Effects of desipramine pretreatment on behavioral and regional neurochemical responses in the mouse forced swimming test: a high resolution in vivo 1H-MRS study at 9.4 Tesla

S-Y. Kim1,2, C-B. Choi1, Y-J. Lee1, H. Kim3, D-W. Lee1, D-C. Woo1, J-H. Chae6, and B-Y. Choe1,2

1Department of Biomedical Engineering, The Catholic University of Korea, Seoul, Korea, Republic of, 2Research Institute of Biomedical Engineering, Seoul, Korea, Republic of, 3Department of Radiology, Kyunghee University Medical Center, Seoul, Korea, Republic of, 4Gachon University of Medicine and Science, Incheon, Korea, Republic of, 5Department of Psychiatry, The Catholic University of Korea, Seoul, Korea, Republic of

INTRODUCTION
Although numerous attempts have been made to create animal models of depression or at least of some pathophysiological aspects, only a few models have been adequately validated. The mouse forced swimming test (FST) is a useful paradigm that is relatively quick and simple to perform. The test is based on the observation that mice, after an initial struggling phase, develop an immobile posture when immersed in cold water in a cylinder without the possibility of escape. Immobility in the FST was originally considered a model of depression [1]. However, no data are currently available about the behavioral and simultaneous non-invasive measurements of neurochemical responses following antidepressant treatment in mice FST model. In the present study, in vivo 1H-MRS at 9.4 T was used to examine the effects of desipramine (DMI) pretreatment on behavioral and regional neurochemical responses of C57BL/6 mice.

MATERIALS AND METHODS
Animal/Animals were randomly assigned into three groups (control+saline: N=10, saline+FST: N=10, DMI+FST: N=10). The mouse FST as shown Fig.1 was essentially similar to that previously described [1]. The DMI (10 mg/kg) was dissolved in distilled water and was injected intraperitoneally (IP) into mice 45 min prior to the FST in a constant volume of 0.5 ml/20 g body weight. To measure active behavior (i.e., climbing and/or swimming) and immobility, a time-sampling technique [2] was used during the last 4 min of the 6 min testing period.

In vivo 1H-MRS Acquisitions and Post-processing
All experiments were conducted on a 9.4 T/20 cm horizontal bore magnet (Bruker BioSpec 94/20, USA, Ettlingen, Germany). For in vivo localized spectroscopy, we used an ultra short echo-time STEAM pulse sequence (TR/TE/TM = 5000/20/2.2 ms; number of data points = 2048; NEX = 384; scan time = 30 min). Postprocessing was carried out semi-automatically using Bruker Topspin 2.0 software. The following procedures were included: (a) Exponential line broadening (3 Hz), (b) zero/first-order phase correction and (c) baseline correction

Absolute Quantitation
The metabolite concentrations were determined relative to Cr+PCr as an internal reference. The concentration of Cr+PCr was assumed to be 6 mM, as similar to the rat brain. The following formula was used: \[ C_{\text{met}} = \frac{A_{\text{met}}}{A_{\text{Cr+PCr}}} \times C_{\text{Cr+PCr}} \] where \( C \) is the concentration, \( A \) is the peak area, and \( N_p \) is the number of protons contributing to the resonance of metabolite \( i \) (\( i = \text{NAA} (N_p = 3), \text{Glx} (N_p = 2), \text{tCho} (N_p = 9), \text{Tau} (N_p = 2), \text{mIns} (N_p = 4) \)).

RESULTS
The administration of DMI to mice 45 min prior to the FST (DMI + FST group) lead to a significant decrease in the immobility mean count (20.4%) as compared to the saline + FST group (\( t = 2.79; \text{df} = 18; P = 0.012 \)) (Fig. 2). The multislice RARE sequence provided high-quality images of the mouse brain with a spatial resolution and contrast that guaranteed precise and reproducible placement of the VOI in the left DLPCF and hippocampus (Fig. 3). In vivo metabolite concentrations obtained from the left DLPCF and left hippocampus of the mice are depicted in Fig. 4. The concentrations of myo-inositol (mlns) and glutamate (Glu) + glutamine (Gln) in the left DLPCF were significantly changed among the three groups (control+saline vs. saline+FST, saline+FST vs. DMI+FST).

DISCUSSION AND CONCLUSION
The present study has demonstrated that the mouse FST induced the reduction in Glx concentrations and elevation in the mlns concentrations in the left DLPCF of the C57BL/6 mouse brain as compared to controls, but not in the left hippocampus. In FST + DMI group, the neurochemical perturbations and behavioral responses reverted to similar levels observed in controls. Our results suggest that glutamatergic activity and glial cell dysfunction contribute to the pathophysiological mechanisms underlying depression and that modulation of synaptic neurotransmitter concentrations represent invaluable targets for antidepressant drug development.

ACKNOWLEDGEMENT
This study was supported by the Korea Health 21 R and D Project, Ministry of Health and Welfare, Republic of Korea (2002-P33-PG6-EV/07-0002) (A081057) and a grant (R01-2007-000-20782-0) from the Purpose Basic Research Grant of the KOSEF and the Korea Research Foundation Grant funded by the Korean Government (KRF-2008-313-D01324 and MEST-2009-0074472) and the program of Basic Atomic Energy Research Institute (BAERI).

REFERENCES