Varying and Testing Compliance of Collagen-Based Gels for Tissue Engineering

by

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ABSTRACT

This study was performed to develop a reliable mechanical testing system to measure the mechanical moduli of variably compliant protein hydrogels. Unconfined compression testing systems need to be developed through optimization of parameters and often through creation of custom parts. Using testing of simple agarose gels, a consistent, straightforward unconfined compression testing system was created. Collagen and gelatin have been used as cell substrates for some time because of their protein structure and thermo-reversible gelation properties. Genipin is a naturally occurring, relatively non-cytotoxic crosslinking agent that binds free amine groups. This study investigated the combination of collagen gels and gelatin gels with a genipin solution to make variably compliant hydrogels. A wash study was done to determine the rate of removal of the potentially cytotoxic crosslinker, genipin. These gels were then tested with a ninhydrin assay to determine the degree to which they were crosslinked, and also were tested with the custom-made unconfined compression testing system. The results showed that genipin did increase the degree to which the gels were crosslinked and this resulted in an increase in the elastic modulus of the gels. The results were more drastic for the gelatin gels as compared to the collagen gels. At the 70 hour timepoint, the collagen gels had become 50% stiffer and the gelatin gels had become 75% stiffer. Our data show that the compression modulus of protein hydrogels can be altered by varying collagen-gelatin concentration. These results lay the foundation for future cellular studies investigating the effect of gel stiffness on cell differentiation and phenotypic expression.