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Chapter 5

Cortical phase changes measured using 7T MRI in subjects with subjective cognitive impairment, and their association with cognitive function

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Abstract

Studies have suggested that in subjects with subjective cognitive impairment (SCI), Alzheimer's disease (AD)-like changes may occur in the brain. Recently, an in vivo study has indicated the potential of ultra-high field MRI to visualize amyloid-β-associated changes in the cortex in AD patients, manifested by a phase shift on T_2^*-weighted MRI scans. The main aim of this study was to investigate whether cortical phase shifts on T_2^*-weighted images at 7T in subjects with SCI can be detected, possibly implicating the deposition of Aβ plaques and associated iron. Cognitive tests and T_2^*- weighted scans using a 7T MRI system were performed in 28 AD patients, 18 subjects with SCI and 27 healthy controls (HC). Cortical phase shifts were measured. Univariate general linear modeling and linear regression analysis were used to assess the association between diagnosis and cortical phase shift, and between cortical phase shift and the different neuropsychological tests, adjusted for age and gender. The phase shift (mean; 1.19, range: 1.00-1.35) of the entire cortex in AD was higher compared to both SCI (mean; 0.85, range: 0.73-0.99, $p<0.001$) and HC (mean; 0.94, range: 0.79-1.10, $p<0.001$). No AD-like changes, e.g. increased cortical phase shifts, were found in SCI subjects compared with HC. In SCI, a significant association was found between memory function (WMS) and cortical phase shift ($\beta = -0.544$, $p = 0.007$). The major finding of the study is that in SCI subjects an increased cortical phase shift measured at high field is associated with poorer memory performance, although as a group, SCI subjects did not show an increased phase shift compared to HC. This increased cortical phase shift related to memory performance might contribute to the understanding of SCI as it is still unclear if SCI is a sign of pre-clinical AD.

Introduction

Subjective cognitive impairment (SCI) is common in the elderly and refers to a subjective decline in levels of cognitive functioning, which cannot be confirmed by neuropsychological evaluation [1]. Longitudinal population-based studies have reported an association with future cognitive decline [2], and dementia [3]. Moreover, neuroimaging studies have shown that in subjects with SCI, Alzheimer's disease (AD)-like changes such as loss of volume in the medial temporal lobe and hippocampus may already occur before cognitive decline becomes evident [4-9]. This suggests that elderly with SCI may be at risk for developing AD. Alternatively, cognitive complaints may be caused by other factors such as depression, anxiety and quality of life [10-13].

Recently, it has been demonstrated that T_2^*-weighted Magnetic Resonance Imaging (MRI) at high field provides indirect evidence of the presence of AD pathology [14,15]. This approach is based on the finding that in autopsy material of AD patients, amyloid-β (Aβ) deposition and neurofibrillary tangles, as well as tau deficiency, were found to co-localize with cortical iron accumulation [16-19]. Based on the high sensitivity of T_2^*-weighted MRI at 7T in detecting small cerebral iron deposits [20,21], we have shown in a previous study that in AD patients the magnitude of the observed cortical phase shift in T_2^*-weighted phase images was correlated with global cognitive functioning, and demonstrated a high sensitivity in differentiating patients with AD from controls [14]. Based on the fact that Aβ plaques and neurofibrillary tangles occur up to 10 to 20 years before cognitive decline can be measured [22], and the findings that SCI predicts future diagnosis of AD [23,24], it could be hypothesized that in subjects with SCI, differences in T_2^*-weighted cortical phase shifts may already be present before cognitive decline can be determined.

The main aim of this study was to investigate whether cortical phase shifts on T_2^*-weighted images at 7T in subjects with SCI can be detected, suggesting the deposition of Aβ plaques and associated iron. The secondary aim was to investigate which cognitive domains are associated with cortical phase shift in these subjects.

Materials and methods

Participants

This study was approved by the local institutional review board. In all cases, informed consent was obtained according to the declaration of Helsinki [25]. In total 28 AD patients with a mean age of 71.2 years (range from 54 to 86 years, 18 male/10 female), 18 SCI subjects with a mean age of 66.5 years (range from 49 to 85 years, 13 male/5 female) and 27 healthy controls (HC) with a mean age of 68.9 years (range from 52 to 80 years, 16 male/11 female) were included.

AD patients and subjects with SCI were recruited from the memory clinic of the Leiden University Medical Center, the VU University Medical Center in Amsterdam, the Bronovo Hospital in The Hague and the Diaconessen Hospital in Leiden. Memory clinic patients were referred to the hospital by their general practitioner or a medical specialist. Prior to the 7 T study all patients underwent a routine clinical protocol, comprising a whole brain clinical MRI and a battery of neuropsychological tests measuring global cognitive functioning (Mini mental state examination (MMSE) [26] and Cambridge Cognitive Examination (CAMCOG) [27]) , memory (Wechsler Memory Scale (WMS) [28] including digit span forward and backward (working memory)), executive function (Trailmaking test (TMT) part B (cognitive flexibility) [29] and STROOP 3 (inhibition) [30]), psychomotor speed (TMT part A) [29] and depression (abbreviated Geriatric Depression Scale (GDS) [31]). A general medical and neurological examinations were performed by a neurologist, psychiatrist or internist-geriatrician. Diagnosis was made in a multidisciplinary consensus meeting using the NINCDS-ADRDA criteria for diagnosing probable AD [32]. SCI was defined by the subjective feeling of memory decline for which these subjects (based on memory complaints) were referred to the memory clinic, however subjects with SCI performed within the normal range on standard neuropsychological tests (i.e. criteria for MCI, dementia or other neurologic or psychiatric disorder were not fulfilled). Participants with the diagnosis 'SCI' and 'probable AD', who were capable of giving informed consent (MMSE ≥ 19), were selected for inclusion. AD patients and subjects with SCI were selected for inclusion in the 7T study either retrospectively within one year after attending the memory clinic, or else prospectively.

HC subjects were recruited as part of the Leiden Longevity Study, the details of which are described elsewhere [33]. So as not to predispose for familial longevity, only partners of offspring were included. Subjects were included if

they lived independently, had an age ≤ 85 years, a Body Mass Index between 23 and 35 kg/m^2, an MMSE ≥ 25 and a GDS ≤ 4. Individuals who were engaged in heavy/intense physical activity, had been immobile for longer than one week in the three preceding months, had shown psychiatric or behavioral problems, or had used either thyroid medication or immunosuppressants, were excluded. Subjects were screened for cognitive deficits by an internist-geriatrician and subjects with the following diseases were excluded: hemorrhagic and ischemic stroke, Parkinson's disease, dementia, mild cognitive impairment (MCI), diabetes type I or II, rheumatoid arthritis, polymyalgia rheumatica, cancer, heart failure, and chronic obstructive pulmonary disease. Prior to the MRI scan all subjects underwent cognitive testing, measuring global cognitive functioning (MMSE), working memory (digit span forward and backward), executive function (TMT part B (cognitive flexibility) and STROOP 3 (inhibition)), and psychomotor speed (TMT part A).

MRI

Image acquisition

MRI was performed on a whole body human 7T MRI system (Philips Healthcare, Best, the Netherlands) using a quadrature transmit and 16-channel receive head coil. Participants were scanned using a 2D flow-compensated transverse T_2^*-weighted gradient-echo scan including the frontal and parietal regions, which are most prone to Aβ deposition, with a total imaging duration of 10 minutes. Positioning of this stack of slices was planned within the frontal and parietal region above the occipital lobe, using the sagittal plane of the survey scan as a reference. The middle of the stack of slices was positioned through the corpus callosum, just above the thalamus. Imaging parameters were: repetition time/echo time 1764/25 ms, flip angle 45°, slice thickness 1.0 mm with a 0.1 mm interslice gap, 20 slices, 240 x 180 x 22 mm field-of-view, 1024 x 768 matrix size – resulting in an in-plane spatial resolution of 0.24 x 0.24 mm^2. The bandwidth per pixel was 46 Hz, corresponding to a readout length of approximately 22 ms. The frequency and phase encoding directions were along the anterior-posterior and right-left axes, respectively. Shimming up to third order was performed using an image based shimming approach [34]. The phase images were subsequently unwrapped by highpass filtering with a 92x92 kernel [35].

Image analysis

Phase values in the cortex were determined using the transverse 2D T_2^*-weighted gradient echo scans. The phase values of the cortical gray matter (GM) were determined on the unwrapped phase images in regions of interest (ROIs) in four different areas of the brain: frontal, parietal, left temporoparietal and right temporoparietal. Because of the laminar variation in the cortical areas [36], histograms perpendicular to the cortex within these regions were created to measure peak GM phase values, over at least 10 cortical regions per slice and per region. To correct for local macroscopic magnetic field inhomogeneities subcortical white matter (WM) phase values were measured and used as an internal reference value. ROI's were selected containing only the GM or WM, avoiding blood vessels, CSF, WM hyperintensities and other abnormalities which could influence the results. Phase values for GM and WM were measured in these four areas per MRI slice for every other slice (ten in total), resulting in 40 phase values for GM and WM separately per subject. Phase values of the different ROIs of the four regions were averaged. Per subject, the overall phase shift between cortical GM and subcortical WM (lobar cortical phase shift) was calculated for each region and expressed in radians. All phase values reported in our manuscript represent these averages [14].

Statistics

Data are, where appropriate, expressed as mean +/- standard deviation. One-way ANOVA was used to assess differences in age, phase shift measurements and neuropsychological tests between AD patients, subjects with SCI and HC. Post hoc independent-samples t-tests were used to assess differences in age, phase measurements, and different neuropsychological test results between groups (two-sample comparison of means, two-sided test, equal variances were assumed). To evaluate differences in gender and education, chi-square tests were performed. Univariate general linear modeling analysis was used to assess the association between diagnosis and cortical phase shifts, adjusted for age and gender. To determine the association between the different neuropsychological tests and phase shift in the cortex, linear regression analysis was used, correcting for age and gender in SCI subjects. All statistical analyses were performed with the Statistical Package of Social Sciences (SPSS, version 20.0; SPSS, Chicago, Ill)

Results

The characteristics of the participants, including the mean scores for different neuropsychological tests per group, are shown in table 1. No differences in age, gender and education were found between groups. A small difference in performance on the digit span backwards test ($p = 0.024$) and TMT A ($p = 0.042$) between subjects with SCI and control subjects was found. For the other tests, no differences between these groups were found. AD subjects demonstrated an overall lower cognitive performance than subjects with SCI and HC.

Figure 1 shows representative magnitude and phase images from an AD patient, a subject with SCI and a HC subject. The mean phase shift of the entire cortex in AD patients (mean; 1.19, range: 1.00-1.35) was higher compared to both subjects with SCI (mean; 0.85, range: 0.73-0.99, adjusted for age and gender: $p<0.001$) and healthy control subjects (mean; 0.94, range: 0.79-1.10, adjusted for age and gender: $p<0.001$). No AD-like changes, e.g. increased cortical phase shifts, were found in subjects with SCI compared with healthy controls. Parcellation into frontal, parietal or temporoparietal subregions did not yield different conclusions.

Table 2 shows the results of the linear regression analysis examining the association between cortical phase shift of the whole brain (including all ROIs) and the scores of neuropsychological testing corrected for age and gender in SCI subjects. A significant association was found between memory function (WMS) and cortical phase shift ($\beta = -0.544$, $p = 0.007$). No significant association was found between other cognitive functions and cortical phase shift. Parcellation into frontal, parietal and temporoparietal subregions showed a highly significant association between cortical phase shift in the frontal region in SCI subjects and memory function (WMS) ($\beta = -0.721$, $p < 0.0001$) and a less significant association with global cognitive functioning (MMSE) ($\beta = -0.519$, $p = 0.021$). Within the parietal region a less significant association was also found with memory function (WMS) ($\beta = -0.406$, $p = 0.041$). Note that these results were not corrected for multiple comparisons and therefore should be interpreted with care, especially those with moderate significance (i.e. 0.05 - 0.01).

Table 1: Mean baseline characteristics and mean scores with standard deviation (SD) and range of different neuropsychological tests of healthy control subjects, subjects with subjective cognitive impairment and Alzheimer's disease patients. In addition, significant p-values between the groups are shown.

	HC (SD; range) (n=27)	SCI (SD; range) (n=18)	AD (SD; range) (n=28)
Age, years	68.9 (8.1; 52-80)	66.5 (11.2; 49-85)	71.2 (8.4; 54-86)
Male ; female	16 ; 11	13 ; 5	18 ; 10
Education (median)	3.3 (3)	3.5 (3)	4.0 (4)
GDS	-	3.3 (2.6; 0-7)	3.3 (2.6; 0-9)
MMSE score	29.0 (1.1; 26-30)	28.3 (1.6; 25-30)	22.4 (2.0; 18-26)**††
CAMCOG	-	92.7 (5.0; 86-100)	74.5 (8.4; 50-87)††
WMS	-	121.12 (15.2; 97-143)	91.1 (9.7; 74-103)††
Digit span forward	5.8 (0.9; 5-7)	4.8 (0.8; 4-6)	5.2 (1.3; 3-8)
Digit span backward	5.2 (1.0; 3-7)	3.8 (1.1; 2-5)*	3.6 (0.9; 2-5)**
TMT part A	35.0 (6.3; 28-46)	44.5 (15.9; 25-77)*	85.3 (80.4; 32-300)*†
TMT part B	79.3 (25.9; 51-131)	100.5 (53.0; 44-208)	226.7 (90.4; 79-300)**††
STROOP card 3	107.9 (31.6; 72-160)	132.3 (60.4; 83-300)	182.9 (70.2; 100-300)*

* = p<0.05 and ** = p<0.001: HC vs SCI or AD
† = p<0.05 and ††= p<0.001: SCI vs AD
Abbreviations: HC = healthy control, SCI = subjective cognitive impairment, AD = Alzheimer's disease, SD = standard deviation, GDS = Geriatric Depression Scale, MMSE = Mini Mental State Examination, CAMCOG = Cambridge Cognitive Examination, WMS = Wechsler Memory Scale, TMT = Trail Making Test. TMT part A and B and STROOP card 3 are given in seconds, other neuropsychological test are given in points.

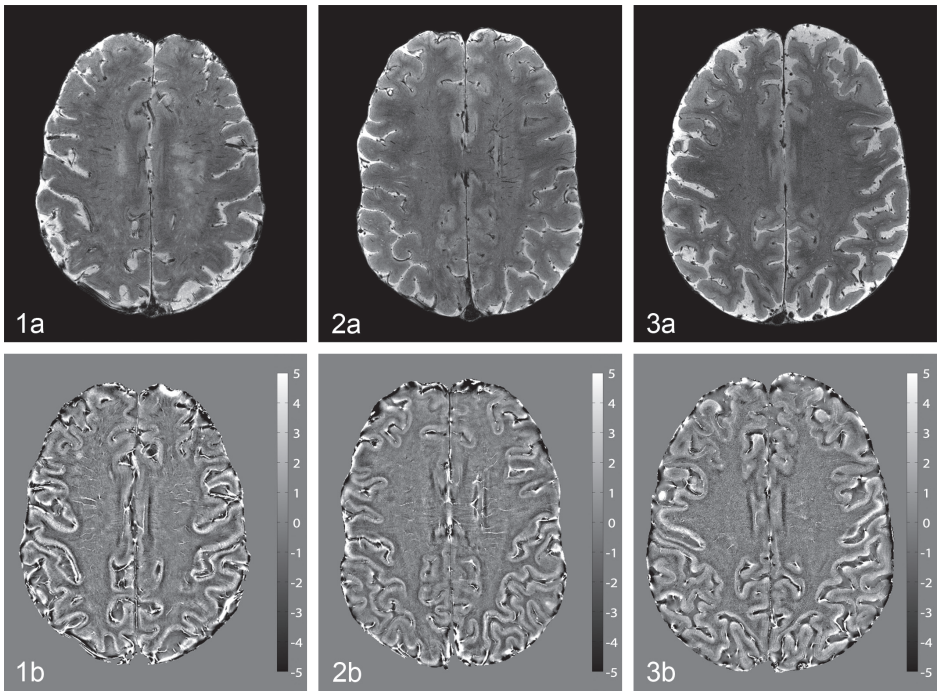

Figure 1: Representative magnitude (a) and phase images (including a scale bar in Hz) (b) acquired from an Alzheimer's disease patient (1), subject with subjective cognitive impairment (2) and a healthy control subject (3).

Table 2: Beta (β) and p-values correlating cortical phase shift with the scores of neuropsychological testing in subjects with subjective cognitive impairment adjusted for age and gender.

Cognitive domain	Cognitive test				
		β	Std. error B	Partial r	p-value
Depression	GDS	-0.007	0.014	-0.009	0.991
Global cognitive functioning	MMSE	-0.238	0.011	-0.278	0.297
Global cognitive functioning	CAMCOG	-0.251	0.004	-0.300	0.320
Memory	WMS	-0.544	0.001	-0.667	0.007*
Psychomotor speed	TMT part A	0.616	0.002	0.445	0.096
Executive function (cognitive flexibility)	TMT part B	0.250	0.001	0.171	0.541
Executive function (inhibition)	STROOP card 3	-0.060	0.000	-0.068	0.863

Abbreviations: GDS = Geriatric Depression Scale, MMSE = Mini Mental State Examination, CAMCOG = Cambridge Cognitive Examination, WMS = Wechsler Memory Scale, TMT = Trail Making Test.

Discussion

The major finding of the study is that in SCI subjects an increased cortical phase shift measured with high field MRI is associated with poorer memory performance, although as a group SCI subjects did not show an increased phase shift compared to HC. The finding of increased cortical phase shift being related to memory performance is important new information, as it is still unclear whether SCI represents a first stage of AD or a sign of pre-clinical AD, and results from previous studies are inconclusive regarding rate of decline and risk for conversion to AD dementia [1]. Previous longitudinal and epidemiological studies have suggested that SCI is a precursor of AD [3,37,38] and therefore we hypothesized that we might find an increased phase shift, related to AD pathology, in SCI subjects compared to controls, but that this phase shift would be lower compared to AD patients. However, in fact we found no increased cortical phase shift in SCI subjects compared to control subjects. Our findings might be explained by studies demonstrating that the majority of SCI subjects do not develop AD as there is only a 20% chance that SCI will progress to dementia in this group [39] and multiple factors contribute to the presence of cognitive complaints, such as depression, anxiety and quality of life [11-13]. Moreover, SCI is a heterogeneous group in which some pre-AD subjects might show an increase in phase, while non-AD cognitive impairment is associated with phase decrease, suggesting perhaps a different mechanism for this group.

Our study shows that cortical phase shift, determined on T_2^*-weighed MRI at 7T, is associated with memory performance in SCI subjects. In contrast, global cognition, executive function as well as psychomotor speed are not associated with cortical phase shift in these subjects. Based on previous studies we expected to find an association between the iron sensitive phase shift and cognition [40]. However, assuming that cortical phase shift is an indirect marker of the amount of amyloid plaques, the association would be more complicated. Previous studies have shown that this association is not straightforward and most likely influenced by the presence of neurofibrillary tangles (NFT) as discussed in a review by Nelson and coworkers [22]. The dynamic biomarkers model of AD states that AD starts with an increase of Aβ in the brain, followed by formation of NFT, and at a later stage memory is impaired when the amount of Aβ is still increasing [41]. Since we found an association between cortical phase shift and memory, and no other cognitive domain, this implies that in SCI subjects cortical phase shift might be a marker to demonstrate very early changes in the brain related to memory function.

Our data demonstrate that subjects with SCI do not show AD-like phase changes in comparison to HC. Although the SCI group performs slightly lower on the cognitive tests than the HC group, our results show also no highly significant differences in cognitive function between the groups. Several explanations for these findings are possible and are partly described above. If SCI is, in fact, an early AD stage in which AD pathology is present without the presence of cognitive symptoms, as is suggested by several studies, then our data indicate that phase changes would only appear in a later stage of the disease when AD pathology is more severe and starts to have an effect on brain function. Another explanation for our results might be that SCI is not strongly associated with AD, since it is related to many factors other than AD, and only a smaller subset of the SCI subjects develop AD [10-12,42,43]. Alternatively, because phase changes are measured at a group level, it might be that as most SCI subjects do not develop AD, the differences between controls and SCI subjects were too small to show significant differences. Investigating cortical phase changes in longitudinal studies in SCI, MCI and AD subjects to evaluate the possibility of detecting early AD pathology over time could give more insight into SCI and the development of AD pathology and the association with cortical phase changes and cognitive performance.

One overall limitation of the study is that phase measurements may partly be influenced by the geometry and orientation of the individual structures [21]. To limit such effects as much as possible, all participants were positioned in the same manner, and for every subject, the phase measurements were performed in the same way and were averaged to cancel out the possible effects of geometry and orientation. Although it is most likely that the difference in phase shift between groups is mainly attributable to iron, other compounds may contribute to a change in phase shift, such as deoxy-hemoglobin, myelin and proteins [36,44].

In conclusion, in this study we show that AD patients demonstrate an increased cortical phase shift indicating greater iron accumulation compared to HC. Moreover, these cortical phase shifts are associated with decreased memory performance in SCI subjects. Although previous studies have suggested that in SCI, AD-like changes may occur before cognitive decline becomes manifest, our data do not show any signs of AD-like high field T_2^*-weighted cortical phase shifts at a group level in subjects with SCI.

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References

1. Jessen F, Amariglio RE, van BM et al. A conceptual framework for research on subjective cognitive decline in preclinical Alzheimer's disease. Alzheimers Dement. 2014.
2. Dik MG, Jonker C, Comijs HC et al. Memory complaints and APOE-epsilon4 accelerate cognitive decline in cognitively normal elderly. Neurology. 2001;57:2217-2222.
3. Jessen F, Wiese B, Bachmann C et al. Prediction of dementia by subjective memory impairment: effects of severity and temporal association with cognitive impairment. Arch Gen Psychiatry. 2010;67:414-422.
4. van der Flier WM, van Buchem MA, Weverling-Rijnsburger AW et al. Memory complaints in patients with normal cognition are associated with smaller hippocampal volumes. J Neurol. 2004;251:671-675.
5. Striepens N, Scheef L, Wind A et al. Volume loss of the medial temporal lobe structures in subjective memory impairment. Dement Geriatr Cogn Disord. 2010;29:75-81.
6. Saykin AJ, Wishart HA, Rabin LA et al. Older adults with cognitive complaints show brain atrophy similar to that of amnestic MCI. Neurology. 2006;67:834-842.
7. Stewart R, Godin O, Crivello F et al. Longitudinal neuroimaging correlates of subjective memory impairment: 4-year prospective community study. Br J Psychiatry. 2011;198:199-205.
8. Jessen F, Feyen L, Freymann K et al. Volume reduction of the entorhinal cortex in subjective memory impairment. Neurobiol Aging. 2006;27:1751-1756.
9. Stewart R, Dufouil C, Godin O et al. Neuroimaging correlates of subjective memory deficits in a community population. Neurology. 2008;70:1601-1607.
10. Eramudugolla R, Cherbuin N, Easteal S et al. Self-reported cognitive decline on the informant questionnaire on cognitive decline in the elderly is associated with dementia, instrumental activities of daily living and depression but not longitudinal cognitive change. Dement Geriatr Cogn Disord. 2012;34:282-291.
11. Montejo P, Montenegro M, Fernandez MA et al. Subjective memory complaints in the elderly: Prevalence and influence of temporal orientation, depression and quality of life in a population-based study in the city of Madrid. Aging Ment Health. 2011;15:85-96.
12. Jorm AF, Christensen H, Korten AE et al. Memory complaints as a precursor of memory impairment in older people: a longitudinal analysis over 7-8 years. Psychol Med. 2001;31:441-449.
13. Knopman DS. Subjective cognitive impairment: fickle but fateful. Neurology. 2012;79:1308-1309.
14. van Rooden S, Versluis MJ, Liem MK et al. Cortical phase changes in Alzheimer's disease at 7T MRI: A novel imaging marker. Alzheimers Dement. 2013.

15. Nakada T, Matsuzawa H, Igarashi H et al. In vivo visualization of senile-plaque-like pathology in Alzheimer's disease patients by MR microscopy on a 7T system. J Neuroimaging. 2008;18:125-129.
16. Duce JA, Tsatsanis A, Cater MA et al. Iron-Export Ferroxidase Activity of beta-Amyloid Precursor Protein Is Inhibited by Zinc in Alzheimer's Disease. Cell. 2010;142:857-867.
17. Lei P, Ayton S, Finkelstein DI et al. Tau deficiency induces parkinsonism with dementia by impairing APP-mediated iron export. Nat Med. 2012;18:291-295.
18. Smith MA, Harris PL, Sayre LM et al. Iron accumulation in Alzheimer disease is a source of redox-generated free radicals. Proc Natl Acad Sci U S A. 1997;94:9866-9868.
19. van Rooden S, Maat-Schieman ML, Nabuurs RJ et al. Cerebral amyloidosis: postmortem detection with human 7.0-T MR imaging system. Radiology. 2009;253:788-796.
20. Duyn JH. The future of ultra-high field MRI and fMRI for study of the human brain. Neuroimage. 2011.
21. Yao B, Li TQ, van Gelderen P et al. Susceptibility contrast in high field MRI of human brain as a function of tissue iron content. Neuroimage. 2009;44:1259-1266.
22. Nelson PT, Braak H, Markesbery WR. Neuropathology and cognitive impairment in Alzheimer disease: a complex but coherent relationship. J Neuropathol Exp Neurol. 2009;68:1-14.
23. Jorm AF, Masaki KH, Davis DG et al. Memory complaints in nondemented men predict future pathologic diagnosis of Alzheimer disease. Neurology. 2004;63:1960-1961.
24. Barnes LL, Schneider JA, Boyle PA et al. Memory complaints are related to Alzheimer disease pathology in older persons. Neurology. 2006;67:1581-1585.
25. Lynoe N, Sandlund M, Dahlqvist G et al. Informed consent: study of quality of information given to participants in a clinical trial. BMJ. 1991;303:610-613.
26. 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;12:189-198.
27. Derix MM, Hofstede AB, Teunisse S et al. [CAMDEX-N: the Dutch version of the Cambridge Examination for Mental Disorders of the Elderly with automatic data processing]. Tijdschr Gerontol Geriatr. 1991;22:143-150.
28. Wechsler DA. A standardized memory scale for clinical use. Journal of Psychology. 1945;19:87-95.
29. Reitan RM. Validity of the Trail Making Test as an indication of organic brain damage. Percept Mot Skills. 1958;8:271-276.
30. Stroop JR. Studies of interference in serial verbal reactions. Journal of Experimental Psychology. 1935;18:643-662.
31. Sheikh JA YJ. *Clinical Gerontology:A Guide to Assessment and Intervention.* New York: Howarth Press; 1986.

32. McKhann G, Drachman D, Folstein M et al. 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;34:939-944.
33. Schoenmaker M, de Craen AJ, de Meijer PH et al. Evidence of genetic enrichment for exceptional survival using a family approach: the Leiden Longevity Study. Eur J Hum Genet. 2006;14:79-84.
34. Schar M, Kozerke S, Fischer SE et al. Cardiac SSFP imaging at 3 Tesla. Magn Reson Med. 2004;51:799-806.
35. Haacke EM, Xu Y, Cheng YC et al. Susceptibility weighted imaging (SWI). Magn Reson Med. 2004;52:612-618.
36. Duyn JH, van Gelderen P., Li TQ et al. High-field MRI of brain cortical substructure based on signal phase. Proc Natl Acad Sci U S A. 2007;104:11796-11801.
37. Jonker C, Geerlings MI, Schmand B. Are memory complaints predictive for dementia? A review of clinical and population-based studies. Int J Geriatr Psychiatry. 2000;15:983-991.
38. Reisberg B, Shulman MB, Torossian C et al. Outcome over seven years of healthy adults with and without subjective cognitive impairment. Alzheimers Dement. 2010;6:11-24.
39. Mitchell AJ. The clinical significance of subjective memory complaints in the diagnosis of mild cognitive impairment and dementia: a meta-analysis. Int J Geriatr Psychiatry. 2008;23:1191-1202.
40. Schroder N, Figueiredo LS, de Lima MN. Role of brain iron accumulation in cognitive dysfunction: evidence from animal models and human studies. J Alzheimers Dis. 2013;34:797-812.
41. Jack CR, Jr., Knopman DS, Jagust WJ et al. Hypothetical model of dynamic biomarkers of the Alzheimer's pathological cascade. Lancet Neurol. 2010;9:119-128.
42. Mitchell AJ. Is it time to separate subjective cognitive complaints from the diagnosis of mild cognitive impairment? Age Ageing. 2008;37:497-499.
43. Knopman DS. Subjective cognitive impairment: fickle but fateful. Neurology. 2012;79:1308-1309.
44. Duyn JH. Study of brain anatomy with high-field MRI: recent progress. Magn Reson Imaging. 2010;28:1210-1215.