Regional Metabolism During Healthy Aging in Mice Brain: A ^{1}H-[^{13}C]-NMR Study

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Introduction: Normal aging is associated with a decline in brain function and is a major risk factor for many neurodegenerative diseases. The brain mitochondrial function has been shown to be altered with healthy aging. Despite key role of neurotransmitter in brain function, brain energy metabolism has not been explored during healthy aging. As glutamatergic and GABAergic neurotransmitter energetics is supported by brain oxidative glucose metabolism[1], normal aging will lead to impairment in glutamate and GABA neurotransmitters pathway. In this study, we have investigated energy metabolism associated with glutamatergic and GABAergic neurons in the cerebral cortex, hippocampus and striatum of young adult and aged C57BL6 mice by using ^{1}H-[^{13}C]-NMR spectroscopy together with infusion of ^{13}C labeled glucose and acetate.

Materials and Methods: Two groups of male C57BL6 mice were studied: Group (i) 6 month old (young adult, n=26), Group (ii) 24 month old (aged, n=26). Overnight fasted mice were anesthetized with urethane (1.5 g/kg, i.p). [1,6-$^{13}C_2$]Glucose was administered via tail vein into mice for different time (10, 20, 35, 60 and 90 min) using a bolus-variable rate infusion protocol[2]. In addition, both groups of mice were infused with [2-^{13}C]acetate[3] for 15 and 120 min to evaluate astroglial metabolism, and the ratio V_{cyc}/V_{tca} for glutamatergic and GABAergic neurons, respectively. At the end of infusion, blood was collected and mice head were frozen in liquid nitrogen. Frozen brain was dissected into the cerebral cortex, hippocampus and striatum, and metabolites were extracted from frozen tissues[4]. The concentration and percent ^{13}C enrichment of metabolites were determined from the ^{1}H-[^{13}C]-NMR spectrum of the tissue extract recorded at 600 Hz Bruker AVANCE spectrometer[5]. The ratio V_{cyc}/V_{tca} for different brain regions was determined from the steady state [2-^{13}C]acetate experiment[6]. ^{13}C Turnover of amino acids from [1,6-$^{13}C_2$]glucose was analyzed using a three compartment metabolic model to determine the glutamatergic and GABAergic fluxes in different brain regions with aging[2].

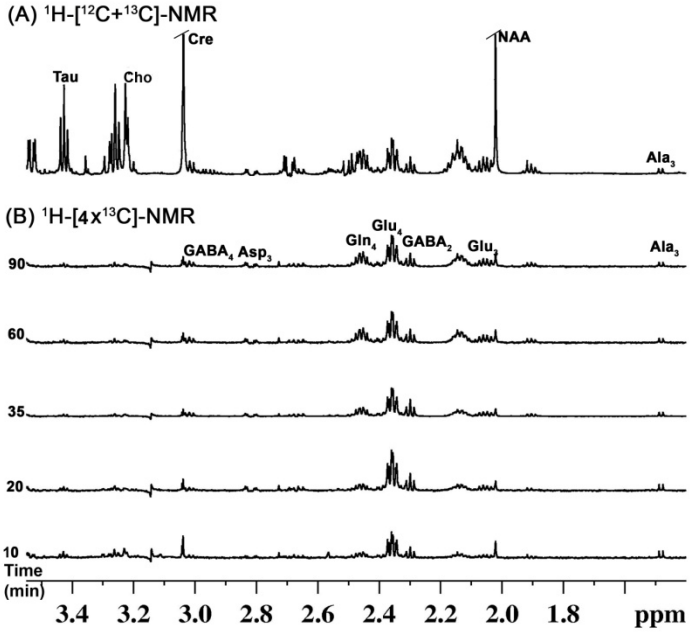

Fig. 1 (A) ^{1}H-[^{12}C+^{13}C]-NMR spectrum, (B) ^{1}H-[4x^{13}C]-NMR spectra from striatum

Results and Discussion: The level of cerebral metabolites was quantified in the tissue extract from the ^{1}H[^{12}C+^{13}C]-NMR spectrum (Fig. 1A). Level of glutamate, aspartate and taurine was found to be reduced significantly ($p<0.05$) in the cerebral cortex and striatum in aged mice. The level of glutamine and choline was elevated in the hippocampus of aged mice as compared with young adult. Analysis of accumulation of ^{13}C label into amino acids from [2-^{13}C]acetate suggest that astroglial metabolism is increased in the aged mice. The increment in astroglial metabolism follows the order: cerebral cortex < hippocampus < straiatum with aging (Fig. 2B). The glutamatergic TCA cycle and neurotransmitter cycle was found to reduced with age and the reduction follows the order hippocampus < cerebral cortex < striatum. The reduction in GABAergic fluxes follows the order cerebral cortex < striatum ~ hippocampus (Fig. 2B). The decreased excitatory and inhibitory fluxes across brain regions may explain the reduced cognitive function with aging.

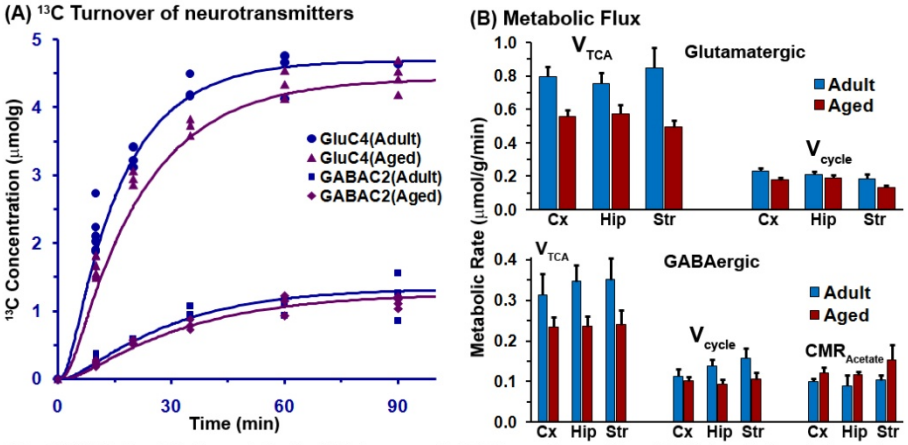

Fig. 2 (A) Fit of metabolic model to the ^{13}C Turnover of striatal neurotransmitters, (B) Metabolic flux during aging

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