495a Investigation of Chemical Effects on Neural Progenitor Cell Adhesion and Differentiation Using Micropatterned Substrates

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Current research indicates that a variety of chemical and physical cues work together to cause progenitor cell differentiation. In vitro, a homogeneous population of adult hippocampal progenitor cells (AHPCs) exposed to the same conditions will differentiate into all three classes of neural cells. Our hypothesis is that individual cells in an AHPC population are diverse in their capability to respond to their environment, and to test this we will expose AHPCs to combinations of signals from diffusible factors and preformed distributions of extracellular matrix (ECM) proteins. Prior results from our laboratory have shown that microgrooved substrates with proteins in the grooves influence neural progenitor differentiation when they are co-cultured with astrocytes. Microgrooved polystyrene substrates have been fabricated and chemically modified with poly-L-lysine and laminin. Rat postnatal type-1 astrocytes and AHPCs cultured on these substrates have aligned well along the grooves of the patterned surface. AHPCs have appeared highly elongated and extended their processes in the direction of the grooves and along groove boundaries. AHPCs have been co-cultured on top of near confluent monolayers of astrocytes on the patterned substrates. As the majority of the AHPCs were not in direct contact with the patterned substrate, it appears that directed outgrowth may be stimulated by properties of the astrocyte surface. Differentiation of the AHPCs in co-culture and growing alone were assessed morphologically and immunocytochemically up to 6 days in vitro using cell type specific antibodies. A significantly higher percentage of the AHPCs expressed neuronal markers in co-culture on micropatterned surfaces than on planar surfaces or growing alone. To create distributions of ECM proteins without the microgrooves, polydimethylsiloxane stamps were created from silicon microdies. A FITC conjugated ECM protein was applied to glass coverslips with the stamps and visualized with fluorescence microscopy. The effect of these ECM protein distributions on neural progenitor cell growth and differentiation was investigated. These results were compared to the effect of ECM proteins in microgrooves on polymer substrates on progenitor growth and differentiation to ascertain the role of diffusion limitations in the microgrooves on neural progenitor cell differentiation.