ACCSCIENCE PUBLISHING

International Journal of Bioprinting

RESEARCH ARTICLE

Bioprinting with adipose stem cells and hydrogel modified with bioactive glass

Supplementary File

In order to study the release of gelatin from the alginate+gelatin (AG) hydrogel used in this study, AG, 1.25G, and 2.5G scaffolds measuring 15 × 15 × 1 mm^3 were fabricated without cells, crosslinked with 0.1 M $CaCl_2$ solution for 10 min, and washed twice with deionized (DI) water. The samples were soaked in DI water in airtight containers under standard culture conditions for up to 7 days. The surrounding DI water collected after 1 day and 7 days, including the $CaCl_2$ solution used for crosslinking, were all analyzed for the presence of gelatin using proton nuclear magnetic resonance (^{1}H-NMR) spectroscopy (Bruker 400 MHz Avance™ III HD, Billerica, MA). First, known quantities of gelatin were dissolved in DI water, and 0.2 mL of gelatin solution was mixed with 0.6 mL of deuterium oxide (99.9 atom %, Sigma Aldrich, St. Louis, MO), and the solution was transferred to NMR tubes (Colorspec®, Sigma Aldrich, St. Louis, MO) for analysis. The area under a unique characteristic gelatin peak at ~1.9 ppm (Figure 2a on main article) on the horizontal axis was calculated and averaged for known quantity of gelatin concentration to obtain a gelatin standard curve as shown in Figure 2b on the main article.

Later, DI water used to soak AG, 1.25G, and 2.5G scaffolds was collected from the containers and analyzed for presence of gelatin. The area under the specific peak was calculated, and standard curve was used to determine the amount of gelatin present in DI water. Figure 14 on the main article shows the cumulative percentage of gelatin that is released from AG, 1.25G, and 2.5G scaffolds to the surrounding DI water with time. Even as the differences are not significant until 24 h, the amount of gelatin that was released from AG scaffolds was significantly high in comparison to gelatin release from 1.25G and 2.5G scaffolds. This result indicates that the addition of B3 glass could possibly impact the adhesion of alginate and gelatin molecules, thereby slowing down the release of gelatin from the hydrogel matrix to surrounding environment.