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(101) Development of a 3-Dimensional *In Vitro* Model to Study Reactive Gliosis Following Nervous System Injury

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Injury to the spinal cord results in the formation of a glial scar which is associated with inhibition of axonal regeneration. One of the major limitations of research into improving repair strategies is the lack of a cell culture model that accurately recapitulates the complex *in vivo* situation. Our aim is to develop an effective model to address this need.

Astrocytes in the undamaged CNS express low levels of GFAP, but following injury exhibit a reactive phenotype exemplified by GFAP up-regulation. Primary glial cell cultures were analysed in 2D monolayers and 3D collagen gels for GFAP expression. In 2D cultures $73.4 \pm 4.0\%$ of cells were GFAP positive, whereas $40.7 \pm 3.5\%$ were immunoreactive for GFAP in 3D collagen gels. As 3D astrocyte cultures more closely modelled the *in vivo* situation we used this model to investigate the response of astrocytes to dorsal root ganglia cells (DRGs). Dissociated DRGs were labelled with CellTracker™, seeded onto astrocyte-populated collagen gels and maintained in culture for 5 days. Astrocytes near the DRG interface showed marked GFAP up-regulation and adopted a reactive morphology which was observed up to 1 mm away.

Astrocytes in 3D culture exhibit a lower basal level of reactivity than cells grown in monolayer, providing a system in which stimulation of activation can be investigated. This model provides a useful tool for investigating triggers of reactive gliosis, as demonstrated by the response observed to cells found at the inhibitory interfaces formed following damage to the spinal cord.