Lentiviral-mediated gene therapy for mucopolysaccharidosis type IIIA

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1 Abstract

Gene therapy is promising for the treatment of monogenetic disorders because it aims to restore overall homeostasis, not by treating disease symptoms, but by targeting the fundamental cause of disease. Viral vectors are valuable tools for mediating the transfer of therapeutic genes to target cells, and lentiviral vectors in particular are well suited to this role as they chromosomally integrate, can enter dividing and non-dividing cells, and evoke little or no immune response.

The aim of the current thesis was to evaluate the potential of using lentiviral-mediated gene therapy for the treatment of mucopolysaccharidosis type IIIA (MPS IIIA), a heritable lysosomal storage disorder affecting the central nervous system (CNS). The chronic and progressive course of MPS IIIA results from lysosomal accumulation of heparan sulphate (a highly sulphated glycosaminoglycan), secondary to deficiency of the lysosomal hydrolase sulphamidase. Accumulation of heparan sulphate within the cells of the reticulo-endothelial system, the monocyte-macrophage system, and neurons, leads to hepatosplenomegaly and severe, progressive neuropathology in affected children, and ultimately to death at around 15 years of age. There are currently no effective treatments for MPS IIIA patients.

The somatic and CNS aspects of pathology in a mouse model of MPS IIIA were addressed in this study using two separate methods of therapeutic gene delivery; intravenous gene delivery, which directs gene transfer to the liver, and gene delivery to the brain *via* the

cerebral lateral ventricles which utilises the cerebrospinal fluid to achieve gene distribution throughout the brain

After intravenous administration of a self-inactivating lentiviral vector expressing murine sulphamidase to young adult MPS IIIA mice, the livers of treated animals were effectively modified to express high levels of therapeutic sulphamidase. The resultant widespread delivery of enzyme secreted from transduced cells to somatic tissues *via* the peripheral circulation corrected most somatic pathology, furthermore, markers of MPS IIIA pathology within the brains of treated mice were significantly reduced. When liver directed gene therapy was repeated in a second cohort of mice, however, similar benefits to the brain were not observed, presumably because the resulting levels of peripherally circulating enzyme were comparatively low. Although, in common with the first study, somatic pathologies still were corrected. Alternatively, lentivirus delivered to the brains of MPS IIIA mice *via* the cerebral lateral ventricles achieved extensive sulphamidase gene distribution and reduced lysosomal storage throughout the brain. Improvements in behaviour were observed for these animals, as was the complete prevention of pathological urine retention.

The blood-brain-barrier (BBB) limits the transfer of therapeutic enzymes from the blood to the brain, therefore gene therapy approaches to treat CNS pathology in the MPS are to either challenge the BBB with high levels of circulating enzyme, or bypass it altogether, by the direct delivery of therapeutic genes into the brain. While both approaches have advantages for the treatment of MPS IIIA, the results presented for the current thesis

suggest that gene delivery to the brain *via* the cerebral lateral ventricles may, at this stage, be the more practical and efficacious approach to the treatment of CNS pathology in MPS IIIA patients.

2 Declaration

This work contains no material which has been accepted for the award of any other degree or diploma in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text.

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