



**Karolinska  
Institutet**

Institutionen för Klinisk Neurovetenskap

# **Immune Cells and Stem Cells in Spinal Cord Injury: Defining Spinal Cord Injury Associated Microglia**

AKADEMISK AVHANDLING

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# ABSTRACT

Traumatic spinal cord injury (SCI) commonly results from falls, sport activities or traffic accidents and most often results in lost sensory, motor and/or autonomic functions below the level of injury. The cellular- and immune response following SCI is complex and has implications for regeneration and recovery. The immune cell response, and the microglial response in particular, has not yet been investigated at single-cell resolution. Furthermore, several questions remain concerning the importance of histocompatibility of transplanted stem cells, their cellular response and their contribution to functional recovery following SCI. This thesis investigated stem cell transplantation as a therapeutic approach for SCI as well as disease-specific transformations of immune cells, focusing on microglia, following SCI.

In the *first project* we investigated the causality between transplantation of neural progenitor cells (NPCs) and recovery of hind limb locomotor function following SCI. Ventricular-subventricular zone (V-SVZ) derived NPCs were transplanted into the injury epicenter of rats subjected to severe contusion SCI. The NPCs were investigated in terms of differentiation, transcriptional profile, effect on neuroinflammation and causal contribution to recovery of hind limb locomotor function. We found that there is causality between transplantation of NPCs and recovery of hind limb locomotor function and that this correlates with their differentiation into oligodendrocytes, enhancement of myelination and suppression of neuroinflammation. In the *second project* we investigated the importance of histocompatibility of mesenchymal stem cells (MSCs) transplanted into SCI. Syngeneic- or allogeneic mouse bone marrow derived MSCs were transplanted into the injury epicenter of mice subjected to severe contusion SCI and the immune- and inflammatory response as well as the contribution to functional recovery was investigated. We found that syngeneic MSCs activate macrophages alternatively and enhance neuronal survival, which correlates with suppression of inflammation and enhancement of hind limb function. Thus, the histocompatibility of transplanted MSCs is of importance for their therapeutic potential in SCI. In the *third project* we investigated the cellular response of MSCs transplanted into SCI. Mouse bone marrow derived fluorescent MSCs were transplanted into the injury epicenter during the acute phase of SCI in mice and isolated one week later. The global transcriptional profile, phenotype and fate of the MSCs was investigated. We found that MSCs transplanted into SCI adopt immune-cell like characteristics by up-regulating expression of genes and surface markers associated with immune cells and immune system functions (phagocytosis/endocytosis, production of cytokines/chemokines). In the *fourth project* we investigated disease-specific transformations of immune cells in SCI at single cell resolution. SCI was induced in mice and CD45<sup>+</sup> immune cells were isolated from the spinal cord at various time points and subjected to single-cell RNA sequencing. The gene expression analysis was supplemented with histological evaluation as well as a depletion model of microglia. We found that following degeneration, demyelination or trauma to the CNS homeostatic microglia undergo a distinct temporal transformation resulting in a disease-associated subtype of microglia, which persists in the chronic phase of the injury and has a beneficial role for functional recovery.

In conclusion, stem cell therapy for SCI shows a great deal of potential but is, at the moment, not sufficient or efficient enough to restore function to a near-normal level. Moreover, following SCI microglia undergo disease-associated transformations, which persists in the chronic phase of the injury, and contribute to functional recovery.