Current Challenges for the Early Detection of Alzheimer's Disease: Brain Imaging and CSF Studies

Rachel Mistur, MS[a]; Lisa Mosconi, PhD[a]; Susan De Santi, PhD[a];
Marla Guzman, BA[a]; Yi Li, MD[a]; Wai Tsui, MS[a,b]; Mony J. de Leon, EdD[a,b]

Department of [a]Psychiatry, New York University School of Medicine, New York, NY, USA
[b]Nathan Kline Institute, Orangeburg, NY, USA

The development of prevention therapies for Alzheimer's disease (AD) would greatly benefit from biomarkers that are sensitive to the subtle brain changes that occur in the preclinical stage of the disease. Reductions in the cerebral metabolic rate of glucose (CMRglc), a measure of neuronal function, have proven to be a promising tool in the early diagnosis of AD. *In vivo* brain 2-[^{18}F]fluoro-2-Deoxy-D-glucose-positron emission tomography (FDG-PET) imaging demonstrates consistent and progressive CMRglc reductions in AD patients, the extent and topography of which correlate with symptom severity. There is increasing evidence that hypometabolism appears during the preclinical stages of AD and can predict decline years before the onset of symptoms. This review will give an overview of FDG-PET results in individuals at risk for developing dementia, including: presymptomatic individuals carrying mutations responsible for early-onset familial AD; patients with Mild Cognitive Impairment (MCI), often a prodrome to late-onset sporadic AD; non-demented carriers of the Apolipoprotein E (ApoE) ε4 allele, a strong genetic risk factor for late-onset AD; cognitively normal subjects with a family history of AD; subjects with subjective memory complaints; and normal elderly followed longitudinally until they expressed the clinical symptoms and received post-mortem confirmation of AD. Finally, we will discuss the potential to combine different PET tracers and CSF markers of pathology to improve the early detection of AD. **J Clin Neurol 2009;5:153-166**

Key Words Alzheimer disease, PET, CSF markers.

Introduction

Alzheimer's disease (AD) is the most common form of dementia in the elderly and is the fourth leading cause of death in individuals over age 65 after heart disease, cancer, and stroke.[1] AD is a progressive neurodegenerative disorder with insidious onset and characterized by severe decline in episodic memory. Instrumental signs include aphasia, apraxia, and agnosia, together with general cognitive symptoms, such as impaired judgment, decision-making, and orientation.[2] Currently, there are no available tests for the definitive diagnosis of AD *in vivo* and the clinical diagnosis of AD remains a behavioral diagnosis after the exclusion of other causes. This greatly limits the potential for early intervention and prevention research. Furthermore, it is estimated that between 50% and 90% of dementia cases are left undiagnosed by standard clinical examinations.[3]

The definitive diagnosis of AD is based on the post-mortem observation of specific pathological lesions: intracellular neurofibrillary tangles (NFT), amyloid beta (Aβ) deposition in the form of extracellular senile plaques and blood vessel deposits, associated with neuronal and synaptic loss, and brain atrophy in specific brain areas.[4,5] Neurodegeneration in AD is estimated to begin as many as 20-30 years before the clinical manifestations become evident.[6-9] During this preclinical phase, plaque and tangle load increase, and the first symptoms appear after specific brain circuits are structurally disrupted through synapse loss and neuronal death (see[10] for review). It is widely recognized that the brain regions most vulnerable in early AD are in the medial temporal lobes [MTL, i.e., hippocampus, and transentorhinal, entorhinal cortex (EC), and subiculum].[6,7,9] The pyramidal cells anatomically connected to the EC and the CA1 and subiculum regions of the hippocampus are particularly prone to NFT formation and degener-

ation, whereas primary sensory-motor, occipital areas, and cerebellum exhibit minimal neuronal loss.[6,7,9] Disruption of the pyramidal neurons in the perforant path is thought to disconnect the hippocampus from the rest of the cortex, strongly contributing to the decline in memory observed in early AD.[11] While there appears to be significant overlap between cerebrovascular disease and AD pathology,[12,13] a neuropathological definition for mixed forms of dementia remains to be established.

There is an association between NFT staging and severity of clinical status in AD. NFTs originate in the MTL, which plays a critical role in the neural control of memory functions, and then begin to cluster in the adjacent inferior temporal and posterior cingulate cortex (PCC) in mild AD, further disrupting episodic and autobiographic memory. In moderate to severe dementia, NFTs develop within the parieto-temporal and prefrontal association cortices, which are involved in the neural control of perception, attention, and language.[6,7] Despite an initial predilection for the neocortex, Aβ depositions are also found in the MTL at later stages of disease.[14-16] For many years, the aggregation of large Aβ fibrils was considered the key event in AD pathogenesis and the main determinant of neuronal degeneration.[17] A recent reformulation of the amyloid cascade hypothesis states that Aβ oligomers (Aβ-derived diffusible ligand, ADDL), not fibrillar Aβ, confer greater neurotoxicity to neurons by disrupting nerve signaling pathways and subsequently causing neuronal cell death.[18]

While the causes of neurodegeneration in AD are under investigation, the early appearance of pathological lesions and the progressive nature of cognitive deterioration in AD indicate a great need for developing biological markers of disease that are sensitive to the brain changes that are expected to occur decades prior to the onset of clinical symptoms. Ideally, a biological measure would be predictive of AD in presymptomatic individuals. Criteria for an ideal biomarker of the disease have been proposed by the Consensus Group on Molecular and Biochemical Markers of AD.[19] In short, a model biomarker for the disease should detect a fundamental characteristic of the neuropathology and be validated in neuropathologically confirmed cases, with sensitivity and specificity of no less than 80%.[19]

Several modalities show promise in the development of early diagnostic tools for AD. These include: magnetic resonance imaging (MRI) measurements, positron emission tomography (PET) imaging of glucose metabolism in the MTL, PET imaging of Aβ deposits, and cerebrospinal fluid (CSF) biomarkers for tauopathy and Aβ. At present, none of these are recommended in any consensus guidelines for the diagnosis of AD, as they are not yet validated by large prospective studies.

Structural imaging, such as MRI, plays an important part in the diagnosis of AD by identifying other causes of dementia, such as tumor, subdural hematoma, and cerebrovascular diseases marked by infarcts and white-matter lesions.[2] Cerebral atrophy, visualized as enlarged ventricles and cortical sulci, can also be identified by CT and MRI, but there is significant overlap with normal aging and other dementias.[20] Nonetheless, several CT and MRI studies have shown that MTL atrophy is an early sign of AD and has value in predicting future dementia in non-demented subjects (see, among others).[21-25] In addition to predicting future AD, hippocampal atrophy is also apparent in vascular cognitive impairment and can be useful in predicting cognitive decline related to vascular dementia.[26]

Ultimately, AD pathology has the effect of impairing neuronal function, which then leads to the clinical symptoms of dementia. Functional neuroimaging offers the unique capability to both visualize the direct effects of neuronal activity and quantify the rates of specific biological processes at the tissue level *in vivo*. PET imaging with 2-[^{18}F]fluoro-2-Deoxy-D-glucose (FDG) has long been used to track AD-related brain changes by providing qualitative and quantitative estimates of the cerebral metabolic rate of glucose (CMRglc).

Glucose is the predominant source of energy for the brain. Early studies showed that glucose utilization is not only measurable, but also serves as an index of neuronal function.[27] Brain energy metabolism is mainly associated with brain glutamate signaling,[28] as more than 80% of brain's neurons are excitatory and 90% of synapses are glutamatergic. Thus, glucose metabolism can be interpreted as an index of synaptic functioning and density.[27,29-31] A recent animal study using FDG-PET showed a significant correlation between CMRglc *in vivo* and levels of synaptophysin, a marker of synaptic density, assessed at *post-mortem*.[31] Additionally, astrocytes in the extraneuronal space have metabolic demands and contribute to the FDG-PET signal.[32,33] Recent studies have shown that coupling between energy metabolism and neuronal activity may also be achieved through astrocytic uptake of glucose and lactate shuttling to neurons.[33]

The present review will provide an overview of FDG-PET studies in AD, with an emphasis on the presymptomatic and preclinical detection of AD.

Alterations of Brain Glucose Metabolism in Alzheimer's Disease

One of the striking features of AD is the drastic reduction of CMRglc in specific brain regions. FDG-PET studies in AD demonstrate consistent and progressive CMRglc reductions, whose extent and topography correlate with symptoms seve-

rity (see[34] for review). FDG-PET studies report that, as compared to age-matched healthy normal controls, AD patients show regional metabolic reductions in the parieto-temporal[35,36] and posterior cingulate cortices,[37] against a background of widespread global metabolic impairment.[38] The severity and topography of the metabolic reductions may also vary according to the age of onset of AD.[39] Late-onset AD patients may be more likely to show mild metabolic reductions in the inferior temporal gyri, rather than extensive hypometabolism in the parieto-temporal regions, which are more typically observed in early-onset AD.[39] CMRglc reductions are also apparent in the frontal lobe in advanced disease.[40] This pattern of hypometabolism is seen in contrast to relative preservation of the primary motor and visual areas. Moreover, the cerebellum, thalamus and basal ganglia nuclei are spared from significant reductions in glucose metabolism.[41] In comparison to AD, normal aging is characterized by subtle and diffuse CMRglc reductions in the frontal cortex and anterior cingulate cortex (ACC), accompanied by slight reductions in global CMRglc.[42]

These FDG-PET findings were initially reported in the early 1980's and have since been replicated numerous times. Furthermore, the pattern of hypometabolism described is largely accepted as a reliable *in vivo* hallmark of AD. Studies show that temporo-parietal abnormalities have high sensitivity in distinguishing AD from normal aging, despite some regional overlap.

These characteristic CMRglc reductions have also been used to differentiate AD from other neurodegenerative diseases, such as frontotemporal and Lewy body dementia (LBD),[43] and cerebrovascular disease.[44]

One of the major challenges of the field remains to disentangle the relative contributions of AD, vascular cognitive impairment and, to a lesser extent, LBD to different stages of cognitive impairment in the general population.

The Early Diagnosis of Alzheimer's Disease

Importantly, MTL CMRglc reductions have been observed on FDG-PET before the onset of AD symptoms and a growing list of observations has highlighted the importance of PET as a tool for the detection of early disease and for estimating an increased risk for future dementia. This list of observations includes (Table 1):

• Presymptomatic individuals carrying autosomal dominant mutations responsible for early-onset familial AD;

• Patients with Mild Cognitive Impairment (MCI), which is in many cases a prodrome to AD;

• Cognitively normal elderly who declined to MCI and AD several years after PET scan acquisition;

• Cognitively normal individuals carriers of the Apolipoprotein E (ApoE) E4 allele, a susceptibility factor for late-onset AD;

• Cognitively normal subjects with subjective memory complaints;

• Cognitively normal subjects with a maternal family history of AD.

The main FDG-PET findings from these studies are reviewed below.

Conversion Studies

Presymptomatic early-onset familial Alzheimer's disease

Autosomal dominant mutations have been identified in three

Table 1. Preclinical diagnostic findings

At-risk group	Control group	PET findings	References
Presymptomatic Early-onset Familial AD	Mutation Non-carriers	• Whole brain hypometabolism • Parieto-temporal, PCC, frontal cortex, and MTL hypometabolism	48, 46, 47
NL ApoE-4 Carriers (ApoE-4 +)	NL ApoE-4 Non-carriers (ApoE-4 -)	• Parieto-temporal, PCC, thalamus, and frontal cortex hypometabolism • Greater CMRglc decline over time	73, 74, 75, 76, 77, 78, 79
NL with Subjective Memory Complaints (SMC +)	NL without Subjective Memory Complaints (SMC -)	• Parieto-temporal and MTL hypometabolism	79
NL with a Maternal Family History of AD (FHm)	Normal Individuals without a Maternal Family History of AD (FH- and FHp)	• Parieto-temporal, PCC, frontal cortex, and MTL hypometabolism • Greater CMRglc decline over time	85, 86
NL-MCI & NL-AD	NL-NL	• MTL hypometabolism when NL • Parieto-temporal and PCC hypometabolism at time of decline • Greater CMRglc decline over time	49, 68, 50

AD: Alzheimer's disease, NL: cognitively normal subjects, MCI: mild cognitive impairment, PCC: posterior cingulate cortex, CMRglc: cerebral metabolic rate of glucose, MTL: medial temporal lobes.

genes, i.e., amyloid precursor protein (APP, on chromosome 21), Presenilin 1 (PS1, on chromosome 14), and Presenilin 2 (PS2, on chromosome 1), which are associated with early-onset familial AD (FAD). FAD accounts for <5% of AD cases in the general population and is characterized by autosomal dominant inheritance with 100% penetrance and a specific early age of symptom-onset for a given pedigree (see[45] for review). Therefore, the study of presymptomatic mutation carriers, close to the expected age of dementia onset provides unique information about preclinical AD-related brain changes in individuals who are destined to develop the disease.

Several FDG-PET studies examining presymptomatic FAD have shown parieto-temporal, posterior cingulate, and frontal cortex hypometabolism in most FAD cases, as compared to age-matched controls.[46,47] A study by Kennedy et al.[47] showed that whole-brain CMRglc in presymptomatic FAD individuals is intermediate between controls and symptomatic FAD patients, suggesting a progression of global CMRglc impairment throughout the course of disease. However, these early studies examined only the neocortex. They did not examine the MTL, nor did they perform partial volume correction of the FDG-PET values. Since these patients also showed significant atrophy on MRI, it remained to be established whether the CMRglc reductions were an effect of an increasing CSF pool or if they reflect functional deficits in the remaining tissue. Unfortunately, CSF is not resolved by the PET camera and therefore cannot be avoided in tissue sampling. As a result, partial volume effects of CSF lower CMRglc values obtained with FDG-PET.

We recently addressed this issue in an FDG-PET and MRI study of presymptomatic PS-1 carriers from families with early-onset FAD, examined an average of 13 years prior to the estimated age at disease onset.[48] Our data show MTL and cortical hypometabolism in presymptomatic FAD, as compared to age-matched non-carriers from the same families. Furthermore, these CMRglc reductions in FAD individuals exceeded tissue loss. Specifically, we compared CMRglc and volumes in several brain regions, including the hippocampus, EC, PCC, parietal and temporal cortices, and the whole-brain. Significant volume reductions in FAD as compared to controls were restricted to the parietal cortex. Conversely, CMRglc reductions on FDG-PET were observed in all regions examined and remained significant after MRI partial volume correction. After partial volume correction, the CMR glc reductions ranged from 13% (whole brain) to 21% (PCC), reflecting true reductions of brain glucose utilization per unit brain volume. After partial-volume correction, MTL CMRglc values were reduced by 12% in the hippocampus and 20% in the EC. Overall, presymptomatic FAD patients displayed widespread CMRglc reductions in the brain regions typically hypometabolic in clinically diagnosed dementia patients. In contrast, structural brain atrophy was relatively absent in these individuals.[48] These results provide definitive evidence that CMRglc reductions precede clinical symptoms as well as gross structural brain changes. Similar observations have been made in the preclinical stages of late-onset AD.[49,50] Fore a more detailed discussion on the value of PET relative to structural MRI, see.[20]

Mild cognitive impairment

A successful strategy to examine the preclinical stages of sporadic AD has been to investigate MCI patients. MCI is recognized by many as a transitional state between healthy aging and dementia, during which individuals are able to perform the usual activities of daily living, but suffer mild memory impairments and/or other cognitive difficulties exceeding those expected on the basis of normal aging. These symptoms reduce the quality of life for the patient and put them at higher risk for developing AD.[51,52] In particular, patients in research settings having salient memory deficits, e.g., amnestic MCI, decline to AD with an estimated conversion rate of 10-30% per year.[51,52]

While parieto-temporal and PCC hypometabolism is consensually recognized as the metabolic signature of AD, there is currently no specific pattern of hypometabolism considered to be a hallmark for MCI, as related to AD (see[20,34] for recent reviews). In keeping with the concept of MCI as an intermediate stage along the hypothesized continuum from normal aging to AD, MCI patients generally present with mild global and regional hypometabolism within the same brain regions typically affected in clinical AD,[37,49,53-62] including an increasing numbers of reports of MTL CMRglc reductions.

The regional patterns of CMRglc reductions in MCI are milder and more variable than those found in AD and correspond to variations in the patterns of cognitive and behavioral abnormalities in individual patients.[62,63] Importantly, FDG-PET studies in MCI showed that hippocampal hypometabolism is a consistent feature of MCI patients regardless of the neuropsychological profile.[49,50,55,61] Studies have shown a more diversified metabolic profile in non-amnestic MCI (i.e., patients with selective deficits in attention and language[51]) who show either an absence of cortical hypometabolism or hypometabolism in brain regions including, but not restricted to the anterior cingulate and parieto-temporal cortices.[49,55,61,64,65] In contrast, amnestic MCI patients more consistently show pronounced abnormalities in the parieto-temporal cortices and PCC.[37,56-58,66] Therefore, early metabolic reductions in the PCC may represent a more specific marker of AD than hippocampal hypometabolism.[58] Given the higher rate of decline to AD in amnestic compared to non-amne-

stic MCI,[52] these data suggest that more severe and spatially extended CMRglc reductions in AD-specific regions may predispose these patients to develop AD in the near future.

In addition to cross-sectional examination of FDG-PET in the differentiation of MCI from normal aging, a growing body of longitudinal FDG-PET examinations has examined the predictive value of these measures in the decline from MCI to AD. These studies, most of which focused on amnestic MCI patients, demonstrated that baseline CMRglc reductions are more pronounced in MCI who progress to AD, as compared to MCI who remained stable. The reported prediction accuracies range from 75% to 100%.[37,53,54,57,60,62] Moreover, there is evidence showing that the metabolic changes in declining MCI patients are progressive and that longitudinal CMRglc measures both predict and correlate with decline to AD.[58]

Decline from normal cognition to mild cognitive impairment and to Alzheimer's disease

Very little work has been done with FDG-PET to monitor the progression from normal aging to sporadic AD. Such studies are limited because of the intrinsic difficulty of observing clinical change for a group with a low incidence of decline (1-3%/year) and a slow progression of cognitive deterioration.[67] Following cognitively normal persons over time until they develop dementia requires large subject samples, long follow-up intervals, and great expense.

Few published FDG-PET studies have monitored decline from normal to MCI[49] or from normal to MCI and dementia.[50,68] In the first study, de Leon et al.[49] studied 67 normal elderly, 48 of whom completed a 3-year follow-up. Within the longitudinal subgroup, 11 declined to MCI and one to AD. The results showed that reduced baseline CMRglc in the EC predicted a future diagnosis of MCI with 83% sensitivity and 85% specificity. Moreover, longitudinal CMRglc reductions were found in the EC, hippocampus, and lateral temporal cortex during the progression to MCI. Importantly, these effects remained significant after correcting the CMRglc values for MRI partial volume effects, suggesting that these early CMRglc reductions in MCI are independent of tissue loss and represent true reductions of glucose consumption per gram of brain tissue. However, considering that many MCI remain stable, it was not established whether the observed CMRglc reductions in MCI were in fact due to AD.

In 2006, Jagust and colleagues[68] also looked at early metabolic changes in cognitively normal individuals studied longitudinally. Sixty subjects were followed for a mean of 4 years and received baseline FDG-PET scans and annual evaluations of global cognition, assessed by the Modified Mini-Mental State Examination (MMSE). Within this cohort, six subjects developed incident dementia or cognitive impairment after the initial visit. The results showed that baseline CMRglc reductions in the angular gyri, left mid-temporal gyrus, and left middle frontal gyrus predicted rate of change on the MMSE, and were associated with faster cognitive decline.[68]

We recently published the first longitudinal FDG-PET study monitoring the conversion from normal cognition to AD. Mosconi et al. examined 77 normal elderly who were followed over 6-14 years and received multiple FDG-PET examinations. Over this interval, 11 baseline normal subjects developed dementia, 6 of whom were diagnosed with AD, and 19 declined to MCI. Decline for both outcome groups occurred, on average, 8 years after the baseline exam. CMRglc in the hippocampus and cortical regions were examined as predictors and correlates of change in clinical status. The baseline hippocampal CMRglc was the only regional predictor of future cognitive decline and predicted decline from normal to AD with 81% accuracy, including two post-mortem confirmed AD cases. The baseline hippocampal CMRglc also predicted decline from normal to MCI with 71% accuracy and from normal to another dementia with 77% accuracy. Hippocampal hypometabolism was also a significant predictor of the time to decline. On survival analysis, for individuals with hippocampal CMRglc $\leq$24 μmol/100 g/min, the predicted time to decline to AD was 7 years. For hippocampal CMRglc of 25-29 μmol/100 g/min, the predicted time to decline was 9.5 years, and for CMRglc $\geq$30 μmol/100 g/min, greater than 14 years. In AD, for every unit decrease in baseline hippocampal CMRglc, the time to decline decreased by 8.7% [95% CI: 3.0-14.1%] [χ^2 (1)=8.6, p=0.003], which corresponds to a time ratio (TR) of 1.1 (95% CI: 1.0-1.4) years; the time to decline to other dementias is decreased by 4.7% (95% CI: 0.3-8.9%) [χ^2 (1)=4.6, p<0.05], for a TR of 1.0 (95% CI: 0.8-1.2) years; the time to decline in MCI is decreased by 7.2% (95% CI: 2.8-11.5%) [χ^2 (1)=9.93, p<0.01], for a TR of 1.08 years (95% CI: 1.03-1.11). Furthermore, normal subjects who later declined experienced greater rates of hippocampal, PCC and temporal cortex CMRglc reductions, as compared to non-declining subjects.[69]

In addition, these FDG-PET data provided direct evidence for a topographical progression of CMRglc abnormalities, which appear to originate in the MTL during the normal stages of cognition, extend to the PCC at the MCI stage, and finally spread to the parieto-temporal cortices in advanced dementia,[50] in keeping with NFT pathology findings.[9]

Overall, our results demonstrated an association between reduced hippocampal CMRglc during normal aging, shorter time intervals until the onset of dementia, and increased risk for cognitive decline, years in advance of the clinical diagnosis.

Risk Factors for Alzheimer's Disease

Apolipoprotein E E4 genotype

The epsilon 4 allele of the ApoE gene on chromosome 19 is a widely recognized genetic risk factor for late-onset AD.[70,71] The ApoE gene codes for the production of lipoproteins, which mediate the transport of lipids throughout bodily fluids such as blood and CSF. ApoE is specifically thought to maintain neuronal homeostasis by transporting lipids such as cholesterol and phospholipids, which are responsible for neuronal plasticity, throughout the central nervous system.[72] ApoE also provides lipids necessary for the repair of damaged nerve cell membranes. There are three common isoforms of the ApoE allele, including ApoE-2, ApoE-3, and ApoE-4. The ApoE-4 genotype has been designated as a risk factor for AD since 40% of AD patients have at least one ApoE-4 allele and there is also a negative association between the dose of ApoE-4 allele and the mean age of onset of AD.[71]

The mechanisms by which the ApoE-4 allele confers increased risk of future AD is under study, although it is believed that ApoE-4 may confer decreased plasticity of neuronal synapses and an inability to repair damaged neurons, as compared to the other allelic variants.[72]

FDG-PET studies examining the effects of the ApoE-4 allele on CMRglc in non-demented individuals reported that, compared to non-carriers, ApoE-4 carriers have mild but definite CMRglc reductions in the same regions as clinically affected AD patients.[73-79] There is evidence in middle-age ApoE-4 carriers that the metabolic reductions are progressive and correlate with reductions in cognitive performance.[73,76] One study showed a 25% decline in CMRglc over an interval of 2 years in cognitively normal persons carrying two ApoE-4 alleles.[76] Moreover, the same pattern of hypometabolism was observed in 20-40 year-old carriers. These CMRglc reductions are considered the earliest brain abnormalities yet found in living persons at risk for AD.[77]

Nonetheless, these studies are limited by the relatively short follow-up intervals and it remains to be established whether the CMRglc reductions in ApoE-4 carriers are predictive of decline to AD.

Subjective memory complaints

Subjective memory complaints (SMC) are widespread in the elderly community with a prevalence of 25-50%. For many, these complaints represent a preclinical sign of incipient dementia.[80] We recently published an FDG-PET study examining CMRglc in cognitively normal individuals with and without SMC.[79] The results showed that normal individuals with SMC have significant CMRglc reductions in several brain regions, including the parahippocampal gyri (PHG),

parieto-temporal and inferior frontal cortices, fusiform gyrus, and thalamus, as compared to demographically matched individuals with no such complaints. Hypometabolism in the PHG region, which anteriorly includes the EC,[81] was the most significant predictor of SMC status. These reductions distinguished subjects with and without SMC with 75% accuracy and an odds ratio of 2.4 (95% CI=1.3-4.8, p<0.001). In other words, normal individuals with SMC are more than twice as likely to have PHG deficits, as compared to those with no complaints.[79] Moreover, we also explored the effects of ApoE genotype on CMRglc and showed a significant interaction between SMC and ApoE status. Among subjects with SMC, carriers of the ApoE-4 genotype had the lowest CMRglc measures in the PHG, temporal and frontal cortices, and thalamus, as compared to the three other subgroups. Again, the CMRglc reductions were most prominent in the PHG (18%).

Maternal family history of Alzheimer's disease

After advanced age, the most prominent demographic risk factor for developing AD is a first-degree family history of late-onset AD.[71] Normal individuals with a first-degree relative affected with AD, particularly those with an affected parent, have a 4- to 10-fold increased risk for developing AD.[82-84] We recently performed the first imaging study to compare individuals with and without a parental history of AD. In this analysis, we also examined the effects of maternal and paternal history of AD on CMRglc in normal individuals.[85] Specifically, we examined the FDG-PET scans of 49 cognitively normal elderly grouped according to their family history of AD. We compared individuals with a maternal (FHm), paternal (FHp), or no family history of AD (FH-). The results showed that FHm individuals have CMRglc reductions in the PCC/precuneus, parieto-temporal and frontal cortices, and MTL, as compared to FHp and FH- individuals.[85] Intriguingly, these brain regions are typically affected in clinical AD patients. The results remained significant after accounting for other potential risk factors for AD, such as age, female gender, ApoE genotype, and presence of subjective memory complaints. The ApoE-4 genotype was represented in 22% of our FHm subjects and the same effects were found in the group of ApoE-4 non-carriers.

As a follow-up to our cross-sectional study, we examined 66 cognitively normal subjects, which included 37 FH-, 9 FHp, and 20 FHm. These groups were controlled for age, education, prevalence of subjective memory complaints, and ApoE genotype.[86] We compared CMRglc across FH groups at baseline, 2-year follow-up, and longitudinally. FHm showed significantly greater reductions in CMRglc in the PCC, parieto-temporal lobes, and MTL at baseline and follow-up

when compared to FHp and FH-.[86] The reductions in FHm increased from 13% at baseline to 23% at follow-up in the same brain regions and this increase in severity was significant. In comparison, there was no significant difference between CMRglc at baseline and follow-up in FHp and FH-. Additionally, the mean annual decline in CMRglc for FHm (-3.14 ± 1.3 μmol/100 g/min) was significantly greater than that observed in FH- (-0.75 ± 1.5 μmol/100 g/min) and in FHp (-0.56 ± 0.91 μmol/100 g/min) ($p<0.001$).[86] The progressive hypometabolism seen in FHm is similar to that seen in ApoE-4 carriers, although the observed CMRglc reductions are independent of ApoE genotype. This evidence suggests an alternative mechanism by which FHm increases vulnerability to brain glucose hypometabolism. According to our previous PET findings, these CMRglc reductions may predispose FHm individuals to developing AD in the future. However, as with the ApoE-4 genotype, it is necessary to longitudinally follow subjects in all family history categories to determine whether the observed CMRglc alterations truly predict the onset of AD.

The biological and genetic mechanisms that underlie the CMRglc reductions in individuals with a maternal family history of AD are not known. With all that is known about the molecular processes involved in glucose metabolism, hypometabolism may be due to, among other factors, a combination of defective mitochondrial function and possible mitochondrial DNA (mtDNA) mutations.[87] mtDNA is entirely maternally inherited in humans and diseases associated with mtDNA mutations often present as sporadic disorders.[87] These pieces of evidence lend support to a mtDNA transmission hypothesis. Although other genetic mechanisms (i.e., epigenetic imprinting, chromosome X transmission, and mutations in nuclear DNA affecting mitochondrial function) could potentially account for our findings, our findings suggest maternal transmission of hypometabolism in normal subjects at risk for AD.

Combining PET Tracers

Overall, the addition of other PET tracers to FDG-PET may be of use in the early detection of AD. An effective strategy to increase the diagnostic accuracy in AD would be to combine the sensitivity of FDG-PET with another modality that provides a disease-specific measure of pathology. Recently, several PET tracers for Aβ plaques have been developed. The best known tracer, N-methyl-[^{11}C]2-(4'-methylaminophenyl)-6-hydroxybenzothiazole, also known as Pittsburgh Compound-B (PIB), binds to Aβ plaques in the brain.[88] Several PIB-PET studies demonstrated significant PIB retention in AD patients as compared to controls, mostly evident in the middle- and pre- frontal cortex, parieto-temporal regions, PCC/precuneus, occipital lobes, thalamus and striatum (Fig. 1).[88-92] These regions are consistent with the known pattern of Aβ plaque deposition observed at post-mortem, and correlate with reductions in CSF Aβ1-42.[93] PIB-PET studies have shown significant PIB retention in the AD patients, and in as many as 61% of MCI[90-92] and 22% of normal elderly.[92] One follow-up PIB-PET publication indicates a lack of longitudinal progression,[94] suggesting that amyloid deposition may plateau at the AD stage. At this early stage of development, PIB-PET has also been shown to facilitate an improved differential diagnosis of different dementias.[92,95] Although, a recent study suggested that PIB may not be specific for dense, classical plaques.[96] PIB appears to bind to a family of amyloid substrates ranging from diffuse plaques to plaques in the vascular system (i.e., cerebral amyloid angiopathy), as well as NFT.[97] These data, together with the observation that many normal elderly have brain amyloid deposits, suggest that PIB-PET imaging may be more suitable to rule out AD, since a patient without PIB uptake is unlikely to have AD.

The 20-minute radioactive decay half-life of ^{11}C limits the use of PIB to centers with an on-site cyclotron, which makes it difficult to employ the tracer in clinical practice. To over-
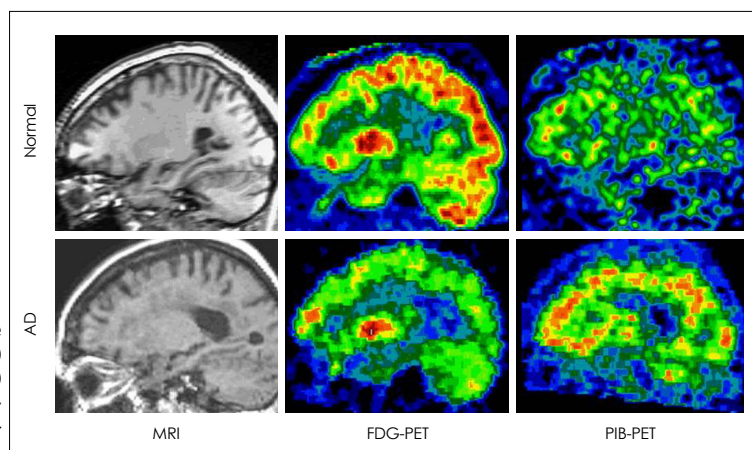

Fig. 1. Two representative cases: magnetic resonance image (MRI, left column), FDG-PET (middle column) and PIB-PET (right column) of a normal control (top row) and an AD patient (bottom row). FDG: 2-[^{18}F]fluoro-2-Deoxy-D-glucose, PIB: Pittsburgh Compound-B, AD: Alzheimer's disease.

come these limitations, amyloid tracers labeled with fluorine-18 (^{18}F, 110 minute half-life) have been developed. The first fluorinated Aβ PET tracer to become available is 2-(1-{6-[(2-[^{18}F]Fluoroethyl)(methyl)amino]-2-naphthyl}ethylidene) malononitrile (^{18}F-FDDNP), which binds with high specificity to both Aβ fibrils and NFTs.[98,99] ^{18}F-FDDNP binding was elevated in AD and MCI patients as compared to healthy elderly, and showed uptake in the parieto-temporal, PCC, and frontal regions, yielding 100% diagnostic separation between AD and controls, and 95% between MCI and controls.[99] Moreover, there was a strong correlation between tracer uptake and scores on tests of memory and global cognition, as well as longitudinal progression for a subgroup of non-demented subjects that deteriorated over 2 years.[99] One potential early diagnostic advantage of ^{18}F-FDDNP over PIB is its capacity to bind NFT in addition to Aβ plaques.[100] It is known that neurofibrillary tangle pathology, unlike Aβ deposition, appears in the early stages of disease in the hippocampal formation[9] and increased NFT load in this region is associated with impairment of cognition.[101] One issue raised is the narrow range of tracer binding across subjects and subsequently low percentage change in binding across groups, ranging from 5-8% increase in patients with MCI and AD relative to controls.[100] Further studies are needed to assess the extent to which ^{18}F-FDDNP increases both early diagnosis and diagnostic specificity.

Another fluorinated tracer for Aβ deposits, *trans*-4-(*N*-methyl-amino)-4'-{2-[2-(2-[^{18}F]fluoro-ethoxy)-ethoxy]-ethoxy}-stilbene (^{18}F-BAY94-9172), has recently become available.[102] ^{18}F-BAY94-9172 binding matched the reported post mortem distribution of plaques, and was consistently higher in AD patients, as compared to healthy controls and FTD patients. Visual interpretation was 100% sensitive and 90% specific for the detection of AD.[102]

At the present time, amyloid imaging studies have not yet reported on early diagnosis, or on predictions of longitudinal clinical change. Additional validation studies are needed before Aβ PET imaging can enter into clinical practice.

CSF biomarkers

CSF is in direct contact with the brain and the molecular composition of CSF reflects biochemical changes in the central nervous system. Consequently, many studies have examined the CSF as a possible source for biomarkers of AD pathology. Several candidate diagnostic biomarkers have emerged. The most widely studied CSF analytes include markers for tau [i.e., total (T-tau) and hyperphosphorylated tau (P-tau) proteins] and Aβ pathology (i.e., peptide fragments of Aβ$_{40}$ and Aβ$_{42}$ amino-acid residues), as well as isoprostane (IsoP, a marker of lipid peroxidation and inflammation).

CSF tau studies

Two types of CSF measures for tau pathology have been used in AD: T-tau and markers for isoforms (X) of P-Tau (P-TauX). T-tau, the first biomarker to be available, is the most widely used. Overall, CSF T-tau reflects both the normal metabolism of tau and the nonspecific release of tau following neuronal damage, whereas P-tau$_{231}$ reflects abnormal tau metabolism that is both sensitive and specific for AD.[103] The evidence consistently demonstrates elevated CSF concentrations of T-tau in AD and in MCI compared to NL controls.[104] CSF T-tau is increased to around 300% in AD as compared to controls, probably as a result of neuronal and axonal degeneration, with a mean sensitivity of 84% and 91% specificity.[105] Moreover, 11 studies show that CSF T-tau (and P-tauX in several studies) alone or in combination with Aβ$_{42}$ predicts the conversion from MCI to AD.[106] Equivalent prediction accuracies for CSF T-tau and P-tau$_{231-235}$ were reported by Arai,[107] by Hansson using P-tau$_{181}$,[108] and by our group[109] with P-tau$_{231}$. However, reports about CSF X-tau levels in normal aging are limited and contradictory; some show age-related elevations in T-tau[110] and others do not.[111,112] Our adult lifespan data shows age-related increases after age 60 for both P-tau$_{231}$ and T-tau, and both demonstrate additive ε4 related effects.[113] The T-tau level is not specific for AD, as CSF T-tau levels are also elevated in other neurodegenerative diseases.[114,115] In acute stroke, the T-tau, but not the P-tau$_{181}$ levels, were increased and later returned to normal.[116] However, P-tau$_{231}$ offers reasonable diagnostic specificity for AD. Hampel and his co-investigators found that the levels of P-tau$_{231}$, but not T-tau, were consistently elevated in AD as compared with frontotemporal dementia (FTD), LBD, vascular dementia, and NL elderly controls.[117] More recent work extended the specificity of P-tau$_{231}$ for AD to major depression[118] and CJD.[119] Similarly, others demonstrated the advantage of P-tau$_{181}$ over T-tau in comparisons between AD with FTD[120,121] and with non-AD dementias.[122,123] A recent longitudinal biomarker study using MRI showed that changes in the levels of P-tau$_{231}$ were longitudinally associated with the change in the hippocampal volume in MCI patients.[124] However, there is very limited evidence for CSF tau levels to increase with clinical progression,[124,125] thus making the CSF measurement a potentially ancillary measurement to be used with modalities that do confer longitudinal information, such as FDG-PET or MRI. At present there are no reports of the combined use of tau biomarkers with FDG-PET in longitudinal designs.

CSF Aβ studies

CSF-Aβ$_{42}$ is reduced in AD to around 50% of control concentrations, which may be due to the deposition of the peptide in brain plaques or to reduced neuronal production. Cross-

sectional CSF Aβ studies consistently show reduced $A\beta_{42}$ levels in both late onset AD[104,106] and MCI,[108,126] but there is a dearth of longitudinal data. The available longitudinal data show $A\beta_{42}$ levels decrease in AD, but the magnitude of change has limited diagnostic value.[115,127] The background effects of aging on CSF turnover and Aβ production and clearance are poorly understood.[128-130] Peskind et al.[131] recently showed that CSF $A\beta_{42}$ reductions were increasingly found with old age and detected at younger ages in ε4 carriers, and our data confirm that observation.[113]

Fagan recently reported that CSF $A\beta_{42}$ reductions are associated with PIB evidence for deposition, but that the relationship is bimodal rather than linear.[93] CSF $A\beta_{42}$ reductions predict decline from MCI to dementia[106] and from CDR=0 to CDR>0 when combined with T-tau or P-tau[181].[132] To date, only one study importantly showed that high CSF T-tau/$A\beta_{42}$ or P-tau[181]/$A\beta_{42}$ ratios, but not X-tau or Aβx measures alone, predicted decline from CDR=0 (normal) to CDR>0 (impaired).[132]

The diagnostic utility of CSF $A\beta_{40}$, a predominant feature of vascular amyloid, as an AD biomarker is less well understood than $A\beta_{42}$. A limited number of reports have shown elevated CSF $A\beta_{40}$ levels with increasing age.[133,134] However, several cross-sectional studies failed to observe differences between AD and NL[135,136] and for this reason the $A\beta_{42/40}$ ratio, controlling for overall Aβ production and clearance, is often used as a diagnostic marker.

CSF isoprostane studies

Oxidative stress is a recognized feature of AD and other neurodegenerative diseases.[137] Postmortem studies by both the Pratico and Montine groups show elevated brain[138] and CSF IsoP levels[139] in AD. IsoP are isomers of enzymatically derived prostaglandins, which are produced by O_2 radical-catalyzed peroxidation of polyunsaturated fatty acids.[140] Most AD work has focused on IsoP's derived from prostaglandin $F_{2\alpha}$, which is a reliable marker of *in vivo* oxidative stress.[141] These studies also demonstrate correlations between neuronal oxidation[138] and Braak staging.[139] There is a growing awareness that IsoP changes are early features of AD that may even precede the development of fibrillar amyloid plaques.[142-144] At postmortem CSF IsoP is elevated in MCI,[144] and there is consensus that the levels are elevated in both AD[145,146] and MCI *in vivo*.[124,147] In collaboration with Pratico, our CSF IsoP data show longitudinal elevations in MCI[124] that accurately predict the decline from MCI to AD.[109] Specifically, IsoP levels do change with clinical progression[109] and with progressive brain damage.[148] While there is no mechanistic basis to expect that elevated IsoP levels are AD-specific, several studies show that CSF IsoP levels are higher in AD than FTD[149,150] and other dementias.[151]

Combining PET with CSF biomarkers

Unfortunately, there is not much literature examining CSF biomarkers and PET. We recently published the first study to examine the relationship in normal subjects with subjective memory complaints (SMC) between hypometabolism on FDG-PET and CSF markers of AD pathology.[79] Across all subjects, CMRglc in the MTL, parieto-temporal and frontal cortices were significantly correlated with CSF levels of T-Tau, P-Tau[231] and IsoP, while no significant relationships with PET were found between CSF $A\beta_{40}$ and $A\beta_{42}$. ApoE-4 carriers with SMC showed the highest CSF T-Tau, P-Tau[231] and IsoP levels, and the lowest MTL CMRglc on PET, compared to all other subgroups. These data indicate a relationship between MTL CMRglc reductions, tau pathology and lipid membrane peroxidation in normal individuals at risk for late-onset AD, and suggest that the relationship between these biomarkers becomes tighter when subjects start to show memory deficits. Longitudinal follow-up examinations of these subjects are needed to determine whether the observed CSF and CMRglc abnormalities foreshadow clinical decline.

Conclusions

In recent years, several FDG-PET studies have shown that brain glucose hypometabolism in the hippocampal formation, combined with cortical abnormalities, is useful in accurately distinguishing AD from normal aging and, to some extent, from other dementias. These MTL changes can be found in both sporadic AD and early onset FAD individuals, even at the stages where patients are cognitively normal. For sporadic AD, this pattern of hypometabolism has also been reported among those at increased risk for AD, such as those with subjective memory complaints, carriers of the ApoE-4 genotype, and those with a maternal history of AD. Consequently, FDG-PET imaging is a candidate modality for detecting subtle brain changes in early AD.

There is considerable evidence that CMRglc is sensitive to progression effects[152] and can be used as outcome measure in long-term treatment studies of AD.[76,153] A major advantage of PET over other clinically based assessments, such as cognitive performance, is the potential for reducing the sample sizes and study duration. It was estimated that, in order to detect a 33% treatment response with 80% power in a typical 1-year, double-blind, placebo-controlled treatment study, using the MMSE as endpoint would require 224 AD patients per group, whereas a minimal 36 patients per group would be needed for an FDG-PET study.[153]

Further longitudinal studies are necessary to examine whether the risk for developing a specific form of dementia can be predicted based on the detection of individual FDG-PET

patterns of CMRglc abnormalities among cognitively normal subjects or subjects with MCI. Currently, the AD Neuro-imaging Initiative (ADNI) is pooling large amounts of longitudinal data which may provide this opportunity in the future (see: http://www.loni.ucla.edu/ADNI/).

There remains a great need to increase the preclinical diagnostic specificity, and to test whether the combination of the disease-sensitive CMRglc measures with pathology-specific biomarkers, such as with CSF measures of Aβ or tauopathy proteins, or amyloid PET imaging, would improve the differential diagnosis of AD, prior to the onset of dementia.

Accurate characterization of the extent and nature of brain damage in individual patients, based on converging evidence from different biomarkers, could play an important role in the prediction of subjects' clinical course. Other potential benefits include the selection of individualized treatment plans and screening of patients with more uniform underlying pathology for targeted research and drug trials. Overall, CMRglc FDG-PET measures predict cognitive decline from normal aging with sensitivity and specificity greater than 80%, and correlate with clinical progression in AD. CMRglc is therefore a promising candidate biomarker for AD.[19] Overall, PET imaging appears to be of great clinical importance to the early and differential diagnosis of Alzheimer's type dementia.

REFERENCES

1. Kung HC, Hoyert DL, Xu J, Murphy SL. Deaths: final data for 2005. *Natl Vital Stat Rep* 2008;56:1-120.
2. 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. *Neurol* 1984;34:939-944.
3. Hebert LE, Scherr PA, Bienias JL, Bennett DA, Evans DA. Alzheimer disease in the US population: prevalence estimates using the 2000 census. *Arch Neurol* 2003;60:1119-1122.
4. Mirra SS, Heyman A, McKeel D, Sumi SM, Crain BJ, Brownlee LM, et al. The Consortium to Establish a Registry for Alzheimer's Disease (CERAD). Part II. Standardization of the neuropathologic assessment of Alzheimer's disease. *Neurol* 1991;41:479-486.
5. Price JL, Morris JC. Tangles and plaques in nondemented aging and "preclinical" Alzheimer's disease. *Ann Neurol* 1999;45:358-368.
6. Braak H, Braak E. Development of Alzheimer-related neurofibrillary changes in the neocortex inversely recapitulates cortical myelogenesis. *Acta Neuropathol* 1996;92:197-201.
7. Delacourte A, David JP, Sergeant N, Buée L, Wattez A, Vermersch P,

et al. The biochemical pathway of neurofibrillary degeneration in aging and Alzheimer's disease. *Neurol* 1999;52:1158-1165.
8. Morris JC, Storandt M, McKeel DW Jr, Rubin EH, Price JL, Grant EA, et al. Cerebral amyloid deposition and diffuse plaques in "normal" aging: Evidence for presymptomatic and very mild Alzheimer's disease. *Neurology* 1996;46:707-719.
9. Braak H, Braak E. Neuropathological stageing of Alzheimer-related changes. *Acta Neuropathol* 1991;82:239-259.
10. Morrison JH, Hof PR. Life and death of neurons in the aging brain. *Science* 1997;278:412-419.
11. Hyman BT, Van Hoesen GW, Damasio AR, Barnes CL. Alzheimer's disease: cell-specific pathology isolates the hippocampal formation. *Science* 1984;225:1168-1170.
12. Etiene D, Kraft J, Ganju N, Gomez-Isla T, Gemelli B, Hyman BT et al. Cerebrovascular Pathology Contributes to the Heterogeneity of Alzheimer's Disease. *J Alzheimers Dis* 1998;1:119-134.
13. Snowdon DA, Greiner LH, Mortimer JA, Riley KP, Greiner PA, Markesbery WR. Brain infarction and the clinical expression of Alzheimer's disease. The Nun Study. *JAMA* 1997;277:813-817.
14. Arriagada PV, Marzloff K, Hyman BT. Distribution of Alzheimer-type pathologic changes in nondemented elderly individuals matches the pattern in Alzheimer's disease. *Neurology* 1992;42:1681-1688.
15. Giannakopoulos P, Hof PR, Mottier S, Michel JP, Bouras C. Neuropathological changes in the cerebral cortex of 1258 cases from a geriatric hospital: retrospective clinicopathological evaluation of a 10-year autopsy population. *Acta Neuropathol* 1994;87:456-468.
16. Ulrich J. Alzheimer changes in nondemented patients younger than sixty-five: possible early stages of Alzheimer's disease and senile dementia of Alzheimer type. *Ann Neurol* 1985;17:273-277.
17. Selkoe DJ. Alzheimer's disease: genotypes, phenotype, and treatments. *Science* 1997;275:630-631.
18. Lambert MP, Barlow AK, Chromy BA, Edwards C, Freed R, Liosatos M, et al. Diffusible, nonfibrillar ligands derived from Abeta1-42 are potent central nervous system neurotoxins. *Proc Natl Acad Sci U S A* 1998;95:6448-6453.
19. Consensus report of the Working Group on: "Molecular and Biochemical Markers of Alzheimer's Disease". The Ronald and Nancy Reagan Research Institute of the Alzheimer's Association and the National Institute on Aging. *Neurobiol Aging* 1998;19:109-116.
20. Nestor PJ, Scheltens P, Hodges JR. Advances in the early detection of Alzheimer's disease. *Nat Med* 2004;10 Suppl:S34-S41.
21. de Leon MJ, McRae T, Tsai JR, George AE, Marcus DL, Freedman M, et al. Abnormal cortisol response in Alzheimer's disease linked to hippocampal atrophy. *Lancet* 1988;2:391-392.
22. de Leon MJ, George AE, Stylopoulos LA, Smith G, Miller DC. Early marker for Alzheimer's disease: the atrophic hippocampus. *Lancet* 1989;2:672-673.
23. Jack CR Jr, Petersen RC, Xu Y, O'Brien PC, Smith GE, Ivnik RJ, et al. Rates of hippocampal atrophy correlate with change in clinical status in aging and AD. *Neurology* 2000;55:484-489.
24. Rusinek H, De Santi S, Frid D, Tsui W, Tarshish C, Convit A, et al. Regional brain atrophy rate predicts future cognitive decline: 6-year longitudinal MR imaging study of normal aging. *Radiology* 2003;229:691-696.
25. den Heijer T, Geerlings MI, Hoebeek FE, Hofman A, Koudstaal PJ, Breteler MM. Use of hippocampal and amygdalar volumes on magnetic resonance imaging to predict dementia in cognitively intact elderly people. *Arch Gen Psychiatry* 2006;63:57-62.
26. O'Sullivan M, Ngo E, Viswanathan A, Jouvent E, Gschwendtner A, Saemann PG, et al. Hippocampal volume is an independent predictor of cognitive performance in CADASIL. *Neurobiol Aging* 2009;30:890-897.
27. Sokoloff L. Relation between physiological functions and energy metabolism in the central nervous system. *J Neurochem* 1977;29:13-26.
28. Magistretti PJ, Pellerin L, Rothman DL, Shulman RG. Energy on de-

mand. *Science* 1999;283:496-497.

29. Attwell D, Iadecola C. The neural basis of functional brain imaging signals. *Trends Neurosci* 2002;25:621-625.

30. Malonek D, Grinvald A. Interactions between electrical activity and cortical microcirculation revealed by imaging spectroscopy: implications for functional brain mapping. *Science* 1996;272:551-554.

31. Rocher AB, Chapon F, Blaizot X, Baron JC, Chavoix C. Resting-state brain glucose utilization as measured by PET is directly related to regional synaptophysin levels: a study in baboons. *Neuroimage* 2003; 20:1894-1898.

32. Magistretti PJ, Pellerin L. The contribution of astrocytes to the 18F-2-deoxyglucose signal in PET activation studies. *Mol Psychiatry* 1996; 1:445-452.

33. Pellerin L, Magistretti PJ. Glutamate uptake into astrocytes stimulates aerobic glycolysis: a mechanism coupling neuronal activity to glucose utilization. *Proc Natl Acad Sci U S A* 1994;91:10625-10629.

34. Mosconi L. Brain glucose metabolism in the early and specific diagnosis of Alzheimer's disease. FDG-PET studies in MCI and AD. *Eur J Nucl Med Mol Imaging* 2005;32:486-510.

35. Friedland RP, Budinger TF, Ganz E, Yano Y, Mathis CA, Koss B, et al. Regional cerebral metabolic alterations in dementia of the Alzheimer type: positron emission tomography with [18F]fluorode-oxyglucose. *J Comput Assist Tomogr* 1983;7:590-598.

36. Frackowiak RS, Pozzilli C, Legg NJ, DuBoulay GH, Marshall J, Lenzi GL, et al. A prospective study of regional cerebral blood flow and oxygen utilization in dementia using positron emission tomography and oxygen-15. *J Cereb Blood Flow Metab* 1981;1:S453-S454.

37. Minoshima S, Giordani B, Berent S, Frey KA, Foster NL, Kuhl DE. Metabolic reduction in the posterior cingulate cortex in very early Alzheimer's disease. *Ann Neurol* 1997;42:85-94.

38. Ferris SH, de Leon MJ, Wolf AP, Farkas T, Christman DR, Reisberg B, et al. Positron emission tomography in the study of aging and senile dementia. *Neurobiol Aging* 1980;1:127-131.

39. Kim EJ, Cho SS, Jeong Y, Park KC, Kang SJ, Kang E, et al. Glucose metabolism in early onset versus late onset Alzheimer's disease: an SPM analysis of 120 patients. *Brain* 2005;128:1790-1801.

40. Foster NL, Chase TN, Mansi L, Brooks R, Fedio P, Patronas NJ, et al. Cortical abnormalities in Alzheimer's disease. *Ann Neurol* 1984;16: 649-654.

41. Mazziotta JC, Phelps ME. Positron Emission Tomography studies of the brain. In: Phelps ME, Mazziotta JC, Schelbert H. *Positron Emission Tomography & Autoradiography: Principles & Applications for the Brain & Heart*. New York: Raven Press, 1986;493-579.

42. Herholz K, Salmon E, Perani D, Baron JC, Holthoff V, Frölich L, et al. Discrimination between Alzheimer dementia and controls by automated analysis of multicenter FDG PET. *Neuroimage* 2002;17: 302-316.

43. Silverman DH, Small GW, Chang CY, Lu CS, Kung de Aburto MA, Chen W, et al. Positron emission tomography in evaluation of dementia: Regional brain metabolism and long-term outcome. *JAMA* 2001; 286:2120-2127.

44. Szelies B, Mielke R, Herholz K, Heiss WD. Quantitative topographical EEG compared to FDG PET for classification of vascular and degenerative dementia. *Electroencephalogr Clin Neurophysiol* 1994; 91:131-139.

45. Tanzi RE, Bertram L. New frontiers in Alzheimer's disease genetics. *Neuron* 2001;32:181-184.

46. Kennedy AM, Newman SK, Frackowiak RS, Cunningham VJ, Roques P, Stevens J, et al. Chromosome 14 linked familial Alzheimer's disease. A clinico-pathological study of a single pedigree. *Brain* 1995; 118:185-205.

47. Kennedy AM, Frackowiak RS, Newman SK, Bloomfield PM, Seaward J, Roques P, et al. Deficits in cerebral glucose metabolism demonstrated by positron emission tomography in individuals at risk of familial Alzheimer's disease. *Neurosci Lett* 1995;186:17-20.

48. Mosconi L, Sorbi S, de Leon MJ, Li Y, Nacmias B, Myoung PS, et al. Hypometabolism exceeds atrophy in presymptomatic early-onset familial Alzheimer's disease. *J Nucl Med* 2006;47:1778-1786.

49. de Leon MJ, Convit A, Wolf OT, Tarshish CY, DeSanti S, Rusinek H, et al. Prediction of cognitive decline in normal elderly subjects with 2-[(18)F]fluoro-2-deoxy-D-glucose/positron-emission tomography (FDG/PET). *Proc Natl Acad Sci U S A* 2001;98:10966-10971.

50. Mosconi L, De Santi S, Li J, Tsui WH, Li Y, Boppana M, et al. Hippocampal hypometabolism predicts cognitive decline from normal aging. *Neurobiol Aging* 2008;29:676-692.

51. Petersen RC, Stevens JC, Ganguli M, Tangalos EG, Cummings JL, DeKosky ST. Practice parameter: early detection of dementia: mild cognitive impiarment (an evidence-based review). Report of the Quality Standards Subcommittee of the American Academy of Neurology. *Neurology* 2001;56:1133-1142.

52. Gauthier S, Reisberg B, Zaudig M, Petersen RC, Ritchie K, Broich K, et al. Mild cognitive impairment. *Lancet* 2006;367:1262-1270.

53. Herholz K, Nordberg A, Salmon E, Perani D, Kessler J, Mielke R, et al. Impairment of neocortical metabolism predicts progression in Alzheimer's disease. *Dement Geriatr Cogn Disord* 1999;10:494-504.

54. Arnaiz E, Jelic V, Almkvist O, Wahlund LO, Winblad B, Valind S, et al. Impaired cerebral glucose metabolism and cognitive functioning predict deterioration in mild cognitive impairment. *Neuroreport* 2001;12: 851-855.

55. 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;22:529-539.

56. Nestor PJ, Fryer TD, Smielewski P, Hodges JR. Limbic hypometabolism in Alzheimer's disease and mild cognitive impairment. *Ann Neurol* 2003;54:343-351.

57. Chételat G, Desgranges B, de la Sayette V, Viader F, Eustache F, Baron JC. Mild cognitive impairment: Can FDG-PET predict who is to rapidly convert to Alzheimer's disease? *Neurology* 2003;60:1374-1377.

58. 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;30:1104-1113.

59. 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;64:1860-1867.

60. 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 (18)F-FDG PET. *J Nucl Med* 2005;46:1625-1632.

61. Mosconi L, De Santi S, Li Y, Li J, Zhan J, Tsui WH, et al. Visual rating of medial temporal lobe metabolism in mild cognitive impairment and Alzheimer's disease using FDG-PET. *Eur J Nucl Med Mol Imaging* 2006;33:210-221.

62. Anchisi D, Borroni B, Franceschi M, Kerrouche N, Kalbe E, Beuthien-Beumann B, et al. Heterogeneity of brain glucose metaboism in mild cognitive impairment and clinical progression to Alzheimer disease. *Arch Neurol* 2005;62:1728-1733.

63. Haxby JV, Grady CL, Koss E, Horwitz B, Heston L, Schapiro M, et al. Longitudinal study of cerebral metabolic asymmetries and associated neuropsychological patterns in early dementia of the Alzheimer type. *Arch Neurol* 1990;47:753-760.

64. Reed BR, Jagust WJ, Seab JP, Ober BA. Memory and regional cerebral blood flow in mildly symptomatic Alzheimer's disease. *Neurology* 1989;39:1537-1539.

65. Berent S, Giordani B, Foster N, Minoshima S, Lajiness-O'Neill R, Koeppe R, et al. Neuropsychological function and cerebral glucose utilization in isolated memory impairment and Alzheimer's disease. *J Psychiatr Res* 1999;33:7-16.

66. 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;63:2332-2340.

67. Petersen RC, Smith GE, Waring SC, Ivnik RJ, Tangalos EG, Kokmen E. Mild cognitive impairment: clinical characterization and outcome. *Arch Neurol* 1999;56:303-308.

68. Jagust W, Gitcho A, Sun F, Kuczynski B, Mungas D, Haan M. Brain imaging evidence of preclinical Alzheimer's disease in normal aging. *Ann Neurol* 2006;59:673-681.

69. Skurk T, Alberti-Huber C, Herder C, Hauner H. Relationship between adipocyte size and adipokine expression and secretion. *J Clin Endocrinol Metab* 2007;92:1023-1033.

70. Corder EH, Saunders AM, Strittmatter WJ, Schmechel DE, Gaskell PC, Small GW, et al. Gene dose of apolipoprotein E type 4 Allele and the risk of Alzheimer's disease in late onset families. *Science* 1993;261: 921-923.

71. Farrer LA, Cupples LA, Haines JL, Hyman B, Kukull WA, Mayeux R, et al. Effects of age, sex, and ethnicity on the association between apolipoprotein E genotype and Alzheimer disease. A meta-analysis. APOE and Alzheimer Disease Meta Analysis Consortium. *JAMA* 1997;278:1349-1356.

72. Laws SM, Hone E, Gandy S, Martins RN. Expanding the association between the APOE gene and the risk of Alzheimer's disease: possible roles for APOE promoter polymorphisms and alterations in APOE transcription. *J Neurochem* 2003;84:1215-1236.

73. Small GW, Mazziotta JC, Collins MT, Baxter LR, Phelps ME, Mandelkern MA, et al. Apolipoprotein E type 4 allele and cerebral glucose metabolism in relatives at risk for familial Alzheimer disease. *JAMA* 1995;273:942-947.

74. Reiman EM, Caselli RJ, Yun LS, Chen K, Bandy D, Minoshima S, et al. Preclinical evidence of Alzheimer's disease in persons homozygous for the epsilon 4 allele for apolipoprotein E. *N Engl J Med* 1996;334: 752-758.

75. Small GW, Ercoli LM, Silverman DH, Huang SC, Komo S, Bookheimer S, et al. Cerebral metabolic and cognitive decline in persons at genetic risk for Alzheimer's disease. *Proc Natl Acad Sci U S A* 2000;97:6037-6042.

76. Reiman EM, Caselli RJ, Chen K, Alexander GE, Bandy D, Frost J. Declining brain activity in cognitively normal apolipoprotein E epsilon 4 heterozygotes: A foundation for using positron emission tomography to efficiently test treatments to prevent Alzheimer's disease. *Proc Natl Acad Sci U S A* 2001;98:3334-3339.

77. Reiman EM, Chen K, Alexander GE, Caselli RJ, Bandy D, Osborne D, et al. Functional brain abnormalities in young adults at genetic risk for late-onset Alzheimer's dementia. *Proc Natl Acad Sci U S A* 2004;101:284-289.

78. 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;44:288-291.

79. 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;63:609-618.

80. Geerlings MI, Jonker C, Bouter LM, Adér HJ, Schmand B. Association between memory complaints and incident Alzheimer's disease in elderly people with normal baseline cognition. *Am J Psychiatry* 1999; 156:531-537.

81. Bobinski M, de Leon MJ, Convit A, De Santi S, Wegiel J, Tarshish CY, et al. MRI of entorhinal cortex in mild Alzheimer's disease. *Lancet* 1999;353:38-40.

82. Cupples LA, Farrer LA, Sadovnick AD, Relkin N, Whitehouse P, Green RC. Estimating risk curves for first-degree relatives of patients with Alzheimer's disease: the REVEAL study. *Genet Med* 2004;6: 192-196.

83. Green RC, Cupples LA, Go R, Benke KS, Edeki T, Griffith PA, et al. Risk of dementia among white and African American relatives of pa-

tients with Alzheimer disease. *JAMA* 2002;287:329-336.

84. Silverman JM, Ciresi G, Smith CJ, Marin DB, Schnaider-Beeri M. Variability of familial risk of Alzheimer disease across the late life span. *Arch Gen Psychiatry* 2005;62:565-573.

85. Mosconi L, Brys M, Switalski R, Mistur R, Glodzik L, Pirraglia E, et al. Maternal family history of Alzheimer's disease predisposes to reduced brain glucose metabolism. *Proc Natl Acad Sci U S A* 2007;104: 19067-19072.

86. Mosconi L, Mistur R, Switalski R, Brys M, Glodzik L, Rich K, et al. Declining brain glucose metabolism in normal individuals with a maternal history of Alzheimer disease. *Neurology* 2009;72:513-520.

87. Lin MT, Beal MF. Mitochondrial dysfunction and oxidative stress in neurodegenerative diseases. *Nature* 2006;443:787-795.

88. Klunk WE, Engler H, Nordberg A, Wang Y, Blomqvist G, Holt DP, et al. Imaging brain amyloid in Alzheimer's disease with Pittsburgh Compound-B. *Ann Neurol* 2004;55:306-319.

89. Rowe CC, Ng S, Ackermann U, Gong SJ, Pike K, Savage G, et al. Imaging beta-amyloid burden in aging and dementia. *Neurology* 2007; 68:1718-1725.

90. Kemppainen NM, Aalto S, Wilson IA, Någren K, Helin S, Brück A, et al. Voxel-based analysis of PET amyloid ligand [11C]PIB uptake in Alzheimer disease. *Neurology* 2006;67:1575-1580.

91. Pike KE, Savage G, Villemagne VL, Ng S, Moss SA, Maruff P, et al. Beta-amyloid imaging and memory in non-demented individuals: evidence for preclinical Alzheimer's disease. *Brain* 2007;130:2837-2844.

92. Mintun MA, Larossa GN, Sheline YI, Dence CS, Lee SY, Mach RH, et al. [11C]PIB in a nondemented population: potential antecedent marker of Alzheimer disease. *Neurology* 2006;67:446-452.

93. Fagan AM, Mintun MA, Mach RH, Lee SY, Dence CS, Shah AR, et al. Inverse relation between in vivo amyloid imaging load and cerebrospinal fluid Abeta42 in humans. *Ann Neurol* 2006;59:512-519.

94. Klunk WE, Mathis CA, Price JC, Lopresti BJ, DeKosky ST. Two-year follow-up of amyloid deposition in patients with Alzheimer's disease. *Brain* 2006;129:2805-2807.

95. Drzezga A, Grimmer T, Henriksen G, Stangier I, Perneczky R, Diehl-Schmid J, et al. Imaging of amyloid plaques and cerebral glucose metabolism in semantic dementia and Alzheimer's disease. *Neuroimage* 2008;39:619-633.

96. Mathis CA, Wang Y, Klunk WE. Imaging beta-amyloid plaques and neurofibrillary tangles in the aging human brain. *Curr Pharm Des* 2004;10:1469-1492.

97. Lockhart A, Lamb JR, Osredkar T, Sue LI, Joyce JN, Ye L, et al. PIB is a non-specific imaging marker of amyloid-beta (Abeta) peptide-related cerebral amyloidosis. *Brain* 2007;130:2607-2615.

98. Agdeppa ED, Kepe V, Liu J, Flores-Torres S, Satyamurthy N, Petric A, et al. Binding characteristics of radiofluorinated 6-dialkylamino-2-naphthylethylidene derivatives as positron emission tomography imaging probes for beta-amyloid plaques in Alzheimer's disease. *J Neurosci* 2001;21:RC189.

99. Small GW, Kepe V, Ercoli LM, Siddarth P, Bookheimer SY, Miller KJ, et al. PET of brain amyloid and tau in mild cognitive impairment. *N Engl J Med* 2006;355:2652-2663.

100. de Leon MJ, Mosconi L, Logan J. Seeing what Alzheimer saw. *Nat Med* 2007;13:129-131.

101. Powell MR, Smith GE, Knopman DS, Parisi JE, Boeve BF, Petersen RC, et al. Cognitive measures predict pathologic Alzheimer disease. *Arch Neurol* 2006;63:865-868.

102. Rowe CC, Ackermann U, Browne W, Mulligan R, Pike KL, O'Keefe G, et al. Imaging of amyloid beta in Alzheimer's disease with 18F-BAY94-9172, a novel PET tracer: proof of mechanism. *Lancet Neurol* 2008;7:129-135.

103. Mitchell A, Brindle N. CSF phosphorylated tau--does it constitute an accurate biological test for Alzheimer's disease? *Int J Geriat Psychiatry* 2003;18:407-411.

104. Brys M, Mosconi L, De Santi S, Rich KE, de Leon MJ. CSF bio-

markers for mild cognitive impairment. *J Aging Health* 2006;2:111-121.

105. Blennow K, Hampel H. CSF markers for incipient Alzheimer's disease. *Lancet Neurol* 2003;2:605-613.

106. Blennow K, de Leon MJ, Zetterberg H. Alzheimer's disease. *Lancet Neurology* 2006;368:387-403.

107. Arai H, Ishiguro K, Ohno H, Moriyama M, Itoh N, Okamura N, et al. CSF phosphorylated tau protein and mild cognitive impairment: a prospective study. *Exp Neurol* 2000;166:201-203.

108. Hansson O, Zetterberg H, Buchhave P, Londos E, Blennow K, Minthon L. Association between CSF biomarkers and incipient Alzheimer's disease in patients with mild cognitive impairment: a follow-up study. *Lancet Neurol* 2006;5:228-234.

109. Brys M, Pirraglia E, Rich K, Rolstad S, Mosconi L, Switalski R, et al. Prediction and longitudinal study of CSF biomarkers in mild cognitive impairment. *Neurobiol Aging* 2009;30:682-690.

110. Büerger née Buch K, Padberg F, Nolde T, Teipel SJ, Stübner S, Haslinger A, et al. Cerebrospinal fluid tau protein shows a better discrimination in young old (<70 years) than in old old patients with Alzheimer's disease compared with controls. *Neurosci Lett* 1999;277:21-24.

111. Hulstaert F, Blennow K, Ivanoiu A, Schoonderwaldt HC, Riemenschneider M, De Deyn PP, et al. Improved discrimination of AD patients using beta-amyloid (1-42) and tau levels in CSF. *Neurology* 1999; 52:1555-1562.

112. Andreasen N, Minthon L, Davidsson P, Vanmechelen E, Vanderstichele H, Winblad B, et al. Evaluation of CSF-tau and CSF-Abeta42 as diagnostic markers for Alzheimer disease in clinical practice. *Arch Neurol* 2001;58:373-379.

113. Glodzik-Sobanska L, Pirraglia E, Brys M, De Santi S, Mosconi L, Rich KE, et al. The effects of normal aging and ApoE genotype on the levels of CSF biomarkers for Alzheimer's disease. *Neurobiol Aging* 2009;30:672-681.

114. Arai H, Morikawa Y, Higuchi M, Matsui T, Clark CM, Miura M, et al. Cerebrospinal fluid tau levels in neurodegenerative diseases with distinct tau-related pathology. *Biochem Biophys Res Commun* 1997; 236:262-264.

115. Mollenhauer B, Bibl M, Trenkwalder C, Stiens G, Cepek L, Steinacker P, et al. Follow-up investigations in cerebrospinal fluid of patients with dementia with Lewy bodies and Alzheimers disease. *J Neural Transm* 2005;112:933-948.

116. Hesse C, Rosengren L, Andreasen N, Davidsson P, Vanderstichele H, Vanmechelen E, et al. Transient increase in total tau but not phospho-tau in human cerebrospinal fluid after acute stroke. *Neurosci Lett* 2001; 297:187-190.

117. Buerger K, Zinkowski R, Teipel SJ, Tapiola T, Arai H, Blennow K, et al. Differential diagnosis of Alzheimer disease with cerebrospinal fluid levels of tau protein phosphorylated at threonine 231. *Arch Neurol* 2002;59:1267-1272.

118. Buerger K, Zinkowski R, Teipel SJ, Arai H, DeBernardis J, Kerkman D, et al. Differentiation of geriatric major depression from Alzheimer's disease with CSF tau protein phosphorylated at threonine 231. *Am J Psychiatry* 2003;160:376-379.

119. Buerger K, Otto M, Teipel SJ, Zinkowski R, Blennow K, DeBernardis J, et al. Dissociation between CSF total tau and tau protein phosphorylated at threonine 231 in Creutzfeldt-Jakob disease. *Neurobiol Aging* 2006;27:10-15.

120. Vanmechelen E, Vanderstichele H, Davidsson P, Van Kerschaver E, Van Der Perre B, Sjögren M, et al. Quantification of tau phosphorylated at threonine 181 in human cerebrospinal fluid: a sandwich ELISA with a synthetic phosphopeptide for standardization. *Neurosci Lett* 2000;285:49-52.

121. Sjögren M, Davidsson P, Tullberg M, Minthon L, Wallin A, Wikkelso C, et al. Both total and phosphorylated tau are increased in Alzheimer's disease. *J Neurol Neurosurg Psychiatry* 2001;70:624-630.

122. Ishiguro K, Ohno H, Arai H, Yamaguchi H, Urakami K, Park JM, et al. Phosphorylated tau in human cerebrospinal fluid is a diagnostic marker for Alzheimer's disease. *Neurosci Lett* 1999;270:91-94.

123. Parnetti L, Lanari A, Amici S, Gallai V, Vanmechelen E, Hulstaert F, et al. CSF phosphorylated tau is a possible marker for discriminating Alzheimer's disease from dementia with Lewy bodies. Phospho-Tau International Study Group. *Neurol Sci* 2001;22:77-78.

124. de Leon MJ, DeSanti S, Zinkowski R, Mehta PD, Pratico D, Segal S, et al. Longitudinal CSF and MRI biomarkers improve the diagnosis of mild cognitive impairment. *Neurobiol Aging* 2006;27:394-401.

125. de Leon MJ, Segal S, Tarshish CY, DeSanti S, Zinkowski R, Mehta PD, et al. Longitudinal cerebrospinal fluid tau load increases in mild cognitive impairment. *Neurosci Lett* 2002;333:183-186.

126. Andreasen N, Blennow K. CSF biomarkers for mild cognitive impairment and early Alzheimer's disease. *Clin Neurol Neurosurg* 2005; 107:165-173.

127. Tapiola T, Pirttilä T, Mikkonen M, Mehta PD, Alafuzoff I, Koivisto K, et al. Three-year follow-up of cerebrospinal fluid tau, B-amyloid 42 and 40 concentrations in Alzheimer's disease. *Neurosci Lett* 2000; 280:119-122.

128. Bading JR, Yamada S, Mackic JB, Kirkman L, Miller C, Calero M, et al. Brain clearance of Alzheimer's amyloid-beta40 in the squirrel monkey: a SPECT study in a primate model of cerebral amyloid angiopathy. *J Drug Target* 2002;10:359-368.

129. Silverberg GD, Levinthal E, Sullivan EV, Bloch DA, Chang SD, Leverenz J, et al. Assessment of low-flow CSF drainage as a treatment for AD: results of a randomized pilot study. *Neurology* 2002;59: 1139-1145.

130. Silverberg GD, Heit G, Huhn S, Jaffe RA, Chang SD, Bronte-Stewart H, et al. The cerebrospinal fluid production rate is reduced in dementia of the Alzheimer's type. *Neurology* 2001;57:1763-1766.

131. Peskind ER, Li G, Shofer J, Quinn JF, Kaye JA, Clark CM, et al. Age and apolipoprotein E*4 allele effects on cerebrospinal fluid beta-amyloid 42 in adults with normal cognition. *Arch Neurol* 2006;63:936-939.

132. Fagan AM, Roe CM, Xiong C, Mintun MA, Morris JC, Holtzman DM. Cerebrospinal fluid tau/beta-amyloid(42) ratio as a prediction of cognitive decline in nondemented older adults. *Arch Neurol* 2007; 64: 343-349.

133. Fukuyama R, Mizuno T, Mori S, Nakajima K, Fushiki S, Yanagisawa K. Age-dependent change in the levels of Abeta40 and Abeta42 in cerebrospinal fluid from control subjects, and a decrease in the ratio of Abeta42 to Abeta40 level in cerebrospinal fluid from Alzheimer's disease patients. *Eur Neurol* 2000;43:155-160.

134. Shoji M, Kanai M, Matsubara E, Tomidokoro Y, Shizuka M, Ikeda Y, et al. The levels of cerebrospinal fluid Abeta40 and Abeta42(43) are regulated age-dependently. *Neurobiol Aging* 2001;22:209-215.

135. Kanai M, Matsubara E, Isoe K, Urakami K, Nakashima K, Arai H, et al. Longitudinal study of cerebrospinal fluid levels of tau, A beta1-40, and A beta1-42(43) in Alzheimer's disease: a study in Japan. *Ann Neurol* 1998;44:17-26.

136. Mehta PD, Pirttilä T, Mehta SP, Sersen EA, Aisen PS, Wisniewski HM. Plasma and cerebrospinal fluid levels of amyloid beta proteins 1-40 and 1-42 in Alzheimer disease. *Arch Neurol* 2000;57:100-105.

137. Markesbery WR. Oxidative stress hypothesis in Alzheimer's disease. *Free Radic Biol Med* 1997;23:134-147.

138. Montine TJ, Markesbery WR, Zackert W, Sanchez SC, Roberts LJ 2nd, Morrow JD. The magnitude of brain lipid peroxidation correlates with the extent of degeneration but not with density of neuritic plaques or neurofibrillary tangles or with APOE genotype in Alzheimer's disease patients. *Am J Pathol* 1999;155:863-868.

139. Montine TJ, Markesbery WR, Morrow JD, Roberts LJ 2nd. Cerebrospinal fluid F2-isoprostane levels are increased in Alzheimer's disease. *Ann Neurol* 1998;44:410-413.

140. Praticó D. F(2)-isoprostanes: sensitive and specific non-invasive indices of lipid peroxidation in vivo. *Atherosclerosis* 1999;147:1-10.

141. Praticó D, Lawson JA, Rokach J, Fitzgerald GA. The isoprostanes in

biology and medicine. *Trends Endocrinol Metab* 2001;12:243-247.

142. Nunomura A, Perry G, Aliev G, Hirai K, Takeda A, Balraj EK, et al. Oxidative damage is the earliest event in Alzheimer disease. *J Neuropathol Exp Neurol* 2001;60:759-767.

143. Praticó D, Uryu K, Leight S, Trojanowski JQ, Lee VM. Increased lipid peroxidation precedes amyloid plaque formation in an animal model of Alzheimer amyloidosis. *J Neurosci* 2001;21:4183-4187.

144. Markesbery WR, Kryscio RJ, Lovell MA, Morrow JD. Lipid peroxidation is an early event in the brain in amnestic mild cognitive impairment. *Ann Neurol* 2005;58:730-735.

145. Montine TJ, Beal MF, Cudkowicz ME, O'Donnell H, Margolin RA, McFarland L, et al. Increased CSF F2-isoprostane concentration in probable AD. *Neurology* 1999;52:562-565.

146. Praticó D, Clark CM, Lee VM, Trojanowski JQ, Rokach J, FitzGerald GA. Increased 8,12-iso-iPF2alpha-VI in Alzheimer's disease: correlation of a noninvasive index of lipid peroxidation with disease severity. *Ann Neurol* 2000;48:809-812.

147. Praticó D, Clark CM, Liun F, Lee VY, Trojanowski JQ. Increase of brain oxidative stress in mild cognitive impairment: a possible predictor of Alzheimer disease. *Arch Neurol* 2002;59:972-976.

148. de Leon MJ, Mosconi L, Li Y, De Santi S, Yao Y, Tsui WH, et al. Longitudinal CSF isoprostane and MRI atrophy in the progression to AD. *J Neurol* 2007;254:1666-1675.

149. Yao Y, Zhukareva V, Sung S, Clark CM, Rokach J, Lee VM, et al. Enhanced brain levels of 8,12-iso-iPF2alpha-VI differentiate AD from frontotemporal dementia. *Neurology* 2003;61:475-478.

150. Grossman M, Farmer J, Leight S, Work M, Moore P, Van Deerlin V, et al. Cerebrospinal fluid profile in frontotemporal dementia and Alzheimer's disease. *Ann Neurol* 2005;57:721-729.

151. Montine TJ, Kaye JA, Montine KS, McFarland L, Morrow JD, Quinn JF. Cerebrospinal fluid abeta42, tau, and f2-isoprostane concentrations in patients with Alzheimer disease, other dementias, and in age-matched controls. *Arch Pathol Lab Med* 2001;125:510-512.

152. Mosconi L, Brys M, Glodzik-Sobanska L, De Santi S, Rusinek H, de Leon MJ. Early detection of Alzheimer's disease using neuroimaging. *Exp Gerontol* 2007;42:129-138.

153. Alexander GE, Chen K, Pietrini P, Rapoport SI, Reiman EM. Longitudinal PET Evaluation of Cerebral Metabolic Decline in Dementia: A Potential Outcome Measure in Alzheimer's Disease Treatment Studies. *Am J Psychiatry* 2002;159:738-745.