10 - ORIGINAL ARTICLE
Anesthesia

Pharmacological restraint of captivity giant Amazonian turtle *Podocnemis expansa* (Testudines, Podocnemididae) with xylazine and propofol[1]

Contenção farmacológica de tartaruga da Amazônia *Podocnemis expansa* (Testudines, Podocnemididae) de cativeiro com xilazina e propofol

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ABSTRACT
Purpose: Identify a technique to induce brief sedation and hypnosis in *Podocnemis expansa*. **Methods:** Twenty commercially bred *P. expansa*, weighing on average 1.2 ± 0.24 kg, were subjected to two protocols: G1 was given 1.5 mg/kg IM of xylazine and 5 mg/kg IV of propofol, while G2 received 1.5 mg/kg IM of xylazine and 10 mg/kg IV of propofol. The drugs were applied, respectively, in the left thoracic member and in the cervical vertebral sinus. Assessments were made of the anesthetic parameters of locomotion, muscle relaxation, response to pain stimuli in the right thoracic members, pelvic members and tail, easy handling and heartbeat, as well as ambient temperature and glycemic level. **Results:** A consistent hypnotic effect was recorded 49.6 ± 22.1 seconds in G2 and after 58.2 ± 55.1 in G1. All the animals of G2 recovered in 198 minutes, and those of G1 in 156 minutes. **Conclusion:** The hypnosis achieved with these associations was satisfactory, and G1 was as efficient as G2, allowing for the pharmacological restraint for the collection of biological samples, physical examinations and minor surgeries on these species.
Key words: Turtles. Xylazine. Propofol.

RESUMO
Objetivo: Identificar uma técnica para se induzir sedação e hipnose em *Podocnemis expansa*. **Métodos:** Vinte *Podocnemis expansa* de criatório comercial, com média de peso 1,2 ± 0,24 kg, foram submetidas a dois protocolos: G1 recebeu xilazina 1,5 mg/kg IM e propofol 5 mg/kg IV e o G2 xilazina 1,5 mg/kg IM e propofol 10 mg/kg IV. As drogas foram aplicadas no membro torácico esquerdo e no seio vertebral cervical, respectivamente. Observaram-se os parâmetros anestésicos: locomoção, relaxamento muscular, resposta aos estímulos dolorosos no membro torácico direito, nos membros pelvinos e na cauda, facilidade de manipulação e freqüência cardíaca, além da temperatura ambiental e glicemia. **Resultados:** Um efeito hipnótico consistente foi observado aos 49,6 ± 22,1 segundos no G2 e 58,2 ± 55,1 segundos no G1. A recuperação de todos os animais do G2 ocorreu em 198 minutos, e em 156 minutos no G1. **Conclusão:** A hipnose obtida por essas associações foi satisfatória e o Grupo 1 foi tão eficiente quanto o Grupo 2, o que possibilita a contenção farmacológica para coleta de amostras biológicas, exames físicos e realização de pequenas cirurgias nesta espécie.
Descritores: Tartarugas. Xilazina. Propofol.

1. Study performed at Wild Animals Research Laboratory, Federal University of Uberlândia (LAPAS – UFU), Minas Gerais, Brazil.

Introduction

Podocnemis expansa, popularly known as the giant Amazonian turtle or Amazon river turtle and, in its own region, called *arau* or *jurará-açu*, is distributed in the Amazon River basin, which encompasses equatorial forest and cerrado ecosystems in the northern and mid-western regions of Brazil[9]. This animal is considered one of the wild species most widely exploited zootechnically for the flavor and quality of its meat, whose market price is high compared to that of traditional domestic animals[4]. CITES[a] and IUCN[b] today classify this animal as being under threat of extinction.

Pharmacological restraint of turtles for routine physical examination and diagnostic or therapeutic procedures is often required in zoological institutions or wildlife and exotic animal practices[3] and on breeding farms. Premedication techniques are employed to sedate patients to facilitate handling, contribute to the analgesia, and reduce the dosage of anesthetic agents[6]; however, the use of tranquilizers on reptiles should be limited as premedication for general anesthesia[10]. Xylazine is a preanesthetic alpha-2 agonist drug frequently used in veterinary medicine, which induces sedation and analgesia[11]. Propofol is the injectable induction agent of choice in reptiles with available vascular access[14].

The aim of this study was to identify a technique to induce brief sedation and hypnosis in *P. expansa*.

Methods

The study involved 20 healthy specimens of *P. expansa* in captivity, 07 males and 13 females, with an average weight of 1.2 ± 0.24 kg, three years old, belonging to the commercial breeding farm Fazenda Moenda do Lago, in the municipality of Nova Crixás, Goiás, Brazil (13° 45' 55" S and 50° 47' 13,3" W – altitude: 223 m), by the IBAMA-RAM license number 28771. The animals were taken from their breeding tanks using nets, weighed and identified individually, and taken to the study site. The air temperature during the experiment was 29.78 ± 3.78ºC, measured with a thermometer showing maximum and minimum temperatures[c].

The specimens were divided into two groups (G1 and G2), which received, sequentially, 1.5 mg/kg IM of xylazine and 5 mg/kg IV of propofol (G1) and 1.5 mg/kg IM of xylazine and 10 mg/kg IV of propofol (G2). After antisepsis, the xylazine was applied in the muscle of the left thoracic member and the propofol in the cervical vertebral sinus, using hypodermic syringes[d] with 25 x 0.70 mm needle.

Hypodermic syringes with 13 x 0.45 mm needle[e], a digital glucosimeter[f], and glucose test strips[g] containing a drop of blood and coupled to the reading device, were used to obtain data about possible glycemic variations in the animals before the medications and 30 minutes after the application of propofol.

The anesthetic parameters were measured at 0, 5, 10, 20, 30, 45, 60, 90, 120, 150 and 180 minutes after application of the drugs. Time 0 (zero) was the moment when propofol was administered, fifteen minutes after administration of xylazine. Subjective scores of one (1) for minimal effects, two (2) for intermediary effects, and three (3) for maximum effect were used for the first three parameters described below. For the pain sensitivity tests, pulling back of the member in response to pinching was given a score of zero (0) and its absence, a score of one (1).

The following parameters were evaluated:

I – Locomotion: (1) animal with normal ability to move, (2) difficulty to move, and (3) absence of movement.

II – Muscle relaxation: (1) the animals kept its head up or retracted, (2) an intermediary situation, and (3) the head, members and tail remained suspended and relaxed.

III – Handling: (1) difficulty in flexing and extending the head, members and tail and in opening the animal's mouth manually, (2) intermediary situation, and (3) no resistance to manipulation of the head, members and tail, or to opening the animal's mouth.

IV – Sensibility to pain in the right thoracic member: applying a pair of 16-cm curved hemostatic Kelly pincers at the second lock on the phalanges of the forepaw, one waits for the response elicited by the removal of the pain stimulus, which is classified as zero (0) or absence of the response, one (1).

V – Sensibility to pain in the pelvic members: the procedure is similar to that of item 4, but the pincers are applied to the phalanges of the hindpaw.

VI – Heartbeat: measurement of the number of heartbeats per minute, using a vascular Doppler[h] at 0, 10, 30, 60, 120 and 180 minutes.

To check for significant differences in the recorded values, Student's *t* test was applied to the normal distributions, and the nonparametric Mann-Whitney U test, with the level of significance set at 0.05.

Results

All the animals survived the experiment, becoming inactive (loss of spontaneous locomotion) and presenting muscle relaxation, easy handling and loss of pain sensitivity in the right thoracic and pelvic members.

A consistent hypnotic effect was recorded 49.6 ± 22.1 seconds after the application of propofol in G2 and after 58.2 ± 55.1 in G1. All the animals of G2 recovered in 198 minutes, and those of G1 in 156 minutes.

[a] Convention on International Trade in Endangered Species of Wild Fauna and Flora
[b] International Union for Conservation of Natures and Natural Resources
[c] Máxima & Mínima, Incoterm, Porto Alegre, RS
[d] 3,0 ml, disposable, sterile, Injex, Ourinhos, SP
[e] 1,0 ml, disposable, sterile, Injex, Ourinhos, SP
[f] Precision-Plus-Abbott, Abbot Laboratories Brasil
[g] Precision-Plus-Abbott, Abbot Laboratories Brasil
[h] DV-20 Model, Microem, Ribeirão Preto, SP

Groups G1 and G2 did not present a statistically significant difference in the parameters of locomotion, muscle relaxation, heartbeat and pain sensitivity in the right thoracic and pelvic members at 0, 5 and 10 minutes. However, statistically significant differences were recorded for the parameters of pain stimuli in the pelvic members at 20, 30, 45 and 60 minutes and in locomotion at 90, 150 and 180 minutes, with the higher values occurring in G2, as indicated in Figures 1 and 2.

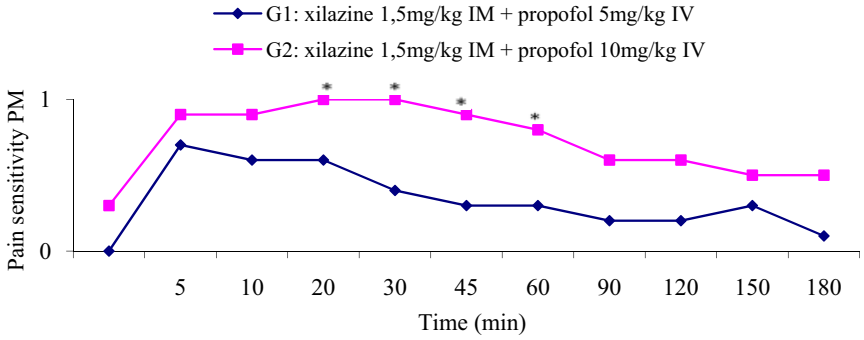

FIGURE 1 – Average scores of pain sensitivity in the pelvic members (PM) at different times, in *P. expansa* anesthetized with 1.5 mg/kg IM of xylazine and 5 mg/kg IV of propofol, and with 1.5 mg/kg IM of xylazine and 10 mg/kg IV of propofol. The asterisks (*) indicate significant differences between the groups ($p < 0.05$)

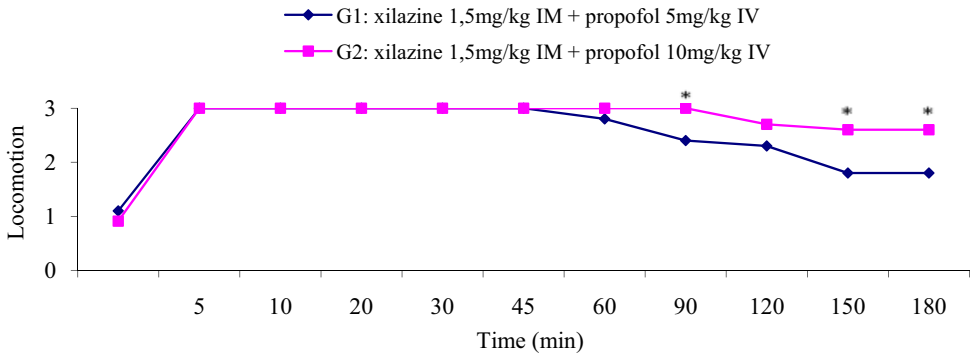

FIGURE 2 – Average scores of locomotion at different times, in *P. expansa* anesthetized with 1.5 mg/kg IM of xylazine and 5 mg/kg IV of propofol, and with 1.5 mg/kg IM of xylazine and 10 mg/kg IV of propofol. The asterisks (*) indicate significant differences between the groups ($p < 0.05$)

The glycemic level showed no statistically significant difference between the groups, but its values showed a discreet increase in all the tested animals, showing an average of 63.28 ± 19.66 mg/dl in the first measurement and 84.42 ± 26.64 mg/dl in the second.

Discussion

Anesthetic induction and recovery times are variable in reptiles, but tend to be longer than in mammals due to the variations in their metabolism in response to air temperature[2]. This study was carried out in the animals' natural environment and the air temperature remained at a gradient considered optimal by the same researcher[2], so the best possible action was expected from the drugs used in the tested dosages.

Because reptiles show anatomical and physiological differences in their circulation, with part of the blood collected from the tail, members and pelvic region flowing to the kidneys and liver and undergoing primary metabolization[7], the drugs were applied in the cranial body.

Xylazine did not produce alterations in the heartbeat of *P. expansa*, like in *Trachemys scripta elegans*[8], but contrary to what has been reported for dogs[11]. Propofol possesses an ultrashort action[14] and, in both groups, favored a minimal time of anesthesia of up to 20 minutes, at which point the statistical differences in pain sensibility in the pelvic members began. This finding is congruent with that of Santana[12], who administered 10 mg/kg of propofol in the cervical venous sinus of *P. expansa* and subjected them to successful esophagotomy. This time marks the beginning of recovery from anesthesia, which, according to Bennett[2], progresses in the cranial-caudal direction, returning in the opposite direction during recovery from anesthesia. The statistical difference in the parameter of locomotion starting from 90 minutes indicates that the lower dose allowed for faster recovery, therefore making it more interesting.

Some authors[1,14] have reported that the administration of propofol in reptiles causes respiratory depression, which was observed in some of the specimens during the experiment, as was their natural subsequent recovery, indicating that chelonians are animals that can withstand periods of apnea[2,14] and

natural recovering.

Contrary to what was reported by Holz[8], who used a combination of ketamine and xylazine without finding any advantage in relation to the use of ketamine alone, in the present study, the combined use of xylazin and propofol promoted a consistent hypnosis that was similar in the two groups, even at half the dosage. In situations where the administration of propofol is preceded by preanesthetic medication, such as opiates, agonists, α_2-adrenergics and phenothiazines, the induction dose can be reduced by 25 to 75%[5]. In the same way that a combination of midazolam and ketamine was utilized in the pharmacological restraint of *Chelydra serpentina*[3], xylazine in combination with propofol can be used for the same purpose in *P. expansa*.

A discreet elevation in the glucose levels was recorded, as also cited by Reyes *et al.*[13], but this elevation was not statistically significant and the values remained within those described for the species[15].

Conclusions

The sedation and hypnosis attained through the association of xylazine and propofol in *Podocnemis expansa* were satisfactory, enabling the species' pharmacological restraint to collect biological samples, make physical examinations and carry out minor surgeries.

The effect of these drugs on G1 was as efficient as on G2, although the dose of anesthetic applied to the former was 50% lower.

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