved recovery parameters, led to an overall improved recovery compared with saline control birds. Recovery parameters, including the variables restraint required

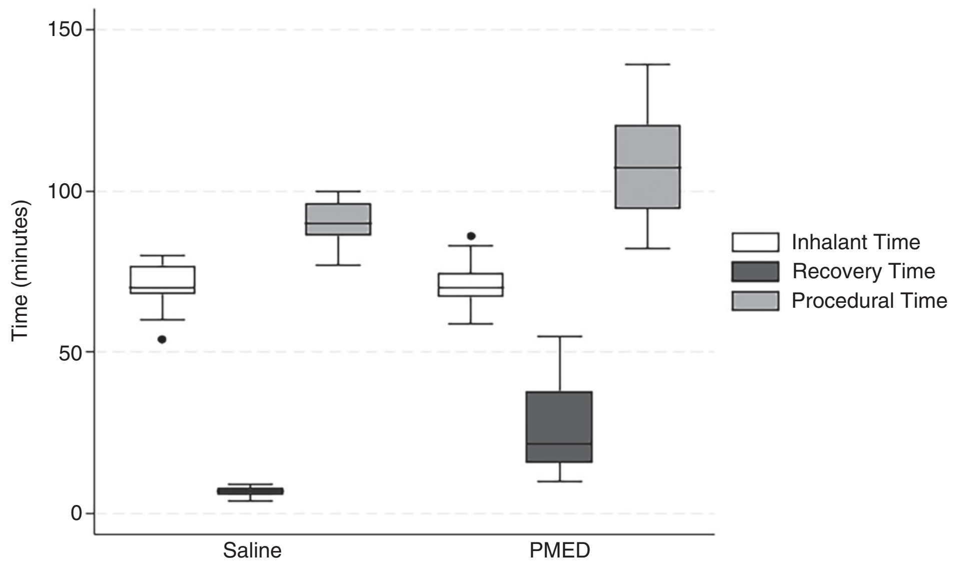

Figure 3. Inhalant, recovery, and procedural time compared between the PMED group (receiving premedication) to the saline control group. The dots below/above Inhalant Time for both Saline and PMED represent outliers.

Table 8. Measure vital parameters during anesthesia at specific time points for saline control compared with PMED

	Saline control	PMED	*P* value
Respiratory rate (breaths/min)			
15 min	10.2 ± 1.79 (*n* = 5)	15.09 ± 5.17 (*n* = 11)	0.0620
20 min	11 ± 4.65 (*n* = 6)	12.08 ± 4.59 (*n* = 13)	0.6418
25 min	9.63 ± 2.92 (*n* = 8)	11 ± 3.56 (*n* = 19)	0.3456
30 min	9.17 ± 3.66 (*n* = 6)	11.07 ± 3.67 (*n* = 14)	0.3009
35 min	8.16 ± 3.43 (*n* = 7)	10.29 ± 3.05 (*n* = 14)	0.1862
Heart rate (beats/min)			
15 min	158.5 ± 30.00 (*n* = 6)	172.5 ± 35.06 (*n* = 14)	0.2663
20 min	158.75 ± 18.16 (*n* = 8)	193.3 ± 23.78 (*n* = 20)	0.0011†
25 min	172.63 ± 27.13 (*n* = 8)	196.45 ± 26.77 (*n* = 20)	0.0437*
30 min	178.33 ± 31.47 (*n* = 6)	195.44 ± 29.33 (*n* = 18)	0.2366
35 min	180.33 ± 30.82 (*n* = 6)	182.36 ± 28.08 (*n* = 14)	0.8873
CO_2 (first recorded value only)			
Minutes from injection	38.5 [36, 41] (*n* = 10)	38 [33, 41] (*n* = 23)	0.6344
Millimeters of mercury	67 [63, 73] (*n* = 10)	71 [67, 81] (*n* = 23)	0.0522

Times for respiratory rate and heart rate are given from injection with premedication/saline control. Heart rate and respiratory rate are presented as mean ± SD (sample size). CO_2 is presented as the median millimeters of mercury at the first CO_2 reading and median minutes from premedication of the first reading. CO_2 data are presented as median [quartile 1, quartile 3] (sample size at that time point). *, $P \leq 0.05$; †, $P \leq 0.01$.

and wing flapping, scored significantly better for the premedication groups than the saline control groups. PMED bird scores for recovery parameters correspond with a smooth, calm, no-restraint-required recovery with minimal wing flapping. Saline control birds scored an average score of 3 by comparison, which corresponds to a less smooth/calm recovery with no restraint required and included minimal wing movement/flapping. A study assessing alfaxalone induction in domestic chickens with and without premedication using a similar score chart also found that premedication significantly improved recovery scores.[4,23] Pharmacokinetic studies[6,12,16,20,36] showed half-lives for MDZ in turkeys ($t_{1/2}$ = 25.2 min) and half-lives for butorphanol in broilers (71.3 min), Hispaniola Amazon parrots ($t_{1/2}$ = 30.6 min), great horned owls ($t_{1/2}$ = 110.4 min), and red-tailed hawks ($t_{1/2}$ = 56.4 min). Based on the variability of these half-lives among different species, ranging from less than to longer than the surgical/inhalant anesthetic period (approximately 70 min for all groups), one could presume the premedication was still providing sedative, anxiolytic, and muscle-relaxation effects during the recovery period.

Overall, throughout all stages of the premedication and recovery process, birds that received premedication (PMED) scored significantly better on parameters that correspond to less stress for the turkeys and the staff handling them. While the birds in our study did not use their beaks as a primary weapon, one or more birds did cause minor scratches and abrasions to staff with their wings and claws. The reduced restraint required, along with minimal to no wing flapping observed in PMED birds, meant fewer opportunities for potential injury to both the birds and their handlers. This risk reduction was demonstrated in a 2014 study[27] of great white pelicans, a significantly larger avian species with greater potential to harm staff or cause self-injury. In the study, MDZ-B premedication improved patient demeanor and handling ability in the pelicans.[27] In the present turkey study, these improvements may have extended into the recovery period, resulting in the improved qualitative recovery scores recorded. While no self-injuries or failure of the surgically repaired tendon were noted in the present study during recovery in either group, it may be a worthy consideration that a smoother recovery would decrease the risk of self-injury in birds used for other models. One point to note in the present study is that standing was not used as the recovery complete parameter. This was due to the likelihood that the surgery and postsurgical casting would affect when and how some birds would be able to achieve and maintain a standing position. Some birds were able to stand almost immediately after extubation and effectively bypass an initial period of sternal recumbency. In those cases, recovery was marked complete at the time of standing.

A potential downside to the longer recovery time and procedural time of premedicated birds is the requirement for additional staff to properly monitor the birds and the equipment required to maximize the number of procedures performed per day. Additional monitoring equipment, anesthesia machines for supplemental oxygen, and recovery kennel space could all be increased with slower recoveries. The prolonged procedural time would be the rate-limiting step in procedures using premedication. One must also consider that a portion of the procedural period is the time from premedication until the initiation of inhalant anesthetic (PS-Inh). PS-Inh would be eliminated in animals that only receive mask induction (i.e., receive no premedication), thus reducing the total hands-on time per bird. The smoother induction and recovery exhibited in premedicated turkeys, however, represent a significant refinement leading to better animal welfare and should, therefore, be implemented even if its use results in fewer total procedures performed per day.

In the current study, physiologic parameters were generally not significantly different between groups. However, heart rate was significantly higher in the premedicated birds at the 20- and 25-min mark postinjection compared with the saline control birds. Reported heart rates in 17- to 21-mo-old female, broad-breasted bronze and Beltsville white turkeys ranged from 219.1 ± 10.3 and 193.1 ± 6.8 bpm, respectively.[37] Six-week-old broad white poults were reported with heart rates of 298 and 309 bpm in another study.[38] Our heart rate data for premedicated birds was generally within the reported normal range of the turkeys from these studies. Saline control birds had lower median heart rates than the reported ranges. Due to our low sample size and the fact that 2 of the 10 saline birds required 3% isoflurane (up from 2.5%) for successful intubation, it is possible that the heart rate data was affected as isoflurane in chickens at higher concentrations has been shown to lower the heart rate in domestic chickens.[39] This finding warrants further study to elucidate if the cause of the lower heart rate in some saline turkeys was the increased isoflurane requirement or if the higher heart rate in PMED birds was related to premedication. The CO_2 millimeters of mercury approached statistical significance; a larger group size may help determine if this difference would have been significant between the two groups. The CO_2 millimeters of mercury were rather high compared with levels generally considered normal (35 to 45 mm Hg) for avian species.[24]

When analyzing the perianesthetic scoring system, there was consistently good agreement (greater than 70%) between rater A (a veterinarian with avian anesthesia experience) and rater B (a surgical lab animal technician with general anesthesia experience). This percentage of agreement outperformed the expected agreement for all variables. The agreement level had a wider range when using Cohen's κ statistic. Categorizing the κ values as previously described by McHugh,[40] the level of agreement was defined as none (0 to 0.200), minimal (0.21 to 0.39), weak (0.40 to 0.59), moderate (0.60 to 0.79), strong (0.8 to 0.90), and almost perfect (above 0.90). There was strong/almost perfect agreement for induction ease and premedication. Moderate agreement was achieved for scores corresponding to premedication restraint required, premedication, muscle relaxation, and recovery tremors. Weak agreement was seen for apnea at induction, recovery restraint required, and recovery wing flapping.[40] One consideration for using this scoring system is that, whenever possible, 2 raters with avian experience should be used to increase the reliability of comparing scores between 2 raters. If only 1 rater is used, as was done in the study from which our scoring system was adapted, then that single experienced rater should evaluate all animals in the study to provide scoring consistency.[4] We elected to take this approach; after the rater agreement had been established, only rater A's data were used in further analysis.

One turkey (no. 30) experienced an unanticipated adverse event that was unrelated to treatment effect; the discovery of a piece of hay lodged within the oropharynx delayed successful intubation by approximately 4 min. An investigation into turkey no. 30 as a potential outlier was performed. The final decision to remove turkey no. 30 from the analysis was made following running the statistical analysis both with and without no. 30 included. While no perianesthetic scores or time range significance/lack of significance changed between the 2 analyses, the authors elected to remove no. 30 to present the most accurate version of the data that corresponds to the study objectives. The delay caused by the piece of hay would increase nearly all time points/ranges, and thus, with turkey 30 being included the median time ranges would be affected and no longer represent the true results for comparison of PMED compared with saline being evaluated. The changes in isoflurane administration required to remove the hay and then resume induction to a depth suitable for intubation may also have caused other changes affecting scores at induction and recovery.

The current study has several limitations. The convenience sampling and resulted in a female-only sex bias and a low sample size; therefore, the findings may be limited to female broad-breasted white turkeys. Physiologic monitoring data (i.e., heart rate, respiratory rate, and CO_2) were not originally intended to be analyzed in this study. Thus, the personnel recording this data followed an anesthetic monitoring standard operating procedure that did not prioritize recording data at exactly 5-min intervals. This resulted in missing data points and an inability to compare the data with a repeated measures statistical method. The physiologic monitoring data were collected retrospectively from the anesthesia records and subsequently manipulated, as described in the methods section. The missing data points may have obscured subtle trends that might have been detected if a more consistent 5-min interval for data collection had been used. The missing data points also limited the statistical tests that could be run. Future studies should be conducted and designed to fully investigate the effects of heart rate and respiratory rate. The duration of oxygen administration changed during recovery, and this is another potential limitation. Extended delivery of oxygen may have affected the duration of time in recovery, but, due to the limited number of animals in the prolonged administration group, this was not an outcome we could explore. This is another outcome that could be further investigated in future studies. The final limitation was the lack of formal live animal training using the scoring rubrics before the start of the study. As outlined in the methods, a single rater (rater A) was responsible for modifying previously used rubrics to create the one used in this study. Rater B discussed the rubrics with rater A before the study started and asked for clarification about what the score should be if they found the rubric unclear before rating any birds. After demonstrating varied rater agreement, we elected to use only the ratings from the more experienced rater A in the analysis, which was less ideal than having 2 raters experienced in avian anesthesia.

While neither premedication strategy, MDZ or MDZ-B, was significantly better than the other, both provided clear advantages to mask induction alone. This study provides evidence of the benefits of premedication as a standard part of all turkey preanesthetic and presurgical protocols due to the improvement in the quality of induction and recovery, while also making turkeys appreciably easier for staff to handle.

Supplementary Materials

Supplementary Table S1. Interrater agreement and Cohen's κ.

Acknowledgments

The authors thank the Texas A&M University Comparative Medicine Program's Health Crew for their support during the procedures and study. They also thank the sponsors of the flexor tendon study for allowing the anesthetic study to be done simultaneously. The authors extend a special thanks to Millie Mattox, Tennille Lamon, MaryBeth Benda, and Jane Costa for providing support throughout various portions of the project and writing process. They also thank Derek Lemons, research statistician, Texas A&M, School of Public Health, for his statistical consultations, especially during the review process.

Conflict of Interest

The authors have no conflicts of interest to declare.

Funding

The external company providing funds for this project had no input or influence on any aspect of the study including study design, data analysis, and conclusions.

References

1. Kadar A, Liu H, Vrieze AM, et al. Establishment of an in vivo turkey model for the study of flexor tendon repair. *J Orthop Res.* 2018;36(9):2497–2505.
2. Molter C, Escobar A, Schroeder C. Backyard poultry and waterfowl sedation and anesthesia. *Vet Clin North Am Exot Anim Pract.* 2022;25(1):163–180.
3. Routh A. Veterinary care of the mute swan. In Practice. 2000; 22(8):426–443. doi:10.1136/inpract.22.8.426.
4. Baldrey V, Stanford M, Bacon H. A comparison of the pharmacodynamic effects of intravenous ketamine–xylazine with alfaxalone

in mute swans (*Cygnus olor*) presenting at a wildlife veterinary hospital. *Vet Anaesth Analg.* 2021;48(5):716–724.
5. Raftery A. Avian anaesthesia. *In Practice.* 2013;35(5):272–278.
6. Doss G, Mans C. Avian sedation. *J Avian Med Surg.* 2021;35(3): 253–268.
7. Echols MS. Backyard poultry and waterfowl anesthesia and surgery. In: *North American Veterinary Conference.* NAVC; 2013.
8. Curro TG. Anesthesia of pet birds. *Semin Avian Exotic Pet Med.* 1998;7(1):10–21.
9. Gunkel C, Lafortune M. Current techniques in avian anesthesia. *Semin Avian Exotic Pet Med.* 2005;14(4):263–276.
10. Zaheer OA, Sanchez A, Beaufrère H. Minimum anesthetic concentration of isoflurane and sparing effect of midazolam in Quaker parrots (*Myiopsitta monachus*). *Vet Anaesth Analg.* 2020;47(3):341–346.
11. Barnes Heller HL. Update on anticonvulsant therapy in the emergent small animal patient. *Vet Clin North Am Small Anim Pract.* 2020;50(6):1423–1431.
12. Cortright KA, Wetzlich SE, Craigmill AL. Plasma pharmacokinetics of midazolam in chickens, turkeys, pheasants and bobwhite quail. *J Vet Pharmacol Ther.* 2007;30(5):429–436.
13. Paul-Murphy J, Hawkins MG. Avian analgesia. In: Miller RE, Fowler M, eds. *Fowler's Zoo and Wild Animal Medicine.* W.B. Saunders; 2012:312–323.
14. Doneley B. *Avian Medicine and Surgery in Practice: Companion and Aviary Birds*. 2nd ed. CRC Press; 2016.
15. Singh PM, Johnson CB, Gartrell B, Mitchinson S, Jacob A, Chambers P. Analgesic effects of morphine and butorphanol in broiler chickens. *Vet Anaesth Analg.* 2017;44(3):538–545.
16. Riggs SM, Hawkins MG, Craigmill AL, Kass PH, Stanley SD, Taylor IT. Pharmacokinetics of butorphanol tartrate in red-tailed hawks (*Buteo jamaicensis*) and great horned owls (*Bubo virginianus*). *Am J Vet Res.* 2008;69(5):596–603.
17. Reim DA, Middleton CC. Use of butorphanol as an anesthetic adjunct in turkeys. *Lab Anim Sci.*1995;45(6):696–697.
18. Sha J, Kongara K, Singh P, Jacob A, Ponnampalam J. Pharmacokinetics and pharmacodynamics of butorphanol and dexmedetomidine after intranasal administration in broiler chickens (*Gallus gallus domesticus*). *Vet Sci.* 2022;9(5).
19. Singh PM, Johnson C, Gartrell B, Mitchinson S, Chambers P. Pharmacokinetics of butorphanol in broiler chickens. *Vet Rec.* 2011;168(22):588.
20. Guzman DS, Flammer K, Paul-Murphy JR, Barker SA, Tully TN Jr. Pharmacokinetics of butorphanol after intravenous, intramuscular, and oral administration in Hispaniolan Amazon parrots (*Amazona ventralis*). *J Avian Med Surg.* 2011;25(3):185–191.
21. Curro TG, Brunson DB, Paul-Murphy J. Determination of the ED50 of isoflurane and evaluation of the isoflurane-sparing effect of butorphanol in cockatoos (*Cacatua* spp.). *Vet Surg.* 1994;23(5):429–433.
22. Martel A, Doss GA, Mans C. Evaluation of the effects of intramuscular injection volume on midazolam-butorphanol induced sedation in domestic pigeons (*Columba livia*). *J Exotic Pet Med.* 2021;37:3–7.
23. Mastakov A, Henning J, de Gier R, Doneley R. Induction of general anesthesia with alfaxalone in the domestic chicken. *J Avian Med Surg.* 2021;35(3):269–279. 11.
24. Divers SJ. Anaesthesia of zoological species (exotic pets, zoo, aquatic, and wild animals). In: Clarke KW, Trim CM, Hall LW, eds. *Veterinary Anaesthesia*. 11th ed. W.B. Saunders; 2014:535–569.
25. Doss GA, Fink DM, Mans C. Assessment of sedation after intranasal administration of midazolam and midazolam-butorphanol in cockatiels (*Nymphicus hollandicus*). *Am J Vet Res.* 2018;79(12):1246–1252.
26. Silverin B. Stress responses in birds. *Poultry Avian Biol Rev.* 1998;9(4):153–168.
27. Horowitz IH, Vaadia G, Landau S, Yanco E, Lublin A. Butorphanol-midazolam combination injection for sedation of great white pelicans (*Pelecanus onocrotalus*). *Israel J Vet Med.* 2014;69:35–39.
28. Sanchez-Migallon Guzman D. Advances in avian clinical therapeutics. *J Exotic Pet Med.* 2014;23(1):6–20.
29. Mutoh T, Kojima K, Takao K, Nishimura R, Sasaki N. Comparison of sevoflurane with isoflurane for rapid mask induction in midazolam and butorphanol-sedated dogs. *J Vet Med A Physiol Pathol Clin Med.* 2001;48(4):223–230.
30. Harvey RC. Precautions when using mask induction. *Vet Clin North Am Small Anim Pract.* 1992;22(2):310–311.
31. Tsukamoto A, Iimuro M, Sato R, Yamazaki J, Inomata T. Effect of midazolam and butorphanol premedication on inhalant isoflurane anesthesia in mice. *Exp Anim.* 2015;64(2):139–145.
32. Kulkarni S, Hickman D. Isoflurane and carbon dioxide elicit similar behavioral responses in rats. *Animals (Basel).* 2020;10(8).
33. Joyner PH, Jones MP, Ward D, Gompf RE, Zagaya N, Sleeman JM. Induction and recovery characteristics and cardiopulmonary effects of sevoflurane and isoflurane in bald eagles. *Am J Vet Res.* Jan 2008;69(1):13–22.
34. Midon M, Escobar A, Yamada DI, de Freitas PO, Minto BW, Barletta M. Isoflurane respiratory anesthetic index in chickens (*Gallus gallus domesticus*). *J Zoo Wildl Med.* 2021;52(1):327–331.
35. Flecknell P, Roughan J, Hedenqvist P. Induction of anaesthesia with isoflurane and sevoflurane in rabbit. *Lab Anim.* 1999;33(1):41–46.
36. Schwartz M, Muñana KR, Nettifee-Osborne JA, Messenger KM, Papich MG. The pharmacokinetics of midazolam after intravenous, intramuscular, and rectal administration in healthy dogs. *J Vet Pharmacol Ther.* 2013;36(5):471–477.
37. Ferguson TM, Miller DH, Bradley JW, Atkinson RL. Blood pressure and heart rate of turkeys, 17–21 months of age. *Poult Sci.* 1969;48(4):1478–1481.
38. Krista L, Burger R, Waibel P. Blood pressure and heart rate in the turkey as measured by the indirect method and their modifications by pharmacological agents. *Poultry Sci.* 1963;42(3):646–652.
39. Deori P, Sarma KK, Nath PJ, Singh CK, Nath R. Physiological alteration, quality of anesthesia and economy of isoflurane in domestic chickens (*Gallus domesticus*). *Vet World.* 2017;10(5):493–497.
40. McHugh ML. Interrater reliability: the kappa statistic. *Biochem Med (Zagreb).* 2012;22(3):276–282.