Development of a gene-activated scaffold for iPSC-neuron-directed spinal cord injury repair

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Background: Spinal Cord Injury (SCI) is a traumatic life-changing injury that induces permanent loss of function and paralysis below the site of injury. A biomaterial scaffold capable of bridging the lesion cavity and delivering human induced pluripotent stem cell (iPSC)-derived projection neurons to the injured cord might offer the potential to help drive functional recovery after SCI. However, few implants are optimally designed to actively support axonal regrowth and the viability of transplanted iPSC-neurons. Therefore, this study aimed to develop a biomimetic gene-activated biomaterial scaffold carrying axon growth-inducing nucleic acid cargoes encapsulated in nanoparticles (NPs) capable of transfecting implanted iPSC-neurons to target SCI repair.

Methods: Non-viral NPs were generated by complexing the GAG-binding enhanced transduction peptide with reporter plasmid DNA and thereafter added to iPSC-neural progenitors (NPCs) and neurons to determine the optimal NP concentration for safe and effective transgene expression. Hyaluronic acid-based biomimetic scaffolds were prepared and functionalised with non-viral NPs containing reporter plasmids to assess NP incorporation, loading efficiency, and release dynamics, and then subsequently seeded with iPSC-NPCs to validate scaffold-mediated transfection.

Results: Results demonstrated successful and sustained non-viral transfection of iPSC-neural progenitors and neurons without inducing changes in cell viability, cytotoxicity or morphology, indicating biocompatibility of the NPs. Scanning electron microscopy demonstrated the successful loading and distribution of NPs throughout the scaffold. NP loading efficiency was greater than 96%, and evaluation of the NP release profile demonstrated a high retention rate, with scaffolds retaining the total loaded genetic cargo over 7 days. Finally, ongoing analysis validates scaffold-mediated transfection, demonstrating enhanced infiltration, survival and outgrowth of iPSC-neurons on the developed growth factor encoding gene-activated scaffold.

Conclusions: In conclusion, we outline the development of an iPSC-neuron optimised gene-activated scaffold containing nanoparticle-encapsulated plasmid DNA capable of trophically supporting neuronal survival and growth. The platform will serve the dual purpose of providing a growth-supportive environment for regrowing axons while also facilitating the formation of new neural connections in the distal cord, thereby increasing the potential for functional recovery after injury.

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