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Blind perineural injection for sciatic and femoral nerve block in cats: anatomical study and *in vivo* application

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ABSTRACT: This study investigated staining of sciatic (SN) and femoral (FN) nerves in feline cadavers following injection of two different volumes of methylene blue, and the application of one chosen volume *in vivo* using lidocaine in healthy sedated cats. Fourteen adult feline cadavers weighing 2.8 ± 1.2 kg and 10 healthy adult cats weighing 3.5 ± 1.2 kg were used in the study. First, an anatomical study of the SN and FN was carried out on six feline cadavers to measure depth of each nerve, followed by injection of 1% methylene blue on another eight cadavers to compare 0.2 with 0.3 mL/kg on each pelvic limb. The volume associated with staining ≥ 2 cm of the nerves was considered adequate for application *in vivo*. Based on the cadaveric study, 10 healthy cats received 1.25% lidocaine on one side and saline on the contralateral side at 0.2 mL/kg to assess efficacy of perineural block. Injections were performed between the semimembranosus and semitendinosus muscles for the SN and in the femoral triangle, next to the iliopsoas muscle, for the FN. Motor assessment included a score of ataxia and patellar reflex testing. Sensory assessment comprised proprioception and skin clamping at the leg. Data were analyzed using paired *t* test. Results showed insufficient *in vivo* block, demonstrated by lack of difference between sides ($P > 0.05$). In conclusion, SN and FN block cannot be successfully performed in cats without the aid of imaging modalities or neurostimulation using low volumes of solution (0.2 mL/kg).
Key words: pelvic limb, peripheral nerves, local anesthesia, nerve blocks, feline, regional anesthesia.

Injeção perineural para bloqueio de nervo isquiático e femoral às cegas em felinos: estudo anatômico e aplicação *in vivo*

RESUMO: O objetivo deste estudo foi investigar a coloração do nervo isquiático (SN) e femoral (FN) em cadáveres felinos após injeção de dois volumes diferentes de azul de metileno, bem como a aplicação de um volume selecionado *in vivo* utilizando lidocaína em felinos saudáveis sedados. Catorze cadáveres de felinos adultos com peso de 2,8 ± 1,2 kg e 10 gatos saudáveis adultos com peso de 3,5 ± 1,2 kg foram incluídos no estudo. Primeiramente, foi conduzido um estudo anatômico do SN e NF em seis cadáveres a fim de mensurar a profundidade de cada nervo, seguido de injeção de azul de metileno 1% em outros oito cadáveres para comparar os volumes de 0,2 e 0,3 mL/kg em cada membro pélvico. O volume associado com coloração de ≥ 2 cm dos nervos foi considerado adequado para aplicação *in vivo*. Com base no estudo cadavérico, 10 felinos saudáveis receberam lidocaína 1,25% em um lado e solução salina no lado contralateral em volume de 0,2 mL/kg, a fim de avaliar a eficácia do bloqueio perineural. As injeções foram realizadas entre os músculos semimembranoso e semitendinoso para o SN e no triângulo femoral, próximo ao músculo iliopsoas, para o FN. A avaliação de bloqueio motor incluiu um escore de ataxia e teste de reflexo patelar. Já a avaliação de bloqueio sensitivo incluiu propriocepção e pinçamento de pele da perna. Os dados foram analisados por meio de teste t pareado. Os resultados demonstraram bloqueio insuficiente *in vivo*, representado por ausência de diferença entre os lados ($P > 0.05$). Concluiu-se que o bloqueio do SN e FN não pode ser realizado com sucesso em felinos sem auxílio de modalidades de imagem ou neuroestimulação utilizando baixos volumes de solução (0,2 mL/kg).
Palavras-chave: membro pélvico, nervos periféricos, anestesia local, bloqueios neurais, felino, anestesia regional.

INTRODUCTION

Local anesthesia is a main component of orthopedic procedures in companion animals. In this context, peripheral nerve blocks are being increasingly used as an alternative to epidural anesthesia for surgery on the pelvic limbs. Epidural anesthesia has been shown to cause hypotension (IFF & MOENS, 2008) and the bilateral motor blockade produced by it can prolong recovery from anesthesia, thereby making perineural injection of local anesthetics an interesting alternative to epidural anesthesia in small animals (CAMPOY et al., 2012).

The femoral nerve arises from the fifth, fourth and sixth lumbar spinal roots (KONIG & LIEBICH, 2004). It supplies the four heads of

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the quadriceps muscle to then give rise to its main sensory branch, the saphenous nerve (MAHLER, 2012). To locate the femoral nerve in the femoral triangle, the main reference is the pulse of the femoral artery (VETTORATO et al., 2012). The sciatic nerve arises from the lumbosacral trunk and leaves the pelvis between the piriformis and deep gluteus muscles, or between the piriformis and genu muscles (CROUCH, 1969; HARRISON, 1969; GHOSHAL, 1972). The nerve then supplies the piriformis, deep gluteus, genu, quadratus femoris, semitendinosus, semimembranosus and biceps femoris muscles. The sciatic nerve can be found immediately underneath the biceps femoris, on the lateral aspect of the thigh (HARRISON, 1969; GHOSHAL, 1972).

Sciatic and femoral nerve blocks have been extensively investigated in dogs (MAHLER & ADOGWA, 2008; CAMPOY et al., 2010; PORTELA et al., 2010; SHILO et al., 2010; COSTA-FARRÉ et al., 2011; CAMPOY et al., 2012; ECHEVERRY et al., 2012; TREIN et al., 2017). However, most studies use ultrasound or neurostimulator guidance to locate the nerves, which might not be the case of most hospitals, clinics or even countries worldwide. Therefore, knowing blind techniques using solely anatomic landmarks remains an important part of anesthesiology in clinical practice.

Only a few studies have been found investigating sciatic and femoral nerve block in cats. One study assessed the visual distribution of bupivacaine using magnetic resonance imaging following perineural injection of 0.1 mL/kg with the aid of a neurostimulator and demonstrated successful sensory and motor blockade (EVANGELISTA et al., 2017). Similarly, another study using 0.3 mL/kg of bupivacaine has demonstrated successful blockade of the sciatic nerve block with neurostimulation in cats (FREITAS et al., 2004). The femoral nerve was effectively blocked in cats with 2% lidocaine at 2 mg/kg. The authors compared ultrasound and neurostimulation for nerve location and demonstrated greater efficacy with the use of ultrasound guidance (HARO et al., 2016).

Anatomical studies that investigated the technique for blind injection of sciatic and femoral nerve in cats had not been found at the time this study was performed. Therefore, the purpose of the present study was to first investigate the distribution of two volumes of methylene blue following perineural injection of the sciatic and femoral nerves in feline cadavers, then to assess the clinical effect of one chosen volume for an *in vivo* study using lidocaine in healthy sedated cats. We hypothesized that, using the volume capable of producing correct staining of the nerves during the cadaveric study, blind sciatic and femoral nerve block could be successfully performed *in vivo*.

MATERIALS AND METHODS

Animals

The study comprised three phases: an anatomical study using six feline cadavers weighing 3.2 ± 0.7 kg for measurement of nerve depth; a preliminary study of methylene blue spread using eight feline cadavers weighing 2.8 ± 1.2 kg; and an *in vivo* study using 10 healthy adult cats weighing 3.5 ± 1.2 kg.

Study design

Phase 1 - Anatomical Study

Phase 1 was carried out on six feline cadavers with the purpose of finding the mean depth of each target nerve for standardization of the technique at subsequent phases of the study. Exposure of each nerve was performed according to MAHLER & ADOGWA (2008).

To expose the sciatic nerve, a skin incision was performed at the lateral side of each thigh following an imaginary line between the great trochanter of the femur and the stifle. A transverse incision was made on end of the first incision to allow reflection of the skin cranially and caudally, thus exposing the biceps femoris muscle. The muscle was then sectioned at its midpoint and reflected proximally and distally to expose the underlying sciatic nerve (Figure 1).

For the femoral nerve, two transversal incisions were performed on the medial thigh, one at the inguinal abdomen and one at the stifle. A third incision connected the transversal incisions and the skin was reflected to both sides. The femoral nerve was then exposed within the iliopsoas muscle, next to the femoral artery within the femoral triangle (Figure 2). The depth of each nerve was measured in centimeters using a digital caliper with accuracy of 0.02 mm (Lee Tools, São Paulo State, Brazil) from a predetermined site of injection (described in Phase 2) on the skin on both pelvic limbs. The means of nerve depth were used as a reference for needle insertion during the next phases of the study.

Phase 2 - Determination of Injected Volume Using 1% Methylene Blue

Phase 2 involved the use of eight cadavers to compare two volumes of 1% methylene blue (hydrated methylene blue, Neon Commercial, SP,

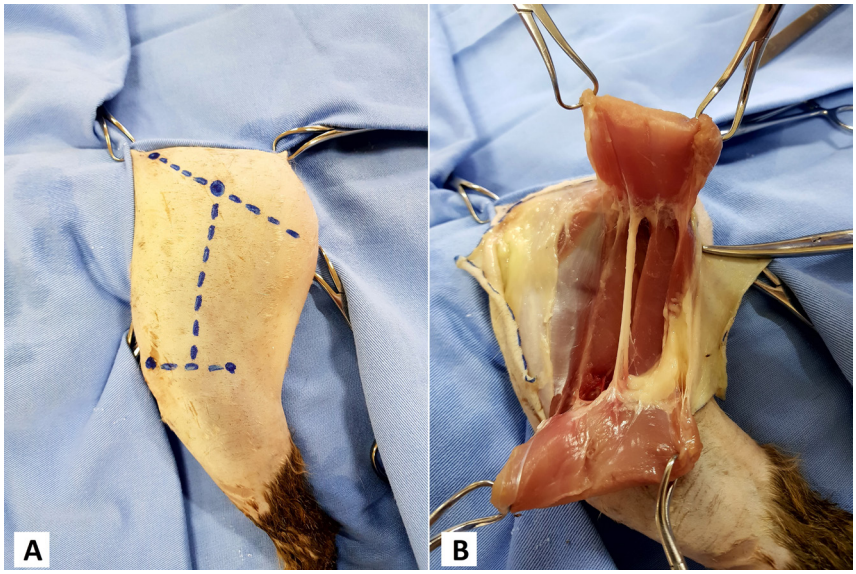

Figure 1 - Dissection window for the left sciatic nerve in a feline cadaver. A) Lines marking the skin incisions from the great trochanter of the femur to the stifle. B) Complete dissection retracting the halves of the biceps femoris muscle to expose the sciatic nerve underneath.

Brazil) solution for nerve staining ≥ 2 cm, which is the minimum required for perineural injection according to CAMPOY et al. (2008). The volumes used were 0.2 and 0.3 mL/kg, respectively G0.2 and G0.3, each one administered to one pelvic limb of each cadaver. Sides were randomly selected using the random number generator on Microsoft Excel (Microsoft Corporation, version 14.0.7143.5000, 2010, USA).

Perineural injections were performed according to CAMPOY et al. (2008) using a 22-gauge hypodermic needle (Descarpack, Biotecmed, São Paulo State, Brazil). Cadavers were placed in lateral

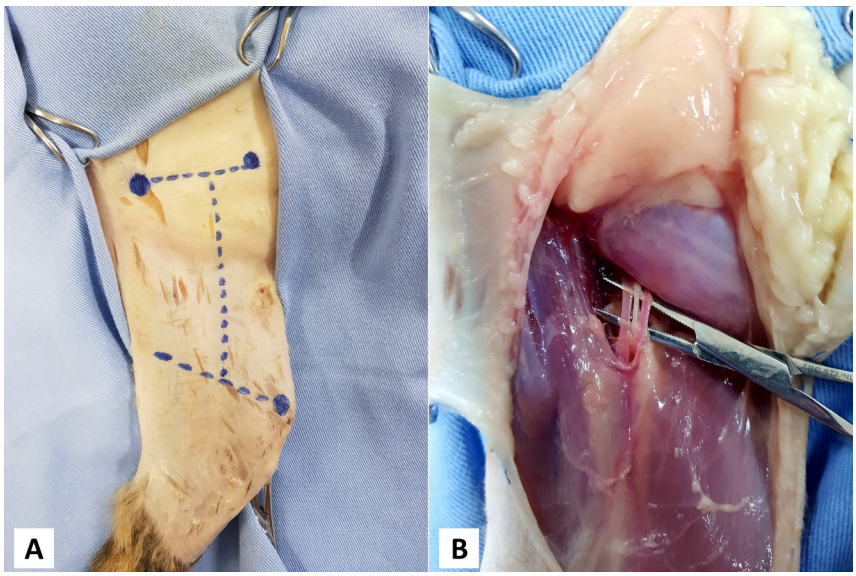

Figure 2 - Dissection window for the left femoral nerve in a feline cadaver. A, Lines marking the skin incisions from the inguinal abdomen to the stifle. B, Complete dissection exposing the femoral artery and vein to the sides and the femoral nerve at the center of the femoral triangle.

recumbency with the limb to be injected uppermost in natural extension for the sciatic nerve and abducted laterally for the femoral nerve. Perineural injection of the sciatic nerve was performed between the semimembranosus and semitendinosus muscles, immediately below the level of the greater trochanter. The needle was advanced between the muscles for approximately 2.3 cm (according to the results of Phase 1). Following sciatic nerve injection, the limb was abducted at 90 degrees and slightly extended for femoral nerve injection at the skin proximal to the stifle, at a 45º angle with the sartorius muscle. The anatomical landmarks of the femoral triangle were used as references and the needle was advanced into the triangle for 2.7 cm (according to Phase 1) through the sartorius and rectus femoris muscles. All injections were performed by the same individual, previously trained in the first phase of the study and unaware of the volume used. The syringe was covered with aluminum foil to prevent the anesthetist from seeing its volume.

Following all four injections (sciatic and femoral nerve on both sides), the same technique previously described for dissection was carried out for assessment of nerve staining with 1% methylene blue. The extension of nerve staining was measured in cm using a digital caliper and was considered adequate for application *in vivo* when ≥ 2 cm according to CAMPOY et al. (2010).

Phase 3 - In vivo study

Ten healthy adult client-owned cats from the local community aged 2.0 ± 1.3 years were included in the study upon informed consent signed by the owners with the purpose of investigating the efficacy of the previously studied volumes of injectate for perineural block *in vivo*. Subjects were deemed healthy based on complete blood count and physical examination including heart rate, respiratory rate, capillary refill time, mucosal coloration, rectal temperature and skin turgor. Only animals with docile demeanor were included in the study. Demeanor was determined based on tolerance to restraint and response to stimulus with food (which would be used later to encourage cats to walk during assessment of the blockade).

Baseline recordings (T_B) comprised motor and sensory assessment of the pelvic limbs through scores, as follows: 0 = normal response, no blockade; 1 = decreased response, partial blockade; 2 = no response, complete blockade. Motor assessment included ataxia and patellar reflex test. Sensory assessment included proprioceptive test and pain assessment tested through skin clamping using a straight 2.2 Kelly forceps closed to the first ratchet for 3 seconds on three points. The three points used for pain assessment corresponded to saphenous, peroneal and tibial nerve blockade, respectively: medial aspect of the thigh (supplied by the saphenous nerve), plantar aspect of the metatarsus (supplied by the peroneal nerve), and skin on the lateral aspect of the fourth digit (supplied by the tibial nerve), according to TREIN et al. (2017).

Prior to the procedures, animals were fasted for 12 hours, and water was withheld for 2 hours. Subjects were sedated with intramuscular xylazine (König, São Paulo, Brazil) at 2% (1 mg/kg). Xylazine was given intramuscularly at the middle to distal third of the semi-tendinous muscle. Following sedation, perineural injections were performed with 1.25% lidocaine or saline immediately below the level of the greater trochanter. Finally, sedation was reversed with intravenous yohimbine (0.1 mg/kg; Drogavet, Paraná State, Brazil). Local anesthesia was performed 20 minutes following sedation using the same injection technique described for Phase 2. Both pelvic limbs were used for the study. One side received 1.25% lidocaine (Bravet, Rio de Janeiro State, Brazil) without epinephrine for each nerve (GL) and the contralateral side received the same volume of saline (GC). Sides were randomly selected using the random number generator on Microsoft Excel (Microsoft Corporation, version 14.0.7143.5000, 2010, USA).

Five minutes after injections (with completion of injections being T_0), motor and sensory assessments were performed with the cats still under sedation (T_S). Yohimbine was then administered and postanesthetic evaluations were carried out at 10-minute intervals (T_{10}, T_{20}, T_{30}, T_{40}, etc.), starting from when cats were able to stand to when all variables returned to baseline.

Statistical analysis

Results from Phase 1 were calculated as mean and standard deviation. Results from Phase 2 were analyzed using Fisher's exact test to assess the relation between staining and each volume of solution. A paired t test was used to compare the extension of nerve staining between groups G0.2 and G0.3.

The duration of motor and sensory blockades was assessed for normality using Shapiro-Wilk's test and then compared among groups GL and GC using Wilcoxon's signed ranks test. The observation of plantigrade stance (motor blockade of the sciatic nerve) and the use of lidocaine was analyzed using Fisher's exact test. Analyses were

performed using a commercial software (GraphPad Prism 6.01, 1999-2012 GraphPad Software, USA) and significance was considered when $P \leq 0.05$.

RESULTS

The mean depth of sciatic and femoral nerves on the six cadavers used in Phase 1 was 2.36 ± 0.46 and 2.75 ± 0.79 cm on the right pelvic limb and 1.87 ± 0.36 and 2.66 ± 0.98 cm on the left pelvic limb, respectively.

Median values (minimum-maximum) of nerve staining during Phase 2 in G0.2 were 0 (0-4.0) cm and 0.25 (0-2.68) cm for sciatic and femoral nerves, respectively. Values in G0.3 were respectively 1.91 (0-6.40) cm and 1.62 (0-2.62) cm (Figure 3 and Figure 4). No differences were found with regard to the extension of staining in cm between groups and there was no association between injected volume and staining ≥ 2.0 cm ($P > 0.05$).

No differences were found between sides on phase 3 of the study regarding duration of ataxia, proprioceptive changes, and loss of reflexes that indicated nerve blockade (Figure 5). Seven cats out of ten (70%) from phase 3 demonstrated plantigrade stance in the limb injected with lidocaine, which was recorded as presence of ataxia during visual scoring. A significant association was found between plantigrade stance and the use of lidocaine ($P = 0.003$). Three cats showed a decrease in proprioception and a few cats showed decreased response during skin clamping (five cats on the medial thigh, three cats on the metatarsus and two cats on the fourth phalanx). However, none of the variables tested in the *in vivo* study differed among groups ($P > 0.05$), demonstrating insufficient blockade of the pelvic limb with the volume tested.

DISCUSSION

This is the first study to completely investigate the effectiveness of sciatic and femoral nerve block in cats without the use of imaging or neurostimulation, which is the most common scenario of clinical practice in many countries. However, our results point to the impossibility of relying on these perineural blocks without the aid of imaging or neurostimulation, since results from Phase 3 showed insufficient block in most subjects.

Sciatic nerve block is expected to result in loss of the muscular support to the stifle and hock. In our study, seven cats (7/10, 70%) showed plantigrade stance (motor blockade), which is a sign of sciatic nerve blockade. However, other variables (sensory blockade tests) indicated that the blockade was only partial (tibial and peroneal nerve tests).

Femoral nerve block results in loss of sensation on the medial side of the leg (TREIN et al., 2017). In the present study, saphenous nerve block proved unsuccessful since there was no difference between groups with regard to duration of decreased responses. Therefore, the change in the response seen on both sides during the first 20 minutes of evaluations can be ascribed to residual sedative

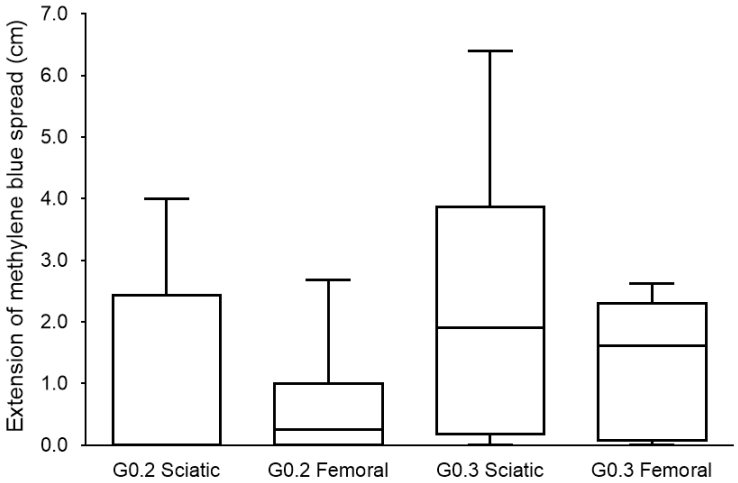

Figure 3 - Median, interquartile range (box), and minimum-maximum (whiskers) of sciatic and femoral nerve staining with 1% methylene blue at 0.2 (G0.2) and 0.3 (G0.3) mL/kg in feline cadavers. Groups do not differ according to Wilcoxon matched pairs signed rank test ($P > 0.05$).

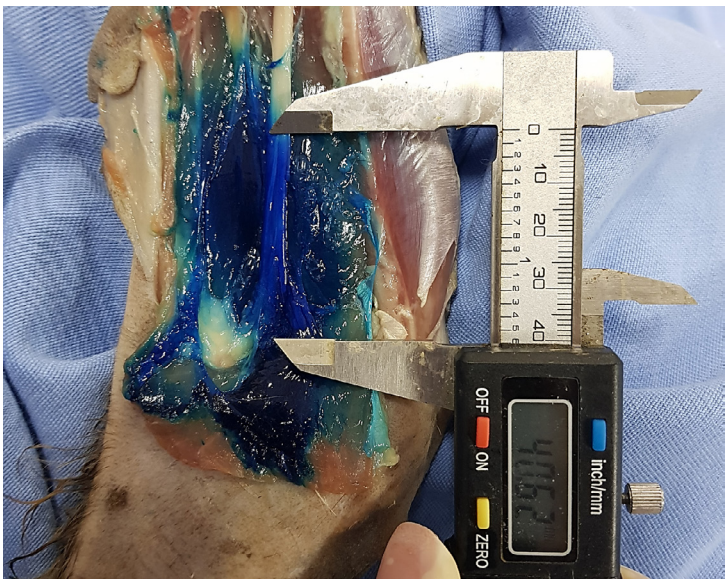

Figure 4 - Demonstration of the measurement of sciatic nerve staining with 1% methylene blue using a digital caliper in a feline cadaver. Staining in the picture is measured as 4.06 cm (1.6 inches).

effects of xylazine, which might also be the case for tibial and peroneal nerves.

The first two phases of our study provided the basis for needle insertion and volume of injectate during phase 3, which were respectively 2.3 cm for the sciatic nerve, 2.7 cm for the femoral nerve and 0.2 mL/kg. The volumes used during phase 2 were selected according to previous studies in small animals (ECHEVERRY et al., 2010; TREIN et al., 2017). However, given the blind technique used in our study, the low volume of 0.1 mL/kg was not considered an option and thus 0.2 and 0.3 mL/kg were used for comparison. The advantages of ultrasound and neurostimulation for perineural blocks have been well established in small animals (MAHLER & ADOGWA, 2008; CAMPOY et al., 2010; SHILO et al., 2010; COSTA-FARRÉ et al., 2011; ECHEVERRY et al., 2012; MAHLER, 2012). Lower volumes (0.1 mL/kg) still resulted in successful anesthesia in those studies due to the more precise identification of the nerves. However, the present study provided an option for perineural anesthesia without any guidance, given the reality of animal anesthesia in third world countries. The results of our study failed to provide a feasible alternative for sciatic and femoral nerve block in healthy cats.

Despite the results of phase 2 showing no relationship between volume of injectate and nerve staining, the volume selected for phase 3 proved insufficient to provide complete blockade of sciatic and femoral nerves in our study. Low volumes (0.1-0.2 mL/kg) of anesthetic solution have shown to provide successful blockade in dogs with the use of electrostimulation and ultrasonography (MAHLER & ADOGWA, 2008; CAMPOY et al., 2010; TREIN et al., 2017).

In cats, the sciatic nerve was successfully anesthetized using bupivacaine at 0.1 mL/kg with ultrasound guidance followed by verification of the spread of the solution using magnetic resonance imaging (EVANGELISTA et al., 2017). A few cats showed signs of partial nerve blockade in our study, but none showed complete blockade of sciatic and femoral nerve with 0.2 mL/kg, which demonstrated that blind techniques should not be advised in the feline species with low volumes of solution despite previous training.

Locoregional anesthesia performed with bone and muscle anatomic references can be impaired by fascial tissue. Previous studies have demonstrated that fasciae can act as a barrier against diffusion of local anesthetics, hindering their dispersion around the nerves (BURCKETT-ST.LAURENT et al., 2017). This can lead to poor absorption by the target nerve and incomplete or ineffective anesthesia, specially in peripheral blocks (KARMAKAR et al., 2018).

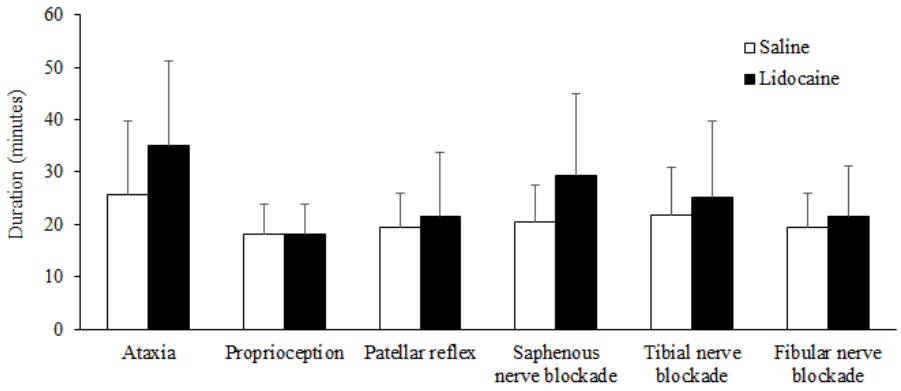

Figure 5 - Means (columns) and standard deviation (whiskers) of the duration (minutes) of ataxia, loss of proprioception, loss of patellar reflex and saphenous, tibial and fibular nerve blockade following perineural injection of saline solution or 1.25% lidocaine at 0.2 mL/kg in healthy cats. Groups do not differ according to paired t test ($P > 0.05$).

Higher volumes, adjuvants and precise identification of fascial planes are commonly used strategies to improve efficacy of perineural blocks (SAKAMOTO et al., 2020).

In the present study, the higher volume of injectate (0.3 mL/kg) was tested only during phase 2 with methylene blue. In phase 3, 1.25% lidocaine was used at 0.2 mL/kg and compared with a control group using the same volume of saline solution. Future studies using 0.3 mL/kg of a local anesthetic should be performed to further investigate the feasibility of a blind technique for sciatic and femoral nerve block in cats with a higher volume to increase the possibility of correctly reaching these nerves. However, it is possible that lidocaine might need to be further diluted to achieve a higher volume of 0.3 mL/kg and this might implicate a higher risk of neural toxicity from lidocaine.

The volume and concentration of lidocaine in the present study accounted for a total of 5 mg/kg per cat (2.5 mg/kg each nerve). The dose of lidocaine was the main reason for the concentration used in our study. Lidocaine at 2% would have resulted in 8 mg/kg per cat, which would be above the toxic dose of lidocaine in the feline species (6 mg/kg) (CERVANTES, 2011). Dilution of the anesthetic agent might be another reason for the incomplete blockade.

In one study using 0.3% bupivacaine and a different approach for blind injection in cats, the volume of 0.33 mL/kg provided successful motor and sensory blockade of the sciatic nerve, which lasted for 165 and 241 minutes, respectively (FREITAS et al., 2004). However, those authors did not test the femoral nerve block using bupivacaine. The femoral nerve can be more difficult to anesthetize without the use of guidance. In addition, the distance between the femoral nerve and the external iliac artery differs significantly among dogs and cats (MOGICATO et al., 2015), which could make femoral nerve block more difficult using solely anatomical references that are more commonly studied in dogs.

These authors recommend using other techniques to have successful pelvic limb anesthesia without ultrasonography or neurostimulation in the feline species,. The initial purpose was to provide an alternative to epidural, so that feline patients could recover from pelvic limb surgeries with one functional limb. However, the results demonstrated that blind perineural injection is not suitable for reliable pelvic limb anesthesia in cats, and thus other techniques should be recommended for pelvic limb desensitization in this species. One main limitation of the study was basing the injected volume solely on a cadaveric study using methylene blue. Despite previous studies discussing the extension of nerve staining ≥ 2 cm as representative of an appropriate spread, this was found in dogs and not cats (CAMPOY et al., 2010). In addition, considering the different species and blindly performed technique, perhaps the use of 0.3 mL/kg would have been more appropriate for the purpose of our study. Another limitation of the study was calculating the depth of the nerves in cadavers with the nerves exposed through dissection. The tissue became deformed following dissection. Perhaps a future study using imaging *in vivo* would provide more precise measures of nerve depth.

CONCLUSION

In conclusion, sciatic and femoral nerve block cannot be successfully performed in cats without the aid of imaging modalities or neurostimulation using low volumes of solution (0.2 mL/kg).

ACKNOWLEDGMENTS

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AUTHORS' CONTRIBUTIONS

All authors contributed equally to the conception and writing of this manuscript. All authors have revised and approved the manuscript in its final version.

BIOETHICS AND BIOSECURITY COMMITTEE APPROVAL

The study was approved by the Committee for Animal Usage at the Centro Universitário das Faculdades Integradas de Ourinhos (protocol No. 017/2017).

DATA AVAILABILITY STATEMENT

All data pertaining this study can be obtained through contacting the author for correspondence at andre.v.soares@ufsm.br.

DECLARATION OF USE OF ARTIFICIAL INTELLIGENCE

No resources of artificial intelligence were used to write or replace authors' activities and skills in this manuscript.

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