Pre-clinical Validation of Superparamagnetic Iron Oxide Nanoparticle-Labeled Neural Stem Cells for In Vivo Tracking and Post-Mortem Identification in the Spinal Cord

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Introduction

Cell-based therapies have been proposed for a wide variety of neurologic disorders in the spinal cord. Clinical trials using stem cells to treat these disorders have shown promising results to date. However, most of these trials lack a feasible approach for tracking the cells *in vivo* and identifying them post-mortem.

Approaches for tracking cells that have been proposed rely on either genetic modification of the cell or physical labeling with intracellular contrast agents. We propose physically labeling cells with ferumoxytol (Feraheme, AMAG Pharmaceuticals, Inc, Lexington, MA, USA), an ultra-small superparamagnetic iron oxide nanoparticle. Ferumoxytol was chosen because it was internalized by the cells, produced ample local magnetic field inhomogeneities that decrease T2 relaxivity for visualization with Magnetic Resonance Imaging (MRI), and it is approved by the US FDA.

The goal of the present study is to develop and validate a rapidly translatable methodology for *in vivo* visualization and post-mortem identification of stem cells transplanted into the spinal cord.

GDNF-Secreting hNPC Neurospheres

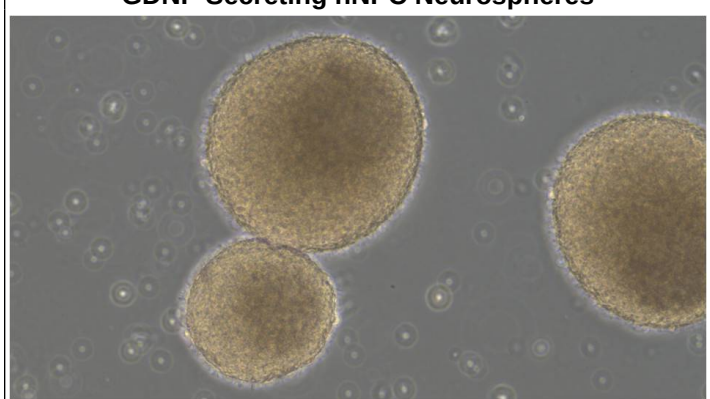

Figure 1: GDNF-secreting hNPC neurospheres visualized with light microscopy (20X Magnification).

In Vitro MR Visualization of GDNF-hNPC

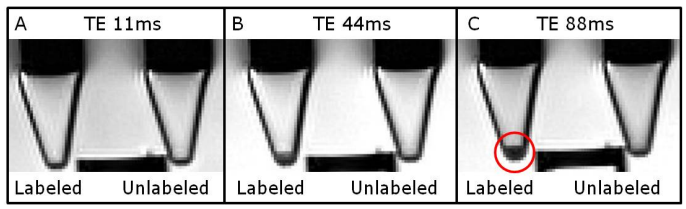

Figure 2: Sagittal MR images through 15 mL conical tubes containg ferumoxytol labeled and unlabeled GDNF-hNPC pellets. Labeled and unlabeled cells imaged at Echo Time (TE) A) 11ms, B) 44ms, and C) 88ms demonstrating significant negative T2 contrast that increases with TE.

Methods

- Glial cell line-derived neurotrophic factor secreting human neural progenitor cells (GDNF-hNPC) grown in neurospheres (Figure 1) were dissociated and labeled with 400 ug/mL ferumoxytol, 10 ug/mL protamine sulfate, and 2 IU/mL heparin sulfate.
- Cell viability was measured with trypan blue and labeling success assessed with MRI.
- MR Images were acquired using a Siemens Trio Trim 3T Full Body MRI.

MR T2 Relaxivity Curve of GDNF-hNPC

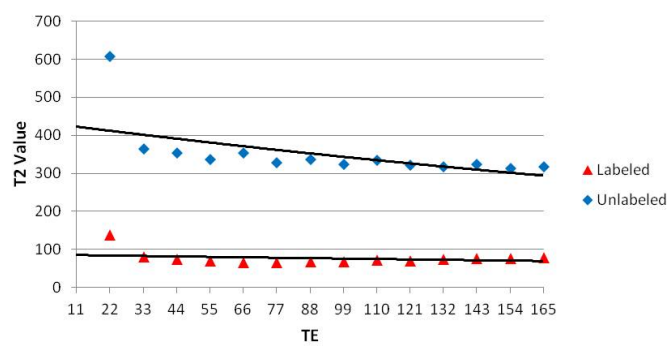

Figure 3: Ferumoxytol labeled and unlabled GDNF-hNPC pellets imaged with MRI. T2 values are significantly reduced in the labeled cells.

Results

- *In vitro* cell viability was not altered by labeling (90.5% labeled vs. 85.2% unlabeled GDNF-hNPCs viable).
- Significant contrast differences between labeled and unlabeled GDNF-hNPC pellets were observed with MRI (Figure 2) and quantified with T2 relaxivity curves (Figure 3).
- *In vitro* detection limit of ferumoxytol labeled cells was 2E4 cells in an agarose phantom (Figure 4).

In Vitro MR Detection Limit of Labeled Cells

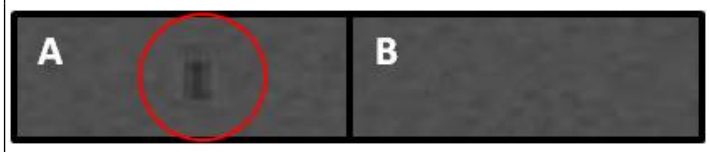

Figure 4: Axial MR images through agarose gel phantom visualizing A) 2E4 ferumoxytol labeled cells, the lowest total labeled cell amount visualized and B) 2E4 unlabeled cells not visualized.

Conclusions

Intracellular MRI contrast agents serve as a valuable tool for tracking neural stem cells in the spinal cord. Additional work must be completed to assess the effect these particles have on cell function and the feasibility of scaling this approach to humans.

References

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