学位論文内容の要旨

Abstract of the dissertation

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学位論文題名

The title of the doctoral dissertation

# Chondrotoxicity of local anesthetics: *In vitro* effects of local anesthetics on cell viability and apoptosis in cultured canine articular chondrocytes

（局所麻酔薬の軟骨毒性に関する研究：

培養軟骨細胞における局所麻酔薬の細胞障害性とアポトーシスについて）

Abstract

Local anesthetics are a group of drugs that reversibly block voltage-gated sodium channels in excitable tissues, which inhibits the influx of sodium ions into the cell and interrupts the conduction of neural impulses. As a result, local anesthetics prevent the propagation of the pain stimulus, making them the only class of drugs that can fully block nociceptive impulses within targeted and/or restricted areas of the body. Bupivacaine, levobupivacaine and ropivacaine are potent, long-acting, amide-type local anesthetics that have many clinical applications including intra-articular administration. In veterinary practice, intra-articular administration of local anesthetics is used, especially in dogs and horses, for pain management during arthroscopic surgery, diagnosis of lameness, and for the control of pain associated with joint diseases such as osteoarthritis. However, studies have demonstrated that bupivacaine, levobupivacaine and ropivacaine all have chondrotoxic effects, which has raised questions on their safety when used for intra-articular injections. Therefore, the present thesis investigated the *in vitro* chondrotoxicity of bupivacaine, levobupivacaine and ropivacaine and evaluated the factors involved in their adverse effects towards cultured canine articular chondrocytes. The objectives of the studies performed were to evaluate their effects on articular chondrocyte viability and to elucidate the biomolecular pathways involved in their chondrotoxicity.

The dissertation is divided into two experimental chapters: The first chapter evaluated the *in vitro* chondrotoxicity of bupivacaine at low concentrations in cultured canine articular chondrocytes. The purpose of this study was to evaluate the effect of bupivacaine on chondrocyte viability at concentrations similar to *in vivo* synovial fluid concentrations at three different

time points. The second chapter evaluated the comparative *in vitro* effects of bupivacaine, levobupivacaine and ropivacaine on cell viability and caspase activity in cultured canine articular chondrocytes in order to elucidate whether they would activate the extrinsic or intrinsic pathways of apoptosis. The findings from the first chapter demonstrated that bupivacaine at low concentrations that are similar to *in vivo* synovial fluid concentrations can negatively affect chondrocyte viability especially with prolonged exposure. The second chapter showed that local anesthetics can induce apoptosis through both the extrinsic and intrinsic pathways, depending on the type of local anesthetic used. The level of chondrotoxicity, the type of caspase activated, and the level of caspase activation was dependent on the type of local anesthetic. Bupivacaine was the most chondrotoxic followed by levobupivacaine, and ropivacaine was the least chondrotoxic.

In conclusion, the *in vitro* chondrotoxicity of local anesthetics in cultured canine articular chondrocytes is dependent on the time of exposure, the concentration of the drug and the type of local anesthetic that the chondrocytes are exposed to during treatment. The cell death pathways induced by local anesthetics may also be dependent on the type of drug. Additionally, ropivacaine may be a safer choice for intra-articular local anesthesia in clinical applications compared to bupivacaine and levobupivacaine. Further studies are required to determine safe protocols and drug dosages for intra-articular local anesthetics; and to clarify the implications of local anesthetic-induced chondrotoxicity and how these adverse effects can be managed in clinical patients.