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

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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 neurochemial responses following antidepressant treatment in mice FST model. In the present study, *in vivo* ^{1}H-MRS at 9.4 T was used to examine the effects of desipramine (DMI) pretreatment on behavioral and regional neurocheimal responses of C57BL/6 mice.

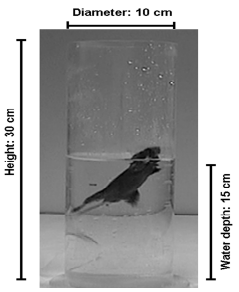

Fig.1. A mouse shows active behavior in an inescapable cylinder during FST procedure.

MATERIALS AND METHODS

Animal A total of thirty C57BL/6 mice were randomly assigned into three groups (contol+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 intraperitonelly (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 ^{1}H-MRS Acquisitions and Post-processing All experiments were conducted on a 9.4 T/20 cm horizontal bore magnet (Bruker BioSpec 94/20, USR, Ettlingen, Germany). For *in vivo* localized spectroscopy, we used an ultra short echo-time STEAM pulse sequence (TR/TM/TE = 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. $[C_i=(A_i/A_{Cr+PCr})*(N_{p,Cr+PCr}/N_{p,i})*C_{Cr+PCr}]$ 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* = NAA (N_p = 3), Glx (N_p = 2), tCho (N_p = 9), Tau (N_p = 2), 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; d.f. = 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 DLPFC and hippocampus (Fig. 3). *In vivo* metabolite concentrations obtained from the left DLPFC and left hippocampus of the mice are depicted in Fig. 4. The concentrations of myo-inositl (mIns) and glutamate (Glu) + glutamine (Gln) in the left DLPFC were significantly changed among the three groups (control+saline vs. saline+FST, saline+FST vs. DMI+FST).

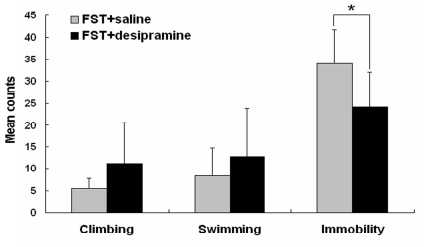

Fig.2. The effects of acute DMI treatment on behavioral responses of C57BL/6 mice exposed to the FST

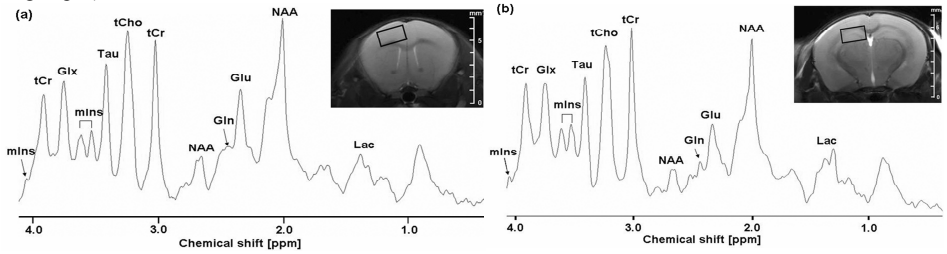

Fig.3. Multislice T_2-weighted axial images of the C57BL/6 mouse brain with the VOI centered in the (a) left DLPFC (1.35 × 2.1 × 2.5 mm^3) and (b) hippocampus (1.3 × 2.0 × 2.5 mm^3) and *in vivo* ^{1}H NMR spectra acquired with an ultra short echo STEAM pulse sequence from the corresponding brain regions are shown.

DISCUSSION AND CONCLUSION

The present study has demonstrated that the mouse FST induced the reduction in Glx concentrations and elevation in the mIns concentrations in the left DLPFC 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.

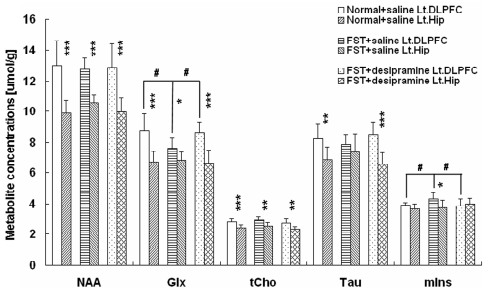

Fig.4. Absolute metabolite concentrations quantified in the left DLPFC and hippocampus of the three groups. Metabolic differences among the groups and regional variations between the left DLPFC and hippocampus are indicated by a crosshatch (#) and asterisk (*), respectively.

ACKNOWLEDGEMENT

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