Design of Alginate/Gelatin Hydrogels for Craniofacial Bone Tissue Engineering: Optimizing Osteogenesis in Dental Pulp Stem Cells Without Compromising Other Cellular Functions

Alginate/gelatin (Alg-Gel) hydrogels have been explored in combination with mesenchymal stromal/stem cells (MSCs) to support bone tissue formation, reconstruction and regeneration. A key challenge for clinical translation is optimizing the stiffness of Alg-Gel hydrogels to selectively enhance osteogenesis. In this study, we investigated the influence of hydrogel stiffness on the adhesion, morphology, proliferation, and osteogenic differentiation of dental pulp stem cells (DPSCs), identifying optimal formulations to decouple osteogenic cues from other cell behaviors. A range of Alg-Gel combinations was cast and cross-linked with 2% $CaCl_2$ to finetune the mechanical properties. Two formulations were selected: a “low-stiffness hydrogel” (2% alginate, 8% gelatin; 11 ± 1 kPa) and a “high-stiffness hydrogel” (8% alginate, 12% gelatin; 55 ± 3 kPa). The hydrogels exhibited distinct swelling and degradation profiles, with the stiffer formulation showing reduced degradation. Both supported robust DPSC adhesion and proliferation. However, osteogenic differentiation, assessed by alkaline phosphatase activity, Alizarin Red staining, and bone nodule formation, was significantly enhanced in the high-stiffness hydrogel. These findings demonstrate that the stiffness of Alg-Gel hydrogels can be fine-tuned to promote osteogenesis without compromising other essential cellular functions. Importantly, this biomaterial is currently being investigated as a scaffold for craniofacial bone tissue engineering applications.