Metabolic Changes in Hippocampus and Thalamus after Sleep Deprivation: An Experimental Proton MRS Study

Iris Y. Zhou[1,2], Abby Y. Ding[1,2], Frank Y. Lee[1,2], and Ed X. Wu[1,2]

[1]*Laboratory of Biomedical Imaging and Signal Processing, The University of Hong Kong, Hong Kong SAR, China, People's Republic of,* [2]*Department of Electrical and Electronic Engineering, The University of Hong Kong, Hong Kong SAR, China, People's Republic of*

INTRODUCTION

Sleep plays a key role in facilitating learning and memory consolidation [1, 2]. Insufficient sleep, with a high prevalence in society today, affects alertness and neurocognitive process in a negative manner [3]. Several studies have shown that deprivation of sleep can result in memory deficits and impaired cognitive performance in humans [4, 5]. The neurobiological alterations underlying these behavioral deficits, especially in regions related to learning and memory such as hippocampus and in the structures involved in alertness and attention such as thalamus, are of interest. Proton MRS (^{1}H MRS) can be used to assess the metabolic changes in living brain [6] and thus could provide biochemical evidence underlying the neural process, such as psychiatric disorders [7]. In this study, we aim to use in vivo ^{1}H MRS to investigate the metabolic changes induced by sleep deprivation (SD) in hippocampus and thalamus.

MATERIALS AND METHODS

Animal Preparation: Male Sprague-Dawley rats (~8 weeks old, N=9) were subjected to rapid eye movement (REM) sleep deprivation (SD) and MR scanned before and after the treatment. ***Sleep deprivation*** [8]: In the multiple small-platform technique employed, five platforms with each 6cm diameter were placed in the middle of a water tank. Platforms were spaced 9cm apart so that rats could easily move between them but could not lie across any two. The water reached up to ~2 cm below the surface of the platform. Food and water were available *ad libitum*. All SD treatments lasted 72 hr under a 12h day/night cycle. Video monitoring was performed throughout the training and was used for later sleep deprivation validation. ***MRI Protocols:*** All MR measurements were performed on a 7 T Bruker MRI scanner using a quadrature surface coil. Under inhaled isoflurane anaesthesia, the animal was kept warm under circulating water at 37 ºC with respiratory monitoring. RARE T2-weighted anatomical images were acquired for voxel localization in ^{1}H MRS. After localized shimming with FieldMap, ^{1}H MRS was performed using a PRESS sequence combined with outer volume suppression (OVS) and with TR/TE=2500/20ms, 2048 data points and 256 averages. A 2×4×2 mm^3 voxel was placed over the left hippocampus and another 3×3×3 mm^3 voxel was centered at the left thalamus. ***Data Analysis:*** MR spectra were processed with jMRUIv4.0 software using simulated metabolites in NMR-SCOPE as prior knowledge. The raw data was apodized with a 15-Hz Gaussian filter and phase-corrected. The residual water signal was filtered out with HLSVD algorithm. Various ratios of metabolites, NAA:Cr, Cho:Cr, Glu:Cr, Lac:Cr, m-Ins:Cr and Tau:Cr were statistically evaluated using two-tailed paired student's t-tests between before and after SD treatment with $p < 0.05$ considered as significant.

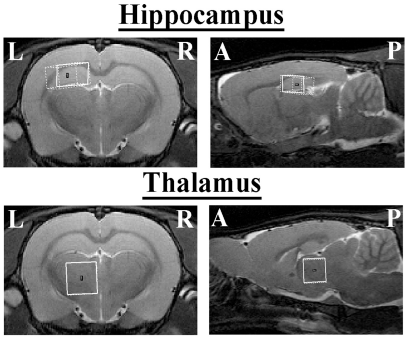

Fig.1 Localization of the voxels for ^{1}H MRS measurement in hippocampus and thalamus.

RESULTS

Fig. 1 illustrates the typical localization of the voxels for ^{1}H MRS measurement of rat hippocampus and thalamus. For each region of interest, ^{1}H MRS spectra before and after SD were averaged from all rats and shown in Fig.2. Reduction of N-acetylaspartate (NAA) level can be clearly observed in the hippocampal spectrum after SD compared to that of before SD. The statistical evaluation (Fig. 3) of the metabolite signal with respect to creatine (Cr) peak revealed that, besides the distinct higher hippocampal NAA level ($p<0.001$), glutamate (Glu) signal was also significantly lower ($p<0.01$) in hippocampus after SD. Meanwhile, Glu:Cr significantly increased ($p<0.05$) in thalamus after SD. Lactate (Lac) level decreased ($p<0.05$) in thalamus after SD.

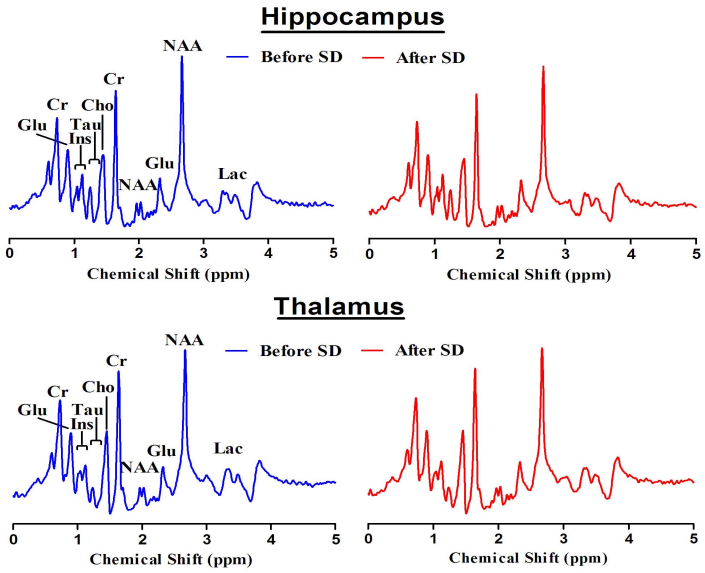

Fig.2 Averaged ^{1}H MRS spectra of all rats (N=9) before and after sleep deprivation (SD) in hippocampus and thalamus.

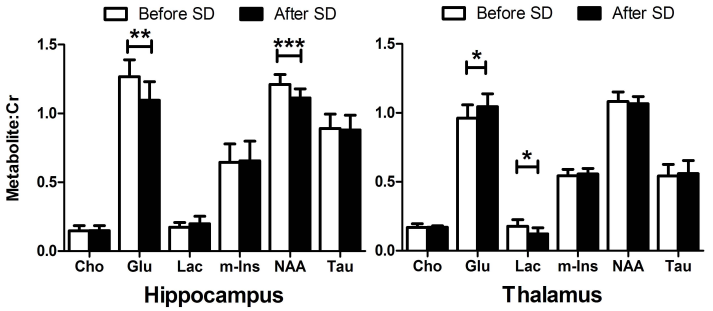

Fig.3 Comparisons of metabolite ratios before and after exposure to sleep deprivation (SD) in hippocampus and thalamus. Paired t-tests were performed with * $p<0.05$, ** $p<0.01$, *** $p<0.001$.

DISCUSSION AND CONCLUSION

Reduction of NAA, a marker of neuronal density, integrity and health [6], indicates neuronal loss and cellular dysfunction [9]. Previous histological study showed that the cell proliferation in the dentate gyrus of hippocampus was suppressed by prolonged (72hr) SD. Therefore, the reduced hippocampal NAA:Cr after SD can be due to neuronal loss. Moreover, using the same SD paradigm, neurophysiological study found severely reduced neuronal excitability in CA1 area of hippocampus [8]. It indicates that, along with neuronal loss, the NAA decrease observed after SD could also result from neuron cell dysfunction. Furthermore, the reduced cellular excitability in hippocampus can be reflected by the reduction of Glu, an amino acid acting as excitatory neurotransmitter in the brain, observed in this study. In thalamus, glucose metabolism decreases after SD [10]. Therefore, Lac, an end product of anaerobic glycolysis, was found to decrease in thalamus after SD. Our finding of Glu:Cr increase in thalamus is in line with previous ex vivo measurement in cats [11], possibly arising from increased glutamine synthetase [12]. In conclusion, the metabolic alterations elicited by sleep deprivation in rat brain are documented in this study using in vivo ^{1}H MRS, providing neurochemical evidence of the behavioral deficits associated with sleep deprivation.

REFERENCES

[1]Maquet P. Science 2001;294:1048-52. [2]Siegel J. M. Science 2001;294:1058-63. [3]Durmer J. S. and D. F. Dinges Semin Neurol 2005;25:117-29. [4]Chee M. W. and Y. M. Chuah Proc Natl Acad Sci U S A 2007;104:9487-92. [5]Goel N., Semin Neurol 2009;29:320-39. [6]Choi J. K., NMR Biomed 2007;20:216-37. [7]Lyoo I. K. and P. F. Renshaw Biol Psychiatry 2002;51:195-207. [8]McDermott C. M., J Neurosci 2003;23:9687-95. [9]Siegmund A., Biol Psychiatry 2009;65:258-62. [10]Thomas M., J Sleep Res 2000;9:335-52. [11]Micic D., Nature 1967;215:169-70. [12]Sallanon-Moulin M., Brain Res Mol Brain Res 1994;22:113-20.