Imaging TCA Cycle Metabolism in a Rat Brain by Hyperpolarization.

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Background: Real time metabolic imaging of the brain opens up exciting applications for early detection and treatment monitoring of stroke, brain tumors and Alzheimer's Disease [1]. The main limitation for imaging agents to be used in the brain is their ability to cross the blood-brain barrier (BBB). We observed limited transport of the charged molecule succinate across the BBB in earlier studies [2]. Neutral and hydrophobic compounds are known to have better transport through the BBB. The neutral compound hyperpolarized diethyl succinate has the potential of being an affective imaging agent for the brain. Diethyl 1-^{13}C 2,3-d_2 succinate is generated through the hydrogenation of diethyl 1-^{13}C 2,3-d_2 fumarate and hyperpolarized by PHIP (parahydrogen induced polarization) method, which increases the ^{13}C MR signal by 5000 fold. We have previously employed hyperpolarized diethyl succinate to image and observe real time metabolism in normal and tumor-bearing mice [3]. Using ^{13}C MRS, metabolism of diethyl succinate in the Tricarboxylic Acid Cycle (TCA cycle) was observed in normal animals after a 10 µmol tail vein injection.

Purpose: The goal of this research is to use hyperpolarized diethyl succinate to detect metabolism in a rat brain as well as to demonstrate that the compound crosses the blood-brain barrier.

Methods: We utilized PHIP to hyperpolarize diethyl 1-^{13}C 2,3-d_2 succinate in a custom-built polarizer [4] and the hyperpolarized solution was injected via the carotid artery of normal male Spraque Dawley rat (N=3) in near physiological concentrations (10-20 µmol). A ^{1}H/^{13}C dual resonance 4 cm ID solenoid volume coil was utilized for ^{13}C hyperpolarized *in vivo* imaging and spectroscopy. ^{13}C FISP sequence with a flip angle of 60^{o}, FOV 6 or 7 cm, and slice thickness of 15.2 mm was used to observe the biodistribution of the compound. ^{13}C CSI with a 1 ms gauss pulse, 200 ms TR, 8 x 8 or 16 x 16 matrix, FOV ranging from 2.64 cm to 4 cm, slice thickness of 8 to 12 mm was used. CSI was processed using 3DiCSI software (Columbia University, Qui Zhao). The flux rate of the compound within the tumor was determined using a simple pulse and acquire ^{13}C sequence. All ^{13}C imaging and spectroscopy was done on a horizontal bore Bruker Avance 4.7T animal scanner.

Results: Real time biodistribution of the hyperpolarized compound reveals that diethyl succinate (DS) is delivered to the brain of the rat by carotid arterial injection (Fig. 1). The hyperpolarized succinate signal from the inflowing blood allowed for ^{13}C imaging and spectroscopy up to 1 minute after injection. The majority of the hyperpolarized signal within the brain of the animal is also observed with ^{13}C CSI (Fig. 2). ^{13}C MR spectroscopy of the brain localized by the coil shows the formation of multiple downstream TCA cycle metabolic products from the injection of the hyperpolarized diethylsuccinate (DS) identified as succinate, malate and glutamate (Figs 3 and 4).

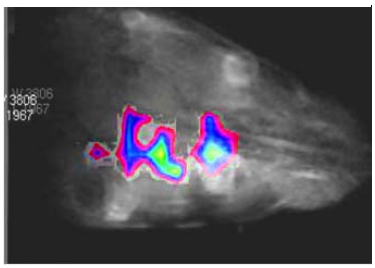

Fig 1

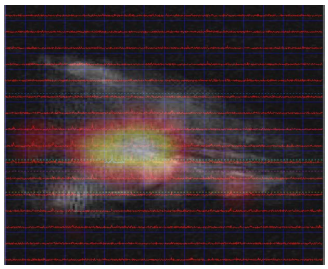

Fig 2

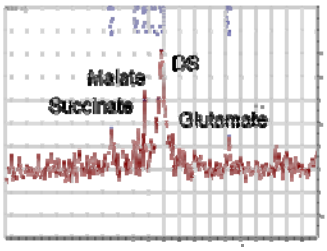

Fig 3

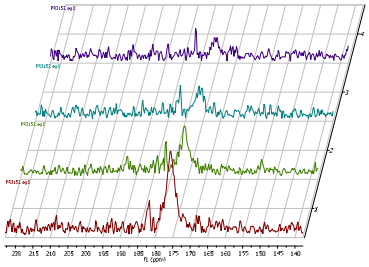

Fig 4

Conclusions: Diethyl succinate crosses the blood brain barrier and is a promising hyperpolarized metabolic imaging agent for studying the Tricarboxylic Acid (TCA) cycle in brain.

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