

**Anaesthesia with sevoflurane in pigeons: minimal anaesthetic concentration
determination and investigation of cardiorespiratory variables at 1 MAC**

Julie Botman¹ (BSc), Fabien Gabriel¹ (DVM), Alexandra Dugdale² (DVM, PhD,
Dip.ECVAA), Jean-Michel Vandeweerd¹ (DVM, PhD, DECVS)

¹Department of Veterinary Medicine, University of Namur, Namur Research Institute for Life Sciences, rue de Bruxelles, 61, 5000, Namur, Belgium.

²Faculty of Health and Life Sciences, University of Liverpool, School of Veterinary Science, Chester High Road, Neston CH64 7TE, United Kingdom.

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Corresponding author: Dr. Alex Dugdale

Email: alex@liv.ac.uk

Abstract

Objective. To determine the minimal anaesthetic concentration (MAC) of sevoflurane in pigeons, and to investigate the effects of 1 MAC sevoflurane anaesthesia on cardiovascular and respiratory variables compared with the awake state.

Design. Prospective, experimental study.

Animals. Seven healthy adult pigeons.

Procedure. After acclimatisation to handling, heart rate (HR), heart rhythm, respiratory rate (f_R), end-expired carbon dioxide tension ($PE'CO_2$), inspired CO_2 tension (iCO_2), indirect systolic arterial blood pressure (SAP) and cloacal temperature (T) were measured to determine baseline, 'awake' values. Pigeons were then anaesthetised with sevoflurane and MAC was determined by the "bracketing" method. The same variables were monitored during a 40 minute period at 1.0 MAC sevoflurane for each bird.

Results. Mean MAC was $3.0 \pm 0.6\%$ for SEVO. During maintenance of anaesthesia at 1.0 MAC, SAP decreased significantly ($P < 0.001$) without any significant change in HR. Although $PE'CO_2$ increased significantly ($P = 0.001$) despite an increase in f_R , awake $PE'CO_2$ values were unexpectedly low. Sinus arrhythmias were detected in 2 birds under sevoflurane anaesthesia. The times to tracheal intubation and to recovery were 2.5 ± 0.7 min and 6.4 ± 1.7 min, respectively. Recovery was rapid and uneventful in all birds.

Conclusions and clinical relevance. Sevoflurane is suitable for anaesthesia in pigeons.

List of abbreviations

f_R = respiratory rate

HR= heart rate

iCO_2 = inspired CO_2 tension

ISO = isoflurane

$PE'CO_2$ = end-expired carbon dioxide tension

SAP= indirect systolic arterial blood pressure

SEVO = sevoflurane

T= cloacal temperature

Introduction

Although avian anaesthesia can be performed with either injectable or inhalation agents, inhalation anaesthesia is often preferred because of rapid anaesthetic induction and recovery, rapid adjustments in anaesthetic depth, minimal biotransformation and minimal myocardial depression (Naganobu and Hagi, 2000; Rival, 2002; Escobar and others, 2009). Sevoflurane (SEVO) has been used for avian anaesthesia, where it is characterized by less airway irritation (Granone and others, 2012) and a lower blood solubility (Edling, 2006) than isoflurane (ISO), and is associated with faster anaesthetic induction and recovery times that could justify its wider use (Korbel, 1998; Joyner and others, 2008; Granone and others, 2012; Phair and others, 2012).

In the pigeon, anaesthetic protocols with SEVO have been described where the delivered concentrations (according to vaporiser dial setting) were 8% for induction and 4% for maintenance of anaesthesia (Korbel, 1998). As bird lungs contain air capillaries instead of alveoli, the term minimal alveolar concentration, as reported in mammals, is inappropriate. Instead, the term minimal anaesthetic concentration (MAC) is used. The MAC of SEVO is unknown in pigeons, as well as cardiorespiratory parameters under SEVO. In contrast, there is more information about the effects of ISO (Fitzgerald and Blais, 1991; Korbel, 1998; Touzot-Jourde and others, 2005; Botman and others, 2016). Protocols administering 3.0–5.0% ISO for induction and 1.5–3.0% for maintenance of anaesthesia have been described (Korbel, 1998; Touzot-Jourde and others, 2005; Botman and others, 2016), and two MAC values of ISO have been reported: $1.51 \pm 0.15\%$ (Fitzgerald and Blais, 1991) and $1.8 \pm 0.4\%$ (Botman and others, 2016). In addition, ISO anaesthesia in pigeons can result in hypercapnia, hypotension, mild hypothermia and second- and third-degree atrioventricular blocks (Botman and others, 2016).

The objectives of the current study were to determine the MAC of sevoflurane and the effects of sevoflurane anaesthesia on the cardiorespiratory variables at 1 MAC sevoflurane.

Materials and methods

Study design

This was a prospective, experimental study; the experimental protocol (**Figure 1**; 14/209/VA) was approved by the ethical committee for animal welfare of the University of Namur.

Birds and baseline monitoring parameters

Seven adult racing pigeons (*Columbia livia*) were used. They had a mean \pm SD (standard deviation) weight of 443 ± 60 g (1.0 ± 0.1 lb). They were housed in an aviary (3 x 2 x 2 m) between interventions. Food and water were available *ad libitum*. All pigeons were acclimatized to handling during a 1 month period before this study. They were selected for good health based on a physical examination and for absence of cardiac arrhythmias in the awake state.

One week prior to the study, baseline heart rate and rhythm were assessed with an electrocardiograph (ECG) (Mindray; PM-9000vet, China) , and systolic blood pressure was determined by Doppler plethysmography (Doppler Vet BP; Mano Medical, France) (**Figure 2**). Furthermore, capnography (Scio Four Oxi Plus; Dräger, Germany) was used to measure respiratory rate, inspiratory carbon dioxide tension and end-expired carbon dioxide tension (PE'CO₂), and cloacal temperature was measured (Mindray; PM-9000vet, China). These measurements were performed in a quiet environment without tranquilization. Pigeons were covered with a light towel. Flat clip electrodes were directly connected to the skin at the patagium of each wing and at the skin fold proximal to the left stifle joint. Pigeons were gently restrained for 15 minutes before recording measurements. To reduce stress, they were kept in an upright position (Lopez Murcia and others, 2005).

For capnography, pigeons breathed from a sectioned blind end of a latex glove finger fixed around the beak proximal to the nares (Botman and others, 2016) (**Figure 3**). The other end of the glove was used to administer a fresh gas flow of 100% oxygen delivered at 500mL/min via a non-rebreathing Bain system. Inspired and expired gases (for measurement of inspired CO₂ and PE'CO₂ tension) were sampled at a sidestream sampling rate of 200 mL/min via a 20-gauge non-blunted needle, inserted into the glove close to the beak and connected, via the sampling tubing, to the capnograph (measuring to 1 decimal place).

Measurements were recorded separately over a 10 minute period.

Induction and maintenance of anaesthesia

Before general anaesthesia, birds were fasted for 6 hours. After 5 minutes of preoxygenation with a facemask (100% oxygen, 1L/min), anaesthesia was induced with SEVO (Sevorane; Abbott, North Chicago, Ill) at an initial vaporiser setting of 7 %, delivered in 100% oxygen (1L/min) via a Bain non-rebreathing system. This SEVO percentage was chosen on the basis of the work of Joyner and others (2008), who compared 4% ISO and 7% SEVO in bald eagles; and we have previously reported the use of 4% ISO in pigeons (Botman and others, 2016). The vaporiser (Tec 7 Vaporizer, Datex-Ohmeda, Finland) was serviced regularly and its calibration was checked before and after the study. Once the bird was sufficiently relaxed, the facemask was removed, the bird's trachea was intubated with a 3 mm noncuffed endotracheal tube and it was positioned in dorsal recumbency (**Figure 4**). Sevoflurane was then delivered at 4% for a 15 min stabilization period before MAC determination began. Oxygen delivery was adjusted to a flow rate of 500mL/min for the remainder of the procedure. Birds were allowed to breathe spontaneously. An insulated mattress and an infrared light were used to maintain body temperature between 40.6 and 41.0°C (105.1-105.8°F).

Monitoring

The ECG was performed as in awake animals, except that birds were in dorsal recumbency. Conductive gel was applied sparingly, to increase electrode conductivity, but to minimize any cooling effect.

To collect end-expired gas samples, the tip of a 20-gauge non-blunted needle was inserted into the lumen of the endotracheal tube near its connection with the breathing system, and its hub was connected to the gas sampling line of a gas analyzer (Scio Four Oxi Plus; Dräger, Germany) for continuous monitoring of the SEVO concentration, inspired CO₂, PE'CO₂ and $\dot{V}R$. The fresh gas flow of oxygen was set at 4 x minute ventilation (minute ventilation was estimated as 200mL/kg/min, such that 100 % oxygen was delivered at 500mL/min via a calibrated oxygen flowmeter on the anaesthetic machine and then through a non-rebreathing Bain system). Gas samples were withdrawn at a sidestream sampling rate of 200 mL/min; withdrawn samples were not returned to the breathing system but were ducted to the waste anaesthetic agent scavenger system. Before the study began, the calibration of the gas analyzer was checked against a standard gas mixture (Quick Cal calibration gas: GE Healthcare) by the service engineers. In addition, before and after each anaesthetic, the analyzer's calibration was checked against room air (zero CO₂).

A Doppler blood flow probe and occlusive cuff with sphygmomanometer (Doppler Vet BP; Mano Medical, France), were used to monitor indirect SAP. A blood pressure cuff (size 2.5cm) (Pedisphyg, CAS Medical Systems Inc., USA) was positioned between the tibiotarsal-tarsometatarsal joint and the stifle joint. The width of the blood pressure cuff was approximately equivalent to 40-50 % of the limb circumference. The Doppler probe was placed, using ultrasound coupling gel to ensure adequate contact, distal to the blood pressure cuff, over the metatarsal artery.

To measure the body temperature an electronic temperature probe (Mindray; PM-9000vet, China) was inserted into the cloaca.

Minimal Anaesthetic Concentration (MAC) determination

After 15 minutes of stabilization and placement of the monitoring instruments, physiological variables were recorded at baseline (start of the period for MAC determination) and observed every five minutes, during MAC determination (**Figure 1**).

The MAC was determined by using the “bracketing” method described by Ludders and others (1989). Briefly, after stabilization at a predetermined end-expired anaesthetic concentration, the jaws of a Rochester-Carmalt forceps were clamped to the first ratchet lock on a digit until either a gross purposeful movement occurred (kicking of the limbs or moving of the wings), or for up to 60 seconds. The end-expired SEVO concentration was decreased by 10% if no motor response occurred. After 15 minutes of equilibration at each new constant end-expired SEVO concentration, this procedure was repeated until the bird reacted to the stimulus. The end-expired SEVO concentration was then increased by 10%, followed by 15 minutes of equilibration, until the reaction disappeared. End-expired SEVO concentration was determined as the average of four samples.

The time between tracheal intubation and the first stimulation for MAC determination (stabilization period) was 30 minutes. The MAC was defined and calculated as “the median value between the maximal end-expired concentration that allowed movement and the minimal end-expired concentration that prevented movement” (Naganobu and Hagi, 2000). Temperature and inspired CO₂ were observed during the whole procedure to ensure that no hypothermia and CO₂ inspiration occurred during MAC determination.

Recording of cardiorespiratory and other variables

Immediately after the MAC was determined, the end- expired SEVO concentration was maintained at 1.0 MAC for a period of 15 minutes for stabilization, followed by a period of 40 minutes during which monitored variables were recorded at 1.0 MAC (**Figure 1**). HR (beats/min), fR (breaths/min), iCO₂ (mmHg), PE'CO₂ (mmHg), SAP (mmHg) and T (°C) were recorded 3 times over each 5 minute epoch and a mean value was determined. ECG was recorded for the entire anaesthetic period, including the earlier periods of stabilization and MAC determination.

Several intervals of interest were measured. Time to induction of anaesthesia was defined as the time from initial delivery of volatile anaesthetic agent to successful tracheal intubation (Mercado and others, 2008). The time between tracheal intubation and the first stimulation for MAC determination (stabilization period) was recorded, as was the time from the start of inhalant anaesthetic administration to successful MAC determination. After the last monitored variable was measured, the vaporiser was turned off and 100% oxygen was administered until the endotracheal tube was removed. Time to tracheal extubation was defined as the time from cessation of volatile agent administration (vaporiser turned off) to the time of coughing, swallowing or shaking of the head, resulting in removal the endotracheal tube (Granone and others, 2012). Recovery time was defined as the time from cessation of volatile anaesthetic administration (vaporiser turned off) until the bird was able to stand and walk unassisted (Botman and others, 2016). The time from the start of anaesthetic administration to the time of cessation of anaesthetic administration was defined as the total anaesthetic time (Phair and others, 2012).

Data analysis

Data were collected in Microsoft Excel and were analyzed using IBM® SPSS® Statistics, version 21.0. Shapiro-Wilk tests were used to examine the normality of data. Parametric data were analysed using the paired Student t-test. The paired Wilcoxon test was used for non

parametric data. Results are presented as mean \pm SD and significance was set at a P-value of <0.05 .

Results

All data were normally distributed except tracheal intubation time.

MAC

The MAC value of SEVO in these seven pigeons was estimated at $3.0 \pm 0.6\%$. The time from the start of inhalant anaesthetic administration to successful MAC determination was 79 ± 26 minutes.

Variables recorded under SEVO anaesthesia

The times to tracheal intubation, tracheal extubation and recovery (to standing/walking unassisted), were 2.5 ± 0.7 min, 3.4 ± 2.1 min and 6.4 ± 1.7 min, respectively. The total anaesthetic time was 124.4 ± 25.7 min. Cloacal temperature remained stable, with a minimum value of 40.6°C (105.1°F) under SEVO. Inspired CO_2 remained at 0 mmHg throughout all the awake and anaesthetic procedures.

Mean (\pm SD) HR was 170 ± 24 beats/min and no arrhythmias were identified in awake pigeons. Under SEVO anaesthesia, HR was 149 ± 24 beats/min, fR was 40 ± 9 breaths/min, SAP was 91 ± 7 mmHg and $\text{PE}'\text{CO}_2$ was 46 ± 14 mmHg. Except for cloacal temperature and HR, these variables differed significantly from initial values in the awake state (**Table 1**). The $\text{PE}'\text{CO}_2$ values recorded from awake birds was, however, unexpectedly low.

The occurrence and progress of arrhythmias in anaesthetised birds is summarized in **Table 2**. Sinus arrhythmias (**Figure 5**) were detected in two pigeons under sevoflurane anaesthesia during MAC determination: fairly persistently in one bird and only transiently in the other. Sinus arrhythmia was determined on one occasion during anaesthetic maintenance at 1 MAC

SEVO in the bird which displayed fairly persistent sinus arrhythmia during MAC determination. All arrhythmias resolved after the discontinuation of anaesthesia.

Discussion

MAC

To date, the MAC value of SEVO in pigeons has not been reported. In the seven pigeons used in the current study, SEVO MAC was $3.0 \pm 0.6\%$, which is similar to the reported sevoflurane MAC of $2.9 \pm 0.1\%$ in guineafowl (Escobar and others, 2012), but higher than in other avian species: $2.21 \pm 0.32\%$ in chickens (Naganobu and others, 2003) and $2.03 \pm 0.32\%$ in crested serpent eagles (Chan and others, 2013). However, in this last study (Chan and others, 2013), body temperature significantly decreased during anaesthesia, which may have caused some underestimation of the MAC value.

The higher MAC value of SEVO in these pigeons, compared with some other avian species, might indicate a higher sensitivity of pigeons to mechanical stimulation applied to a toe. The MAC value, however, may vary considerably among avian species (Mercado and others, 2008) and can be influenced by many physiological factors (body temperature, metabolic rate, blood CO₂ tension and large variations in blood pressure (Quasha and others, 1980)). In the current study, as in several other studies determining MAC values in birds (Ludders and others, 1989; Naganobu and Hagio, 2000; Naganobu and others, 2003; Kim and others, 2011; Phair and others, 2012; Escobar and others, 2012), body temperature remained stable during all the anaesthetic procedures. Consequently, body temperature was unlikely to have influenced these results.

Inspired CO₂ remained zero although mild hypercapnia developed under SEVO anaesthesia (according to end expired CO₂ values), suggesting a degree of hypoventilation. Systolic arterial blood pressure during the MAC determination period was always higher than 78 mmHg indicating that only mild-to-moderate hypotension occurred. At the present time, there is no published information on how changes in CO₂ tension and arterial blood pressure may

affect MAC.

Characteristics of SEVO anaesthesia

SEVO produced a rapid and smooth anaesthetic induction in this series of pigeons. Time to induction (tracheal intubation) was shorter with 7% SEVO (2.5 ± 0.7 min) than reported with 4% ISO in a similar experimental set-up (4.3 ± 1 min; Botman and others, 2016). Anaesthetic induction with 7% SEVO in this study was, unsurprisingly, slower than in another study which used 8% delivered SEVO (95 ± 9 sec; Korbelt, 1998), for anaesthetic induction in racing pigeons (Sakai and others, 2005). Comparison of anaesthetic induction times was not an aim of the present study; nevertheless, anaesthetic induction times using different anaesthetic agent percentages would be interesting to investigate under the same experimental conditions.

Time to tracheal extubation (3.4 ± 2.1 min) was shorter than that reported following ISO anaesthesia under a similar experimental set-up (8.7 ± 1.8 min; Botman and others, 2016).

The time to recovery was also shorter with SEVO (6.4 ± 1.7 min) compared with ISO under similar experimental conditions (17.6 ± 3.4 min; Botman and others, 2016). These results are similar to previous studies reporting faster recovery times with SEVO compared to ISO in avian species (Korbelt, 1998; Joyner and others, 2008; Granone and others, 2012; Chan and others, 2013). These findings could be explained by the lower blood gas partition coefficient of SEVO (Patel and Goa, 1996), that is, a lower blood solubility of SEVO compared to ISO (Edling, 2006). This could be an advantage of SEVO over ISO for prolonged anaesthetic periods or in debilitated birds (Joyner and others, 2008). However, it is also possible that after prolonged anaesthesia with SEVO, the recoveries can be prolonged as body fat acts as a sink/reservoir for SEVO, despite the potential for more hepatic metabolism of SEVO than

ISO, which should help to offset this effect to some degree (Eger and others, 1997; Sakai and others, 2005; McKay and others, 2010).

In this series of pigeons, there was no significant difference in HR between initial, awake values (170 ± 24 beats/min) and those under SEVO anaesthesia (149 ± 24 beats/min). HR was well maintained under SEVO anaesthesia, as was previously reported under ISO anaesthesia in similar conditions (135 ± 28 beats/min; Botman and others, 2016). This is consistent with other studies in psittacines (Quandt and Greenacre, 1999), red-tailed hawks (Granone and others, 2012) and crested serpent eagles (Chan and others, 2013), but is at odds with a study in bald eagles in which HR was significantly higher under ISO anaesthesia than SEVO anaesthesia (Joyner and others, 2008).

Mild-to-moderate hypotension was identified under SEVO anaesthesia (SAP 91 ± 7 mmHg); similar to that which occurred under ISO anaesthesia in a similar experimental set-up (87 ± 11 mmHg; Botman and others, 2016). In other studies, no differences were reported in arterial blood pressure during ISO or SEVO anaesthesia in bald eagles (Joyner and others, 2008), and red-tailed hawks (Granone and others, 2012). Hypotension can be explained by a decrease of the systemic vascular resistance following the vasodilator effect of these inhalant agents (Souza and others, 2005; Schnellbacher and others, 2012). Hypotension can also be observed in association with cardiac arrhythmias (Lichtenberger, 2005).

The respiratory rate under SEVO anaesthesia in the present study (40 ± 9 breaths/min) was statistically significantly higher than initial values in the awake state (33 ± 5 breaths/min). In contrast, in birds of different species, the respiratory rate under SEVO anaesthesia in other studies was lower than in awake birds (Escobar and others, 2009; Phair and others, 2012; Chan and others, 2013). For measurements in the awake state in this study, pigeons were very calm and handled very gently to avoid any stress. They were also acclimatized to handling 1

month before this study, and for the measurements, they were covered with a light towel in an upright position and were allowed to relax for 15 minutes before recording measurements.

Alternatively, pigeons in the present study may have become more stressed during the induction of anaesthesia and therefore presented a higher respiratory rate compared with the awake state without any stress.

PE'CO₂ under SEVO anaesthesia demonstrated a mild hypercapnia (46 ± 14 mmHg), despite the higher respiratory rate, similar to previously reported values under ISO anaesthesia (52 ± 7 mmHg; Botman and others, 2016). Such mild hypercapnia is commonly observed during anaesthesia with spontaneous ventilation in birds (Ludders and others, 1989; Joyner and others, 2008; Escobar and others, 2011; Escobar and others, 2012; Botman and others, 2016). Hypercapnia can be explained by hypoventilation due to dorsal recumbency, muscle relaxation and the effects of the volatile anaesthetic agents on the central and peripheral chemoreceptors (Gleed and Ludders, 2001). In this study, the PE'CO₂ values recorded in awake pigeons were lower than expected (and lower than had been achieved using the same technique in a previous study (Botman and others, 2016)), perhaps due to sample-dilution, as can be associated with sidestream sampling techniques (Bilbrough, 2006). With this in mind, it is hard to draw firm conclusions regarding the observed changes in PE'CO₂ that occurred with SEVO anaesthesia.

In birds, the sinoatrial node determines the normal rhythm of the heart (Lumeij and Ritchie, 1994). Arrhythmias have been described with variability in their occurrence. No arrhythmias were recorded in 64 awake spanish pouler pigeons in an upright position without tranquilization (Lopez Murcia and others, 2005). In contrast, sinus arrhythmias have been reported in association with normal respiratory cycles and are considered to be physiologic in racing pigeons (as in other species); in addition, 24% of the birds presented with second-degree AV block (Lumeij and Ritchie, 1994). In another study, second-degree

atrioventricular blocks were identified and interpreted as a physiologic phenomenon in five percent of trained racing pigeons (Lumeij and Stokhof, 1985) . Second-degree atrioventricular heart blocks observed in another study resolved with increasing HR (Rüther, 1998).

In the current study, no arrhythmias were observed in the awake state. Sinus arrhythmias occurred in two birds under SEVO anaesthesia, and only transiently in one of these. In a previous study, arrhythmias occurred relatively frequently under ISO anaesthesia (Botman and others, 2016). During ISO anaesthesia, the most frequent arrhythmias were second-degree atrioventricular blocks (Botman and others, 2016), whereas in the present study, only sinus arrhythmias were detected with SEVO anaesthesia. Arrhythmias associated with ISO (mostly atrioventricular heart blocks) have also been reported in bald eagles (Aguilar and others, 1995; Joyner and others, 2008) and canvasback ducks (Machin and Caulkett, 2000). One comparative study reported arrhythmias (second-degree heart blocks, atrial premature contractions, ventricular premature contractions), with SEVO anaesthesia in birds, but to a lesser extent than with ISO anaesthesia (Joyner and others, 2008). The cause of the sinus arrhythmias observed might have been physiological, i.e. because of increased parasympathetic tone, although the cause and significance of this is unknown.

In conclusion, SEVO is a suitable volatile anaesthetic agent for use in pigeons and provides more rapid anaesthetic induction and recovery than previously reported for ISO anaesthesia. Furthermore, SEVO anaesthesia is associated with minimal cardiac rhythm disturbance.

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Table 1. Mean (\pm SD) heart rate (HR), systolic arterial blood pressure (SAP), respiratory rate (f_R), end-expired carbon dioxide tension (PE' CO₂), and temperature (T) in seven pigeons in the awake state and anaesthetised with 1 MAC sevoflurane. P values from paired t-tests are shown; asterisks show where values are statistically different ($P < 0.05$) within the row.

Variable	Awake state	Sevoflurane Anaesthesia	P values
HR (beats/min)	170 \pm 24	149 \pm 24	0.141
SAP (mmHg)	136 \pm 16*	91 \pm 7*	<0.001
f_R (breaths/min)	32 \pm 5*	40 \pm 9*	0.034
PE' CO ₂ (mm Hg)	17 \pm 6*	46 \pm 14*	0.001
PE' CO ₂ (kPa)	2.3 \pm 0.8*	6.1 \pm 1.9*	0.001
T° (°C)	41.2 \pm 0.5	40.9 \pm 0.2	0.198

Figure legends

Figure 1.

Study design: MAC determination using the bracketing method

Figure 2.

Indirect blood pressure measurement in an awake pigeon

Figure 3.

Sidestream capnography with sampling port at the hub of a 20-gauge needle inserted into a sectioned finger of a latex glove which was placed over the beak, proximal to the nares.

Figure 4.

Placement of ECG electrodes, temperature probe and blood pressure cuff in a pigeon anaesthetised with SEVO

Figure 5.

Sinus arrhythmia in a pigeon anaesthetised with SEVO

Figure 1

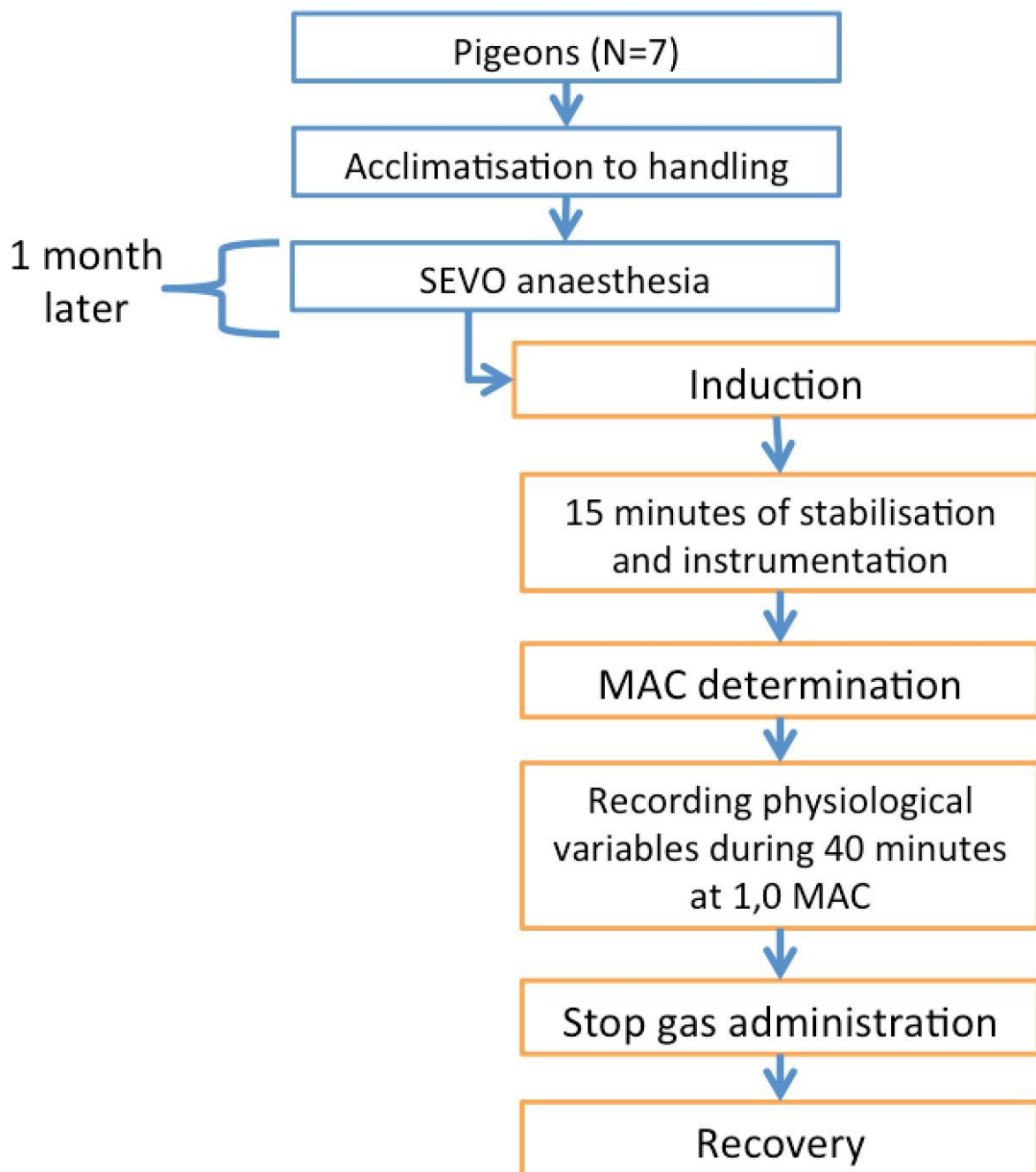


Figure 2



Figure 3



Figure 4



Figure 5

