Blinded, randomized, placebo-controlled study of the efficacy of bupivacaine liposomal suspension using static body weight distribution and subjective pain scoring in dogs after tibial plateau leveling osteotomy (TPLO) surgery

by

Lauren A. Aldrich

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D.V.M., North Carolina State University, 2017

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Approved by:

Major Professor
David A. Upchurch DVM, MS, DACVS-SA

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Abstract

Objective: To compare the analgesic effect of surgical wound infiltration with liposomal bupivacaine (LB) to saline placebo in dogs after tibial plateau leveling osteotomy (TPLO).

Study Design: Blinded, randomized, placebo-controlled clinical prospective study

Animals: 15 client-owned dogs receiving liposomal bupivacaine and 17 dogs receiving an equivalent volume of saline placebo, all with confirmed unilateral cranial cruciate ligament insufficiency.

Methods: Preoperatively and up to 48 hours after surgery, Glasgow Composite Measure Short Form (CMPS-SF) pain scores were assigned and using a weight distribution platform, static body weight distribution ($\%BW_{dist}$) to the operated limb was measured. Postoperatively, dogs also received carprofen 2.2 mg/kg subcutaneously every 12 hours. Rescue analgesia was provided. Treatment success was defined as not requiring rescue analgesia over the 48 hour postoperative period.

Results: There was no difference between treatment success, postoperative opioid consumption, CMPS-SF pain scores, or $\%BW_{dist}$ in dogs that received surgical wound infiltration with LB compared with those receiving saline placebo, following TPLO. There was no linear correlation between CMPS-SF pain scores and $\%BW_{dist}$.

Conclusion: For the population of dogs that underwent TPLO and received postoperative carprofen at our institution, LB did not provide an analgesic effect discernable by success/failure analysis, CMPS-SF pain scores, or $\%BW_{dist}$ measurement using a weight distribution platform, compared with saline placebo.

Clinical Significance (or Impact): LB may not provide detectable analgesia for dogs recovering from TPLO and receiving postoperative carprofen.

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WSAVA – World Small Animal Veterinary Association

NSAID – Non-steroidal anti-inflammatory drug

FDA – U.S. Food and Drug Administration

TPLO – Tibial plateau leveling osteotomy

OPR – Opioid pain relievers

LA – Local anesthetic

IVRA – Intravenous regional anesthesia

PEG – Polyethylene glycol

MVL – Multivesicular liposome

AUC – Area under the curve

RCTs – Randomized clinical trials

CMPS-SF - Short-form Glasgow Composite Measure Pain Scale

CSU-CAPS – Colorado State University Canine Acute Pain Scale

PNT - Pressure nociceptive threshold

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Chapter 1 - Liposomal Bupivacaine Literature Review

An unmet need for bridging analgesia

Maintaining adequate analgesia as veterinary patients are transitioned from hospital to home care is challenging. In hospital, specialized equipment and trained staff enable implementation of multimodal postoperative analgesia protocols. For dogs undergoing orthopedic surgery, the World Small Animal Veterinary Association (WSAVA) Global Pain Council recommends that analgesic protocols include local/regional anesthesia, non-steroidal anti-inflammatory drugs (NSAIDs) and opioids used in the pre- and postoperative periods. The WSAVA also recommends adjunct analgesia, including preoperative alpha-$_2$ agonists, a lidocaine patch or diffusion catheter, and cold therapy.[1]

Provision of up to 24 hours of regional analgesia is possible with single-dose epidurals using morphine-bupivacaine 0.5% [2,3] or morphine-ropivacaine 1%[4], as well as by femoral and sciatic blockade with 0.5% bupivacaine[2] and saphenous or sciatic blockade with ropivacaine 1%.[5] As pain relief from epidural anesthesia or nerve blockade fades and the patient is weaned from injectable opioids, clinicians and owners may continue to use U.S. Food and Drug Administration (FDA) approved NSAIDs[6] for dogs who remain appropriate candidates:[1,7] those that continue to eat and drink normally, and that are not experiencing potential side effects.[8]

For dogs that are not NSAID candidates, there are limited options for oral medications with evidence of efficacy in controlling acute orthopedic pain. A systematic review and meta-analysis of the efficacy of tramadol for postoperative pain management in dogs concluded with moderate certainty that, compared with no treatment, the drug probably results in a reduction in the need for rescue analgesia.[9] Among the oral medications tested in dogs after undergoing tibial plateau leveling osteotomy (TPLO) surgery specifically, pain scores in patients receiving either

hydrocodone-acetaminophen or tramadol were similar but were not compared to placebo or an FDA-approved positive control; 29% of all patients enrolled in that study required rescue analgesia.[10] Another study found that hydrocodone provided inferior analgesia compared to firocoxib in dogs following TPLO surgery, with 50% of patients receiving extended-release hydrocodone requiring rescue analgesia vs. 11% needing rescue analgesia within the firocoxib group. Pain scores were higher and percent body weight supported by the operated limb was lower for dogs receiving hydrocodone.[11] To date, there are no published analgesic efficacy studies of oral codeine in dogs.

Transdermal fentanyl delivery products come close to providing an ideal solution for postoperative pain relief in an outpatient setting. A single dose, sustained release, topical fentanyl solution gained FDA approval and became available in the United States in 2012[12]. The product provided therapeutic plasma levels of fentanyl within 2 to 12 hours of application, sustained for approximately four days[13] and was shown to provide non-inferior analgesia compared with repeated injection of oxymorphone in a population of dogs undergoing soft-tissue or orthopedic surgeries.[14] Unfortunately, the manufacturer no longer markets this product. Currently, clinicians may opt for extra-label use of transdermal fentanyl patches approved for use in humans. There is evidence for fentanyl patches providing sustained postoperative analgesia in dogs,[15-17] but drawbacks of fentanyl patches include the delay of approximately 12-24 hours before reaching therapeutic plasma concentrations, potential variability in systemic absorption,[18] the possibility of overdose with oral transmucosal or enteral absorption,[19,20] and the risk of accidental or intentional misuse by owners or visitors to the home.

The challenge of providing safe, effective postoperative bridging analgesia is not unique to veterinary medicine. While human physiology is such that oral opioid pain relievers (OPRs)

are effective for moderate to severe pain,[21] their associations with adverse events, persistent use beyond the time of need, and addiction[22] have led to an urgent call for the substitution of non-opioid analgesics.

A long-acting local anesthetic (LA) could provide a solution to this One Health need for safe, targeted and well-tolerated analgesic products that complement or substitute for opioids and NSAIDs in patients transitioning to home care.

Local anesthetic agents: mechanism of action, pharmacokinetics and toxicity

Development of a long-acting LA product begins with an understanding of the molecular properties that influence mechanism of action, potency, duration of action and safe use. LAs provide temporary relief from pain by reversibly blocking sensation and motor function of peripheral nerves. Their primary mechanism of action is to block voltage-gated Na+ channels that exist in nerve cell membranes. In the presence of a therapeutic concentration of a LA, a stimulated nociceptor cannot depolarize enough to generate an action potential, so the sensation of pain will not propagate to the central nervous system to be modulated or perceived.[23] LAs are the only class of analgesics that can block transmission of pain,[24] preventing repeated stimulation of primary afferent neurons, thereby preventing central sensitization.[25]

The mammalian LA receptor site (Na_{III}) exists within the inner pore of the Na+ channel. Some antiarrhythmic and anticonvulsant drugs also interact with Na_{III},[26] and our understanding of how these structurally diverse drugs can interact with a single binding site is still evolving.[24,27] The authors of a recent molecular dynamics study of diverse sodium channel blockers proposed a common pharmacophore, or set of general molecular features that a ligand must possess in order to interact with a receptor. The pharmacophore that describes known Na_{III} ligands consists of a cationic moiety (part of a molecule) and an aromatic moiety linked by an intermediate chain that is either an amide or an ester. It is the cationic moiety that interacts with Na_{III}. This binding interaction can be achieved directly by an ammonium functional group or by an electroneutal functional group that clamps a nearby sodium ion into the receptor.[27] Drugs used historically[28] (such as cocaine) and currently (lidocaine, bupivacaine, ropivacaine) as LAs possess an ammonium functional group. On the opposite end of the molecule, the aromatic moiety of a LA

interacts with a hydrophobic region of the inner Na+ channel pore, helping the ligand dock into the channel.[27]

Physicochemical properties of LAs influence how these drugs move through the body from the site of injection. Lipophilicity, degree of protein binding and vasoactivity are the major physiochemical properties that influence systemic absorption, and in turn, duration of effect. Highly lipophilic LAs tend to associate with the lipid-rich neural membrane and surrounding subcutaneous fat, resisting systemic absorption. Lipophilicity is a determinant of LA potency as well, as a more lipophilic drug is better able to permeate and diffuse across the neuron cell membrane, reaching its embedded sodium channels. The primary determinant of lipophilicity is the extent to which a molecule exists in a neutral, non-ionized state at physiologic pH – as opposed to its ionized, conjugate base state – as described by its partition coefficient.[24,29] Among modern LAs commonly used in small animals, bupivacaine is more lipophilic than ropivacaine, which is more lipophilic than lidocaine.[24,30,31]

A greater degree of protein binding is associated with longer duration of action, which is thought to be a result of a drug's affinity for proteins within the sodium channel.[31] Lidocaine has a moderate degree of protein binding, while bupivacaine/levobupivacaine, and ropivacaine are highly protein-bound.[24]

Uptake of a drug into systemic circulation from its site of administration is slower if the local tissue is relatively avascular or if the local vasculature undergoes vasoconstriction. Local anesthetics induce vasoconstriction at low doses and vasodilation at high doses. At therapeutic tissue concentrations, all aminoamide LAs produce some degree of vasoconstriction. Vasoconstriction potency is influenced primarily by lipophilicity, and to lesser degrees drug potency, pKa and molecular weight. The vasoconstriction potency is greatest for

levobupivacaine, followed by ropivacaine and lidocaine.[32] This effect contributes to the longer duration of action for levobupivacaine and ropivacaine compared with lidocaine.

LAs can be administered by infiltration of local tissues, injection around peripheral nerves, or neuraxial injection. LAs will also produce motor blockade when used peripherally or neuraxially[24] – for example, when lumbosacral epidurals result in hind limb paralysis[33] – which has intraoperative benefits but postoperative disadvantages in terms of patient mobility. Tissue infiltration of the surgical site with a LA provides targeted analgesia while avoiding motor blockade.[34]

All local anesthetics have the potential to cause systemic effects, with signs of neurotoxicity preceding cardiovascular compromise. Most severe toxicity events result from inadvertent intravascular injection of an appropriate dosage of local anesthetic, but it is possible for toxicity to follow large-volume tissue infiltration of LA.[35] Humans may experience numbness of the tongue, lightheadedness, visual disturbances and muscle twitching initially. As plasma levels of local anesthetic increase, signs progress to seizure, coma and cardiac arrest.[36] A similar progression is reported in animals.[24] Bupivacaine is more cardiotoxic than lidocaine and ropivacaine. This is thought to be due primarily to its blockade of calcium channels in cardiomyocytes, slowing cardiac depolarization, shortening of the refractory period and decreasing contractility.[37] [35] Ropivacaine has lower potential for neurologic and cardiotoxicity in humans and animals compared to bupivacaine.[24,36]

An ideal LA candidate for long acting, targeted postoperative pain relief without motor blockade would be lipophilic, highly protein-bound, and well tolerated. However, research and development efforts have not yet yielded a LA with an intrinsic duration of action beyond bupivacaine/levobupivacaine's outer limit of 10 hours.[24] Extension of the analgesia provided by

LA tissue infiltration therefore requires modification of the local tissue environment, use of delivery systems or development of extended-release LA formulations.

Prolonging duration of analgesia of local anesthetics

One of the first techniques developed to prolong the duration of effect of a LA involves physically restricting its systemic absorption by use of a tourniquet, now known as intravenous regional anesthesia (IVRA).[38] Bier first described this technique for use in human limb surgery in 1908, but IVRA did not become widespread until Holmes reintroduced it with some modifications in 1963.[39] In dogs, the most recently reported use of an IVRA technique for surgery involved exsanguination of the forelimb with an elastic bandage applied distally to proximally, application of a non-pneumatic, pediatric tourniquet over the mid-antebrachium, then removal of the elastic bandage to establish an ischemic tissue environment. Regional anesthesia was produced by injection of lidocaine 0.5% solution (3 mg/kg) into the cephalic vein, 10 minutes before performing pancarpal arthrodesis. This study showed that during an approximately one hour surgery, IVRA provided analgesia similar to traditional brachial plexus block with lidocaine/ropivacaine, without complications. The study also demonstrated that within about 25 minutes of removing the tourniquet at the end of surgery, dogs began perceiving a pinching stimulus to their toes.[40] IVRA modestly extends the duration of analgesia provided by a LA during surgery, but is not recommended beyond 90 minutes due to the risk of ischemic injury.[41] IVRA is best used to extend analgesia for anesthetized animals, as awake animals do not tolerate tourniquets, and is not intended to provide bridging analgesia.

A chemical tourniquet effect can be achieved in peripheral (as well as neuraxial sites) by the addition of a vasoconstricting drug such as epinephrine[42,43] to LAs. [39,40] The mechanism of action is to decrease local blood flow via α_1 receptor agonism. Addition of epinephrine to lidocaine for tissue infiltration in humans provided complete blockage of a pinprick sensation at 5 hours in 50% of participants, while the lidocaine-only treatment failed to block the sensation in

all participants.[42] In horses, the duration of analgesia for procaine was increased significantly (from 3 to 4 hours) by the addition of epinephrine in a hoof withdrawal heat latency model of analgesic efficacy.[43] Studies are limited in veterinary species, and the addition of epinephrine to bupivacaine for local tissue infiltration has not been described.

Drug delivery systems have been developed to overcome the limited duration of action of single-dose LA tissue infiltration. Continuous wound infiltration with local anesthetics using flexible, indwelling catheters has shown benefits similar to epidural and intravenous analgesia in a meta-analysis of humans recovering from laparotomy and sternotomy.[44] The use of wound soaker catheters in postoperative dog and cat patients (mainly undergoing limb amputation) was described in a single retrospective case series as feasible and well-tolerated, with the most common complication being inadvertent disconnection of the catheter from the extension set (7.7%), followed by incisional infection (5.3%). These patients received either continuous infusion with lidocaine or intermittent boluses of bupivacaine.[45] While wound soaker catheters can be used to provide analgesia beyond the 24 hour outer limit of an epidural or nerve blockade, most clinicians would consider them inappropriate for continued use at home.[45]

FDA-approved patches coated with lidocaine-infused adhesive material have been developed for relief of local pain in people with postherpetic neuralgia, a complication involving allodynia after *Herpes zoster* infection.[46] Lipoderm[47] is one of a handful of FDA-approved patches that are labeled for application to intact skin for up to 12 hours of continuous wear per day. The patch is meant to be applied directly over the area of skin that is most painful, providing analgesia by direct diffusion of lidocaine.[47] Minimal systemic absorption of lidocaine is described after application of lidocaine patches to humans,[47] cats[48] and dogs.[49,50] There are a limited number of clinical studies of the patch's efficacy in relieving acute postoperative pain in

humans, with no overall difference in pain scores, opioid consumption or length of hospital stay in treated patients.[51] Application of peri-incisional 5% lidocaine patches did not result in lower post-operative pain scores compared to placebo patches in dogs that underwent ovariohysterectomy.[52] In dogs undergoing hemilaminectomy for single acute compressive thoracolumbar intervertebral disc extrusion, application of 5% lidocaine patches along both sides of the surgical incision did not reduce rescue analgesia requirement or pain scores compared to placebo.[53] Although lidocaine patches are easily placed, and are tolerated by dogs and cats for up to 72 hours without toxicity, this delivery system has no demonstrated analgesic efficacy to date.

Evolution of liposomal drug delivery

Liposomal delivery of LAs may meet the needs for safe, targeted, bridging post-surgical analgesia. The basic unit of a liposomal delivery system is the liposome, a microscopic vesicle composed of one or more concentric spherical phospholipid bilayers that enclose an aqueous central space. Having lipid and aqueous regions, a liposome is able to carry either lipophilic (associated with the lipid membrane) or hydrophilic drugs (trapped within the internal aqueous compartment). When drugs are carried by liposomes, they are released as the vesicles eventually break down and are protected from early degradation and inactivation. The liposome itself is pharmacologically inert, non-immunogenic and minimally toxic, as it is typically composed of natural phospholipids that are similar to mammalian cell membranes.[54,55]

Liposomal delivery systems were initially developed and launched for clinical use in the late 1980s and early 1990s to enhance the safety of intravenously delivered drugs with a narrow therapeutic index, such as doxorubicin and amphotericin. This first generation of liposomes is now referred to as conventional liposomes. They consist of simple lipid bilayer vesicles, less than 1 micron in diameter, designed to carry an internal payload of water-soluble drugs.[54] In studies involving mice and humans, liposomal doxorubicin was shown to have a longer circulation time and decreased uptake by the heart compared to the free drug, resulting in lower cardiotoxicity.[56,57] _ENREF_53

Conventional liposomes were found to be eliminated by plasma opsonization (adsorption of proteins to the phospholipid surface) with subsequent phagocytosis by resident macrophages in the reticuloendothelial systems of the liver, spleen, kidneys, bone marrow, lungs and lymph nodes.[55] This targeting of liposomes to monocytes and macrophages was inevitably turned to advantage – for instance, with therapeutic bombing of bacteria-laden macrophages with

liposomal antibiotics[58] and administration of liposomal chemotherapeutics to patients with tumors of the liver and spleen.[57]

Subsequent generations of liposomes have been modified to evade phagocytosis and make drug targeting even more specific.[54] Incorporation of polyethylene glycol (PEG) into the liposome shell helps the vesicles resist opsonization and evade recognition by macrophages, to prolong the duration of drugs in circulation, in order to improve accumulation in diseased tissues and reduce side effects. [59-61] Manipulation of the size and charge of the lipid membranes, addition of ligands such as antibodies, proteins and carbohydrates into the phospholipid bilayers allow more targeted drug delivery for chemotherapeutics, vaccines[62] and pain medication.[55,62-65]

Liposome-based analgesia was introduced in 2004, with the FDA approval of extended release epidural morphine injection (DepoDur).[66] Given as a single-dose lumbar epidural administration, this formulation provides 48 hours of analgesia (and sometimes nausea) following lower abdominal and lower extremity surgery in humans.[67,68] DepoDur's extended release of morphine was made possible by a novel liposomal delivery system, DepoFoam, which was developed to serve as a depot for sustained release of drugs into extravascular spaces. The basic unit of DepoFoam is the multivesicular liposome (MVL), a conglomerate of non-concentric, closely-packed lipid bilayers. The DepoFoam MVL is a 10 to 30 micron diameter complex of liposomes that constantly translate, merge and divide.[69,70]

The modification that led to the development of the MVL was the addition of a neutral lipid (a triglyceride) to the standard amphipathic phospholipid solution that undergoes double emulsification to produce liposomes. The triglyceride fills the tiny triangular spaces where the spherical lipid bilayers touch, stabilizing the junctions between individual vesicles. Without this triglyceride filler, the emulsification process yields concentric spheres of liposomes that degrade

much more quickly than the honeycombed MVL liposomes. Encapsulated drug is released by two mechanisms: by permeation through the outermost bilayer membranes and by sudden escape as the outermost vesicles degrade. The rate of release of the active drug can be modified by altering the ratio of long-chain (more stabilizing for slow release) to short-chain (fast release) triglycerides added during production. Another means of slowing the rate of release of the active drug is to increase the osmolarity of the aqueous phase in which the drug is dissolved.[69] DepoFoam enabled the next leap in liposome-based analgesia: the first FDA-approved long-acting, non-opioid analgesic.

Safety and Pharmacokinetics of liposomal bupivacaine

Introduced in 2011 and still used widely for post-surgical pain in people, Exparel,[71] provides slow release of bupivacaine (13.3 mg/mL) from a DepoFoam carrier. Initial approval was for analgesia in adults after single-dose surgical site infiltration.[70] The FDA has recently approved expansion of Exparel's use for surgical site infiltration to patients over 6 years old, making it the first and only long-acting non-opioid analgesic available to pediatric patients.[71] In 2016, the FDA approved Nocita,[72] a seemingly identical liposomal bupivacaine (13.3 mg/mL) product labeled for single-dose surgical site infiltration following cranial cruciate ligament surgery in dogs older than 5 months of age.[73] In 2018, Exparel gained FDA approval for interscalene brachial plexus nerve block in adults; that year, Nocita gained FDA approval for use in peripheral nerve blockade before onychectomy in cats.[74] A review of liposomal bupivacaine for regional anesthesia is beyond the scope of this thesis.

In its safety and tolerability study for original FDA approval, Exparel was infiltrated by a moving needle technique into the deep and superficial subcutaneous tissues of rabbits and dogs. The tissue layers were closed over polypropylene mesh in an inguinal hernia repair model. Exparel dosages ranged from 9 to 30 mg/kg, compared with a control group receiving 9 mg/kg non-liposomal bupivacaine. One rabbit died after receiving a 9 mg/kg dose of Exparel, which was attributed to systemic toxicity in a particularly sensitive species. On histopathologic analysis, there was no evidence of local tissue toxicity in rabbits or dogs receiving Exparel. In 8/24 rabbits receiving Exparel, surgical site tissues sampled at 15 days showed minimal to mild granulomatous inflammation, consistent with a foreign body-type reaction. The authors called this a normal reaction to the liposomal component of the drug, rather than an adverse reaction. All surgical wounds healed as expected.[75]

In this safety study, pharmacokinetics of Exparel was favorable. Peak plasma concentrations resulting from Exparel at 9 mg/kg were lower than from non-liposomal bupivacaine at 9 mg/kg (about 1.7 and 5.8 times lower in rabbits and dogs, respectively). The peak plasma concentration of bupivacaine in animals receiving 30 mg/kg bupivacaine Exparel was lower than that of animals receiving 9 mg/kg non-liposomal bupivacaine. Based on pharmacokinetic area under the curve (AUC) comparisons, plasma concentrations were indeed sustained for animals receiving Exparel. These data demonstrated that Exparel provides sustained release of bupivacaine after tissue infiltration in rabbits and dogs, with greater morbidity and mortality in rabbits than in dogs. [75]

In people, plasma concentration of Exparel was characterized as bimodal, with an initial peak at ¼ to 2 hours and a second peak at 12 to 24 hours. The initial peak was thought to be due to the known presence of a small fraction of free (extraliposomal) bupivacaine in the suspension.[76] Although peak plasma concentrations were higher and half-life was longer in patients with moderate hepatic impairment, the differences were not clinically significant and dose adjustments were not recommended.[70] In contrast to the post-injection granulomatous inflammation described in rabbits, similar pathology has not been reported in humans dosed with Exparel.[77]

The safety of Nocita for surgical site infiltration was evaluated in 123 dogs following either tibial plateau leveling osteotomy, lateral suture stabilization or tibial tuberosity advancement. A control group of 59 dogs received an equal volume of saline placebo. For treated dogs, the three most common adverse events were: discharge from the incision (3.3% incidence), gross incisional inflammation (2.4%), and vomiting (2.4%). For dogs receiving placebo, no patients experienced those events; the three most common adverse events were

surgical limb edema +/- erythema (5.1%), soft stool/diarrhea (1.7%) and inappetence (1.7%). Statistical significance of these events was not described.[73]

In a separate safety study evaluating effects of repeated dosing, Nocita tissue infiltration twice weekly at doses of 5.3, 16 and 26.6 mg/kg did not result in signs of systemic toxicity nor electrocardiogram abnormalities in the study dogs. Granulomatous inflammation was observed in at least one dog in each of the Nocita groups, and this was described as a normal tissue response to the liposome component of the drug. The FDA determined Nocita to be safe for single-dose surgical site infiltration in dogs undergoing surgery for cranial cruciate ligament rupture.[73]

Clinical efficacy of liposomal bupivacaine in humans and animals

Clinical efficacy of LB has been extensively studied in humans. A 2021 systematic review of 63 randomized clinical trials (RCTs) evaluated the post-surgical analgesic efficacy of liposomal bupivacaine used for local tissue infiltration or nerve block in humans. The distribution of studies by surgical specialty was: 33 orthopedic-related studies, 10 for general surgery, 9 for obstetric/gynecology, 4 for oral/maxillofacial surgery and 7 others. The review concluded that LB did not significantly reduce pain scores compared with placebo, standard bupivacaine or standard-of-care, non-bupivacaine analgesic agent in 74.58% of studies. In 4 out of 10 studies of analgesic efficacy comparing LB to placebo, pain scores were not significantly different between the two groups. In 20 out of 29 studies comparing LB to standard bupivacaine, pain scores were not significantly lower for people receiving LB. Pain scores did not differ between treatment groups in any of the 16 studies comparing LB with non-bupivacaine analgesic.[78]

In this review, surgical specialty was associated with the probability of LB providing superior pain relief. LB performed particularly poorly with respect to total knee arthroplasty, with a reduction of pain in only 2 out of 20 studies – one using LB vs. standard bupivacaine for local tissue infiltration[79] and another using LB vs. placebo for femoral nerve block.[80]

Among the 56 studies in the review that evaluated opioid use, LB did not result in significantly decreased postoperative opioid consumption in 85.71%, regardless of the comparative agent (placebo, standard bupivacaine or non-bupivacaine analgesia).[78]

Furthermore, this review found that clinical trials disclosing a financial or employee relationship with the manufacturer of LB were over 14 times more likely to be associated with a

superior treatment effect of LB (OR: 14.31 [95% CI, 2.8, 73.10], P = 0.0001) and over 12 times more likely to report decreased opioid consumption in patients receiving LB (OR: 12.35 [95% CI 1.40, 109.07], P = 0.0237). In light of these findings, the authors expressed concern about financial conflict of interest possibly having an influence on outcomes of RCTs, however they could not test causation.

Another concern expressed by these authors was the underreporting of trial results. Of the total number government-registered RCTs that were started, 46.7% were either completed without published results or were never completed. Publication bias for studies with positive outcomes is recognized in anesthesia literature[81] and across other disciplines.[82] The authors warned practitioners to consider publication bias when interpreting results indicating superiority of LB.[78] Since Exparel costs more than standard bupivacaine, its use cannot be justified in humans if it does not provide superior analgesia, reduce opioid consumption or reduce hospital readmissions in comparison to placebo or standard of care protocols.

In dogs, the analgesic efficacy of surgical site infiltration with Nocita has been reported in two clinical trials. In a randomized, placebo-controlled, masked study,[83] LB was shown to provide local analgesia in dogs following lateral retinacular suture placement with arthrotomy. Outcome measures included subjective pain scores based on the short-form Glasgow Composite Measure Pain Scale (CMPS-SF)[84] and success/failure analysis. Pain scores were lower for dogs receiving LB versus saline placebo at all time points (0 to 60 hours) post-operatively except at 72 hours, at which time only two dogs remained in the placebo group. Treatment success, defined as percent of dogs not requiring rescue analgesia, was significantly higher for dogs receiving LB versus placebo over the 0-24, 0-48 and 0-72 hour post-operative intervals. This study was funded

by the manufacturer of Nocita, the corresponding author was a paid consultant and two of the authors were employees of said manufacturer.[83]

A subsequent randomized, masked clinical study evaluated analgesic efficacy of surgical site infiltration with LB compared with standard bupivacaine in dogs recovering from tibial plateau leveling osteotomy (TPLO) with arthrotomy. In addition to pain scoring with the CMPS-SF and the Colorado State University Canine Acute Pain Scale[85] (CSU-CAPS), this study used pressure nociceptive threshold (PNT) testing as an objective outcome measure of pain. A commercially available pressure algometer was used to apply force to over the medial joint space of the stifle, measuring maximum tolerated pressure before and at various times after surgery. Over the 48-hour postoperative period, dogs administered LB at wound closure were less likely to need rescue analgesia. Total opioid consumption was reduced in the LB group as well.[86] The opioid-sparing benefit conferred by LB was attributed to the longer duration of effect[83] for that drug compared with standard bupivacaine.[87] Interestingly, neither pressure threshold measurements nor pain scores differed significantly between treatment groups at any time point. This study reported no financial conflicts of interest.

Conclusions and rationale for further research

There is a need within human and veterinary medicine for safe, targeted and well-tolerated analgesic products that complement or substitute for opioids and NSAIDs in patients transitioning to home care. Standard formulations of LAs and current LA delivery systems fall short of meeting this need for bridging analgesia.

Both clinical studies of LB efficacy in dogs[83,86] found that LB reduced the requirement for rescue analgesia, however the more recent study did not find that LB had a significant effect on pain scores or PNT values. There were differences in methodology between these studies that may have contributed to the difference in results. First, TPLO surgery performed in the second study may have incited pain at a deeper anatomic location than the pain inflicted by the lateral suture stabilization performed in the first surgery. Since LB is not administered into bone marrow or periosteum, perhaps the analgesia provided by LB could be less complete for dogs recovering from TPLO. Second, LB was not compared to placebo in the more recent study, so it is possible that the treatment effect of LB and standard bupivacaine were not different enough to be discriminated by pain scoring or PNT. Third, dogs in the second study received twice-daily carprofen as adjunct analgesia, while the dogs in the first study had received no adjunct analgesia postoperatively. The background analgesia provided by carprofen may have blunted the treatment effect detected by pain scoring and PNT.

As TPLO is commonly performed orthopedic procedures performed in dogs, finding a positive treatment effect of LB in dogs recovering from TPLO could benefit many animals. Conversely, finding a negative treatment effect could save owners considerable expense. To date, there are no reported randomized, placebo-controlled, masked clinical trials without financial conflicts of interest that evaluate the efficacy of LB in dogs.

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Chapter 2 - Blinded, randomized, placebo-controlled study of the efficacy of bupivacaine liposomal suspension using static body weight distribution and subjective pain scoring in dogs after tibial plateau leveling osteotomy (TPLO) surgery

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"Blinded, randomized, placebo-controlled study of the efficacy of bupivacaine liposomal suspension using static body weight distribution and subjective pain scoring in dogs after tibial plateau leveling osteotomy (TPLO) surgery"

Lauren A. Aldrich, DVM

David A. Upchurch, DVM, MS, DACVS-SA

James K. Roush, DVM, MS, DACVS

Department of Clinical Sciences, College of Veterinary Medicine, Kansas State University, Manhattan, Kansas. Dr. Aldrich's present address is BluePearl Specialty and Emergency Pet Hospital, 9500 Marketplace Rd., Fort Myers, FL 33912.

Correspondence

David Upchurch, 1800 Denison Ave., Manhattan, KS 66506.

Email: dupchuda@vet.k-state.edu

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Author Contributions:

Aldrich, LA, DVM: Study conception, study design, acquisition and collation of raw data, writing and drafting of the work, critical revision and final approval of the manuscript, and agreement to be accountable for all aspects of the work.

Upchurch, DA, DVM, MS, DACVS-SA: Study design, interpretation of the data, critical revision and final approval of the manuscript, and agreement to be accountable for all aspects of the work.

Roush, JK, DVM, MS, DACVS: Statistical interpretation of data, critical revision and final approval of the manuscript, and agreement to be accountable for all aspects of the work.

Abstract

Objective: To compare the analgesic effect of surgical wound infiltration with liposomal bupivacaine (LB) to saline placebo in dogs after tibial plateau leveling osteotomy (TPLO).

Study Design: Blinded, randomized, placebo-controlled clinical prospective study

Animals: 15 client-owned dogs receiving liposomal bupivacaine and 17 dogs receiving an equivalent volume of saline placebo, all with confirmed unilateral cranial cruciate ligament insufficiency.

Methods: Preoperatively and up to 48 hours after surgery, Glasgow Composite Measure Short Form (CMPS-SF) pain scores were assigned and using a weight distribution platform, static body weight distribution ($\%BW_{dist}$) to the operated limb was measured. Postoperatively, dogs also received carprofen 2.2 mg/kg subcutaneously every 12 hours. Rescue analgesia was provided. Treatment success was defined as not requiring rescue analgesia over the 48-hour postoperative period.

Results: There was no difference between treatment success, postoperative opioid consumption, CMPS-SF pain scores, or $\%BW_{dist}$ in dogs that received surgical wound infiltration with LB compared with those receiving saline placebo, following TPLO. There was no linear correlation between CMPS-SF pain scores and $\%BW_{dist}$.

Conclusion: For the population of dogs that underwent TPLO and received postoperative carprofen at our institution, LB did not provide an analgesic effect discernable by success/failure analysis, CMPS-SF pain scores, or $\%BW_{dist}$ measurement using a weight distribution platform, compared with saline placebo.

Clinical Significance (or Impact): LB may not provide detectable analgesia for dogs recovering from TPLO and receiving postoperative carprofen.

Introduction

A challenge of the immediate postoperative period is maintaining adequate analgesia as veterinary patients are transitioned from hospital to home care. As pain relief from local anesthetic tissue infiltration, epidural anesthesia or nerve blockade fades and the patient is weaned from injectable opioids, FDA-approved non-steroidal anti-inflammatory drugs (NSAIDs) are available for continued analgesia.[1] For dogs that are not NSAID candidates,[2-4] oral narcotics and lidocaine patches are available, but evidence of their efficacy in controlling acute postoperative pain is lacking.[5-9] There is evidence for fentanyl patches providing sustained postoperative analgesia in dogs[10-12] but drawbacks include a delay of about 12-24 hours to reach therapeutic plasma concentrations, variability in systemic absorption,[13] the possibility of overdose with oral transmucosal or enteral absorption,[14,15] and the risk of accidental or intentional misuse by owners or visitors to the home.[16,17]

The need for safe, targeted and well-tolerated analgesic products for postoperative pain relief in humans and animals has led to the development of FDA-approved liposomal bupivacaine (LB) products that provide extended release of the local anesthetic into infiltrated tissues.[18,19] The efficacy of LB in reducing pain scores and opioid consumption in people recovering from surgery was evaluated in a 2021 systematic review of 63 randomized clinical trials (RCTs). LB did not significantly reduce pain scores compared with placebo, standard bupivacaine or non-bupivacaine analgesic agent in 74.58% of studies measuring pain. LB failed to reduce postoperative opioid consumption in 85.71% of studies evaluating opioid use. Furthermore, this review found that clinical trials disclosing a financial or employee relationship with the manufacturer of LB were 14.31 times more likely show pain relief and 12.35 times more likely to report decreased opioid consumption in patients receiving LB.[20]

In dogs, the analgesic efficacy of single-dose surgical site infiltration with LB (Nocita; Elanco Animal Health, Greenfield, Indiana) has been reported in two clinical trials. The first was a randomized, placebo-controlled, blinded study of LB use in dogs after lateral retinacular suture placement with arthrotomy. Dogs receiving LB were less likely to require rescue analgesia and had lower pain scores between 0 and 60 hours after surgery, based on the short-form Glasgow Composite Measure Pain Scale (CMPS-SF).[21,22] This study disclosed financial relationships with the manufacturer of Nocita.[21]

A subsequent randomized, masked clinical study evaluated the efficacy of surgical site infiltration with LB compared with standard bupivacaine in dogs receiving carprofen after tibial plateau leveling osteotomy (TPLO) with arthrotomy. In addition to pain scoring with the CMPS-SF and the Colorado State University Canine Acute Pain Scale (CSU-CAPS),[23] this study used a pressure algometer to measure mechanical thresholds as an objective measure of pain. Over the 48-hour postoperative period, dogs administered LB were less likely to need rescue analgesia and had reduced opioid consumption. However, neither pain scores nor pressure nociceptive threshold measurements were different between treatment groups at any time point. This study reported no financial conflicts of interest.[24]

Further exploration of an objective outcome measure sensitive to acute postoperative orthopedic pain is therefore warranted. An emerging objective measure of limb pain in dogs is $\%BW_{dist}$, the percentage of total body weight supported by a given limb at a natural stance. Measurements of $\%BW_{dist}$ have shown consistency over time, sensitivity to limb lameness, and changes in limb use after TPLO and total hip arthroplasty. [25,26] [27-31] To our knowledge, no study has compared $\%BW_{dist}$ with subjective pain scores in dogs; this seems a rational next step in evaluating whether $\%BW_{dist}$ is an appropriate outcome measure of acute postoperative pain.

As TPLO is one of the most commonly performed orthopedic procedures performed in dogs, finding a positive treatment effect of LB in dogs recovering from TPLO could benefit many animals. Conversely, finding a negative treatment effect could eliminate unnecessary client expenses. To date, there are no reported randomized, placebo-controlled, masked clinical trials that evaluate the efficacy of LB in dogs undergoing TPLO.

The prospective study reported here was conducted to compare outcomes of client-owned dogs receiving carprofen and single-dose tissue infiltration with LB or saline placebo after undergoing TPLO. Our objectives were to (1) compare CMPS-SF pain scores between treatment groups at time points up to 48 hours postoperatively; (2) compare the need for rescue analgesia between groups during the first 48 hours postoperatively; (3) compare the number of rescue opioid doses relative to treatment group size (4) compare $\%BW_{dist}$ between treatment groups up to 48 hours postoperatively; and (5) describe the statistical relationship between CMPS-SF pain scores and $\%BW_{dist}$. The null hypotheses we tested were that: no significant difference would exist between CMPS-SF pain scores or $\%BW_{dist}$ for the treatment groups at any time point; no difference would exist between treatment groups over the duration of the study for rate of treatment success or the number of rescue opioid doses relative to group size: and that no meaningful statistical relationship would exist between $\%BW_{dist}$ and CMPS-SF pain scores.

Materials and Methods

Dogs

Client-owned dogs scheduled for TPLO between July 2019 and July 2020 for confirmed unilateral cranial cruciate ligament (CCL) insufficiency of any duration were eligible for inclusion in the study. The study was approved by our institution's animal care and use committee. Written informed owner consent was obtained for all enrollees. Screening was completed on the day before surgery, and candidates were subject to physical and orthopedic examinations, brief sedation for tibia/fibula radiographs for surgical planning, and hematologic analysis (complete blood cell count and serum biochemistry). Exclusion criteria were: age less than 1 year; current or historic bilateral CCL insufficiency; other clinically evident orthopedic disease; neurologic disease; uncontrolled diagnosed or clinically suspected systemic disease; any surgery within the previous 14 days; short acting corticosteroid use within the previous 7 days or repository steroid use within the previous 2 months; NSAID use other than carprofen within the previous 7 days; use of other analgesics within the previous 48 hours; and temperament that might interfere with subjective pain scoring or stance analysis. Demographic information gathered included unique patient identifier, age, sex, breed, body weight, affected hind limb, and estimated duration of lameness.

Experimental Design

This was a prospective, blinded, randomized, placebo-controlled analgesic efficacy study with CMPS-SF pain scores and $\%BW_{dist}$ the primary outcome measures. Dogs were allocated into LB treatment and saline control groups by randomized stratification. Dogs presenting to our institution for TPLO have commonly received a recent dose of the NSAID, carprofen. To avoid a

confounding effect on postoperative pain measurement, we stratified enrollees by carprofen use into "recent carprofen" (within the last 40 hours, representing approximately 5 terminal plasma half-lives)[32] or "no recent carprofen" groups. Dogs within these groups were randomly assigned to either the LB or placebo treatment subgroups using a computerized random selection generator (randomizer.org).

Data from pain scoring and $\%BW_{dist}$ measurement were collected preoperatively (before sedation for TPLO planning radiographs) and over the 48-hour postoperative study duration by two investigators (L.A., I.O.) who were blinded to treatment assignment.

CMPS-SF

The CMPS-SF is a subjective clinical metrology instrument designed for efficient assessment of acute postoperative pain in dogs in a clinical setting.[22] Through a combination of observation and interaction across six behavioral categories, a total pain score is assigned between 0 and 24 for ambulatory patients, or between 0 and 20 for patients who cannot walk without assistance. Two investigators (L.A. and I.O.) trained together in use of the CMPS-SF for several days before the study began, to achieve subjective interobserver consistency in scoring of postoperative orthopedic surgery patients. Pain scores for study participants were assigned by one of these two investigators preoperatively (baseline) and at 2, 4, 8, 12, 20, 24, 32, 40, and 48 hours postoperatively, where 0 hours was time of extubation.

Static body weight distribution

Data from body weight distribution measurement were collected by one of two investigators (L.A. and I.O.) at baseline and at 4, 12, 24 and 48 hours postoperatively using a

weight distribution platform (PetSafe Stance Analyzer, Companion Animal Health, Newark, DE, USA). At the start of each session, the platform and associated software were calibrated to zero weight. Based on previous studies of the repeatability of stance analysis,[25,27] the patient was walked at a velocity of approximately 1 m/s onto the soft plastic platform, led on a short neck leash by an investigator positioned to the animal's right. The dog was abruptly stopped as the investigator moved in front of the animal to discourage additional forward movement. This procedure was repeated if the dog sat, lay down, or did not otherwise assume a natural, square stance with one foot upon each of the quadrants of the platform and its head held approximately on midline. After the dog maintained a square stance for appoximately 5 seconds, data collection was started, with multiple measurements of body weight distribution to each limb captured at approximately 0.5 to 1 second intervals, using a handheld remote control. Outlier measurements resulting from aberrant body movements were immediately discarded and 6 valid measurements per session were obtained, from which mean $\%BW_{dist}$ was calculated for the operated limb. When pain scoring and $\%BW_{dist}$ measurement were scheduled at concurrent time points, pain scoring was completed first.

Rescue analgesia, success/failure determination, and mean rescue opioid doses

Dogs were administered a single dose of hydromorphone 0.08 mg/kg subcutaneously as rescue analgesia if they were assigned a CMPS-SF pain score of 6/24 or 5/20 or greater during any of the scheduled or unscheduled (clinically indicated) pain assessments based on previous recommendations.[13] After a dog received rescue analgesia, all subsequent pain scores and $\%BW_{dist}$ measurements were excluded from statistical analysis to avoid a confounding effect on results.

Anesthesia, analgesia and surgery

All dogs were premedicated with IM administration of acepromazine (0.01 to 0.02 mg/kg) and hydromorphone (0.08 to 0.1 mg/kg). Anesthesia was induced with intravenous propofol and maintained with isoflurane in oxygen, both given to effect. No regional adjunct anesthetic techniques such as lumbosacral epidural or nerve blockade were permitted. All dogs received cefazolin (22 mg/kg) IV and IM at induction of anesthesia, as well as intravenous fluid therapy during the anesthetic period. Treatments for systemic hypotension under anesthesia included fluid therapy, antimuscarinics and adrenergic agonists, as needed.

Routine TPLO[33] was performed by one of eight primary surgeons, including residents, ACVS board-eligible and ACVS board-certified surgeons. Based on surgeon preference regarding stifle joint exploration and meniscal cartilage treatment, any of the following procedural variations were permitted: craniomedial parapatellar arthrotomy, cranial cruciate ligament debridement, meniscal debridement, or midbody outside-to-inside medial meniscal release.[34]

After stabilization of the osteotomy and closure of the joint capsule, the surgical wound was infiltrated with either undiluted LB (5.3 mg/kg;0.4 mL/kg) or an equal volume of sterile saline (0.4 mL/kg), based on the patient's random group assignment. The infiltrate was administered using the moving needle technique[21] using a sterile syringe fitted with a 1.0 to 1.5 inch, 22-gauge needle. The entire volume was distributed into three tissue layers as described in previous LB efficacy studies[21,24] with approximately 25%, 50% and 25% injected into the superficial tissues of the closed joint capsule, the closed fascial tissue, and the subcuticular tissue, respectively, before skin closure. No surgeon found it necessary to dilute the LB with

saline to achieve full coverage of the surgical site. The investigator who performed TPLO surgery in addition to postoperative pain assessment (L.A.) remained blinded to the treatment by exiting the surgical suite after stabilizing the osteotomy and closing of the joint capsule, allowing an alternate surgeon to perform tissue infiltration and closure.

When skin apposition was complete, the patient received a single dose of IV hydromorphone (0.08 mg/kg). Postoperative radiographs were performed routinely under general anesthesia. During the 48-hour postoperative study period, all dogs received subcutaneous carprofen (2.2 mg/kg) every 12 hours, beginning at time of extubation (0 hours). When a carprofen dose was scheduled concurrently with pain scoring and $\%BW_{dist}$ measurement, data were collected before carprofen was administered. Dogs experiencing significant dysphoria upon recovery were permitted to receive a single IV dose of dexmedetomidine (1 mcg/kg) at the discretion of the supervising anesthesiologist.

Postoperative care included leash walks every 4 hours, free access to water, meals offered every 12 hours, and icing of the incision site every 4 hours between 7am and 11pm. The surgical incision was protected with a bandage consisting of a non-adherent dressing with an adhesive covering until time of discharge. Dogs wore an Elizabethan collar at all times. Adverse events were noted and addressed.

Sample size calculation

Sufficient raw data are not previously reported describing static body weight distribution in dogs in the acute postoperative period, making a priori power analysis for sample size calculation a challenge. The kinetic variable, peak vertical force (PVF), and $\%BW_{dist}$ were shown to have similar sensitivity to detecting change in hind limb use before and approximately

4 months after total hip replacement.[27] To estimate an expected effect size of $\%BW_{dist}$ for LB compared to placebo, we calculated an effect size of changes in PVF ratios reported in a previous analgesic efficacy study.[35] Pain intensity for the induced synovitis model used in that study was expected to peak at 2-3 hours after urate crystal injection.[36] At 3 hours after urate crystal injection, PVF ratios were significantly different between firocoxib and placebo groups[35] and the effect size was large (Cohen's d = 1.4, effect size index for two-tailed t-test of means). Using that effect size, a priori power analysis yields a sample size of 30 dogs (15 in each treatment group), at a power (1-β) of 0.95 and significance (α) of 0.05.

Statistical analysis

Comparisons of pertinent treatment group variables including recent carprofen use and frequencies of arthrotomy and meniscal debridement were performed using a Chi-square test. Pain scores were compared between treatment groups preoperatively and at 2, 4, 8, 12, 20, 24, 32, 40 and 48 hours postoperatively with a nonparametric Mann-Whitney U test. Treatment success for a patient was defined as not requiring any rescue analgesia within the entire 48 hour postoperative period. The proportion of successes vs. failures between treatment groups was compared with a Chi-square test. A comparison between the number of required rescue opioid doses relative to treatment group size was made using a nonparametric Mann-Whitney U test. Data describing $\%BW_{dist}$ for the operated hind limb were determined to be normally distributed ($p = 0.001$) by use of the Anderson-Darling test. $\%BW_{dist}$ values were compared between treatment groups preoperatively and at 4, 12, 24, and 48 hours postoperatively with a Student's T test. Linear relationship between pain score and $\%BW_{dist}$ was assessed using Pearson's

correlation coefficient. Pain Scores and %BW values were excluded from analysis after rescue analgesia as described. Significance was set at $P < 0.05$ for all tests.

Results

Thirty-two dogs were enrolled and all completed the study. Fifteen (15) dogs were treated with LB and 17 with saline placebo. Frequencies of recent carprofen use ($P = 0.39$), arthrotomy ($P = 0.54$), and meniscal debridement ($P = 0.31$) (Table 1) were not different between treatment groups.

Pain scores

Median CMPS-SF pain scores at preoperative baseline were 1 (range 1-5) and 1 (range 1-3) for the LB and saline groups, respectively ($P = 0.82$). Median pain scores did not differ between treatment groups at any postoperative time point (Table 2).

Success/failure analysis

Overall treatment success was not different between dogs that received LB and those that received placebo (chi square $P = 0.27$) (Table 3). Two out of 15 dogs in the LB group required rescue analgesia, both at 2 hours postoperatively. Five out of 17 dogs in the placebo group required rescue analgesia: 4 dogs at 2 hours postoperatively, and 1 dog at 8 hours postoperatively.

Rescue opioid doses

The number of rescue opioid doses did not differ between the treatment groups, with the LB group (n = 15) receiving 3 total opioid doses and the placebo group (n = 17) receiving 10 total opioid doses ($P = 0.41$).

%BW$_{dist}$

Mean %BW$_{dist}$ values for the operated hind limb at preoperative baseline did not differ between the LB (6.7 ± 4.0%) and placebo (7.5 ± 4.7%) ($P = 0.61$) any postoperative time point. (Table 4). %BW$_{dist}$ data was unable to be collected at three postoperative time points for the 25 dogs that did not require rescue analgesia. One dog was too sedate to stand at 4 hours postoperatively and two dogs repeatedly chose to sit or lay down upon reaching the weight distribution platform at 48 hours postoperatively and could not be encouraged to stand. For the 7 dogs that required rescue analgesia, concurrent and subsequent %BW$_{dist}$ values were excluded from statistical analyses.

Relationship between pain score and %BW$_{dist}$

Analysis of linear correlation between CMPS-SF pain scores and %BW$_{dist}$ pooled to include all study participants did not demonstrate a statistically significant relationship at any time point, with Pearson's r values of 0.11, 0.23, 0.02, -0.24, -0.19 at preoperative baseline, and 4, 12, 24 and 48 hours, respectively.

Adverse events

Adverse postoperative events were observed in 5 dogs during the study; 3 in the LB group and 2 in the placebo group. Within the LB group, two dogs had incisional complications. One dog was noted to have bandage strikethrough at 4, 8 and 20 hours postoperatively that resolved with placement of a soft padded compression bandage. Another dog had focal serosanguineous discharge at 24 hours postoperatively that resolved with placement of a single surgical staple to improve skin apposition. One dog in the LB group became mildly cage

aggressive and less cooperative over time, however we were able to complete pain scoring and body weight distribution measurements. Within the placebo group, one dog regurgitated at 42 hours postoperatively and another dog was noted to have soft but formed stool.

Post hoc sample size calculation

The size of the treatment effect observed for $\%BW_{dist}$ was smaller than estimated before the study was initiated. Effect size (Cohen's d) for $\%BW_{dist}$ ranged from 0.27 to 0.75; at a power ($1-\beta$) of 0.80 and significance (α) of 0.05, a total of 58 to 436 dogs would have been needed to detect differences in $\%BW_{dist}$ between treatment groups.

Discussion

Results of this study show no difference between treatment success, postoperative rescue opioid consumption, CMPS-SF pain scores, or $\%BW_{dist}$ in dogs receiving carprofen and single-dose surgical wound infiltration with either LB or saline placebo, after TPLO. Therefore, we could not reject any of our null hypotheses. In the absence of any observed treatment effect, our first concern is to address the possibility that a type II error was committed. To calculate sample size prior to the study, we used an estimated treatment effect for %BWdist that turned out to be much greater than the actual treatment effect that we observed for $\%BW_{dist}$. Enrolling between 58 to 436 dogs into the study within a reasonable time frame would have been very challenging.

Using 46 and 29 dogs, respectively, two previous clinical studies[21,24] both found that dogs administered LB were less likely to require rescue analgesia after stifle surgery. The first was a pilot, randomized, placebo-controlled, masked study of dogs undergoing lateral retinacular suture placement with arthrotomy. The percent of dogs requiring rescue analgesia was significantly lower for dogs receiving LB versus placebo over 0-24, 0-48 and 0-72 hour post-operative intervals.[21] Although the extent of the soft tissue approach is similar between TPLO and lateral retinacular suture placement, TPLO involves greater surgical trauma. It is possible that periosteal and bone marrow pain are not well controlled by LB, especially if it does not penetrate deeper than the soft tissues into which it is injected. In addition, dogs in the pilot study received no scheduled analgesia following a single dose of hydromorphone given before induction of anesthesia. In our study, carprofen was administered every 12 hours, postoperatively. The clinical benefit of LB for TPLO-induced pain in dogs already receiving carprofen therefore comes into question.

A subsequent randomized, blinded study[24] incorporated carprofen into its postoperative analgesia protocol for dogs that received either LB or 0.5% bupivacaine surgical site infiltration for TPLO with arthrotomy. There was an opioid-sparing benefit of LB in the face of background treatment with carprofen. Over the 48 hour postoperative period, dogs administered LB at wound closure were less likely to need rescue analgesia and consumed a lower total morphine equivalent for rescue analgesia.[24] The opioid-reducing benefit conferred by LB was attributed to its longer duration of effect compared with standard bupivacaine. Although this study showed a clinical benefit of LB beyond that achieved by carprofen alone, we cannot directly compare their treatment success analysis with our own. This is because the decision to provide rescue analgesia in that study was based on pain scores using assigned by the CSU-CAPS, rather than the with the CMPS-SF. The authors argued[24] that CMPS-SF scores can be increased by signs of anxiety in dogs. For this reason, the CMPS-SF analgesic intervention score remains controversial. It is worth noting that while the LB pilot study used the CMPS-SF to identify patients needing rescue analgesia, the intervention level was raised from a suggested 6/24 to 8/24 based on investigator experience.[21]

Subjective pain scales are limited in their ability to describe the magnitude of pain relief provided by the treatment compared to placebo, but they are regarded as the current gold standard for evaluating pain in animals. The original, longer form of the CMPS[37] has been shown to have criterion validity, demonstrating sensitivity to acute post-operative pain in dogs in a clinical setting.[38] While the CMPS-SF was derived from the CMPS to help clinicians more efficiently identify acute pain and implement rescue analgesia in dogs, it has not undergone criterion validation. In this and previous studies, background analgesia has been minimized to improve the sensitivity of the CMPS-SF to treatment effect. Similar to findings in the previous

LB for TPLO study[24], pain scores in our study were not different for dogs receiving LB compared to control at any time point. The background effect of the carprofen given to the dogs in these studies may have obscured the ability to detect a clinical benefit of LB. We could have eliminated the background analgesic effect of the NSAID in the present study, but we considered it clinically valuable to determine whether LB might provide detectable analgesia beyond that of carprofen. In addition, while it was appropriate to exclude from analysis the subsequent pain scores of dogs that received rescue analgesia, this also decreased our sample population size and the power of the study over time. Another limitation of the study with respect to pain scoring was that we did not statistically test interobserver agreement between the two investigators during the pre-study training period.

It is interesting that objective means of pain assessment in the present and previous LB for TPLO study[24] failed to demonstrate a difference in outcome for dogs receiving LB. In the previous TPLO study, mechanical nociceptive threshold values did not differ between dogs that received LB compared with 0.5% bupivacaine. It is possible that the treatment effects of LB and standard bupivacaine were truly not different enough to be discriminated by pain scoring or pressure algometry. It is also possible that individual variability in responses to algometry[39] or learned aversion to the algometer with repeated use [24,40] contributed to the insensitivity of the instrument to pain.

Measurement of $\%BW_{dist}$ as a means of describing limb use or presumed limb pain has been described in the literature. Static body weight distribution was first evaluated using pressure sensitive walkway equipment. In normal dogs, measurements of $\%BW_{dist}$ were consistent from one week to the next, provided handling technique was consistent.[25] In dogs recovering from total hip replacement, $\%BW_{dist}$ to the operated limb increased at 3, 6 and 12

months after surgery, although without simultaneous pain scoring, it was not possible to conclude whether this change was related to a decrease in limb pain over time or simply a change in limb use. In another study, $\%BW_{dist}$ was shown to be as sensitive as traditional ground reaction forces, vertical impulse and peak vertical force, for evaluating limb use in dogs before and months after total hip replacement.[27]

More recent research measuring $\%BW_{dist}$ has made use of a weight distribution platform as a smaller and less expensive alternative to pressure sensitive walkway equipment. Measurement of $\%BW_{dist}$ using a weight distribution platform was found to be accurate compared to a pressure sensitive walkway,[28] sensitive to and specific for limb lameness and orthopedic disease[29], and repeatable for paired same-day or next-day measurements in dogs with hind limb lameness.[41] To our knowledge, the present study is the first to statistically compare subjective pain scores with $\%BW_{dist}$. We found no linear correlation between these outcome measures. There are several possible explanations for this.

Once a dog off-loads its limb to the point of being non-weight bearing, we lose the ability to assign any number besides zero to quantify their level of pain. We observed many dogs to be non-weight bearing after surgery, and some remained non-weight bearing for the remainder of our study. It would be inaccurate to assume that non-weight bearing dogs, all described by $\%BW_{dist} = 0$, are equally painful in those limbs. During individual stance analysis sessions, we observed some variability in a patient's willingness to bear weight on their operated limb. A limitation of the study is that we did not attempt to investigate this phenomenon. Another limitation was the relatively infrequent postoperative measurement of $\%BW_{dist}$ (4, 12, 24 and 48 hours) compared with the frequency of pain scoring (2, 4, 8, 12, 20, 24, 32, 40 and 48 hours). In this way, we may have missed opportunities for $\%BW_{dist}$ to describe pain. Finally, we observed

in some dogs a preference for sitting or laying down on the weight distribution platform, which was slightly elevated and softer than the surrounding floor. For these dogs, obtaining valid stance data required many passes over the platform. This brings into question the practicality of using this equipment to measure $\%BW_{dist}$. Using a weight distribution platform that is less tempting to dogs as a place of rest could make data collection more efficient.

We conclude that in this population of dogs recovering from TPLO and receiving postoperative carprofen, there was no difference between overall treatment success, relative rescue analgesia requirement, CMPS-SF pain scores, or $\%BW_{dist}$ in dogs that received surgical wound infiltration with LB compared with saline placebo. $\%BW_{dist}$ holds promise as an objective outcome measure for acute orthopedic surgical pain and the design of a firmer, lower-profile weight distribution platform is warranted.

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Table 1 Population frequencies for variable relevant to postoperative pain assessment in the LB and saline treatment groups

	LB (*n* = 15)	Saline placebo (*n* = 17)	Chi square p-value
Recent carprofen use (# of dogs)	4	7	0.39
Stifle arthrotomy performed (# of dogs)	10	13	0.54
Meniscal debridement performed (# of dogs)	5	3	0.31

(abbreviations: LB, liposomal bupivacaine)

Table 2 Median (range) pain scores assigned by use of the CMPS-SF for dogs receiving LB (*n*=15) or saline placebo (*n* = 17)

	LB (*n* = 15)		Saline placebo (*n* = 17)		
	Pain score	Number of dogs	Pain score	Number of dogs	p value
Preoperative baseline	1 (1-5)	15	1 (1-3)	17	0.82
Time after extubation (h)					
2	4 (1-11)	15	3 (1-11)	17	0.76
4	3 (2-4)	13	3 (1-5)	13	0.76
8	2 (1-5)	13	2 (1-12)	13	0.49
12	2 (1-5)	13	2 (1-4)	12	0.37
20	2 (1-4)	13	1.5 (1-4)	12	0.94
24	1 (1-2)	13	1 (1-3)	12	0.48
32	1 (1-2)	13	1 (1-3)	12	0.91
40	1 (1-2)	13	1 (1-2)	12	0.98
48	1 (1-2)	13	1 (1-2)	12	0.79

(abbreviations: CMPS-SF, Glasgow composite mean pain score, short form; LB, liposomal bupivacaine)
For dogs requiring rescue analgesia, subsequent pain scores were excluded from statistical analyses.

Table 3 Success/failure analysis results, where dogs requiring rescue analgesia at any postoperative timepoint (0 to 48 hours) were defined as treatment failures

	LB (*n* = 15)	Saline placebo (*n* = 17)	Chi square p-value
Success (*n*)	13	12	0.27
Failure (*n*)	2	5	

(abbreviations: LB, liposomal bupivacaine)

Table 4 Mean (standard deviation) for %BW dist values for dogs receiving LB (*n* = 15) or saline placebo (*n* = 17)

	LB (*n* = 15)		Saline placebo (*n* = 17)		
	$\%BW_{dist}$	Number of dogs	$\%BW_{dist}$	Number of dogs	P value
Preoperative baseline	6.7 (4.0)	15	7.5 (4.7)	17	0.61
Time after extubation (h)					
4	3.9 (4.0)	13	1.6 (1.8)	12[a]	0.08
12	2.9 (5.0)	13	1.8 (2.8)	12	0.07
24	3.3 (3.4)	13	2.4 (3.1)	12	0.50
48	2.4 (2.5)	11[b]	4.8 (3.7)	12	0.37

(abbreviations: $\%BW_{dist}$, percent of total body weight distributed to the operated leg; LB, liposomal bupivacaine) For dogs requiring rescue analgesia, concurrent and subsequent measurements of $\%BW_{dist}$ were excluded from statistical analyses. [a]Data point missing for one dog who was too sedate to stand but did not require rescue analgesia. [b]Data points are missing for two dogs that refused to stand (in favor of sitting) on the weight distribution platform but did not require rescue analgesia.