Cerebral Metabolite Assessment in low and high capacity running rats using 1H-MRS

S. R. Roys[1,2], A. Murugundla[3], S. Xu[1], A. Anderson[3], J. Zhuo[1], M. Limsam[3], J. Choi[3], L. Koch[4], S. Britton[4], K. Chandrasekaran[3], P. Yarowsky[5], J. Russell[3], and R. P. Gullapalli[1,2]

[1]Diagnostic Radiology & Nuclear Medicine, University of Maryland School of Medicine, Baltimore, MD, United States, [2]Core for Translational Research in Imaging @ Maryland (C-TRIM), [3]Neurology, University of Maryland School of Medicine, Baltimore, MD, United States, [4]Anesthesiology, University of Michigan, Ann Arbor, MI, [5]Experimental Therapeutics and Pharmacology, University of Maryland School of Medicine, Baltimore, MD, United States

Introduction

There has been a recent interest in understanding the CNS complications due to diabetes mellitus both clinically and in animal models.[1] Very little is understood regarding CNS changes that lead to various cognitive impairments among diabetics. Even less is understood regarding the differences between high performing diabetic patients versus the low performing diabetics. The purpose of this study was to examine the neurochemical profile differences between low capacity runner rats (LCR) and high capacity runner rats (HCR) using proton magnetic resonance spectroscopy at 7.0 Tesla. LCR rats manifest metabolic risk factors resembling human impaired glucose tolerance (IGT) and are a polygenic rat developed based on their reduced running capacity compared to HCR rats.[2]

Materials and Methods

LCR and HCR Models

There were a total of 9 aged animals (>21 months) in each of the groups. Animals were fed regular chow and maintained with a regular sleep/wake cycle. The experimental protocol was approved by the Committee for the Welfare of Laboratory Animals of the University of Maryland.

In Vivo ^1H MRS

All experiments were performed on a Bruker Biospec 7.0 Tesla 30 cm horizontal bore scanner using Paravision 5.0 software. A Bruker ^1H surface coil array was used as the receiver and a Bruker 72 mm linear-volume coil as the transmitter. Proton density-weighted MR images were taken using a 2D rapid acquisition with relaxation enhancement (RARE) sequence (TR/TE=5500/9.5 ms) for anatomic reference. A point-resolved spectroscopy (PRESS) pulse sequence (TR/TE=2500/20 ms) was used for data acquisition from a 3 x 3 x 3 mm^3 voxel centered around the hippocampus, and superior thalamic structures as shown in Figure 1 (inset). For each spectrum, 300 acquisitions were averaged for a total of 13 min. At all times during the experiment, the animal was under 1-2% isoflurane anesthesia and 1 L/min oxygen administration. Respiratory monitoring was performed and the animal was maintained at 36-37 ^oC during the entire experiment.

Proton MRS data was fitted using the LC Model package using only metabolites with standard deviations (SD) % < 20 were included for further analysis. The ratios of *in vivo* mean metabolite concentrations of γ-aminobutyric acid (GABA), Glucose (Glc), Glutamine (Gln), glutamate (Glu), myo-inositol (Ins), glycerophosphorylcholine (GPC) and phosphorylcholine (PCh)., N-acetylaspartate (NAA) and Taurine (Tau) relative to total creatine (tCr) were computed for each voxel and a paired one-tail Student t-test was performed to compare the difference between the neurochemical profiles of the LCR and the HCR rats.

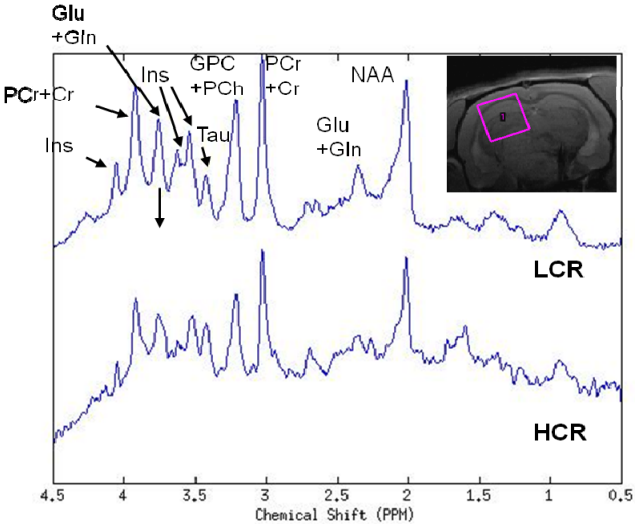

Figure 1. Example spectra from a low capacity rat (top) and a high capacity rat (bottom) obtained at the level of the hippocampus (inset)

MET/Cr+PCr	LCR	HCR	P
GABA	0.69±0.18	0.72±0.22	0.34
Glc	0.66±0.21	0.78±0.27	0.13
Gln	0.81±0.27	0.93±0.19	0.08
Glu	1.01±0.12	0.87±0.16	**0.0057**
Ins	1.17±0.14	1.02±0.14	**0.0012**
NAA	0.747±0.098	0.76±0.085	0.302
Tau	0.7±0.098	0.52±0.07	**9.22E-06**
GPc+PCh	0.242±0.024	0.22±0.026	**0.0074**
NAA+NAAG	0.92±0.12	0.95±0.13	0.24
Glu+Gln	1.82±0.27	1.79±0.29	0.32

Table 1. Neurochemical profile of Low capacity and High capacity diabetic rats

Results & Discussion: Figure 1 shows example spectrum obtained from a LCR and an HCR rats. Table 1 provides a summary of the neurochemcial profile of the LCR and the HCR rats along with the differences between the two cohorts. Significant increases ($p<0.05$) in glutamate, taurine, inositol and choline containing compounds were observed in the LCR rats compared to the HCR rats. A decreasing trend was observed in glutamine ($p<0.08$) for the LCR rats compared to the HCR rats. No significant elevation of glucose was observed in the LCR rats compared to the HCR rats indicating no difference in glucose uptake. As an excitatory neurotransmitter, glutamate has a functional role in synaptic plasticity and learning. An elevation of glutamate has been observed in neurodegenerative disease such as Alzheimer's.[3] The excess glutamate among the LCR rats may also be a result of impaired glial function. The increases in myo-inositol may be related to accumulation of the osmolyte associated with diabetes and increased choline containing compounds may be consistent with the increased vascular/ischemic basis to gliosis among the LCR rats.[4] Taurine has antioxidant properties and is known to act as an osmoregulator and regulator of mitochondrial function. The elevation of taurine among the LCR rats indicates acceleration of glucose uptake and glycogen synthesis in the liver providing an insulin-like effect.

Conclusion: Our results indicate significant differences between the cerebral metabolic profile in aged LCR compared to HCR rats. Although there is no significant difference in glucose uptake, the LCR rat has elevated levels of taurine. Myo-inositol and choline containing compounds were elevated consistent with diabetes, gliosis and membrane turnover. The elevation of glutamate may indicate a neurodegenerative process in the LCR rats. Further work is necessary to understand the effect of the long-term elevation of these metabolites in the LCR rats.

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