Phosphocreatine and Neurotransmitters Changes in the Newborn Rat during Acute Hypoglycemia measured by *in vivo* ^{1}H NMR Spectroscopy

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INTRODUCTION

The developing hippocampus is particularly vulnerable to hypoxia-ischemia, perinatal iron deficiency, and acute hypoglycemia (1-4). Perturbations in the normal development of hippocampus may result in cognitive abnormalities in infants. The purpose of this study was to determine the neurochemical changes in the developing rat hippocampus during acute hypoglycemia using *in vivo* ^{1}H NMR spectroscopy.

METHODS

All NMR measurements were performed at 9.4 T (Varian/Magnex). All 1st- and 2nd-order shims were adjusted by FASTMAP (5). STEAM (TE = 2 ms) combined with OVS and VAPOR water suppression was used for localization (6). The positions of VOIs were determined using multi-slice RARE imaging. Metabolite concentrations were quantified using LCModel with macromole-cule spectra included in the basis set (7). The unsuppressed water signal was used as an internal reference. Sprague-Dawley rat pups (14 day old, n = 8) were anesthetized (N_2O : O_2 = 1 : 1 with 0.5 - 1.5% isoflurane) and different levels of hypoglycemia were induced by i.p. injection of an insulin dose (2 – 6 IU/kg). Plasma glucose was measured immediately before and after the NMR study.

RESULTS AND DISCUSSION

The high spectral resolution (Fig. 1) allowed reliable quantification of 14 brain metabolites (Cramer-Rao lower bound < 10% for Cr, PCr, Glu, NAA, PE, Tau, GPC + PCho, CRLB < 20% for Asp, GABA, Gln, GSH, Ins, Lac, and NAAG) in each measured spectrum. Concentrations of most of the metabolites were independent plasma glucose (e.g., NAA in Fig. 2). However, concentrations of Glu, Cr, and PCr were highly dependent on concentration of the plasma glucose (Fig. 2). In addition, aspartate had a tendency to increase (R = -0.5, p < 0.05) with decreasing plasma Glc. The sum of Asp and Glu concentrations was constant within the experimental error. Increased blood flow during hypoglycemia resulted in reduced line width of 8 – 9 Hz (8) that allowed the reliable quantification of both Cr and PCr directly by ^{1}H NMR (CRLB ≤ 7%). The decreased [PCr]/[Cr] ratio at low Glc levels (< 2 mM) was in agreement with the literature (3-4), indicating insufficient energy production to maintain the basic metabolism. The consumption of Glu was most likely due to transamination reaction, which was consistent the total concentration of Asp and Glu independent of the plasma Glu levels, except for the lowest values (< 1 mM). Interestingly, brain lactate was not affected by lowered plasma Glc, even though Lac consumption presumably increased, which implied increased brain glycogenolysis.

In conclusion, high field *in vivo* ^{1}H NMR spectroscopy can provide detailed and spatially selective information about changes in the neurochemical profile, particularly about the energy metabolism, during hypoglycemia in a developing rat brain.

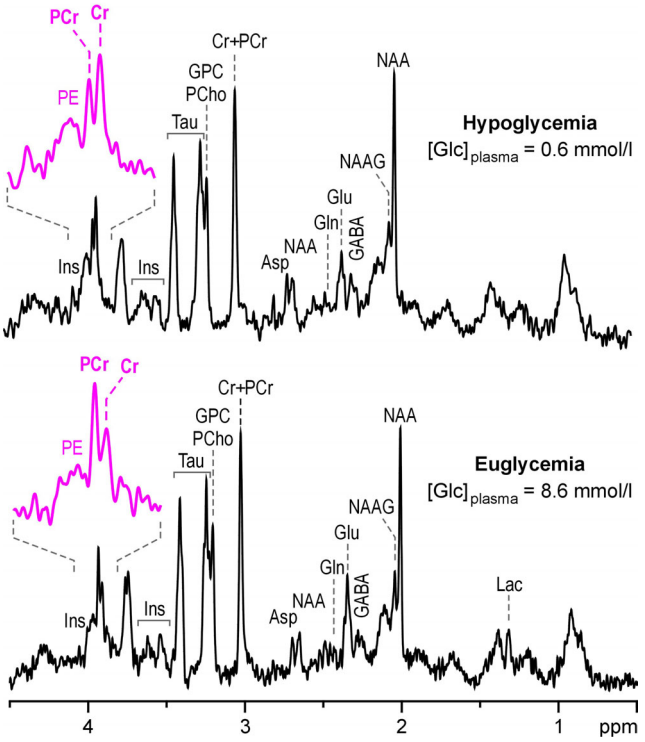

Fig. 1 *In vivo* ^{1}H NMR spectra from the hippocampus of 14-day old rat during euglycemia and hypoglycemia. STEAM, TE = 2 ms, TR = 5s, VOI = 10 µl, NT = 160.

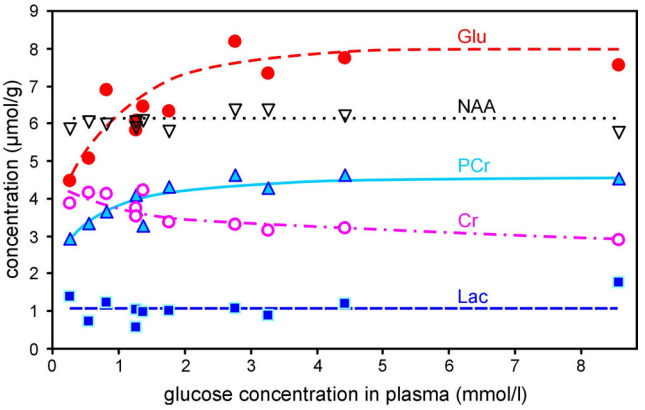

Fig. 2 Dependence of concentrations of selected metab-olites in the rat hippocampus on the plasma glucose concentration.

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Supported by WM Keck Foundation and NIH grant P41 RR08079.