



Anaesthetic and Cardiopulmonary Effects of Dexmedetomidine, Buprenorphine, Dexmedetomidine – Buprenorphine on Alfaxalone in Adult Rabbits – A comparative Study



CrossMark

Sogebi, Emmanuella and Cliff, Ann Iyabode

Department of Veterinary Medicine and Surgery, College of Veterinary Medicine, Federal University of Agriculture, Alabata, Nigeria.

ALFXALONE is a neuroactive steroid used as a general anaesthetic in several species including dogs, cats, ferrets and rabbits. This study evaluates the anaesthetic, analgesic, sedative and cardiopulmonary effects of Alfaxalone (alfaxan®) constant rate of infusion in Dexmedetomidine (Dexdomitor®) and buprenorphine premedicated rabbits. A blinded, randomized crossover study with a two - week washout period was carried out on six adult mixed breed rabbits with an average weight of 2.0 ± 0.5 kg. Each rabbit received a dose of Dexdomitor® (0.3 mgkg⁻¹), buprenorphine (0.1 mgkg⁻¹) or Dexdomitor® and buprenorphine as premedicants intramuscularly (IM), induced with a loading dose of 4mgkg⁻¹ and maintained with constant rate infusion of alfaxan at 9 mgkg⁻¹hr⁻¹ for an hour. Blood glucose levels were measured prior to and after treatments. Anaesthetic indices (duration and quality of anaesthesia, analgesia and the quality of sedation) were recorded. Cardiorespiratory variables (heart rate, respiratory rate, pulse rate, peripheral capillary oxygen saturation) and rectal temperature were measured at 0, thereafter 15, 30, 45 and 60 minutes after treatment. Data obtained were expressed as Mean \pm SEM, and subjected to ANOVA. Anaesthetic indices and blood glucose levels were analyzed with student's t- test. $P \leq 0.05$ was considered significant. There were no significant differences between the groups in the pulse rate but a level of significance within the groups over time in some cardiopulmonary parameters. Dexdomitor® improved the anaesthetic and analgesic potentials of alfaxalone and produced more stable cardiopulmonary effects compared to the buprenorphine- alfaxan® and Dexdomitor®-buprenorphine-alfaxan® combinations.

Keywords: Anaesthetic, Analgesic, CRI alfaxalone, Rabbits, Sedative.

Introduction

General anaesthesia (from Greek *an-*, “without”; and *αἴσθησις*, *aisthēsis*, “sensation”), traditionally meant the condition of having sensation (including the feeling of pain) blocked or temporarily taken away [1]. The triad of general anaesthesia includes analgesia, paralysis (muscle relaxation) and unconsciousness [1,2]. However, no single agent can accomplish these hence, polypharmacy is

central to the practice of anaesthesia in both humans and animals [3]. The three main phases of general anaesthesia are the induction phase, the maintenance phase, and the recovery phase. The induction of general anaesthesia is commonly accomplished with a primary injectable induction agent (such as propofol or alfaxalone) alone or in combination with co-induction agents such as a benzodiazepine, lidocaine, or an opioid bolus [4].

Alfaxalone is commonly used in veterinary

*Corresponding author : Sogebi, Emmanuella, E.mail: eaosogebi@gmail.com, Tel. 08034481316

(Received 27/04/2020, accepted 12/07/2020)

DOI. 10.21608/ejvs.2020.28770.1170

©2020 National Information and Documentation Center (NIDOC)

practice as an induction agent. Though it is relatively more expensive than other induction agents, it is often preferred due to the lack of depressive effects on the cardiovascular system [5]. The most common side effect is respiratory depression. A new formulation of alfaxalone has been employed producing light anaesthesia lasting between 30 minutes in rabbits and ferrets, to few hours in cats and dogs, depending on the dose employed, and fast recovery with minimal or limited cardiovascular effects [6-11]. However, a lack of analgesia was observed, and analgesic support should be provided in painful procedures [12]. Moreover, in rabbit, the poor degree of analgesia and the respiratory depression that is produced at high dose rates limit the usefulness of this drug [13].

Premedication (administering sedative, analgesia drugs prior to induction) increases the potency of alfaxalone as an induction agent [14]. Pre-anaesthetic drugs include alpha-2 adrenergic agonists (xylazine, medetomidine, dexmedetomidine), opioids (buprenorphine, butorphanol, pentazocine) and benzodiazepines (diazepam, midazolam) [15, 16].

Constant rate infusions (CRIs) of different drugs can provide analgesia and increase anaesthetic depth during surgical interventions, reducing the requirement for inhalant anaesthetics [17]. Controlled intravenous administration of alfaxalone offers a wide safety margin and an excellent cardiovascular profile, making it an excellent choice for anaesthetic induction and maintenance of rabbits [16]. This study compared the constant rate infusion (CRI) of alfaxalone with other medications for sedation and anaesthesia in adult rabbits.

Materials and Methods

The study was approved by the Ethics Committee (FUNAAB/COLVET/CREC/011/19) of the College of Veterinary Medicine, Federal University of Agriculture, Abeokuta (FUNAAB), Nigeria.

Experimental animals

Six adult, mixed breed, rabbits of both sexes (three males and three females) with an average weight of 2.0 ± 0.5 kg were obtained locally and were housed individually in locally fabricated cages made from wire mesh and wood. A commercially formulated pelleted ration and water were given *ad libitum*. All animals were considered healthy based on physical examination and complete full blood count.

Experimental design

In a prospective controlled crossover blinded study, drugs were administered as premedicants on six rabbits in three protocols, with a two-week 'washout' interval between the protocols. The six rabbits received 0.3 mgkg^{-1} Dexdomitor® (DEX) (Orion Pharma Ltd, Bangladesh) – P. I, 0.1 mgkg^{-1} Buprenorphine (BUP) (Dechra Pharma, USA) - P. II, a combination of 0.3 mgkg^{-1} Dexdomitor® and 0.1 mgkg^{-1} Buprenorphine (DEX + BUP) – P. III. The drugs were administered in separate syringes at five minutes interval intramuscularly (IM). Induction was achieved with a loading dose of intramuscular injection of 4 mgkg^{-1} Alfaxan® (ALFA) (Jurox Limited, West Sussex, UK) and maintained with a constant rate infusion (CRI) at $9 \text{ mgkg}^{-1} \text{ hr}^{-1}$ for an hour using a syringe pump (PractiVet®, Phoenix). Venous access was achieved through the auricular vein and lactated ringer's solution (LRS) was instituted at 5 ml/kg/hr to maintain homeostasis during anaesthesia using an infusion pump (Hawkmed™, Wellkang Ltd., China). The CRI of Alfaxan® was sustained for one hour and all rabbits that received Dexdomitor® and Dexdomitor® combination received intramuscular injection of 0.15 mgkg^{-1} Antipam® (Dechra, The Netherlands) after one hour as reversal agent.

Blood glucose levels were measured prior to (0 minute) and after treatments using a glucometer (On Call Plus, ACON Labs, USA). Anaesthetic indices (duration and quality of anaesthesia, analgesia and quality of sedation) were determined. The quality of sedation was assessed using the Inova Sedation Scale (ISS). The noxious stimulus was performed by pinching the web of the skin between the toes with forceps every 5 minutes post treatment to determine both the onset and duration of analgesia. Movement of the hind limb was considered as a negative response and lack of response as positive.

Cardiorespiratory variables [heart rate, respiratory rate, pulse rate, peripheral capillary oxygen saturation (SpO_2)] and rectal temperature of each rabbit were monitored individually and measurements were taken at 0, 15, 30, 45 and 60 minutes post treatment using the multi-parameter patient monitor (General Meditech Inc., China), and charted on the anaesthetic sheet. The monitoring was continued until full recovery from anaesthesia.

Data analysis

Data obtained were expressed as mean \pm Standard Error of Mean (SEM), and comparisons within treatments made using repeated-measures Analysis of Variance (ANOVA). Anaesthetic indices and glucose levels were analyzed using Student's t-test and nonparametric data analyzed using the Mann-Whitney U-test.

Results

General observations

Four animals recovered fully from anaesthesia and were included in the data analysis. Two rabbits had bilateral bulgy eyeballs (exophthalmos) few minutes into anaesthesia in P II (Fig. 1).



Fig. 1. One of the rabbits with the bulgy eyeball in P. II.

Anaesthetic responses

Duration of anaesthesia was significantly longer, 65.0 ± 1.5 minutes, in P. III; then, 62.0 ± 1.5 minutes in P. II, compared to protocol I, 54.0 ± 1.5 minutes. The duration of analgesia was significantly longer in P. III, 45.0 ± 5.0 minutes; then, 40.0 ± 5.0 minutes in P. I compared to P. II, 30.0 ± 5.0 minutes (Table 1).

Protocols I and II recorded a significantly decreased blood glucose values post-treatment, 73.00 ± 25.74 mg dL⁻¹ and 49.50 ± 28.50 mg dL⁻¹ respectively compared with the pre-treatment values, 134.25 ± 14.89 mg dL⁻¹ and 102.25 ± 7.17 mg dL⁻¹ respectively. There was a sharp spike in the blood glucose post-treatment in P. III (Fig. 2).

TABLE 1. Effects of Dexdomitor®-Alfaxan®, Dexdomitor®-Alfaxan® and Dexdomitor®-Buprenorphine-Alfaxan® combinations on the anaesthetic indices.

Time/ Treatment	Duration of Anaesthesia (mins)	Duration of Analgesia (mins)	Quality of Recovery
BUP + ALF	62.0 ± 1.5	30.0 ± 5.0	Moderately smooth
DEX + ALF	54.0 ± 1.5	40.0 ± 5.0	Poor
DEX + BUP + ALF	$65.0 \pm 1.5^*$	45.0 ± 5.0	Fair

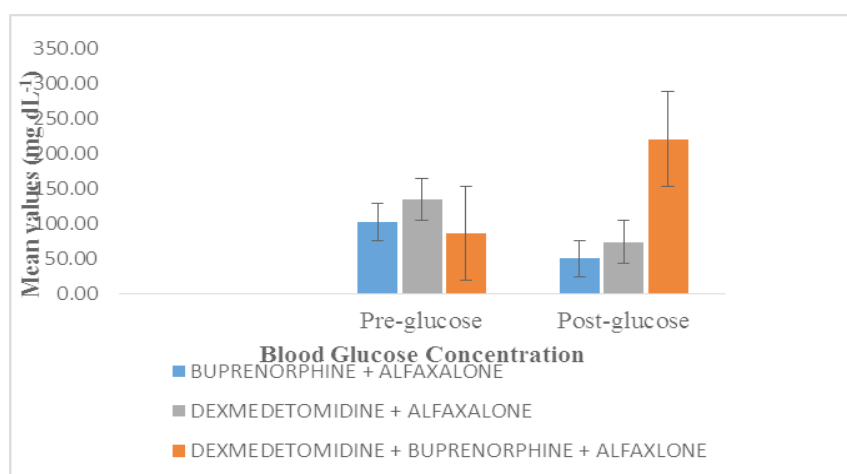


Fig. 2. Effects of Dexdomitor®-Alfaxan®, Buprenorphine-Alfaxan® and Dexdomitor®-Buprenorphine-Alfaxan® combinations on Blood glucose concentration.

The level of sedation was recorded to be deepest in protocol P. III. In P. I, rabbit (A) was observed to be alert and (C) observed to be occasionally drowsy, easy to arouse while in the P. II, the rabbit (B) was observed to be dozing intermittently (Fig. 3).

Cardiopulmonary responses

Dexdomitor®-buprenorphine-Alfaxan® combination caused a significant ($P < 0.05$) decrease in the heart rate, the respiratory rate

and rectal temperature as compared to other protocols. There were significant differences in the SpO_2 between the drug combinations, with marked ventilatory depression in P. I. There were significant differences within the P. I in the SpO_2 and in the heart rate at all times in P. III. Significant increase in the respiratory rate was exhibited within the P. III, no significant difference in the pulse rate within the protocols and slight significant increase in the rectal temperature at 0th and 15th minutes in P. III (Fig.4).

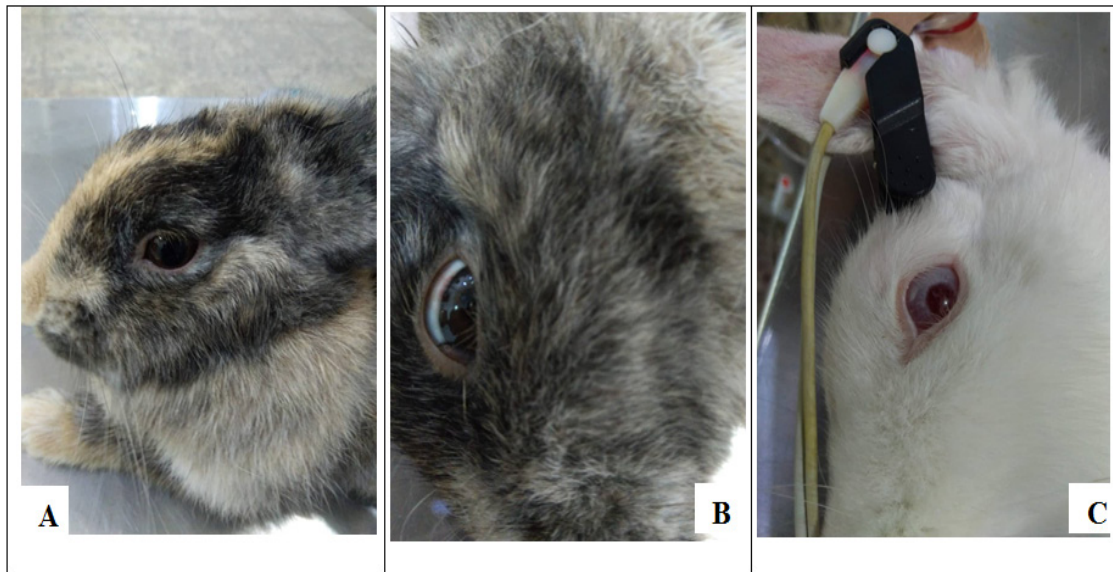


Fig. 3. The various levels of sedation in the rabbits for either of Dexdomitor®-Alfaxan®, Buprenorphine-Alfaxan® and Dexdomitor®-Buprenorphine-Alfaxan® combinations

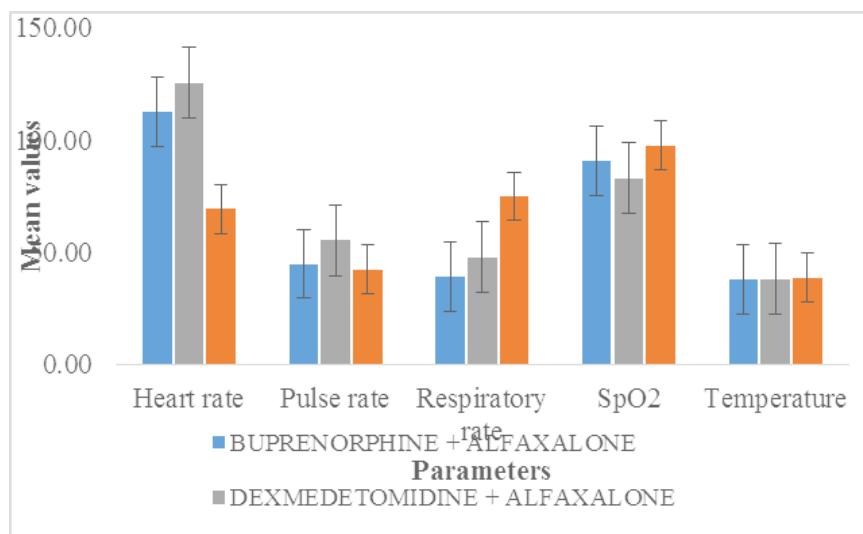


Fig. 4. Effects of Dexdomitor®-Alfaxan®, Buprenorphine-Alfaxan® and Dexdomitor®- Buprenorphine-Alfaxan® combinations on cardiopulmonary indices.

Discussion

This study has evaluated the effects of buprenorphine and Dexdomitor® premedicants on constant rate infusion (CRI) dose of Alfaxan® which was carried out on six rabbits. Beneficial anaesthetic and cardiopulmonary effects of Alfaxan® and Alfaxan®-based combinations were observed.

Although alfaxalone has no known antinociceptive properties, the administration of either Dexdomitor®, buprenorphine or a combination of both increased duration of sedation compared to the use of Alfaxan® alone and produced short-term analgesia in this study. This agrees with the findings of Navarrete-Calvo et al. [16] who stated that alfaxalone administration (though subcutaneously), produced uncomplicated sedation that may be recommended for non-painful procedures that do not require complete immobility. The addition of Dexdomitor® and buprenorphine increased the duration of sedation and immobility but did not result in general anaesthesia in the CRI dose used in this study. The administration of Dexdomitor® and combination of Dexdomitor® and buprenorphine protocol may be useful for non-painful procedures requiring extended immobility. It also agrees with the findings of Perkowski [21] who also stated that the combinations of opioids (buprenorphine) and alpha2-adrenoceptor agonists (Dexdomitor®) can lead to supra-additive or synergic antinociceptive and sedative effects in the canine species.

The shorter duration of anaesthesia observed in the second protocol could be due to the administration of the reversal agent- Atipam® after the drug (CRI) was discontinued. This agrees with Pinelas et al. [12] who stated that in the scenario of prolonged sedation, pharmacologic reversal of Dexdomitor® with atipamezole should expedite recovery. Quality of recovery was judged to be moderately smooth and no adverse reactions with the Dexdomitor® - Alfaxan® protocol. It was poor with a few adverse reactions such as shivering and staggering movements observed in the buprenorphine- Alfaxan® protocol, and fair in the Dexdomitor®-buprenorphine- Alfaxan® protocol with a few staggering. This outcome contrasts the findings by Pinelas et al. [12] who reported that in the Dexmedetomidine-treated group several animals had recoveries that were characterized by paddling, strong startle responses, and multiple attempts to right themselves. The quality of recovery is important, because recovery from

anaesthesia can be stressful for animals, and providing a smooth recovery improves overall animal welfare related to the anaesthetic event. In the Dexdomitor®-buprenorphine- Alfaxan® protocol, there was marked increase in the blood glucose concentration. This could be due to the potential side effects of dexmedetomidine which include polyuria and interference with insulin production, contributing to hyperglycemia [10].

Anaesthetic drugs are usually associated with cardiovascular and respiratory depression closely related with the drugs involved but also their interactions. These effects are a safety concern during anaesthesia. Dexdomitor®-Buprenorphine-Alfaxan® combination produced significant reduction in heart rate and this agrees with the findings by Perkowski [21] who reported that Dexdomitor® in combination with any of the opioids would result in cardiovascular depression, compared with results for administration of Dexdomitor® alone. Besides, buprenorphine, a mu opioid receptor agonist, also produces bradycardia [18]. Thus, both buprenorphine-Alfaxan® and Dexdomitor®- Buprenorphine- Alfaxan® combinations produced a marked decrease in heart rate. The combination of Dexdomitor® and Alfaxan® produced more ventilatory depression, determined by marked decrease in SpO₂. Significant increase in the respiratory rate was observed in the Dexdomitor®-buprenorphine-Alfaxan® treatment group as opposed to the cardiopulmonary depression observed with this combination as reported by Perkowski [21].

Postmortem examination was carried out on the two carcasses. The outcome of the postmortem revealed frothy exudates in the trachea. These could be caused by aspiration of gastric contents leading to obstruction of the airway, and consequently death. This agrees with the findings by Tietjen et al. [23] who reported that acute airway obstruction leading to arterial hypoxaemia may cause immediate death. Thus, prompt removal of inhaled particles, oxygenation of the patient and most importantly, prevention of further aspiration by tracheal intubation are essential for survival.

Most of the studies reported with CRI Alfaxan® are done in dogs and cats using it in combination of either of drug groups used in the study. To the best of our knowledge, this is the first report in rabbits.

Conclusion

This study has evaluated that Dexdomitor®-Alfaxan® is a more suitable drug combination in rabbit anaesthesia as it provides better analgesic, and more stable cardiopulmonary effects. Clinical studies should evaluate the feasibility of these anaesthetic combinations in surgical procedures. The effects on relevant parameters such as blood pressure or capnography should be considered to better assess the true impact of Alfaxan® and Alfaxan®-based combinations.

Acknowledgements

The authors wish to appreciate the technical support of Mr. J.O Oyela of the department of Veterinary Medicine and Surgery, College of Veterinary Medicine, Federal University of Agriculture, Abeokuta, Nigeria, before, during and after the study.

The donation of a syringe pump (PractVet®, Phoenix, USA) by Stephen Cital RVT used in conducting the study, is also appreciated. Stephen Cital is the current Chair for the Committee of Veterinary Technician Specialties, USA. This donation has assisted in making the conduct of the study possible.

References

1. Everett, T. and Tattersall, M., Maintenance of Anaesthesia., *Clinical Anaesthesia*, **15** (2), 51-55 (2014).
2. Bajwa, S.J., Bajwa, S.K. and Kaur, J., Comparison of Two Drugs Combination in Total Intravenous Anaesthetics: Propofol-Ketamine and Propofol-Fentanyl, *Saudi Journal of Anaesthesia*, **4** (2), 72-79 (2010).
3. Acktermann W., Stoicea N. and Bergese S., Polypharmacy and Perioperative Management, *Austin Journal of Anaesthesia and Analgesia*, **2** (2), 1013 (2014).
4. Mandal, M, Opioids as Co-induction Agents- the Pros and Cons., *International Journal of Critical Illness & Injury Science*, **7** (2), 77- 78 (2017).
5. Rezende, M., Reintroduced Anaesthetic Alfaxalone, *Clinician's Brief*. Retrieved August 19, 2019 (2015).
6. Gil, A.G., Silvan, G., Villa, A. and Illera, J.C., Heart and Respiratory Rates and Adrenal Response to Propofol or Alfaxalone in Rabbits, *Veterinary Record*, **170** (17), 444-448(2012). doi: 10.1136/vr.100573.
7. Giral, M., Garcia- Olmo, D.C., Gómez- Juárez, M. and Gómez de Segura, I.A., Anaesthetic Effects in the Ferret of Alfaxalone Alone and in Combination with Medetomidine or Tramadol- A Pilot Study, *Laboratory Animal*, **48** (4), 313-320 (2014).
8. Goodwin, W.A., Keates, H.L. and Pasloske, K., The Pharmacokinetics and Pharmacodynamics of the Injectable Anaesthetic Alfaxalone in the Horse. *Veterinary Anaesthesia and Analgesia*, **38**, 431-438 (2011).
9. Grint, N.J., Smith, H.E. and Senior, J.M., Clinical Evaluation of Alfaxalone in Cyclodextrin for the Induction of Anaesthesia in Rabbits, *Veterinary Record*, **163** (13), 395-396 (2008).
10. Pypendop, B.H., Ranasinghe, M.G. and Pasloske, K., Comparison of two intravenous anaesthetic infusion regimes for alfaxalone in cat, *Veterinary Anaesthesia and Analgesia*, **45** (4), 459 – 466 (2018).
11. Hunt, J.R., Goff, M., Jenkins, H., Harris, J., Knowles, T.G., Lascelles, B.D.X., Mendl, M., Whay, H.R. and Murrell, J.C., Clinical measurements performed during alfaxalone total intravenous anaesthesia for radiography and neurophysiological investigations in dogs, *Veterinary Anaesthesia and Analgesia*, **46** (4), 483-487 (2019).
12. Pinelas, R., Alibhai, H.I. and Mathis, A., Effects of Different Doses of Dexmedetomidine on Anaesthetic Induction with Alfaxalone – a Clinical Trial, *Veterinary Anaesthesia and Analgesia*, **41**, 378-385 (2014).
13. Flecknell, P.A., Special Techniques, In: Laboratory Animal Anaesthesia, 3rd edition., pp. 109-137 (2009).
14. Clarke, K.W., Trim, C.M. and Hall, L.W., Anaesthesia for Obstetrics. In: Veterinary Anaesthesia, 11th edition. Oxford: Saunders, W.B. pp. 587-598, (2014).
15. Tutunaru, A.C., Sonea, A., Drion, P., Serteyn, D. and Sandersen, C., Anaesthetic Induction with Alfaxalone may Produce Hypoxaemia in Rabbits Premedicated with Fentanyl/ Droperidol, *Veterinary Anaesthesia and Analgesia*, **40** (6), 657-659 (2013).
16. Navarrete-Calvo, R., Gómez-Villamandos, R.J., Morgaz, J., Manuel Domínguez, J., Fernández-Sarmiento, A., Muñoz-Rascón, P., López Villalba, I. and Del Mar Granados, M., Cardiorespiratory, Anaesthetic and Recovery Effects of Morphine

- Combined with Medetomidine and Alfaxalone in Rabbits, *Veterinary Record*, **174** (4), 95-99 (2014). doi: 10.1136/vr.101293.
17. De Amorim Cerejo, S., De Mattos-Jr, E., Nishimura, L.T., Quarterone, C. and Franco, L.G., Effects of Constant Rate Infusion of Anaesthetic or Analgesic drugs on General Anaesthesia with Isoflurane: A retrospective study in 200 Dogs, *Semina: Agrarian Sciences*, **34** (4), 1807-1822 (2013).
 18. Doerning, C.M., Bradley, M.P., Lester, P.A. and Nowland, M.H., Effects of Subcutaneous Alfaxalone Alone and in Combination with Dexmedetomidine and Buprenorphine in Guinea pigs (*Cavia porcellus*), *Veterinary Anaesthesia and Analgesia*, **45** (5), 658-666 (2018).
 19. Nishimura, L.T., Auckburally, A., Santilli, J., Vieira, B.H.B., Oliveira, Garcia, D.O., dos Santos Honscho, C.S. and de Mattos-Junior, E., Effects of Dexmedetomidine Combined with Commonly Administered Opioids on Clinical Variables in Dogs, *American Journal of Veterinary Research*, **79** (3), 267-275 (2018).
 20. Bradley, M.P., Doerning, C.M., Nowland, M.H. and Lester, P.A., Intramuscular Administration of Alfaxalone Alone and in Combination for Sedation and Anaesthesia of Rabbits (*Oryctolagus cuniculus*), *Journal of the American Association for Laboratory Animal Science*, **58** (2), 216-222 (2019).
 21. Perkowski, S., Sedation of the Critically Ill Patient. In: Silverstein, D.C., Hopper, K., editors. *Small Animal Critical Care Medicine*, Saunders Elsevier, St. Louis (MO). pp. 700 -704 (2009).
 22. Hu, C., Flecknell, P.A. and Liles, J.H., Fentanyl and Medetomidine Anaesthesia in the Rat and its Reversal Using Atipamezole and Either Nalbupine or Butorphanol, *Laboratory Animal*, **26**, 15-22 (1992).
 23. Tietjen, P.A., Kaner, R.J. and Quinn, C.E., Aspiration Emergencies, *Clinics in Chest Medicine*, 15:117-135 (1994).
 24. Huynh, M., Poumeyrol, S., Pignon, C., Le Teuff, G. and Zilberstein, L., Intramuscular Administration of Alfaxalone for Sedation in Rabbits, *Veterinary Record*, **176** (10), 255 (2015). doi: 10.1136/vr.102522.