

Manganese-Enhanced MRI and ¹H MR Spectroscopy of the hypothalamus in a model of dehydration induced anorexic rats

N. Just^{1,2}, and R. Gruetter^{1,2}

¹LIFMET, CIBM, EPFL, Lausanne, Switzerland, ²Department of Radiology, UNIL and HUG, Lausanne and Geneva, Switzerland

Introduction: Neurons of the hypothalamic paraventricular nucleus (PVN) regulate the hypothalamic pituitary-thyroid axis (HPT) and are involved in the control of food intake. The model of dehydration-induced anorexia (DIA) (1) leads to decreased voluntary food intake. The aim of the present work was to investigate the possibility of measuring the hypothalamic responses of female rats submitted to DIA and of normally fed rats using Manganese-Enhanced MRI (MEMRI) and proton MR spectroscopy at 14.1T.

Materials and methods: 5 female SD rats weighing 220-225g on day 0 (D0) were used. 3 rats received a 2.5% NaCl (SIGMA, USA)(1) solution as drinking liquid and food ad libitum (DIA group) during 7 days. The 2 remaining rats received water and food ad libitum (N: Control group). A 30 mg/kg dose of 100mM MnCl₂ solution prepared according to(2) was infused in the tail vein of 2 DIA rats and 1 N rat 3 days (D3) after submitting the DIA rats to the dehydration regime. The last DIA and control rats were infused on D6. During the MR experiments, rats were anesthetized with 2% isoflurane. Their body temperature was maintained at 37.5±0.5°C using a temperature-controlled water circulation and a rectal probe. Their respiration rate was also monitored throughout the experiment. Imaging and spectroscopy were performed on D4 and D7 24 hours after MnCl₂ infusion. The rats were positioned in a dedicated holder with head fixation. All the experiments were performed on a 14.1T/26cm horizontal bore magnet (Magnex, Varian). Field homogeneities were adjusted using FASTMAP(3). A quadrature T/R 17-mm surface coil was used. For MEMRI, a 3D gradient echo sequence (TR/TE=20/5ms, Flip angle=70°, FOV=25x25x25mm, matrix size=256x256x128, axial slices, BW=25KHz) was used. Localized Proton spectroscopy was performed using SPECIAL(4) in a 50μl VOI localized in the same hypothalamic region for all the rats and metabolite concentrations were calculated using LCmodel(5).

Results: The rats under DIA lost between 9% and 15% of their initial weight over seven days whereas the normally fed rats gained 13% of their initial body weight during the same period. We also noticed an increased NaCl-loaded water intake for the DIA rats as reported before (6). Figure1 shows 3D gradient echo images in 2 control rats: one without MnCl₂ (1A) and one 24h after MnCl₂ infusion (1B) and in a DIA rat (1C). All were imaged on D3. The signal to noise ratio was calculated for each rat for the entire brain slice and ROIs in the lateral hypothalamus and in the pituitary gland. SNR further increased in the DIA rats (SNR~45 in the lateral hypothalamus) compared to the control rats (SNR~28). The arrow in the DIA rat (fig2B) indicates enhancement of the paraventricular hypothalamic nuclei next to the 3rd ventricle. No enhancement was seen in the control group (Fig2A). The results of proton MR spectroscopy are reported in fig.3 where mean metabolite concentrations were calculated in 2 controls and 2 DIA rats at D7. Increases were detected for Asc, Pcr, Ins, Glu and Gln in DIA rats compared to control rats at D7.

Conclusion: The stimulus of cellular dehydration like the taste of salt induced a response of the HPT axis detectable at 14.1T using MEMRI and proton MRS.

References:(1) Watts AG et al. J Neurosci,19,6111-6121; (2):Silva AC et al NMR in Biomed, 2004,17,532-43 (3):Gruetter R et al. MRM,29 :804,1993; (4):Mlynarik V et al. MRM,56 :965,2006 ;(5) Provencher SW.MRM,30 :672,1993 ;(6)Jaimes-Hoy L et al. Hormones&Behavior,53 :366,2008

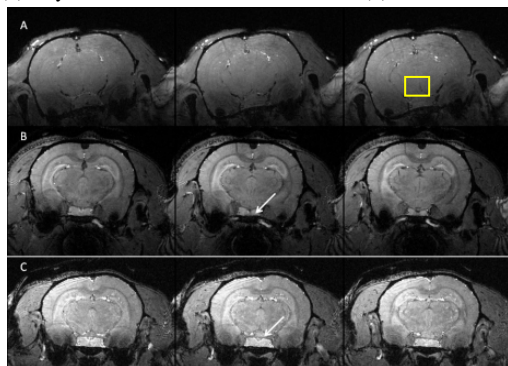


Figure1: Contrast comparison in 3 rats at D3:A. Control rat with no MnCl₂. The VOI for ¹H MRS is shown. B: Control rat 24h after MnCl₂ infusion. C DIA rat 24h after MnCl₂ infusion. Increased contrast can be seen in the pituitary gland (arrows)

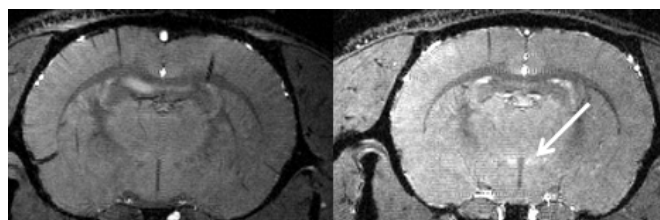


Fig2: A. Control rat and B. DIA rat on D3 after MnCl₂ infusion. The arrow shows enhancement in the paraventricular hypothalamic nucleus (PaAM,PaAP) in the DIA rat but not in the control rat

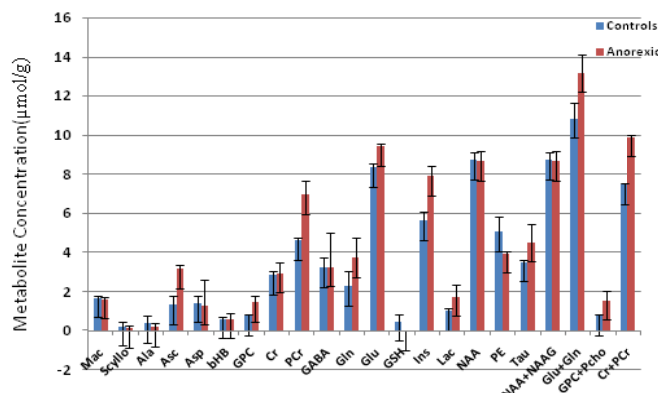


Figure3. Metabolite Concentrations in 2 controls and 2 DIA rats at D7.

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