Carbon Nanotubes Direct Neurite Growth: A Substrate for Neural Repair?

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Introduction: Failure of central nervous system (CNS) regeneration following injury occurs for many reasons, including the presence of inhibitory factors associated with myelin, glial scar formation, and lack of guidance cues for regenerating axons. In particular, there is increasing evidence that a highly aligned supporting substrate may be needed to promote accurate regeneration. Carbon nanotubes (CNT) consist of very densely packed, aligned bundles of tubular carbon molecules. The aligned structure could serve to guide growth in a particular direction. CNT can theoretically be produced with suitable geometry to allow the entry and growth of neurites (~1um diameter) while excluding the entry of larger cell bodies (~10um)such as astrocytes and fibroblasts that contribute to scarring at CNS injury sites.

Methods: Sympathetic chain ganglia and cortical neurons from E9-10 chick embryos were harvested and plated on CNT bundles of varying diameters and orientations to assess their ability to support neuronal attachment and neurite outgrowth. CNT arrays with various spacing between bundles were also used as templates for growth to further examine the effects of geometry and topography. Living neurons and their processes were stained with a vital dye and imaged with an inverted fluorescence microscope.

Results: Imaging revealed neuronal attachment to CNT with extensive aligned neurite outgrowth in the direction of the CNT. Neurite fascicles formed on CNT when clusters of several cell bodies were present. Neuronal attachment and outgrowth was also achieved on non-aligned CNT bundles, but the neurites did not show obvious orientation.

Discussion: The contrast between growth on aligned and unaligned CNT bundles was striking. Growth on aligned bundles appeared extensive, parallel, and with minimal branching. Incident light imaging confirmed that growth was in the direction of alignment. Growth on unaligned CNT, such as on the top of a vertical array, appeared randomly oriented. This pattern was seen in cultures of both cortical and sympathetic neurons. The results suggest that aligned carbon nanotubes might be a suitable material for supporting axonal regeneration.

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