Serial Proton MRS Evaluation of Neurometabolism in a Mouse Model of Systemic Lupus Erythematosus (SLE)

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INTRODUCTION

The involvement of the central nervous system (CNS) in systemic lupus erythematosus (SLE) is a common manifestation that affects 14–75% of patients depending on both the population studied and the methodology of assessment. Owing to the current limitation in knowledge of underlying pathogenetic mechanisms, the lack of effective imaging, the paucity of histopathologic findings and the poor correlation between instrumental data and clinical manifestations, neuropsychiatric SLE (NPSLE) remains a major unsolved diagnostic problem. Although a wide range of neuroimaging tools have been used to evaluate CNS complaints, no single technique has proven to be definitive in NPSLE. This impedes both clinical diagnosis and drug discovery for the treatment of this condition [1, 2]. *In vivo* localized magnetic resonance spectroscopy (MRS) provides a noninvasive method to investigate neurochemical metabolites and can provide insight into unique pathological processes at the molecular or cellular level [3-5]. The specific aim of the current study was to detect and determine the neurochemical changes associated with the NPSLE condition, by comparing quantitative MRS measurements in lupus MRL/*lpr* mice during disease progression with control (B6) mice.

MATERIALS AND METHODS

Experiments were conducted on 5 male B6 control mice and 5 male MRL/*lpr* lupus mice. The study was performed, in 3 intervals, over an 8 week period, started when the animals were 7 weeks in age. All MR experiments were performed on a 7T small animal MRI system (Bruker BioSpin, Ettlingen, Germany), equipped with an actively-shielded gradient set, with a maximum gradient strength of 400 mT/m. Signal excitation and reception were accomplished with a 25 mm Litz RF coil (Doty Scientific, Columbia, SC). Conventional T_2-weighted RARE images (TE = 60 ms, TR = 4000 ms, slice thickness = 1 mm, matrix = 256 x 192) were acquired in coronal (FOV = 2.5 x 2.5 cm^2) and axial (FOV = 2.2 x 2.2 cm^2) orientations to assess differences in anatomy and to assist in positioning of the MRS voxels. Spectra were obtained with a double spin-echo (PRESS) sequence (TE = 20 ms, TR = 2500 ms) with and without VAPOR water suppressions. For each individual water-suppressed spectrum, data were acquired with a spectral width of 4 KHz, and 192 averages. Localized shimming was achieved by using the FASTMAP technique [6]. Identical voxels (3 x 3 x 3 mm^3) were prescribed in a central location of the forebrain in the left hemisphere, below the cortex, to include tissue from both the striatum and hippocampus (figure 1). The individual spectra were processed and quantified using LCModel [7].

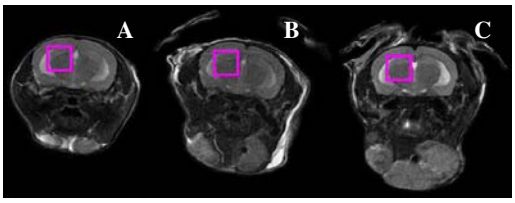

Figure 1: MRS voxel location in a lupus (MRL/*lpr*) mouse, at 7 weeks (A), 11 weeks (B) and 15 weeks (C)

RESULTS

Satisfactory spectra were acquired, at each of the three time points, from both sets of mice. In the 10 mouse brains, neither visible lesions nor any obvious changes of image intensities were seen in the T_2-weighted images, at any of the three time points. Before each acquisition of water spectra as the reference scan, each localized voxel was shimmed until the water peak had a linewidth (full width at half maximum) of <10 Hz. All examined metabolites were reliably detected, with mean %SDs (Cramer-Rao lower bounds) less than the previously determined reliability cutoff of 20% [7], and were compared for the two groups to investigate metabolic differences present in the brain tissues. Using LCModel, each detectable metabolite's concentration was quantified as the ratio to total Creatine (tCr for Creatine + Phosphocreatine) concentration, in order to reduce systematic variations. Table 1 lists the results of quantified metabolite peak ratios to tCr (proportional to their concentrations) for Choline-containing compounds (tCho), N-Acetyl Aspartate (NAA), Glutamate + Glutamine (Glx), *myo*-Inositol (mI) and Taurine (Tau). No significant changes were seen in the ratios of tCho/tCr, for any of the three time points, between the control and lupus mice. The ratios of NAA/tCr were significantly higher in the lupus mice at both the 7 week ($P < 0.01$) and 15 week ($P < 0.05$) evaluation periods. A significant decrease was seen in the Glx/tCr ratios of the lupus mice for both the 11 week ($P < 0.05$) and 15 week ($P < 0.05$) time points. The ratio of mI/tCr was significantly higher in the lupus mice at only the 11 week ($P < 0.005$) evaluation period. The ratio of Tau/tCr was significantly higher in the lupus mice at both 7 weeks ($P < 0.005$) and 15 weeks ($P < 0.01$).

Metabolite Ratios	B6 (Control) (mean ± SD)			MRL/*lpr* (Lupus) (mean ± SD)		
	7 (weeks)	**11 (weeks)**	**15 (weeks)**	**7 (weeks)**	**11 (weeks)**	**15 (weeks)**
tCho / tCr	0.28 ± 0.06	0.27 ± 0.03	0.27 ± 0.03	0.26 ± 0.05	0.29 ± 0.03	0.27 ± 0.04
NAA / tCr	0.82 ± 0.06	0.83 ± 0.09	0.82 ± 0.1	0.92 ± 0.05	0.90 ± 0.12	0.94 ± 0.09
Glx / tCr	1.74 ± 0.1	1.74 ± 0.08	1.74 ± 0.26	1.59 ± 0.29	1.49 ± 0.25	1.41 ± 0.28
mI / tCr	0.68 ± 0.05	0.66 ± 0.08	0.69 ± 0.13	0.65 ± 0.09	0.84 ± 0.07	0.84 ± 0.05
Tau / tCr	0.70 ± 0.12	0.77 ± 0.09	0.64 ± 0.11	0.89 ± 0.03	0.89 ± 0.18	0.86 ± 0.09

Table 1: Summary of metabolite concentration ratio measurements

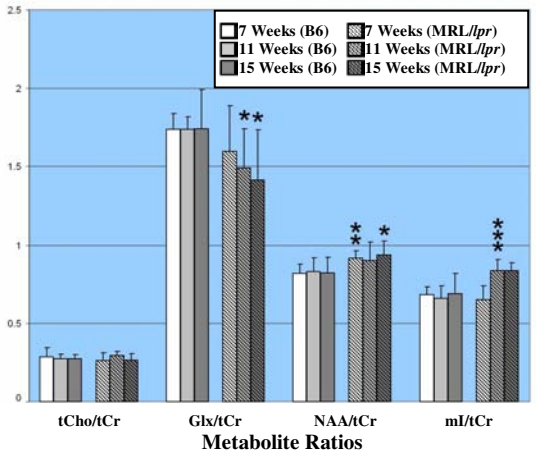

Figure 2: MRS metabolite ratio quantitation of control and lupus mice (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.005$)

DISCUSSION

The statistical correlation between the metabolite concentration ratios shown in Figure 2 indicate that Glx, mI and NAA signals all may serve as sensitive metabolic markers related to CNS involvement in lupus progression. These results suggest that the measured neuron-metabolites via MRS could provide useful neurochemical information into both the stage and severity of the condition. Among these, the most interesting finding is the age-dependent decreases of the known neurotransmitter Glx, along with the increases seen in NAA, which is commonly thought of as a reliable marker of neuronal viability. These findings are currently being correlated with behavioral studies to provide a better understanding of the mechanism of the neuropsychiatric symptoms in lupus. This information could lead to more precise methods of clinical diagnosis, and eventually, advances in drug discovery.

REFERENCES

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