

Use of Structural Magnetic Resonance Imaging to Predict Who Will Get Alzheimer's Disease

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We used magnetic resonance imaging (MRI) measurements to determine whether persons in the prodromal phase of Alzheimer's disease (AD) could be accurately identified before they developed clinically diagnosed dementia. Normal subjects ($n = 24$) and those with mild memory difficulty ($n = 79$) received an MRI scan at baseline and were then followed annually for 3 years to determine which individuals subsequently met clinical criteria for AD. Patients with mild AD at baseline were also evaluated ($n = 16$). Nineteen of the 79 subjects with mild memory difficulty "converted" to a diagnosis of probable AD after 3 years of follow-up. Baseline MRI measures of the entorhinal cortex, the banks of the superior temporal sulcus, and the anterior cingulate were most useful in discriminating the status of the subjects on follow-up examination. The accuracy of discrimination was related to the clinical similarity between groups. One hundred percent (100%) of normal subjects and patients with mild AD could be discriminated from one another based on these MRI measures. When the normals were compared with the individuals with memory impairments who ultimately developed AD (the converters), the accuracy of discrimination was 93%, based on the MRI measures at baseline (sensitivity = 0.95; specificity = 0.90). The discrimination of the normal subjects and the individuals with mild memory problems who did not progress to the point where they met clinical criteria for probable AD over the 3 years of follow-up (the "questionables") was 85% and the discrimination of the questionables and converters was 75%. The apolipoprotein E genotype did not improve the accuracy of discrimination. The specific regions selected for each of these discriminations provides information concerning the hierarchical fashion in which the pathology of AD may affect the brain during its prodromal phase.

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The discrimination of those destined to develop Alzheimer's disease (AD) from the larger pool of individuals with mild memory loss is of increasing importance, as strategies for the prevention or delay of dementia are developed. Early detection of AD is, however, complicated by the fact that the disease progresses in an insidious manner, generally starting with mild memory impairments that often go unrecognized for years, until a more generalized cognitive decline prompts a diagnosis of dementia. Moreover, mild memory impairments are quite common in the elderly, and not everyone with subtle memory difficulties progresses to develop AD. Current diagnostic techniques have limited usefulness in predicting which individuals with mild memory impairments will progress over time.

Modern imaging techniques offer one potential method of early detection. In addition, by identifying the magnetic resonance imaging (MRI) measurements useful in the discrimination of prodromal AD, one might learn more about which brain regions are the most affected in the earliest stage of AD.

Several studies have demonstrated that structural MRI measurements of the medial temporal lobe, particularly the hippocampal formation and adjacent temporal horn, differentiate patients with mild AD from controls and from patients with other neurological and psychiatric disorders.¹⁻⁹ Medial temporal lobe resection, to date, has been the primary focus of measurement because these brain regions show the greatest damage in end-stage AD.¹⁰⁻¹² Moreover, a memory

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deficit is most commonly the earliest cognitive symptom of the disorder^{13–15} and medial temporal lobe structures are essential for normal memory.¹⁶

Recent neuropathological studies have provided more detailed information about which specific brain regions within this general area are selectively affected in the earliest stage of AD. The initial neuronal lesions of AD (eg, the neurofibrillary tangles and neuritic plaques) appear to occur in the entorhinal cortex, a portion of the anterior parahippocampal gyrus that receives projections from widespread limbic and association areas and gives rise to the perforant pathway, the major cortical excitatory input to the hippocampus itself.^{12,17} Some layers of the entorhinal cortex undergo 40% to 60% neuronal depopulation even in the earliest phase of AD, when memory impairments and patient complaints are subtle and the symptoms do not reach threshold for a diagnosis of AD.¹⁷ The superior temporal sulcus region, a higher order multimodal association area of the cortex, also appears to be affected early in the disease process, although slightly later than the entorhinal cortex¹⁸; neuropathological examination shows a marked volume loss in both the entorhinal cortex and the superior temporal sulcus region.^{17,18} Only rare cases, with an atypical presentation of AD (eg, the spatial presentation or so-called Balint's syndrome) appear not to demonstrate this pattern of neuropathology.¹⁹

The entorhinal cortex and the superior temporal sulcus, along with the volume of the hippocampus, were recently measured by MRI in a group of mild AD patients and controls, demonstrating highly significant differences between the groups.²⁰ Hippocampal measurements can identify a significant, but modest, number of cases of AD before meeting criteria for dementia²¹; but the use of the entorhinal cortex and the superior temporal sulcus as a predictor of AD during its prodromal phase is, to our knowledge, unknown.

The present study was therefore undertaken, to determine whether MRI measures of the entorhinal cortex and the banks of the superior temporal sulcus could identify subjects with AD in the prodromal phase, that is, when they had evidence of deficits in recent memory but did not meet clinical criteria for dementia or AD. It was hypothesized that MRI measurements of brain regions that show the greatest neuronal loss in the earliest stage of AD (eg, the entorhinal cortex and the banks of the superior temporal sulcus) would be more accurate in identifying subjects in the prodromal phase than more general measurements of the medial temporal lobe (eg, the temporal horn) or measurements that tend to reflect the generalized atrophy that is evident during the middle or late phase of the disease (eg, the third ventricle).¹¹ It was also hypothesized that the cingulate gyrus, particularly the caudal portion of the anterior cingulate (Brodmann area 24), would be a sen-

sitive marker of the earliest stage of AD. This prediction was based primarily on functional imaging data, using single-photon emission computed tomography, obtained from prodromal cases of AD.²²

To test these hypotheses, we acquired MRI measures at baseline in 119 subjects, 79 of whom met criteria for "questionable" AD when the study began (ie, they had progressive difficulty with cognitive function, but did not meet clinical research criteria for AD). After 3 years of follow-up, 19 of these individuals had progressed to the point where they met clinical research criteria for probable AD.²³ It was therefore possible to determine whether MRI data obtained at baseline could be used to predict the status of the subjects by the end of the next 3 years.

Subjects and Methods

Subjects

The subjects in the study consisted of 119 elderly individuals, all of whom provided informed consent consistent with institutional guidelines. Of these, 103 were participants in a longitudinal study examining preclinical predictors of AD. The longitudinal study participants had been recruited through the print media (rather than from a clinic or other medical referral source) and underwent a multistage screening procedure. To be included in the study, participants had to be 65 or older, free of significant underlying medical, neurological, or psychiatric illness (based on standard laboratory tests and a clinical evaluation), and meet the Clinical Dementia Rating (CDR)²⁴ criteria described below. (The CDR scale was designed to stage individuals according to their functional ability, from 0 representing normal function to 5 representing the terminal phase of dementia.)

At baseline, these 103 subjects were divided into two groups, based on their functional status. One group consisted of 24 subjects with normal cognition (CDR = 0.0) and one group consisted of 79 subjects with "questionable AD" (CDR = 0.5). The age of the two groups was equivalent (71.8 and 72.0 years, respectively), as was the mean Mini-Mental State Examination²⁵ score (29.2 and 29.1, respectively). After enrollment, these individuals were evaluated annually; the current follow-up rate of the cohort is 99%.

After 3 years of follow-up, the 103 participants in the longitudinal study were divided into three groups, based on their functional status at baseline and at follow-up. Their general characteristics are briefly described below; additional details concerning the sample are described elsewhere.^{22,26}

Sixteen subjects, not part of the longitudinal study, are also included in this report. These were individuals who met clinical research criteria for probable AD²³ when the MRI data were collected. The diagnosis of probable AD was based on the evaluation of a neurologist, psychiatrist, and neuropsychologist; and included standard laboratory tests to exclude medical causes known to produce dementia (eg, SMA-20 [Sequential Multiple Analysis-20 tests], VDRL [Venereal Diseases Research Laboratories (test)], and structural imaging studies). These individuals are included for comparison with the participants in the longitudinal study and with studies that have only examined patients with mild AD.

GROUP 1: NORMALS. This group consisted of subjects who met the CDR criteria for normal cognition (CDR = 0.0) both at baseline and after 3 years of annual follow-up evaluations (n = 24). Thus, these subjects entered the study with normal cognition and remained cognitively intact for 3 years. Their mean age was 71.8 years, and the group consisted of 10 males and 14 females.

GROUP 2: QUESTIONABLES. This group contained subjects who met CDR criteria for questionable AD (CDR = 0.5) at both baseline and follow-up (n = 60). Thus, these subjects entered the study with evidence of mild memory impairments, and after 3 years of follow-up none had progressed to the point where they met clinical criteria for probable AD. The mean age of this group was 71.5, and the group consisted of 24 males and 36 females.

GROUP 3: CONVERTERS. This group consisted of subjects who met CDR criteria for questionable AD (CDR = 0.5) when they were first examined (at baseline), but within 3 years of follow-up their cognitive difficulties had progressed to the point where they met NINCDS/ADRDA (National Institute of Neurological and Communicative Disorders and Stroke/Alzheimer's Disease and Related Disorders Association) criteria for probable AD (n = 19).²³ Thus, like the questionable subjects, the converters had entered the study with evidence of mild memory problems, but unlike the participants in Group 2, those in Group 3 had progressed to meet clinical criteria for AD within the follow-up period. The annual medical, neurological, psychiatric, and laboratory evaluation was augmented, as needed, to exclude other potential causes of cognitive decline in these subjects. It should be noted that the rate of conversion to AD (which in the present sample was approximately 6% per year) appears to be strongly related to the level of functional difficulty at baseline.²⁶ The mean age of the group was 73.5 years, and the group consisted of 9 males and 10 females.

GROUP 4: MILD AD. This group consisted of patients, not part of the longitudinal study, who met NINCDS/ADRDA criteria for probable AD²³ at the time the MRI data were collected (n = 16). These patients were mildly impaired (CDR = 1), as reflected by a mean Mini-Mental State Examination²⁵ score of 24.8. Their mean age was 67.8, and the group consisted of 7 males and 9 females.

MRI Procedures

The MRI data presented here consist of regions of interest (ROIs) derived from three-dimensional T1-weighted gradient echo scans of the brain (repetition time [TR] = 35 msec; echo time [TE] = 5 msec; field of view = 220; flip angle = 45°; slice thickness = 1.5 mm; matrix size = 256 × 256). Each ROI was adjusted by the total intracranial volume on the scan, calculated by a semiautomated computer program.²⁷

Two types of MRI measures were used—a set that was manually drawn and a set that was defined by an automated algorithm. Both types of measures used images that were normalized so that they could be resliced in standard planes. The reformatting procedure required the operator to identify five fixed anatomical landmarks, using a computer mouse:

the anterior commissure (AC), the posterior commissure (PC), and three points along the interhemispheric fissure. Head rotation from front to back was adjusted by a horizontal line that connected the AC and PC points. Head rotation from left to right was adjusted by a sagittal plane that was generated from the three points on the interhemispheric fissure. The coronal plane could then be generated from these two planes of adjustment (because the coronal plane is 90° to the sagittal plane and 90° to the AC-PC line).²⁷

The first set of ROIs consisted of five manually drawn ROIs that were obtained from the MRI images, with the operator “blinded” to the group status of the subject. These ROIs were selected because the neuropathological or functional neuroimaging data, mentioned above, strongly suggested their involvement in the early stage of AD. They included the following: the volume of the entorhinal cortex (EC), the volume of the banks of the superior temporal sulcus (STS) (both of which were calculated on three consecutive coronal slices), and the volume of cingulate gyrus, which was subdivided into three sections, as follows: the rostral portion of the anterior cingulate (CING1), the caudal portion of the anterior cingulate (CING2), and the posterior cingulate (CING3). Figures 1, 2 and 3, and their accompanying legends, present the guidelines the operators followed in measuring these ROIs.

To determine the reliability with which the ROIs were measured, we calculated the percent agreement and interrater reliability for two independent operators, blinded to subject status. The mean difference between the raters was modest; for the EC, it was 0.33 (95% confidence interval [CI], -0.35–1.0), and for superior temporal sulcus, it was 0.10 (CI, -1.08–0.90). The cingulate was divided into three contiguous regions, as described above, and the mean differences between the raters for these regions were 0.085 (CI, 0.58–0.11), 0.17 (CI, 0.12–0.22), and 0.11 (CI, 0.07–0.15). Interrater reliabilities were also high (EC_{mean}: $r = 0.96$; STS_{mean}: $r = 0.92$; CING1_{mean}: $r = 0.99$; CING2_{mean}: $r = 0.99$; CING3_{mean}: $r = 0.99$). The reliability of the program used to calculate the volumes of each region, once it has been manually outlined, has been previously established.²⁶

The second set of ROIs consisted of six automated measures of cerebrospinal fluid spaces that either provided a reflection of the integrity of the medial temporal lobe (eg, the temporal horn) or assessed generalized atrophy that is evident in the middle and late stage of disease (eg, the third ventricle). These latter ROIs, which have been previously used in other studies,²⁷ were as follows: the temporal horns, the suprasellar cisterns, the third ventricle, the lateral ventricles, the sylvian fissures, and the interhemispheric fissure. Some of these measures, particularly the temporal horn, have previously been shown to be sensitive to the presence of disease in mild AD cases.²⁷ The semiautomated procedures used to calculate the volume of the ROIs are described in detail elsewhere.²⁸

Genetic Assessment

The apolipoprotein E (ApoE) gene was also examined in the participants because the E4 allele of this gene is overrepresented in AD patients compared with the general popula-

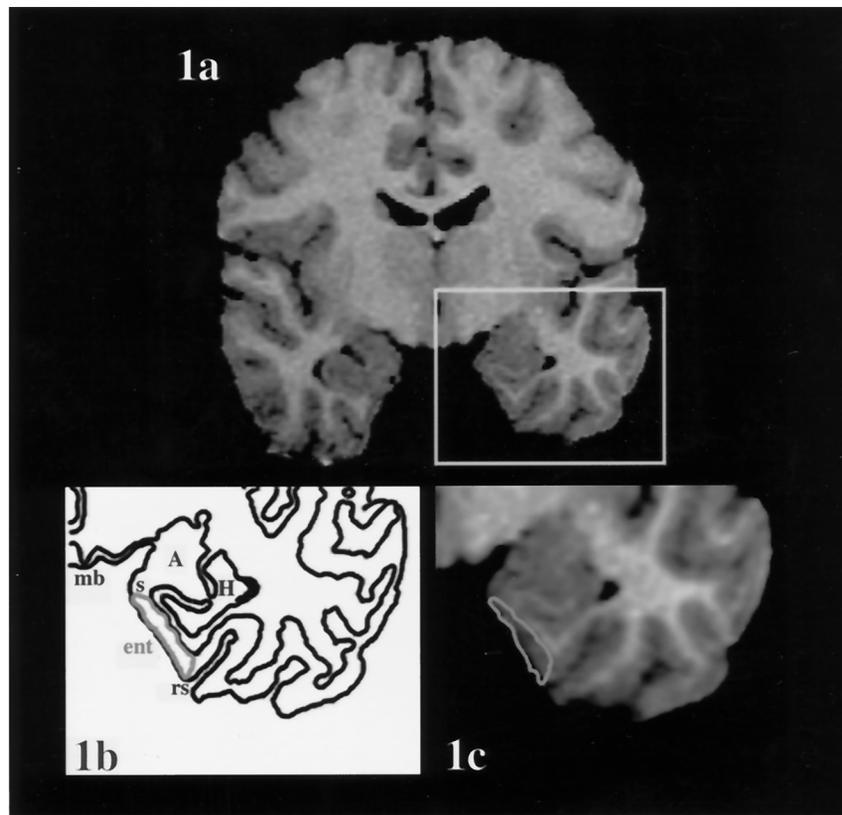


Fig 1. (a) A coronal magnetic resonance imaging (MRI) scan, at the level on which the entorhinal cortex was measured. The area outlined represents the portion of the scan used to orient the operator to the landmarks of the entorhinal cortex. A box has been placed over the region of interest in one hemisphere. (b) A diagram of the entorhinal cortex (ent) and adjoining medial temporal lobe regions, including the rhinal sulcus (rs), the hippocampus (H), the amygdala (A), the subiculum (S), and the mammillary bodies (mb). (c) The region of the entorhinal cortex measured in the present study, as delineated on the MRI scan of a control subject. The entorhinal cortex is located inframedially on the surface of the brain. It is fused with the subiculum of the hippocampus medially and extends to the rhinal sulcus laterally. The entorhinal cortex was outlined on three consecutive normalized coronal images centered at the level of the mammillary bodies. The outline of the region began at the angle formed by the junction of the rhinal sulcus and the surface of the brain (the posterior end of this region may be continuous with the collateral sulcus in some subjects). The outline then transected this angle, cutting across the gray matter to the level of the white matter. Then the edge of the white matter was followed to the inferior surface of the hippocampus. The outline then followed the surface of the brain back to the starting point. (This protocol was based on the fact that the most anterior and posterior edges of entorhinal cortex and the banks of the superior temporal sulcus are difficult to define with reliability. Moreover, unbiased stereological data from human brain tissue indicate that the neuron counts within a single section of these brain regions are excellent predictors of total volume.^{17,18})

tion,²⁹ and is now widely recognized as a risk factor for AD. We therefore sought to determine whether ApoE status was useful as a predictor of which individuals were likely to “convert” to AD over time, either alone or in combination with the MRI measures.

Data Analysis

The MRI data were analyzed by Student’s *t* test and discriminant function analysis.³⁰ Sensitivities and specificities³¹ and confidence intervals were also calculated, where appropriate. The MRI data presented here were obtained at baseline, but the groups for the individuals in the longitudinal study are based on subject status after three annual follow-up visits.

Results

Comparison of Normals, Questionables, and Converters

The mean values and standard deviations of the MRI measures in the Normals, Questionables, and Converters were examined first. In preliminary analyses performed to check the appropriateness of summing the measures from the left and the right hemispheres, significant differences in volume on the basis of laterality were apparent in the measures related to the cingulate gyrus. Thus, these measures were expressed as the difference between the volumes in the right and the left hemispheres. Whereas, all other measures were ex-

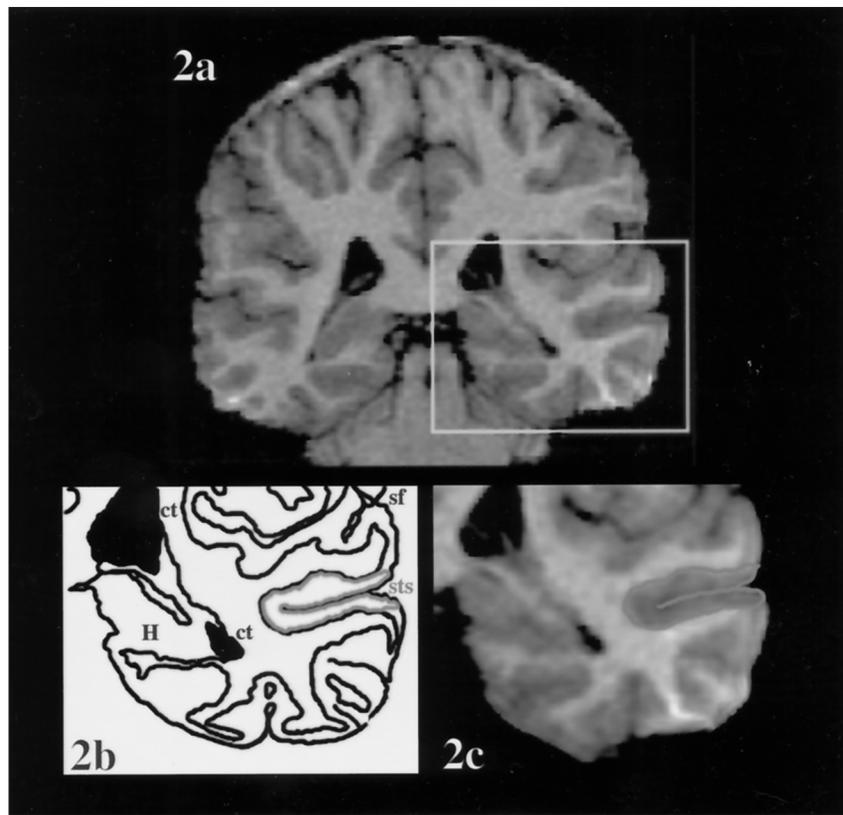


Fig 2. (a) A coronal magnetic resonance imaging (MRI) scan, at the level on which the superior temporal sulcus region was measured. The area outlined represents the portion of the scan used to orient the operator to the landmarks of the banks of the superior temporal sulcus. A box has been placed over the region of interest in one hemisphere. (The apparent asymmetry of the angle of the left sylvian fissure is the result of the angle of cut). (b) A diagram of the superior temporal sulcus (sts) region and adjoining temporal lobe areas, including the collateral trigone (ct), the sylvian fissure (sf), and the hippocampus (H). (c) The region of the banks of the superior temporal sulcus measured in the present study, as delineated on the MRI scan of a control subject. The gray matter of the upper and lower banks of the superior temporal sulcus was measured on three consecutive normalized coronal slices centered on the level of the rostral tip of the collateral trigone. The outline of the region began at the medial most point of the lower bank of the sulcus and continued along the edge of the white matter. The profile of the sulcus was then projected across the gray matter to the surface of the brain. The outline then continued along the edge of the brain to the depth of the sulcus. This process was then repeated for the upper bank of the sulcus.

pressed as the mean value of the sum of the right and left hemisphere.

The 11 MRI measures were then examined individually, using t tests, to see if they differed among groups. To accomplish this, Groups 1, 2, and 3 were compared pairwise (ie, Normals vs Converters, Normals vs Questionables, and Questionables vs Converters). The results of these individual t tests are presented in the Table. They are intended as descriptive statistics only, to highlight the major differences among the groups. Three of the 11 variables were significantly different in two or more of the group comparisons: the entorhinal cortex, the banks of the superior temporal sulcus, and the caudal portion of the anterior cingulate. An additional four variables were significantly different in one of the group comparisons. For example, the volume of the entorhinal cortex was 36% smaller in the

Converters (Group 3) in comparison with the Normals (Group 1) ($t = 5.65$; $p < 0.0001$). The superior temporal sulcus region was 24% smaller in the Converters in comparison with the Normals ($t = 5.49$; $p < 0.001$). The caudal portion of the anterior cingulate was also considerably smaller in the Converters in comparison with the Normals ($t = 2.09$; $p < 0.04$).

To examine the manner in which the 11 ROIs, in combination, best differentiated the groups, several discriminant function analyses were then performed. This method of analysis was used to avoid the selection of variables that appeared significantly different by t test alone, thus, capitalizing on chance. The first discriminant analysis was conducted to determine whether the 11 ROIs, together, significantly differentiated the Normals, Questionables, and Converters. Of the 11 ROIs, five were manually drawn, and six were identified by

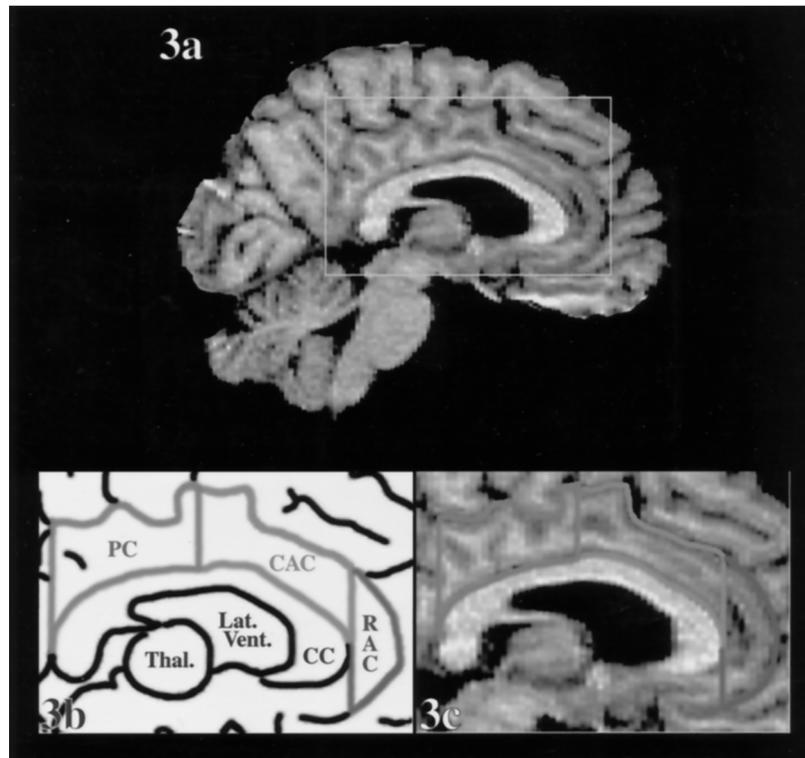


Fig 3. (a) A sagittal reconstruction of a coronally acquired magnetic resonance imaging (MRI) scan, at the level on which the cingulate gyrus was measured. The area outlined represents the portion of the scan used to orient the operator to the landmarks of the cingulate. A box has been placed over the region of interest in one hemisphere. (b) A diagram of the cingulate gyrus divided into the rostral portion of the anterior cingulate (RAC), the caudal portion of the anterior cingulate (CAC), and the posterior cingulate (PC). Adjoining landmarks include the corpus callosum (CC), the lateral ventricle (Lat. Vent.), and the thalamus (Thal.). (c) The region of the cingulate gyrus measured in the present study, as delineated on the MRI scan of a control subject. The cingulate gyrus was measured on consecutive normalized coronal slices from the rostral most extent to the marginal ramus of the cingulate sulcus. First, the sagittal view was used to demarcate the cingulate sulcus in each hemisphere. This information was then transferred to the coronal image. Second, the cingulate gyrus was outlined; this began at the lateral most point of the corpus callosum sulcus, continued along the surface of the callosum medially to the interhemispheric fissure, then dorsally to the cingulate sulcus, and then laterally to the lateral most extent of the cingulate sulcus; to surround the portion of the cingulate gyrus to be measured, a straight line was then drawn connecting the lateral most extent of the cingulate and the corpus callosum sulci. Third, the outlined region was then divided into three sections by two lines: the rostral portion of the anterior cingulate (RAC) was identified by drawing a line in the coronal plane at the tip of the corpus callosum, and the caudal portion of the anterior cingulate (CAC) was differentiated from the posterior cingulate (PC) by drawing a line through the cingulate gyrus in the coronal plane at the level of the mammillary bodies (white matter tracts representing the end of the fornix are used to mark the middle of the mammillary bodies in this plane).

semiautomated procedures that quantified cerebrospinal fluid-filled spaces, as described above. The discriminant function analysis included a total of 14 variables: each of the 11 ROIs and a measure of age, sex, and intracranial volume for each subject. The latter three variables were included to adjust for any possible differences between the groups based on these variables. This overall discriminant function was highly statistically significant ($\chi^2 = 86.03$; $p < 0.00001$). It should be noted that the statistical power of such an analysis is based on the overall sample size, rather than the size of individual groups.³⁰ Moreover, the homogeneity of the group variances was examined and found to be satisfactory (assuring that the small size of some of the groups did not compromise the analysis).

We then performed a stepwise discriminant function analysis, to select the MRI variables that best differentiated the Normals, Questionables, and Converters. The term *best* refers here to the process by which each variable enters the algorithm in the order by which it improves the significance of the overall function. In the stepwise analysis, age, sex, and intracranial volume were entered at the first step, and then the algorithm of the discriminant function selected the variables that, when combined, best differentiated the groups from one another. Three of the 11 variables were selected as the best discriminators between the Normals, Questionables, and Converters ($\chi^2 = 72.91$; $p < 0.00001$). They were (1) the entorhinal cortex, (2) the banks of the superior temporal sulcus, (3) the cau-

Table. MRI Measures in Normals, Questionables, and Converters ($ml\ 10^{-2}$)

Region of Interest	Normals	Questionables	Converters
n	24	60	19
Age	71.6	71.5	72.8
Entorhinal cortex	4.30 ^{a,b}	2.90 ^b	2.85 ^a
Temporal sulcus	5.32 ^{a,b}	4.76 ^{b,c}	4.15 ^{a,c}
Anterior cingulate 1	-1.30	0.20	-2.93
Anterior cingulate 2	5.14 ^a	5.73 ^c	-2.45 ^{a,c}
Posterior cingulate	-4.00 ^b	2.36 ^b	1.09
Sylvian fissure	14.7	14.0	16.0
Suprasellar cistern	42.3	40.3 ^c	44.7 ^c
Third ventricle	4.69	4.39 ^c	5.34 ^c
Lateral ventricle	91.6	93.8	113.5
Temporal horn	2.12	2.35	2.58
Interhemispheric fissure	54.3	48.5	49.1

^aSignificant difference between normals and converters.

^bSignificant difference between normals and questionables.

^cSignificant difference between questionables and converters.

dal portion of the anterior cingulate (CING2), referred to below as the anterior cingulate. The covariates entered at the first step (age, sex, and intracranial volume) were not significant.

Three separate discriminant functions were then performed in which these three variables were entered and the three groups were compared pairwise. Age, sex, and intracranial volume were entered at the first step in each of these analyses, despite that the overall discriminant function indicated that they did not significantly contribute to the discrimination. This was done to assure that any subtle, though nonsignificant, effect of these variables was eliminated as much as possible. It should be noted that the covariate method of adjustment for intracranial volume, which was used here, has been recommended^{31,32} because it permits a correction to occur only when there is a significant correlation between intracranial volume and the volume of a particular ROI. The results of the stepwise discriminant functions can be summarized briefly as follows:

NORMALS VS CONVERTERS. The discriminant function that compared the Normals and the Converters was highly significant ($\chi^2 = 38.48$; $p < 0.00001$). Two of the three variables were significant contributors to the discrimination: the volume of the entorhinal cortex ($p < 0.0001$) and the volume of the banks of the superior temporal sulcus ($p < 0.0009$). The overall accuracy of the discrimination of the Normals versus the Converters was 93% (sensitivity = 95%; specificity = 90%).

NORMALS VS QUESTIONABLES. The discriminant function that compared the Normals and the Questionables was highly significant ($\chi^2 = 50.34$; $p < 0.00001$). The same two MRI variables that were significant in the

differentiation of the Normals and Converters contributed to the discrimination of the Normals and the Questionables, that is, the volume of entorhinal cortex ($p < 0.00001$) and the volume of banks of the superior temporal sulcus ($p < 0.015$). The overall accuracy of the discrimination of the Normals and Questionables was 85% (sensitivity = 82%; specificity = 96%).

QUESTIONABLES VS CONVERTERS. The discriminant function that compared the Questionables and the Converters was also highly significant ($\chi^2 = 18.57$; $p < 0.005$). However, in this comparison, a different set of two variables contributed to the discrimination. The volume of the banks of the superior temporal sulcus ($p < 0.003$) and the volume of anterior cingulate ($p < 0.009$) contributed significantly to the discrimination. The accuracy of the discrimination between the Questionables and the Converters was 75% (sensitivity = 68%; specificity = 48%).

Discriminant function analyses were then used to examine the contribution of ApoE genotype, alone or in combination with the MRI variables, to the discrimination of the Normals, Questionables, and Converters. ApoE status was coded as E4 present or absent. When ApoE status alone was used to differentiate these three groups, the discriminant function was not statistically significant. ApoE status was then added to each of the discriminant function analyses in which the MRI data had been examined. Addition of ApoE status did not significantly improve the discrimination of the three groups above that of the MRI data alone.

Normals vs Mild AD Patients

The MRI data of the Normals and the mild AD patients were then compared by a discriminant function in which a measure of the entorhinal cortex, the banks of the superior temporal sulcus, and the caudal portion of the anterior cingulate were included. This analysis was highly significant ($\chi^2 = 80.7$; $p < 0.00001$). The variables that contributed to the discrimination were the entorhinal cortex ($p < 0.00001$) and the banks of the superior temporal sulcus ($p < 0.00001$). The accuracy of discrimination between the Normals and the mild AD patients was 100%. The discrimination accuracy between the mild AD patients and the Converters was 82.4%, and for the mild AD patients versus the Questionables it was 94.7%.

Discussion

In this study, we asked whether MRI measures at baseline could discriminate between three groups of individuals—subjects with mild memory impairments who convert to AD within 3 years, individuals with mild memory problems who do not progress sufficiently in 3 years to warrant a clinical diagnosis of AD, and normal age-related controls. Our data demonstrate that

MRI measures of the entorhinal cortex, the banks of the superior temporal sulcus, and the caudal portion of the anterior cingulate are useful in discriminating these groups. The greatest accuracy is found in the comparison of the Normals with the Converters, which is 93%. The comparison of the Normals and Questionables and the comparison of the Questionables and the Converters is lower but still robust (85% and 75%, respectively). This is perhaps not surprising because based on previous experience, it is anticipated that over subsequent years of follow-up more questionable subjects will progress to the point where they meet criteria for AD. Thus, some of the subjects who are currently categorized as Questionable are in the prodromal phase of AD, making differentiation from the individuals who have already converted to AD particularly challenging. It will be important to follow the individuals who remained questionable after 3 years of evaluation, to determine the earliest point at which MRI measures can be used to discriminate individuals who are destined to later develop AD. It should also be noted that patients with mild AD could be differentiated from controls with 100% accuracy, using these MRI measures. This is consistent with a previous study that used comparable measures to compare the same subject groups.²⁰ It is also consistent with data showing that subjects with a rating of questionable AD have smaller hippocampal volumes than patients with mild AD.³² Taken as a whole, these findings are of potential clinical importance.

These findings are also informative on a theoretical level. They confirm the involvement of the entorhinal cortex and the banks of the superior temporal sulcus in prodromal AD, and suggest that these MRI measures are likely to be a reflection of known underlying AD neuropathology. The degree of difference in the measure of the entorhinal cortex between the Normals and the Converters (37%) is, in fact, remarkably similar to the amount of neuronal loss measured in neuropathological studies of this region in similar subjects (32%).^{12,17} These data also support the hypothesis that MRI measures of brain regions that demonstrate neuropathological changes in the early stage of AD are better at differentiating patients with AD who are in the prodromal phase than measurements that reflect alterations that develop later in the course of the disease (eg, atrophy affecting the third ventricle).

The current data also confirm the involvement of the caudal portion of the anterior cingulate in an early stage of AD. This brain region is known to develop severe neuronal loss in AD,³³ but the stage at which this occurs is not yet known. The present findings suggest that the neuronal loss begins early in the disease and may, in part, be responsible for the cognitive impairments seen at the early stage of AD. The entorhinal cortex is part of a memory-related neural system in the

brain,³⁴ which is consistent with the fact that most patients with AD present with a progressive memory impairment, as noted above. The superior temporal sulcus region is a multimodal association area, and it seems necessary for holding information during a delay and has thus been hypothesized to play a role in memory or the attentional capacities necessary for normal memory.^{35,36} The anterior cingulate is strongly and reciprocally connected with memory-related structures, including the entorhinal cortex.^{33,37} However, it is also strongly and reciprocally connected with the prefrontal cortex. It has been hypothesized that the anterior cingulate plays a major role in executive function abilities, primarily through its reciprocal connections with the prefrontal cortex.³⁸ The present MRI data suggest that the executive function impairment observed in mild AD patients³⁹ may be related to neuronal loss in the caudal portion of the anterior cingulate.

These MRI data also suggest the hierarchical fashion in which the pathology of AD may affect the brain during its prodromal phase, because it seems likely that the brain regions that best differentiate the Normals from the other subject groups are those that are affected first. The entorhinal cortex and the banks of the superior temporal sulcus were selected as the best discriminators when the comparison was between the Normals and either of the other two groups (the Converters or the Questionables). Neuropathological data suggest that the entorhinal cortex is affected before the superior temporal sulcus.^{17,18} In contrast, one of the brain regions selected for the discrimination between the Questionables and the Converters was the caudal portion of the anterior cingulate, which suggests that its involvement may correspond to the point at which the clinical symptoms progress to the point where an individual meets criteria for probable AD. Alternative explanations for these findings are that there is a moderate reduction in size of the entorhinal cortex in persons with normal memory problems or that some normal individuals have lower neuronal reserve in the banks of the superior temporal sulcus and the caudal portion of the anterior cingulate. Because neuropathological data suggest that neuron number is remarkably consistent in the entorhinal cortex and the banks of the superior temporal sulcus among normal individuals who are 60 to 90 years of age,^{17,18} the latter two possibilities seem less likely.

The negative findings regarding ApoE status as a predictor of conversion to AD are consistent with several recent studies, including a large multicenter study that examined the use of ApoE genotype as a diagnostic test for AD.⁴⁰ In this study, the sensitivity and specificity of ApoE alone in discriminating autopsy-proven cases of AD from those of cases with other forms of dementia (eg, Pick's disease and cerebrovascular disease) was 65% and 68%, respectively. In the

present study, the accuracy of discriminating controls from converters was not significant.

There is still much to be learned about the natural history of individuals in the prodromal phase of AD. In the present study, volume loss in the entorhinal cortex, the superior temporal sulcus, and the caudal portion of the anterior cingulate provided highly accurate information that concerned those who would develop clinical changes consistent with probable AD during the next 3 years from those who would remain cognitively normal. The discrimination of subjects with memory problems who would develop AD within 3 years from those who would remain questionable was more difficult, but the discriminant function still correctly predicted 75% of these individuals. Improvement in this prediction rate may well occur if these MRI measures can be supplemented with additional measures based on either MRI or other relevant domains.

Identification of individuals in a "preclinical" or prodromal phase will be critical to test existing therapies for their ability to alter the course of the illness and for the development of novel strategies to prevent or delay dementia. To perform this work in an efficient and cost-effective manner, it will be particularly important to develop methods for the identification of populations of individuals "enriched" with those highly likely to develop AD within a short period of time. Structural MRI can potentially be used to characterize the natural history of patients with AD, even in the earliest stages of disease, as well as to act as a quantitative and biologically meaningful end point in therapeutic trials.

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