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Comparison of two anesthetic inhalant agents (isoflurane and sevoflurane) on induction and recovery from anesthesia, physiological effects, hematocrit and biochemistry profiles in domestic pigeons (*columba livia domestica*)

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A B S T R A C T

Isoflurane and sevoflurane are among the newer inhalant anaesthetics that have shown fewer adverse effects than their older counterparts; as a result, they are used more frequently in avian practice. Determination of the differences between induction and recovery time of isoflurane and sevoflurane in pigeons (*Columba Livia Domestica*). In addition, the anaesthetic effects of each on their physiological parameters, hematocrit and plasma chemistry values were evaluated. A total of 20 male pigeons were allocated to two groups of 10 undergoing anaesthesia by 5% isoflurane and 6% sevoflurane in oxygen in a crossover, randomised design. Three data sets were collected pre-anaesthesia, at the end of anaesthesia with the cessation of medications, and 24 hours post-anaesthesia. Different physiological and biochemistry parameters were evaluated. It was shorter in pigeons treated with isoflurane, and they experienced a longer RT than the other group. No significant difference was observed between isoflurane and sevoflurane regarding RT. HR decreased in both groups at the end of anaesthesia, which was more significant in pigeons anaesthetised with isoflurane. Although RR and T decreased at the end of anaesthesia in both groups, they were lower within the group treated with sevoflurane. Both anaesthetics decreased plasma biochemistry factors in most samples except creatinine phosphokinase and albumin, which almost returned to the initial state 24 hours post-anaesthesia. However, isoflurane is considered the most commonly inhaled anaesthetic in avian practice; sevoflurane is recommended for anaesthesia in pigeons.

Keywords: *Anaesthesia, Avian, Isoflurane, Pigeon, Sevoflurane.*

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1 Introduction

Many techniques developed for anaesthesia in small animals are also used in avian medicine. Applying these techniques in birds is burdened by limitations attributed to the physiological and anatomical differences between avian and small animals(1). However, avian anaesthesia can be implemented by either injectable or inhalation agents. Inhalation anaesthesia is frequently preferred for clinical examination, diagnostic research, surgical processes, and investigation and management approaches (2).

Isoflurane and sevoflurane are among the newer inhalant anaesthetics that have shown fewer adverse effects than their older counterparts; as a result, they are used more frequently in practice (3). Isoflurane has traditionally been selected as the safest inhalant anaesthetic for diagnosing birds at high risk and in a critical state. Isoflurane anaesthesia is known for having minimal negative cardiovascular effects, thus providing rapid induction times and short recovery times; however, it can contribute to periods of anxiety and hyperactivity during induction and recovery. Apnea or cardiac arrhythmias have been reported in raptors due to the presence of a minimal concentration of this gas in the alveolae (4, 5).

Sevoflurane, a novel inhalant gas, may have potential benefits over isoflurane. Several types of research have demonstrated sevoflurane to be an appropriate inhalant gas for induction and quick recovery in mammals and birds (5, 6). In some species of birds, sevoflurane has shorter anaesthetic induction and recovery times (IT and RT), and there is normal physiological behaviour during recovery. There are relatively mild circulatory and respiratory adverse effects and a decreased outbreak of cardiac arrhythmias compared to effects arising from isoflurane anaesthesia. The respiratory suppression arising from volatile anaesthetics is one of the main concerns in spontaneously breathing avian patients.

The myorelaxation (muscle relaxation) induced by inhalant anaesthetics may cause respiratory suppression by attenuating the respiratory muscles; therefore, evaluation of the respiratory rate during anaesthesia is another important factor besides heart rate to consider (7). Progressive respiratory depression has been described in different species of birds, suggesting that avian species have particular cardiorespiratory response variations to inhalant anaesthetics. During anaesthesia, hypothermia occurs due to decreased basal metabolic rate and muscular activity, exposure to cold surfaces, and extra thermal energy needed to heat and humidify cold and dry anaesthetic gases moving through the respiratory system (8). Hypothermia can also cause a decrease in heart rate, a decrease in the induction time, and a decrease in the reversal of anaesthesia. The variations of hematocrit and plasma biochemistry factors were assessed during and after anaesthesia with different inhalant gases in various avian species and other animals (9-13). The purpose of this study was to compare isoflurane and sevoflurane as two common inhalational anaesthetic gases by evaluating the physiological and behavioural effects of the gasses and to determine which one would create more satisfying anaesthesia with more rapid induction and recovery times for pigeons.

2 Methods and Materials

2.1 Animals

This study was approved by the Animal Ethics and Use Committee of the Faculty of Veterinary Medicine of Tehran University (No. 013/2015). Twenty domestic male mature pigeons were obtained from a private breeder and kept in similar conditions for one week before anaesthesia was used. The birds were fed a commercial pigeon diet and tap water *ad libitum*. Pigeons had a mean ± SD weight of 342 ± 0.3 g. Routine clinical check-ups by a veterinarian during the experiment duration revealed no signs of malnutrition, dehydration, or any other disorder. All birds were determined to be healthy by physical examination, haematology, and plasma biochemistry profile results before the beginning of the study.

2.2 Experimental design

The pigeons were off-feed for 24 hours pre-anaesthesia and were randomly assigned to two treatment groups. Each of the groups, including ten pigeons, received isoflurane (Piramal healthcare®, Telangana, India) and sevoflurane (Piramal healthcare®, Telangana, India) separately with specific concentrations, and they were intubated with 12-F Cole endotracheal tube (Ruschelit®, Willy Rusch AG, Kermen, Germany). We used an isoflurane vaporiser (Everest Veterinary Anaesthetic Vaporizer, England) and a sevoflurane vaporiser (Vip3000; Matrix, Orchard Park, NY, USA), and McKinley Anaesthesia WorkStation (McKinley ONE®, Everest Veterinary Technology, England). This project was confirmed by the Animal Care and Use Committee of Ferdowsi University in Mashhad, Iran.

2.3 Experimental study

General anaesthesia was induced for every pigeon with 5% isoflurane and 6% sevoflurane concentration and in 100% oxygen at a rate of 0.7 L minute-1 released by mask with a vaporiser. Afterwards, the anaesthesia procedure was maintained at the concentration of 2% of each gas with 100% oxygen for 30 minutes. During the experiment, every pigeon was positioned in dorsal recumbency on cotton cloth by keeping their extended wings to the table with tape and covering the birds in a flow of warm-water blanket (TP-500 T/Pump Heat Therapy Pump, Gaymar Industries Inc, Orchard Park, NY, USA) set at 40ºC (104ºF) to provide conductive heat. The McKinley Anaesthesia WorkStation (McKinley ONE®, Everest Veterinary Technology, England) is an inhalant anaesthetic delivery system that utilises a circle system for delivery and is designed for application in animals weighing 50 g to 60 kg. The McKinley ONE system was adjusted based on the manufacturer's instructions.

2.4 Physiological alterations and designation of induction and recovery specifications

Every pigeon was kept under surgical plane anaesthesia with negative pedal withdrawal reflex, palpebral reflex, jaw tone, and wing tone. The corneal reflex was maintained, and pupils remained constricted. The presence of these reactions and plucked feathers on the ventral abdomen was used to evaluate the level of anaesthesia. Time to induction was described as the time from the primary transfer of gaseous anaesthesia up to a medium rate of anaesthesia. This was attained as specified by observing desirable muscle relaxation and no voluntary blinking. Recovery time was described as the time from terminating gaseous anaesthesia until the bird could hold its head erect while unassisted (14).

Following termination of anaesthesia, the birds were administered 100% oxygen via an endotracheal tube for 1 minute. Heart rate (HR), respiratory rate (RR), peripheral capillary oxygen saturation (SpO2), and body temperature (BT) were measured with an electronic probe (Dixtal 2010, Dixtal, Manaus, Amazonas 69077-000, Brazil) placed on the basilic vein at different times, including before anaesthesia, for obtaining the baseline values simultaneously after the termination of anaesthesia and 24 h post-anaesthesia.

2.5 Evaluation of hematocrit and plasma chemistry

The basilic vein obtained blood samples (1 ml) from venipuncture. Blood sampling was performed before the birds were anaesthetised (baseline), at the time of anaesthesia termination, and 24 hours post-anaesthesia. The blood specimens were transferred into a lithium heparin tube (Mirotainer®, Becton Dickinson). After being transferred to the laboratory, they were kept on ice for 2-3 minutes at room temperature. The hematocrit (HCT) was measured by centrifugation of whole blood at 2000 g for 5 minutes. An Automated Clinical Chemistry Auto-analyzer (Roche Hitachi 912, Kyoto, Japan) measured plasma chemistry values within 30 minutes. This included the following nine parameters: plasma urea nitrogen (BUN), uric acid (UA), alanine aminotransferase (ALT), aspartate aminotransferase (AST), creatine phosphokinase (CPK), albumin (ALB), total protein (TP), triglyceride (TG) and cholesterol (CHO).

2.6 Statistical analysis

Results were evaluated by one-way analysis of variance for repeated measures to compare time-related variables within two anaesthetic groups. Time effect, time ×group reciprocal effect, and zero time sampling (before anaesthesia) were considered covariates. When the time ×group reciprocal effect was significant, the comparison was made for each time between the two groups using the Nonparametric Mann-Whitney U test because the data were not normally distributed. Differences were considered to be significant at $P < 0.05$.

3 Results

3.1 Induction and recovery from anaesthesia

All pigeons did not scramble during the induction period with each anaesthetic inhalant gas, nor did we notice any unfavourable responses during the intubation. The statistical analysis indicated that IT and RT were significantly longer in the sevoflurane and isoflurane groups, respectively ($P<0.05$). This finding demonstrated that the pigeons were anaesthetised faster by isoflurane and recovered faster from anaesthesia with sevoflurane (Figure 1 and Figure 2). Indeed, no significant difference was observed between the two anaesthetic inhalant agents regarding RT, not IT.

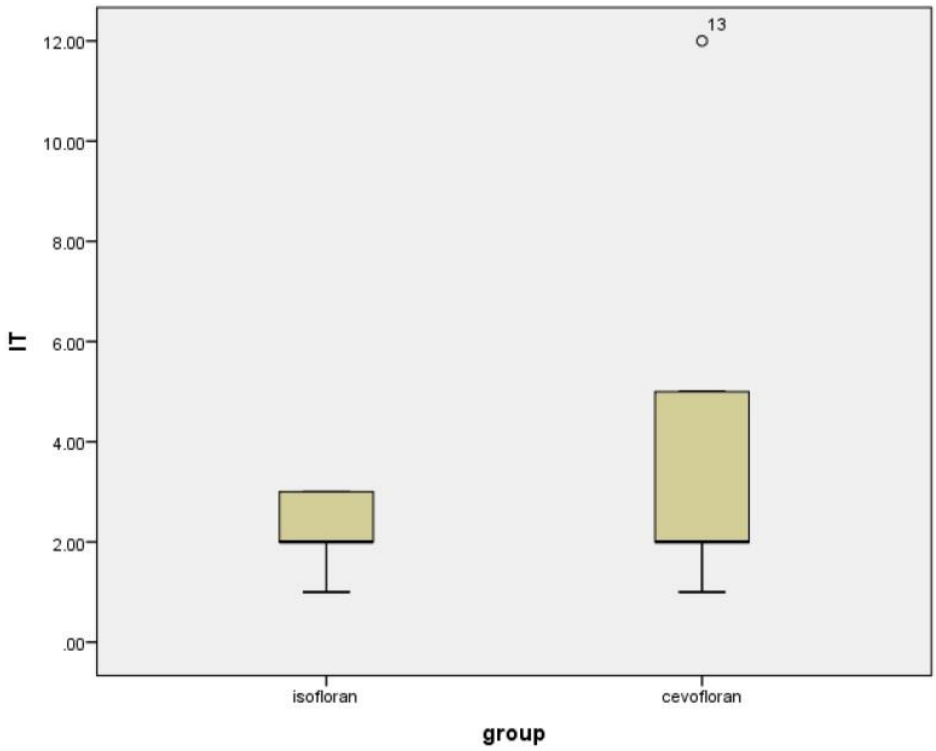

Figure 1. Induction time from isoflurane and sevoflurane anaesthesia in pigeons. Each column represents the mean. IT median of 2 min is similar in both groups. One sample in the sevoflurane group showed the longer IT of 5 min; n = 10 for each group.

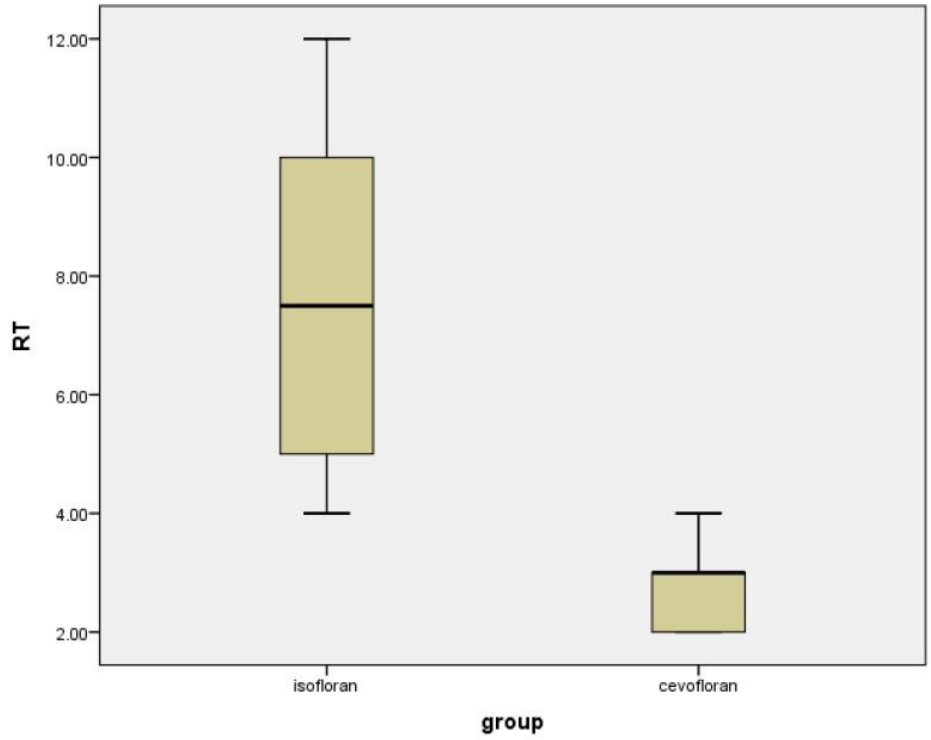

Figure 2. Recovery time from isoflurane and sevoflurane anaesthesia in pigeons. Each column represents the mean; a considerable difference is observed in RT between the isoflurane and sevoflurane groups with medians of 7 and 3 min, respectively. n = 10 for each group.

3.2 *Variations in the physiological effects of the two anaesthetics*

According to the findings, both sevoflurane and isoflurane reduced HR. In both groups, with simultaneous cessation of anaesthesia (after 30 minutes), HR decreased by the same amount in both groups. HR also returned to normal within 24 hours post-anaesthesia (Figure 3). This study demonstrated that RR declined in both groups during anaesthesia. However, the decrease caused by sevoflurane was more dramatic than isoflurane. Furthermore, RR returned to normal 24 h post-anaesthesia in the isoflurane group, while in the sevoflurane group, it did not return to the initial state despite the increasing trend. Moreover, in three subjects treated with sevoflurane, a decline in respiratory rate was observed 24 hours post-anaesthesia (Figure 4).

Regarding the impact of the two anaesthetics on BT, we observed its decline with both anaesthetic agents. BT was measured after the cessation of anaesthesia and returned to normal 24 hours post-anaesthesia. A more remarkable decrease was observed in the sevoflurane group (Figure 5). Results indicated that the SpO2 percentage remained stable, between 91 to 93 percentages, without any changes at all three stages.

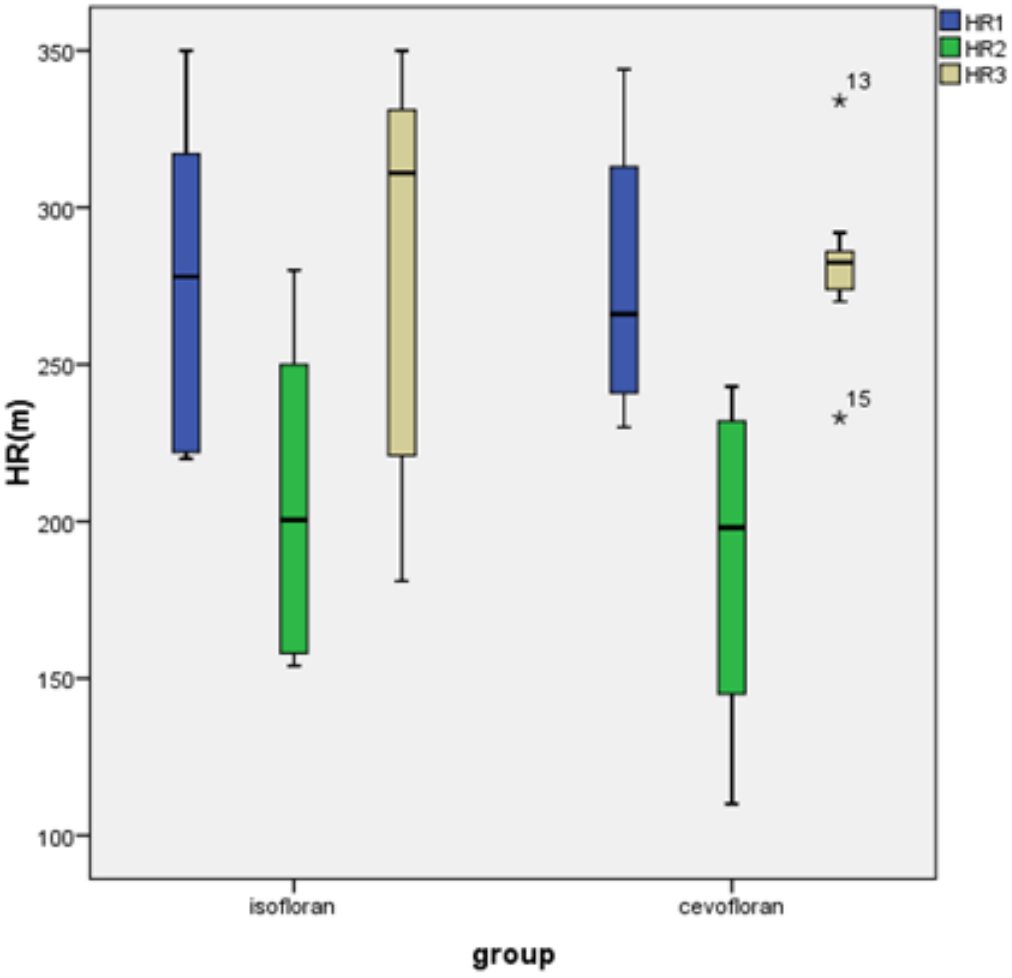

Figure 3. Comparison of the changes in heart rate (HR) (median and percentiles) between the two groups of isoflurane and sevoflurane during the study (blue: before anaesthesia induction, green: simultaneous with anaesthetic cessation, yellow: 24 h post-anaesthesia).

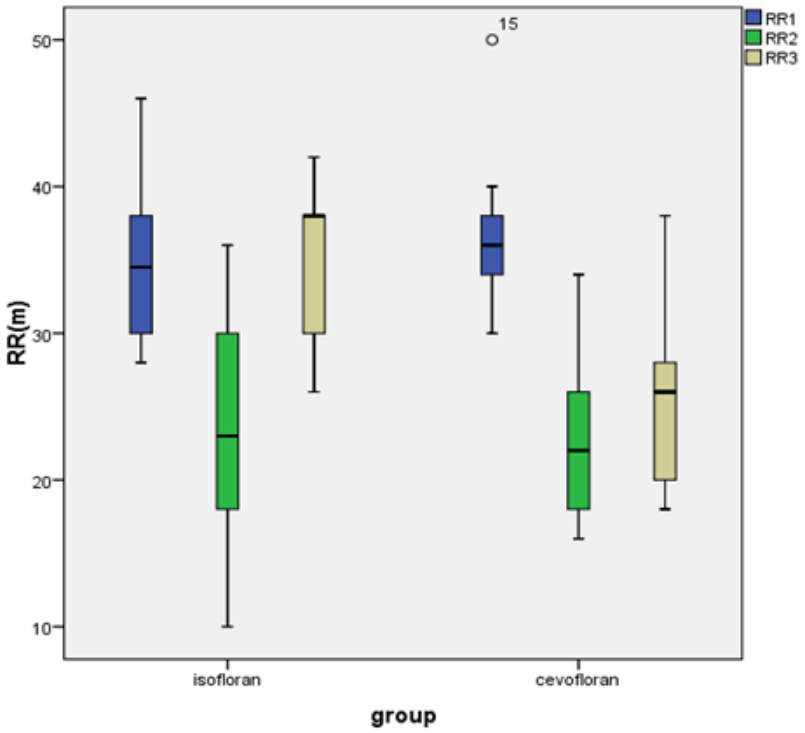

Figure 4. Comparison of the changes in respiratory rate (RR) (median and percentiles) between the two groups of isoflurane and sevoflurane during the study (blue: before anaesthesia induction, green: simultaneous with anaesthetic cessation, yellow: 24 h post-cessation).

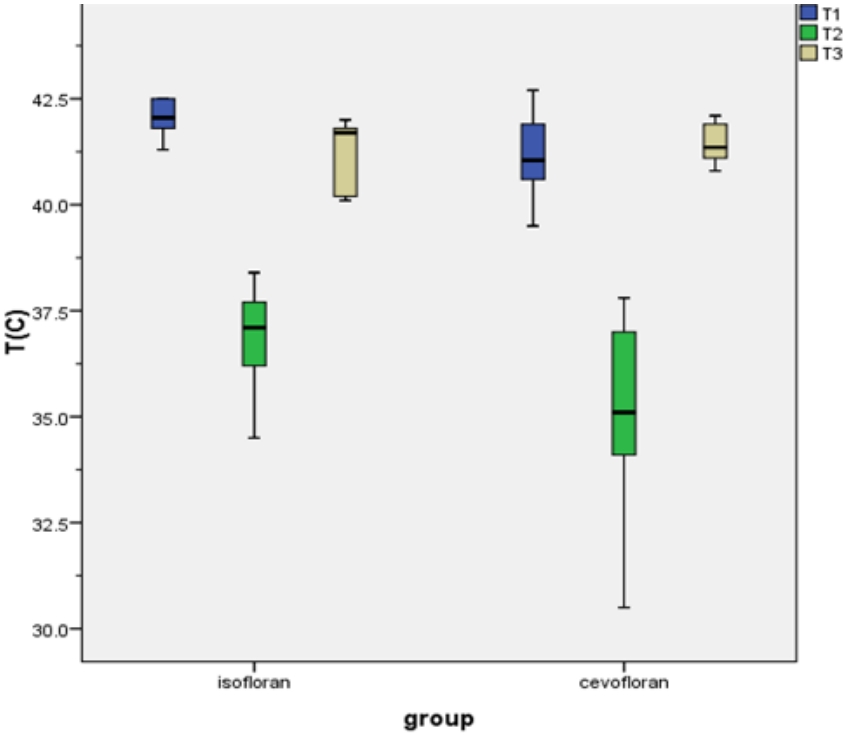

Figure 5. Comparison of the changes in body temperature (BT) (median and percentiles) between the two groups of isoflurane and sevoflurane during the study (blue: before anaesthesia induction, green: simultaneous with anaesthetic cessation, yellow: 24 h post-cessation).

3.3 HCT and plasma biochemical value findings

Comparisons demonstrated that at all three sampling times, significant differences were observed between the two treatment groups regarding HCT, ALT, AST, BUN, CPK, TG, CHOL, UA, TP, and ALB (P<0.05). The impact showed a decreasing trend toward ALT, AST, TG, CHO, UA, TP, and HCT. In contrast, CPK activity increased during anaesthesia with both anaesthetics and the ALB level was unaffected in both groups. It should be noted that ALB concentration increased unexpectedly in the third sampling time (24 h post-cessation of anaesthesia) for the isoflurane group, but it was unchanged at other sampling stages. A significant difference was only observed for BUN levels between pre-anaesthesia and cessation of anaesthesia (P<0.05). In contrast, CPK variation significantly differed between the two treatment groups at only 24 h post-cessation of anaesthesia (P<0.05). There was a significant difference between the two groups at all three sampling times for the other remaining factors.

4 Discussion

This study provides information about variations in the induction and recovery time of anaesthesia, its physiological effects, and HCT and chemistry profiles in pigeons anaesthetised with isoflurane and sevoflurane. Both inhalation anaesthetic gases created a smooth, rapid, and uneventful induction and recovery from anaesthesia in pigeons, consistent with previous investigations in pigeons, bald eagles, chickens, and psittacines (15-17). Concerning IT and RT, our results revealed that IT was similar in both groups, with a median of two minutes. The median RT of three minutes for the sevoflurane group was significantly lower than the isoflurane group with seven minutes, suggesting that sevoflurane allows for a faster recovery. In this study, induction time was more rapid with isoflurane. However, this difference was insignificant, in contrast to previous studies that reported a shorter anaesthesia induction time with sevoflurane (5, 12). They also reported significantly shorter recovery times with sevoflurane anaesthesia in eagles. This agrees with our study in which we observed a faster recovery time with sevoflurane. The pigeons had an excitatory response as indicated by uncoordinated wing flapping during recovery; however, this excitatory response was minimal overall. In a study, the researchers reported no significant difference in recovery time between sevoflurane and isoflurane in psittacine birds; the birds seemed to become alert faster and were less ataxic with sevoflurane (15). In one current study, the increased frequency of activity observed in eagles during recovery from sevoflurane versus isoflurane may be related to the shortened recovery time. A faster recovery time may be favourable following extensive anaesthetic periods or in debilitated pigeons. In another study performed on red-tailed hawks, though, the difference in IT and RT between isoflurane, sevoflurane, and desflurane was insignificant (3). The time required for the hawks to track a moving object post-anaesthesia was remarkably shorter when the anaesthesia was performed with sevoflurane and desflurane than with isoflurane. These results are in line with the current study. In our study, during the thirty minutes of anaesthesia, we did not observe morbidity or mortality.

Apnea has been reported in psittacines, cranes, and waterfowl (18, 19). In several birds, particularly waterfowl, periods of apnea and bradycardia can happen at the time of induction of anaesthesia because of a physiological response termed a dive response (18). We did not observe any apnea or dive response in our pigeons at 5% isoflurane and 6% sevoflurane concentrations.

Inhalant anaesthetics are familiar for the induction of dose-dependent respiratory suppression in all species. The avian respiratory system is anatomically and physiologically different compared with the mammalian system. According to this study's findings, sevoflurane and isoflurane reduced HR. This agrees with previous investigations in which there was a significant decrease of HR during anaesthesia with isoflurane and sevoflurane in bald eagles, crested serpent eagles, common buzzards, and thick-billed parrots (5, 12, 20, 21). No significant reduction in HR was demonstrated in several studies conducted in crested caracaras and pigeons (17). Botman *et al.* (2016) reported no significant decrease in HR from the wake to the sleep state under isoflurane in pigeons (17). This contradiction with our study may be due to different concentrations of isoflurane at induction and maintenance and the difference in significant levels that were used for the two studies (P<0.001).

According to the study conducted by Chan *et al.* (2013) on the effects of isoflurane and sevoflurane in crested serpent eagles, the researchers concluded that no significant differences in HR were observed in the two groups (12). However, their findings appear to be contrary to our results; they administered concentrations of 1.46% and 2.03% of isoflurane and sevoflurane, respectively, and maintained a 3% concentration. Also, they used a lower dose of sedatives in the induction stage compared to the current study;

therefore, all these reasons may cause contradictory HR variation.

Before anaesthesia was administered to alert the mode of pigeons, a baseline respiratory rate of 30 breaths per minute was reported (17). A similar value was observed in this study (isoflurane group: 29-39 breaths per minute and sevoflurane group: 33-38 breaths per minute). The *P* value differed significantly between the two groups at all three sampling stages, in contrast with Chan *et al*. (2013) study in which the degree of respiratory depression during sevoflurane anaesthesia did not significantly differ from that of isoflurane anaesthesia (12). Both isoflurane and sevoflurane led to a decline in RR in pigeons with significant differences. Our findings were in accordance with former investigations in bald eagles, red-tail hawks, crested serpent eagles, crested caracaras, goats, sheep, and cats (3, 12, 21-25). In a similar study, the authors reported no decrease in the respiratory rate of pigeons anaesthetised with isoflurane (17), and their findings contrast with ours. The reason may be due to the dose-dependent manner of isoflurane as a ventilation depressor, as described in a previous study (26). This could explain the difference between two similar studies.

The optimum body temperature of an avian is between 40°C and 44.4°C (104ºF-111.92ºF) (27). Throughout the time of avian anaesthesia, a heat supplement is recommended to compensate for the reduction of body temperature over time. In the current study, despite a supplemental heat source, BT values of the pigeons progressively decreased with both inhalation and anaesthetic agents. This finding is in relationship with previous studies (5, 12, 21, 23, 27, 28). Before anaesthesia, BT in both groups was between 41°C to 42°C (105.8ºF-107.6ºF). During anaesthesia, this value significantly decreased. The isoflurane group was maintained at 36°C (96.8ºF), and the sevoflurane group was maintained at 33°C-37°C (91.4ºF-98.6ºF). This variation exceeded more than 2°C during the period of anaesthesia. This is in contrast with the other similar study performed on pigeons. The researchers did not observe large temperature differences while using isoflurane anaesthesia, so that this difference could be due to diverse concentrations of this gas and heat management protocol during anaesthesia. We discovered that pigeons under isoflurane had a significantly higher BT than those under sevoflurane. Chan's study found that crested serpent eagles under sevoflurane anaesthesia produced a significantly higher BT than those in the isoflurane treatment group (12). In addition, Granone *et al*. (2012) demonstrated hypothermia during anaesthesia, but insignificant differences in BT were found in red-tailed hawks following the application of isoflurane, sevoflurane, and desflurane (3). Consistent with our study, the decreasing trend of BT in pigeons during sevoflurane and isoflurane anaesthesia was similar to previous reports in crested caracaras, cockatoos, and bald eagles under isoflurane or sevoflurane anaesthesia (5, 27, 29). The authors concluded that none of the anaesthetic delivery systems suppressed a remarkable reduction in the core BT of pigeons. It was maintained at the same range as in our study (30).

In the current study, the effects of anaesthesia by isoflurane and sevoflurane on HCT, as well as biochemical markers, including ALT, AST, BUN, UA, TG, CHOL, TP, CPK, and ALB were evaluated for most factors. The impact demonstrated a decreasing trend. In contrast with our study, Chan *et al.* (2013) did not report any changes in HCT and biochemical factors, including AST, ALT, CPK, CHO, and BUN in the crested serpent eagle. Notably, the latter study measured the aforementioned factors one-hour post-anaesthesia. Consequently, the discrepancy in results could be attributed to the differences in the species and the delay, which allowed sufficient time to eliminate the possible induced alterations.

Similar to our study, studies performed on sheep and goats found that HCT was decreased in groups under isoflurane and sevoflurane anaesthesia. Their result revealed no significant change in AST, ALT, BUN, and CPK (9, 22). Serum biochemical values such as TP, CHO, ALB, BUN, and UA evaluated in ostriches anaesthetised with isoflurane revealed no significant difference. In the present study, ALB was only unaffected, and the concentration of the rest of the biochemical markers was changed. In addition, our results regarding SpO2 percentage indicated that pulse oximeter readings were from 93% to 96%. This finding is similar to the previous studies (3, 12).

5 Conclusion

According to the findings of this study, isoflurane and sevoflurane created a smooth, rapid induction and recovery from anaesthesia in pigeons. However, anaesthesia with sevoflurane was shown to have a shorter RT than isoflurane. The two anaesthetics were not significantly different regarding IT. Biochemical markers returned to normal at 24 h post sevoflurane anaesthesia, but the values did not completely return to baseline levels after 24 h post isoflurane anaesthesia. The cardiopulmonary effects of sevoflurane were in accordance with those of isoflurane. Values related

to the HCT and plasma biochemistry panel following sevoflurane usage did not vary from those of isoflurane. As a result, considering the evaluated effects of these two anaesthetic agents, sevoflurane is recommended for administering anaesthesia in pigeons.

Conflict of Interest

The authors declare that they have no conflict of interest.

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Author Contributions

M.R. Emami and M. Mohri coordinated the study, F. Noori and P. Nakhaee provided a draft manuscript, and all authors contributed to approving the final manuscript.

Data Availability Statement

Data are available from the first author upon reasonable request.

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