

Local Anesthetics Induce Chondrocyte Death in Bovine Articular Cartilage Disks in a Dose- and Duration-Dependent Manner

Ian K. Y. Lo, M.D., F.R.C.S.C., Paul Sciore, Ph.D., May Chung, B.Sc., Sherri Liang, B.Sc., Richard B. Boorman, M.D., F.R.C.S.C., Gail M. Thornton, Ph.D., Jerome B. Rattner, Ph.D., and Kenneth Muldrew, Ph.D.

Purpose: The purpose of this study was to evaluate the effect of various local anesthetics on chondrocyte viability in articular cartilage by use of a bovine disk model. **Methods:** Full-thickness bovine cartilage disks were isolated from the condylar surfaces of the radial-carpal joint by use of a 4-mm biopsy punch and were incubated in various concentrations of local anesthetics (e.g., bupivacaine) for varying amounts of time and stained for membrane integrity by use of ethidium bromide and SYTO 13 stain (Molecular Probes, Carlsbad, CA). Cell and nuclear morphology was assessed by transmission electron microscopy. **Results:** The addition of local anesthetics (i.e., 0.25% bupivacaine, 1% lidocaine, and 0.5% ropivacaine) to bovine articular cartilage disks had a negative effect on chondrocyte viability. Culturing bovine articular cartilage disks for increasing periods of time decreased chondrocyte viability for each of the local anesthetics, with significant negative correlations being shown between time of exposure to the drug and chondrocyte viability. These effects were also affected by the presence or absence of epinephrine in local anesthetic preparations. **Conclusions:** Our results suggest that local anesthetics (i.e., bupivacaine, lidocaine, or ropivacaine) can have a detrimental effect on chondrocyte viability in bovine articular cartilage disks in a dose- and duration-dependent manner. **Clinical Relevance:** After arthroscopic surgery, it has been common practice to inject various local anesthetics into the joint for pain relief. Because adult chondrocytes have little or no capacity to regenerate, these results suggest that high-dose, long-term intra-articular administration of local anesthetics should be performed with caution. **Key Words:** Bovine—Chondrocyte viability—Local anesthetics—Cytotoxicity—Chondrolysis.

Arthroscopic joint surgery is one of the most commonly performed procedures in surgical practice. In the United States arthroscopic procedures are 3 of the top 5 most commonly performed procedures by ortho-

paedic surgeons taking part II of the American Board of Orthopaedic Surgery oral examinations.¹ One major advantage of arthroscopic surgery is its minimally invasive nature, which limits surgical wounds, bleeding, and in particular, postoperative pain. Despite this, however, postoperative pain can still be problematic. In an effort to decrease postoperative pain, various treatments have been used including narcotics, anti-inflammatory drugs, regional and local blocks, and local anesthetics, both subcutaneously and intra-articularly. These methods used alone or in combination have allowed most arthroscopic procedures to be performed on an outpatient basis. With superior pain control, the ability to perform procedures in a minimally invasive outpatient manner has resulted in a significant reduction in cost per case.²⁻⁴

In an effort to improve postoperative pain control and limit systemic side effects, the use of intra-articular local

From the Department of Surgery, Faculty of Medicine (I.K.Y.L., P.S., M.C., S.L., R.B.B., G.M.T.), and Department of Anatomy and Cell Biology (J.B.R., K.M.), University of Calgary, Calgary, Alberta, Canada.

Supported by the Canadian Institutes of Health Research. The authors report no conflict of interest.

Received November 20, 2008; accepted March 30, 2009.

Address correspondence and reprint requests to Ian K. Y. Lo, M.D., F.R.C.S.C., Department of Surgery, University of Calgary, 3330 Hospital Dr NW, Calgary, Alberta, Canada. E-mail: ikylo@ucalgary.ca

© 2009 by the Arthroscopy Association of North America

0749-8063/09/2507-8654\$36.00/0

doi:10.1016/j.arthro.2009.03.019

anesthetics (e.g., bupivacaine, lidocaine, and ropivacaine) has gained popularity. Local anesthetic use after arthroscopic surgery is routinely performed as a single-dose intra-articular injection (e.g., 20 mL of 0.25% bupivacaine). Furthermore, to provide even longer-term relief, continuous infusion of intra-articular local anesthetics has been commonly performed for up to 48 to 72 hours.⁵⁻¹¹ In this method a small catheter is placed intra-operatively into the joint, and local anesthetic is continuously infused into the intra-articular environment. Many authors have reported superior pain relief with intra-articular continuous infusion of local anesthetics compared with placebo or other methods of pain relief.⁵⁻¹¹ In addition, some authors have reported devastating complications with their use (mostly associated with catheter placement).⁵⁻⁹ Although intra-articular local anesthetic use after surgery is a common indication, intra-articular local anesthetics are also commonly used in the outpatient setting. They have been used for pain relief, to address diagnostic challenges, and in combination with other agents (methylprednisolone) for therapeutic use. Despite these common practices, the relative safety of a single dose or continuous infusion of local anesthetics is unclear. In particular, the constant “bathing” of cartilage and articular chondrocytes in a foreign environment (i.e., local anesthetics) may be of concern. Recently, however, several authors have reported cases of acute chondrolysis after arthroscopic shoulder surgery.¹²⁻¹⁵ In many cases the result has been a complete denuding of cartilage leading to end-stage arthritis. These cases are particularly devastating because postsurgical arthritis in the young population has no good treatment option. Although many of these cases were associated with the use of cautery or thermal capsulorrhaphy (a potential cause of acute chondrolysis resulting from increased joint temperatures during surgery), there have been increasing concerns that the use of postoperative, intra-articular local anesthetic infusion may be associated with cytotoxicity and chondrolysis.¹⁶⁻²⁰ The purpose of this study was to evaluate the effect of local anesthetics (i.e., bupivacaine, lidocaine, and ropivacaine) on chondrocyte viability. Using bovine articular cartilage disks as a model system, we hypothesized that local anesthetics would have a negative effect in a dose- and duration-dependent manner on chondrocyte viability.

METHODS

To evaluate the effects on articular cartilage chondrocytes, a bovine cartilage disk model was used. Disks were chosen because chondrocytes *in situ* are more representative of the *in vivo* state compared with isolated

chondrocytes. This model was used because of the relative inaccessibility of normal human articular cartilage and the maintenance of an intact pericellular and extracellular matrix surrounding articular cartilage chondrocytes. Bovine articular cartilage was obtained from a local abattoir. Full-thickness cartilage disks were isolated from the condylar surfaces of the radial-carpal joint by use of a 4-mm biopsy punch and were used immediately after harvest. Disks were washed twice with sterile phosphate-buffered saline solution (PBS), and 3 disks per well were placed in a 24-well tissue culture plate and were cultured under various conditions (e.g., concentrations and times) with 3 commonly used local anesthetics: 0.25% bupivacaine, 1% lidocaine, and 0.5% ropivacaine. Furthermore, to study the effect of epinephrine, 0.25% bupivacaine with epinephrine (1:200,000 epinephrine, 5 $\mu\text{g/mL}$) and 1% lidocaine with epinephrine (1:200,000 epinephrine, 5 $\mu\text{g/mL}$) were used as prepared solutions available from the manufacturer. To evaluate the combined effects of ropivacaine with epinephrine, exogenous epinephrine was added to each preparation to simulate concentrations found in premade solutions of bupivacaine with epinephrine (i.e., 1:200,000 final epinephrine concentration) and solutions of lidocaine with epinephrine that are obtained directly from the manufacturer. Ropivacaine does not normally come prepared with epinephrine because ropivacaine itself may cause vasoconstriction.

Concentrations of each local anesthetic (i.e., 0.25% bupivacaine with or without epinephrine, 1% lidocaine with or without epinephrine, and 0.5% ropivacaine with or without epinephrine) were prepared in sterile PBS. For example, to prepare 10 mL of a 50% solution of bupivacaine (supplied at 0.25% by the manufacturer), 5 mL of bupivacaine was added to 5 mL of sterile PBS to yield a 50% solution of the original stock. Each of the concentrations of local anesthetics used in the study was prepared in this manner by use of sterile PBS. To determine the effect of dose, articular cartilage disks were cultured in 1 mL of 0%, 2.5%, 5%, 10%, 25%, 50%, and 100% solutions of each local anesthetic in PBS at 37°C for 12 hours. To determine the effect of duration, articular cartilage disks were cultured in 50% and 100% solutions (as percents of the manufacturer's concentrations) of each local anesthetic for 1, 3, 5, 8, and 12 hours. For each experiment, 3 articular cartilage disks were cultured in each well with 1 mL of solution to approximate the relative surface area-to-joint volume ratio in the human shoulder.^{21,22} In addition, disks were cultured in 1:200,000 concentrations of epinephrine in sterile PBS (both at 100% and 50% concentra-

tions, prepared as described previously by use of PBS) to serve as epinephrine controls. All experiments were repeated 3 times to ensure reproducibility. Cartilage disks were also incubated in PBS as controls, because previous work has shown that cells in cartilage remain viable in PBS, even with extended incubation.²³ After incubation, disks were cut into 70- μ m sections by use of a TPI Vibratome 1000 Sectioning System (TPI, St Louis, MO). Cell viability was determined by use of SYTO 13 green fluorescent nucleic acid stain (Molecular Probes, Carlsbad, CA) and 3.3-ng/mL ethidium bromide (Sigma-Aldrich, Oakville, ONT, Canada). Sections were viewed under a fluorescence microscope and scored for cell viability. Cell viability was determined by a blinded separate observer with significant experience in cell viability analysis. Nine cartilage slices were scored for each of the treatment groups at the given time points and for the given treatments. Digital images were projected, and green and red cells were counted manually, with care taken to include the full thickness of articular cartilage in each field. In addition, for each of the incubation regimens, cartilage disks were isolated for assessment of cell and nuclear morphologic changes and chromatin organization by transmission electron microscopy. In brief, disks were isolated and fixed in 0.1-mol/L sodium cacodylate-buffered 1.5% electron microscopy-grade glutaraldehyde, 1% paraformaldehyde, and 0.4% ruthenium hexamine trichloride. After fixation, disks were set in 1% osmium tetroxide and embedded in Poly/Bed 812 (Polysciences, Warrington, PA). Thin sections of the disks were obtained with an ultramicrotome and were stained with 2% ethanolic uranyl acetate and lead citrate, and electron micrographs were obtained with a Hitachi H-7000 transmission electron microscope (Hitachi High Technologies Canada Inc., Toronto, ONT, Canada). Statistical analysis was performed by use of the Pearson or rank correlation coefficient to determine the degree of correlation between variables.

RESULTS

The addition of local anesthetics (i.e., 0.25% bupivacaine, 1% lidocaine, and 0.5% ropivacaine) to bovine articular cartilage disks had a negative effect on chondrocyte membrane integrity. Representative data for bupivacaine are shown in Fig 1. The results for varying doses of bupivacaine, lidocaine, and ropivacaine are summarized in Fig 2. At a concentration of 50%, chondrocyte membrane integrity was less than 30% at 12 hours when disks were cultured in any of the local

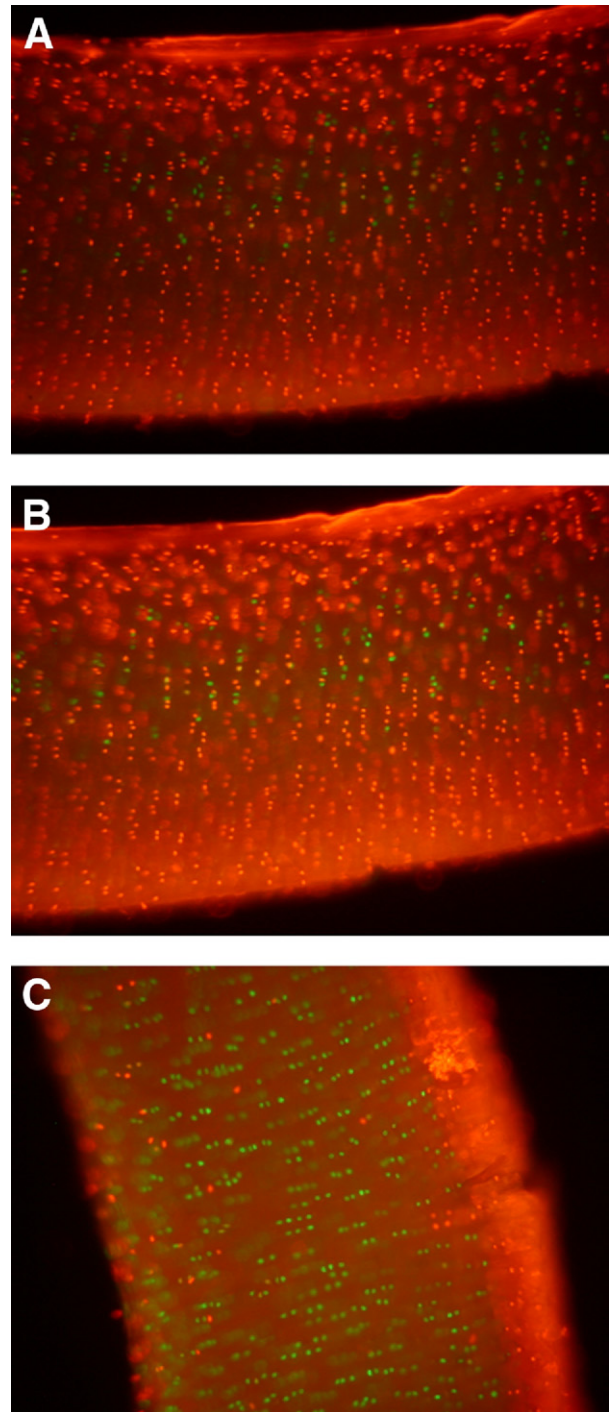


FIGURE 1. Fluorescence microscopy of bovine articular cartilage disks cultured for 12 hours in differing solutions of 0.25% bupivacaine: (A) 100%, (B) 50%, and (C) 0% (PBS). Magnification 150 \times . The decrease in cell viability with increased bupivacaine concentration should be noted. Green fluorescence indicates live articular chondrocytes, and red fluorescence indicates dead articular chondrocytes.

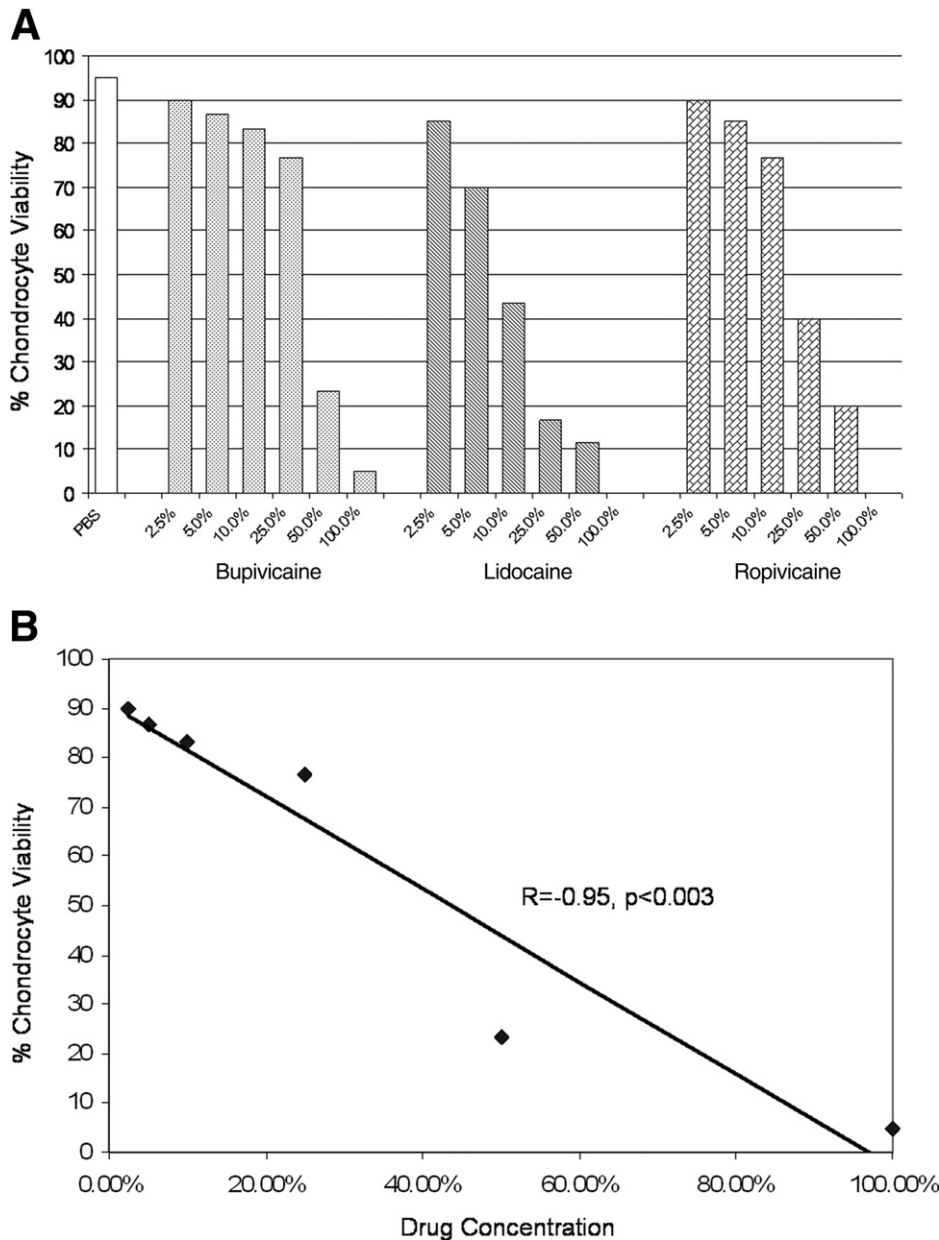


FIGURE 2. (A) Effect of varying concentrations of 0.25% bupivacaine, 1% lidocaine and 0.5% ropivacaine on chondrocyte viability in bovine articular cartilage disks cultured for 12 hours. (B) Scatter plot and Pearson correlation coefficient of chondrocyte viability in bovine articular cartilage disks cultured in increasing concentrations of 0.25% bupivacaine.

anesthetics. Culturing bovine articular cartilage disks in increasing concentrations of each local anesthetic for 12 hours resulted in significant decreases ($P < .05$) in chondrocyte membrane integrity (Fig 2A), with a significant negative correlation between chondrocyte membrane integrity and drug concentration. A representative scatter plot of this correlation is shown for bupivacaine in Fig 2B ($R = -0.95, P < .003$). Similar negative correlations were obtained for lidocaine ($R = -1, P < .003$) and ropivacaine ($R = -0.93, P < .007$).

The results of culturing bovine articular cartilage disks in 50% and 100% solutions of 0.25% bupivacaine, 1% lidocaine, and 0.5% ropivacaine for increasing periods of time are summarized in Figs 3A, 3B, and 3C, respectively. Culturing bovine articular cartilage disks for increasing periods of time decreased chondrocyte membrane integrity for each of the local anesthetics, with significant negative correlations being shown between time of exposure to the drug and chondrocyte membrane integrity (100% bupivacaine: $R = -0.93, P = .007$; 50% bupivacaine: $R = -0.96,$

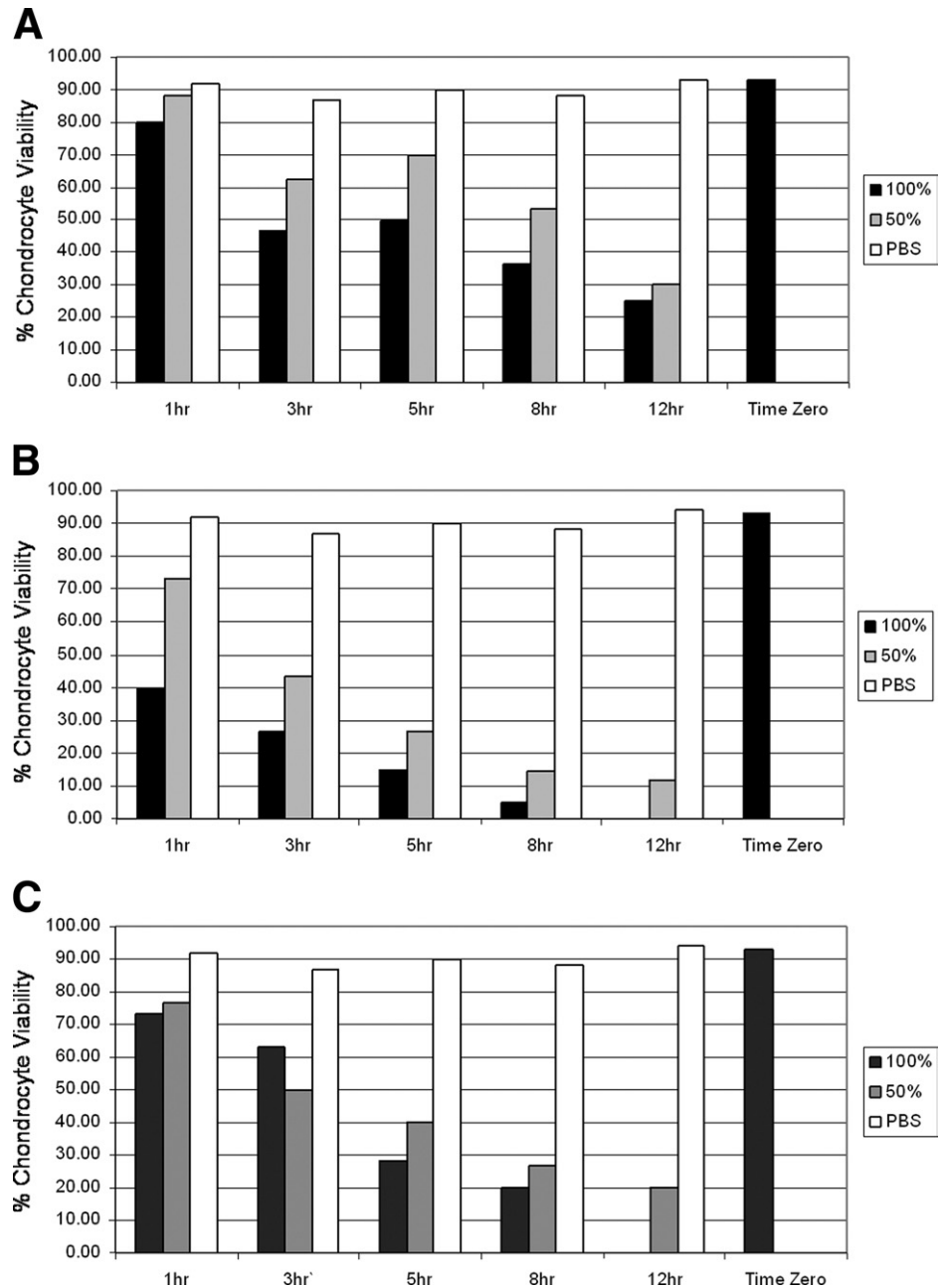


FIGURE 3. Effect of increasing periods of culturing on chondrocyte viability in bovine articular cartilage disks cultured in 50% solutions and 100% solutions of (A) 0.25% bupivacaine, (B) 1% lidocaine, and (C) 0.5% ropivacaine.

$P = .002$; 100% lidocaine: $R = -1$, $P < .003$; 50% lidocaine: $R = -1$, $P < .003$; 100% ropivacaine: $R = 0.84$, $P < .05$; and 50% ropivacaine: $R = -1$, $P < .003$.

The results of culturing bovine articular cartilage disks in 50% solutions of 0.25% bupivacaine with and without epinephrine for 8 and 12 hours are summarized in Fig 4. Culturing bovine articular cartilage disks in 50% or 100% solutions of epi-

nephrine alone did not have a significant effect on chondrocyte membrane integrity when compared with time 0 ($P > .05$) or PBS controls ($P > .05$) (Fig 4A). However, the presence of epinephrine in the bupivacaine solution (whether in the solution provided by the manufacturer or when added exogenously to bupivacaine without epinephrine) had significant negative effects on chondrocyte membrane integrity ($P < .008$) (Fig 4B).

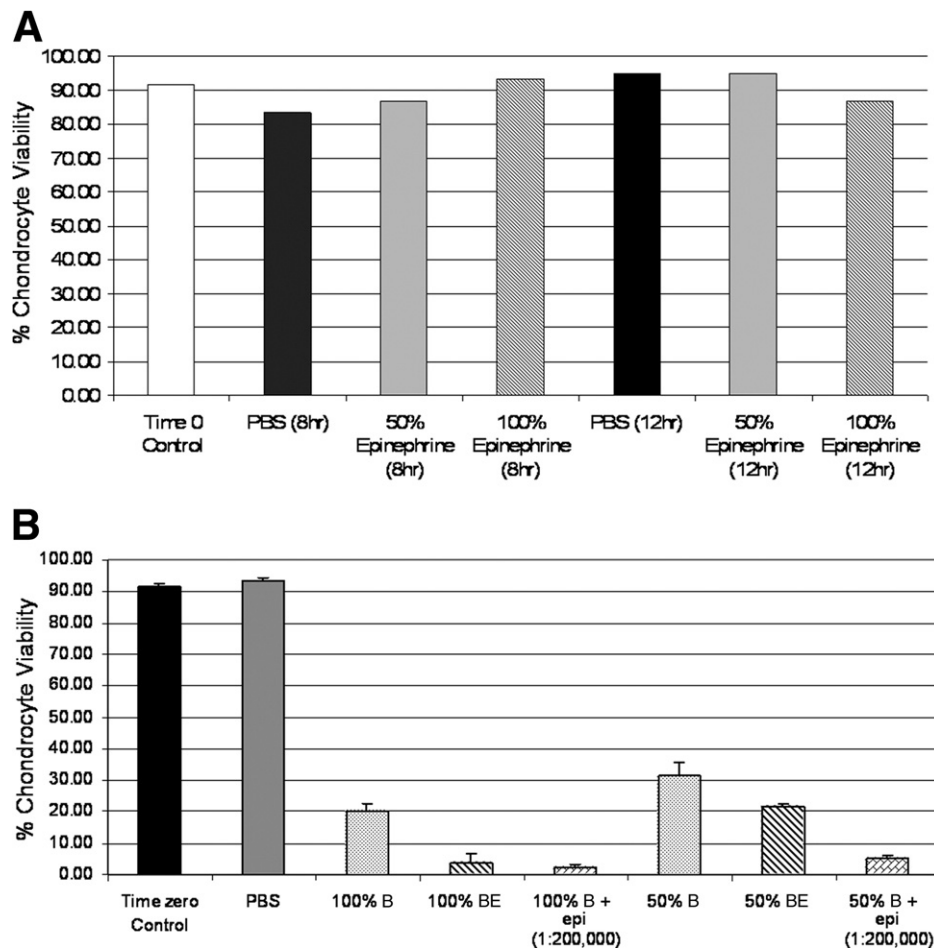


FIGURE 4. Effect of culturing bovine articular disks in (A) 50% and 100% solutions of 1:200,000 epinephrine for 8 and 12 hours and (B) 50% and 100% solutions of 0.25% bupivacaine with and without epinephrine at 8 and 12 hours. [B, bupivacaine; BE, bupivacaine with epinephrine as provided by manufacturer; B + epi (1:200,000), exogenous epinephrine added to bupivacaine to final concentration of 1:200,000.]

The results of the assessment of cell morphology (representative data showing exposure to bupivacaine) are shown in Fig 5. As shown, nuclear changes appear after only 1 hour of treatment, with more pronounced changes occurring at the 5-hour time point. These changes include alterations in nuclear morphology as well as changes in chromatin organization. In addition, the cell membrane remains intact. These results show that cells exposed to bupivacaine undergo visible morphologic changes in a short period of time. Similar ultrastructural changes were observed with all local anesthetics tested.

DISCUSSION

The results of this study show that the commonly used local anesthetics bupivacaine, lidocaine, and ropivacaine have a negative effect on chondrocyte viability in cultured bovine articular cartilage disks. Using a well-characterized model, this study showed

that the effect of exposure to bupivacaine, lidocaine, and ropivacaine occurred in both a dose- and duration-dependent fashion. In 2006 Chu et al.¹⁹ reported that exposure of 0.5% bupivacaine to bovine articular cartilage chondrocytes was cytotoxic. They reported that after exposure of chondrocytes with 0.5% bupivacaine for 1 hour, fewer than 1% of chondrocytes were viable using a chondrocyte–alginate bead culture system. Furthermore, they reported that exposure of bovine articular cartilage cores for 30 minutes with 0.5% bupivacaine decreased chondrocyte viability. This effect was augmented when the superficial 1 mm of cartilage was removed. Although our results are in concordance with those of Chu et al., they studied the effect of a single concentration of 0.5% bupivacaine, and it was unclear whether this was specific to bupivacaine or could be seen with other local anesthetics. Similarly, Gomoll et al.²⁰ studied the in vivo effects of a continuous infusion of 0.25% bupivacaine with and without epinephrine into the rabbit's shoulder. They

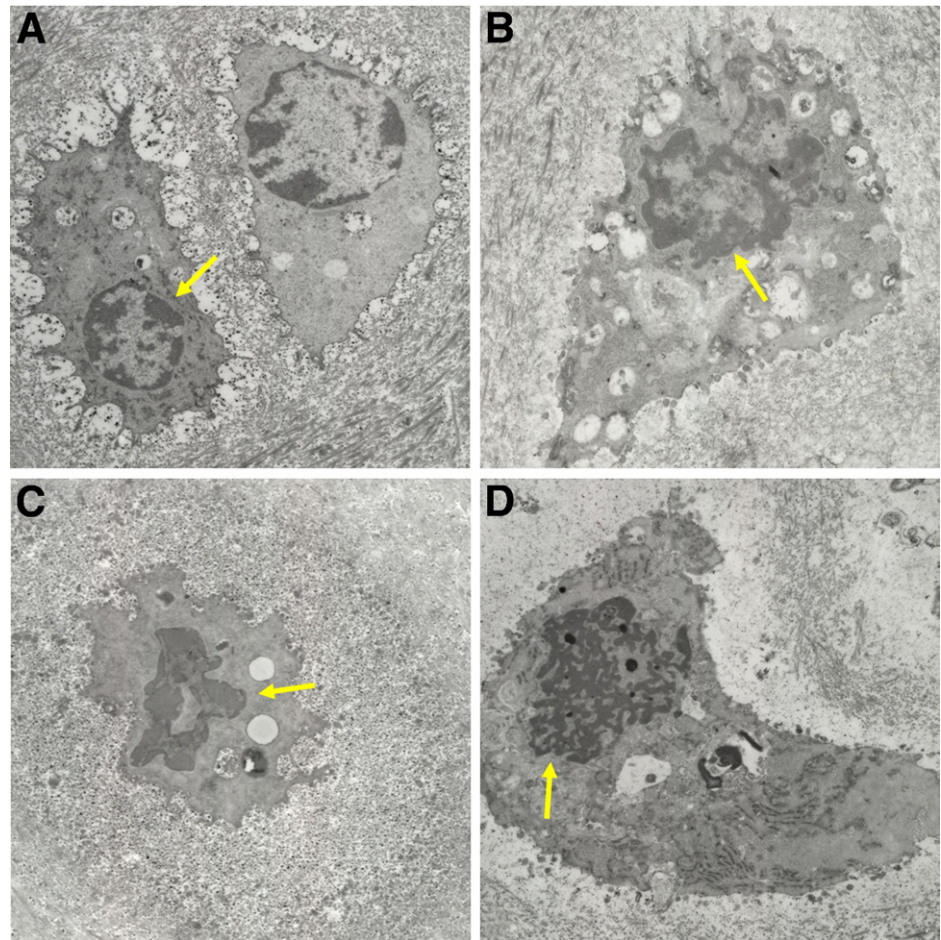


FIGURE 5. Effect of 100% solution of 0.25% bupivacaine on bovine chondrocyte morphology (cell and nuclear) as studied by transmission electron microscopy. Magnification 3500 \times . The arrows mark the nucleus. The changes in chromatin organization and nuclear membrane invaginations with exposure to local anesthetic should be noted. (A) PBS control (5-hour incubation). (B) One-hour incubation. (C) Three-hour incubation. (D) Five-hour incubation.

showed that infusion of bupivacaine or bupivacaine with epinephrine into the rabbit's shoulder decreased sulfate uptake, cell viability, and histologic scores. Our results are in concordance with those reported by Chu et al. and Gomoll et al. and suggest that bupivacaine has a negative effect on chondrocyte viability in both in vitro and in vivo animal models. In addition, we were able to examine the effects of other commonly used local anesthetics, and we showed that lidocaine and ropivacaine have a similar detrimental effect. This may be a result of the related structure of these agents and therefore a general drug class effect.²³ Furthermore, our study shows that ultrastructural nuclear changes were found in chondrocytes treated with local anesthetics. These changes in nuclear integrity as well as an intact cell membrane are consistent with observed phenotypic outcomes found in apoptotic cells. We are currently assessing whether specific apoptotic pathways are associated with chondrocyte death in this model system. Our preliminary results

have shown that local anesthetics (bupivacaine, ropivacaine, and lidocaine) induce apoptosis (as determined by transmission electron microscopy) and necrosis in bovine cartilage disk cells. The membrane integrity dyes indicate cell death due to both apoptosis and necrosis.

Importantly, other factors that have been implicated were also studied. In our study we determined that the addition of epinephrine to a final concentration of 5 $\mu\text{g/mL}$, similar to the final concentrations of epinephrine found in the local anesthetics tested (1:200,000; prepared in PBS), did not significantly or consistently increase or decrease chondrocyte viability when used in isolation. Interestingly, although the addition of epinephrine decreased chondrocyte viability when used in combination with bupivacaine, there was no significant effect when used in combination with lidocaine or ropivacaine. These results suggest that although epinephrine may augment the cytotoxicity of bupivacaine, the drug itself (i.e., bupivacaine, lidocaine, or ropivacaine) may also be cytotoxic. This portion of the study is important because,

clinically, some intra-articular catheter companies have continued to market their products and have suggested that the addition of epinephrine may be the culprit and that bupivacaine alone (or lidocaine and ropivacaine) can be safely used.

Another factor that may be related to cytotoxicity is the carrier solution itself. Local anesthetic solutions are commonly stored and used at an acidic pH. This pH varies between 4.0 and 5.7 and prevents precipitation of the active ingredient during long-term storage. Cells exposed to acidic stress are known to have changes in cell metabolism and cell function, with these changes possibly leading to a decrease in cell viability. Thus it is possible that the acidity of the carrier solution itself and not the active local anesthetic was cytotoxic to cells. However, in our study although acidity may further enhance cytotoxicity, all solutions were prepared by use of PBS and allowed to equilibrate for 1 hour before usage. In each case the pH of the solution was tested before usage, and in all solutions except the 100% solutions (for which no PBS was added), the pH was between 6.7 and 7.0 before usage and the pH was not significantly different at the end of the culturing duration. Thus the pH of the solution did not significantly affect the results. Of note, neutralization of the pH did not cause precipitation of the drug for the short durations evaluated in this study.

In this study we also showed that exposure to any of the local anesthetics occurred in a duration-dependent manner. Similarly, in the study by Chu et al.,¹⁹ the effect of duration of exposure was studied in bovine articular chondrocyte-alginate beads. In their study exposure of bovine articular chondrocyte-alginate beads for 15, 30, or 60 minutes to 0.5% bupivacaine decreased chondrocyte viability to less than 1% and was not significantly different for each of the exposure times. These results may be because of the differences in model systems, whereby the alginate bead system does not maintain the complex 3-dimensional pericellular and extracellular architecture of articular cartilage and likely allows more rapid and complete penetration into the bead. Thus the chondrocytes in the alginate bead system used by Chu et al. likely had exceptionally high concentrations of bupivacaine leading to almost complete death of cells even after only 15 minutes of exposure. These results highlight the importance of understanding each model system and the impetus for further evaluation in human articular cartilage.

Our study determined that the effect of chondrocyte viability was also dose dependent. Although almost all doses studied did affect chondrocyte viability, lower

concentrations did not affect chondrocyte viability as much as higher concentrations. These results suggest that, clinically, the effect of postoperative bleeding may dilute the toxic effects of local anesthetics.

Despite the collective results of this study and previous studies,¹⁶⁻²⁰ the reported incidence of complications from a single dose or continuous infusion of intra-articular local anesthetics is comparatively low. However, no scientifically rigorous clinical evaluations have been performed to objectively study this effect. Furthermore, single-dose injections of local anesthetics are commonly given with other drugs (methylprednisolone) that may be potentially cytoprotective (or cytotoxic) or in joints already affected with osteoarthritis. In addition, local anesthetic chondrocyte cytotoxicity postoperatively may also be affected by joint specificity, the dilution effect of bleeding, and extra-articular extravasation of local anesthetic. Furthermore, the pharmacokinetics and clearance of intra-articular local anesthetics are unclear and may be accelerated during the initial inflammatory response after surgery. In fact, our dosage and duration results suggest that the cytotoxic effects of local anesthetics may be affected by the initial concentration of drug given, the dilution caused by bleeding, and the duration of exposure (which is affected by postoperative pharmacokinetics of the drug). Clinically, the low incidence of complications related to local anesthetic use is in part likely directly related to its dose- and duration-dependent characteristics. In most cases its use clinically is likely below the threshold of cytotoxicity but may reach and exceed this threshold in cases of acute chondrolysis. However, to our knowledge, the threshold for cytotoxicity in human articular cartilage has not been determined, and therefore intra-articular local anesthetics should be used with caution. Although we have shown that bupivacaine, lidocaine, and ropivacaine have detrimental effects on chondrocyte viability in a dose- and duration-dependent manner, this was determined in bovine articular cartilage disks. A limitation of this study is that an *in vitro* bovine articular cartilage disk model was used. Furthermore, these results may not be generalizable to large human joints. Consequently, the ability to absolutely translate our findings into clinical practice remains limited.

There is evidence that adult chondrocytes have little or no capacity to regenerate,^{24,25} and therefore our results suggest that high-dose, long-term intra-articular administration of local anesthetics should be performed with caution.

CONCLUSIONS

Our results suggest that local anesthetics (i.e., bupivacaine, lidocaine, and ropivacaine) can have a detrimental effect on chondrocyte viability in bovine articular cartilage disks in a dose- and duration-dependent manner.

REFERENCES

- Harner CD, Vogrin TM. Specialty update. What's new in sports medicine. *J Bone Joint Surg Am* 2002;84:1095-1099.
- Bergstrom R, Hamberg P, Lysholm J, Gillquist J. Comparison of open and endoscopic meniscectomy. *Clin Orthop Relat Res* 1984;133-136.
- Pettrone FA. Meniscectomy: Arthrotomy versus arthroscopy. *Am J Sports Med* 1982;10:355-359.
- Pruzansky ME, Remer S, Freedman B. The effect of arthroscopic surgery of the knee on hospital utilization. *Health Serv Res* 1989;24:685-692.
- Alford JW, Fadale PD. Evaluation of postoperative bupivacaine infusion for pain management after anterior cruciate ligament reconstruction. *Arthroscopy* 2003;19:855-861.
- Barber FA, Herbert MA. The effectiveness of an anesthetic continuous-infusion device on postoperative pain control. *Arthroscopy* 2002;18:76-81.
- Chao D, Young S, Cawley P. Postoperative pain management for arthroscopic shoulder surgery: Interscalene block versus patient-controlled infusion of 0.25% bupivacaine. *Am J Orthop* 2006;35:231-234.
- Harvey GP, Chelly JE, AlSamsam T, Coupe K. Patient-controlled ropivacaine analgesia after arthroscopic subacromial decompression. *Arthroscopy* 2004;20:451-455.
- Hoenecke HR Jr, Pulido PA, Marris BA, Fronek J. The efficacy of continuous bupivacaine infiltration following anterior cruciate ligament reconstruction. *Arthroscopy* 2002;18:854-858.
- Klein SM, Nielsen KC, Martin A, et al. Interscalene brachial plexus block with continuous intraarticular infusion of ropivacaine. *Anesth Analg* 2001;93:601-605.
- Klein SM, Steele SM, Nielsen KIC, et al. The difficulties of ambulatory interscalene and intra-articular infusions for rotator cuff surgery: A preliminary report. *Can J Anaesth* 2003; 50:265-269.
- Jerosch J, Aldawoudy AM. Chondrolysis of the glenohumeral joint following arthroscopic capsular release for adhesive capsulitis: A case report. *Knee Surg Sports Traumatol Arthrosc* 2007;15:292-294.
- Larsen MW, Higgins L, Basamania CJ. Severe glenohumeral chondrolysis following shoulder arthroscopy: A series of 6 cases treated with hemiarthroplasty. *J Shoulder Elbow Surg* 2006;16:e47.
- Levine WM, Clark AM Jr, D'Alessandro DF, Yamaguchi K. Chondrolysis following arthroscopic thermal capsulorrhaphy to treat shoulder instability. A report of two cases. *J Bone Joint Surg Am* 2005;87:616-621.
- Petty DH, Jazrawi LM, Estrada LS, Andrews JS. Glenohumeral chondrolysis after shoulder arthroscopy: Case reports and review of the literature. *Am J Sports Med* 2004;32:509-15.
- Gomoll AH, Yank AB, Kang RW, et al. Long-term effects of bupivacaine on cartilage in a rabbit shoulder model. *Am J Sports Med* 2009;37:72-77.
- Piper SL, Kim HT. Comparison of ropivacaine and bupivacaine toxicity in human articular chondrocytes. *J Bone Joint Surg Am* 2008;90:986-991.
- Dragoo JL, Korotkova T, Kanwar R, Wood B. The effect of local anesthetics administered via pain pump on chondrocyte viability. *Am J Sports Med* 2008;36:1484-1488.
- Chu CR, Izzo NJ, Papas NE, Fu FH. In vitro exposure to 0.5% bupivacaine is cytotoxic to bovine articular chondrocytes. *Arthroscopy* 2006;22:693-699.
- Gomoll AH, Kang RW, Williams JB, Bach BR, Cole BJ. Chondrolysis after continuous intra-articular bupivacaine infusion: An experimental model investigating chondrotoxicity in the rabbit shoulder. *Arthroscopy* 2006;33:813-819.
- Graichen H, Jakob J, von Eisenhart-Roth R, Englmeier KH, Reiser M, Eckstein F. Validation of cartilage volume and thickness measurements in the human shoulder with quantitative magnetic resonance imaging. *Osteoarthritis Cartilage* 2003;11:475-482.
- Victoroff BN, Deutsch A, Protomastro P, Barber JE, Davy DT. The effect of radiofrequency thermal capsulorrhaphy on glenohumeral translation, rotation and volume. *J Shoulder Elbow Surg* 2004;13:138-145.
- Hunter S, Timmermann S, Schachar N, Muldrew K. The effects of hypothermic storage on chondrocyte survival and apoptosis in human articular cartilage. *Cell Preserv Technol* 2006;4:82-90.
- Schachar NS, Novak K, Hurtig M, et al. Transplantation of cryopreserved osteochondral Dowel allografts for repair of focal articular defects in an ovine model. *J Orthop Res* 1999; 17:909-920.
- Muldrew K, Hurtig M, Novak K, Schachar N, McGann LE. Localization of freezing injury in articular cartilage. *Cryobiology* 1994;31:31-38.