

# Development of Bioartificial Bone Tissue through 3D-Bioprinting with Sepiolite, Eggshell, Gelatin Biocomposite and Mesenchymal Stem Cells

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Conventional medicine can currently treat many bone-related diseases, but problems including complex fractures and severe bone loss still remain largely unresolved. In this effort, under bone tissue engineering, the regenerative approach of 3D-bioprinting was embraced in order to respond to aforementioned concerns by developing bioartificial bone tissue capable of stimulating defective osseous regions to regenerate, with the first-time use of sepiolite and eggshell in the respective approach. In this study; composite bioinks comprising of eggshell, sepiolite, gelatin methacrylate and bone marrow mesenchymal stem cells were extruded through a 3D-bioprinter to build up bioartificial draft bone tissue. Cellular bioprintings from bioinks with different ratios of eggshell and/or sepiolite were conducted and then transferred to cell culture for investigations. Findings revealed that bioprinted models could maintain their geometric structural form in cell culture over 7-8 days via pH studies and microscopic observations. Moreover, in all experimental groups, it was determined that the number of cells increased between 30-82% during that time interval with AlamarBlue cell viability studies. The highest cell proliferation rate was detected in the composite model containing 5% eggshell and sepiolite, where the initial cell number  $4.67 \times 10^6 \pm 0.14 \times 10^6$  increased to  $9.12 \times 10^6 \pm 1.02 \times 10^6$  in 7 days. Ultimately, while promoting waste utilization and eco-friendliness, this study showed that the 3D-bioprinted models reinforced the viability of cells. Furthermore, evidence was laid out to prove the theoretical backbone of the study's vision for a regenerative and personalized treatment model that can potentially cope with high-level osseous problems.