



Published in final edited form as:

Alzheimers Dement. 2012 ; 8(2): 105–113. doi:10.1016/j.jalz.2011.05.2416.

Fornix integrity and hippocampal volume predict memory decline and progression to AD

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Abstract

Background—The fornix is the predominant outflow tract of the hippocampus, a brain region known to be affected early in the course of Alzheimer’s disease (AD). The aims of the present study were to: 1) examine the cross-sectional relationship between fornix DTI measurements (fractional anisotropy (FA), and mean (MD), axial (DA) and radial (DR) diffusivities), hippocampal volume, and memory performance, and 2) compare fornix DTI measures to hippocampal volumes as predictors of progression and transition from amnesic mild cognitive impairment (MCI) to AD dementia.

Methods—23 MCI participants with baseline hippocampal volumetry and diffusion tensor imaging received detailed evaluations at baseline, 3, 6, 12 months, and 2.5 years. Six participants converted to AD over the follow-up. Fornix and posterior cingulum DTI measurements and hippocampal volumes were ascertained using manual measures. Random effects models assessed each of the neuroimaging measures as predictors of decline on the MMSE, CDR-Sum of boxes and Memory z-scores; ROC analyses examined the predictive value for conversion to AD.

Results—There was a significant correlation between fornix FA and hippocampal volumes. However, only the fornix measurements (FA, MD, DR, DA) were cross-sectionally correlated with memory z-scores. Both fornix FA and hippocampal volumes were predictive of memory decline. Individually, fornix FA and MD and hippocampal volumes were very good predictors of progression with likelihood ratios >83, and better than 90% accuracy.

Conclusion—Fornix FA both cross-sectionally correlated with and longitudinally predicted memory decline and progression to AD. Manually-drawn fornix ROI shows comparable promise to hippocampal volume as a predictive biomarker of progression and warrants replication in a larger study.

Keywords

Fornix; Hippocampus; Mild Cognitive Impairment; Biomarker; Diffusion tensor imaging

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The corresponding author had full access to all the data in the study and had final responsibility of the decision to submit for publication

All other authors have no disclosures.

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1. Introduction

Individuals with amnesic mild cognitive impairment (aMCI) have an increased risk of progressing to dementia due to Alzheimer's disease (AD-dementia) [1], and recent studies indicate that the majority of aMCI cases have AD pathology in their brains on autopsy [2, 3]. However, while a diagnosis of aMCI clearly increases the risk for progression to AD-dementia, many individuals with aMCI do not progress. Clinic-based longitudinal studies of aMCI cohorts suggest rates of instability (e.g. returning to normal cognition) for the aMCI diagnosis on the order of 10% but epidemiological studies show instability rates as high as 25–40% [4–6]. MCI cases that improve or do not decline (i.e. “non-progressors”) may not have Alzheimer's pathology, or they have a milder form of the disease, and are, therefore, less likely to benefit from current and newly-developing AD-specific therapies. Thus, there is an inherent need to identify which aMCI cases will progress to AD-dementia.

Cerebrospinal fluid (CSF) measures of amyloid-beta(1-42) and phospho-tau, and amyloid plaque imaging (e.g. 11C-PIB PET, AV-45) may be good diagnostic and predictive markers of conversion [7–9]. However, their utility as biomarkers in the population is limited by their costs and invasiveness. More recently, less costly and invasive magnetic resonance (MR) imaging measures of hippocampal and global cortical atrophy have been found to be as good as, or superior to, CSF measures in predicting aMCI progression [10, 11]. Hippocampal atrophy also correlates with memory performance [12–15], although the strength of the association can vary based on the diagnostic group(s) examined (e.g. normal control, MCI, AD-dementia) and on the type of memory tests incorporated [12, 13, 16]. However, hippocampal atrophy has not been clinically utilized as a single AD diagnostic marker due to the large variability within diagnostic groups and because atrophy can be found in other dementias as well as normal aging.

The fornix is the predominant outflow tract of the hippocampus, connecting the hippocampus with the septal nuclei and with the mammillary bodies in the hypothalamus. An initial study examining the fornix in PS1 and PS2 mutation carriers reported that, compared to non-carriers, presymptomatic mutation carriers had significantly reduced fornix fractional anisotropy (FA), measured by diffusion tensor imaging (DTI) [17]. While the cingulum bundle FA was also reduced in the presymptomatic carriers, this finding was not significant, suggesting the fornix may be a more sensitive, early pathological marker. Similarly, we have previously shown that among a group of cognitively normal controls, aMCI and early AD-dementia cases, that fornix FA is reduced with increasing severity of the disease [18]. In light of these findings and the need to identify a biomarker to predict progression from aMCI to AD, the aims of the present study were to: 1) examine the cross-sectional correlation between the fornix and posterior cingulate white matter integrity using DTI, hippocampal volumes measured with structural MR methods, and memory performance; 2) compare the imaging parameters as indicators of cognitive and functional decline; and 3) compare neuroimaging and other AD-related variables in predicting progression from aMCI to AD-dementia.

2. Methods

2.1. Participants

Participants were community-dwelling volunteers who enrolled in a longitudinal study examining the utility of neuroimaging measures as biomarkers of AD progression. Recruiting methods have previously been described [18]. Briefly, participants were recruited from the Clinical Core of the Johns Hopkins Alzheimer's Disease Research Center (ADRC) and from memory clinics associated with Johns Hopkins Medicine. Participants were diagnosed with Amnesic Mild Cognitive Impairment (MCI) if they were non-demented

participants with mild memory problems, Clinical Dementia Rating (CDR)=0.5, and met Mayo criteria for amnesic MCI, single or multiple domains [19]. All participants were required to be older than 55 years, have no history of a neurological disease or of a major psychiatric illness, and have an informant who could provide information about their daily function. Twenty-five MCI participants completed the baseline examination, 23 of whom had available both hippocampal volumes and DTI scans and comprised the sample for the present analyses. Informed consent was obtained prior to the initiation of the study in accordance with the requirements of the Johns Hopkins Institutional Review Board. Consent procedures followed the guidelines endorsed by the Alzheimer's Association for participation of cognitively impaired individuals [20].

2.2. Assessments

Each participant received a detailed medical evaluation at baseline, three months, six months and one-year later, including: (1) a medical, psychiatric and neurologic history; (2) a neuropsychological battery; (3) a physical, psychiatric and neurological examination; (4) an assessment of clinical severity using the CDR scale [21]; (5) MRI scan; and (6) a blood draw. An additional in-person clinical examination was conducted approximately 2.5 years after baseline for the purpose of determining clinical progression from MCI to AD dementia, based on a consensus diagnosis by ADRC investigators.

The neuropsychological battery included the Mini-Mental State Exam (MMSE) [22]; Alzheimer's Disease Assessment Scale – cognitive portion (ADAS-Cog) [23]; California Verbal Learning Test (CVLT) [24]; the Logical Memory Story A from the Wechsler Memory Scale (WMS) [25]; Trail Making Test (TMT) Parts A (TMT-A) and B (TMT-B) [26]; and the total score on the Controlled Oral Word Association test for category and letter fluency [27]. A composite memory score ("Memory z-scores") was computed using the baseline means and standard deviations for the cognitively healthy subjects enrolled in this study. The memory z-scores included the following tests: CVLT-total correct trials 1 to 5, CVLT-short delayed free recall, CVLT-long delayed free recall, WMS-immediate memory, and WMS-delayed memory.

The CDR [21] examines functioning in six domains: memory, orientation, judgment/problem solving, community affairs, home/hobbies, and personal care. Scores include a composite score (CDR-composite) and Sum of Boxes (CDR-Sum), which is the sum of ratings in each of six domains with a range of 0 (no impairment) to 30 (maximum impairment in all domains). CDR-SB was chosen herein, instead of the composite, because of its greater range and demonstrated sensitivity to change in MCI and AD demented (e.g. [28]).

2.3. MRI acquisition

MRI images were acquired on a 3.0 Tesla (3T) scanner (Philips Medical Systems, Best, The Netherlands) at the F.M. Kirby Research Center for Functional Brain Imaging at the Kennedy Krieger Institute. A Magnetization Prepared Rapid Gradient Recalled Echo (MPRAGE) scan and a DTI scan was acquired. The MPRAGE scan was conducted according to the protocol of the Alzheimer's Disease Neuroimaging Initiative (ADNI).

DTI images were acquired using a SENSE head coil on the 3T scanner, equipped with Dual Quasar gradients (up to 80 mT/m). For acquisition, an eight-element arrayed RF coil, converted to six-channel to be compatible with the six-channel receiver, was used. For DTI acquisitions, a single-shot spin echo - echo planar sequence (SE-EPI) was used, with diffusion gradients applied in 32 non-collinear directions and $b = 700 \text{ s/mm}^2$. Five additional reference images with least diffusion weighting ($b = 33 \text{ s/mm}^2$) were also acquired. Fifty to

sixty axial slices were acquired to cover the entire cerebral hemispheres and the cerebellum, parallel to the AC-PC line. The field of view, the size of the acquisition matrix, and the slice thickness were 212×212 mm/ $96 \times 96 / 2.2$ mm. Other imaging parameters were: TR > 7,000 ms and TE = 80 ms; and SENSE reduction factor = 2.5. To improve the signal-to-noise ratio, two datasets were acquired, leading to a total acquisition time of 7 minutes.

2.3.1. Hippocampal Volume Measurement—Right and left hippocampal volumes were calculated by voxel counting on binary hippocampal images segmented on T1-MRI data as previously described [29]. A template-based segmentation algorithm was used to segment the binary hippocampal images for the baseline scans. Briefly, a representative elderly normal subject was selected as the template and left and right hippocampi were segmented manually. A total of 38 landmarks were placed on the hippocampus similar to the procedure described in Csernansky et al [30]. First, the head and the tail landmarks were placed to identify the principal axis of the structure. Nine slices at equal distance from each other and perpendicular to this principal axis were then identified and a total of four landmarks were placed at the anterior, inferior, posterior, and superior midpoints in each slice. Using these landmarks, the subvolume images were then created around the hippocampi and the landmarks were used to calculate a rigid transformation [31] between the template and individual subject subvolumes. The large deformation diffeomorphic metric mapping (LDDMM) landmark matching algorithm [32] was used to register the subvolumes further. These transformations were subsequently used to move the template hippocampi segmentation onto each subject's MR scan and to create an initial hippocampus segmentation for the subject. The alternating kernel mixture (AKM) [33] method was used to segment the subject subvolume image into Grey Matter (GM)/white matter (WM)/CSF regions. Using this AKM segmentation, any mislabeled WM and CSF voxels were removed from the initial hippocampus segmentation. The right and left hippocampi were initially examined separately. However, as there were no differences in the correlations or predictive performances for the right vs. left hippocampus, the two volumes were averaged. In order to control for head size, the outcome measure was the ratio of the total hippocampal volume to the intracranial volume (ICV).

2.4 DTI Data Processing

The DTI datasets were transferred to a personal computer running a Windows platform and were processed using DtiStudio (mri.kennedykrieger.org or www.DtiStudio.org) [34]. Images were first realigned by affine transformation using Automatic Image Registration [35], in order to remove any potential small bulk motion and Eddy-current distortion. The six elements of the diffusion tensor were calculated for each pixel using multivariate linear fitting. After diagonalization, three eigenvalues and eigenvectors were obtained. For the anisotropy map, fractional anisotropy (FA) was used [36]. The eigenvector (v_1) associated with the largest eigenvalue was used as an indicator for fiber orientation. A 24-bit color-coded orientation map was created by assigning red, green, and blue channels to the x (right-left), y (anterior-posterior), and z (superior-inferior) components of the v_1 and its intensity was modulated by FA. The mean diffusivity (MD) was calculated as an average of the three eigenvalues. The parallel, or axial, diffusion (DA) was identical to the first eigenvalue, and the radial diffusion (DR) was defined as an average of the second and third eigenvalues.

2.5 DTI regions of interest

Protocols were developed to identify specific fiber tracts and to manually delineate regions of interest (ROI) within the fiber tracts using the in-house software MriStudio/RoiEditor (www.MriStudio.org or mri.kennedykrieger.org), as previously described [18]. Eight ROIs were originally identified and manually drawn with high reliability (mean interclass correlation = 0.87; range = 0.82–0.95) using standardized guidelines based on location,

color, and size. For the purpose of the present study, we focused on efferent tracks of the hippocampus, including the fornix and posterior cingulum. These ROIs were identified as follows (Fig. 1): 1) fornix: the body of the fornix drawn in two adjacent axial slices using the ventral midbrain and splenium of the corpus callosum as anatomic landmarks; 2) cingulum bundle: drawn on the posterior portion of the cingulum bundle on the same axial slices as the body of the fornix. As there was little difference between consecutive slices of the fornix or posterior cingulum in relation to outcomes, we took the average of the two slices. Left and right sides of the posterior cingulum were initially examined separately. As there was little difference in the results, the DTI measurements (FA, MD, DA, DR) of the two sides was averaged for the analyses presented here. Thus, we had single measures of FA, MD, DA, and DR for both the fornix and the posterior cingulum.

2.6 Statistical analysis

The demographic and health-related characteristics were examined by MCI converter status (stable vs. converter) using Fischer's Exact Test for dichotomous variables and students t-tests for continuous variables. Individuals who progressed to a clinical consensus-conferenced diagnosis of AD-dementia over the follow-up were considered converters; those who remained with a diagnosis of MCI were considered to be stable. None of the participants reverted from a diagnosis of MCI to a cognitive normal control. MD, DA, DR and the hippocampal volume/ICV ratio were transformed by multiplying by 10^2 in order to have comparable units to the FA. Partial correlation coefficients were used to examine the cross-sectional baseline correlations between the fornix and posterior cingulum FA, MD, DA and DR values and hippocampal volume, adjusting for baseline age; and between the fornix and posterior cingulate FA, MD, DA, and DR values, hippocampal volume, and memory z-scores adjusting for age and years of education. Random effects models were used to examine the baseline neuroimaging measures and decline on the MMSE, CDR-sum of boxes (CDR-Sum), and memory z-score. This approach allows us to assess the effects of key fixed factors, such as age, on the average rate of change in each outcome while accounting for the dependence between within-subject repeated measures. Multivariate models controlled for age and education. We initially examined time as a random effect, but this variable did not contribute to the model in terms of log-likelihood values so only a random intercept was incorporated in the final models.

The predictive values of the fornix and posterior cingulate FA, MD, DA, and DR and hippocampal/ICV ratio were calculated using receiver operating characteristics (ROC) analyses. The area under the ROC curve (AUC) and 95% confidence intervals were estimated and sensitivity, specificity, and accuracy (percent correctly classified) were computed. The *a priori* p-value was set at $p < 0.05$. All analyses were conducted using STATA Version 10.0 (StataCorp, College Station, TX).

3. Results

All 23 MCI participants were followed for at least one year; 20 were followed up to 2.5 years. There were no demographic or clinical differences between the three participants without a follow-up at 2.5 years and the 20 participants with a follow-up. Six of the 23 MCI participants (26.1%) progressed to a diagnosis of AD over the 2.5 years. Those who progressed had lower MMSE and memory z-scores at baseline compared to those who did not progress (Table 1). There were no differences in demographics such as age, education or APOE E4 allele status by progression status, nor was there a significant difference for CDR-Sum.

Examining the baseline partial correlation coefficients among the DTI variables and hippocampal/ICV ratio, controlling for age, only a higher fornix FA strongly and

significantly correlated with a larger hippocampal/ICV ratio ($r = 0.438$, $p = 0.041$). Higher fornix MD, DA, and DR also correlated with smaller hippocampal/ICV ratios, but these results did not reach significance at the $p < 0.05$ level (Table 2). The posterior cingulate DTI measures did not significantly correlate with the total hippocampal volume/ICV ratio ($p > 0.05$). Examining the partial correlations between all neuroimaging measures and memory z-scores, controlling for age and education, only fornix FA ($r = 0.441$, $p = 0.051$), MD ($r = -0.464$, $p = 0.039$), DA ($r = -0.448$, $p = 0.048$), and DR ($r = -0.458$, $p = 0.043$) correlated with memory z-scores (Table 2). Neither the hippocampal/ICV ratio nor the posterior cingulate DTI measures trended towards an association with memory z-scores.

The baseline neuroimaging measures were next examined as predictors of progression on the MMSE, CDR-sum and memory z-scores using random effects models. In multivariable analyses, controlling for age and education, only fornix FA ($b = 7.62$, 95% CI: 0.91–14.35), DA ($b = -18.52$, 95% CI: -34.31, -2.74) and DR ($b = -20.56$, 95% CI: -37.37, -3.76) were associated with better memory z-scores at baseline. Fornix DA and DR were also associated with CDR-Sum at baseline (Table 3). In longitudinal analyses, both the low fornix FA and low hippocampal/ICV ratio were predictive of decline on the MMSE, CDR-sum, and memory z-scores over 2.5 years of follow-up (Table 3). Fornix MD was predictive of decline on the MMSE and CDR-sum, but not on memory z-scores and the fornix DA was only predictive of MMSE decline. Interestingly, the posterior cingulate MD ($b = -0.04$, 95% CI: -0.06 – -0.01) and DR ($b = -0.06$, 95% CI: -0.12, -0.01), but not FA or DA, was also predictive of memory decline but not decline on the MMSE or CDR-sum.

Lastly, we examined the classification accuracy of the neuroimaging measures and other factors with respect to progression from MCI to AD using ROC analyses. Both the fornix FA and MD and hippocampal/ICV ratio were very good predictors of progression with AUC > 0.90 , likelihood ratios (LR) > 83 , and better than 90% accuracy (Table 4). Comparison of the fornix (FA, MD, DA or DR) and hippocampal AUC scores did not show statistically significant differences. Similarly, because the AUC and LRs were so high, the combination of the fornix FA or MD and hippocampus did not improve on the predictive value of each measure in isolation. It is important to point out, however, that the predictive value of these neuroimaging measures using ROC analyses only trended towards significance ($p = 0.1$) to be better than the predictive value of the MMSE and CDR-sum. This is likely due to the small sample size and large confidence intervals because the comparison of likelihood ratios clearly indicate that the fornix FA and MD and hippocampal/ICV ratio are superior to the clinical measures.

4. Discussion

This study examined the relationship between MRI measures including hippocampal volume and fornix and posterior cingulum (PC) white matter integrity, their relation to memory, and their predictive value for cognitive decline and progression from MCI to AD dementia. Cross-sectionally, fornix white matter integrity (FA, MD, DA, and DR) correlated more strongly with memory performance than the hippocampal/ICV ratio or PC white matter integrity. Longitudinally, both fornix FA and hippocampal/ICV ratio were predictive of decline in memory performance and progression from MCI to AD dementia over 2.5 yrs. Fornix white matter integrity (FA, MD, DA, and DR) was as good as the hippocampal volume in predicting progression from MCI to dementia. Since the fornix (FA, DA and DR) also cross-sectionally correlated with memory performance, the fornix may be a better correlate of early disease progression, and a clinically useful biomarker. Additional studies with larger sample sizes are warranted to confirm these results.

The disruption of white matter, including axonal and dendritic integrity, occurs early in the AD process, and has been demonstrated in both animal and *in vivo* human studies [37–39]. One evolving hypothesis is that disruption of white matter tracts reflects the susceptibility of late-myelinating regions to the effects of aging and AD. These white matter changes make individuals particularly vulnerable to the clinical manifestations of AD [39] and occur before neuronal degradation and atrophy are detectable. DTI was developed to measure the integrity of white matter fiber bundles in the nervous system [40, 41]. It is thought to be especially sensitive to microstructural alterations in nerve fibers [42], and a sensitive early indicator of Alzheimer pathology [43]. There are multiple indices that can be measured with DTI. FA is a measure of anisotropic water diffusion and reflects the degree of directionality of cellular structures within the fiber tracts and, therefore, the structural integrity of tracts [44]. MD, a measure of randomized mean water diffusion, represents a loss of barriers restricting the water motion, partial breakdown of tissue cytoarchitecture [44] or demyelinating processes [39]. However, FA and MD alterations could be due to changes of diffusion either parallel or perpendicular to the principal direction of the tensor. Thus, in the present study we also examined DA and DR. Axial diffusivity (DA) is a measure of parallel diffusivity and is thought to be an indicator of axonal damage while radial diffusivity (DR) is a measure of perpendicular diffusivity and thought to be an indicator of myelin breakdown [45].

Previous DTI studies using the ROI approach have focused on hippocampal or posterior cingulum FA, MD, or apparent diffusion coefficient (ADC) (e.g. [46–48]). These studies suggest that hippocampal MD and ADC is higher, and FA lower, in MCI cases compared to cognitively normal controls. One study that compared hippocampal volumetry and DTI measures reported poor correlation between the volumetry and DTI measures [48], and that left hippocampal MD was the strongest independent correlate of poor verbal memory performance and also best separated MCI cases from normal controls. Another study reported higher hippocampal ADC values in MCI cases who converted to AD dementia compared to those who did not convert over two years [49].

In contrast to these studies, we focused directly on the fornix, the predominant outflow tract of the hippocampus that connects it with the septal nuclei and mamillary bodies in the hypothalamus. The posterior fornix fibers further connect to anterior thalamic nuclei and to cingulate cortex. Prior studies have demonstrated that limbic projections and pathways (e.g. fornix) connecting to the frontal lobes are preferentially affected early in the course of AD [39, 50]. An initial study examining the fornix in PS1 and PS2 mutation carriers reported that, compared to non-carriers, presymptomatic mutation carriers had significantly reduced fornix FA [17]. While the cingulum bundle FA was also reduced in the presymptomatic carriers, this finding was not significant, suggesting the fornix may be a more sensitive, early pathological marker. Our results, among MCI cases, are congruent with this finding such that the fornix, but not the PC, correlated cross-sectionally with memory performance, and predicted longitudinal decline in memory and progression to AD. While the PC MD and DR were predictive of memory decline, the fornix was a much better predictor of clinical conversion to AD dementia and is likely a more useful correlate of disease progression.

As mentioned, the majority of DTI studies to date have primarily reported on FA and MD. Of those using a ROI approach, FA has been the most sensitive [18, 51] although another study reported that MD was more sensitive [46]. It has been suggested that ROI-based approaches may inflate the significance of FA over MD [52]. Indeed, a recent cross-sectional voxel-wise analysis of 25 AD patients and 13 elderly controls using tract-based spatial statistics reported alterations in diffusivity (MD, DA and DR) were more significant and more sensitive than FA in temporo-parietal white matter, posterior cingulum, splenium and fornix [52]. However, another recent cross-sectional study, using an atlas-based

tractography approach, of 25 AD patients, 19 MCI, and 15 cognitively normal controls reported group differences that varied by DTI measurement and tract [53]. FA was significantly different among groups only in the fornix; DA and MD in all white matter tracts except the fornix; and DR was altered in the fornix and interior longitudinal fasciculus. These data suggest that the sensitivity of the DTI measurements may vary by region and tract of interest.

In the present ROI-based study of participants with MCI, we also examined DA and DR in addition to FA and MD. While DA and DR were more strongly associated with cognitive performance cross-sectionally (Table 2), fornix FA and MD were more strongly predictive of longitudinal decline. Thus, it is also possible that the sensitivity of each DTI measures varies by diagnostic group and over time. Ultimately, a longitudinal study of normal controls, MCI and AD participants using various approaches (ROI-based, voxel-based and tract-based) are needed to better understand these conflicting results and to determine the relationship of the methodology to any differences in findings between FA, MD, DA and DR. This is particularly important in further understanding the pathology of AD in terms of whether damage to white matter is due to secondary degeneration or primary myelin damage [45].

Limitations of this study warrant consideration. First, while the expected percentage (10–15%/year) of individuals with MCI converted to AD, the overall sample size was small. Second, a possible confounder in this study is that atrophy of the fornix could also contribute to decreased FA. The smaller the fornix, the more CSF could be included in a given voxel size. Since CSF has FA near 0, the partial volume effect of increased CSF in a voxel would cause the mean FA within that voxel to be decreased. While we cannot completely eliminate the partial volume effect, we did use high-resolution MPRAGE and Large Deformation Diffeomorphic Metric Mapping (LDDMM) analysis, in an attempt to reduce the likelihood of this problem.

A recently-published phase I clinical trial evaluating the effects of fornix deep brain stimulation (F-DBS) in AD patients supports the relevance of the fornix to the pathophysiology of AD [54]. Thus, this biomarker could be a useful measure to enrich a clinical trial with those most likely to progress.

Acknowledgments

The authors would like to thank Dr. Gwenn Smith for comments on the manuscript. This research was funded in part by grants from GlaxoSmithKline, the National Institute on Aging (P50-AG005146, P50-AG021334, R21AG033774, and R21NS060271-01) and the National Institute of Research Resources (NCRR, P41-RR15241). NCRR is a component of the National Institutes of Health (NIH). The manuscript's contents are solely the responsibility of the authors and do not necessarily represent the official view of NCRR or NIH.

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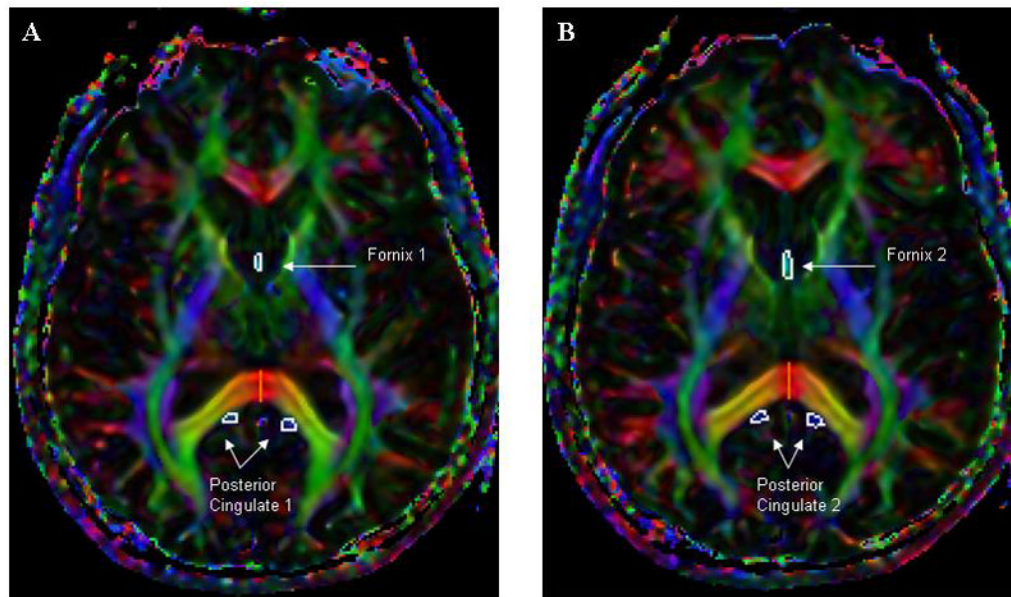


Figure 1. Regions of interest on two consecutive slices (A and B) for the fornix and posterior cingulate.

Table 1

Demographic and Cognitive Characteristics of the MCI participants and by Converter Status

Variable	All (n=23) mean(SD)/N(%)	Stable MCI (n=17)	Converted to AD (n=6)	p-value
		mean(SD)/N(%)	mean(SD)/N(%)	
Age (yrs)	75.6 (5.5)	74.5 (5.8)	78.7 (2.9)	0.112
Female	7 (30.4%)	5 (29.4%)	2 (33.3%)	0.618
Education (yrs)	15.6 (3.1)	15.3 (3.1)	16.3 (3.4)	0.497
APOE E4 allele	10/22 (45.5%)	9/16 (56.3%)	1/6 (16.7%)	0.162
Dementia Med	4/22 (18.2%)	2/16 (12.5%)	2/6 (33.3%)	0.292
CDR-Sum	1.3 (0.8)	1.2 (0.8)	1.7 (0.7)	0.229
MMSE	26.7 (2.1)	27.2 (2.0)	25.0 (1.3)	0.018
Memory Z-score	-2.0 (1.2)	-1.5 (1.0)	-3.2 (0.3)	0.001
CVLT-total	34.2 (12.9)	38.6 (12.1)	22.5 (5.5)	0.006
CVLT-sdfr	5.4 (3.7)	7.1 (2.6)	0.7 (0.8)	<0.001
CVLT-ldfr	5.5 (3.9)	6.8 (3.5)	1.8 (2.3)	0.004
Wechsler-Imm	8.2 (3.1)	8.8 (3.3)	6.5 (1.8)	0.128
Wechsler-Del	6.0 (4.2)	7.1 (4.1)	2.8 (2.7)	0.031
GDS	1.5 (1.2)	1.5 (1.1)	1.2 (1.6)	0.609

Abbreviations: MCI=amnesic mild cognitive impairment; AD=Alzheimer's disease; CDR-Sum = Clinical Dementia Rating, Sum of Boxscores; MMSE=Mini-mental State Examination; CVLT=California Verbal Learning Test; GDS=15-item Geriatric Depression Scale

* Fischer's Exact test for categorical variables; Students t-test for continuous variables

Table 2

Cross-sectional Correlations Between the Fornix, Posterior Cingulate, Hippocampal/ICV Ratio, and Memory Z-score

	Total Hippocampal Volume/ICV*		Memory Z-score*	
	<i>corr</i>	<i>p-value</i>	<i>corr</i>	<i>p-value</i>
<i>Fornix</i>				
Fractional Anisotropy (FA)	0.438	0.041	0.441	0.051
Mean Diffusivity (MD)	-0.281	0.205	-0.464	0.039
Axial Diffusivity (DA)	-0.209	0.350	-0.448	0.048
Radial Diffusivity (DR)	-0.212	0.344	-0.458	0.043
<i>Posterior Cingulate</i>				
Fractional Anisotropy (FA)	0.360	0.100	-0.120	0.615
Mean Diffusivity (MD)	-0.088	0.697	0.009	0.969
Axial Diffusivity (DA)	0.200	0.371	-0.267	0.255
Radial Diffusivity (DR)	-0.324	0.141	0.105	0.659
<i>Hippocampal Volume/ICV</i>			0.182	0.444

ICV = intracranial volume

* Controlling for age (imaging) and age and education (memory Z-score)

Table 3

DTI measures and hippocampal volumes predict decline in MCI over 2.5 years

	Univariate			Multivariate*		
	b (95% CI)	p	b* time (95% CI)	p	b* time (95% CI)	p
Fornix FA						
mmse	14.45 (3.48, 25.41)	0.010	0.03 (0.01, 0.04)	< 0.001	0.03 (0.01, 0.04)	< 0.001
cdrsom	-3.36 (-7.84, 1.12)	0.141	-0.01 (-0.02, -0.01)	< 0.001	-0.01 (-0.02, -0.01)	< 0.001
memory Z	10.06 (3.93, 16.19)	0.001	0.01 (0.001, 0.01)	0.020	0.01 (0.001, 0.01)	0.020
Fornix MD (x10⁻²)						
mmse	-14.07 (-24.54, -3.60)	0.008	-0.02 (-0.04, -0.01)	0.001	-0.02 (-0.03, -0.01)	0.001
cdrsom	3.64 (-0.43, 7.73)	0.079	0.01 (0.005, 0.02)	0.001	0.01 (0.005, 0.02)	0.001
memory Z	-8.74 (-14.81, -2.67)	0.005	-0.004 (-0.01, 0.001)	0.151	-0.004 (-0.009, 0.001)	0.158
Fornix DA (x10⁻²)						
mmse	-37.54 (-62.16, -12.93)	0.003	-0.04 (0.07, -0.01)	0.022	-0.04 (-0.07, -0.005)	0.023
cdrsom	10.36 (0.43, 20.28)	0.041	0.01 (-0.001, 0.03)	0.060	0.01 (-0.001, 0.03)	0.057
memory Z	-23.85 (-37.80, -9.90)	0.001	-0.0004 (-0.01, -0.01)	0.945	-18.52 (-34.31, -2.74)	0.947
Fornix DR (x10⁻²)						
mmse	-40.54 (-69.44, -11.64)	0.006	-0.02 (-0.06, 0.01)	0.160	-0.02 (-0.06, 0.01)	0.156
cdrsom	14.04 (3.70, 24.38)	0.008	0.01 (-0.002, 0.03)	0.092	15.40 (3.95, 26.85)	0.089
memory Z	-26.52 (-42.07, -10.98)	0.001	0.002 (-0.01, 0.01)	0.721	-20.56 (-37.37, -3.76)	0.726
Posterior Cingulate FA						
mmse	-8.36 (-31.77, 15.05)	0.484	0.01 (-0.02, 0.03)	0.709	-5.26 (-23.29, 12.78)	0.568
cdrsom	0.68 (-7.92, 9.28)	0.876	-0.002 (-0.01, 0.01)	0.753	0.55 (-7.92, 9.04)	0.898
memory Z	-3.69 (-16.35, 8.96)	0.567	0.004 (-0.004, 0.01)	0.372	-2.86 (-14.07, 8.35)	0.617
Posterior Cingulate MD (x10⁻²)						
mmse	-2.39 (-52.93, 48.16)	0.926	-0.02 (-0.06, 0.03)	0.428	-9.48 (-48.21, 29.26)	0.632
cdrsom	2.61 (-15.54, 20.77)	0.778	0.02 (-0.005, 0.04)	0.129	3.26 (-14.80, 21.31)	0.724
memory Z	10.26 (-17.49, 38.01)	0.469	-0.04 (-0.06, -0.01)	0.006	8.98 (-16.00, 33.96)	0.481
Posterior Cingulate DA (x10⁻²)						
mmse	-170.16 (-335.79, -4.54)	0.044	0.01 (-0.17, 0.19)	0.903	-114.15 (-245.63, 17.33)	0.089
cdrsom	53.06 (-8.67, 14.79)	0.092	-0.01 (-0.09, 0.08)	0.868	49.70 (-12.40, 111.79)	0.117

	Univariate			Multivariate*		
	b (95% CI)	p	b* time (95% CI)	b (95% CI)	p	b* time (95% CI)
memory Z	-67.43 (-159.22, 24.36)	0.150	-0.01 (-0.08, 0.05)	-47.52 (-131.28, 36.24)	0.266	-0.01 (-0.08, 0.05)
Posterior Cingulate DR (x10 ⁻²)						
mnse	7.05 (-173.83, 188.13)	0.938	-0.16 (-0.33, 0.01)	23.07 (-115.20, 161.33)	0.744	-0.15 (-0.32, 0.01)
cdrsum	-12.40 (-78.51, 53.70)	0.713	0.03 (-0.05, 0.11)	-17.52 (-82.33, 47.28)	0.596	0.03 (-0.05, 0.11)
memory Z	13.23 (-84.41, 110.88)	0.790	-0.06 (-0.12, -0.01)	23.31 (-62.56, 109.18)	0.595	-0.06 (-0.12, -0.01)
Hippocampal/ICV Ratio (x10 ⁻²)						
mnse	13.31 (-14.51, 41.14)	0.348	0.11 (0.07, 0.15)	-4.41 (-30.32, 21.52)	0.739	0.11 (0.07, 0.15)
cdrsum	-2.44 (-12.98, 8.11)	0.650	-0.05 (-0.06, -0.03)	-2.89 (-14.98, 9.21)	0.640	-0.05 (-0.06, -0.03)
memory Z	11.90 (-3.99, 27.78)	0.142	0.03 (0.01, 0.04)	4.37 (-12.95, 21.70)	0.621	0.03 (0.01, 0.04)

* Controlling for age and education

Table 4
 Receive Operating Characteristic (ROC) Analyses for Predictors of Conversion from MCI to AD

Predictor	AUC [95% CI]	Optimal Cut-off	Sensitivity	Specificity	Accuracy	Likelihood Ratio
Age						
Higher Age (years)	0.71 (0.48, 0.93)	>80	66.70%	70.60%	69.60%	2.27
CDR-Sum						
Higher CDR-Sum	0.73 (0.46, 0.99)	>2	50.00%	88.30%	78.30%	4.27
MMSE						
Lower MMSE	0.82 (0.65–0.99)	<24	50.00%	88.24%	78.26%	4.25
Posterior Cingulate						
Low FA	0.62 (0.32, 0.92)	<0.43	50.00%	82.35%	73.90%	2.83
High MD ($\times 10^{-2}$)	0.73 (0.46, 0.99)	>0.248	83.33%	76.47%	78.26%	3.54
High DA ($\times 10^{-2}$)	0.58 (0.34, 0.82)	>0.134	16.67%	82.35%	65.22%	0.94
High DR ($\times 10^{-2}$)	0.73 (0.43, 1.00)	>0.61	83.33%	82.35%	82.61%	4.72
Fornix						
Low FA	0.95 (0.85, 1.00)	<0.435	83.30%	100.00%	95.70%	>83.00*
High MD ($\times 10^{-2}$)	0.92 (0.79, 1.00)	>0.41	83.30%	94.12%	91.30%	14.17
High DA ($\times 10^{-2}$)	0.79 (0.60, 0.98)	>0.22	66.67%	82.35%	78.26%	3.78
High DR ($\times 10^{-2}$)	0.86 (0.71, 1.00)	>0.098	100.00%	82.35%	86.96%	5.67
Hippocampal Volume						
Low Volume/ICV ratio ($\times 10^{-2}$)	0.98 (0.93, 1.00)	<0.13	83.30%	100.00%	95.70%	>83.00*

* The likelihood ratio (Sensitivity/1-Specificity) could not be calculated since the denominator was 0. For purposes of estimation, the denominator was replaced with 1.

FA = fractional anisotropy; MD = mean diffusivity; DA = axial diffusivity; DR = radial diffusivity