

# Live Cell Analysis of Collagen Deposition and Cell Proliferation: Indicator of a Promising Osteoarthritis Treatment

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Hypertonic Dextrose (HD) has been clinically suggested to regenerate cartilage, yet the underlying cellular mechanisms are not well understood. The single published in vitro study, which employs an indirect measurement method, does not find evidence that HD induces cell proliferation or collagen deposition. To address this clinical-in vitro inconsistency, this study investigates HD effects on mouse cartilage (ATDC5) cells cultured in a custom-prepared normoglycemic media. Across multiple trials, I used advanced light microscopy to directly measure proliferation, metabolism, morphology, and collagen deposition. To quantify differences between HD and controls at baseline and follow-ups, I used single cell analysis (SCA) such as logistic regression and survival analysis, as well as aggregate cell analysis (ACA). I used Mannitol to assess effects of hypertonicity and Phosphate Buffered Saline (PBS) as an isotonic and non-glycemic control. HD-treated chondrocytes proliferated more quickly ( $p < 0.01$ ) and deposited more collagen ( $p < 0.05$ ) than mannitol and PBS treated controls. Shortly after treatment (HD) administration, cells began to shrink and undergo apoptosis relative to PBS treated controls. ACA showed that HD initially increased glycolytic metabolism relative to both mannitol and PBS controls across trials. The novel use of a mannitol control reveals that, as a treatment, HD first stresses chondrocyte cells via a hypertonic mechanism and then acts as a metabolic resource for chondrocyte cells to revitalize themselves. Study results align with clinical literature, thereby strengthening the argument that HD is a promising therapeutic option meriting further research. These study results focus attention on chondrocyte deposition of Collagen II.