Effect of Nifedipine and Ketotifen on Joint Capsule Cell Based Collagen Gel Contraction

Kristi Billard, BSc; Mei Zhang, MD; Prism S. Schneider, MD, PhD; Dave Hart, PhD; Paul T. Salo, MD; ***Kevin Hildebrand, MD, FRCSC***
University of Calgary, Calgary, Alberta, CANADA

Purpose: Previous studies have shown that the calcium channel blocker Nifedipine (NF) and the mast cell stabilizer Ketotifen Fumarate (KF) may be used to manipulate the myofibroblast-mast cell-neuropeptide (MMN) axis that underlies joint capsule fibrosis in posttraumatic contractures. We tested our hypothesis that joint capsule (JC) cell-mediated collagen gel contraction will be decreased more when KF and NF are combined compared to either compound in isolation.

Methods: Posterior JCs from the contracture knee of 6 adult New Zealand white rabbits were harvested and transferred to a collagen gel assay using previously described methods. The gels were treated with assorted combinations of human mast cells (HMCs) 0 or 2.5×10^5, KF, and NF. KF was added at a concentration of 10^{-6} prior to gel casting overnight and NF was added at concentrations of 10^{-4}, 10^{-6}, 10^{-8}, and 10^{-10} to the JC cell/collagen gel mixture. After a 12-hour initial culture, the gels were released and photographed at 0-72 hours post-release. The contraction of collagen gels was measured and the area of the gel calculated using Image J image processing system. The gel contraction was expressed as a percentage of the gel diameter at 0 hours. Statistical comparisons used a 2-way (treatment, time) analysis of variance with a post-hoc Tukey test. Significance was set at $P \leq 0.05$.

Results: Collagen gel contraction increased significantly over time 6 hours postrelease and beyond. NF inhibited the contraction of a JC/collagen gel in a dose-dependent manner with statistically significant decreases when compared to controls at concentrations of 10^{-4} to 10^{-8}. The addition of mast cells partially reverses the inhibitory effect of NF. Ketotifen had no effect on the gel contraction if HMCs were not present. However, when KF was added to HMC and NF, KF enhanced the inhibiting effects of NF on the JC at concentrations of 10^{-4} to 10^{-8}.

Conclusion: Nifedipine resulted in a dose-dependent inhibition of JC (myofibroblast-mediated) collagen gel contraction. Ketotifen has no direct effect on the JC. In the current study it was demonstrated that mast cells must be present in order for KF and NF to have additive inhibitory effects. It appears that NF works directly on the JC while KF modifies JC gel contraction indirectly through its effects on the mast cell in this in vitro model of joint contractures. These results add to the growing body of evidence that drugs such as KF and NF can manipulate the MMN axis to control the severity of posttraumatic joint contractures and may lead to new safe and cost-effective treatment options.

See pages 401 - 442 for financial disclosure information.