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Article

The Determination of the Minimum Anaesthetic Concentration of Halothane in the Rock Dove (*Columba livia*) Using an Electrical Stimulus

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Simple Summary: An ethical way to examine nociception in animals is to do so when they are anaesthetised. Nociception is the process by which the nervous system perceives a noxious stimulus, and in the conscious animal, this could result in pain. This nociceptive process can still be active in anaesthetised animals, and the resulting electrical activity in the brain can be measured by electroencephalography (EEG). By using this technique, it is possible to quantify the nociceptive response of an animal. To allow comparisons between animals and studies using this model, a method of standardising anaesthetic concentration is required for each species examined. The volatile agent halothane has been used successfully for nociception assessment in both mammals and birds. For volatile anaesthetics, the standardised level is known as the minimum anaesthetic concentration (MAC). As the MAC of halothane is not known in Rock Dove (*Columba livia*), and this study aims to determine this using electrical stimulus.

Abstract: This study aims to determine the minimum anaesthetic concentration (MAC) of halothane in the Rock Dove using electrical stimulus. Seven Rock Doves are anaesthetised with halothane, and the MAC is determined using the bracketing method. An electrical stimulus (two single pulses and two five-second stimuli, all separated by five-second pauses; 30 Hz, 30 V, 7.5 ms) is applied to the legs via subcutaneous electrodes. A maximum of eight periods of electrical stimulation, each with a preceding 15 min stable phase, is applied to each bird. If the non-reflexive movement occurred following stimulation, the end-tidal halothane (Fe'Hal) is increased by 10% before the next stimulus delivery. If no movement occurred, Fe'Hal is decreased by 10%. The MAC is the average of the highest concentration that allowed movement and the lowest that prevented movement. Physiological variables and ventilatory settings are recorded every five minutes. The current delivered is calculated offline. The mean ± SD MAC of halothane is 1.62 ± 0.29%, calculated from five birds. During the entire anaesthesia, all birds had cardiac arrhythmias —with three having sporadic recurrent periods of prolonged ventricular standstill followed by marked sinus tachycardia. The mean recorded voltage and calculated current and resistance are 27.6 ± 2.7 V, 20.3 ± 7.3 mAmp and 1.6 ± 0.9 kΩ, respectively. The advantage of halothane for prolonged anaesthesia in Rock Doves may be limited when noxious stimulation is used, due to the development of severe ventricular arrhythmias.

Keywords: avian; Rock Doves; *Columba livia*; halothane; minimum anaesthesia concentration; cardiac arrhythmias; neurophysiology

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1. Introduction

Ornithological research in the field and laboratory may often require the use of anaesthesia. It is important that the significant impact of anaesthesia on outcome variables

is considered when standardising research techniques [1]. This is particularly relevant for variables related to the central nervous system, the target of general anaesthesia. This standardisation of anaesthesia originates with species, drug and dose considerations. The Rock Dove (*Columba livia*) has been an often-used species in laboratories investigating neurological responses, including memory loss and cognition [2,3]. Halothane is an inhalant anaesthetic agent recommended for anaesthesia in some electrophysiological studies, particularly those exploring brain and cortical responses to noxious stimuli. In this context, halothane is superior to the newer commonly used inhalant agents, such as isoflurane and sevoflurane, as it preserves cortical responses to a much greater degree, at least in chickens and various mammals [4,5]. The MAC of halothane in Rock Dove has not yet been reported.

The minimum alveolar concentration (MAC) of an inhalant anaesthetic provides the basis for standardising inhalant anaesthetic administration in both research and clinical settings [6]. The basic MAC of any given agent is also known as the ED_{50}, a level where the dose is effective in 50% of the given population. Many MAC and ED levels are reported for various purposes along a dose-response curve. These range from the level required for verbal command in humans, to that required to suppress any autonomic responses in the tested population. The reported MAC of inhalant anaesthetics in birds is limited to a small number of the 10,000 bird species and agents, with techniques extrapolated from those used in mammalian research. In birds, MAC is specifically known as minimum anaesthetic concentration as birds do not possess alveoli, rather gas exchange occurs in the parabronchi. Some isoflurane MAC values reported in birds include chickens (isoflurane 1.25 $\pm$ 0.13%), ducks (isoflurane 1.30 $\pm$ 0.23%) and some parrot (isoflurane, 2.09 $\pm$ 0.17%) and hawk species (isoflurane, 2.05 $\pm$ 0.45%). [7–11] Notably some of the reported MAC values of Rock Doves for isoflurane (1.8% $\pm$ 0.4%) and sevoflurane (3.0 $\pm$ 0.6%) are considerably higher than those of chickens (1.25 $\pm$ 0.13%, 2.21 $\pm$ 0.32%) [9,10]. As yet, the MAC for halothane in Rock Doves has not been reported, and it is anticipated to be much higher than that reported for chickens. The objective of this study was to describe the MAC of halothane in the Rock Dove using an electrical stimulus.

2. Materials and Methods

The study was a prospective, experimental, descriptive study. Seven adult Rock Doves (*Columbia livia*) of undetermined sex, weighing 341 $\pm$ 34 g, were included in the study. The number of animals chosen was due to previous pigeon MAC studies using the same number and the bracket method being accurate in small sample sizes [12–14]. The birds were obtained during a pest population cull in Wellington, New Zealand and transported the same day to the housing at Massey University Veterinary Tower by ventilated private vehicle with visual assessment each 30 min. On arrival at the laboratory the birds were assessed to be in good health by physical examination by a wildlife veterinarian, then weighed and leg-banded with unique colours for identification. Prophylactic parasiticide administration via crop syringe was carried out given the unknown parasite load of the birds (0.2 mg kg^{-1} moxidectin, Cydectin Long Acting Endectocide for Cattle and Sheep 1 mg mL^{-1}, Zoetis, Auckland, New Zealand; 25 mg kg^{-1} toltrazuril, Baycox Coccidiocide for Piglets 50 mg mL^{-1}, Bayer, Auckland, New Zealand). Additionally, 10–15 mL of standard saline solution were administered to ensure hydration status following capture and transport. The birds were housed in two parrot cages (Avi One, Ingleburn, Australia), four birds per cage, in a temperature (18–22 °C) and artificial light-controlled room. The cages were lined with clean newspaper that was replaced every 24 h. The room was set to a 12 h light-dark cycle with unregulated humidity and was an open system setup. Ad-libitum access to a mixed seed diet and doxycycline-treated water (180 mg L^{-1}, Doxyvet, The Australian Pigeon Company, Bayswater Australia) was allowed during lighted hours with both food and water sources renewed every 24 h. Feed only was removed in the evening and replaced each morning. An acclimatisation period of at least seven days was allowed prior to commencing experimental procedures.

All procedures were approved by the Massey University Animal Ethics Committee (protocol 18/90) and performed at near sea level.

Birds were removed from housing prior to morning feeding on the day of anaesthesia. The order in which the birds were sampled was randomised using an out-of-the-hat method, with one bird per day. Prior to induction of anaesthesia, each bird was weighed (Navigator® portable scale, Ohaus, NJ, United States) and its crop palpated to ensure it was empty. With the bird held gently in a towel, a custom-made face mask with an eye shield was placed over the bird's face and connected to a T-piece breathing circuit. One minute of pre-oxygenation with 2 L min^{-1} was followed by the incremental introduction of halothane (Halothane-Vet, Piramal Enterprises Limited, Mumbai, India). The vaporiser setting was increased by 1% every 60 s until spontaneous eyelid movement was abolished. The trachea was then intubated with a 3.0 mm internal diameter cuffed endotracheal tube (Jorvet, Loveland, CO, USA), the cuff inflated, and connected to the T-piece circuit of a small animal ventilator (SAV04, Vetronic Services, Staines-upon-Thames, UK).

A 5 Fr silicone feeding tube (Paediatric Feeding Tube, Mallinckrodt, UK) that had been passed through the side stream port of the endotracheal tube adaptor and situated in the distal endotracheal tube prior to intubation was connected to the sampling line of a multi-parameter monitor (Carescape B650 Anaesthetic Monitor; GE Healthcare, Helsinki, Finland). End-tidal gases were measured continuously using a sampling rate of 200 mL min^{-1}. This monitor was calibrated as per the manufacturer's recommendations using a calibration mix of 2.0% desflurane and 5.01% carbon dioxide (Quick Calibration Gas, GE Healthcare, Helsinki, Finland). The multi-parameter monitor was also used to measure pulse rate (via pulse oximetry), peripheral arterial oxygen haemoglobin saturation (SpO_2), respiratory rate (fr) and respiratory gases [end-tidal expired carbon dioxide ($Fe'CO_2$, partial pressure), end-tidal halothane concentration (Fe'Halo, percentage) and end-tidal inspired oxygen fraction (FiO_2, percentage)].

The lead II ECG trace was recorded using 30-gauge 7 mm stainless steel electrodes (subdermal needle electrode XS, Technomed, Maastricht, Netherlands) placed subcutaneously on the proximal cranial wing margin and medial thigh. The data were digitised (Powerlab 7/35; AD Instruments, Sydney, Australia) and continuously recorded (LabChart Pro v8.1.13; AD Instruments, Sydney, Australia). A LabChart channel was programmed to detect ventricular depolarisation (QRS complexes) and display the heart rate (bpm) in real-time. Physiological values and ventilation and anaesthesia settings were recorded every five minutes. Cloacal temperature (°C) was recorded every 15 min with a rectal thermometer (target 42 °C).

Immediately following tracheal intubation, mechanical ventilation was initiated. The peak inspiratory pressure (PIP), expiratory time and fresh gas flow (FGF) were adjusted to target normocapnia ($Fe'CO_2$ of 30–40 mmHg, 4–5.3 kPa). The FGF was reduced to the minimal rate causing no rebreathing of carbon dioxide. During anaesthesia 5 mL kg^{-1} h^{-1} of Hartmann's solution was delivered via a 22-gauge 25 mm catheter (BD Insyte, Becton Dickinson Infusion Therapy, Franklin Lakes, NJ, USA) inserted into a medial metatarsal vein. Two birds did not receive intravenous fluid therapy as an intravenous catheter could not be placed. Thermal support was provided using a forced warm-air blower, infrared light and space blanket. The times of mask placement, halothane start, intubation and stimulation period start (application of first electrical stimulus at each halothane concentration) were recorded.

Two 30-gauge 7 mm stainless steel electrodes (subdermal needle electrode XS, Technomed, Maastricht, Netherlands) were placed subcutaneously 10 mm apart over the lateral tibia or femur. These electrodes were attached to a constant voltage generator (S48 Square Pulse Stimulator, Astro-Med Inc. Grass Product Group, West Warwick, RI, USA) set to deliver an electrical stimulus of 30 V at 30 Hz with a pulse duration of 7.5 ms. The stimulation sequence was two single impulses followed by two five-second stimuli, each separated by a five-second pause. The sequence was manually programmed for every application. The pattern of the stimulus delivery was adapted by that described by Valverde et al. [15].

Stimulation current and electrode impedance were calculated from the potential difference across a 150 Ω resistor placed in series with the electrodes. This potential difference was measured using a digital oscilloscope (QC 1934 100 mhz digital storage oscilloscope, Digitech, Osaka, Japan) placed in parallel with the resistor. Electrode position was changed between left and right tibia and femur at each stimulation. Each location was used a maximum of two times. Electrodes were replaced or repositioned to maintain impedance $\leq$ 4 kΩ.

The MAC of halothane was determined using the bracketing method [16]. At each halothane concentration assessed, the end-tidal halothane was kept stable for 15 min prior to stimulation. A period of electrical stimulation was then applied, and the bird observed for gross movement. If movement occurred (a positive response) the end-tidal halothane was increased by approximately 10% for the next stimulation. If no movement occurred (a negative response), it was decreased by approximately 10%. This process was repeated up to eight times for each bird. The starting concentration for each bird was dictated by the final halothane concentration tested for the preceding bird. Based on the difference between reported MAC values of isoflurane and sevoflurane in chickens and Rock Doves, the initial halothane concentration tested for the first bird was 1.1% [8]. A positive movement response was considered to be any non-reflexive movement of the extremities, excluding the stimulated leg. These include head or neck lifting, wing flapping or leg kicking [8]. Each stimulation period was applied to completion or terminated immediately if a positive response was seen. After the completion of the MAC measurements, the birds were deeply anaesthetised with halothane prior to being euthanised with IV sodium pentobarbital, given in 500 mg increments until all cardiac electrical activity ceased as determined by the ECG.

Halothane MAC was calculated using the mathematical averaging method. The MAC was defined as the average of two consecutive inhalant concentrations, one resulting in movement and one not, in response to a noxious stimulus [17]. When multiple MAC values were obtained in an individual, these values were averaged. Individual MAC values were then averaged for the group of birds to give a final population MAC value.

Values for the physiological variables and equipment settings recorded at the end of each 15 min stabilisation period, prior to the stimulation, were pooled and averaged across all stimulation periods. These were then presented as the median. The ECG traces were visually assessed offline for the presence of arrhythmias; their appearance was described in relation to when the MAC bracket was obtained during data collection. The total times for anaesthesia (intubation to completion of last stimulation period) and stabilisation (intubation to the first stimulus) were calculated from the anaesthetic records following completion of data collection.

Data were tested for normality using Shapiro-Wilk assessment and by visual distribution assessment and central tendencies expressed as mean ± standard deviation if normally distributed or median (range) if not. The statistical package Prism 8 for macOS (Version 8.1.0 (221), 3 April 2019, GraphPad Software, San Diego, CA USA, www.graphpad.com) was used for all data analysis.

3. Results

All seven birds completed the study. Average bodyweight at the time of procedure was 342 ± 26 g. All birds were successfully intubated within five minutes of halothane administration. Total anaesthesia duration was 219 ± 34 min with 158 ± 38 min from the first stimulation period to completion of the last stimulation period. The physiological variables and equipment settings recorded immediately prior to stimulus application are shown in Tables 1 and 2.

During anaesthesia a number of cardiac arrhythmias were detected; second-degree atrioventricular block, atrioventricular dissociation and prolonged (maximum 20 s) ventricular standstill followed by sinus tachycardia. Three birds had cardiac arrhythmias during the pre-stimulation 15 min stable period (Table 3). Additionally, arrhythmias were detected

through the remainder of the anaesthesia when not apparent during the pre-stimulation stabilisation period. Five birds had an intermittent second-degree atrioventricular block. Three birds had sporadic recurring periods of prolonged (<20 s) ventricular standstill followed by marked sinus tachycardia that worsened over the course of the experiment. The cyclical reappearance of the ventricular arrhythmias caused the physiologically stable periods to shorten over the anaesthetic period. In one bird the ventricular arrhythmia occurred before the application of stimulation period number seven prior to a bracket being obtained.

Table 1. The mean ± standard deviation or median (range) values for physiological variables, recorded prior to each stimulation period of seven halothane-anaesthetised Rock Doves.

Variable	Value
HR (bpm)	118 (55–208)
f_R (bpm)	20 (11–32)
$P_E'CO_2$ (mm Hg)	35 ± 3
SpO_2 (%)	100 (96–100)
Cloacal Temperature (°C)	40.2 (39.4–41.7)

Table 2. The mean ± standard deviation or median (range) values for ventilator settings, recorded prior to each stimulation period of seven halothane-anaesthetised Rock Doves.

Setting	Value
FGF (L min^{-1})	1.0 (0.8–2.0)
PIP (cm H_2O)	3 (2–4)
Expiratory time (s)	1.4 (0.8–2.0)
FIO_2 (%)	96 (95–97)

Table 3. Occurrence and type of cardiac arrhythmias noted during the determination of halothane MAC in seven Rock Doves. Arrhythmias observed during the pre-stimulation stabilisation period are listed.

Stabilisation Period	Bird Number						
	1	**2**	**3**	**4**	**5**	**6**	**7**
1	0	0	0	0	0	1	0
2	0	0	0	0	0	1	0
3	0	0	0	1	0	1, 2	0
4	0	0	0	1	0	1, 2	0
5	0	0	0	1	0	1, 2	0
6	-	1	0	1	0	1, 2	0
7	-	1	0	1	0	1, 2	0
8	-	1	0	1	0	1, 2	0

0 = none, 1 = Second degree atrioventricular block and/or atrioventricular dissociation, 2 = Ventricular standstill followed by sinus tachycardia.

The MAC of halothane was determined as end-tidal halothane of 1.62 ± 0.29%, calculated from five birds (Table 4). Two birds did not contribute to the MAC calculation as they had only either positive or negative responses (i.e., no brackets achieved). The range of MAC values for the five birds was 1.15% to 1.95%. A total of 53 stimulation periods were recorded. The first bird had five repetitions of the stimulus applied, due to logistical constraints, and the remaining six birds had eight. The in-situ impedance of the stimulus electrodes could not be reduced to ≤4 kΩ on 14 of 53 (26%) occasions. The average recorded voltage and calculated current and resistance were 27.6 ± 2.7 V, 20.3 ± 7.3 mAmp and 1.6 ± 0.9 kΩ, respectively. The non-reflexive movement observed during all positive responses was wing flapping.

Table 4. The starting end-tidal halothane concentrations for each of seven Rock Doves during MAC determination, the number of MAC brackets achieved for each bird and the individual MAC values.

Bird Number	Starting End-Tidal Halothane (%)	MAC Brackets Achieved	Individual MAC
1	1.1	0	-
2	0.95	0	-
3	1.7	2	1.67
4	1.5	4	1.6
5	1.7	3	1.15
6	1.2	1	1.95
7	1.9	3	1.72

4. Discussion

The present study reports a halothane MAC for Rock Dove. The halothane MAC value determined for Rock Doves using electrical stimulus was 1.62 ± 0.29%. Severe ventricular arrhythmias were noted in a number of birds later in the anaesthetic procedure.

The variation of MAC values attributed to species is typically quoted as 10–20%, with between-species variation less at around 10% [12]. This has not been maintained in previous pigeon studies where considerably more variability in both individual and group MAC values have been reported [13,14,18]. The present study further challenges this concept, with both intra- and interspecies variation much higher than reported. Individual bird MAC values in the current study ranged from 1.15% to 1.95%, a spread of nearly 60%. Chicken and pigeon studies using comparable study designs demonstrated MAC of isoflurane in Rock Doves up to 56% higher than that of chickens [10,14]. This marked difference is mirrored in the present study with nearly double the MAC of halothane in the Rock Doves compared to that reported for chickens (0.85% ± 0.09%) [8]. The MAC values reported in other free-flying bird species are much closer to those of Rock Doves. For example, red-tailed hawks have an approximately 10% greater isoflurane MAC than that reported for Rock Doves [7,19].

The bracketing method is most commonly used for MAC determination in both mammals and birds, suitable for use with small sample sizes [7,19,20]. The noxious stimulus in reported MAC studies in birds has been either electrical or mechanical [7,14]. A recent review of MAC in dogs suggests the intermittent electrical stimulus is now the more commonly used method of noxious stimulation [15,21].

Physiological status and experimental setup may also impact determined MAC values. Sustained adverse physiological states that can occur during general anaesthesia, including hypoventilation, hypoxaemia, hypotension and hypothermia, can significantly alter the MAC value obtained [12]. In the present study, potential limitations included those variables that were not controlled or measured, specifically body temperature, blood pressure and cardiac rhythm. Hypothermia has been shown to reduce MAC of halothane by approximately 5% for every 1 °C decrease of body temperature in dogs, rats and children [22–24]. Birds in this study exhibited mild hypothermia, with a range of 39.4 to 41.7 °C during anaesthesia. The normal body temperature of a conscious pigeon is approximately 42 °C [14]. Though the impact of hypothermia on MAC determination has not been established in Rock Doves, given the consistent effect across species, including goldfish, it seems reasonable to assume a similar impact [25].

Thus, birds with a mean temperature 2 °C below this value may have as much as a 10% underestimation of MAC. All reasonable efforts were taken to maintain a normal body temperature in the present study, including the bird resting on a hot-water mat and forced warm-air blower blanket, covered with an aluminium space blanket and the use of an infrared lamp when the bird was uncovered. During the stimulation periods, all heating devices were removed to allow visual access to the bird and minimise ECG interference. A temperature decrease was observed during these times.

Hypotension has also been shown to significantly affect MAC values, at least in dogs [26]. In the present study, we were unable to effectively measure or monitor arterial blood pressure, due to the difficulty of placing an arterial catheter in these sized birds. During anaesthesia with halothane, ducks and chickens were normotensive [8,27]. The use of Doppler blood pressure monitoring in recent pigeon MAC studies on larger bodyweight birds is reported, however the reliability and precision of this method of blood pressure measurement have not been validated in Rock Doves [14]. A possible refinement may be the investigation of catheterisation of the femoral or carotid artery in these small-sized animals.

Hypoventilation, as characterised by elevated end-tidal carbon dioxide tension, was not detected. Though the gold-standard assessment of oxygenation is an arterial partial pressure, this was unable to be assessed in the current study, due to the size of the birds used. The level of arterial blood haemoglobin saturation with oxygen detected by pulse oximetry and the arterial partial pressure of oxygen in anaesthetised Rock Doves has been assessed [28]. At high pulse-oximetry readings, as seen in the current study, it indicates that the Rock Doves had an arterial partial pressure of oxygen of at least 100 mmHg, such that none would have been hypoxaemic.

Cardiac arrhythmias were observed in all birds during anaesthesia, however, the type of arrhythmia or timing of application of the electrical stimulus meant that the risk of these events impacting MAC measurement was minimised. The second-degree atrioventricular block was regularly observed, similar to reports in isoflurane and sevoflurane anaesthetised Rock Doves [13,14]. One bird also had atrioventricular dissociation, an arrhythmia not previously reported in Rock Doves. Atrioventricular dissociation is characterised by independent atrial and ventricular pacemakers at very similar or equal rates [29]. Both rhythms are considered physiologically benign, as there were no observable impacts on the bird's cardiovascular stability. Thus, these kinds of arrhythmias are unlikely to have impacted MAC measurements made during their occurrence.

In contrast, three birds had severe ventricular arrhythmias as the anaesthetic period progressed. Halothane is recognised to have catecholamine-enhanced arrhythmogenic effects in both animals and humans, and such arrhythmias can often be seen during noxious stimulus [30]. Ludders reported similar ventricular arrhythmias to those seen in the current study with halothane use in ducks, which in one case was fatal [27]. Though they reported successful treatment with lidocaine, this drug is known to reduce MAC in dogs, rats and humans [12], so this was not attempted. The real-time clinical assessment suggested that these birds' cardiac output was markedly reduced during these arrhythmias with precipitously decreasing expired carbon dioxide tensions noted. When the abnormal rhythm reverted to sinus rhythm, these birds rapidly regained physiological stability. Thus, when these arrhythmias were observed, the electrical stimulation was applied only during periods of normal sinus rhythm to minimise their effect of on the MAC value. To understand the degree to which these arrhythmias have an impact on the MAC value, a much larger number of animals would be needed, as would the more invasive assessment of cardiac function.

As this study was terminal, it was not established if the arrhythmias decreased or stopped once anaesthesia was discontinued, as previously noted in halothane-anaesthetised ducks [27]. Previous reports are conflicting on the natural occurrence of arrhythmias in conscious Rock Doves [14,31]. However, as conscious rhythm assessment was not done, it is unknown whether the birds in the present study had arrhythmias prior to anaesthesia.

A number of different modalities have been used in MAC studies of animals and humans to provide the noxious stimulus. Providing that the stimulus is supramaximal, a similar MAC will be achieved, regardless of the modality used [12]. If the stimulus is not supramaximal, some of the intra- and inter-individual variability in the MAC value may be attributable to the stimulus intensity [17]. In the case of electrical stimulation, the intensity of the stimulus is determined by the current—which, therefore, determines its sub- or supramaximal nature. In the present study, the current delivered using a constant 30 V stimulus was calculated to be around 20 mA. This voltage was chosen because

reports from red-tailed hawk MAC studies showed evidence of tissue damage at the site of electrode insertion when using a 50 V stimulus, with subsequent studies utilising 30 V [7,11]. However, the delivered current is influenced by naturally varying impedance between the stimulating electrodes and so can be reduced, despite the voltage being held constant [32]. To reduce the risk of delivering a sub-maximal stimulus, recent studies have utilised constant-current stimulation as opposed to constant-voltage [32,33]. No information is available on the voltage or current intensity required for a supramaximal stimulus in birds. Furthermore, a study investigating the MAC of sevoflurane in thick-billed parrots demonstrated a significantly higher MAC resulting from electrical versus mechanical stimulus [34]. This supports the use of electrical stimulus in the current study as a presumptive supramaximal stimulus.

The present study is the first to report the electrical current measured during a MAC study in birds. Constant-current stimulation is recommended in future studies for improved consistency of the electrical stimulation [32]. Using electrical stimulation is not without disadvantages. Rat data suggest that repeated testing in the same location can result in desensitisation and a falsely decreased tendency for movement [35]. In the present study, electrode positions were changed for each successive stimulation to protect against desensitisation.

The form in which the electrical stimulus has been applied in previous MAC studies has varied [21]. Commonly the electrical stimuli have been applied for an arbitrary period, usually 60 s. The stimulus has been terminated when there is gross movement or when the time has elapsed with no movement noted. This technique is still used regularly in both mammals and birds [7,33]. A technique developed and validated in dogs and rabbits by Valverde [15] indicated that a shorter discontinuous stimulation pattern, performed over 20 s to 30 s, gave the same MAC values as the longer, potentially more traumatic, 60 s period. A recent review of MAC determination in dogs indicated that this method had become a common electrical stimulus used in MAC studies. [21] This is supported by its use in multiple recent studies in sheep and dogs [36–38]. A final limitation of the study is the use of the single concentration calibration gas mixture containing desflurane. It is appreciated that the most scientifically robust method of calibrating such equipment is to use a minimum of three different concentrations of the target agent. However, this study utilised a clinically available agent monitor and used the manufacturer's recommendations for calibration.

5. Conclusions

This is the first report of a MAC of halothane in Rock Doves, as well as selected physiological variables in halothane-anaesthetised Rock Doves, and current delivered during constant-voltage determination of MAC in birds. The development of severe ventricular arrhythmias may limit halothane use in prolonged anaesthesia in Rock Doves, particularly those undergoing noxious stimulation.

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