LUCIA VICTORIA BEL

EVALUATION OF THE EFFICENCY OF ROCURONIUM IN GENERAL ANESTHESIA OF THE RED EARED TERRAPIN (TRACHEMYS SCRIPTA ELEGANS)

(SUMMARY OF PhD THESIS)

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INTRODUCTION

In the last years, an important number of reptiles have become pets, *Trachemys scripta elegans* being probably one of the most common turtle kept as a new companion animal.

Physiological and anatomical characteristics of turtles, including the ability of retracting their heads, not breathing for long periods of time and other particularities such as cardiac shunts, the portal-renal system, anaerobic metabolism and the influence of slow metabolism on substances absorption, are some of the factors that cause difficulty in the induction and maintenance of general anesthesia.

It has been proven that neuromuscular blocking agents can determine immobilization for endotracheal intubation in the case of *Podocnemis expansa* (Bosso et al., 2009) and *Terrapene carolina major* (Kaufman et al., 2003) turtles, with the advantage that these substances can be pharmacologically antagonized. On the other hand, an anesthetic protocol using just a neuromuscular blocking agent, could not be used for surgical procedures, due to the fact that these substances do not determine analgesia, thus the necessity of elaborating new anesthetic protocols, for use both in handling these species and performing surgical procedures.

1. GOAL AND STRUCTURE OF THE PhD THESIS

Being that, as far as we are aware, the nondepolarising blocking agent Rocuronium has not been previously used in *Trachemys scripta*
elegans, the goal of the PhD thesis was to elaborate an anesthetic protocol that would include this substance.

The objectives of the thesis were:

- To identify an efficient dose of Rocuronium for endotracheal intubation in *Trachemys scripta elegans*;
- To evaluate the efficiency of Neostigmine, a product used in different mammals as an reversal agent of the neuromuscular blocking agent Rocuronium in *Trachemys scripta elegans*;
- To identify with the ASTRUP method, changes in venous blood gas and acid base values determined by general anesthesia with Rocuronium;
- To elaborate an anesthetic protocol for surgical procedures using Rocuronium for induction, Sevoflurane for maintenance and Tramadol for analgesia.

The PhD thesis has 6 chapters, starting with the bibliographical study. Chapters 2, 3, 4, and 5 describe original research regarding the efficiency of Rocuronium in general anesthesia of the red eared terrapin (*Trachemys scripta elegans*) and chapter 6 presents original data regarding the evaluation on clinical cases of a new anesthetic protocol for use in surgical procedures.

2. RESULTS

The objective of chapter 2, „Evaluation of the effects of rocuronium in the induction of anesthesia of *Trachemys scripta*
*Trachemys scripta elegans* was to identify the dose of Rocuronium that would determine a neuromuscular block for endotracheal intubation, by evaluating 4 different protocols, using 0.25 mg/kg, 0.3 mg/kg, 0.4 mg/kg and 0.5 mg/kg of the agent. Each group was represented by 6 turtles of the subspecies.

In the case of group A (dose 0.25 mg/kg), 17% of the individuals lost their palpebral reflex, with no possibility of intubation. In the case of group B (dose 0.3 mg/kg) 67% of the individual lost their palpebral reflex and 50% could be intubated, in the case of group C (dose 0.4 mg/kg), although 100% of the individuals lost their palpebral reflex, only 83% could be intubated. In the case of group D (dose 0.5 mg/kg) 100% of the individuals lost the palpebral reflex and they could all be intubated.

Following the scores of T1 (time from Rocuroniumului administration to loss of palpebral reflex), analyzed with the Kruskal-Wallis H test, statistic relevance was seen between groups A and B (p=0.02, $\chi^2 =5.28$), A and C (p=0.001, $\chi^2 =9.65$), A and D (p=0.001, $\chi^2 =9.90$), B and D (p=0.029) and B and C (p=0.04), without any significance between group C and D (p=0.2). For these two groups T4 (time form Rocuronium administration to intubation) was statistically relevant, with p=0.02 and $\chi^2 =5.25$. For this analysis, only these two groups were taken into account, due to the fact that only in the case of these individuals 100% lost their palpebral reflex.

The objective of chapter 3, “Evaluation of the reversal effect of neostigmine in NMB anesthesia *Trachemys scripta elegans*” was to determine weather or not Neostigmine could be used as a reversing agent for the neuromuscular blocking agent Rocuronium. For this purpose, 14 turtles were divided into two groups, one received Neostigmine, the other
received Saline, as placebo. To monitor the neuromuscular block and to assess the timing of the administration of Neostigmine/NaCl 0.9%, the train of four monitoring method was used (TOF).

Mean time to return of the palpebral reflex from the administration of Neostigmine/Placebo was 24.71±9.19 minutes (SD) for the Neostigmine group, compared to 72.71±41.51 minutes (SD) for the saline group (p=0.001 and χ² =9.8), mean time to detubation from the administration of Neostigmine/Placebo was 20.71±8.42 minutes for the Neostigmine group, compared to 48.71±38.51 minutes for the saline group (p=0.02 and χ² =5.0). Mean time to recovery from the administration of Neostigmine/Placebo was 38.00±7.87 minutes for the Neostigmine group, compared to 87.14±40.80 minutes for the saline group (p=0.001 and χ² =9.8).

Mean time to recovery from Neostigmine/Placebo administration was 49.14 minutes shorter in the case of the Neostigmine group, with the duration of the anesthetic protocol itself being 53.57 minutes shorter.

The objective of chapter 4, „Blood gas and acid-base evaluation in Trachemys scripta elegans anesthetised with rocuronium”, was to identify possible alteration in blood gas and acid-base values due to the anesthetic protocol. Venous blood gas samples were collected from the central venous plexus, from 15 turtles under general anesthesia with rocuronium, at 3 different moments, and analyzed with the ASTRUP method.

In the case of PO₂ and PCO₂, M0 and M1, M0 and M2 respectively, were statistically relevant for PO₂ χ² =13.34, p=0.0003 (M0-M1), χ² =13.19, p=0.0003 (M0-M2), for PCO₂ χ² =5.39, p=0.02 (M0-
M1), \( \chi^2 =4.82, p=0.02 \) (M0-M2), with no relevance at M1-M2. Mean pH at M0 was 7.54±0.13, at M1, 7.62±0.1, at M2 7.66±0.05, statistically relevant T0-T2 (p=0.03, \( \chi^2 = 4.59 \)).

Based on the Colton scale, at M0, there was good and very good correlation (p<0.05, r between 0.5-0.75 and 0.75-1, valid for negative values also) between pH and PCO\(_2\) (r=-0.86), pH and HCO\(_3\) (r=0.6), pH and Lactate (r=-0.81) and PO\(_2\) and SO\(_2\) (r=0.78); at M1 between pH and PCO\(_2\) (r=-0.78), pH and PO\(_2\) (r=0.52), pH and HCO\(_3\) (r=0.77), pH and SO\(_2\) (r=0.73), PO\(_2\) and SO\(_2\) (r=0.55), PCO\(_2\) and SO\(_2\) (r=-0.87), and at M2 between pH and HCO\(_3\) (r=0.62), PO\(_2\) and SO\(_2\) (r=0.67) and HCO\(_3\) and SO\(_2\) (r=0.62).

Due to ventilation with oxygen throughout the anesthetic protocol there was an increase in the values of SO\(_2\) and PO\(_2\) and a decrease of PCO\(_2\). At M1, the turtles use an anaerobic metabolism, thus the increase of the lactate value and the decrease of the HCO\(_3\) value, compared to M0.

Rocuronium anesthesia did not induce any alterations in Na, K, Ca and glucose.

The objective of chapter 5, „Evaluating the analgesic effect of Tramadol in Rocuronium anesthesia of Trachemys scripta elegans” was to determine if Tramadol, administered at 5 mg/kg (n=30, 15 turtles received Tramadol, 15 saline) would provide sufficient analgesia for surgical procedures. Due to the neuromuscular block, alterations in the cardiac frequency at the time of incision were considered a clinical marker of pain.

Mean heart rate for the group that received Tramadol (23.93 ± 0.39 beats per minute) increased with 1.13±0.61 beats per minute at the time of
incision, compared to the group that received NaCl 0.9% (27.45 ± 0.63 beats per minute), as placebo, in which case it increased with 14.6±5.3 beats per minute (Fig. 1), proof to the fact that Tramadol, administered at 5 mg/kg determines analgesia in *Trachemys scripta elegans* (p<0.001, χ² =22.05).

Chapter 6, „Evaluation of a new anesthetic protocol for surgical anesthesia in *trachemys scripta ssp.“ describes 11 clinical cases of turtles that were anesthetised with Tramadol (5 mg/kg), Rocuronium (0.5 mg/kg), Atropine (0.03 mg/kg), Neostigmine (0.05 mg/kg) and Sevoflurane (3%) for different surgical procedures.

![Fig. 1. Mean heart rate (in BPM) at the time of incision](image)

**3. GENERAL CONCLUSIONS**
• From the 4 doses of Rocuronium tested for the induction of anesthesia of *Trachemys scripta elegans*, only the 0.5 mg/kg dose proved effective for endotracheal intubation of all the individuals, while lower doses proved less effective.

• Neostigmine, administered at 0.05 mg/kg proved effective as a reversal agent of Rocuronium, the time of recovery from the neuromuscular block being much shorter in the case of this group, compared to the placebo one.

• Rocuronium anesthesia did not determine alterations of Na, K, Ca, glucose, blood gas and acid-base values and did not affect the heart rate.

• Tramadol, administered at 5 mg/kg proved effective as an analgesic for surgical procedures in the anesthetic protocol using Rocuronium, Neostigmine and Sevofluran in the case of *Trachemys scripta elegans*.

**Recommended anesthetic protocol** for surgical procedures in *Trachemys scripta elegans*:

- Tramadol administered IM, 5 mg/kg;
- After 15 minutes, administration of Rocuroniumului, IM, 0.5 mg/kg;
- Endotracheal intubation;
- Maintenance with Sevofluran 3%, on IPPV, with 0.6-1 l/minute, 4-6 respirations/minute.
- Administration of Atropine, IM, 0.03 mg/kg;
• After 3 to 5 minutes, administration of Neostigmine, IM, 0.05 mg/kg;

References
