Eur J Nucl Med Mol Imaging (2008) 35:2191–2202
DOI 10.1007/s00259-008-0869-z

ORIGINAL ARTICLE

Principal component analysis of FDG PET in amnestic MCI

Flavio Nobili · Dario Salmaso · Silvia Morbelli · Nicola Girtler · Arnoldo Piccardo · Andrea Brugnolo · Barbara Dessi · Stig A. Larsson · Guido Rodriguez · Marco Pagani

Received: 13 March 2008 / Accepted: 1 June 2008 / Published online: 22 July 2008

Abstract
Purpose The purpose of the study is to evaluate the combined accuracy of episodic memory performance and ^{18}F-FDG PET in identifying patients with amnestic mild cognitive impairment (aMCI) converting to Alzheimer's disease (AD), aMCI non-converters, and controls.
Methods Thirty-three patients with aMCI and 15 controls (CTR) were followed up for a mean of 21 months. Eleven patients developed AD (MCI/AD) and 22 remained with aMCI (MCI/MCI). ^{18}F-FDG PET volumetric regions of interest underwent principal component analysis (PCA) that identified 12 principal components (PC), expressed by coarse component scores (CCS). Discriminant analysis was performed using the significant PCs and episodic memory scores.
Results PCA highlighted relative hypometabolism in PC5, including bilateral posterior cingulate and left temporal pole, and in PC7, including the bilateral orbitofrontal cortex, both in MCI/MCI and MCI/AD vs CTR. PC5 itself plus PC12, including the left lateral frontal cortex (LFC: BAs 44, 45, 46, 47), were significantly different between MCI/AD and MCI/MCI. By a three-group discriminant analysis, CTR were more accurately identified by PET-CCS + delayed recall score (100%), MCI/MCI by PET-CCS + either immediate or delayed recall scores (91%), while MCI/AD was identified by PET-CCS alone (82%). PET increased by 25% the correct allocations achieved by memory scores, while memory scores increased by 15% the correct allocations achieved by PET.
Conclusion Combining memory performance and ^{18}F-FDG PET yielded a higher accuracy than each single tool in identifying CTR and MCI/MCI. The PC containing bilateral posterior cingulate and left temporal pole was the hallmark of MCI/MCI patients, while the PC including the left LFC was the hallmark of conversion to AD.

Keywords ^{18}F-FDG PET · Episodic memory · Mild cognitive impairment · Alzheimer's disease

F. Nobili (✉) · N. Girtler · A. Brugnolo · B. Dessi · G. Rodriguez
Clinical Neurophysiology, Department of Endocrinological and Medical Sciences, University of Genoa,
Viale Benedetto XV, 6,
16132 Genoa, Italy
e-mail: flaviomariano.nobili@hsanmartino.it

F. Nobili · N. Girtler · A. Brugnolo · B. Dessi · G. Rodriguez
Alzheimer Evaluation Unit, S. Martino Hospital,
Genoa, Italy

F. Nobili · N. Girtler · A. Brugnolo · B. Dessi · G. Rodriguez
Head–Neck Department, S. Martino Hospital,
Genoa, Italy

D. Salmaso · M. Pagani
Institute of Cognitive Sciences and Technologies, CNR,
Rome, Italy

D. Salmaso · M. Pagani
Institute of Cognitive Sciences and Technologies, CNR,
Padua, Italy

S. Morbelli
Nuclear Medicine Unit, Department of Internal Medicine,
University of Genoa,
Genoa, Italy

A. Piccardo
Nuclear Medicine Unit, Department of Imaging Diagnostics,
Galliera Hospital,
Genoa, Italy

S. A. Larsson · M. Pagani
Department of Nuclear Medicine, Karolinska Hospital,
Stockholm, Sweden

Introduction

The need to accurately identify those patients with amnestic mild cognitive impairment (aMCI) who are already affected by Alzheimer's disease (AD) but are not demented yet is emerging [1, 2]. Potentially effective disease-modifying drugs are currently under active investigation and need to be tested in AD patients as early as possible. In fact, the aMCI population has been proven to have a high risk to develop AD within few years [3]. However, the aMCI population is highly heterogeneous, including both patients who remain stable over time and who revert to normalcy [4].

aMCI is currently identified by both a reported and an objective memory impairment, either associated (amnestic multi-domain) or not (amnestic single-domain) with a slight impairment in other cognitive areas [5, 6], although daily functions are mainly preserved [5]. The deficit in episodic memory is the neuropsychological core and can be ascertained by tools assessing immediate and delayed verbal recall, which have been shown to be useful in discriminating aMCI converters from non-converters [7]. However, although rather sensitive, impaired episodic memory is not specific for early AD since it can be also found in other forms of cognitive impairment, such as depression [8], subcortical dementia and fronto-temporal dementia [9]. Moreover, floor effects on memory tests in patients with aMCI may make recall measures relatively insensitive to longitudinal changes [1].

It is a common opinion that supportive features are needed to improve the diagnostic accuracy of pre-dementia AD. Such features have been identified in some 'biomarkers', mainly including increased phosphorylated tau protein and decreased Aβ 1–42 amyloid cerebrospinal fluid levels, hypometabolism in associative cortex in ^{18}F-fluorodeoxyglucose positron emission tomography (^{18}F-FDG PET, simply 'PET' from now on) and evidence of medial temporal lobe atrophy in magnetic resonance imaging (MRI). The core criteria of gradual onset and progressive impairment of episodic memory during more than 6 months must be sustained by significant changes in at least one of these biomarkers for the diagnosis of AD before dementia becomes 'probable' [2].

The challenge is open on which biomarker can add more value to episodic memory tests in giving the most accurate identification. Anchisi et al. [10] utilised PET and the California Verbal Learning Test-long delay free recall together and showed an improvement in the detection of aMCI converters to AD, as compared to each of the two data as taken alone. Good discriminative value has been obtained by combining neuropsychology and MRI atrophy analyses [11] or by combining neuropsychology, PET and apolipoprotein E (ApoE) genotype [12]. However, the added value of neuropsychology to PET has not been confirmed by other studies [13].

As for PET studies in aMCI converters and non-converters to AD, only some investigations employed normal controls [10, 14–16]. However, these studies were based on voxel-by-voxel comparison of metabolic data and assessed differences based on two-group comparisons (various combinations of MCI non-converters, MCI converters and controls), while a three-group analysis would be more meaningful, better analyzing possible areas of overlap. In fact, the heterogeneous population of MCI patients tends to overlap with normal ageing on the one hand and with AD on the other hand [6]. There is growing evidence that the relationship between normal aging and AD-related changes could be considered on a *continuum* [17]. Given current evidence, it is likely that the mechanisms underpinning cognitive decline in aging vs AD are only partially distinct [18]. For instance, the influential cholinergic hypothesis suggests that changes to acetylcholine functioning and other cholinergic system abnormalities contribute to normal cognitive decline and electroencephalography (EEG) alterations, as well as to AD [19]. Moreover, the pre-symptomatic patterns of cognitive decline are not reliably distinguished from 'normal' variation in cognitive function in late life. Finally, the neuropathological evidence of the hypothetical discontinuity between normal ageing and dementia is lacking; the best available evidence suggests that there is no boundary at all [17]. Thus, a control group of healthy elderly seems of relevance in this context.

With the aim to verify both the independent and the combined accuracy of PET and episodic memory performance in the prediction of AD, we followed up a series of aMCI patients and of healthy controls over time. Both the individual and the combined discriminant power of episodic memory scores and PET were assessed in confirmed controls (CTR), aMCI patients who remained aMCI (MCI/MCI) and aMCI patients who developed AD (MCI/AD) in a three-group analysis. For this purpose, PET data were submitted to principal component analysis (PCA) applied to volumetric regions of interest (VROI) based on Brodmann areas.

Materials and methods

Patients The study included outpatients with memory complaints in whom an objective memory deficit was demonstrated by means of neuropsychological tests. Dementia was excluded on the basis of clinical interview with the patient and caregiver, using the Mini-Mental State Examination (MMSE) [20] for general cognition, questionnaires for the Activities of Daily Living (ADL) [21] and

instrumental ADL (IADL) [22] and Clinical Dementia Rating (CDR) scale that was 0.5 in all patients.

Patients underwent a standard battery of blood count, blood chemical examinations and urinalysis, according to the commonly followed rules to exclude secondary causes of cognitive impairment. Presence of analphabetism, major vision disturbances, psychiatric illnesses, epilepsy, major head trauma, Parkinsonism, previous stroke or TIA and brain masses were other exclusion criteria. A mild depressive trait, as ascertained by the 15-item Geriatric Depression Scale (GDS), was not an exclusion criterion. Neuropsychiatric symptoms were assessed by interviewing the informant with the Neuropsychiatric Inventory (NPI) [23]. Patients scoring higher than 0 on the delusion and the hallucination NPI items were excluded.

MRI was performed in all patients by means of a 1.5-T equipment. Only patients with MRI evidence of major stroke were excluded, while white matter hyperintensities, leukoaraiosis and lacunae were not exclusion criteria. Medial temporal lobe (MTL) atrophy and subcortical cerebrovascular disease were visually rated by means of the five-point Scheltens' scale [24] and the Age-Related White Matter Changes (ARWMC) scale [25], respectively. As for ARWMC, a global score for each subject was computed by summing up the scores obtained in each brain region. The same rater, who was blind to clinical diagnosis, scored both MTL atrophy and ARWMC.

The initial group comprised 36 aMCI patients (22 women, 14 men; mean age: 76.0±5.5 years). These patients underwent a neuropsychological battery, including evaluation of (1) verbal episodic memory (immediate and delayed recall, IR and DR from now on) by the six-trial Selective Reminding test (SRT) [26]; (2) visuo-motor abilities, divided and attentional shifting by the Trail-making test, forms A (TMT-A) and B (TMT-B); (3) categorical verbal fluency (2′ test for animals); (4) visuo-constructional abilities by the copying figures test, including simple copy and copying with guiding landmarks of the Mental Deterioration Battery [27]; (5) abstract and logical reasoning by the Raven's PM38 matrices (set A–D, according to Spinnler and Tognoni [28]); (6) executive attention by the Stroop color-word test (correct items achieved in 30 s, according to Barbarotto et al. [29]). The Clock completion test (as evaluated according to Watson et al. [30]) was used as a mixed measure of visuospatial abilities and executive functions and the Symbol-Digit test [31] as a mixed measure of working memory and executive functions.

A z score lower than −1.5, computed on the normative database of each test, corrected for age and education, was established for impairment in a specific cognitive domain. According to the Petersen's criteria [5], patients with a z score lower than −1.5 either on the IR or DR of the SRT (single-domain aMCI) as well as patients scoring less than −1.5 both on SRT and in other cognitive domains (multi-domain aMCI) were considered.

All patients were carefully treated for systemic co-morbidity; drugs known to depress brain synaptic transmission, such as benzodiazepines and tryciclic antidepressant, were withdrawn. Then, patients started to be followed up with clinical examination (also including MMSE, ADL and IADL questionnaires and CDR) every 6 months. A follow-up time of at least 1 year was available in all patients. During the follow-up period, two patients no longer showed any cognitive objective deficit after 26 and 35 months, respectively, and were excluded from the study. Another patient developed fronto-temporal dementia, according to the current criteria [32], after 1 year and was excluded.

Thus, the final study group included 33 patients (Table 1). Twenty (61%) patients were affected by mild to moderate hypertension and were on anti-hypertensive drugs; 16 (48%) presented hypercholesterolemia, and five of them received statins administered orally. Ischaemic heart disease was present in four (12%) patients and diabetes mellitus in two (6%). Family history of dementia in first-degree relatives was positive in 17 (52%) patients. ApoE genotype was available in 22 patients; heterozygous ApoE ε4 genotype was present in nine (41%). The modified Hachinski ischaemic scale [33] was ≤2 in all patients.

During the follow-up (mean 21.1±10.9 months), 11 patients (MCI/AD) developed dementia of the AD type, according to the NINCDS-ADRDA [34] and DSM-IV criteria, while 22 patients were confirmed to have aMCI (MCI/MCI). The mean annual conversion rate to AD was approximately 17% (11/36=31% in 21.7 months mean follow-up time; Table 1).

Controls The protocol received the approval of the local ethics committee. Control subjects were healthy volunteers who gave their informed consent, recruited during university courses dedicated to elderly people. Their healthy condition was carefully checked by means of general medical history, clinical examination and the same exclusion criteria as for patients, with the exception of cognitive complaints. MMSE was performed, and only subjects with a normal score (i.e. ≥26) were considered. Moreover, only subjects with a CDR of 0 were included. These subjects underwent the same neuropsychological battery as patients and brain MRI [all but four underwent CT because of metallic devices (n=2) or claustrophobia (n=2)]. Seventeen subjects (four men and 13 women, aged 62–83 years, mean 70.6±7.1) were included.

Control subjects (CTR) were asked to continue their evaluation over time. Two of them disclosed significant

Table 1 Main baseline demographic and clinical features of the three groups, as identified at follow-up visit

	Groups			*p*			
	CTR	MCI/MCI	MCI/AD	Group effect	CTR vs MCI/MCI	CTR vs MCI/AD	MCI/MCI vs MCI/AD
N	15	22	11				
Age (years)	70.0±7.0	74.6±5.4	77.3±4.8		–*	–*	
Sex M/F	4/11	11/11	2/9	n.s.			
Apo ε4 allele	n.a.	5/14 (36%)	4/8 (50%)	n.s.			
Education (years)	11.0±4.4	8.8±4.7	8.5±3.9	n.s.			
GDS	3.3±2.3	3.8±2.9	3.5±2.6	n.s.			
NPI	n.a.	8.8±8.7	7.2±9.1	n.s.			
Follow-up time (months)	20.2±8.5	20.6±10.3	22.2±12.4	n.s.			
Baseline MMSE	29.0±1.1	27.4±2.0	27.6±1.4		–*	–*	
MMSE at follow-up	28.9±1.0	27.5±1.6	24.0±1.7		–*	–*	–*
MTL atrophy score	0.53±0.64	2.14±0.94	2.73±0.65		–*	–*	–*
ARWMC score	2.13±3.23	5.05±4.36	6.18±3.60		–*	–*	

GDS 15-item Geriatric Depression Scale, *NPI* Neuropsychiatric Inventory, *MMSE* Mini-Mental State Examination, *MTL* medial temporal lobe, *ARWMC* Age-Related White Matter Changes, *n.a.* not available, *n.s.* not significant
*$p<0.05$

impairment in two neuropsychological tests (episodic memory and visuoconstruction) at follow-up visit and a CDR of 0.5; thus, they were excluded from the study. At last, the CTR group consisted of 15 subjects, ranging in age from 58 to 83 years. A mild to moderate hypertension was present in four (27%) subjects who were on anti-hypertensive drugs; mild hypercholesterolemia was found in eight (53%) subjects, four of whom were receiving statins administered orally. A history of ischaemic heart disease was found in one subject (6%). Four (27%) subjects had a positive family history for dementia in first-degree relatives.

Tables 1 and 2 report the main baseline demographics and mean neuropsychological scores of the three groups of 15 CTR subjects, the 22 MCI/MCI and of the 11 MCI/AD patients.

^{18}F-FDG PET PET was performed within 3 months from the clinical–neuropsychological examination (mean 29.9 days in patients and 29.8 days in CTR). Subjects fasted for at least 6 h. Before radiopharmaceutical injection, blood glucose was checked and was <140 mg/dl in all cases. After a 10-min rest in a silent and obscured room,

Table 2 Baseline neuropsychological test scores (mean ± SD) of the three groups, as identified at follow-up visit

	Groups			*p*		
	CTR	MCI/MCI	MCI/AD	Group effect	CTR vs MCI/MCI	CTR vs MCI/AD
N	15	22	11			
CCT	1.1±1.9	2.6±2.6	2.8±2.6	n.s.		
SRT IR	48.4±9.4	30.0±4.8	28.7±2.2		–*	–*
SRT DR	7.0±2.2	1.6±1.3	1.4±1.0		–*	–*
Verbal fluency	27.6±5.4	17.7±4.8	17.3±4.2		–*	–*
TMT-A (s)	46.3±12.4	69.7±28.8	59.2±12.6		–*	–*
TMT-B (s)	128.6±51.2	228.0±117.7	234.7±91.7		–*	–*
Figure copying: simple	9.0±1.8	8.7±1.3	8.8±0.8	n.s.		
Figure copying: with guiding landmarks	65.6±3.8	64.5±5.8	67.9±1.8	n.s.		
Stroop color	39.3±7.6	34.0±6.3	34.7±7.2		–*	
Stroop color-word	17.1±4.9	12.1±4.6	12.2±4.0		–*	–*
Raven's PM38	31.1±8.9	26.7±7.6	26.1±8.5	n.s.		
Digit symbol	38.2±9.7	25.0±9.0	24.6±1.8		–*	–*

There was no significant neuropsychological test difference between MCI/MCI and MCI/AD.
CCT Clock completion test, *SRT* Selective Reminding test, *IR* immediate recall, *DR* delayed recall, *verbal fluency* categorical fluency for animals (2-min test), *TMT-A* Trail-making test, form A, *TMT-B* Trail-making test, form B, *n.s.* not significant
*$p<0.05$

with eyes closed and ears unplugged, subjects were injected with approximately 370 MBq of ^{18}F-FDG via a venous cannula, according to the guidelines of the European Association of Nuclear Medicine [35]. They remained in the room for 30 min after injection, then they were moved to the PET room where scanning started approximately 45 min after injection and lasted 20 min. A polycarbonate head holder was used to reduce head movements during the scan. Images were acquired by a 'Discovery ST' PET-CT equipment (GE Healthcare, USA) on a 128×128×64 matrix (isotropic voxel of 2.34 mm) in a two-dimensional mode with a total axial field of view of 15 cm and no interplane gap space. Images were reconstructed by a OSEM algorithm, 16 subsets and two iterations. Dicom files were exported and converted to Analyse files.

Standardisation software Computerized Brain Atlas (CBA; Applied Medical Imaging©, Uppsala, Sweden) is a software tool for analysis of neuroimaging data [36]. All image sets were spatially normalised into the stereotactic space of the atlas by using the global polynomial transformation [37]. It consists of translations, rotations and linear scaling along and around each of the three image axes. It also contains 18 nonlinear shape-deforming parameters, which make it possible to individualize the brain shape. In this study, a fully automatic method was used in a first step, in which all scans were registered to a dedicated PET template [38]. Then, further manual adjustments were performed, when needed, to optimise the fitting of the CBA contours to both external and internal (brain ventricles) borders.

For evaluation and statistical analysis of the reformatted data sets, 25 VROIs were selected in each hemisphere in order to cover most of the cortical and subcortical brain structures likely involved in MCI-early AD, on the basis of current literature [39, 40]. These regions correspond to Brodmann areas (BA) and numeration in prefrontal (BA9, BA10, BA46), orbitofrontal (BA11, BA47), frontal (BA6, BA8, BA44, BA45), parietal (BA5, BA7, BA39, BA40) and temporal (BA21, BA37, BA38, hippocampus) cortex. Four regions, representing primary and associative auditory cortex (AUD=BA22+41+42+52) were merged into one VROI. The remaining regions corresponded to anterior (BA24, BA32) and posterior (BA31) cingulate, occipital (BA17) cortex, basal ganglia and thalami.

CBA has the advantages of including in the analysis areas sharing anatomo-functional characteristics (the VROIs that correspond to BA were originally classified according to the brain cyto-architectonics) and of producing data for processing that contain fewer independent variables. In the case of the present work, it allowed to further analyse the 50 VROIs focussing information in the principal components (PCs).

In order to obtain a set of normalised relative metabolic data, a scaling factor was computed, averaging all brain voxels data and setting the global brain average to the conventional value of 50 'uptake units'. All regional uptake values were then related to this value.

Statistical analysis After adaptation and definitions of VROIs using CBA, the ^{18}F-FDG uptake data of all subjects were exported to a statistical package (Systat 10, 2000) for statistical analysis.

PCA was performed on all the 48 subjects and was based on the 50 VOIs. Briefly, PCA is a data-driven technique (i.e. there is no a priori model or hypothesis) that transforms the original variables (VROI uptake values in this case) by clustering them into factors (PCs; see [41] for a discussion). The PCs are new variables, defined as linear functions of the original variables, solving the issue of 'multicollinearity', that is, the inter-correlation between the original variables. VROI values with higher loadings within a PC are highly correlated to one another and give insight into what the PC represents. The PCs are uncorrelated to one another, and each of them explains a different part of the total variance of data set. PC values can be computed for each subject as factor scores, better expressed as coarse component scores (CCS). The stability of the PCA was evaluated by means of the T2 Hotelling's test. Further methodological details are reported in Appendix 1. PCA applied to SPECT and PET data was proven to yield meaningful results in neurodegenerative diseases both using VROI- [42–44] and voxel-based [45, 46] approaches.

ANOVA was applied to PET-CCS values to test the statistical significance of PET differences, considering the 'group' as a between-subject variable. Significance level was set to $p<0.05$. In a first analysis, the two groups of 33 aMCI and 15 CTR subjects, based on the baseline diagnostic classification, were considered. Instead, in a second analysis, the variable group included the three diagnostic categories at follow-up, i.e. MCI/MCI, MCI/AD and CTR.

Moreover, different sets of discriminant analysis were performed to estimate the concordance between groupings carried out on the basis of clinical diagnosis and PET data. In a first step, the two groups of 33 aMCI and 15 CTR subjects, based on the baseline diagnostic classification, were considered. Then, in a second step, group membership was analysed according to the clinical diagnosis at follow-up, i.e. MCI/AD, MCI/MCI and CTR. In the latter case, predictor variables were IR, DR and those PET-CCS yielding a significant difference between groups.

The outcome of discriminant analysis resulted in discriminant functions that are the linear combinations of variables included in the analysis. The relevance of a discriminant function is given by its canonical correlation, i.e. the total variance explained by each discrimi-

nant function. On the basis of the scores computed for each subject, a classification matrix was computed to evaluate the efficiency of the analysis. Significance of discriminant analysis was tested by means of the 'approx. *F* value'.

Results

PC structure PCA identified 12 PCs accounting for the 81% of the total variance. The T2 Hotelling test showed no outliers ($p<0.01$). The VROIs with the higher factor loading for each PC are shown in Table 3; they were used for interpreting the meaning of each PC and computing the PET-CCS values. Concerning the factor loadings of each PC (data not shown), in four instances (PC4, PC6, PC8 and PC11), there was a negative correlation between VROIs and PCs, expressing an inverse relationship with the overall metabolism. Three VROIs were excluded by PCA in the final solution (BA08R, BA21L, BA46R). Furthermore, all the correlations between PET-CCS and the corresponding component scores were highly significant ($r=0.832$; $p<0.001$).

Comparison between CTR and aMCI according to baseline classification Based on the diagnostic classification at baseline, two PCs disclosed significantly lower PET-CCS values in the 33 aMCI patients vs the 15 CTR subjects. PC5 included bilateral BA31 and left BA38 (mean value in aMCI vs CTR: 0.50 vs 0.72; $F(1, 46)=13.849$, $p=0.001$) and PC7 including orbitofrontal cortex (bilateral BA11 and the right BA47; mean value in aMCI vs CTR: 0.58 vs 0.80; $F(1, 46)=14.169$, $p<0.001$). The discriminant analysis on PET-CCS data of PC5 and PC7 yielded an 87% specificity and a 79% sensitivity in allocating the 33 aMCI patients and the 15 CTR to the correct group (approx. $F(2, 45)=13.848$, $p<0.001$; canonical correlation=0.617).

Comparison among CTR, MCI/MCI and MCI/AD according to follow-up classification (Tables 3 and 4) A significant group effect was found, as in the previous comparison, for both PC5 and PC7. Considering these two PCs, both MCI/MCI and MCI/AD groups showed lower PET-CCS values than CTR. Moreover, significantly lower PET-CCS values were found in MCI/AD as compared to MCI/MCI for PC12, including the left lateral frontal cortex (LFC; BA44, BA45, BA46, BA47) and for

Table 3 PCA structure, mean and SD of PET-CCS values and statistical differences among the three groups according to the diagnosis at follow-up

PC	VROI with high loading on the PC	PET-CCS values						Group effect		Tukey HSD multiple comparisons		
		CTR		MCI/MCI		MCI/AD				CTR vs MCI/MCI	CTR vs MCI/AD	MCI/MCI vs MCI/AD
		Mean	SD	Mean	SD	Mean	SD	$F(2, 45)$	p	p	p	p
1	BA9R BA10R BA10L	0.51	0.29	0.57	0.22	0.55	0.20					
2	AUDL BA6R BA6L BA8L BA9L BA37L	0.67	0.12	0.63	0.19	0.63	0.25					
3	BA39L BA39R BA40L BA40R	0.57	0.26	0.49	0.18	0.39	0.21					
4	PUTL PUTR THALL THALR	0.57	0.26	0.59	0.21	0.39	0.31					
5	BA31L BA31R BA38L	**0.72**	**0.16**	**0.58**	**0.15**	**0.36**	**0.19**	**15.129**	**0.000**	**0.004**	**0.000**	**0.002**
6	BA05L BA5R BA7L BA7R	0.47	0.27	0.61	0.20	0.59	0.26					
7	BA11L BA11R BA47R	**0.80**	**0.16**	**0.58**	**0.22**	**0.58**	**0.15**	**6.931**	**0.002**	**0.003**	**0.005**	
8	HIPL HIPR	0.48	0.18	0.49	0.24	0.55	0.18					
9	BA24L BA24R BA32L BA32R CAUDL CAUDR	0.54	0.14	0.55	0.24	0.58	0.25					
10	AUDR BA21R BA38R BA44R BA45R	0.66	0.19	0.58	0.21	0.53	0.28					
11	BA19L BA19R BA37R	0.47	0.19	0.51	0.26	0.52	0.27					
12	BA44L BA45L BA46L BA47L	**0.54**	**0.22**	**0.63**	**0.18**	**0.34**	**0.25**	**6.869**	**0.002**			**0.002**

The 12 PCs explain the 81% of total variance. The *F* and the significance *p* values are reported for group comparisons (the three PC yielding significant differences are in bold). PCA excluded from the final solution BA08R, BA21L, BA46R

Caud Caudate nucleus, *Put* putamen, *Thal* thalamus, *Hip* hippocampus, *Aud* auditory cortex, *BA* Brodmann area, *L* left, *R* right

Table 4 Results of the three-group discriminant analysis, employing episodic memory test alone (either immediate or delayed recall), ^{18}F-FDG PET alone (coarse component scores of the significant PCs) and the combined analysis with the two tools

Clinically assessed group membership	Predicted group membership				% subjects correctly classified	Statistic and canonical correlations (CC) for DF1 and DF2
		CTR	MCI/MCI	MCI/AD		
Immediate recall (IR)						Approx. $F(2, 45)=38.58$, $p<0.001$; CC=0.795
CTR	CTR	12	3	0	80	
Patients	MCI/MCI	1	13	8	59	
	MCI/AD	0	4	7	64	
Delayed recall (DR)						Approx. $F(2, 45)=53.11$, $p<0.001$; CC=0.838
CTR	CTR	14	1	0	93	
Patients	MCI/MCI	1	10	11	45	
	MCI/AD	0	5	6	55	
FDG PET (CCS values)						Approx. $F(6, 86)=9.49$, $p<0.001$; CC=0.708; 0.523
CTR	CTR	12	2	1	80	
Patients	MCI/MCI	4	16	2	73	
	MCI/AD	0	2	9	82	
PET-CCS + IR						Approx. $F(8, 84)=20.09$, $p<0.001$; CC=0.903; 0.601
CTR	CTR	14	1	0	93	
Patients	MCI/MCI	0	20	2	91	
	MCI/AD	0	2	9	82	
PET-CCS + DR						Approx. $F(8, 84)=18.96$, $p<0.001$; CC=0.892; 0.613
CTR	CTR	15	0	0	100	
Patients	MCI/MCI	0	20	2	91	
	MCI/AD	0	2	9	82	

For each comparison, the percentage of correct allocations is reported. Used alone, ^{18}F-FDG PET and DR showed the highest allocation value for CTR, ^{18}F-FDG PET and either IR or DR for MCI/MCI patients and ^{18}F-FDG PET for MCI/AD patients. Used together, classification values substantially increases for both CTR and MCI/MCI patients.
DF Discriminant function.

PC5 (Table 3; Figs. 1 and 2). There was no significant correlation between age and each of the three PCs, i.e. PC5, PC7 and PC12.

In the three-group analysis, the discriminant analysis (Table 4) using episodic memory IR score correctly allocated 80% of CTR, 59% of MCI/MCI and 64% of MCI/AD (canonical correlation=0.795). Using episodic memory DR score, the corresponding values were 93% in CTR, 45% in MCI/MCI and 55% in MCI/AD (canonical correlation=0.838). With PET-CCS, the correct allocations were 80% in CTR, 73% in MCI/MCI and 82% in MCI/AD (canonical correlations, discriminant function 1=0.708; discriminant function 2=0.523).

When PET-CCS were used together with IR score, the percentage of correct allocations raised to 91% in MCI/MCI (canonical correlations, discriminant function 1=0.903, discriminant function 2=0.601). The best figures were obtained by combining PET-CCS and DR score: 100% in CTR, 91% in MCI/MCI and 82% in MCI/AD (canonical correlations, discriminant function 1=0.892, discriminant function 2=0.613). Thus, the combined use of episodic memory and PET increased correct allocations from 93% to 100% in CTR and from 73% to 91% in MCI/MCI, yielding about 25% mean increased rate of correct allocations as compared to episodic memory tests alone and about 15% increase as compared to PET alone.

Looking at Table 4 (both PET-CCS + IR and PET-CCS + DR), mis-classification only concerned the two patient groups because two MCI/MCI patients were classified within the MCI/AD group and vice versa. On the contrary, separation between CTR and patients as a whole was total (100% accuracy).

Given the known heterogeneity of MCI/MCI group, discriminant analysis was also applied just to the direct comparison between the more homogenous CTR and MCI/AD groups. This yielded an 87% specificity and a 91% sensitivity with PET-CCS data (PC5 and PC7) as used alone [approx. $F(2, 23)=41.45$, $p<0.0001$; canonical

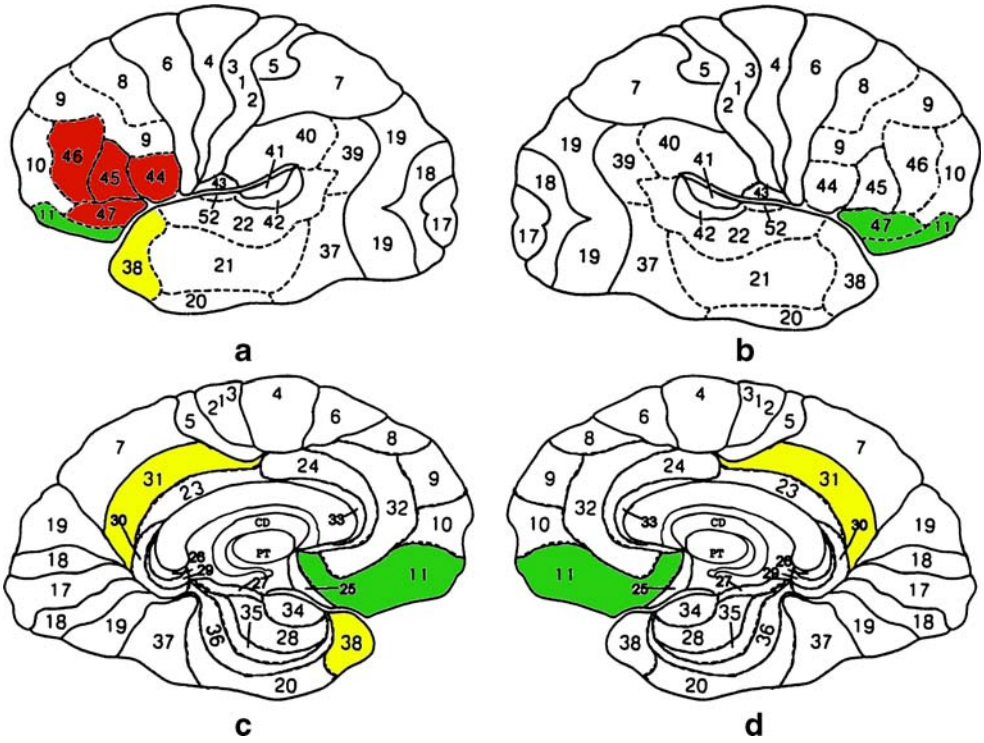

Fig. 1 The three PCs showing a significant effect at three-group comparison. In **a** and **b**, the lateral aspect of the left and right hemispheres is shown, respectively; in **c** and **d**, their medial aspect. In *yellow*, PC5 (bilateral posterior cingulate gyrus and left temporal pole); in *green*, PC7 (bilateral BA 11 and right BA47) that were significantly different between CTR and both MCI/MCI and MCI/AD; moreover, PC5 also significantly differed between MCI/MCI and MCI/AD patients. In *red*, PC12 (left lateral frontal cortex) that was significantly different between MCI/AD and MCI/MCI

correlation=0.78] and a 100% sensitivity and specificity by the combined used of PET-CCS and memory test scores [either IR or DR; PET + IR approx. $F(3, 22)=41.46$, $p<0.0001$; canonical correlation=0.92; PET + DR approx. $F(3, 22)=41.22$, $p<0.0001$; canonical correlation=0.93).

Figure 3 shows the plot of individual discriminant scores for discriminant function 1 against discriminant function 2. The discriminant analysis for the significant PET-CCS and IR gave a canonical correlation of 0.903 for discriminant function 1 and of 0.601 for discriminant function 2. The first function explained the 89% of variance and the canonical correlation yielded an 81.5% of total variance explained by our grouping. Centroid data indicated that discriminant function 1 effectively discriminated the three groups, whereas function 2 added further value in discriminating MCI/MCI and MCI/AD. IR score was the most important variable for discriminant function 1 and PC12 for discriminant function 2.

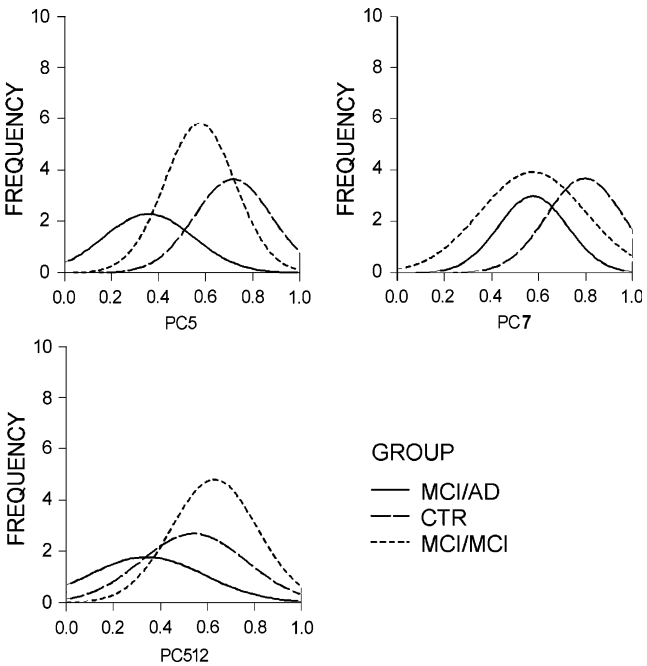

Fig. 2 Density distribution (normal curves) for the three significant PCs. PC5 (*top left*), PC7 (*top right*), PC12 (*bottom*). The number of subjects is reported on the *ordinate axis*, while PET-CCS values are on the *abscissa axis*. PC5 distinguished between MCI/AD and both MCI/MCI and CTR and MCI/MCI from CTR. PC7 identified both MCI/AD and MCI/MCI from CTR but was not different between MCI/MCI and MCI/AD. PC12 was able to distinguish MCI/AD from MCI/MCI

Discussion

This study shows that combining episodic memory evaluation and PET improved correct allocations of CTR and MCI/MCI in a three-group discriminant analysis, while in MCI/AD allocation, episodic memory did not add value to PET. The finding of a better performance of the two tools used in combination confirms the results of another study

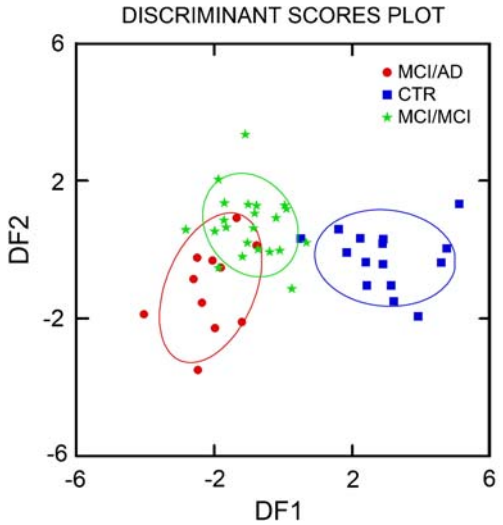

Fig. 3 Individual discriminant scores, derived from episodic memory test (immediate recall) and PET-CCS data, for discriminant function 1 (*DF1*) plotted against those of discriminant function 2 (*DF2*). There is a partial overlap between MCI/AD and MCI/MCI

[10]. Episodic memory scores as taken alone were rather ineffective in discriminating between MCI/MCI and MCI/AD patients, whereas PET reached considerably higher figures (Table 4). As a matter of fact, PET added a 7% discriminant power to memory tests in controls but a 31% in MCI/MCI and 18% in MCI/AD, yielding about 25% mean increased rate of correct allocations as compared to memory tests alone. On the other hand, the mean gain added by memory tests to PET recognition was lower but not trivial (about 15%). The deriving concept is that combining the discriminant power of two methods may improve correct allocations because each tool can detect functional impairment in some subjects in whom the other tool is not accurate enough. This is an emerging strategy, which is increasingly applied to the combination of other biomarkers, including PET, MRI and CSF biomarkers, either with or without neuropsychology [10–13, 47].

The three-group approach adopted here reflects the continuum model to represent the relationship between the functional decline in normal aging and AD. Correlative data between cognitive measures and neurophysiological data (notably quantitative EEG and event-related potentials) supports such a model [48]. Moreover, the rate of hippocampal volume loss on MRI correlates with clinical progression in the cognitive continuum from normal aging to MCI and to AD [49]. In the non-demented elderly population, the entorhinal cortex and the basal forebrain display diffuses plaques as well as neurofibrillary tangles or pre-tangle tau pathology. It has been suggested that these lesions also contribute to cognitive aging [50]. Adding weight to these arguments is the premise that age is the strongest risk factor for AD despite the complex array of genetic and environmental factors that have also been implicated [51]. By adopting a three-group comparison, the overlap between MCI patients and healthy controls on the one hand, and AD patients on the other hand, was taken into account.

The three-group comparison can justify the slightly lower accuracy figures reached here than in some other PET studies employing control subjects in a two-group comparison approach [10, 15] or not employing controls at all [12, 52]. When discriminant analysis was applied to the direct comparison between MCI/AD and CTR, which are more stable groups, about 90% accuracy was achieved by PET, as used alone, and 100% accuracy was yielded by the combination of PET and memory test scores. Moreover, it is to note that the annual conversion rate (about 17%) to AD in the present study is closer to epidemiological data [53], while in some PET studies, it was higher, raising from 22% [12] to 29% [13] and to 36% [15]. These figures may suggest a higher severity of impairment of aMCI patients in those series, explaining accuracy values exceeding the 90% in identifying converters and non-converters aMCI [10, 12, 52].

Identification of controls was excellent with episodic memory DR score and was total with the combined use of DR score and PET. Two out of the 22 MCI/MCI patients were ‘mis-classified’ within the MCI/AD group, but this could be just an apparent inaccuracy. In fact, these two patients could still clinically be in the pre-dementia phase but already showing the biological hallmarks of AD. This interpretation is in keeping with previous PET data already showing hypometabolism in associative cortex in asymptomatic subjects at high risk to develop AD [54]. Since the follow-up time was limited to about 21 months, some MCI/MCI patients may be early, not-yet-converted AD. This well-known heterogeneity of MCI/MCI group might have affected PCA composition in CTR and MCI/AD, the other two more stable groups. However, a distinct PCA only in these two last groups is not justified, given the limited number of subjects (i.e. 26) as compared to the relatively high number of variables (i.e. 50). The mis-identification of two out of 11 MCI/AD patients may be explained by considering the left lateralization of PC12, yielding differentiation between MCI/MCI and MCI/AD patients together with PC5. In fact, although AD is a diffuse neurodegenerative disease, strong asymmetries are often found in the earliest stages. The PET scans of both the mis-classified MCI/AD patients were checked after these results, and actually one of the two patients showed a strong prevalence of impairment in the right hemisphere, including the LFC.

As for the regional metabolic impairment, as highlighted by PCA, metabolic level in the posterior cingulate expressed by PC5 is confirmed as a sensitive marker of aMCI, both in converters and non-converters. Moreover,

PCA highlighted PC7 (bilateral orbitofrontal cortex), showing a reduction of values in aMCI vs CTR. Grey matter atrophy in orbitofrontal cortex has been reported in aMCI non-converters vs controls [55], and fractional anisotropy of white matter fibres has been recently reported to be affected mainly in the fornix and in the orbitofrontal regions in preclinical familial AD [56]. Orbitofrontal cortex has also been found to participate to the AD-conversion hypometabolic pattern by a PET study [52]. Our findings are consistent with studies demonstrating that limbic projections and pathways connecting the frontal lobes are early affected in the course of AD [57, 58].

The discrimination between MCI/MCI and MCI/AD groups was expressed again by hypometabolism in bilateral posterior cingulate (and left temporal pole, included in the same PC5) and in PC12, including an extended area of the left LFC. The finding of the left LFC is in keeping with Drzezga et al. [15] who re-examined eight converters and 12 non-converters with PET after 1 year. They found that the metabolic decline in LFC was a specific marker of cognitive deterioration and included this area in their 'typical' pattern of converting MCI patients [52]. Interestingly, a specific covariance hypoperfusion (CBF-PET) pattern has been found in early AD, involving the SRT (the same episodic memory test used here), cingulate, inferior parietal lobule, middle and inferior frontal, supramarginal and precentral gyri [45]. In a PET study in 37 aMCI patients [12], the hypometabolic pattern in left LFC and anterior cingulate gave a similar accuracy (87% vs 84%) as hypometabolism in right BA40 in separating aMCI converters and non-converters, but a much higher sensitivity (75% vs 38%). Moving to the pre-symptomatic stage, the left LFC and the bilateral lateral temporal cortex were shown to predict early cognitive decline in a sample of healthy elderly subjects at high risk for AD [54], and this further points to this areas as an early hallmark of neurodegeneration. On the contrary, Anchisi et al. [10] found that left LFC hypometabolism was rather a feature of stable MCI patients over time.

The left LFC contains the language areas and, moreover, is involved in working memory, episodic memory encoding and semantic memory retrieval [59, 60]. Conversion to AD mainly entails a worsening episodic and working memory that significantly impairs the everyday functional autonomy, which is consistent with the idea that metabolic failure in these areas is a main PET hallmark of conversion. According to the pathophysiological model of Braak and Braak [57], these regions receive long axons from the hippocampi and entorhinal cortex and are early functionally affected by the AD process. It might be argued that decreased metabolism in frontal cortex is due to advancing age, but this seems unlikely because age difference between MCI/AD and MCI/MCI was not statistically significant, and there was no correlation between age and significant PCs.

PC5 (posterior cingulate and left temporal pole) values showed a progressive reduction across CTR, MCI/MCI and MCI/AD, while metabolism in PC12 (left LFC) decreased in MCI/AD only. This finding is in keeping with those of a recent paper on age-associated cognitive decline [16] and might suggest a compensatory recruitment in LFC in MCI/MCI patients, which fails in those who convert to AD. Therefore, the posterior cingulate seems to show a more linear metabolic reduction from normal ageing to AD [12, 15, 16], whereas the LFC (especially on the left hemisphere) would be firstly recruited in a sort of compensatory effort that still allows preserved everyday functioning, while its failure would allow IADL impairment and diagnosis of dementia.

No effect was observed in the hippocampi (PC8), which are indeed frequently missed by PET studies using automatic voxel- or VROI-based analysis. It is likely that partial volume effect, which affects MTL structures, is responsible of these findings and that hippocampal hypometabolism can only be highlighted by means of accurate anatomical segmentation [61].

A limitation of this study is the small number of MCI/AD, due to the time needed to detect conversion, which is common to all the PET studies to date, reporting small samples of 'converters' in the range of 1–2 years [10, 12, 15, 52].

In conclusion, we showed that combining the episodic memory and PET data increased the discrimination power among controls, aMCI converters and non-converters. Among PET data, two PC, including the bilateral posterior cingulate and the left temporal pole (PC5) and the left LFC (PC12), discriminated aMCI patients converting to AD after a mean time of 21 months.

Acknowledgments This study complies with the current Italian laws and received ethical approval.

We thank Dr. Giampiero Villavecchia for supervising PET acquisition and Dr. Alessandra Piccini for performing ApoE evaluation.

Appendix 1

PCs may be treated as new variables, and their values can be computed for each subject. These values are known as factor scores, or component scores (CS), and are a linear combination of the variables included in the analysis. They should be used both to re-evaluate group differences and as predictor variables in diagnostic research. However, in the latter case, it is preferable to use an imperfect estimate (CCS) generated by the algebraic sum of all the VROIs with higher loading in a given factor. Therefore, as CCS

take into account the sign of factor loadings, they can deeply differ from the individual VROI values belonging to each PC. Unlike CS, they are not a linear combination of each variable, but an estimate of PCs. Like CS, they are essentially uncorrelated to one another. An advantage of using CCS is that they can be more easily computed and interpreted than CS and they can also be compared among studies [38].

The number of factors was determined by the number of eigenvalues greater than one. Variables with an absolute factor loading greater than 0.5 were considered as representative of a given factor. This is an arbitrary value, but it is commonly used since it explains a moderate part of the variance of the factor. By increasing the value further, some variables may be eliminated from the calculation of CCS, thus reducing the variance explained by these scores. CCS were standardised to a 0–1 scale. The stability of the PCA was evaluated by means of the T2 Hotelling's test. Hotelling's T2 is a measure of the multivariate distance of each observation from the centre of the data set. When PCA is done, T2 and PROB can be saved. PROB is the upper-tail probability of T2. The robustness of the PCA can be assessed looking at the outliers.

References

1. Cummings JL, Doody R, Clark C. Disease-modifying therapies for Alzheimer disease: challenges to early intervention. Neurology 2007;69:1622–34.
2. Dubois B, Feldman HH, Jacova C, DeKosky ST, Barberger-Gateau P, Cummings J, et al. Research criteria for the diagnosis of Alzheimer's disease: revising the NINCDS-ADRDA criteria. Lancet Neurol 2007;6:734–46.
3. Bennett DA, Wilson RS, Schneider JA, Evans DA, Beckett LA, Aggarwal NT, et al. Natural history of mild cognitive impairment in older persons. Neurology 2002;59:198–205.
4. Loewenstein DA, Acevedo A, Agron J, Duara R. Stability of neurocognitive impairment in different subtypes of mild cognitive impairment. Dement Geriatr Cogn Disord 2007;23:82–6.
5. Petersen RC. Mild cognitive impairment as a diagnostic entity. J Intern Med 2004;256:183–94.
6. Feldman HH, Jacova C. Mild cognitive impairment. Am J Geriatr Psychiatry 2005;13:645–55.
7. Sarazin M, Berr C, De Rotrou J, Fabrigoule C, Pasquier F, Legrain S, et al. Amnestic syndrome of the medial temporal type identifies prodromal AD. A longitudinal study. Neurology 2007;69:1859–67.
8. Fossati P, Harvey PO, Le BG, Ergis AM, Jouvent R, Allilaire JF. Verbal memory performance of patients with a first depressive episode and patients with unipolar and bipolar recurrent depression. J Psychiatr Res 2004;38:137–44.
9. Pasquier F, Grymonprez L, Lebert F, Van der Linden M. Memory impairment differs in frontotemporal dementia and Alzheimer's disease. Neurocase 2001;7:161–71.
10. Anchisi D, Borroni B, Franceschi F, Kerrouche N, Kalbe E, Beuthien-Beumann B, et al. Heterogeneity of brain glucose metabolism in mild cognitive impairment and clinical progression to Alzheimer disease. Arch Neurol 2005;62:1728–33.
11. Visser PJ, Scheltens P, Verhey FRJ, 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.
12. Mosconi L, Perani D, Sorbi S, Herholz K, Nacmias B, Holthoff V, et al. MCI conversion to dementia and the *APOE* genotype. A prediction study with FDG-PET. Neurology 2004;63:2332–40.
13. Chetelat G, Eustache F, Viader F, De La Sayette V, Pelerin A, Mezegne F, et al. FDG-PET measurement is more accurate than neuropsychological assessments to predict global cognitive deterioration in patients with mild cognitive impairment. Neurocase 2005;11:14–25.
14. Mosconi L, Tsui WH, Pupi A, De Santi S, Drzezga A, Minoshima S, et al. 18F-FDG PET database of longitudinally confirmed healthy elderly individuals improves detection of mild cognitive impairment and Alzheimer's disease. J Nucl Med 2007;48:1129–34.
15. Drzezga A, Lautenschlager N, Siebner H, Riemenschneider M, Willoch F, Minoshima S, et al. Cerebral metabolic changes accompanying conversion of mild cognitive impairment into Alzheimer's disease: a PET follow-up study. Eur J Nucl Med Mol Imaging 2003;30:1104–13.
16. Hunt A, Schönknecht P, Henze M, Seidl U, Haberkorn U, Schröder J. Reduced cerebral glucose metabolism in patients at risk for Alzheimer's disease. Psychiatry Res: Neuroimaging 2007;155:147–54.
17. Whalley LJ. Brain ageing and dementia: what makes the difference? Br J Psychiatry 2002;181:369–71.
18. Vandenberghe R, Tournoy J. Cognitive aging and Alzheimer's disease. Postgrad Med J 2005;81:343–52.
19. Dringenberg HC. Alzheimer's disease: more than a "cholinergic disorder"—evidence that cholinergic–monoaminergic interactions contribute to EEG slowing and dementia. Behav Brain Res 2000;115:235–49.
20. Folstein MF, Folstein SE, McHugh PR. "Mini-Mental State": a practical method for grading the cognitive state of patients for the clinician. J Psychiatr Res 1975;12:189–98.
21. Katz S, Downs TD, Cash HR, Grotz RC. Progress in development of the index of ADL. Gerontologist 1970;10:20–30.
22. Lawton MP, Brody EM. Assessment of older people; self-maintaining and instrumental activities of daily living. Gerontologist 1969;9:179–86.
23. Cummings JL, Mega M, Gray K, Rosenberg-Thompson S, Carusi DA, Gornbein J. The Neuropsychiatric Inventory: comprehensive assessment of psychopathology in dementia. Neurology 1994;44:2308–14.
24. Scheltens P, Leys D, Barkhof F, Huglo D, Weinstein HC, Vermersch P, et al. Atrophy of medial temporal lobes on MRI in "probable" Alzheimer's disease and normal ageing: diagnostic value and neuropsychological correlates. J Neurol Neurosurg Psychiatry 1992;55:967–72.
25. Wahlund LO, Barkhof F, Fazekas F, Bronge L, Augustin M, Sjogren M, et al. A new rating scale for age-related white matter changes applicable to MRI and CT. Stroke 2001;32:1318–22.
26. Masur DM, Fuld PA, Blau AD, Thal LJ, Levin HS, Aronson MK. Distinguishing normal and demented elderly with selective reminding test. J Clin Exp Neuropsychol 1989;11:615–30.
27. Carlesimo GA, Caltagirone C, Gainotti G. The Mental Deterioration Battery: normative data, diagnostic reliability and qualitative analysis of cognitive impairment. The group for the standardization of the Mental Deterioration Battery. Eur Neurol 1996;36:378–84.
28. Spinnler H, Tognoni G. Standardizzazione e taratura Italiana di test neuropsicologici. Ital J Neurol Sci 1987;6(Suppl. 8):1–120.
29. Barbarotto R, Laiacona M, Frosio R, Vecchio M, Farinato A, Capitani E. A normative study on visual reaction times and two Stroop colour-word tests. Ital J Neurol Sci 1998;19:161–70.

30. Watson YI, Arfken CL, Birge SJ. Clock completion: an objective screening test for dementia. J Am Geriatr Soc 1993;41:1235–40.
31. Amodio P, Wenin H, Del Piccolo F, Mapelli D, Montagnese S, Pellegrini A, et al. Variability of trail making test, symbol digit test and line trait test in normal people. A normative study taking into account age-dependent decline and sociobiological variables. Aging Clin Exp Res 2002;14:117–31.
32. Knopman DS, Boeve BF, Parisi JE, Dickson DW, Smith GE, Ivnik RJ. Antemortem diagnosis of frontotemporal lobar degeneration. Ann Neurol 2005;57:480–8.
33. Loeb C, Gandolfo C. Diagnostic evaluation of degenerative and vascular dementia. Stroke 1983;14:399–401.
34. McKhann G, Drachman D, Folstein M, Katzman R, Price D, Stadlan EM. Clinical diagnosis of Alzheimer's disease: report of the NINCDS-ADRDA Work Group under the auspices of Department of Health and Human Services Task Force on Alzheimer's Disease. Neurology 1984;34:939–44.
35. Bartenstein P, Asenbaum S, Catafau A, Halldin C, Pilowski L, Pupi A, et al. European Association of Nuclear Medicine procedure guidelines for brain imaging using [18F]FDG. Eur J Nucl Med 2002;29:BP43–BP48.
36. Greitz T, Bohm C, Holte S, Eriksson L. A computerized brain atlas: construction, anatomical content, and some applications. J Comput Assist Tomog 1991;15:26–38.
37. Thurfjell L, Bohm C, Bengtsson E. CBA—an atlas based software tool used to facilitate the interpretation of neuroimaging data. Comput Methods Programs Biomed 1995;4:51–71.
38. Andersson JLR, Thurfjell L. Implementation and validation of a fully automatic system for intra- and inter-individual registration of PET brain scans. J Comput Assist Tomogr 1997;21:136–44.
39. Poulin P, Zakzanis KK. In vivo neuroanatomy of Alzheimer's disease: evidence from structural and functional brain imaging. Brain Cogn 2002;49:220–5.
40. Van Heertum RL, Tikofsky RS. Positron emission tomography and single photon emission computed tomography brain imaging in the evaluation of Dementia. Semin Nucl Med 2003;33:77–85.
41. Pett MA, Lackey NR, Sullivan JJ. Making sense of factor analysis in health care research: a practical guide. London: Sage; 2003.
42. Rodriguez G, Nobili F, Copello F, Vitali P, Gianelli MV, Taddei G, et al. 99mTc-HMPAO regional cerebral blood flow and quantitative electroencephalography in Alzheimer's disease: a correlative study. J Nucl Med 1999;40:522–9.
43. Pagani M, Salmaso D, Rodriguez G, Nardo D, Larsson SA, Nobili F. Principal component analysis in mild and moderate Alzheimer's Disease. Psychiatry Res: Neuroimaging; in press.
44. Varrone A, Pagani M, Salvatore E, Salmaso D, Sansone V, Amboni M, et al. Identification by [99mTc]ECD SPECT of anterior cingulate hypoperfusion in progressive supranuclear palsy, in comparison with Parkinson's disease. Eur J Nucl Med Mol Imaging 2007;34:1071–81.
45. Scarmeas N, Habeck CG, Zarahn E, Anderson KE, Park A, Hilton J, et al. Covariance PET patterns in early Alzheimer's disease and subjects with cognitive impairment but no dementia: utility in group discrimination and correlations with functional performance. NeuroImage 2004;23:35–45.
46. Salmon E, Kerrouche N, Perani D, Lekeu F, Holthoff V, Beuthien-Baumann B, et al. On the multivariate nature of brain metabolic impairment in Alzheimer's disease. Neurobiol Aging 2007; July 23; epub ahead of print.
47. Mosconi L, De Santi S, Brys M, Tsui WH, Pirraglia E, Glodzik-Sobanska L, et al. Hypometabolism and altered cerebrospinal fluid markers in normal apolipoprotein E E4 carriers with subjective memory complaints. Biol Psychiatry 2008;63:609–18.
48. Liddell BJ, Paul RH, Arns M, Gordon N, Kukla M, Rowe D, et al. Rates of decline distinguish Alzheimer's disease and mild cognitive impairment relative to normal aging: integrating cognition and brain function. J Integr Neurosci 2007;6:141–74.
49. Kantarci K, Jack CR Jr. Neuroimaging in Alzheimer disease: an evidence-based review. Neuroimaging Clin N Am 2003;13:197–209.
50. Mesulam MM, Shaw P, Mash D, et al. Cholinergic nucleus basalis tauopathy emerges early in the aging-MCI-AD continuum. Ann Neurol 2004;55:815–28.
51. Blennow K, De Leon M, Zetterberg H. Alzheimer's disease. Lancet 2006;368:387–403.
52. Drzezga A, Grimmer T, Riemenschneider M, Lautenschlager N, Siebner H, Alexopoulus P, et al. Prediction of individual clinical outcome in MCI by means of genetic assessment and 18F-FDG PET. J Nucl Med 2005;46:1625–32.
53. DeCarli C. Mild cognitive impairment: diagnosis, prognosis, aetiology, and treatment. Lancet Neurol 2003;2:15–21.
54. de Leon MJ, Convit A, Wolf OT, Tarshish CY, DeSanti S, Rusinek H. Prediction of cognitive decline in normal elderly subjects with 2-[18F]fluoro-2-deoxy-D-glucosey positron-emission tomography (FDG PET). Proc Natl Acad Sci USA 2001;98:10966–71.
55. Chetelat G, Landeau B, Eustache F, Mezenge F, Viader F, de la Sayette V, et al. Using voxel-based morphometry to map the structural changes associated with rapid conversion in MCI: A longitudinal MRI study. NeuroImage 2005;27:934–46.
56. Ringman JM, O'Neill J, Geschwind D, Medina L, Apostolova LG, Rodriguez Y, et al. Diffusion tensor imaging in preclinical and presymptomatic carriers of familial Alzheimer's disease mutations. Brain 2007;130:1767–76.
57. Braak H, Braak E. Development of Alzheimer-related neurofibrillary changes in the neocortex inversely recapitulates cortical myelogenesis. Acta Neuropathol (Berl) 1996;92:197–201.
58. Bartzokis G, Sultzer D, Lu PH, Nuechterlein KH, Mintz J, Cummings JL. Heterogeneous age-related breakdown of white matter structural integrity: implications for cortical "disconnection" in aging and Alzheimer's disease. Neurobiol Aging 2004;25:843–51.
59. Cabeza R, Nyberg L. Imaging cognition. II. An empirical review of 275 PET and fMRI studies. J Cogn Neurosci 2000;12:1–47.
60. Simic G, Bexheti S, Kelovic Z, Kos M, Grbic K, Hof PR, et al. Hemispheric asymmetry, modular variability and age-related changes in the human entorhinal cortex. Neuroscience 2005;130:911–925.
61. Mosconi L, Tsui WH, De Santi S, Li J, Rusinek H, Convit A, et al. Reduced hippocampal metabolism in MCI and AD: automated FDG-PET image analysis. Neurology 2005;64:1860–7.