495b Application of Micropatterning Techniques to Co-Culture Systems for Hepatic Tissue Engineering

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Micropatterned culture systems may be appropriate in vitro models for application to hepatic tissue engineering, such as drug screening and cellular interactions studies. It was reported that hepatocyte co-culture with other cell types improved hepatocyte functionality and life span in culture. In co-culture systems, heterotypic cell-cell interactions are important for the maintenance of liver-specific functions. Microfabrication techniques can be used for the spatial control of cells in culture. We previously reported micropatterning of hepatocyte and fibroblasts in co-culture to study the role of homotypic and heterotypic cell-cell interactions. In this study, we introduce three different types of patterns using hepatocytes, 3T3 fibroblasts, and endothelial cells in co-culture systems to better understand the role of cellular interactions. In addition, to increase the heterotypic interface between hepatocytes and fibroblasts, hepatocytes were patterned on fibroblast feeder layers and compared to other types of patterns. We fabricated PDMS stencils to pattern circular holes with 500-1000 micrometer diameter sizes. Different types of cell patterns using hepatocytes, 3T3 fibroblasts, and endothelial cells, were employed to evaluate and characterize the cellular interactions in co-culture systems. Medium samples were collected for functional analysis. The patterned cells were characterized by immunofluorescence and immunohistochemistry. In our preliminary studies, high expression of albumin and bromodeoxyuridine (BrdU) positive hepatocytes at the boundaries between hepatocytes and 3T3 fibroblasts were observed. Higher liver-specific functions were observed with increasing heterotypic interface between hepatocytes and fibroblasts. Further characterization of patterned cells will be reported.