An investigation of the mechanisms underpinning the effect of anti-inflammatory drugs on neural stem cells

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Abstract

Anti-inflammatory drugs such as corticosteroids (CSs) and minocycline (MINO) are widely used in the treatment of a range of clinical conditions and to suppress graft rejection in stem cell transplantation therapy. However, such treatment is associated with adverse effects on brain development. The effects of anti-inflammatory drug on neural stem cells (NSCs) are largely unknown and the molecular mechanisms underlying these effects are poorly documented. The focus of this project is to systematically investigate the effects of different anti-inflammatory drug at different concentrations on the fate of NSCs using two different *in vitro* models.

In this thesis, it is shown that all three types of CSs (dexamethasone, prednisone and methylprednisolone) affect NSCs propagated in monolayers and neurospheres. Comparison of the monolayer and neurosphere growth formats for NSCs following CS treatment revealed that CS decreased NSCs proliferation and neuronal differentiation while accelerated the maturation of oligodendrocytes without concomitant effects on cell viability and apoptosis. The findings suggest that the difference in the physical format of NSCs does not impact on CS influences on these cells with similar results obtained for both culture systems.

Further, label-free quantitative proteomics was used to study methylprednisolone effects on NSCs at the cellular and molecular levels in monolayer cultures. Proteomics and bioinformatics analyses revealed that methylprednisolone induced downregulation of growth associated protein 43 and matrix metallopeptidase 16 with upregulation of the cytochrome P450 family 51 subfamily A member 1. These

findings support the hypothesis that neurological deficits associated with CS treatment mediated via effects on NSCs, and highlight putative target mechanisms underpinning such effects.

Finally, the organotypic spinal cord slice model was used to investigate the efficacy of MINO as a combinatorial therapy with transplanted NSCs. The data from neurosphere culture showed that MINO had no direct effect on key regenerative properties of NSCs such as proliferation and differentiation. While, the findings from organotypic spinal cord slice culture showed the astrogliosis and activated microglia were reduced and the outgrowth of the nerve fibres was increased following a combinatorial therapy. This study demonstrates the utility of the organotypic spinal cord slice model to test the efficacy of MINO as a combinatorial therapy with transplanted NSCs.