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Development Of 3-d Cell Culture Methods For Modeling Long Term Neuron Culture And The Blood-brain Barrier

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Most strategies for mammalian cell culture involve attachment of dissociated cells to planar substrates. Such strategies are relatively easy to perform, allow observations of individual cells that are difficult to make in vivo, and have provided information on dynamic cellular processes for over a century. However, cells cultured in two dimensions behave radically different from cells in their native three dimensional environments. We hypothesize that three-dimensional (3-D) cell cultures will behave differently from two-dimensional (2-D) cultures and will more closely mimic in vivo function and organization. To test this hypothesis, 3-D scaffolds will be made from hydrogels. Scaffolds will be developed capable of holding cells in three dimensions. The effects of encapsulation on the viability and functionality of individual primary cultures of astrocytes and neurons grown within or on the surface of a variety of substrates has been investigated. Substrates include poly-(ethylene) glycol diacrylate (PEGDA), hydroxyethyl methacrylate (HEMA), and alginate hydrogel matrices. Peptides were incorporated into the gels by covalent tethering to hydrogel monomers or by bulk polymerization within hydrogels to promote cell growth and proliferation. Cell viability was assessed by nuclear labeling with vital stains. Proliferative capacity of astrocytes, microglia, and endothelial cells was monitored by Bromodeoxy Uridine (BrdU) incorporation. Cellular differentiation was monitored by observing changes in cell morphology. Two cell culture models will be developed using 3-D culture technology: long term neuron cultures and the blood-brain barrier.