ABSTRACT

The physiology of a variety of different cell types has been shown to be remarkably different within 3-dimensional (3D) environment compared to 2-dimensional (2D) monolayer culture environment. Solid 3D scaffolds made from synthetic polymers offer various structural and mechanical features such as optimum surface to volume ratio, open pore structure, and optimum geometry for stem cell specific applications. Concomitantly, visualization of 3D growth and differentiation should be done in a minimally destructive manner to better assess cellular structure. This creates the need for rapid and easy visualization of 3D cultures using non-invasive technologies. The objective of the study is to compare the osteogenic differentiation of Ad-MSCs in 2D and 3D geometries and 3D visualization using the full field optical coherence imager, Light-CT. Mesenchymal stem cells (MSCs) derived from fat (Ad-MSCs) are an excellent source of stem cells due to waste tissue availability (liposuction) and abundance of fat tissue associated stem cells. Ad-MSCs have the potential to become a major source of MSCs for Tissue Engineering of bone and cartilage. The assay was carried out using 3D PS scaffolds with fiber diameter of 150-um and fiber-to-
fiber spacing of 200-um. Ad-MSCs were seeded at 11,000-cells/cm² on 48-well plates and 48-well PS scaffolds. The cultures were supplemented with osteogenic differentiation media (ODM) for 1, 2, and 3 weeks. The degree of osteogenic differentiation was assessed using the Alizarin Red staining method, which shows positive for the presence of calcium. The degree of calcium deposition in 3D PS scaffolds was greater than in 2D PS plates at each time point. Particularly, calcium mineralization in 3D was 7-fold higher than in 2D at week 1 of osteogenic differentiation. Light-CT images were taken at the 3D matrix surface and at different depths into the 3D porous structure. Extensive bone-like matrix was found in the porous structure in between the polystyrene fibers in the scaffold at different depths. This demonstrates that the Light-CT can be an enabling tool to image cellular and tissue-like structures on 3D polymer scaffolds for potential Tissue Engineering and Regenerative Medicine in vitro applications.