INTRODUCTION

The increased popularity and success of metal-on-metal (MOM) hip implants has resulted in the emergence of similar MOM prostheses for use in total disc replacement (TDR). However, wear debris generated from such metal prostheses can lead to adverse tissue reactions as reported in recent studies[1, 2]. Consequently, adequate evaluation of the cellular responses to metal wear debris generated by artificial intervertebral discs is warranted. Hence, the aim of this study was to investigate the biological effects of clinically relevant metal nanoparticles on cells isolated from the dura mater, a tissue in close proximity to spinal implants.

METHODS

Wear Particles were generated in a 6 station pin-on-plate wear rig under a force of 80 N. The bearing combination studied comprised medical grade wrought cobalt-chrome (Co-Cr) alloy ASTM F1537 pins and plates with smooth counterparts (Ra: 0.01-0.02µm). Wear particles were then sterilised at 180°C for 4 hours and were characterized using FESEM and Image Pro-plus as described previously [3]. Porcine dural fibroblasts and dural epithelial cells were isolated from the dura mater. The cells were cultured with Co-Cr nanoparticles at particle volumes ranging from 0.06 µm³ to 121 µm³ per cell. The resulting biological effects were evaluated using a range of cell-based assays. Cell viability was assessed using the ATP-lite™ assay at 24 hour intervals over 4 days. Production of the proinflammatory cytokines IL-8 and IL-6 in the culture supernatants was determined by enzyme-linked immunosorbent assay (ELISA) and oxidative stress was assessed using 5- (and - 6) - carboxy-2', 7'- dichlorodihydrofluorescein diacetate (carboxy - HDCFDA) as a fluorescent probe after a 24 hour exposure of the cells to a particle volume of 50 µm³ per cell.

RESULTS

Wear particles were observed to be uniformly round to oval in shape and 10-80 nm in size. Differential effects of the Co-Cr nanoparticles were observed on the dural fibroblasts and epithelial cells. The particles significantly reduced the viability of the epithelial cells (p<0.05 ANOVA) in a dose and time-dependent manner but not the fibroblasts when compared to the cell only negative control (Figures 1 & 2). Both the fibroblasts and epithelial cells were induced to secrete IL-8 when cultured with the particles at doses of either 60.5 or 121 µm³ per cell or both (p<0.05 ANOVA). No significant release of IL-6 was observed in both cell types at all doses.

DISCUSSION

The survival of the dural fibroblasts at doses cytotoxic to the epithelial cells suggests novel differences in the resistance of the two cell types to metal nanoparticle-induced toxicity which may be regulated, in part, by a difference in their susceptibility to reactive oxygen species. Moreover, secretion of IL-8 by the epithelial cells and fibroblasts upon culture with CoCr nanoparticles indicates the inflammatory potential of the wear particles.

SIGNIFICANCE

The results generated from this study contribute to a greater understanding of the potential risks associated with the use of MOM total disc prostheses.

REFERENCES

2. Guyer et al., Spine (2011)

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