ELSEVIER

Neurobiology of Aging 25 (2004) 913–924

NEUROBIOLOGY OF AGING

www.elsevier.com/locate/neuaging

Structural correlates of mild cognitive impairment

Henrike Wolf [a,d,*], Anke Hensel [a], Frithjof Kruggel [b], Steffi G. Riedel-Heller [a], Thomas Arendt [c], Lars-Olof Wahlund [d], Hermann-Josef Gertz [a]

[a] *Department of Psychiatry, University of Leipzig, Leipzig, Germany*
[b] *Max-Planck-Institute of Cognitive Neuroscience, Leipzig, Germany*
[c] *Paul-Flechsig Institute for Brain Research, University of Leipzig, Leipzig, Germany*
[d] *Neurotec Department, Division of Clinical Geriatrics, Karolinska Institutet, Huddinge University Hospital, S-14186 Huddinge, Sweden*

Received 22 October 2002; received in revised form 29 July 2003; accepted 28 August 2003

Abstract

The structural correlates of mild cognitive impairment (MCI) were examined in 105 elderly subjects whose cognitive function ranged from intact to demented, including 38 subjects with MCI. Hippocampal volumes (left and right HcV), brain volume (BV), and grey matter volume (GMV) and white matter volume (WMV) were segmented from high resolution magnetic resonance data sets and normalised to intracranial volume (ICV). Hippocampal volume reductions, but not global brain, white or grey matter atrophy, were associated with MCI. White matter lesion severity did not differ over cognitive states. In multiple logistic regression models, normalised HcV and ICV (indicating premorbid brain volume) were significant predictors of MCI versus normality. Normalised BV and ICV significantly predicted dementia versus MCI. Absolute volumetric measures of HcV and BV yielded comparable classification accuracies. Hippocampal atrophy may be the crucial step for the transition from normality to MCI. Widespread brain atrophy may be the step to determine the transition from MCI to dementia. Brain volume reserve effects appear to be involved in both of these steps.

Keywords: Ageing; Cognition; Brain; Hippocampus; MRI; Volumetry; Normalisation; Mild cognitive impairment; Brain reserve; Apolipoprotein E; Correlation

1. Introduction

Cognitive deficits that are not severe enough to fulfil dementia criteria are a common phenomenon in elderly subjects. This transitional zone between normal ageing and dementia has been referred to in broad terms as “mild cognitive impairment” (MCI). Despite various definitions and concepts used in this research field, it is generally agreed that subjects with MCI are at increased risk of dementia. Alzheimer’s disease (AD) appears to be the far most common cause of progressive cognitive decline in elderly subjects, but MCI may also be prodromal in other dementia disorders [45]. In a varying but not trivial proportion of MCI subjects, the cognitive impairment may either improve or remain stable over time [7,43,50].

Subjects with MCI are being used in a growing number of neuroimaging studies. Many of these have focussed on possible early predictors of AD in subjects with MCI. Therefore, the brain structures that are known to be affected early by Alzheimer pathology [5] have been most intensively studied in MCI.

Atrophy of the hippocampus has been identified as the most consistent finding in a handful of studies applying structural imaging with volumetry to heterogeneous populations with MCI [10,11,13,14,34,57,60,64]. Atrophy of other limbic structures, such as the entorhinal cortex and/or parahippocampal gyrus [10,11,13,14,34,64], amygdala [16] and cingulate cortex [20,33] may also be associated with MCI. However, fewer studies and some controversial results [14,34,64] exist with regard to these structures.

Only few studies have focussed on other brain structures and more widespread atrophy patterns in MCI. Despite occasional reports of reduced global grey matter volume (GMV) [14] or atrophy in multiple neocortical areas [8,33], the magnitude of these studies imply that global brain, white or grey matter atrophy is not usually pronounced in MCI [8,10,11,19,56,60]. However, atrophy in neocortical temporal lobe areas [9] as well as more widespread (“global”) brain atrophy [20], may be the steps that determine the decline to dementia in subjects suffering from MCI.

* Corresponding author. Tel.: +46-8-585-89783; fax: +46-8-585-85470.
E-mail address: henrike.wolf@neurotec.ki.se (H. Wolf).

0197-4580/$ – see front matter © 2003 Elsevier Inc. All rights reserved.
doi:10.1016/j.neurobiolaging.2003.08.006

Volumetric studies in the field of MCI have usually interpreted volume differences over cognitive states as an indication of atrophy. However, some recent studies suggested that such findings may also reflect premorbid differences in absolute size of these structures [12,39]. The vast majority of neuroimaging studies tend to normalise absolute volume measurements, most commonly by division by intracranial size or similar procedures. This is done to correct for inter individual variations in head size that may considerably confound the volume of brain structures [29]. Normalisation has been shown to decrease the variability of hippocampal measurements [22] and to provide a considerable advantage to neuroimaging studies with small sample size, because it makes volumetric measurements from males and females comparable [3,29]. However, normalisation may "neutralise" the possible influence of premorbid brain size.

The present study aimed to characterise the structural correlates of late-life cognitive impairment in a cross-sectional study. Subjects were collected to represent the cognitive continuum from normality to mild to moderate dementia. The study was restricted to subjects in a narrow age range of 75–85. A combination of clinical and psychometric criteria was used for the definition of MCI. Groups were not pre selected based on a priori defined diagnostic criteria.

The following hypotheses were tested:

(1) Hippocampal and global brain volumes are closely correlated with memory and global cognitive functions.
(2) The hippocampal volume distinguishes elderly without cognitive impairment from elderly subjects with mild cognitive deficits, while the degree of global brain atrophy distinguishes between MCI and dementia.
(3) Relative brain volumes, i.e. volumes "normalised" by division by ICV, are superior predictors of cognitive state as compared to absolute volumes.

2. Materials and methods

2.1. Subjects

All subjects and/or their legal caregivers gave informed written consent to participate in this study that was approved by the local ethics committee. The subjects were collected to represent a characteristic sample of the elderly with a cognitive continuum from normality to mild/moderate dementia within a narrow age range of 75–85 years. Subjects were recruited from two sources: (1) the Leipzig Longitudinal Study of Aging (LEILA 75+) [48] and (2) referrals to the local Memory Clinic.

The vast majority of non-demented subjects ($n = 68$) were participants of LEILA 75+ who were consecutively recruited for this study according to MMSE strata as previously described [60]. They represent approximately 10% of the non-demented LEILA 75+ sample in the same age range. Due to the sampling procedure along a cognitive continuum, subjects with mild cognitive deficits are over-represented in this neuroimaging study as compared to the total LEILA 75+ population [6]. Five additional non-demented subjects with mild cognitive deficits were recruited from referrals to the local memory clinic. They did not differ from the LEILA 75+ subjects with regard to demographic features, general health status and neuropsychological performance. The non-demented subjects in this study did not differ from the corresponding LEILA 75+ population with regard to the prevalence of myocardial infarction (9.5% versus 10.5%, $P = 0.8$), diabetes mellitus (20% versus 22%, $P = 0.4$), and stroke (9% versus 6.4%, $P = 0.3$).

An age-matched "reference group" with mild to moderate dementia was recruited from patients attending the local Memory Clinic who had recently been diagnosed with a dementia disorder. The memory clinic receives referrals from general practitioners and specialists in the fields of Neurology, Psychiatry and Internal Medicine in the larger city area of Leipzig, including the areas covered by LEILA 75+. The study sample included 38 subjects that were used in a previously published study [60].

Exclusion criteria in this study were defined broadly as conditions leading to the inability to participate in a neuroimaging study and/or inability to complete neuropsychological tests. More specifically, such conditions were clearly defined contraindications for MRI (i.e. pacemaker or recent heart/brain surgery), as well as severe physical handicap, medical, psychiatric and neurological diseases or severe sensory deficits. A history or presence of cancer, diabetes, heart disease, stroke, Parkinson-like features and mild depressive symptoms did not lead to exclusion from the study. None of the subjects enrolled in this study suffered from temporal lobe epilepsy, brain tumours likely to cause cognitive deficits or major vessel infarcts.

According to clinical diagnostic criteria (described below), 38 subjects in this population were assigned to the subgroup MCI, i.e. their cognitive function was not normal, but they did not fulfil dementia criteria. Thirty-two were classified as "dementia".

2.2. Methods

2.2.1. Clinical and neuropsychological assessment

All subjects were clinically assessed as described previously [60]. Neuropsychological assessment was based on the SIDAM (Structured Interview for the Diagnosis of Dementia of Alzheimer type, Multi-infarct dementia and dementias of other aetiology according to ICD-10 and DSM-III-R) [66]. The SIDAM test performance part consists of a neuropsychological screening battery with 55 questions, including all items of the Mini-Mental State Examination (MMSE; [17]). It yields a maximum score of 55 (*SI*DAM *sco*re or SISCO). The SISCO can be subdivided into several cognitive domains (orientation, immediate recall, delayed recall, long term memory, intellectual abilities,

verbal abilities/calculation, visuo-spatial function, and aphasia/apraxia). The composite sub score "memory" is derived by summing the performance in the sub domains "immediate recall" (of numbers and words), "delayed recall" (of verbal and figural material) and "long term memory" (biographical memory). The maximum score in "memory" is 20. Age- and education-specific norms for the SISCO sub scores were recently established using the LEILA 75+ population [6].

The presence of a dementia syndrome was defined in accordance with ICD-10 research criteria [63]. Diagnoses of dementia disorders were made according to ICD-10 research criteria [63]. Because of insufficient operationalisation within the ICD-10, the NINDS–AIREN [51] criteria for vascular dementia were additionally applied in all cases with a dementia syndrome.

The presence of MCI was defined as evidence of objective cognitive impairment in the absence of dementia. This was based on either a Clinical Dementia Rating (CDR) of 0.5 ("questionable dementia") [28] and/or impaired test performance on SISCO, as defined by scores of at least 1 S.D. below age- and education-adjusted means in one or more cognitive sub domains. Hence, the psychometric criteria are consistent with those suggested for ageing-associated cognitive decline [38]. Clinical and psychometric assessments were made independently. Subjective memory complaint was disregarded as a criterium for MCI. As in LEILA 75+ [7], memory complaint was only poorly associated with objective cognitive performance: 20 subjects classified as MCI in this study did not report any memory problems.

2.2.2. Neuroanatomical imaging and analysis

2.2.2.1. MR aquisition. Three-dimensional (3D) T1-weighted high resolution MRI brain data sets were obtained on a Siemens Vision 1.5 T scanner using a 3D T1-weighted sequence (MPRAGE, TR 11.4 ms, TE 4.4 ms, 128 slices, matrix 256 × 256, voxel size 0.9 mm × 0.9 mm × 1.5 mm). In addition, T2-weighted MR images were obtained (TR 5016 ms, TE 132 ms, matrix 357 × 512, 19 slices, 5-mm slices, gap 1.5 mm, field of view 255 mm × 255 mm, transversal).

2.2.2.2. MR analysis. Collected data sets were analysed with the BRIAN system [36]. All analyses were performed blind to knowledge of the cognitive state or other clinical data on the subjects. First, the T1-weighted data sets were aligned with the stereotactical coordinate system and interpolated to an isotropical voxel size of 1 mm using fourth-order b-spline interpolation. Second, automated segmentations of brain compartments were performed from the whole 3D brain data set. The intracranial compartments, GMV, white matter volume (WMV), internal and external cisterns (cerebrospinal fluid = CSF compartments) were automatically determined using a boundary-guided region-growing procedure, as described previously [27,60]. The brain volume (BV) was defined as the sum of the GMV and WMV. The intracranial volume (ICV) was defined as the sum of the BV and the CSF volume (Fig. 1). Validation and reliability studies demonstrated high validity and re-scan and re-measure reliability (ICC 0.99 for summed volumes on repeated scans from 12 subjects) (for details see [61]).

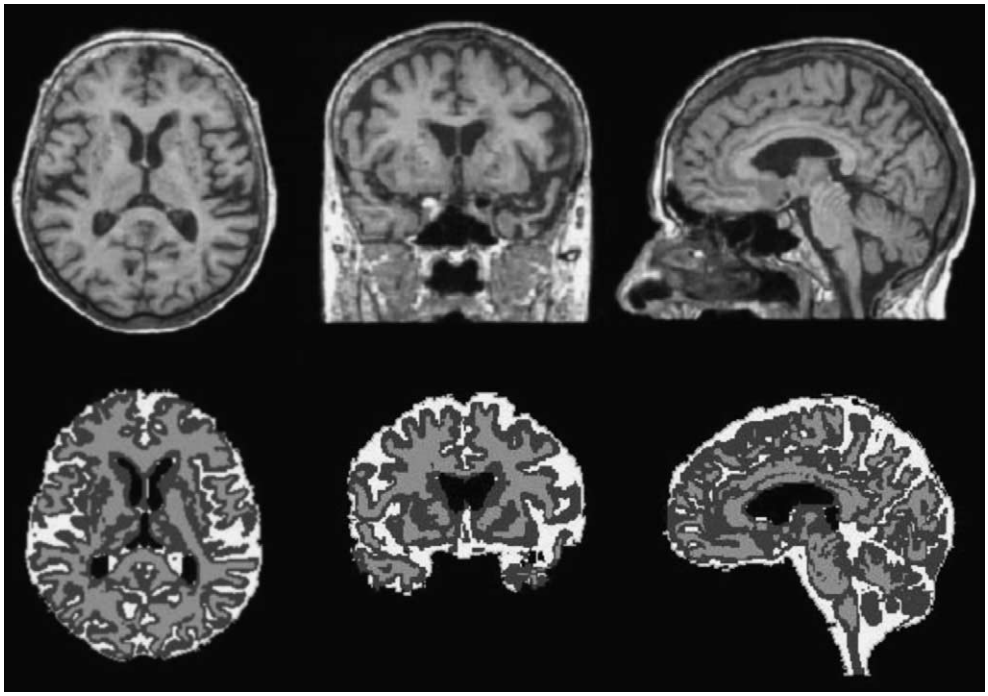

Fig. 1. Boundary guided region growing. Example segmentation in a 78-year-old women with mild dementia in Alzheimer's disease (MMSE 22). The brain volume was defined as the sum of white matter plus grey matter volume. The ICV was defined as the sum of internal (ventricular = black) and external (cortical = white) cerebrospinal fluid spaces plus the brain volume.

Third, six cross-sections of the *hippocampus* at 3-mm intervals were outlined manually with a mouse driven cursor in the coronal plane (perpendicular to AC–PC line) on both hemispheres. The first two hippocampal slices were placed in the posterior part of the hippocampal head. The following four slices were placed in the body of the hippocampus (for details see [60]). All hippocampal measures in this study were performed by one experienced rater who was blind to the clincial status of the subjects (AH). Areas 1–6 were multiplied by slice thickness (3 mm) and summed on each side to yield the left versus right hippocampal volume estimate (left versus right HcV).

A high inter-rater reliability of this method has been demonstrated [60]. Unlike other morphometric approaches, our protocoll uses a fixed number of slices. Our method is based on the assumption that atrophy along the longitudinal axis of the hippocampus is relatively small as compared to atrophy in the coronal plane. This assumption was confirmed by an experiment that measured the anterior–posterior (AP) extension of the hippocampus in a random sample of 56 (30 males) subjects spanning all cognitive states. There were no statistically significant differences in the AP extension of the hippocampus over cognitive state ($F = 0.3$, $P = 0.75$) and gender ($F = 0.3$, $P = 0.58$). The mean distance (S.D.) between area 1 and the posterior disappearance of hippocampal grey matter was 26.8 mm (2.9) in subjects without cognitive impairment (NC), 26.3 (2.4) in MCI and 26.5 (2.7) in dementia.

The measurement techniques for brain compartments and hippocampal volumes are demonstrated in Figs. 1 and 2.

2.2.2.3. Normalisation. To account for inter individual variation in head size and to yield comparable estimates of atrophy, the segmented brain volumes (BV, WMV, GMV) as well as the hippocampal volume estimates (left HcV, right HcV, total HcV) were normalised according to the following formula:

$$\text{RV}\,(\%) = \frac{V\,(\text{cm}^3)}{\text{ICV}\,(\text{cm}^3)} \times 100$$

where RV is the relative volume and V is the absolute volume of a brain structure.

2.2.2.4. Visual assessments. All visual assessments were performed blind to knowledge of the cognitive state or other clinical data on the subjects by one rater (HW). A 4-point scale (0: absence; 1: mild; 2: moderate; 3: severe) was used to yield a white matter severity index for the features periventricular and deep white matter hyperintensities (PVH, DWMH), dilated perivascular spaces (DPS) (from T2-weighted images), and multiple small lesions (ML) (from T1-weighted images). Lacunar infarcts >4 mm diameter were identified and counted using both T1- and T2-weighted images. Hippocampal atrophy was rated on T1-weighted images using a 5-point scale (0: absent; 1: very mild; 2: mild; 3: moderate; 4: severe) and then dichotomised as either significant (score >2) or absent (scores 0, 1 or 2). Intra rater experiments, which were performed several months after the initial rating, demonstrated a high reliability with kappa values of 0.88 for VRS, 0.92 for DWMH, 0.95 for PVH, and 0.91 for hippocampal atrophy.

2.2.3. Apolipoprotein E (ApoE) genotype

A number of previous studies suggest that the ApoE genotype, the main known genetic susceptibility factor for Alzheimer's disease, has effects on hippocampal size, atrophy and hemispheric lateralisation [23,46,55]. ApoE was therefore considered as a confounder of hippocampal volumes in this study. The ApoE genotype was analysed in a sub sample of 60 patients. Genomic DNA was extracted from blood leukocytes (30 μl whole blood) and processed as previously described [54].

2.2.4. Statistical analysis

The data were analysed with SPSS for windows (Version 10.0.7).

The BV and both (left and right) hippocampal volumes were considered as the main predictor variables of cognitive states. The white and grey matter volumes were defined

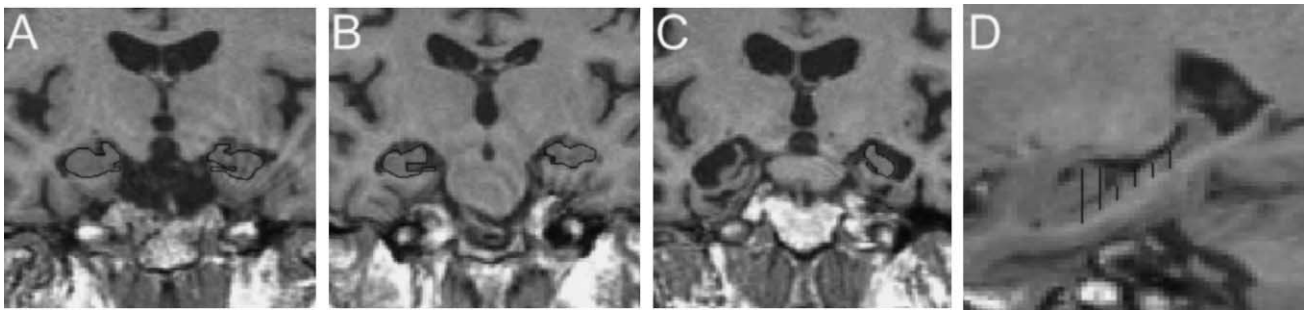

Fig. 2. Hippocampal outlining, showing examples of hippocampal outlining in three cases in the coronal plane. (A) Non-atrophied hippocampus (first outlined area) in a 80-year-old woman with normal cognition (MMSE 28). (B) Mildly atrophied hippocampus (first outlined area) in a 75-year-old woman with MCI (MMSE 27). (C) Severely atrophied hippocampus (second outlined area) in a 84-year-old mildly demented woman with Alzheimer's disease (MMSE 21). (D) Sagittal aspect of the hippocampus. Levels of hippocampal area measurements 1–6 (anterior–posterior) are indicated from left to right. The first two measurements were placed in the hippocampal head, the following four in the hippocampal body. Panel D was taken from [60].

as secondary predictors. Both, absolute volumes and relative volumes (percentage of ICV) were considered in the analyses. To make absolute measures comparable in men and women, ICV, BV, WMV, GMV and left and right HcV were transformed into percentile rank scores separately for each gender. The ICV—as an indicator of pre-morbid brain volume—was considered as a confounder of regional brain volumes. Further, age, education and gender were used as confounders in multivariate analyses. The significance level was set to $P = 0.05$ in all analyses.

Hypothesis 1 was examined using explorative statistics and Spearman's rank correlation. In addition, partial correlation analyses were calculated to remove the effect of global brain atrophy and ICV. Correlation comparisons for dependent samples were calculated to show whether normalised and absolute hippocampal volumes were similarly correlated with cognitive performance [52]. To test Hypothesis 2, we first assessed differences over cognitive states and gender using two-way multivariate ANOVA with all predictor variables, followed by Tukey post-hoc statistics. To identify the variables which were responsible for the transition of cognitive states, logistic regression models (backward conditional) with brain volumes and possible confounds of cognitive state were calculated. To show whether normalisation of brain measures yields an advantage over raw (absolute) volumes (Hypothesis 3), separate logistic regression models were calculated for absolute measures (represented by BV, respectively WMV and GMV, and left and right HcV) and normalised measures (represented by relative brain volume, respectively WMV and GMV, and relative left and right HcV), followed by McNemar's χ^2 statistics to verify whether the differences in classification were significant. Collinearity was checked for, and variables that correlated by more than $r = 0.85$ with each other were not entered together. Individual predicted probability values from each regression model were saved. The accuracy of classification was calculated based on every predicted probability value and represented as area under the receiver operator characteristic (ROC) curves. Sensitivities were calculated at a given specificity of 80%. To examine the validity of the regression models for distinct subgroups, area under the curves (AUCs) and their confidence intervals were additionally calculated for diagnostic subgroups (CDR 0.5, AACD, amnestic MCI, AD, dementia with CVD).

3. Results

3.1. Baseline characteristics of the sample

Table 1 shows that all groups were well matched according to age and sex. In women, differences in the educational level occurred between NC and MCI/De. Men with MCI showed trends for higher performance on MMSE, global SISCO, and—not shown—SIDAM sub scores. Results from our field study do not support gender differences in test performance on the SISCO [7]. Hence, we must presume the presence of selection bias towards more mildly impaired men.

Table 1
Baseline characteristics of the sample

Cognitive state Gender proportion		NC ($n = 35$) 21 F/14 M, mean (S.D.)	MCI ($n = 38$) 26 F/12 M, mean (S.D.)	De ($n = 32$) 20 F/12 M, mean (S.D.)
Age	Male	78.9 (2.8)	79.2 (2.7)	77.6 (3.2)
	Female	78.6 (2.5)	79.5 (3.1)	77.9 (2.4)
MMSE	Male	28.7 (0.7)	27.4 (1.6)	20.1 (5.3)[a]
	Female	29.1 (0.70)	26.0 (1.7)	20.7 (3.0)[b]
SISCO	Male	51.1 (2.4)	48.2 (2.8)	32.8 (8.3)[b]
	Female	51.6 (2.2)	44.7 (3.8)	32.6 (5.6)[b]
Years of education	Male	11.9 (2.4)	11.4 (1.7)	12.5 (2.2)
	Female	12.5 (3.3)	10.0 (1.4)	10.4 (1.2)[c]

Abbreviations: NC, no cognitive impairment; MCI, mild cognitive impairment; De, dementia; F, number of female subjects; M, number of male subjects.

[a] NC, MCI > De.

[b] NC > MCI > De.

[c] NC > MCI, De (ANOVA, Tukey post-hoc test, $P \leq 0.005$).

Two independent methods were applied to identify subjects with MCI in this study. Naturally, the two criteria CDR 0.5 and AACD overlapped considerably (79%), but not completely. Three subjects fulfilled the psychometric AACD criteria, but were clinically judged to have a CDR 0. Five subjects despite having received a CDR of 0.5 did not fulfil the psychometric criteria for AACD. Among the subjects classified as AACD, eight (seven females) performed at least 1.5 S.D. below age- and education-adjusted means in short term memory, i.e. they would fulfil the psychometric criteria for "amnestic MCI" (if the criteria "subjective memory complaint" and "absence of global cognitive impairment" are disregarded). The general level of impairment in our MCI subjects was very mild, as also indicated by a mean CDR sum of boxes score of 1 (S.D. 0.88) in this group.

Among the patients with dementia, 25 had CDR scores of 0.5 and 1, indicating mild dementia. The remaining seven patients received a CDR of 2 (moderate dementia).

Thirty patients received an ICD-10 diagnosis of Alzheimer's disease (10 with atypical/mixed form), and two had vascular dementia. According to NINDS–AIREN criteria, none of the 32 demented patients fulfilled the criteria for "probable vascular dementia (VaD)" (two had possible vascular dementia, six had "AD with CVD", 24 no diagnosis). For subgroup analyses, the eight patients fulfilling NINDS criteria for "possible VaD" and "AD with CVD" are referred to as "dementia with CVD" subgroup.

Common medical diseases in our sample included arterial hypertension (61%), diabetes mellitus (20%), coronary artery disease (18%) and a history of stroke (9%). The prevalences did not differ over cognitive states, except for coronary artery disease, which tended to be more common in MCI and dementia (χ^2-test, $P = 0.024$). In none of the examined white matter features (DWMH, PVH, DPS, ML, la-

cunar infarcts), were there observed significant differences in frequency or severity over cognitive states and gender (χ^2 statistics and Kruskal–Wallis tests, all P values > 0.3). In contrast, the severity of visually assessed hippocampal atrophy differed significantly over the three cognitive states ($P < 0.0005$). Significant hippocampal atrophy (score > 2) was observed in seven cognitively normal subjects (20%), 14 subjects with MCI (34%), and 19 (59%) subjects with dementia ($\chi^2 = 11.0$, $P = 0.004$). Other incidental findings included small meningeomata without therapeutic consequences (one female with NC, one female with dementia). In eight subjects, a pronounced ventricular enlargement was found (one female with NC, three females, one male with MCI, two males and one female with dementia). In none of the latter subjects could the suspicion of "normal pressure hydrocephalus" be confirmed after thorough clinical investigations and follow-up.

3.2. Volumetric measurements

Absolute and normalised brain volumetric measurements across cognitive states and gender, and results of the corresponding statistical analyses are shown in Table 2.

All hippocampal measures differed significantly between NC and MCI. All global brain volumes (BV, WMV, GMV) differed between NC and De and MCI and De. There were significant cognitive group × gender interaction effects in absolute BV and WMV, indicating a steeper decline with decreasing cognitive state in women. However, no such interaction effects were seen with normalised BV and WMV, which suggests that this finding represents differences in premorbid brain size between men and women rather than differential atrophy patterns. The main effects and interaction effects remained unchanged when the analysis was controlled for the ApoE genotype in a sub sample of 60 subjects with a known ApoE status. Because some previous studies, including our own [60], suggested an asymmetrical involvement of the hippocampus in the earliest stages of AD, we tested the possibility of pronounced or decreased lateralisation effects in a repeated measure ANOVA. The right HcV was larger than the left over cognitive groups and gender ($F(1, 99) = 15.8$, $P < 0.0005$). There were no significant interaction effects in this model to prove hippocampal asymmetry effects on cognitive state ($F(2, 99) = 1.31$, $P = 0.276$).

Table 2
Brain volumetric measurements over cognitive state and gender

Predictor	Gender	Cognitive state			Between-subject effects[a]		
		NC ($n = 35$)	MCI ($n = 38$)	De ($n = 32$)	Gender (d.f. 1)	Cognitive group (d.f. 2)	Interaction (d.f. 2)
ICV (cm^3)	M	1548 (107)	1561 (126)	1540 (96)	M > F	NC [>] MCI, De	Gender × Cog
	F	1492 (133)	1386 (125)	1364 (110)	$F = 31.5$; $P < 0.0005$	$F = 2.8$; $P < 0.068$	$F = 2.8$; $P = 0.067$
BV (cm^3)	M	1088 (100)	1116 (96)	1028 (63)	M > F	NC, MCI > De	Gender × Cog
	F	1069 (113)	1003 (76)	916 (45)	$F = 22.2$; $P < 0.0005$	$F = 14.1$; $P < 0.0005$	$F = 3.3$; $P = 0.04$
RBV (% of ICV)	M	70 (5.2)	72 (2.5)	67 (5.0)	M = F	NC, MCI > De	–
	F	72 (3.8)	73 (4.5)	67 (3.2)	$F = 1.1$; $P = 0.3$	$F = 11.2$; $P < 0.0005$	
WMV (cm^3)	M	451 (56)	467 (40)	427 (37)	M > F	NC, MCI > De	Gender × Cog
	F	451 (63)	413 (41)	370 (27)	$F = 15.6$; $P < 0.0005$	$F = 11.7$; $P < 0.0005$	$F = 4.0$; $P = 0.022$
WMV (% of ICV)	M	29 (3.8)	30 (1.5)	28 (2.8)	M = F	NC, MCI > De	–
	F	30 (2.4)	30 (2.6)	27 (2.6)	$F = 0.05$; $P = 0.8$	$F = 7.6$; $P = 0.001$	
GMV (cm^3)	M	637 (64)	650 (64)	602 (37)	M > F	NC, MCI > De	–
	F	618 (58)	590 (40)	546 (29)	$F = 20.2$; $P < 0.0005$	$F = 11.2$; $P < 0.0005$	
GMV (% of ICV)	M	41 (2.7)	42 (2.2)	39 (2.8)	M [<] F	NC, MCI > De	–
	F	42 (2.6)	43 (2.4)	40 (2.0)	$F = 2.8$; $P = 0.09$	$F = 8.6$; $P < 0.0005$	
Right HcV (cm^3)	M	1.62 (0.22)	1.48 (0.3)	1.20 (0.3)	M = F	NC > MCI > De	–
	F	1.62 (0.17)	1.37 (0.19)	1.27 (0.2)	$F = 0.1$; $P = 0.8$	$F = 22.8$; $P < 0.0005$	
Left HcV (cm^3)	M	1.53 (0.12)	1.43 (0.21)	1.2 (0.24)	M [>] F	NC > MCI > De	–
	F	1.51 (0.2)	1.25 (0.15)	1.2 (0.27)	$F = 2.9$; $P = 0.09$	$F = 19.6$; $P < 0.0005$	
Right HcV (% of ICV)	M	0.10 (0.015)	0.096 (0.020)	0.078 (0.019)	M < F	NC > MCI > De	–
	F	0.11 (0.014)	0.099 (0.015)	0.094 (0.018)	$F = 5.4$; $P = 0.022$	$F = 13.2$; $P < 0.0005$	
Left HcV (% of ICV)	M	0.099 (0.010)	0.092 (0.014)	0.079 (0.018)	M = F	NC > MCI, De	–
	F	0.10 (0.012)	0.091 (0.013)	0.088 (0.021)	$F = 1.2$; $P = 0.3$	$F = 9.6$; $P < 0.0005$	

Abbreviations: M, male; F, female; Gender × Cog, Gender × Cognitive group interaction; NC, no cognitive impairment; MCI, mild cognitive impairment; De, dementia; ICV, intracranial volume; BV, brain volume; HcV, hippocampal volume; WMV, white matter volume; GMV, grey matter volume; RBV, relative brain volume; [>] trend, i.e. $0.05 < P$-value < 0.1.

[a] Two-way multivariate ANOVA with gender and cognitive state as factors.

Table 3
Spearman rank correlation coefficients of hippocampal volume (absolute and normalised) with global cognitive function (SISCO) and memory (memory sub scale of the SISCO)

	Spearman correlation coefficients		Partial correlation coefficients controlled for RBV		Partial correlation coefficients controlled for ICV
	HcV ranked	HcV %	HcV ranked	HcV %	HcV ranked
Non-demented					
Total memory	0.56 ($P < 0.0005$)	0.32 ($P = 0.006$)[a]	0.55 ($P < 0.0005$)	0.32 ($P = 0.006$)[a]	0.47 ($P < 0.0005$)
SISCO	0.60 ($P < 0.0005$)	0.34 ($P = 0.001$)[a]	0.58 ($P < 0.0005$)	0.39 ($P = 0.001$)[a]	0.51 ($P < 0.0005$)
Demented					
Total memory	−0.01 ($P = 0.99$)	−0.06 ($P = 0.77$)	−0.06 ($P = 0.73$)	−0.16 ($P = 0.49$)	−0.03 ($P = 0.86$)
SISCO	−0.17 ($P = 0.34$)	−0.23 ($P = 0.21$)	−0.13 ($P = 0.40$)	−0.21 ($P = 0.26$)	−0.10 ($P = 0.61$)

Abbreviations: HcV ranked, gender-specific percentile rank of hippocampal volume; ICV, intracranial volume; HcV %, relative HcV (ratio of ICV).
[a] Differences in correlation coefficients between ranked HcV vs. normalised HcV were significant ($P < 0.005$).

3.3. Relationship between brain structures and continuous measures of cognitive functions

Explorative analyses revealed that the relationship between global cognitive performance (SISCO) and hippocampal volume was not ideally linear (Fig. 3). A bend in the regression line occurred at total SISCO scores between 35 and 45 (MCI range). The total memory score showed a similar non-linear relationship with hippocampal measures with a bend occurring between 12 and 14 (MCI range). The same pattern was observed for both sides and for both absolute and normalised volumes. In contrast, the relationships between cognitive measures and BV, WMV and GMV were linear (not shown). Considering only non-demented subjects also revealed a nonlinear relationship between hippocampal and cognitive measures. Table 3 lists the correlation coefficients between hippocampal volume and memory and SISCO separately for non-demented and demented subjects. Significant correlations in the expected direction were only found in non-demented subjects. They remained stable after controlling for global brain atrophy (RBV) and ICV.

3.4. Cross-sectional predictors of cognitive state

The main results of the regression analyses are summarised in Table 4. There were no statistically significant differences in the classification accuracy between absolute and relative models.

When relative GMV and relative WMV were entered instead of relative BV, both remained in the final model together with ICV. Yet, the classification accuracy was not higher than that achieved by the model based on relative BV only. This indicates that, as a diagnostic tool, the global

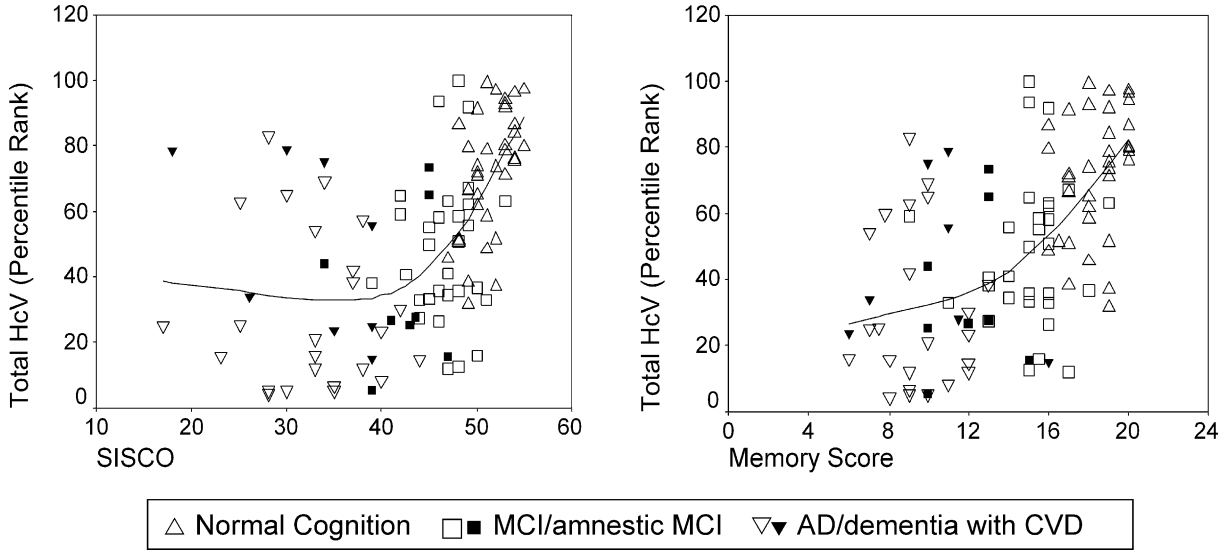

Fig. 3. Scatter plot. Hippocampal volume (left plus right percentile rank of absolute volumes) against global cognition (SISCO) and memory function (memory sub domain of the SIDAM) in 105 elderly subjects with a continuum of cognitive functions. Locally weighed trend curves (70%). Spearman rank correlation coefficients in the total sample were $r = 0.58$ for total memory and $r = 0.56$ for SISCO. These correlations remained stable after correction for brain atrophy (RBV) (partial correlation coefficent $r = 0.52$, $P < 0.0005$ for memory, and $r = 0.46$, $P < 0.0005$ for SISCO).

Table 4
Logistic regression models

	Transition from NC to MCI ($n = 73$)						Transition from MCI to De ($n = 70$)					
	Model 1 ("absolute")			Model 2 ("relative")			Model 1 ("absolute")			Model 2 ("relative")		
	Predictor	*B*	*P* value	Predictor	*B*	*P* value	Predictor	*B*	*P* value	Predictor	*B*	*P* value
Final model	Left HcV	−0.049	<0.0005	ICV	−0.035	0.01	BV	−0.067	0.0005	ICV	−0.048	0.005
	Education	−0.294	0.03	L. HcV %	−109.4	0.001	Age	−0.298	0.01	RBV	−47.4	<0.0005
				Education	−0.302	0.04	Education	0.341	0.09	Age	−0.317	0.013
Accuracy (%)[a]	78			75			78			78		
Sensitivity (%)[b]	76			74			69			85		
Specificity (%)[b]	80			80			80			80		
AUC (95% CI)	0.831 (0.738–0.924)			0.868 (0.786–0.950)			0.871 (0.788–0.953)			0.878 (0.797–0.960)		
LR	3.8			3.7			3.45			4.25		

The following predictor variables were entered into "absolute models": BV/[ICV], left HcV, right HcV, age, gender, education, education × gender. The following predictor variables were entered into "relative models": RBV, relative left HcV, relative right HcV, age, gender, education × gender, ICV sensitivity and specificity in the models contrasting MCI and dementia were based on prediction of dementia, /[ ] indicates that alternative models were calculated. Abbreviations: NC, no cognitive impairment; MCI, mild cognitive impairment; De, dementia; HcV, hippocampal volume; BV, brain volume; ICV, intracranial volume; HcV %, relative HcV (ratio of ICV); RBV, relative brain volume (ratio of ICV); L., left; LR (likelihood ratio) = sensitivity/(1 − specificity); AUC, area under the curve.

[a] Classification accuracy, derived from individual group classification asigned by the logistic regression model, note that arbitrary cut-offs on probability values are chosen.

[b] Derived from ROC curves based on individual probability values from the regression models.

brain volume alone seems to be sufficient to separate MCI and dementia.

Thus, of all brain structures measured, only the hippocampus was able to distinguish NC from MCI. As predicted, global brain volume, but not the hippocampus, was responsible for the transition from MCI to De. When entered as absolute volumes, HcV and BV alone were sufficient predictors of cognitive state. When entered as normalised volumes, the ICV was required as an additional predictor. The hypothesis that normalised measurements are superior to absolute measurements was disproved.

Finally, we considered the effect of our prediction models on possible subtypes among our cases with dementia and MCI. Receiver–operator statistics showed that the diagnostic models derived from absolute brain volumes for classification of MCI and NC also had a high predictive value for amnestic MCI (AUC = 0.819; 95% CI: 0.684–0.954), CDR 0.5 (AUC = 0.836; 95% CI: 0.743–0.930), and AACD (AUC = 0.743; 95% CI: 0.627–0.859) versus all other non-demented subjects. Likewise, the model for MCI and De also had significant predictive value to classify "dementia plus CVD" versus MCI (AUC = 0.744; 95% CI: 0.622–0.866). It should be noted that even though cerebrovascular lesions were frequent in our population, none of the demented subjects met the criteria for probable vascular dementia, i.e. Alzheimer's disease must be considered as the main etiological background of dementia in our study.

4. Discussion

We investigated structural correlates of MCI based on high-resolution MRI in a well characterised elderly sample within a narrow age range of 75–85. Post-mortem studies revealed a high prevalence of Alzheimer pathology in this age group. By the age of 85 virtually everyone will have some neurofibrillary tangles in their cerebral cortex, yet not everyone is demented [24,49]. Based on results from our own epidemiological study, the prevalence of dementia and MCI in this age group approximates 10 and 18%, respectively [47]. About 30% with MCI based on modified AACD criteria have been shown to develop dementia over 2.6 years in this elderly population [7].

In line with previous studies and our hypothesis, we found a close correlation between hippocampal volume and memory as well as global cognitive performance (Hypothesis 1). Further, in line with our expectations (Hypothesis 2), of the chosen volumetric measurements the hippocampal volume uniquely distinguished MCI from normal cognition. Measures of global brain atrophy distinguished best between MCI and dementia. This was due to both white and grey matter reduction. Contrary to the prediction that normalisation of volumetric measurements yields an advantage over raw volumes, normalised and absolute measurements yielded comparable classification accuracies. In models based on normalised measures, the ICV was required as an additional predictor of cognitive state. We thus rejected Hypothesis 3.

Some aspects of our results need to be discussed in more detail in the following sections.

4.1. Hippocampal volumes in MCI

In accordance with previous studies [10,11], we found that of the brain volumetric measurements examined, the HcV uniquely distinguished MCI from normal. The mean differences in HcV between MCI and NC based on HcV volumes is comparable to the 7–16% reported in other studies [10,11,13,14,57,58,60,64]. Our accuracy of classification of MCI and NC was comparable to the 63–73% that were reported by similar studies [10,14,64]. Yet, it should be noted that hippocampal volumes overlapped considerably over cognitive states in our study. Given that 50% classification can be achieved by chance in the regression models, classification rates of 75–85% are only moderate. On the other hand, the transitional character of MCI as a diagnostic category means that a 100% correct classification is unrealistic.

The finding of "specific hippocampal volume reductions" [10] in MCI fits well into the concept of AD as a "hippocampal dementia" [2]. As shown by Braak and Braak [5], neurofibrillary pathology in AD evolves in a distinct hierarchical pattern. The initial clinical characterisation of the stageing model suggested that the transentorhinal stages (stages 1 and 2) were the "clinically silent" period of AD, while the limbic stages (stages 3 and 4) represent incipient AD with first objective symptoms, and the neocortical stages (stages 5 and 6) fully developed dementia. Subsequently, clinico-pathological studies reported a high prevalence of AD pathology in subjects with MCI and a high sensitivity of hippocampal atrophy to detect AD, even in non-demented subjects [26,41]. However, there was also considerable variability of cognitive performance beyond the severity of AD lesions [25,31,49].

Diagnostic confirmation in the grey zone of MCI is usually based on the subsequent conversion to dementia and AD. However, conversion rates vary widely across studies and tend to be higher in clinic-based populations as compared to population-based samples [50]. If one considers the demographic and clinical heterogeneity of samples with MCI across published studies, the homogeneity of findings concerning hippocampal volumes is rather surprising. In the absence of pathological diagnostic confirmation in MCI subjects as in our and many other studies, the presumed impact of AD pathology on hippocampal volume loss cannot be proved. Other degenerative diseases and ischemia have to be considered as alternative causes of hippocampal volume loss.

The global brain atrophy in our study was no more pronounced in MCI as compared to controls. This finding is in accordance with the majority of cross-sectional studies on MCI that analysed either global brain atrophy or region-of-

interest analyses in neocortical areas [10,11,19,56,60]. In our MCI group, the normalised brain volume and grey matter volume even tended to be larger than normal. Overall, a higher variability may be present in MCI regarding the degree of neocortical atrophy. The global brain volume, but not the hippocampal volume, was the volumetric measurement to distinguish MCI from dementia. Hence, the degree of global brain atrophy may be a more important factor in the transition from MCI to dementia than is hippocampal atrophy.

4.2. The relationship between HcV and cognitive functions

If one assumes that hippocampal atrophy and neurofibrillary pathology are correlated [42], the stageing model of AD implies that hippocampal atrophy can be used as a global indicator of disease severity in AD. The physiological function and the early disconnection of the hippocampus with different cortical areas in AD [44] imply correlations between HcV and memory as well as global cognitive functions, even in non-demented subjects. This could be confirmed by our study and is in accordance with previous studies [10,41]. The relationship between HcV and global cognition as well as memory was not ideally linear. To all of our knowledge, this finding has never been reported previously in a comparable neuroimaging study. Our findings would be consistent with a floor effect in hippocampal atrophy during the degenerative process, i.e. a more pronounced hippocampal atrophy in the earliest disease stages that involve the transition to MCI may reach a plateau in the dementia stage. This has been suggested by the results of serial studies of MCI and pre clinical AD [21,32,37,59,65]. One pathological study reported floor effects in hippocampal neurofibrillary pathology in the more advanced AD stages [25]. In our cross-sectional study, possible bias due to selective survival, scale properties of the SIDAM scores, and selection bias in our demented cases have to be considered as alternative explanations for these findings.

4.3. Normalised versus absolute volumetric measures

Considering both normalised and absolute measures, there was little indication in our study that normalisation yields an advantage over raw measures with regard to cross-sectional classification accuracy as well as structural–functional correlations. Models based on normalised measures additionally required the ICV (as an indicator of pre-morbid brain size) to yield comparable classification accuracy as absolute HcV and BV. In fact, had we entered only normalised measures of hippocampal and brain volume without the ICV, the accuracy of classification would have been considerably lower than that achieved with absolute HcV and BV alone (not shown). Further, significant gender differences were present even after normalisation. Such findings call for caution with regard to the use of “standard” adjustment procedures.

In our sample, relatively large ICV differences were found over cognitive states, particularly in women. In light of two recent negative reports on ICV differences over cognitive states [15,30], this may appear as an atypical finding. We carefully excluded the possibility that measurement errors caused this result. Most convincingly, easy-to-obtain measurements of head size yielded the same results [61]. In fact, this is not a new finding. Significant differences in intracranial size over cognitive states were occasionally found in other neuroimaging studies [10,18,62]. Evidence is accumulating from pathological [40], epidemiological [4,53] and large-scale imaging studies [12,39] to support a role of premorbid brain size on late-life cognitive functions and the clinical expression of AD. In women as compared to men, more pronounced effects of small head size [53] and other indicators of early-life growth [35] on the risk of dementia and AD have been occasionally reported. However, the situation is inconclusive.

The pre-morbid brain volume may either influence cognitive performance as part of a general performance factor [39] and/or by providing reserve against degenerative processes and thus modifying the clinical expression of dementia diseases [31,40].

Our findings raise the further questions of whether it is only the degree of hippocampal atrophy that distinguishes cognitively unimpaired elderly subjects from those with MCI, or whether those with MCI may have started with smaller hippocampi earlier in life. The latter is implied by the finding that the ICV was not required as an additional predictor when absolute hippocampal volumes were used. This finding may indicate an effect of premorbid hippocampal volume rather than whole brain volume. Since absolute measurements reflect atrophy and total pre-morbid size of a structure to an unknown degree, such questions cannot be answered sufficiently based on cross-sectional results. Some findings from serial neuroimaging and neuropsychological studies in MCI would be consistent with this situation [1,32].

In summary, our findings suggest that neuroimaging studies should consider the ICV or other estimates of premorbid brain size as confounders of regional brain volumes and cognitive function.

4.4. Limitations

The main limitations are those of a cross-sectional study. No follow-up with post-mortem confirmation of diagnoses is available in our sample. The validity of MCI as a diagnostic category is not yet proven. Due to the sampling procedure and the broad MCI concept applied, our group with MCI is probably etiologically heterogeneous. The results have to be interpreted carefully with regard to etiological assumptions. Our study did not address the question of whether the hippocampus may be superior to other medial temporal lobe structures to predict MCI. In particular, the entorhinal cortex may be early and severely affected by neurofibrillary pathology [5].

4.5. Conclusions

Hippocampal volume reductions, but not global brain, white or grey matter atrophy, were associated with MCI in this study. From the limited angle of a cross-sectional study our findings suggest that hippocampal atrophy may be the step that determines the transition from normality to MCI. Widespread brain atrophy may be the crucial factor to determine the transition from MCI to dementia in AD, while hippocampal atrophy plays a less important role at this stage. Brain volume reserve effects may be involved in both of these steps.

Acknowledgments

This paper was supported by Interdisziplinäres Zentrum für Klinische Forschung (IZKF) at the University of Leipzig (Projekt C8), and by a research stipend from the German Research Council to Henrike Wolf. The authors wish to thank Richard Cowburn (Ph.D.), Neurotec, Huddinge, for linguistic revision of the manuscript, and Götz Gelbrich (Ph.D.), Coordination Center for Clinical Trials Leipzig (KKSL), Leipzig, for statistical consulting.

References

[1] Bäckman L, Small BJ, Fratiglioni L. Stability of the preclinical episodic memory deficit in Alzheimer's disease. Brain 2001;124:96–102.
[2] Ball MJ, Fisman M, Hachinski V, Blume W, Fox A, Kral VA, et al. A new definition of Alzheimer's disease: a hippocampal dementia. Lancet 1985;1:14–6.
[3] Bhatia S, Bookheimer SY, Gaillard WD, Theodore WH. Measurement of whole temporal lobe and hippocampus for MR volumetry: normative data. Neurology 1993;43:2006–10.
[4] Borenstein Graves A, Mortimer JA, Bowen JD, McCormick WC, McCurry SM, Schellenberg GD, et al. Head circumference and incident Alzheimer's disease: modification by apolipoprotein E. Neurology 2001;57:1453–60.
[5] Braak H, Braak E. Neuropathological stageing of Alzheimer-related changes. Acta Neuropathol (Berl) 1991;82:239–59.
[6] Busse A, Aurich C, Zaudig M, Riedel-Heller S, Matschinger H, Angermeyer MC. Age- and education-specific reference values for the cognitive test of the SIDAM (Structured Interview for the Diagnosis of Dementia of the Alzheimer type, Multi-infarct dementia and dementias of other etiology according to ICD-10 and DSM-IV). Z Gerontol Geriatr 2002;35:565–74.
[7] Busse A, Bischkopf J, Riedel-Heller SG, Angermeyer MC. Mild cognitive impairment: prevalence and incidence according to different diagnostic criteria. Results of the Leipzig Longitudinal Study of the Aged (LEILA 75+). Br J Psychiatry 2003;182:449–54.
[8] Chételat G, Desgranges B, de la Sayette V, Viader F, Eustache F, Baron JC. Mapping gray matter loss with voxel-based morphometry in mild cognitive impairment. Neuroreport 2002;13:1939–43.
[9] Convit A, de Asis J, de Leon MJ, Tarshish CY, De Santi S, Rusinek H. Atrophy of the medial occipitotemporal, inferior, and middle temporal gyri in non-demented elderly predict decline to Alzheimer's disease. Neurobiol Aging 2000;21:19–26.
[10] Convit A, de Leon MJ, Tarshish C, De Santi S, Tsui W, Rusinek H, et al. Specific hippocampal volume reductions in individuals at risk for Alzheimer's disease. Neurobiol Aging 1997;18:131–8.
[11] De Santi S, de Leon MJ, Rusinek H, Convit A, Tarshish CY, Roche A, et al. Hippocampal formation glucose metabolism and volume losses in MCI and AD. Neurobiol Aging 2001;22:529–39.
[12] DeCarli C, Massaro J, Au R, D'Agostino E, Wolf PA. Cranial cavity size and cognitive performance amongst the offspring of the Framingham Heart Study. Neurobiol Aging 2002;(Suppl):1323.
[13] Dickerson BC, Goncharova I, Sullivan MP, Forchetti C, Wilson RS, Bennett DA, et al. MRI-derived entorhinal and hippocampal atrophy in incipient and very mild Alzheimer's disease. Neurobiol Aging 2001;22:747–54.
[14] Du AT, Schuff N, Amend D, Laakso MP, Hsu YY, Jagust WJ, et al. Magnetic resonance imaging of the entorhinal cortex and hippocampus in mild cognitive impairment and Alzheimer's disease. J Neurol Neurosurg Psychiatry 2001;71:441–7.
[15] Edland SD, Xu Y, Plevak M, O'Brien P, Tangalos EG, Petersen RC, et al. Total intracranial volume: normative values and lack of association with Alzheimer's disease. Neurology 2002;59:272–4.
[16] Fischl B, Salat DH, Busa E, Albert M, Dieterich M, Haselgrove C, et al. Whole brain segmentation: automated labeling of neuroanatomical structures in the human brain. Neuron 2002;33:341–55.
[17] Folstein MF, Folstein SE, McHugh PR, Albert M. "Mini-Mental State". A practical method for grading the cognitive state of patients for the clinician. J Psychiatr Res. 1975;12:189–98.
[18] Förstl H, Burns A, Jacoby R. Quantitative CT scan analysis in senile dementia of the Alzheimer type. I: Computerized planimetry of cerebrospinal fluid areas. Int J Geriatr Psychiatry 1991;6:709–13.
[19] Förstl H, Zerfass R, Geiger-Kabisch C, Sattel H, Besthorn C, Hentschel F. Brain atrophy in normal ageing and Alzheimer's disease. Volumetric discrimination and clinical correlations. Br J Psychiatry 1995;167:739–46.
[20] Fox NC, Crum WR, Scahill RI, Stevens JM, Janssen JC, Rossor MN. Imaging of onset and progression of Alzheimer's disease with voxel-compression mapping of serial magnetic resonance images. Lancet 2001;358:201–5.
[21] Fox NC, Warrington EK, Freeborough PA, Hartikainen P, Kennedy AM, Stevens JM, et al. Presymptomatic hippocampal atrophy in Alzheimer's disease. A longitudinal MRI study. Brain 1996;119(Pt 6):2001–7.
[22] Free SL, Bergin PS, Fish DR, Cook MJ, Shorvon SD, Stevens JM. Methods for normalization of hippocampal volumes measured with MR. AJNR Am J Neuroradiol 1995;16:637–43.
[23] Geroldi C, Laakso MP, DeCarli C, Beltramello A, Bianchetti A, Soininen H, et al. Apolipoprotein E genotype and hippocampal asymmetry in Alzheimer's disease: a volumetric MRI study. J Neurol Neurosurg Psychiatry 2000;68:93–6.
[24] Gertz HJ, Xuereb J, Huppert FA, Brayne C, McGee MA, Paykel ES, et al. Examination of the validity of the hierarchical model of neuropathological staging in normal aging and Alzheimer's disease. Acta Neuropathol (Berl) 1998;95:154–6.
[25] Gertz HJ, Xuereb JH, Huppert FA, Brayne C, Kruger H, McGee MA, et al. The relationship between clinical dementia and neuropathological staging (Braak) in a very elderly community sample. Eur Arch Psychiatry Clin Neurosci 1996;246:132–6.
[26] Gosche KM, Mortimer JA, Smith CD, Markesbery WR, Snowdon DA. Hippocampal volume as an index of Alzheimer neuropathology: findings from the Nun Study. Neurology 2002;58:1476–82.
[27] Hojjatoleslami A, Kruggel F, von Cramon DY. Segmentation of white matter lesions from volumetric MR-images. In: Taylor C, Colchester A, editors. Medical image computing and computer-assisted intervention. Heidelberg: Springer; 1999. p. 52–61.
[28] Hughes CP, Berg L, Danziger WL, Coben LA, Martin RL. A new clinical scale for the staging of dementia. Br J Psychiatry 1982;140:566–72.
[29] Jack Jr CR, Twomey CK, Zinsmeister AR, Sharbrough FW, Petersen RC, Cascino GD. Anterior temporal lobes and hippocampal formations: normative volumetric measurements from MR images in young adults. Radiology 1989;172:549–54.

[30] Jenkins R, Fox NC, Rossor AM, Harvey RJ, Rossor MN. Intracranial volume and Alzheimer disease: evidence against the cerebral reserve hypothesis. Arch Neurol 2000;57:220–4.
[31] Katzman R, Terry R, DeTeresa R, Brown T, Davies P, Fuld P, et al. Clinical, pathological, and neurochemical changes in dementia: a subgroup with preserved mental status and numerous neocortical plaques. Ann Neurol 1988;23:138–44.
[32] Kaye JA, Swihart T, Howieson D, Dame A, Moore MM, Karnos T, et al. Volume loss of the hippocampus and temporal lobe in healthy elderly persons destined to develop dementia. Neurology 1997;48:1297–304.
[33] Killiany RJ, Gomez-Isla T, Moss M, Kikinis R, Sandor T, Jolesz F, et al. Use of structural magnetic resonance imaging to predict who will get Alzheimer's disease. Ann Neurol 2000;47:430–9.
[34] Killiany RJ, Hyman BT, Gomez-Isla T, Moss MB, Kikinis R, Jolesz F, et al. MRI measures of entorhinal cortex vs. hippocampus in preclinical AD. Neurology 2002;58:1188–96.
[35] Kim JM, Stewart R, Shin IS, Yoon JS. Limb length and dementia in an older Korean population. J Neurol Neurosurg Psychiatry 2003;74:427–32.
[36] Kruggel F, Lohmann G. BRIAN—a toolkit for the analysis of multimodal brain datasets. In: Lemke HU et al., editors. Computer assisted radiology; 1996. p. 323–8.
[37] Laakso MP, Lehtovirta M, Partanen K, Riekkinen PJ, Soininen H. Hippocampus in Alzheimer's disease: a 3-year follow-up MRI study. Biol Psychiatry 2000;47:557–61.
[38] Levy R. Aging-associated cognitive decline. Int Psychogeriatr 1994;6:63–8.
[39] MacLullich AM, Ferguson KJ, Deary IJ, Seckl JR, Starr JM, Wardlaw JM. Intracranial capacity and brain volumes are associated with cognition in healthy elderly men. Neurology 2002;59:169–74.
[40] Mortimer J, Snowdon D, Markesbery W. Head circumference, education and risk of dementia: findings from the nun study. J Clin Exp Neuropsychol 2003;25:671–9.
[41] Nagy Z, Hindley NJ, Braak H, Braak E, Yilmazer-Hanke DM, Schultz C, et al. Relationship between clinical and radiological diagnostic criteria for Alzheimer's disease and the extent of neuropathology as reflected by 'stages': a prospective study. Dement Geriatr Cogn Disord 1999;10:109–14.
[42] Nagy Z, Jobst KA, Esiri MM, Morris JH, King EM, MacDonald B, et al. Hippocampal pathology reflects memory deficit and brain imaging measurements in Alzheimer's disease: clinicopathologic correlations using three sets of pathologic diagnostic criteria. Dementia 1996;7:76–81.
[43] Palmer K, Wang HX, Backman L, Winblad B, Fratiglioni L. Differential evolution of cognitive impairment in nondemented older persons: results from the Kungsholmen Project. Am J Psychiatry 2002;159:436–42.
[44] Pearson RC, Esiri MM, Hiorns RW, Wilcock GK, Powell TP. Anatomical correlates of the distribution of the pathological changes in the neocortex in Alzheimer disease. Proc Natl Acad Sci USA 1985;82:4531–4.
[45] Petersen RC, Doody R, Kurz A, Mohs RC, Morris JC, Rabins PV, et al. Current concepts in mild cognitive impairment. Arch Neurol 2001;58:1985–92.
[46] Reiman EM, Uecker A, Caselli RJ, Lewis S, Bandy D, de Leon MJ, et al. Hippocampal volumes in cognitively normal persons at genetic risk for Alzheimer's disease. Ann Neurol 1998;44:288–91.
[47] Riedel-Heller SG, Busse A, Aurich C, Matschinger H, Angermeyer MC. Incidence of dementia according to DSM-III-R and ICD-10: results of the Leipzig Longitudinal Study of the Aged (LEILA 75+), Part 2. Br J Psychiatry 2001;179:255–60.
[48] Riedel-Heller SG, Schork A, Matschinger H, Angermeyer MC. Recruitment procedures and their impact on the prevalence of dementia. Results from the Leipzig Longitudinal Study of the Aged (LEILA 75+). Neuroepidemiology 2000;19:130–40.
[49] Riley KP, Snowdon DA, Markesbery WR. Alzheimer's neurofibrillary pathology and the spectrum of cognitive function: findings from the Nun Study. Ann Neurol 2002;51:559–66.
[50] Ritchie K, Artero S, Touchon J. Classification criteria for mild cognitive impairment: a population-based validation study. Neurology 2001;56:37–42.
[51] Roman GC, Tatemichi TK, Erkinjuntti T, Cummings JL, Masdeu JC, Garcia JH, et al. Vascular dementia: diagnostic criteria for research studies. Report of the NINDS–AIREN International Workshop. Neurology 1993;43:250–60.
[52] Sachs L. Angewandte statistik (Applied statistics). Berlin: Springer; 1999.
[53] Schofield PW, Logroscino G, Andrews HF, Albert S, Stern Y. An association between head circumference and Alzheimer's disease in a population-based study of aging and dementia. Neurology 1997;49:30–7.
[54] Stieler J, Lederer C, Brückner MK, Wolf H, Holzer M, Gertz HJ, et al. Impairment of mitogenic activation of peripheral blood lymphocytes in Alzheimer's disease. Neuroreport 2001;12:3969–72.
[55] Tohgi H, Takahashi S, Kato E, Homma A, Niina R, Sasaki K, et al. Reduced size of right hippocampus in 39- to 80-year-old normal subjects carrying the apolipoprotein E epsilon4 allele. Neurosci Lett 1997;236:21–4.
[56] van der Flier WM, van den Heuvel DM, Weverling-Rijnsburger AW, Bollen EL, Westendorp RG, van Buchem MA, et al. Magnetization transfer imaging in normal aging, mild cognitive impairment, and Alzheimer's disease. Ann Neurol 2002;52:62–7.
[57] Visser PJ, Scheltens P, Verhey FR, Schmand B, Launer LJ, Jolles J, et al. Medial temporal lobe atrophy and memory dysfunction as predictors for dementia in subjects with mild cognitive impairment. J Neurol 1999;246:477–85.
[58] Visser PJ, Verhey FR, Hofman PA, Scheltens P, Jolles J. Medial temporal lobe atrophy predicts Alzheimer's disease in patients with minor cognitive impairment. J Neurol Neurosurg Psychiatry 2002;72:491–7.
[59] Wahlund LO, Basun H, Almkvist O, Julin P, Axelman K, Shigeta M, et al. A follow-up study of the family with the Swedish APP 670/671 Alzheimer's disease mutation. Dement Geriatr Cogn Disord 1999;10:526–33.
[60] Wolf H, Grunwald M, Kruggel F, Riedel-Heller SG, Angerhofer S, Hojjatoleslami A, et al. Hippocampal volume discriminates between normal cognition; questionable and mild dementia in the elderly. Neurobiol Aging 2001;22:177–86.
[61] Wolf H, Kruggel F, Hensel A, Wahlund LO, Arendt T, Gertz HJ. The relationship between head size and intracranial volume in elderly subjects. Brain Res 2003;973:74–80.
[62] Wolf H, Wahlund LO, Julin P, Kruggel F, Riedel-Heller S, Hensel A, et al. Is bigger better? Evidence for the cerebral reserve hypothesis of dementia in two large, independently examined samples. Neurobiol Aging 2002;(Suppl):1688.
[63] World Health Organisation. ICD-10 classification for mental and behavioural disorders. Diagnostic criteria for research. Geneva: WHO; 1993.
[64] Xu Y, Jack Jr CR, O'Brien PC, Kokmen E, Smith GE, Ivnik RJ, et al. Usefulness of MRI measures of entorhinal cortex versus hippocampus in AD. Neurology 2000;54:1760–7.
[65] Yamaguchi S, Meguro K, Shimada M, Ishizaki J, Yamadori A, Sekita Y. Five-year retrospective changes in hippocampal atrophy and cognitive screening test performances in very mild Alzheimer's disease: the Tajiri Project. Neuroradiology 2002;44:43–8.
[66] Zaudig M, Mittelhammer J, Hiller W. SIDAM—a Structured Interview for the Diagnosis of Dementia of the Alzheimer type, Multi-infarct dementia and dementias of other aetiology according to ICD-10 and DSM-III-R. Psychol Med 1991;21:223–5.