Multimodal MRI and PET Imaging to Study Rat Model of Traumatic Brain Injury

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Introduction The present pre-clinical multi-modal imaging integrates the emerging molecular imaging (PET) and MRI techniques to assist the traumatic brain injury (TBI) research using the rat model of TBI. We propose to investigate the post-injury metabolism changes in injured brains using the deoxyglucose (^{18}F) (FDG) micro PET imaging (μPET). MR imaging techniques include the T_1/T_2/T_2*/diffusion/perfusion MRI, susceptibility weighted imaging (SWI), and MR spectroscopy (MRS). These provide comprehensive information at the structural and functional levels that can help us investigate imaging markers of disease progression, white matter degeneration, vascular abnormalities, and micro-hemorrhages following TBI. PET and MR images are spatially co registered. Correlations between changes in glucose metabolism and MRI perfusion and diffusion in various brain regions and in lesion and perilesional regions in rats that undergo mild~moderate TBI are investigated.

Methods

Subjects and Injury Design: The study employs a controlled cortical impact (CCI, focal injury) TBI model. Rats in the CCI group were anesthetized with isoflurane and receive a moderate injury using the Leica CCI Impactor. Injury were induced using a deformation depth of 2-2.5mm for moderate injury, at an impact speed of 5m/s and a 4mm impactor tip, with a dwell time of 200msec. Injury were inflicted through a 5mm craniotomy located 3 mm caudal and 2.5 mm medial to Bregma.

Male Sprague Dawley rats (150 gram, 6 groups, imaging at different times after CCI, n=4 per group) will undergo a baseline PET and MRI session, and another cross sectional session at the same day and 1, 3, 7, 10, and 21 days post CCI for PET scans, and the next day for MRI scans, respectively, to wait for the FDG clearance. All experiments were approved by the IACUC of USUHS.

PET Protocol Rats were injected about 1.0-1.5 mCi of FDG via tail vein. They were constantly under anesthesia (isoflurane 2% and oxygen 2L/min) during the injection, uptake, and imaging sessions. Animals were maintained body temperature, and breath and heart rate were monitored to minimize the variability that may affect the FDG uptake. PET/CT images were acquired with Siemens Inveon Multimodality Micro PET/CT scanner (Siemens) with a 12 cm small bore, and ~1.5 mm spatial resolution. The FDG uptake is 45 minutes followed by a 30 minutes PET scan. A 10-minute CT scan is conducted before PET scan for attenuation correction.

MRI Protocol The MR measurements including T_1, multi-echo T_2, SWI, DWI, perfusion (ASL), and MR spectroscopy were performed on the Bruker BioSpec system (Bruker NMR, Inc., Billerica, MA) that consists of a 7-Tesla (T), 20-cm horizontal bore, and superconducting magnet (Magnex Scientific, Abingdon UK), and equipped with an 86mm quadrature transmit coil, and a dedicated phased array head coil. Anesthesia were delivered using isofluorane (2%) and oxygen/air mixture, the body temperature are maintained, and the respiration rate, heart rate, and blood O_2 level were monitored during MRI. The total MRI scan time is about 2 hours.

Multi spin echo images were acquired with TR=12s, and TE=20, 60, 100, 140ms, to localize the brain injury, and to measure T_2 in the tissue. Multi-echo SWI is based on the 3D multi-echo gradient echo method with TR = 30ms, TE = 3, 7, 11, 15, 19ms, and 400um slice thickness. The longer echoes represent T_2* weighted image. The 3D multi-echo data will be analyzed for quantitative susceptibility measurement. Cerebral blood flow (CBF) is an important physiological variable associated with the blood oxygen level dependent contrast and local brain metabolism. Continuous arterial spin labeling (CASL) [1] was used for measuring CBF non-invasively. CASL was implemented using a four-short EPI with TR=3s, TE=15ms, 3 slices positioned at the injury level, and 8 repetitions of the interleaved control and labeling acquisitions. DWI was implemented using an eight-shot EPI sequence with TR = 6250ms, TE = 26ms, three diffusion directions (x, y, z), and b = 100, 200, 400, 800, 1600 s/mm. All images are acquired with 100um^2 in-plane resolution. Lastly, localized MR spectroscopy was acquired using the single voxel PRESS method with TR = 2500ms and TE = 25ms. The voxel (3mm^3) was localized at the hippocampus region on the ipsilateral and contralateral side of the CCI injury.

Ex vivo MRI and *Histopathology* Animals were euthanized after the last *in vivo* MRI scan session and brains were harvested for ex vivo MRI and histological analysis.

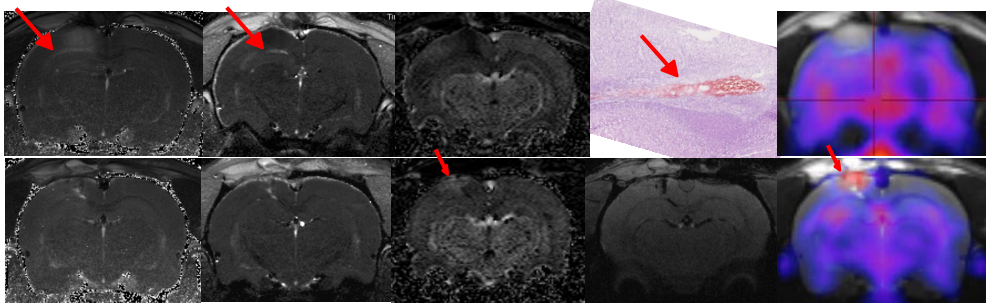
Image Analysis and Multimodal Data Integration SWI are generated using the magnitude and phase information. Multi-channel phase images were combined, and T_2, ADC, and CBF maps were generated using an in-house developed image analysis software. MRS spectra were processed using the XSOSNMR software (X. Mao & D. Shungu, Laboratory for Advanced MRS Research, Cornell University) for baseline correction and curve fitting to obtain peak areas of NAA (2.02 ppm), Cho (3.2 ppm), Cr (3. 02ppm), Glx (2.1~2.5ppm), and Lipid/Lac (1.33pm). Our current study primarily examined alterations in NAA and Cho, which include the estimations of the peak areas, and the ratios of NAA/Cr and Cho/Cr. Finally, PET and CT images, T_2, CBF, and ADC maps were re-sampled and co registered, and rat brain ROIs were segmented using the VivoQuant software (inviCRO, LLC, Boston MA). The brain regions were defined by the VivoQuant rat atlas. The rat atlas was non-linearly fit to the PET and MR images for each individual rat, for regional and quantitative PET/MRI data analysis. Injury area (lesion) were segmented manually, and the lesion ROI was also flipped and applied to the contralateral side of CCI for comparison. The FDG uptake values were normalized to the FDG uptake in the cerebellum.

Results CCI results in brain injury that are detectable by quantitative MRI and ^{18}FDG PET. The brain ROI analysis show changes of T_2, ADC, CBF and glucose metabolism in the lesion and perilesional regions at different phases post CCI (24 hours ~ 3 weeks). Figure 1 shows example of rats scanned at 1 day (MRI, top) and 4 days (MRI, bottom) post CCI. A bleeding region (red arrow) around corpus callosum is confirmed by the histology (H&E stain, Fig 1d, top). ROI analysis showed differences between the ipsilateral and contralateral regions of CCI. Decrease of ADC and CBF, and increase of glucose metabolism were found at the acute phases post CCI (Fig. 2), particularly in cortex, hypothalamus, olfactory, hippocampus, and corpus callosum regions. An inverse correlation is found between the ADC and glucose metabolism especially at the acute phases. Moderate CCI caused metabolic depression within the injured brain tissue (Fig 1e, top). On the other hand, glucose metabolism were found increase around lesion coupled with increase of CBF in the area (Fig. 1c&1e, bottom). In general, changes of glucose metabolism and CBF depend on the severity and phase of injury. Finally, reductions of NAA were detected in some of the CCI rats by MRS, indicating the neuronal damage due to the CCI.

Conclusions and discussions MRI and MicroPET are non-invasive systems that eliminate the need for biopsies and allow serial and longitudinal studies to be performed for TBI research and for monitoring the effects of interventions on disease progression and outcomes. We hope to identify neuroimaging techniques that can provide better assessment of structural and functional abnormalities and can be translatable to clinical TBI studies.

Reference **1.** Silva AC, Kim SG. Magn. Reson. Med. 42: 425-429, 1999.

Acknowledgement We would like to thank Xianling Mao and Dr. Dikoma Shungu, Weill Medical College of Cornell University, for their technical support and use of the XSOSNMR software package for MRS data analysis.



(a) T_2 map (b) ADC map (c) CBF map (d) Upper: H&E staining; Lower: SWI (e) PET overlay

Figure 1. Multi-modal MRI and PET images of CCI rats. From left to right: T2, ADC, CBF, histology/SWI, and PET

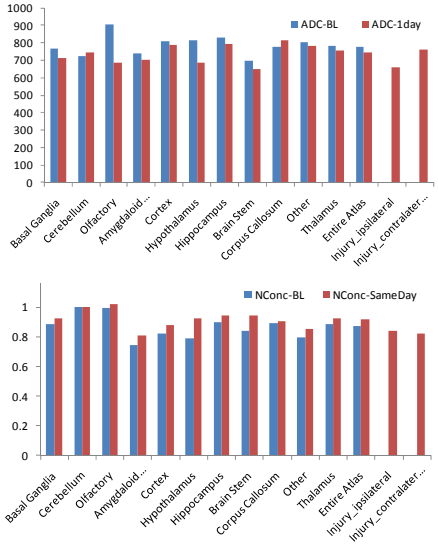


Figure 2. Regional ADC and normalized uptake