Introduction: Investigation of preclinical stem cell donor efficacy is imperative to translational efforts in stroke treatment. Here, treatment efficacy between different donors of human mesenchymal stem cells (MSC) were assessed after acute application to a rat model of transient ischemia. Biochemical markers were measured longitudinally over 21 d using sodium chemical shift imaging (CSI), relaxation enhanced MR spectroscopy (RE-MRS) and T2-weighted proton imaging. Applying these techniques at ultra-high field (21.1 T) provide increased sensitivity, enabled insight into ionic and metabolic homeostasis, and demonstrated the extent of tissue recovery dependent on donor metrics.

Methods: Two human donors were evaluated in this study, one considered to be compromised (Donor 1) and one with demonstrated therapeutic efficacy (Donor 2). Cell assays for population doubling time, colony forming, immunomodulation and senescence were completed through passage 8 (P8) for both donors prior to in vivo experiments. At P5, 3D aggregates approximately 400-µm in diameter were induced in ultra-low attachment well plates, and subsequently dissociated and incubated with 7.47 µg Fe/mL micron-sized particles of iron oxide prior to injection. A transient MCAO model1 was instituted in Sprague-Dawley rats to induce striatal ischemia followed by immediate arterial administration of ~1 mil dissociated MSCi in 50-µL saline or saline only (control). Behavioral characterization was conducted concurrently with MR scanning to 21-d post-surgery. All data were acquired using the ultra-wide bore 21.1-T, 900-MHz vertical magnet at NHMFL with a linear birdcage double-tuned 23Na/1H radio frequency coil. Imaging was acquired 1, 3, 7 and 21 d post-ischemia to assess tissue recovery and overall treatment efficacy. Successful cell administration was confirmed with 50x50-µm in-plane resolution gradient recalled echo (GRE) images. 3D 23Na CSI was acquired at 1-mm isotropic resolution (TR = 60 ms) and zero-filled to 0.5-mm isotropic resolution in MatLab for image analysis in Amira 3D Visualization Software. RE-MRS2 evaluated metabolites in a 3-mm isotropic voxel in both ischemic and contralateral hemispheres. 4-kHz bandwidth excitation pulses were used to target lactate, creatine, choline and N-acetyl aspartate, while avoiding water. Spatial selectivity was achieved by an adiabatic selective refocusing (LASER)2 pulse sequence. T2W images acquired with a 1H Fast Spin Echo (FSE) sequence and 100x100-µm in-plane resolution enabled anatomical reference to the ischemic lesion.

Results and Discussion: MR assessments with respect to 23Na/1H lesion volume and signal as well as metabolite concentrations provide reliable indication of compromised donor cells from Donor 1, even when implanted at P5. Cell assays indicated normal activity at P5, but at P8, demonstrated increased population doubling time, immunomodulatory activity, SA-β-gal activity and overall senescence for Donor 1, which were absent in cell assays of Donor 2 with passage. Lesion volume as determined by 23Na signal is significantly reduced on day 3 for Donor 2 compared to saline and Donor 1 (Fig.1). Rather than the typical elevated lesion volume seen in uncontrolled stroke on day 3, Donor 2 resulted in an immediate decrease in stroke lesion following treatment as assessed via 23Na CSI. Additionally, percent change in both 23Na (Fig.1) and 1H (Fig.2) signal within the ischemic lesion is significantly reduced on day 3 for Donor 2 compared to Donor 1 and controls. RE-MRS supports MRI findings with reduced lactate levels on day 3 only for Donor 2, indicating energetic recovery.

Acknowledgements: All work has been conducted in accordance with the FSU Animal Care and Use Committee. Funding provided by NIH (RO1-NS102395). NHFML is supported by the National Science Foundation through the NSF (DMR-1644779) and State of Florida.