Nanofibrous Materials for Tissue Engineering
Investigating the Cellular Response to Multiwalled Carbon Nanotubes

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Background

A number of surgical interventions against traumatic injury of cartilage and bone require the use of graft material to fill defects and progress healing. It soon may be possible to fabricate synthetic tissue scaffolds that replicate the structure and function of natural extracellular matrix (ECM), and expose encapsulated cells to growth factors and other cytokines, with the goal of tailoring the process of tissue regeneration (1). Many challenges must first be overcome – including further understanding of the cellular response to nanofibrous synthetic structures.

Primary aim: to investigate the response of osteoblast cells to multi-wall carbon nanotubes (MWCNTs) of similar diameter to collagen fibres – a common constituent of the ECM.

Methods

Fabrication: MWCNT arrays were fabricated using chemical vapour deposition (2).

Cell culture: Three cell types were investigated - human osteosarcoma cells (MG63), primary osteoblasts at third passage (POB) and human lung epithelial carcinoma cells (A549).

SEM Imaging: Characterisation of the MWCNT surfaces (Fig.1) and cell interaction (Fig.2) was investigated using a LEO Gemini FEG SEM.

Cell Studies: Cells were stained with fluorophors to reveal morphology and spreading. A commercially available assay (MTS: Promega, UK) was used to assess cell proliferation at 1 and 7 days.

Results

Nanotube diameter: 35nm ±10nm

- Cells were not able to penetrate into the MWCNT arrays (Fig.2).
- Although different cell morphologies were seen on all surfaces, only point focal contacts were observed on the MWCNT arrays for all cell types (Fig.3).
- Cells proliferated on the nanotube surfaces (Fig.4), but this was reduced in comparison to the glass surfaces (data not shown).
- Cell spreading was reduced on the nanotube surfaces in comparison to the glass and highly ordered pyrolytic graphite (HOPG) controls (Fig.5).
- Fine filopodia were seen to extend from cells cultured on the MWCNTs (Fig.6)

Conclusions

A network of MWCNTs, as a model nanofibrous system, provides a planar environment that cells are unable to penetrate or significantly alter, and is accessible to standard 2D culture techniques. In this study (3), cells reacted to the MWCNTs with reduced spreading and displayed a range of morphologies. A number of POB cells were found to display reduced widening but increased elongation. Only smaller sized focal contacts were found on the nanotube substrates, which may have affected cell migration, and proliferation.

References