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Reduced hippocampal glutamate in Alzheimer disease☆

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Abstract

Altered neurometabolic profiles have been detected in Alzheimer disease (AD) using ^{1}H magnetic resonance spectroscopy (MRS), but no definitive biomarker of mild cognitive impairment (MCI) or AD has been established. This study used MRS to compare hippocampal metabolite levels between normal elderly controls (NEC) and subjects with MCI and AD. Short echo-time (TE = 46 ms) ^{1}H spectra were acquired at 4 T from the right hippocampus of 23 subjects with AD, 12 subjects with MCI and 15 NEC. Absolute metabolite levels and metabolite ratios were compared between groups using a multivariate analysis of covariance (covariates: age, sex) followed by post hoc Tukey's test ($p < 0.05$ significant). Subjects with AD had decreased glutamate (Glu) as well as decreased Glu/creatine (Cr), Glu/myo-inositol (mI), Glu/*N*-acetylaspartate (NAA), and NAA/Cr ratios compared to NEC. Subjects with AD also had decreased Glu/mI ratio compared to MCI. There were no differences between subjects with MCI and NEC. Therefore, in addition to NAA/Cr, decreased hippocampal Glu may be an indicator of AD.

Keywords: Alzheimer disease; MCI (mild cognitive impairment); MRS; Spectroscopy; Hippocampus; Glutamate

1. Introduction

Alzheimer disease (AD) is the most common form of dementia, and is characterized by progressive loss of cognitive function as well as a distinct pathological profile of neurofibrillary tangles and amyloid plaques that begins in the mediotemporal lobe (hippocampus and enthorhinal cortex) and limbic areas as early as decades before clinical diagnosis (Braak and Braak, 1994). Mild cognitive impairment (MCI) is an intermediate clinical stage along the cognitive spectrum between healthy aging and dementia that many consider to be prodromal AD. Subjects with MCI progress to AD at a rate of up to 15%/year compared to 2%/year for normal elderly to AD (Solfrizzi et al., 2004). The diagnosis of AD and monitoring of disease progression typically involves cognitive assessments to detect changes in memory, language, visuo-spatial and executive function.

The molecular neuropathology of Alzheimer disease is thought to precede structural brain alteration by several years. Hence, measurements of tissue metabolism may be sensitive biomarkers of very early disease processes. Proton magnetic resonance spectroscopy (^{1}H MRS) provides a non-invasive method of assessing brain metabolites in vivo. Short echo-time proton MRS is capable of detecting several metabolic by-products, including *N*-acetylaspartate (NAA), glutamate (Glu), glutamine (Gln), myo-inositol (mI), choline (Cho) and creatine (Cr). ^{1}H MRS has been applied previously to the study of AD and has most consistently detected decreased NAA or NAA/Cr in the parietal and occipital cortex (Miller et al., 1993), gray matter (Adalsteinsson et al., 2000; Moats et al., 1994), hippocampus (Dixon et al., 2002; Schuff et al., 1997), and posterior cingulate (Kantarci et al., 2000) as well

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as increased mI in parietal and occipital cortex (Miller et al., 1993), gray matter (Moats et al., 1994), and posterior cingulate (Kantarci et al., 2000).

Although Glu has been less well studied, it is the principal excitatory neurotransmitter involved in learning, memory and cognition and can be detected directly by short echo-time MRS. Previous MRS studies have reported decreased Glu levels in the cortex and hippocampus of transgenic AD mice (Marjanska et al., 2005) but only relative decreases in the sum of Glu and glutamine over creatine in subjects with AD in the cingulate cortex (Antuono et al., 2001; Hattori et al., 2002) and posterior cingulate gyrus, precuneus, and portions of the cuneus (Hattori et al., 2002).

The purpose of this study was to compare hippocampal metabolite levels measured by high magnetic field MRS, particularly Glu, NAA and mI, in subjects with MCI, AD, and normal elderly controls (NEC). The secondary objective was to correlate these metabolite measures with cognitive test results.

2. Methods

2.1. Subjects

All study participants (30 probable AD, 13 MCI and 17 NEC) were recruited from the Aging Brain and Memory Clinic in London, Ontario, Canada. This study was approved by the University of Western Ontario Health Sciences Research Ethics Board. Informed consent was acquired according to the Declaration of Helsinki. Several subjects did not complete their MRI scan due to discomfort or claustrophobia (2 AD, 1 MCI, 1 NEC) and several datasets were excluded due to poor spectral quality (5 AD, 1 NEC) attributable to a poor shim or patient motion during the scan. The remaining subjects (23 probable AD, 12 MCI and 15 NEC) were included in all statistical analyses.

All subjects with AD had probable Alzheimer disease as diagnosed by the Diagnostic and Statistical Manual for Mental Disorders (DSM) IV criteria for dementia and the National Institute of Neurological and Communicative Disorders and Stroke/Alzheimer's Disease and Related Disorder's Association (NINCDS/ADRDA) criteria for Alzheimer disease. Subjects with MCI were diagnosed according to the Petersen criteria (Petersen et al., 2001) of: (1) objective memory impairment based on age and education, (2) memory complaint corroborated by a third party, (3) normal general cognitive function, (4) no dementia and (5) intact activities of daily living (ADL). For inclusion in the MCI group, subjects required a global Clinical Dementia Rating (CDR (Morris, 1997)) scale score of 0.5 and intact activities of daily living assessed using the Lawton–Brody Physical Self-Maintenance Scale and Instrumental ADL Scale (Lawton and Brody, 1969). Depression was also ruled out as the cause of memory impairment using the Geriatric Depression Scale-Short Form (GDS-SF (Sheikh et al., 1991)) and the Cornell Scale for Depression in Dementia (Alexopoulos et al., 1988). Cognitive function was assessed in the AD, MCI, and NEC groups using the Mini-Mental State Exam (MMSE (Folstein et al., 1975)) and in the NEC and AD groups using the Dementia Rating Scale-II (DRS-II (Smith et al., 1994)). NEC subjects had: (1) no memory complaints or impairment, (2) normal instrumental ADL, and (3) age and education appropriate scores on the MMSE, and DRS-II.

The exclusion criteria for all subjects included contraindications to MRI, clinical depression, substance abuse, diagnosis of another dementia or the presence of significant vascular disease or cerebrovascular infarcts. NEC and subjects with MCI had no history of medications for Alzheimer disease. All subjects with AD were on a stable dose of a cholinesterase inhibitor (donepezil [10 mg] or galantamine [16 mg]) drug therapy for at least six months prior to data collection, and also had no history of treatment with memantine.

2.2. Data acquisition

Cognitive testing was performed on subjects within two weeks of their MRI scan. The MMSE, scored using WORLD backwards, was conducted on all subjects, while the DRS-II was performed on NEC and subjects with AD only.

To ensure consistency, all data acquisition and post-processing was performed by the same operator (R.R.). MR imaging and spectroscopy were performed on a whole body 4 Tesla Varian (Palo Alto, CA) MRI scanner with a Siemens (Erlangen, Germany) Sonata gradient coil. Prior to imaging, magnetic field homogeneity was optimized using a manual shim over the whole head using linear and Z^2 shim coils (FWHM < 24 Hz). Three-dimensional (3D) T_1-weigthed fast low angle shot (FLASH) (Frahm et al., 1986) volumetric images (TE/TR/TI = 5/9.5/500 ms, FOV = 24 cm, 2.5 mm thickness, 16 slices, 256 × 256 in-plane acquisition matrix) were acquired parallel to the long axis of the hippocampus for voxel positioning (Fig. 1A). Coronal T_1-weighted 3D-FLASH volumetric images (TI/TR/TE = 500/510/5.3 ms, FOV = 22 cm × 22 cm × 20 cm, 256 × 256 × 80 acquisition matrix) were also acquired over the whole brain and used for the segmentation of brain tissue components within the spectroscopy voxel.

Short echo-time single voxel spectra were localized by adiabatic selective refocusing (LASER (Garwood and DelaBarre, 2001), 500 μs dwell time, 2 kHz receiver bandwidth, τ_{cp} = 6 ms, TE = 46 ms) in the right hippocampus of each patient (Fig. 1A). The acquisition of full spectra (128 averages, TR = 2.2 s), macromolecule spectra (128 averages, TI_1 = 2.2 s, TI_2 = 0.7 s, TR = 4.2 s), and water unsuppressed spectra (8 averages, TR = 2.2 s) as well as all spectral processing were performed as previously described (Kassem and Bartha, 2003). The acquisition of all spectra took ∼15 min out of a total MR scan time of 60 min per subject. Voxels dimensions were chosen to maximize voxel volume while staying largely within the hippocampus of each subject and ranged