Age transformation of combined hippocampus and amygdala volume improves diagnostic accuracy in Alzheimer’s disease

Harald Hampel a,*, Stefan J. Teipel a, Wolfram Bayer a, Gene E. Alexander b, Robert Schwarz a, Mark B. Schapiro c, Stanley I. Rapoport d, Hans-Jürgen Möller a

a Dementia Research and Neuroimaging Section, Memory Clinic, Department of Psychiatry, Ludwig-Maximilian University, Nussbaumstr. 7, 80336 Munich, Germany
b Arizona Alzheimer’s Research Center and Department of Psychology, Arizona State University, Tempe, AZ, USA
c Department of Pediatric Neurology, Children Hospital Medical Center, Cincinnati, OH, USA
d Brain Physiology and Metabolism Section, National Institute on Aging, National Institutes of Health, Bethesda, MD, USA

Received 26 June 2001; received in revised form 20 September 2001; accepted 5 November 2001

Abstract

Objective: The specificity of magnetic resonance imaging (MRI)-based hippocampal measurements in detecting Alzheimer’s disease (AD) pathology is reduced by an age-related reduction of the hippocampus volume. We propose an adjustment for this age effect to increase the diagnostic accuracy of hippocampal volumes in AD.

Method: Using an orthogonal rotational transformation of the coordinate system, values of MRI-determined volumes of hippocampus–amygdala formation (HAF) were transformed according to the age effect in 27 AD patients and 28 age- and sex-matched healthy control subjects.

Results: The age transformation increased the diagnostic accuracy of HAF volumes in the study sample and in an independent sample from the literature. The age-transformed HAF volume predicted AD in a subject with mild cognitive impairment (MCI) with later biopsy-confirmed AD.

Conclusion: Age transformation may provide an easily applicable method to increase the clinical diagnostic accuracy of hippocampal measurements by considering the effect of aging on hippocampus volume.

Keywords: Alzheimer’s disease; Diagnosis; Hippocampus; Magnetic resonance imaging; Discriminant analysis; ROC analysis; Age-transformation

1. Introduction

Atrophy of the hippocampus and amygdala has consistently been shown in Alzheimer’s disease (AD) even in preclinical stages [1–3]. There remains an overlap between AD patients and healthy elderly controls. Many studies have reported age-related reductions of the hippocampus in healthy subjects and AD patients [1,4–7]. Reduction of hippocampus volume with age may therefore reduce our ability to use this volume to distinguish AD patients from healthy elderly controls. In a previous study, age-associated normal percentiles of hippocampus volume have been established in healthy elderly subjects [8]. It may however be useful to determine age-specific thresholds of hippocampus atrophy that can be applied to a single subject, even if the subject is at the lower or upper limit of the age range of the comparison group, and that can be applied within a given sample independently from the measurement protocol of a reference study.

To investigate this possibility, we made an age adjustment, using an orthogonal rotational transformation, of hippocampal volumetric data from magnetic resonance imaging (MRI) examinations of 27 clinically diagnosed AD patients and 28 healthy age- and sex-matched control subjects.

2. Patients and methods

2.1. Patient selection

We investigated 27 patients (mean age: 69.2 ± 9.3 years; 13 females and 14 males) with the clinical diagnosis of...
probable AD according to the NINCDS-ADRDA criteria [9]. This group encompassed one patient with the clinical diagnosis of mild cognitive impairment (MCI) [10] with subsequent progressive cognitive decline, who was later confirmed by biopsy to have AD. For comparison, 28 healthy volunteers (mean age: 68.2 ± 8.5; 15 females and 13 males) were selected. Groups were matched in age and gender.

Cognitive impairment in the AD patients was assessed using the Mini-Mental State Examination (MMSE) [11]. Nine patients showed severe (MMSE < 10), nine patients moderate (10 ≤ MMSE < 20) and nine patients mild (MMSE ≥ 20) dementia. The mean score of the MMSE was 13.5 ± 8.7 (ranging between 0 and 27) in AD patients and 29.8 ± 0.4 (ranging between 29 and 30) in healthy control subjects.

All subjects had been screened to exclude significant medical comorbidity including hypertension according to a previously outlined protocol [12].

The protocol of the study was approved by the National Institute on Neurological Disease and Stroke’s Institutional Review Board. All subjects or the holders of their Durable Power of Attorney signed consent forms to undergo MRI and neuropsychological assessment for clinical investigation and research.

2.2. MRI

An axially oriented double-echo sequence (slice thickness: 6 mm, TR/TE 2000/80 and 2000/20, respectively) and a sagittally oriented T1-weighted volumetric sequence (slice thickness: 2 mm, in-plane resolution: 1 × 1 mm, TR/TE 20/6, flip angle: 45°) were obtained on a 0.5-T MR tomograph (Picker Instruments, Cleveland, OH).

2.3. Volume measurements

The volumes of the right and left hippocampus–amygdala formation (HAF) were measured by one investigator (WB) blinded to clinical data of the subjects using the sagittally oriented T1-weighted volumetric MRI sequence and ANALYZE™ software (Biomedical Imaging Resource, Rochester, MN) on an SGI (Silicon Graphics, Palo Alto, CA) workstation.

First, all sagittal slices were manually aligned perpendicular to the AC–PC plane [13] to reduce measurement variability from different head positions. The rostral border of the hippocampus was identified on the first sagittal slice that showed the hippocampal grey matter in its whole extension, referring to a hippocampus atlas [14]. In this slice, the occipital border of the hippocampus also was identified at the transition from the hippocampal grey matter to the fibers of the fornix. Measurement of the hippocampus then started with the first orthogonally reconstructed coronal plane, cutting its anterior border on the sagittal view. Because image resolution did not always allow reliable discrimination of the head of the hippocampus from the amygdala grey matter, the grey matter from the amygdala was measured together with the hippocampus, view from inferior–anterior.

Fig. 1. Measurement of hippocampus and amygdala formation. (a) Determination of the anterior and posterior border of hippocampus in the first sagittal slice showing the hippocampus formation in its full extension. (b) Delineation of hippocampal grey matter in the coronal slice. (c) Reconstructed left and right hippocampus, view from inferior–anterior.
campal grey matter in each scan. This protocol excluded the most anterior portion of the amygdala. The hippocampus–amygdala formation (HAF) thus comprised the most posterior portion of the amygdala and the hippocampus proper (Fig. 1).

The number of pixels was summed over all slices and multiplied by the voxel size (1 × 1 mm in plane by 2 mm transverse) to estimate the volume of HAF in cubic millimeters (mm³). The intra-class correlation coefficient (ICC) was 0.98 for two independent raters measuring HAF volumes in 10 scans and 0.99 for one rater measuring HAF volumes twice in 10 randomly chosen scans. The ICC does not only take into account correlations between the measures but is also sensitive towards systematic differences in absolute values of measures between the ratings [15]. For two raters, the ICC can range between 0 and 1, with 1 indicating perfect agreement. The coefficient of variation for the two raters was 1.8% for right HAF and 1.9% for left HAF. For one investigator measuring HAF twice in 10 randomly chosen subjects, the coefficient of variation was 2.0% for the right and 2.0% ± 0.9% for the left HAF volumes.

Fig. 3. Mathematical derivation of age transformation. The coordinates \((X,Y)\) of point \(P\) in the orthogonal coordinate system \(x–y\) are transformed into the coordinates \((X',Y')\) in the system \(x'–y'\), with axis \(x'\) parallel to \(R\). Given are \(X,Y\) and \(m\) (the slope of the regression line \(R\)).

\[(I)\] \(\cos(c) = \frac{Y'}{d} \Rightarrow Y' = d\cos(c)\)

\[(II)\] \(d = \sqrt{(X^2 + Y^2)}\)

\[(III)\] \(c = \frac{\pi}{2} - |a| - b = \frac{\pi}{2} + a - b\), for \(a < 0\)

\[(IV)\] \(a = \arctan(m)\)

\[(V)\] \(\tan(b) = \frac{Y}{X} \Rightarrow b = \arctan\left(\frac{Y}{X}\right)\)

\[(II), (IV)\) and \((V)\) in \((I)\) : \(Y' = \sqrt{(X^2 + Y^2)}\cos\left(\frac{\pi}{2} - \arctan\left(\frac{Y}{X}\right) + \arctan(m)\right)\)

\(\Rightarrow Y' = \sqrt{(X^2 + Y^2)}\sin\left(\arctan\left(\frac{Y}{X}\right) - \arctan(m)\right)\).
2.4. Statistics

Age-transformed values for HAF (Y′) were calculated according to the following equation:

\[ Y' = \frac{Y}{X} \sin(\arctan(\frac{Y}{X}) - \arctan(m)) , \]

where \( Y \) is equal to the HAF volume, \( X \) is equal to the age and \( m \) is equal to the slope of the regression line (Fig. 2). The equation describes an orthogonal rotation of the y- and \( x \)-axes of the coordinate system into axes \( y' \) and \( x' \) (Fig. 2a). The axes are rotated until the new axis \( x' \) is parallel to the regression line of age on HAF volume in the AD group. Fig. 3 details the trigonometric derivation.

For \( m \) being negative and \( Y/X \) being positive, \( \arctan(Y/X) - \arctan(m) \) is always positive. Because the sine function \( \sin(z) \) is positive and constantly increasing for all \( z > 0 \), age-transformed values are positive and increasing with HAF volume when age is kept constant.

A cut-off value to discriminate the AD patients and controls was derived from a receiver-operating characteristic (ROC) curve analysis constrained to the maximize sum of sensitivity and specificity.

3. Results

The mean bilateral HAF volume was 2910.8 mm\(^3\) ± 705.0 mm\(^3\) in AD patients and 4629.4 mm\(^3\) ± 776.3 mm\(^3\) in healthy controls. The HAF volumes were significantly reduced by 37% in AD patients (\( p < 0.001 \)) compared with the controls. The HAF volume was significantly correlated with age in AD patients (\( r = -0.42, p < 0.03 \)) and in control subjects (\( r = -0.61, p < 0.001 \)) (Fig. 1a). In an analysis of covariance, there was no significant effect of gender or of a gender by age interaction on HAF volumes (\( p = 0.38 \)). The statistical significance of these results remained unchanged after normalization of HAF volume to the total intracranial volume (the ratio of HAF volume divided by the total intracranial volume).

ROC analysis of absolute HAF volumes revealed sensitivity and specificity levels of 89%. Taking into account the effect of age on HAF volume, the cut-off derived from the ROC analysis of the transformed values resulted in 96% sensitivity and 89% specificity. For HAF volumes normalized to the total intracranial volume, age transformation raised the sensitivity from 92.6% to 96.3% at 96.4% specificity.

We also compared a subgroup of 9 patients with mild AD (MMSE>19) and 28 control subjects. Before the age adjustment, the ROC analysis of HAF volume revealed sensitivity and specificity levels of 89%. After the age adjustment of the absolute HAF data, the ROC analysis resulted in a sensitivity of 100% and a specificity of 86%. For normalized HAF volumes, the sensitivity was raised from 88.9% to 100% at 96.4% specificity in mild dementia.

To investigate the effect of age transformation in an independent sample, we reanalyzed the data presented by Jack et al. [6], who studied 20 patients with the clinical diagnosis of probable AD in mild to moderate stages of the disease and 22 healthy age- and sex-matched control subjects. Hippocampus volumes and corresponding ages were derived from a scatterplot using graph paper. Mean and standard deviations of the derived data were in good agreement with the values given in the paper for the hippocampus (1.98 ± 0.5 and 2.8 ± 0.3 compared to 2.0 ± 0.5 and 2.8 ± 0.3 for AD patients and controls, respectively) and age (72.6 ± 10.5 and 75.9 ± 11.5 compared to 73.5 ± 10.6 and 76.3 ± 11.3 for AD patients and controls, respectively). Areas under the ROC curve were 0.91 before and 0.98 after the age adjustment. Sensitivity and specificity rates derived from the ROC curves under the constraint to maximize the sum of sensitivity and specificity were 85% and 82% before and 90% and 91% after the age adjustment.

To illustrate the potential application of age transformation, we studied the effect of age transformation on the group classification of one patient with MCI and early onset of cognitive impairment (55 years). This patient had a mean HAF volume of 4119 mm\(^3\) and an age-transformed value of HAF of 212. The age transformation and ROC analysis here were based on the sample after exclusion of this subject. Thresholds for HAF and age-transformed HAF determined from the ROC analysis were 3770 mm\(^3\) and 218. Thus, this subject, later confirmed as AD, was within the normal range for HAF (the HAF volume is larger than the threshold) but within the pathological range for the age-transformed HAF (the age-transformed HAF is smaller than the threshold).

Similarly, we assessed the oldest subject of the control group (84 years) having an HAF volume of 3596 mm\(^3\) and a transformed volume of 197. The thresholds determined from the ROC analysis after exclusion of this subject were 3951 mm\(^3\) and 189, respectively. Thus, after age transformation, this subject was assigned to the correct group based on HAF volume.

4. Discussion

In the present study, we report the extent to which hippocampus–amygdala formation (HAF) volumes can discriminate AD patients from healthy elderly controls. Using an orthogonal rotation of the coordinate system according to the age effect in the AD group, the sensitivity of HAF volumes was raised from 89% to 96% in early to severe AD at a sensitivity level of 89%, and at a sensitivity of 100%, the specificity was raised in early AD from 64% to 86%. Reanalysis of an independent sample [6] revealed sensitivity and specificity of 85% and 82% before and 90% and 91% after age adjustment, supporting the findings of our study.

The clinical application of this technique will particularly be helpful in the diagnosis of subjects at the lower end...
(increasing sensitivity) or at upper end (increasing specificity) of the age range of the reference groups. This potential application has been exemplified in two subjects. The HAF volume of the young subject with minimal cognitive impairment, being in the normal range, was correctly assigned to the pathological range after the age transformation. Alternatively, a comparison group in the age range of the young subject could have been used to obtain the same result. On the other end of the age range, age transformation identified the HAF volume of the oldest healthy control subject correctly as normal. Both these examples encourage a further study on the usefulness of age transformation in groups of subjects with relatively low or high age compared to reference groups. Furthermore, because the age adjustment is based on the numerical values of a given study, it does not require the identical reproduction of the imaging and measurement protocol of a reference study.

One approach to increase the sensitivity of structural MRI for diagnosing AD is to combine the assessments of several predictor variables in a discriminant analysis [16]. Discriminant analysis puts several constraints on data distribution and the covariance matrix of predictor variables. This analysis performs worse when the assumption of independence among the predictor variables is violated [17]. The significant correlation between age and HAF volumes violates the prerequisite for the valid interpretation of a discriminant function with age and HAF volumes as predictor variables for the diagnosis. In contrast, the approach presented here relies in a hypothesis-driven manner on the correlation between a volumetric measure and a subject characteristic such as age. Taking into account the effect of head size through a single ratio of HAF volume divided by the total intracranial volume increased the accuracy of group discrimination. Therefore, the combined evaluation of both confounding variables, age and intracranial volume, may be useful to increase the diagnostic accuracy of a single measure.

The easily applicable technique presented here to increase the diagnostic accuracy for a single subject by taking into account the independent effect of age on hippocampus and amygdala volume may be particularly beneficial in clinical settings. Additionally, the technique, if validated in independent samples, may be applied to other markers, which are affected by disease-independent individual characteristics.

Acknowledgements

We thank Dr. Arun Bokde, PhD for the critical review of the manuscript.

Part of this work was supported by a grant of Eisai (Frankfurt) and Pfizer (Karlsruhe), Germany, to H.H. and S.J.T and by a grant of the Medical Faculty of the Ludwig-Maximilian University, Munich, Germany, to S.J.T.

Part of the presented data originated from the doctoral thesis of Wolfram Bayer (Ludwig-Maximilian University, Munich, Germany; in preparation).

References