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Effect of temperature-responsive hydrogel on femoral and sciatic nerve blocks using bupivacaine in Beagle dogs

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Abstract

Objectives: To compare the duration of regional anesthesia of the pelvic limb using bupivacaine with and without a temperature-responsive hydrogel (TRH) in dogs.

Methods: Under anesthesia using medetomidine (10 $\mu g \cdot kg^{-1}$), alfaxalone (2 $mg \cdot kg^{-1}$), and isoflurane, seven healthy male Beagles received four injections of 0.5% bupivacaine (1 $mg \cdot kg^{-1}$ with 5 $\mu g \cdot ml^{-1}$ epinephrine) to block the femoral and sciatic nerves bilaterally via ultrasound with nerve stimulation guidance. Bupivacaine was used on one pelvic limb (Bup treatment), and bupivacaine with TRH was used on the contralateral limb (Bup-TRH treatment). The nerve block was considered successful upon the absence of responses to pinching the digital pads and mid-tibial skin of both pelvic limbs with mosquito forceps; the pinch, proprioception, and locomotion tests were performed before (baseline) and at each hour after the nerve block until sensory and motor functions returned to baseline. The effect of TRH on nerve blocks was analyzed using a linear mixed model.

Results: The duration of the sensory nerve block at the digital pads and mid-tibial skin was longer with Bup-TRH (8.0 ± 1.6 h and 10.9 ± 1.6 h, respectively) than with Bup treatment (3.7 ± 2.0 h and 8.0 ± 1.6 h, respectively). Motor block times of proprioception and locomotion were longer with Bup-TRH (9.3 ± 1.6 and 12.7 ± 1.5 h, respectively) than with Bup treatment (4.6 ± 1.9 and 9.6 ± 1.5 h, respectively). No complications were observed.

Clinical significance: TRH extended the duration of regional anesthesia of the pelvic limb using bupivacaine.

KEYWORDS
canine, hydrogel, nerve block, nerve stimulator, regional anesthesia, ultrasound-guided

1 | INTRODUCTION

Drug delivery systems for local anesthesia have been developed to extend the duration of a single injection of local anesthetic agents and to overcome the complications of multi-injection or constant-injection devices, including infection via a catheter-type device and required resource for long-term pain management, especially in animals that are unable to communicate and self-manage (Abelson et al. 2009).

Drug delivery systems are typically categorized as injectable particles (nanoparticles and liposomes), injectable liquids (cyclodextrins, injectable liquid polymers, and hydrogels), and hybrid formulations (Santamaria et al. 2017). Multivesicular liposome, Nocita, is now a commercially available drug delivery system in veterinary medicine (Grubb & Lobprise 2020).

PF-72 has recently been developed as a drug delivery system and is based on Pluronic F 127 20% w/v and sodium hyaluronate 0.5% w/v (Seol et al. 2013; Oh et al. 2019). Unlike other hydrogels with high viscosities that are challenging to inject and may require surgical implantation (Santamaria et al. 2017), PF-72, one of the recent developed temperature-responsive hydrogel (TRH), is an injectable, viscous fluid at 2–8°C that becomes a gel at 37°C. The in vitro and in vivo biocompatibilities and preclinical feasibility tests of PF-72 are favorable, and minimal inflammatory reactions and no negative impacts on wound healing have been reported in rats (Kim et al. 2015; Oh et al. 2019; Seol et al. 2013). The use of PF-72 as an injectable and degradable hydrogel-based system in the joint cavity also has a therapeutic effect on osteoarthritis in humans (Ayhan et al. 2014). The effect durations of bupivacaine and ropivacaine with PF-72 have been reported as 24–72 h when administered subcutaneously or intra-articularly in rats and humans (Choi et al. 2019; Kim et al. 2015; Oh et al. 2019). For regional anesthesia, a rat study comparing the duration of sciatic nerve blocks using 0.5% bupivacaine with and without another hyaluronic acid-based drug delivery system reported that the effect duration almost doubled from 135 min to about 262 min, a 94% increase (Jia et al. 2004; Weiniger et al. 2010).

To evaluate the use of PF-72 for regional anesthesia in dogs, this study compared the duration of regional anesthesia of the pelvic limb using 0.5% bupivacaine to that when using 0.5% bupivacaine with a PF-72. After performing femoral and sciatic nerve blocks using these agents, the sensory blockade of the digits and mid-tibial skin and the motor blockade (proprioception and locomotion) were compared in Beagle dogs. The hypothesis was that PF-72 would prolong the effects of regional anesthesia in the pelvic limb compared with bupivacaine alone.

2 | MATERIALS AND METHODS

2.1 | Study design and ethical approval

This prospective, blinded, randomized study was approved by the Institute Animal Care and Use Committee of the Seoul National University (SNU-200728-4).

2.2 | Animals

Seven healthy male Beagles with a mean weight of 8.8 ± 1.3 kg and a mean age of 14.3 ± 3.9 months were included in this study. They were obtained from a previous study unrelated to the pelvic limb. No abnormalities were found on physical examination, thoracic radiographs, or blood laboratory studies. All dogs were fasted for 12 h prior to the study period and were given free access to water.

2.3 | Anesthetic procedure

The dogs were sedated with intramuscular injections of medetomidine (10 $\mu g \cdot kg^{-1}$; Tomidine, Provet Veterinary Products, Ltd., Istanbul, Turkey). An intravenous (IV) catheter (Sewoon Medical Co., Seoul, Republic of Korea) was placed in the cephalic vein, and Hartmann solution (JW Pharmaceutical, Seoul, Republic of Korea) was administered IV (5 $ml \cdot kg^{-1}\ h^{-1}$). The dogs were induced with IV alfaxalone (2 $mg \cdot kg^{-1}$; Alfaxan, Jurox, Australia) and intubated. The spontaneously breathing dogs were anesthetized using a rebreathing circuit with isoflurane (I-Fran Liquid, Hana Pharm Co., Ltd., Hwaseong, Republic of Korea) in 100% oxygen (1.5 $L \cdot min^{-1}$) at a target of 1.5% end-tidal isoflurane concentration (F_E'Iso). The physiological parameters were monitored continuously. After all procedures, atipamezole (50 $\mu g\ kg^{-1}$; Reversal, Provet Veterinary Products, Ltd., Istanbul, Turkey) was used to reverse the sedation.

2.4 | Preparation of local anesthetic solution

The 0.5% bupivacaine (Bupivacaine HCl, Myeongmoon Pharm. Co., Hwaseong, Republic of Korea) used in this study included epinephrine (5 $\mu g \cdot ml^{-1}$; Epinephrine Injection, Dai Han Pharm. Co., Ansan, Republic of Korea). The TRH (PF-72, TGel Bio, Co., Ltd., Republic of Korea), a lyophilized powder, was mixed with 10 ml of 0.5% bupivacaine and refrigerated at 2–8°C for one day prior to use according to the manufacturer guideline (Kim et al. 2015).

2.5 | Regional anesthesia for the pelvic limb

The dogs were positioned in lateral recumbency. The skin from the sacrum to the mid-femur and inguinal region was clipped and prepared aseptically, bilaterally. Each dog underwent femoral and sciatic nerve blocks using 1 $mg \cdot kg^{-1}$ (0.2 $ml \cdot kg^{-1}$ per nerve) of 0.5% bupivacaine in the right or left pelvic limb (Bup treatment) and femoral and sciatic nerve blocks using 1 $mg \cdot kg^{-1}$ of 0.5% bupivacaine mixed with TRH (PF-72) in the contralateral pelvic limb (Bup-TRH treatment). The total injected dose of bupivacaine was 4 $mg \cdot kg^{-1}$ in each dog. The pelvic limb, nerve block order, and drug types were randomized via drawing lots. All nerve blocks were performed by one experienced investigator who is skilled in ultrasound-guided nerve blocks. Though the drug types were randomized, the requirement of increased injection pressure of TRH prevented blinding of the investigator due to clear differences in the handling properties of the test treatment.

The ultrasound-guided perineural femoral and sciatic nerve blocks were performed as follows: the femoral nerve was blocked in the iliopsoas muscle before its division into the sartorius and quadriceps femoris muscles using a ventral suprainguinal approach and the

inguinal nipple as a landmark (Echeverry et al. 2012). The sciatic nerve was blocked at the area immediately distal to the greater trochanter and the ischiatic tuberosity (Campoy et al. 2010). Ultrasonographic equipment (Samsung Medison H60, Samsung Medison Co., Ltd., Seoul, Republic of Korea) with a linear transducer was used for the visualization of the needle with an in-plane technique for the approach. An echogenic insulated needle with a short bevel (20 gauge, 150 mm, Sonoplex Nanoline Stim Cannula, Pajunk, Geislingen, Germany) was inserted in the long axis of the ultrasound transducer and advanced toward the target nerve. A nerve stimulator (EZstim II ES400, Life-Tech, Stafford, TX, USA), set at a current of 0.3 mA, a frequency of 2 Hz, and a pulse duration of 0.1 ms, was connected to the insulated needle and confirmed the proximity of the needle tip to the target nerve. To avoid an IV injection, a negative aspiration test was performed before injection. The longitudinal distribution of the local anesthetic agent along the femoral or sciatic nerve was observed using transverse ultrasound images while moving the transducer along the nerves. The same needle, equipment, and method were used for both injections.

2.6 | Evaluation and data collection

A binary outcome (the presence/absence of sensory response and motor function deficit) was used for all assessments. For the sensory blockade assessment, the dogs were placed in a natural standing position and gently restrained by an investigator standing in front of and distracting the dogs. Another investigator who was blinded to the treatment of each limb stood behind the dog being assessed and pinched the digital pads and mid-tibial skin of the pelvic limbs using mosquito forceps (H112-22012; Hermann Medizintechnik, Germany) when the dog looked forward so that the pinching could not be seen by the dog. The presence of responses to pinching the digits and mid-tibial skin was ascertained by the dog turning its head, actively looking at the stimulated area, and/or vocalization. Complete nerve block (i.e. both limbs) was considered to be successful if there were no responses to pinching all four digital pads and all four points of mid-tibial skin in both pelvic limbs and if there was loss of proprioception and locomotion for at least 1 h following administration of the nerve blocks. If no reactions were observed, the first ratchet was closed for one second for equal pressure and released. If a baseline sensory response was noted before closing the ratchet, the ratchet was released immediately. The tests were repeated each hour after the nerve block, and they were stopped when pain responses were observed. The duration of the sensory block was defined as the time from the nerve block to the last timepoint with no sensory responses in pinching points.

The motor blockade was assessed via tests for proprioception and locomotion for each limb. Proprioception was considered as the immediate replacement of the digit on its plantar surface when the dorsal surface was placed on the ground, and it was confirmed to be normal bilaterally before the nerve blocks. Delayed or no reposition of the limb was considered a negative result. Locomotion was evaluated in terms of weight bearing on the limb and the ability to walk in a straight line. Control and assessment locomotion tests were recorded on video (iPhone 8 and 11; Apple, CA, USA) for 2–3 min of walking. Full weight bearing on the limb and the ability to walk without missteps was considered as the baseline. Partial or no weight bearing on the limb or the presence of missteps was considered to be the presence of a motor-function deficit. The duration of the motor block was defined as the time from the nerve block to the last timepoint with a motor-function deficit.

Prior to anesthesia, control pinching responses, normal proprioception, and normal locomotion were measured. After full recovery from anesthesia, the tests were repeated every hour until all control values of all categories had returned. Between evaluations, the dogs rested in a cage to prevent skin damage caused by dragging of the legs. Meloxicam (0.2 $mg{\cdot}kg^{-1}$; Metacam, Boehringer Ingelheim, Barcelona, Spain) was administered subcutaneously after the study period.

2.7 | Statistical analysis

All statistical analyses were performed using SPSS Statistics for Windows, Version 26 (IBM Corporation, NY, USA). A linear mixed model was used with pinching levels, pinching points, and types of drug solutions to assess the influence of different variables on the sensory blockade duration of bupivacaine. The analysis of the duration of the motor blockade (proprioception and locomotion) was performed using a linear mixed model. Akaike's information criterion was used to compare different variance-covariance structures for the mixed model. A quantile-quantile plot was used to assess the distribution of the residuals. A type III analysis of variance was performed with a fixed model. Bonferroni correction was performed for *post hoc* comparisons. The pinching point and type of drug measurements were presented as the mean ± standard deviation. A value of $p \leq 0.05$ was considered statistically significant.

3 | RESULTS

3.1 | Nerve block procedures

On the ultrasound images, the doughnut sign, indicating the distribution of local anesthetic around the nerve, was observed at the injection level in all dogs. Non-circumferential distribution was observed as the anesthetic spread from the injection site. The measured spreading distances of the local anesthetic agent along the nerve were 2.5—4.0 cm around the femoral nerve and 2.3—6.0 cm around the sciatic nerve for the Bup treatment and 2.0—4.0 cm around the femoral nerve and 2.5—7.0 cm around the sciatic nerve for the Bup-TRH treatment.

3.2 | Duration of nerve blocks

The overall duration of complete sensory blockade at the digital pad and mid-tibial skin was longer ($p \leq 0.001$) with the Bup-TRH treatment (8.0 ± 1.6 h and 10.9 ± 1.6 h, respectively) than with the Bup

TABLE 1 Complete sensory blockade duration of femoral and sciatic nerve blocks with bupivacaine (0.5% with 5 μg ml^{-1} epinephrine) in the right or left pelvic limb (Bup treatment) and bupivacaine (0.5% with 5 μg ml^{-1} epinephrine) with temperature-responsive hydrogel in the contralateral pelvic limb (Bup-TRH treatment)

Level	Point	Bup treatment duration (hours)	Bup-TRH treatment duration (hours)
Digit	Second	3.7 ± 2.1	8.1 ± 1.9
	Third	3.7 ± 2.1	8.0 ± 1.6
	Fourth	3.7 ± 2.1	8.0 ± 1.6
	Fifth	3.7 ± 2.1	8.0 ± 1.6
	Overall	3.7 ± 2.0	8.0 ± 1.6
Mid-tibia	Medial	8.9 ± 1.2	11.1 ± 1.6
	Lateral	7.9 ± 1.6	10.7 ± 1.8
	Cranial	7.7 ± 1.4	11.0 ± 1.6
	Caudal	7.7 ± 1.5	10.7 ± 1.8
	Overall	8.0 ± 1.4	10.9 ± 1.6
Linear mixed model	Treatment	$p < 0.001$	
	Level	$p = 0.001$	
	Point	$p = 0.011$	

Data are presented as the mean ± standard deviation

TABLE 2 Complete motor blockade duration of femoral and sciatic nerve blocks with bupivacaine (0.5% with 5 μg ml^{-1} epinephrine) in the right or left pelvic limb (Bup treatment) and bupivacaine (0.5% with 5 μg ml^{-1} epinephrine) with temperature-responsive hydrogel in the contralateral pelvic limb (Bup-TRH treatment)

Level	Bup treatment duration (hours)	Bup-TRH treatment duration (hours)	*p*
Proprioception	4.6 ± 1.9	9.3 ± 1.6	$p < 0.001$
Locomotion	9.6 ± 1.5	12.7 ± 1.5	$p = 0.002$

Data are presented as the mean ± standard deviation

treatment (3.7 ± 2.0 h and 8.0 ± 1.6 h, respectively). The duration of the sensory blockade was significantly different between the four points at each level ($p = 0.011$) (Table 1). The recovery times of proprioception ($p < 0.001$) and locomotion ($p = 0.002$) were also longer with the Bup-TRH treatment (9.3 ± 1.6 and 12.7 ± 1.5 h, respectively) than with the Bup treatment (4.6 ± 1.9 and 9.6 ± 1.5 h, respectively) (Table 2).

3.3 | Complications

No signs of cardiovascular or central nervous system toxicities related to the local anesthetic agents, such as hypotension, seizure, respiratory depression, or coma, were observed. No abnormalities in behavior or gait were observed a day and a week after the procedure.

4 | DISCUSSION

PF-72 significantly prolonged the sensory and motor nerve block effects in regional anesthesia of the pelvic limb. The results support the hypothesis that PF-72 would last longer the duration of the digital and mid-tibial skin sensory blockade and the proprioception deficits caused by regional anesthesia of the pelvic limb using 0.5% bupivacaine. Significant differences were found in the sensory nerve block durations at the four points of each level. These differences may be due to the different durations of the femoral and sciatic nerve blocks, as the femoral nerve is mainly responsible for the sensory innervation of the medial aspect of the pelvic limb while the sciatic nerve is responsible for the lateral aspect (Evans & De Lahunta 2013). The relatively long duration of the femoral nerve block is similar to findings of previous studies that used bupivacaine for femoral and sciatic blocks in dogs (O Cathasaigh et al. 2018; Portela et al. 2010). Motor nerve block effect was longer than sensory nerve block effect. Too long motor blockade limiting locomotion would be undesirable as it may hinder post-operative recovery. As the extended duration presented in this study was 12.7 ± 1.5 h, it is expected to be resolved in a half-day without interrupting post-operative recovery.

The evaluation of PF-72 as a drug delivery system requires proper methods to administer a reproducible nerve block and to objectively assess pain. In most dogs, femoral and sciatic nerve blocks are achieved using reliable anatomical landmarks that are consistently reproducible (Campoy & Mahler 2013). The use of ultrasound and nerve stimulator guidance improves the success rate of these procedures. The nerve block procedures used in this study both have a low learning curve, low anatomical risks, and a high success rate for complete motor and sensory blocks in dogs (Campoy et al. 2010; Echeverry et al. 2012; O Cathasaigh et al. 2018). Both femoral and sciatic nerve blocks were necessary for regional anesthesia of the pelvic limb and facilitated a complete motor blockade (Marolf et al. 2019).

Considering the objective of the drug delivery system, the extended durations of complete sensory blockade at the digital pad and mid-tibial skin when using TRH (8.0 ± 1.6 h and 10.9 ± 1.6 h, respectively) may be considered insufficient compared with previous reports. However, the blockade duration may be evaluated differently according to the evaluating methods, injection methods, patient species, and the size of related nerves. A human study reported that ropivacaine with PF-72 administered directly to the surgical sites relieved pain for up to 72 h based on patient self-reported pain scores (Choi et al. 2019). AUC of pain score used in the human study should not be equated to the binary outcome at a single timepoint used in this study, and the use of a binary outcome for excluding subjective or biased assessment in nonverbal animals may result in an underestimation of the analgesic duration in this study. An in vivo rat study demonstrated significant pain relief for at least two days based on the weight-load and paw-withdrawal thresholds when bupivacaine with PF-72 was injected into the joint cavity (Kim et al. 2015). PF-72 with ropivacaine injected subcutaneously in the area of the sciatic nerve in rats resulted in pain relief for 24 h based on the paw-withdrawal threshold (Oh et al. 2019). As PF-72 was injected by different methods, different release profiles and the

proximity of the gel to the desired site should be expected. The sensory duration of a partial block is approximately 1.5–2 times longer than that of a complete block, as reported in studies using bupivacaine or ropivacaine to achieve femoral and sciatic nerve blocks in dogs (Portela et al. 2010; Trein et al. 2017). Therefore, when TRH is used with local anesthetic agents for post-operative pain relief, the duration of pain relief, including complete and partial nerve blocks, would be longer than reported by the current study. Further studies that include both partial and complete blockades are needed to determine the period of significant pain relief with PF-72 in a clinical setting.

The volume and concentration of local anesthetic agents are important determinants of the success and duration of complete peripheral nerve blocks (Portela et al. 2010), and a high success rate was needed for complete block of all four nerves in this study. A previous study reported that 0.25% of bupivacaine was inadequate for complete femoral and sciatic nerve blocks and that complete nerve blocks were achieved with 0.15 ml·kg^{-1} of 0.5% bupivacaine (O Cathasaigh et al. 2018). Other studies demonstrated that 0.2 ml·kg^{-1} of 0.5% bupivacaine was adequate for sensory blockade of the sciatic nerve even with ultrasound guidance and it increases the success rate, minimize variable outcomes, and is reproducible (Futema et al. 2002; Marolf et al. 2019). In a preliminary experiment, various concentrations and volumes were tested to achieve complete pelvic-limb blocks. While 0.1 ml·kg^{-1} of 0.25%, 0.1 ml·kg^{-1} of 0.5%, and 0.2 ml·kg^{-1} of 0.25% bupivacaine were noted to have a low possibility of a complete block, 0.2 ml·kg^{-1} of 0.5% bupivacaine resulted in a complete block with a relatively high success rate when the doughnut sign was observed, similar to previous studies. This may be due to the circumferential spread of the local anesthetic agent around the sciatic nerve, and the higher volume of anesthetic agent may lead to more consistent reproducibility (Shilo et al. 2010). In addition, the local anesthetic agent in contact with > 2 cm of the nerve indicates a clinically effective peripheral nerve block because the length of the nerve in contact with the local anesthetic agent determines the success of the nerve block (Campoy et al. 2008). Although measuring the whole distances of local anesthetic agents around the target nerves were limited by bone shadows on the ultrasound images as moving the transducer upward, the local anesthetic agent in contact with > 2 cm of the nerve was observable in this study when using 0.2 ml·kg^{-1} of the local anesthetic agent. The used volumes and concentrations of bupivacaine in this study were chosen to achieve the conditions of complete blockade, which was a requirement for data collection. The results indicate that the concentration and volume of local anesthetic agents used were adequate for sensory and motor blockade.

In this study, the total dose of bupivacaine used per dog was 4 mg·kg^{-1}, which exceeded the clinical dose for cardiotoxicity (2 mg·kg^{-1}). To compare the duration of the effect of a local anesthetic agent by two different treatments on the same dog at the same time, both femoral and sciatic nerve blocks were required in the pelvic limbs. Since the dose of bupivacaine per nerve was fixed as mentioned earlier, the total dose of bupivacaine used per individual increased. If a clinical application is performed based on the findings herein, the total dose of bupivacaine per dog would not exceed the toxic dose because, in a clinical environment, surgery is typically performed on one pelvic limb. Although the blood concentration of the local anesthetic was not measured here, the estimated blood concentration resulting from a 4 mg·kg^{-1} bupivacaine injection was estimated to be 4.16 μg·ml^{-1} based on a previous study using bupivacaine for femoral nerve blocks in dogs; the peak plasma concentration of bupivacaine was 0.78 μg·ml^{-1} when administering 0.75 mg·kg^{-1} of bupivacaine for femoral nerve blocks in dogs without epinephrine (O Cathasaigh et al. 2018). This concentration is lower than that reported to cause cardiovascular collapse in dogs (5.7 μg·ml^{-1}) (Groban et al. 2001). Furthermore, most known convulsing and cardiovascular toxic doses are related to IV injection, but even relatively high plasma concentrations caused by large doses do not usually, because systemic toxicity if a significant dose of local anesthetic is not deposited intravascularly (Rosenberg et al. 2004). The addition of 5 μg·ml^{-1} epinephrine (1:200,000) into the local anesthetic agent reduces its peak plasma concentration without prolonging the duration of its effect (Rosenberg et al. 2004). Although the peripheral application of epinephrine may worsen nerve injury due to physical nerve damage or local anesthetic neurotoxicity in animals, the risk of nerve damage from peripheral block is extremely low in normal conditions (Neal 2003). Though systemic exposure would likely differ depending on the route of administration, subcutaneously injected PF-72 delays the release of the local anesthetic agent, resulting in 60% lower blood concentration of the local anesthetic agent and significantly lower concentration in various tissues compared with the free-form local anesthetic agent in rats (Oh et al. 2019). Epidural administration of lidocaine with hyaluronic acid, the ingredient of TRH, reduced the mean plasma lidocaine concentration by 50% compared with the administration of plain lidocaine in dogs (Doherty et al. 1996). The perineural injection site, addition of epinephrine, and characteristics of TRH suggest that the plasma concentration of bupivacaine may be lower than the toxic dose in dogs even with the administration of 4 mg·kg^{-1} of bupivacaine, although further studies regarding the application of epinephrine in TRH are needed. Fortunately, despite the high dose of bupivacaine used in this experiment for distinct objective evaluation, no related complications were observed. Nonetheless, a lower dose of bupivacaine should be used for further clinical trials and applications as a high dose of bupivacaine may lead to critical complications.

PF-72, which offsets the disadvantages of highly viscous hydrogels, is still relatively viscous and requires increased injection pressure. Although this may be compensated with the use of a large-gauge needle with a short length, an intraneural injection cannot be ruled out owing to the injection pressure. Ultrasound guidance is necessary to prevent an intraneural injection and was used in this study.

This study is not without limitations. This study did not include a liposomal bupivacaine treatment group as an active comparator, and it requires further investigation. This study was confined to regional anesthesia in Beagle dogs; thus, more clinical studies regarding the duration of significant pain relief in various dog breeds and other types of regional anesthesia are necessary. As PF-72 is on the market in Korea as a form that can be mixed with desirable local anesthetics and used in post-operative pain relief in laparoscopic abdominal surgery

in human medicine (Choi et al. 2022), more diverse clinical applications in veterinary medicine also would be expected. Although the objectivity of the assessment has been ensured as much as possible, there is still a limitation of only a single investigator having assessed the dogs. In addition, the instability in the limb that recovered slower affected the weight bearing and walking abilities of the opposite limb and resulted in the overestimation of the locomotor function of the limb that recovered faster.

5 | CONCLUSION

PF-72 extended the duration of regional anesthesia of the pelvic limb achieved using bupivacaine for digital sensory blockade from 3.7 ± 2.0 h to 8.0 ± 1.6 h, for mid-tibial skin sensory blockade from 8.0 ± 1.4 h to 10.9 ± 1.6 h, and for proprioception from 4.6 ± 1.9 h to 9.3 ± 1.6 h. PF-72 is an applicable drug delivery system for extending the duration of regional anesthesia of the pelvic limb using bupivacaine in dogs.

AUTHOR CONTRIBUTIONS

Jiyoung Kim: Conceptualization; Data curation; Formal analysis; Investigation; Methodology; Writing – original draft. Dalhae Kim: Investigation. Donghwi Shin: Investigation. Taehoon Sung: Investigation. Suehyung Rhee: Investigation. Minha Kim: Investigation. Inhyung Lee: Resources; Supervision; Validation. Won-gyun Son: Conceptualization; Data curation; Funding acquisition; Investigation; Methodology; Project administration; Resources; Supervision; Validation; Visualization; Writing – review & editing

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CONFLICT OF INTEREST

No conflicts of interest have been declared.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

STUDY DESIGN AND ETHICAL APPROVAL

This prospective, blinded, randomized study was approved by the Institute Animal Care and Use Committee of the Seoul National University (SNU-200728-4).

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PEER REVIEW

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