Ex vivo MRI of Neuronal Progenitor Cells in the Subventricular Zone of the Lateral Ventricle

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Abstract:
In the adult mammalian brain, neural progenitor cells (NPCs) reside in the subventricular zone (SVZ) of the lateral ventricles. These neuronal progenitors differentiate into new neurons that migrate to populate the olfactory bulb (OB). This migration occurs along a distinct migration pathway called the Rostral Migratory Stream (RMS). This study will utilize intracerebroventricular (ICV) injection to apply a super-paramagnetic iron oxide (SPIO) contrast agent. We show that the MR signal detection is limited to the ventricle and that non-specific diffusion of the contrast agent occurs. Any extraventricular signal detection would be result from migrating NPCs labeled with contrast. Successful labeling of these NPCs will provide the opportunity to conduct longitudinal in vivo studies of neurogenesis and migration in traumatic brain injury (TBI).

Materials and Methods:
Single unilateral ICV injections were carried out using a 50-µL Hamilton Microliter Syringe connected to a temporary cannula. 5 µL of 50 and 75 µg/mL of SPIO (Feridex, Bayer, Inc) solution were injected into the lateral ventricle. Animals were sacrificed 120 hours after injection and were transcardially perfused with 0.9% saline followed by 4% paraformaldehyde (PFA).

MRI Equipment:
11.75 T, 500-MHz WB vertical magnet at CoE, FAMU-FSU, equipped with a Bruker Advance console 6.5 & Micro-2.5 gradients and Bruker 11.7-T birdcage RF coil resonant at the H NMR frequency.

Imaging Protocol: 3D Gradient-Recall Echo (GRE) with T/TE/TR=7/5/1000 ms with an isotropic resolution of 50 µm. Scan time was ~9 h with 2 averages.

Introduction:
In the mammalian brain, the SVZ of the lateral ventricle provides a continual source of NPC throughout adulthood. After subsequent differentiations, these cells become new neurons, which migrate to and populate the OB. This neural migration occurs along the RMS, which is a well-described migration pathway (Shapiro et al. 2006). Upon relocation to the OB, these new neurons differentiate into granule cell neurons and periglomerular neurons (Lois and Alvarez-Buylla, 1994). It has been shown that experimentally induced TBI stimulates SVZ proliferation of neural progenitors and provokes deviation in the migration pattern of the RMS (Salman et al., 2004). This phenomenon is of particular importance because similar NPCs have been discovered in non-human primates (Kornack and Rakic, 2001) as well as in humans (Salman et al., 2004).

SPIO contrast agents are endocytosed by NPC, allowing for continuous MR tracking during their migration to the OB. Coupled with highly sensitive SPIOs, high field MRI is a non-invasive imaging modality that offers an excellent spatial resolution and allows for continuous longitudinal and tracking of NPC during their migration (Adams, et al. 1999).

Discussion:
The MR images validated the previously described ICV delivery technique. It has been shown that over the course of 5 days, the SPIO diffused throughout the ventricle, thereby making it available to neuronal progenitors located in the SVZ on the lateral wall. Furthermore, it was shown that the Feridex was effectively contained within the ventricle for the duration of the experiment and that no detectable amounts of SPIOs were observed beyond the ventricle, meaning that SPIOs in the ventricle are not susceptible to passive diffusion throughout the surrounding tissues. Thus, it can be concluded that any extra-ventricular hyperintense MR signal detection would be the result of cells actively endocytosing the SPIOs and migration away from the ventricle to another location. This observation is important in confirming the validity of future deductions. Future studies will include longer incubation periods, in vivo analysis and longitudinal studies coupled with experimentally-induced TBI to track deviations in migration patterns.

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