HHS Public Access

Author manuscript

Neurobiol Aging. Author manuscript; available in PMC 2016 April 01.

Published in final edited form as:

Neurobiol Aging. 2015 April ; 36(4): 1765.e7–1765.e16. doi:10.1016/j.neurobiolaging.2014.12.028.

Association of Alzheimer disease GWAS loci with MRI-markers of brain aging

Ganesh Chauhan[a,*], **Hieab H.H. Adams**[b,c,*], **Joshua C Bis**[d], **Galit Weinstein**[e,f], **Lei Yu**[g,h], **Anna Maria Töglhofer**[i], **Albert Vernon Smith**[j,k], **Sven van der Lee**[b], **Rebecca F Gottesman**[l,m], **Russell Thomson**[n], **Jing Wang**[f,o], **Qiong Yang**[f,o], **Wiro J. Niessen**[c], **Oscar L Lopez**[p,q], **James T Becker**[p,q,r], **Thanh G Phan**[s], **Richard J Beare**[s,t], **Konstantinos Arfanakis**[g,u], **Debra Fleischman**[g], **Meike W. Vernooij**[b,c], **Bernard Mazoyer**[v], **Helena Schmidt**[i], **Velandai Srikanth**[s,n], **Dave S Knopman**[w], **Clifford R Jack Jr**[x], **Philippe Amouyel**[y,z,aa], **Albert Hofman**[b], **Charlie DeCarli**[ab], **Christophe Tzourio**[ac,ad], **Cornelia M van Duijn**[b,ae,af], **David A Bennett**[g,h], **Reinhold Schmidt**[ag], **William T Longstreth Jr**[ah], **Thomas H Mosley**[ai], **Myriam Fornage**[aj], **Lenore J Launer**[ak], **Sudha Seshadri**[e,f,§], **M Arfan Ikram**[b,c,ae,§], and **Stephanie Debette**[a,e,al,am,§]

[a]INSERM U740 (Paris 7 University) and U708 (Bordeaux University), France [b]Department of Epidemiology, Erasmus Medical Center, Rotterdam, Netherlands [c]Department of Radiology, Erasmus Medical Center, Rotterdam, Netherlands [d]Cardiovascular Health Research Unit, Department of Medicine, University of Washington, Seattle, Washington, USA [e]Department of Neurology, Boston University School of Medicine, Boston, Massachusetts, USA [f]The Framingham Heart Study, Boston, Massachusetts, USA [g]Rush Alzheimer's Disease Center, Rush University Medical Center, Chicago, IL, USA [h]Department of Neurological Sciences, Rush University Medical Center, Chicago, IL, USA [i]Institute of Molecular Biology and Biochemistry, Centre for Molecular Medicine, Medical University of Graz, Austria [j]Icelandic Heart Association, Kopavogur Capital Region, Iceland [k]Department of Medicine, University of Iceland, Reykjavik, Iceland [l]Department of Neurology, Johns Hopkins School of Medicine, Baltimore, USA [m]Department of Epidemiology, Johns Hopkins Bloomberg School of Public Health, Baltimore, USA [n]Menzies Research Institute Tasmania, University of Tasmania, Hobart, Tasmania, Australia [o]Department of Biostatistics, Boston University School of Public Health, Boston, Massachusetts, USA [p]Department of Neurology, University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, USA [q]Department of Psychiatry, University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, USA [r]Department of Psychology, University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, USA [s]Stroke and Ageing Research Centre, Southern Clinical School, Department of Medicine, Monash University, Melbourne, Victoria, Australia

Corresponding Author: Dr. Stéphanie Debette, MD, PhD, INSERM U740; 10 Avenue de Verdun; 75010 PARIS, Tel: +33 (0)6 84 07 72 53; Fax: +33 (0)1 57 27 85 94, stephanie.debette@lrb.aphp.fr; sdebette@bu.edu.

*These authors contributed equally to the manuscript

§These authors jointly directed this work

[t]Developmental Imaging Group, Murdoch Childrens Research Institute, The Royal Children's Hospital, Parkville, Victoria, Australia [u]Department of Biomedical Engineering, Illinois Institute of Technology, Chicago, IL, USA [v]CNRS-CEA UMR5296, Université Bordeaux Segalen, Bordeaux, France [w]Department of Neurology, Mayo Clinic College of Medicine, Rochester, MN, USA [x]Department of Radiology, Mayo Clinic College of Medicine, Rochester, MN, USA [y]Department of Epidemiology and Public Health, Pasteur Institute of Lille, Lille, France [z]INSERM, U744, Lille, France [aa]Université Lille 2, Lille, France [ab]Department of Neurology, University of California at Davis, Davis, California, USA [ac]Department of Neuroepidemiology, INSERM U708, Bordeaux, France [ad]Université Victor Segalen Bordeaux 2, France [ae]Netherlands Consortium for Healthy Aging, Leiden, Netherlands [af]Center for Medical Systems Biology, Leiden, Netherlands [ag]Department of Neurology, Clinical Division of Neurogeriatrics, Medical University of Graz, Austria [ah]Departments of Neurology and Epidemiology, University of Washington, Seattle, Washington, USA [ai]Department of Medicine-Geriatrics/Gerontology, University of Mississippi Medical Center, Jackson, Mississippi, USA [aj]The Human Genetics Center and Institute of Molecular Medicine, The University of Texas Health Science Center at Houston, Houston, USA [ak]Laboratory of Epidemiology and Population Sciences, Intramural Research Program, National Institute on Aging, National Institutes of Health, Bethesda, Maryland, USA [al]Department of Neurology, Lariboisière Hospital, Paris 7 University, DHU Neurovasc Paris Sorbonne, Paris, France [am]University of Versailles Saint-Quentin-en-Yvelines, France

Abstract

Whether novel risk variants of Alzheimer's disease (AD) identified through genome-wide association studies (GWAS) also influence MRI-based intermediate phenotypes of AD in the general population is unclear. We studied association of 24 AD risk loci with intracranial volume (ICV), total brain volume (TBV), hippocampal volume (HV), white matter hyperintensity (WMH) burden, and brain infarcts in a meta-analysis of genetic association studies from large population-based samples (N=8,175–11,550). In single-SNP based tests, AD risk allele of *APOE* (rs2075650) was associated with smaller HV (p=0.0054) and *CD33* (rs3865444) with smaller ICV (p=0.0058) In gene-based tests, there was associations of *HLA-DRB1* with TBV (p=0.0006) and *BIN1* with HV (p=0.00089). A weighted AD genetic risk score was associated with smaller HV (beta±SE= −0.047±0.013, p=0.00041), even after excluding the *APOE* locus (p=0.029). However, only association of AD genetic risk score with HV, including *APOE*, was significant after multiple testing correction (including number of independent phenotypes tested). These results suggest that novel AD genetic risk variants may contribute to structural brain aging in non-demented older community persons.

Keywords

Alzheimer; MRI-markers; genetic risk score; GWAS; hippocampal volume

1. INTRODUCTION

Alzheimer's disease (AD) is the leading cause of dementia and represents a major public health burden (Ballard, et al., 2011). Converging evidence suggests that pathological

processes leading to this progressive neurodegenerative disorder start many years before clinical diagnosis of dementia (Sperling, et al., 2011). MRI-markers of brain aging, including total brain volume (TBV) and hippocampal volume (HV), and markers of vascular brain injury, including white matter hyperintensities (WMH) and brain infarcts, are powerful predictors of dementia and may, at least in part, represent intermediate markers reflecting pathological processes leading to AD (Debette and Markus, 2010, Jack, et al., 2013, Jack, et al., 2010, Kaye, et al., 1997, Sperling, et al., 2011, Vermeer, et al., 2007). Intracranial volume (ICV), an imaging marker reflecting brain growth during development and maturation, was suggested to be correlated with resilience to brain damage (Negash, et al., 2013).

Recently, large scale genome-wide association studies (GWAS) and candidate gene based studies have identified novel susceptibility loci for late-onset AD (Boada, et al., 2013, Carrasquillo, et al., 2009, Harold, et al., 2009, Hollingworth, et al., 2011, Jonsson, et al., 2012, Jonsson, et al., 2013, Lambert, et al., 2009, Lambert, et al., 2013, Naj, et al., 2011, Seshadri, et al., 2010). These AD risk variants have recently been used to examine the genotypic overlap between AD and other types of dementia (Carrasquillo, et al., 2014). Some of these variants have been studied with respect to various MRI measures in a mixed study sample of AD patients, mildly cognitive impaired and healthy controls (Biffi, et al., 2010, Furney, et al., 2011). They could also be implemented to explore the impact of genetic determinants of AD on MRI-markers of structural brain changes in non-demented community persons. Indeed, this could provide important information on the disease mechanisms through which these genes affect the risk of AD, and could be of interested for the design of preventative interventions. Whether all previously and newly discovered AD risk loci influence brain structure in advance of clinically detectable dementia has never been systematically investigated in large community samples to our knowledge. Our aim was to study association of known AD GWAS loci with ICV, TBV, HV, WMH burden and brain infarcts in non-demented participants from 10 population-based studies.

2. MATERIALS and METHODS

2.1. Population

Analyses were performed on 8,175 to 11,550 dementia free participants of European ancestry with quantitative brain MRI and genome-wide genotypes (N=8,175 for ICV, N=8,673 for TBV, N=11,550 for HV, N=9,361 for WMH burden and N=9,401 for brain infarcts), from up to 10 population-based cohort studies participating in the Cohorts of Heart and Aging Research in Genomic Epidemiology (CHARGE) consortium: Aging Gene-Environment Susceptibility (AGES)–Reykjavik Study, Atherosclerosis Risk in Communities Study (ARIC), Austrian Stroke Prevention Study (ASPS), Cardiovascular Health Study (CHS), Framingham Heart Study (FHS), Rotterdam Study (RS), Erasmus Rucphen Family (ERF) study, Religious Order Study (ROS) & Rush Memory and Aging Project (MAP), Tasmanian Study of Cognition and Gait (TASCOG) and the 3C-Dijon study. Each study secured approval from institutional review boards, and all participants provided written informed consent for study participation, brain MRI, and use of DNA for genetic research. Individual studies are described in the Supplementary Appendix.

2.2. MRI scans

In each study, MRI scans were performed and interpreted in a standardized fashion, without reference to clinical or genetic information. Details on MRI parameters and phenotype definition are provided in the Supplementary Appendix. Briefly, automated or semi-quantitative post-processing software was used to measure ICV and TBV. TBV was expressed as percentage of ICV to correct for differences in head size (Ikram, et al., 2012). HV was evaluated using operator-defined boundaries drawn on serial coronal sections or automated methods (Bis, et al., 2012). WMH burden was estimated on a quantitative scale using custom-written computer programs in AGES-Reykjavik, ASPS, FHS, and RS; in ARIC and CHS, WMH burden was estimated on a semi-quantitative scale (Fornage, et al., 2011). Brain infarcts were defined as areas of abnormal signal intensity in a vascular distribution that lacked mass effect, ≥ 3–4 mm, distinct from dilated perivascular spaces (Debette, et al., 2010).

2.3. AD GWAS loci

We manually scanned the GWAS catalog (http://www.genome.gov/gwastudies/) and Alzgene (www.alzgene.org/) for GWAS on AD. We only chose studies performed on European subjects, including a replication stage, examining single marker based associations and having loci reaching genome wide significance ($P<5.0\times10^{-8}$). This led to the identification of 24 independent loci. Effect estimates for SNPs with the lowest p-value in each locus (defined as the index SNP of the locus) are presented in Supplementary Table 1. We included the *CD33* locus (rs3865444) despite absence of replication in the latest AD GWAS meta-analysis;(Lambert, et al., 2013) this locus was previously replicated in several AD GWAS,(Hollingworth, et al., 2011, Naj, et al., 2011) and recent functional studies provide strong evidence for involvement of rs3865444 and *CD33* in AD pathology (Bradshaw, et al., 2013). For the *APOE*-ε polymorphism we used rs2075650 as a proxy ($r^2=0.48$ with rs429358, the *APOE*-ε SNP), because *APOE*-ε genotypes cannot be reliably imputed on commercial genome-wide chips. The AD risk variants near *HLA-DRB1*(Lambert, et al., 2013), *ATP5H*/*KCTD2* (Boada, et al., 2013), in *TREM2*,(Jonsson, et al., 2013), and *APP*(Jonsson, et al., 2012) were not included for single-SNP based association and genetic risk score based association as no index SNP or proxy ($r^2>0.3$) was available among the genome-wide genotypes for MRI-markers of brain aging.

2.4. Power calculation

Quanto software (Gauderman, 2002a, Gauderman, 2002b) was used to compute power of of the five MRI marker studies assuming additive model of inheritance at $\alpha=0.0025$ (Supplementary Figure 1). Power for the quantitative traits (ICV, TBV, HV, WMH burden) was computed for different percentage variance explained while for brain infarcts, a dichotomous trait, it was computed for different odds ratios at different allele frequencies.

2.5. Correlation between phenotypes and equivalent number of independent phenotypes

Correlation between the five MRI phenotypes in 3C-Dijon and FHS was calculated based on Pearson's correlation using the "rcorr" function in R. These correlations were used to compute the equivalent number of independent phenotypes using the online tool matSpDlite

(http://neurogenetics.qimrberghofer.edu.au/matSpDlite/). MatSpDlite which is based on the same principles used to identify number of independent SNPs in a locus, gives the equivalent number of independent variables in a correlation (r) matrix, depending upon the ratio of observed eigenvalue variance (after spectral decomposiiton) to its theoretical maximum (Nyholt, 2004).

2.6. Association Analyses

Three analytical approaches were taken to examine the associations of interest.

2.6.1. Single-SNP based association analysis—We tested for association of AD GWAS loci with MRI-markers of brain aging using association estimates obtained from meta-analyses of GWAS for ICV(Ikram, et al., 2012), TBV(Ikram, et al., 2012), HV(Bis, et al., 2012), WMH burden (Fornage, et al., 2011) and brain infarcts (Debette, et al., 2010) using genotypes imputed on the HapMap2 CEU reference panel. AD risk alleles, as described in the latest AD GWAS meta-analysis,(Lambert, et al., 2013) were modeled as the effect alleles for associations with MRI-markers of brain aging. Logistic (brain infarcts) or linear (ICV, TBV, HV and WMH burden) regression was performed within each study, adjusting for age, gender, and principal components of population stratification, and for familial relationships or study center if relevant. For WMH burden, data was log transformed to achieve normal distribution and associations were additionally adjusted for ICV (except for studies measuring WMH burden on a semi-quantitative visual scale, visual grades being inherently normalized for brain size)(Fornage, et al., 2011). For most phenotypes (ICV, TBV, HV, and brain infarcts) meta-analyses were performed using fixed effects inverse variance weighted meta-analysis. For WMH burden, meta-analysis was performed using effective sample size weighted meta-analysis, because WMH burden was measured on different scales across studies. If the lead SNP at a specific AD GWAS locus was not available, a proxy SNP (r^2>0.70 in 1000G CEU) of the lead SNP was used to check single-SNP based association results (Supplementary Table 1). After Bonferroni correction for testing 20 independent loci, $p<0.0025$ was considered significant for single-SNP based associations. However, application of a more stringent threshold additionally accounting for the number of independent phenotypes tested led to a Bonferroni correction of $p<0.000625$.

2.6.2. Gene-based association analysis—Gene-based association tests can be more powerful in comparison to single-SNP based association tests when there are many causal variants in a gene with small effects (Liu, et al., 2010). Single-SNP based association results from the respective MRI-marker GWAS meta-analysis were used to compute gene-based association results using the Versatile Gene-Based Association Study2 (VEGAS2) software (https://vegas2.qimrberghofer.edu.au/) (Liu, et al., 2010). The gene annotations and LD calculation in VEGAS2 are based on 1000 genomes (phase 1 version 3). This tool annotated all but one gene (*MS4A4E*) within 50KB of the index SNPs. The test incorporates information from all markers within a gene and accounts for linkage disequilibrium (LD) between markers by using simulations from the multivariate normal distribution. Gene-based association analyses were performed for all protein coding genes (N=65 genes) which lie within a 50kb distance of index SNP of the AD risk loci. Gene boundaries were defined as 50kb upstream and downstream of the start and end of gene (Liu, et al., 2010). The choice

of 50 KB boundary to cover a gene was chosen as a trade-off between a longer boundary which would have caused excess overlap between nearby genes and a shorter boundary which would have ignored potential regulatory regions (Liu, et al., 2010). Maximum permutation limits were set to 1000,000. After correcting for the number of genes (N=65) tested the multiple testing threshold was $p<0.00077$. A more stringent correction additionally accounting for number of independent phenotypes (N=4) tested, lead to a multiple testing threshold of $p< 0.00019$ for gene based association.

2.6.3. Construction of genetic risk score—We constructed a genetic risk score comprising all selected AD risk variants from 20 independent AD risk loci to estimate joint effect of these SNPs on MRI-markers of brain aging. Methods have been recently developed to apply a genetic risk score to meta-analysis summary estimates without requiring access to raw data from individual studies (Dastani, et al., 2012). For each MRI-marker of interest, the beta-coefficient for a given SNP, as obtained from the GWAS meta-analysis for this MRI-marker, was weighted with the published AD beta-coefficient for the given SNP. The weighted sum of beta-coefficients for all 20 SNPs (Formula-i(a)) was used as the beta-coefficient of the genetic risk score. Similarly, for each MRI-marker of interest, the inverse of the variance for a given SNP (from the GWAS meta-analysis for this MRI-marker) was weighted by the square of the published AD beta-coefficient for the given SNP. These weighted inverse of variances were then summed and the inverse of this sum was used as the variance of the genetic risk score (Formula-i(b)). The Wald statistic was used to test for significance of associations between the genetic risk score and each MRI-marker (Dastani, et al., 2012). For WMH burden, betas and standard errors were estimated from Z-statistics provided by the effective sample size weighted meta-analysis using Formula-ii. AD beta-coefficients used as weights for the score were all drawn from the discovery stage of the recent largest AD GWAS meta-analysis (17,008 AD cases and 37,154 controls, Supplementary Table 1) (Lambert, et al., 2013). Associations with $p<0.05$ were considered significant for genetic risk score based associations.

$$\beta_{\text{grs}}=\frac{\sum_1^m w\beta SE^{-2}}{\sum_1^m w^2\, SE^{-2}} \quad \text{Formula – i(a)}$$

$$SE^2{}_{grs}=\frac{1}{\sum_1^m w^2\, SE^{-2}} \quad \text{Formula – i(b)}$$

β_{grs}=beta of genetic risk score; SE_{grs}=SE of genetic risk score; *w*=weight applied (=SNP-specific beta of AD GWAS); β=SNP specific beta of association with MRI-phenotype; SE= SNP-specific SE of association with MRI-phenotype

$$SE\sim=\sqrt{VP/(ES\times 2pq)} \quad \text{Formula – ii(a)}$$

$$Beta=SE\times Z \quad \text{Formula – ii(b)}$$

VP=phenotypic variance (approximated to 1); ES=Effective sample size; p=Minor allele frequency; q=Major allele frequency.

After correcting for four independent phenotypes tested, the multiple testing threshold for genetic risk score association was P<0.0125.

3. RESULTS

3.1. Correlation and heritability of the five MRI traits

Based on data from two studies which were part of the original meta-analysis the two MRI markers of structural brain aging, ICV and TBV showed high correlation with each other but were only moderately correlated with HV (Supplementary Table 2). The two MRI markers of vascular brain aging WMH burden and brain infarcts showed low correlation with each other and very little or no correlation with the three markers of structural brain aging. Depending upon this correlation the equivalent number of independent phenotypes calculated using matSpDlite was four for both studies. Published literature showed that the five MRI markers had moderate to high heritability (Supplementary Table 3).

3.1. Single-SNP based associations

In total 9 out of 20 AD risk variants that could be analyzed showed association with at least one MRI-marker at p<0.05 (Table 1). With only 2 exceptions (*CD33* locus with brain infarcts (p=0.048) and *PTK2B* locus with ICV (p=0.028)), betas were in the expected direction i.e. the AD risk allele was associated with increased risk for brain infarcts and with lower ICV, TBV and HV. The most significant associations were for *APOE*-rs2075650 with HV (beta±SE=−0.042±0.015, p=0.0054) and *CD33*-rs3865444 with ICV (beta±SE= −5.209±1.886, p=0.0058) (Table 1). However, none of the single-SNP based associations were significant after correcting for multiple testing. None of the AD risk variants showed associations with WMH burden.

3.2. Gene-based associations

Out of the 24 loci investigated, 23 had at least one protein coding gene within 50kb distance. Only rs3851179 (11q14) had no protein coding gene within 50kb and was not represented in the gene-based association analysis (nearest genes: *PICALM* 87.72kb downstream and *EED* 86.95kb upstream). In total, 65 protein coding genes from 23 independent loci were assessed for gene-based association analyses (Supplementary Table 4).

A total of 27 protein coding genes within 50kb of 15 index SNPs were associated with ICV, TBV, HV or brain infarcts at p<0.05 (Table 2). For ICV we observed association with 13 genes within 50kb of five index SNPs (*MEF2C, NME8, PILRB, PILRA, ZCWPW1, MEPCE, PPP1R35, C7orf61, MS4A6A, PVRL2, TOMM40, APOE, APOC1*; p-range: 0.04–0.0078). Eight genes within 50kB of six index SNPs were associated with TBV (*CR1, HLA-DRB1, HLA-DQA1, HLA-DQB1, TAS2R60, SCARA3, ICT1, CD33*; p-range: 0.047–0.0006). *BIN1, TREML1* and *MS4A6A* were associated with HV (p=0.00089, 0.03 and 0.048, respectively) while *MEF2C, AURKA, CSTF1 and TAS2R60*showed association with brain infarcts (p-range: 0.049–0.033). For WMH burden we observed association with three genes from two

loci (*HLA-DQB1*, *HMHA1* and *ABCA7*; p=0.01, 0.046 and 0.049 respectively). If we correct for the number of genes tested the association of HLA-DRB1 with TBV remains significant but if we additionally correct for the number of phenotypes tested this association is not significant.

3.3. Genetic risk score based associations

The AD genetic risk score was associated with smaller HV (beta±SE=−0.047±0.013, p=0.00041) (Table 3). This association was also observed after removing the *APOE* locus from the AD genetic score (beta±SE=−0.050±0.023, p=0.029). There was also nominal association of the AD genetic risk score with smaller TBV (beta±SE =−0.127±0.064, P=0.046) but this association was not significant after excluding the *APOE* locus from the genetic risk score (P=0.13). Only association of the AD genetic risk score with HV including *APOE* locus was significant after correcting for the number of independent phenotypes tested.

4. DISCUSSION

We investigated associations of 24 genome-wide significant AD risk loci with five MRI-markers of brain structure and aging (ICV, TBV, HV, WMH burden and brain infarcts), in over 8,000 dementia free older community participants from the CHARGE consortium. Although no single SNP-based association met the significance threshold after correction for multiple testing, index AD risk variants mapping to eight of the 21 AD risk loci showed nominal association with at least one MRI-marker, the most interesting being association for *APOE* (rs2075650) with smaller HV and for *CD33* (rs3865444) with smaller ICV. In gene-based association analyses *HLA-DRB1* was significantly associated with TBV after correction for number of genes tested. A weighted AD genetic risk score was significantly associated with smaller HV.

In Single-SNP based associations none of the associations were significant after correcting for multiple testing. Nominally significant associations of an *APOE* risk variant with HV (P=0.0054) and a *CD33* variant with ICV (P=0.0058) were observed. Since the mid 1990's (Supplementary Table 5) some studies have described significant associations between the *APOE*-ε4 allele and smaller HV (den Heijer, et al., 2002, Lehtovirta, et al., 1995, Lehtovirta, et al., 1996, Lind, et al., 2006, Liu, et al., 2014, Lu, et al., 2011, Morra, et al., 2009, O'Dwyer, et al., 2012, Plassman, et al., 1997, Schuff, et al., 2009, Soininen, et al., 1995), however other studies did not find such an association (Ferencz, et al., 2013, Khan, et al., 2014, Reiman, et al., 1998, Schmidt, et al., 1996). Using the largest sample size to date (N=11,550), as previously reported by our group, our findings are supportive of an association of the *APOE*-ε4 locus with smaller HV (Bis, et al., 2012). The rs3865444 (*CD33*) AD risk allele association with smaller ICV could perhaps be suggestive of an involvement of this locus in brain maturation and brain reserve. Recent reports suggest that rs3865444 influences *CD33* expression, including in young adults in their twenties (Bradshaw, et al., 2013), and is associated with diminished internalization of amyloid β_{42} peptide, and accumulation of neuritic amyloid pathology and fibrillar amyloid in vivo (Bradshaw, et al., 2013).

Gene-based analyses revealed significant associations of *HLA-DRB1* (index SNP rs9271192) with TBV. The *HLA-DRB1* locus was recently identified to be associated with AD in the largest meta-analysis of AD (Lambert, et al., 2013). This locus is part of the major histocompatibility complex, class II, and our findings add support to the role of autoimmunity in AD. The findings also suggest that the locus may be playing a role in pre-symptomatic stages of the disease, as we observe association with smaller brain volumes in non-demented older community persons.

When combined in a weighted genetic risk score, AD risk variants were associated cumulatively with decreased HV. Interestingly the association was maintained with a similar effect size, although less significant, after removing the *APOE* locus from the analysis, suggesting that, in aggregate, novel AD risk loci are associated with smaller HV in non-demented older community persons. The AD genetic risk score also showed nominal association with smaller TBV. Although this association was no longer significant after removing the *APOE* locus, other loci were contributing to this association, as the *APOE* risk variant alone was not significantly associated with TBV.

There were fewer associations with WMH burden and brain infarcts. Most associations with AD risk variants were observed for ICV, TBV, and HV. This may indicate that, even though they are strong predictors of dementia risk,(Debette and Markus, 2010, Vermeer, et al., 2007) MRI-markers of vascular brain injury could have less shared genetic determinants with AD than MRI-markers of brain growth and brain atrophy, as suggested by others (Biffi, et al., 2010). Noteworthy, our study only tested for overlap of genome-wide significant AD risk variants, did not explore shared heritability and may have been underpowered for less common variants with smaller effect size (Supplementary Figure 1).

Our study has limitations. The 24 AD risk loci do not reflect the full spectrum of genetic susceptibility to AD and the index SNPs used may not be causal variants. The five GWAS of MRI-markers, although the largest of their kind, have fewer samples compared to the AD GWAS from which the loci have been obtained (Bis, et al., 2012, Debette, et al., 2010, Fornage, et al., 2011, Ikram, et al., 2012, Lambert, et al., 2013). These five GWAS of MRI-markers were performed using imputed genotypes based on the HapMap2 panel, which has fewer markers with limited LD information, does not cover rare variants and has lower imputation accuracy, especially for lower allele frequencies, compared to the more recent 1000 genomes reference panels. We therefore couldn’t analyze rare AD risk variants in the present study and we cannot exclude that the more limited LD information might have introduced some bias in the results of the gene-based analyses. In addition, despite major efforts to harmonize phenotype definitions across studies, there may be some residual heterogeneity in methods for quantifying MRI-markers of brain aging. These elements could have reduced our power to detect associations of AD GWAS loci with MRI-markers of brain aging. The choice of 50 KB window for a gene based test does not account for potential regulatory effects on more distant genes. Our findings cannot be generalized to populations of non-European ancestry. Ongoing, larger multi-ethnic GWAS of MRI-markers of brain aging, as well as sequencing projects searching for rare variants associated with AD risk and MRI phenotypes may enable us to expand our findings in the future.

5. Conclusion

In conclusion, we have shown that novel AD genetic risk variants are associated with MRI-markers of structural brain aging in older, non-demented community persons. In aggregate, novel AD genetic risk variants were associated with smaller brain volumes, especially HV. Significant gene-based associations and suggestive single SNP-based associations with ICV, TBV and HV also provide interesting hypotheses for mechanisms underlying genetic associations with AD

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

The authors thank the staff and participants of all participating study for their important contributions.

Aging Gene-Environment Susceptibility-Reykjavik Study: The research has been funded by NIA contract N01-AG-12100 with contributions from NEI, NIDCD and NHLBI, the NIA Intramural Research Program, Hjartavernd (the Icelandic Heart Association), and the Althingi (the Icelandic Parliament).

The Atherosclerosis Risk in Communities Study: The research is carried out as a collaborative study supported by National Heart, Lung, and Blood Institute contracts (HHSN268201100005C, HHSN268201100006C, HHSN268201100007C, HHSN268201100008C, HHSN268201100009C, HHSN268201100010C, HHSN268201100011C, and HHSN268201100012C), R01HL087641, R01HL59367 and R01HL086694 and R01HL7825; National Human Genome Research Institute contract U01HG004402; and National Institutes of Health contract HHSN268200625226C. The authors thank the staff and participants of the ARIC study for their important contributions. Infrastructure was partly supported by Grant Number UL1RR025005, a component of the National Institutes of Health and NIH Roadmap for Medical Research. Funds for this projects were also supported by grant HL093029 to MF.

Cardiovascular Health Study: This CHS research was supported by NHLBI contracts HHSN268201200036C, HHSN268200800007C, N01HC55222, N01HC85079, N01HC85080, N01HC85081, N01HC85082, N01HC85083, N01HC85086, N01HC15103; and NHLBI grants HL080295, HL087652, HL105756 with additional contribution from the National Institute of Neurological Disorders and Stroke (NINDS). Additional support was provided through R01AG20098, R01AG15928, and R01AG023629 from the National Institute on Aging (NIA). A full list of CHS investigators and institutions can be found athttp://chs-nhlbi.org/.

The provision of genotyping data was supported in part by the National Center for Advancing Translational Sciences, CTSI grant UL1TR000124, and the National Institute of Diabetes and Digestive and Kidney Disease Diabetes Research Center (DRC) grant DK063491 to the Southern California Diabetes Endocrinology Research Center.

The Austrian Stroke Prevention Study: The research reported in this article was funded by the Austrian Science Fond (FWF) grant number P20545-P05 and P13180. The Medical University of Graz supports the databank of the ASPS. The authors thank the staff and the participants of the ASPS for their valuable contributions. We thank Birgit Reinhart for her long-term administrative commitment and Ing Johann Semmler for the technical assistance at creating the DNA-bank.

Erasmus Rucphen Family Study: This study is financially supported by the Netherlands Organization for Scientific Research (NWO), the Internationale Stichting Alzheimer Onderzoek (ISAO), the Hersenstichting Nederland (HSN), and the Centre for Medical Systems Biology (CMSB) in the framework of the Netherlands Genomics Initiative (NGI). We thank the participants from the Genetic Research in Isolated Populations, Erasmus Rucphen Family, who made this work possible.

Framingham Heart Study: This work was supported by the Framingham Heart Study's National Heart, Lung, and Blood Institute contract (N01-HC-25195) and its contract with Affymetrix, Inc for genotyping services (Contract No. N02-HL-6-4278). A portion of this research utilized the Linux Cluster for Genetic Analysis (LinGA-II) funded by the Robert Dawson Evans Endowment of the Department of Medicine at Boston University School of Medicine

and Boston Medical Center. It was also funded by grants from the National Institute on Aging (R01 AG08122, AG033193) and the National Institute of Neurological Disorders and Stroke (R01 NS17950).

The Religious Order Study (ROS) and Rush Memory and Aging Project (R-MAP): The R-MAP and ROS data used in this article was supported by National Institute on Aging grants P30AG10161, R01AG17917, and R01AG15819, and the Illinois Department of Public Health.

The Rotterdam Study: The GWA database of the Rotterdam Study was funded through the Netherlands Organization of Scientific Research NWO (nr. 175.010.2005.011). This study was further supported by the Netherlands Genomics Initiative (NGI)/Netherlands Organisation for Scientific Research (NWO) project nr. 050-060-810. The Rotterdam Study is supported by the Erasmus Medical Center and Erasmus University, Rotterdam; the Netherlands organization for scientific research (NWO), the Netherlands Organization for the Health Research and Development (ZonMw; Veni-grant 916.13.054), the Research Institute for Diseases in the Elderly (RIDE), the Ministry of Education, Culture and Science, the Ministry for Health, Welfare and Sports, the European Commission (DG XII), the Municipality of Rotterdam, and the Internationale Stichting Alzheimer Onderzoek.

The Tasmanian Study of Gait and Cognition (TASCOG) is supported by Project Grants from the National Health and Medical Research Council (NHMRC IDs 403000, 491109, 606543), and a grant from the Wicking Dementia Education and Research Centre, Hobart. Velandai Srikanth is supported by an NHMRC/National Heart Foundation Career Development Fellowship (ID 606544).

Three City Study (3C): We thank the staff and the participants of the 3C Study for their important contributions. The 3C Study is conducted under a partnership agreement between the Institut National de la Santé et de la Recherche Médicale (INSERM), the Victor Segalen–Bordeaux II University, and Sanofi-Aventis. The Fondation pour la Recherche Médicale funded the preparation and initiation of the study. The 3C Study is also supported by the Caisse Nationale Maladie des Travailleurs Salariés, Direction Générale de la Santé, Mutuelle Générale de l'Education Nationale (MGEN), Institut de la Longévité, Conseils Régionaux of Aquitaine and Bourgogne, Fondation de France, and Ministry of Research–INSERM Programme "Cohortes et collections de données biologiques." Lille Génopôle received an unconditional grant from Eisai. We thank A. Boland (Centre National de Génotypage) for her technical help in preparing the DNA samples for analyses. This work was supported by the National Foundation for Alzheimer's Disease and Related Disorders, the Institut Pasteur de Lille and the Centre National de Génotypage. Ganesh Chauhan and Stéphanie Debette are supported by a grant from the Fondation Leducq and the Agence Nationale de la Recherche (Chaire d'Excellence).

We also thank Josée Dupuis (Department of Biostatistics, Boston University School of Public Health, Boston, Massachusetts, USA) and Toby Johnson (Department of Medical Genetics, University of Lausanne, Lausanne, Switzerland) for their statistical advice.

Abbreviations

AD	Alzheimer's disease
AGES	Aging Gene-Environment Susceptibility
ARIC	Atherosclerosis Risk in Communities Study
ASPS	Austrian Stroke Prevention Study
CHARGE	Cohorts of Heart and Aging Research in Genomic Epidemiology
CHS	Cardiovascular Health Study
ERF	Erasmus Rucphen Family
FHS	Framingham Heart Study
GWAS	Genome-wide association studies
HV	Hippocampal volume
ICV	Intra-cranial volume
LD	linkage disequilibrium

MAP	Rush Memory and Aging Project
ROS	Religious Order Study
RS	Rotterdam Study
TASCOG	Tasmanian Study of Cognition and Gait
TBV	Total brain volume
VEGAS	Versatile Gene-Based Association Study
WMH	White matter hyperintensity

References

Ballard C, Gauthier S, Corbett A, Brayne C, Aarsland D, Jones E. Alzheimer's disease. Lancet. 2011; 377(9770):1019–31.10.1016/S0140-6736(10)61349-9 [PubMed: 21371747]

Biffi A, Anderson CD, Desikan RS, Sabuncu M, Cortellini L, Schmansky N, Salat D, Rosand J. Genetic variation and neuroimaging measures in Alzheimer disease. Arch Neurol. 2010; 67(6):677–85.10.1001/archneurol.2010.108 [PubMed: 20558387]

Bis JC, DeCarli C, Smith AV, van der Lijn F, Crivello F, Fornage M, Debette S, Shulman JM, Schmidt H, Srikanth V, Schuur M, Yu L, Choi SH, Sigurdsson S, Verhaaren BF, DeStefano AL, Lambert JC, Jack CR Jr, Struchalin M, Stankovich J, Ibrahim-Verbaas CA, Fleischman D, Zijdenbos A, den Heijer T, Mazoyer B, Coker LH, Enzinger C, Danoy P, Amin N, Arfanakis K, van Buchem MA, de Bruijn RF, Beiser A, Dufouil C, Huang J, Cavalieri M, Thomson R, Niessen WJ, Chibnik LB, Gislason GK, Hofman A, Pikula A, Amouyel P, Freeman KB, Phan TG, Oostra BA, Stein JL, Medland SE, Vasquez AA, Hibar DP, Wright MJ, Franke B, Martin NG, Thompson PM, Nalls MA, Uitterlinden AG, Au R, Elbaz A, Beare RJ, van Swieten JC, Lopez OL, Harris TB, Chouraki V, Breteler MM, De Jager PL, Becker JT, Vernooij MW, Knopman D, Fazekas F, Wolf PA, van der Lugt A, Gudnason V, Longstreth WT Jr, Brown MA, Bennett DA, van Duijn CM, Mosley TH, Schmidt R, Tzourio C, Launer LJ, Ikram MA, Seshadri S. Common variants at 12q14 and 12q24 are associated with hippocampal volume. Nat Genet. 2012; 44(5):545–51.10.1038/ng.2237 [PubMed: 22504421]

Boada M, Antunez C, Ramirez-Lorca R, Destefano AL, Gonzalez-Perez A, Gayan J, Lopez-Arrieta J, Ikram MA, Hernandez I, Marin J, Galan JJ, Bis JC, Mauleon A, Rosende-Roca M, Moreno-Rey C, Gudnasson V, Moron FJ, Velasco J, Carrasco JM, Alegret M, Espinosa A, Vinyes G, Lafuente A, Vargas L, Fitzpatrick AL, Launer LJ, Saez ME, Vazquez E, Becker JT, Lopez OL, Serrano-Rios M, Tarraga L, van Duijn CM, Real LM, Seshadri S, Ruiz A. ATP5H/KCTD2 locus is associated with Alzheimer's disease risk. Molecular psychiatry. 201310.1038/mp.2013.86

Bradshaw EM, Chibnik LB, Keenan BT, Ottoboni L, Raj T, Tang A, Rosenkrantz LL, Imboywa S, Lee M, Von Korff A, Morris MC, Evans DA, Johnson K, Sperling RA, Schneider JA, Bennett DA, De Jager PL. CD33 Alzheimer's disease locus: altered monocyte function and amyloid biology. Nature neuroscience. 2013; 16(7):848–50.10.1038/nn.3435

Carrasquillo MM, Khan QU, Murray ME, Krishnan S, Aakre J, Pankratz VS, Nguyen T, Ma L, Bisceglio G, Petersen RC, Younkin SG, Dickson DW, Boeve BF, Graff-Radford NR, Ertekin-Taner N. Late-onset Alzheimer disease genetic variants in posterior cortical atrophy and posterior AD. Neurology. 2014; 82(16):1455–62.10.1212/WNL.0000000000000335 [PubMed: 24670887]

Carrasquillo MM, Zou F, Pankratz VS, Wilcox SL, Ma L, Walker LP, Younkin SG, Younkin CS, Younkin LH, Bisceglio GD, Ertekin-Taner N, Crook JE, Dickson DW, Petersen RC, Graff-Radford NR. Genetic variation in PCDH11X is associated with susceptibility to late-onset Alzheimer's disease. Nat Genet. 2009; 41(2):192–8.10.1038/ng.305 [PubMed: 19136949]

Dastani Z, Hivert MF, Timpson N, Perry JR, Yuan X, Scott RA, Henneman P, Heid IM, Kizer JR, Lyytikainen LP, Fuchsberger C, Tanaka T, Morris AP, Small K, Isaacs A, Beekman M, Coassin S, Lohman K, Qi L, Kanoni S, Pankow JS, Uh HW, Wu Y, Bidulescu A, Rasmussen-Torvik LJ, Greenwood CM, Ladouceur M, Grimsby J, Manning AK, Liu CT, Kooner J, Mooser VE,

Vollenweider P, Kapur KA, Chambers J, Wareham NJ, Langenberg C, Frants R, Willems-Vandijk K, Oostra BA, Willems SM, Lamina C, Winkler TW, Psaty BM, Tracy RP, Brody J, Chen I, Viikari J, Kahonen M, Pramstaller PP, Evans DM, St Pourcain B, Sattar N, Wood AR, Bandinelli S, Carlson OD, Egan JM, Bohringer S, van Heemst D, Kedenko L, Kristiansson K, Nuotio ML, Loo BM, Harris T, Garcia M, Kanaya A, Haun M, Klopp N, Wichmann HE, Deloukas P, Katsareli E, Couper DJ, Duncan BB, Kloppenburg M, Adair LS, Borja JB, Wilson JG, Musani S, Guo X, Johnson T, Semple R, Teslovich TM, Allison MA, Redline S, Buxbaum SG, Mohlke KL, Meulenbelt I, Ballantyne CM, Dedoussis GV, Hu FB, Liu Y, Paulweber B, Spector TD, Slagboom PE, Ferrucci L, Jula A, Perola M, Raitakari O, Florez JC, Salomaa V, Eriksson JG, Frayling TM, Hicks AA, Lehtimaki T, Smith GD, Siscovick DS, Kronenberg F, van Duijn C, Loos RJ, Waterworth DM, Meigs JB, Dupuis J, Richards JB, Voight BF, Scott LJ, Steinthorsdottir V, Dina C, Welch RP, Zeggini E, Huth C, Aulchenko YS, Thorleifsson G, McCulloch LJ, Ferreira T, Grallert H, Amin N, Wu G, Willer CJ, Raychaudhuri S, McCarroll SA, Hofmann OM, Segre AV, van Hoek M, Navarro P, Ardlie K, Balkau B, Benediktsson R, Bennett AJ, Blagieva R, Boerwinkle E, Bonnycastle LL, Bostrom KB, Bravenboer B, Bumpstead S, Burtt NP, Charpentier G, Chines PS, Cornelis M, Crawford G, Doney AS, Elliott KS, Elliott AL, Erdos MR, Fox CS, Franklin CS, Ganser M, Gieger C, Grarup N, Green T, Griffin S, Groves CJ, Guiducci C, Hadjadj S, Hassanali N, Herder C, Isomaa B, Jackson AU, Johnson PR, Jorgensen T, Kao WH, Kong A, Kraft P, Kuusisto J, Lauritzen T, Li M, Lieverse A, Lindgren CM, Lyssenko V, Marre M, Meitinger T, Midthjell K, Morken MA, Narisu N, Nilsson P, Owen KR, Payne F, Petersen AK, Platou C, Proenca C, Prokopenko I, Rathmann W, Rayner NW, Robertson NR, Rocheleau G, Roden M, Sampson MJ, Saxena R, Shields BM, Shrader P, Sigurdsson G, Sparso T, Strassburger K, Stringham HM, Sun Q, Swift AJ, Thorand B, Tichet J, Tuomi T, van Dam RM, van Haeften TW, van Herpt T, van Vliet-Ostaptchouk JV, Walters GB, Weedon MN, Wijmenga C, Witteman J, Bergman RN, Cauchi S, Collins FS, Gloyn AL, Gyllensten U, Hansen T, Hide WA, Hitman GA, Hofman A, Hunter DJ, Hveem K, Laakso M, Morris AD, Palmer CN, Rudan I, Sijbrands E, Stein LD, Tuomilehto J, Uitterlinden A, Walker M, Watanabe RM, Abecasis GR, Boehm BO, Campbell H, Daly MJ, Hattersley AT, Pedersen O, Barroso I, Groop L, Sladek R, Thorsteinsdottir U, Wilson JF, Illig T, Froguel P, van Duijn CM, Stefansson K, Altshuler D, Boehnke M, McCarthy MI, Soranzo N, Wheeler E, Glazer NL, Bouatia-Naji N, Magi R, Randall J, Elliott P, Rybin D, Dehghan A, Hottenga JJ, Song K, Goel A, Lajunen T, Doney A, Cavalcanti-Proenca C, Kumari M, Timpson NJ, Zabena C, Ingelsson E, An P, O'Connell J, Luan J, Elliott A, Roccasecca RM, Pattou F, Sethupathy P, Ariyurek Y, Barter P, Beilby JP, Ben-Shlomo Y, Bergmann S, Bochud M, Bonnefond A, Borch-Johnsen K, Bottcher Y, Brunner E, Bumpstead SJ, Chen YD, Chines P, Clarke R, Coin LJ, Cooper MN, Crisponi L, Day IN, de Geus EJ, Delplanque J, Fedson AC, Fischer-Rosinsky A, Forouhi NG, Franzosi MG, Galan P, Goodarzi MO, Graessler J, Grundy S, Gwilliam R, Hallmans G, Hammond N, Han X, Hartikainen AL, Hayward C, Heath SC, Hercberg S, Hillman DR, Hingorani AD, Hui J, Hung J, Kaakinen M, Kaprio J, Kesaniemi YA, Kivimaki M, Knight B, Koskinen S, Kovacs P, Kyvik KO, Lathrop GM, Lawlor DA, Le Bacquer O, Lecoeur C, Li Y, Mahley R, Mangino M, Martinez-Larrad MT, McAteer JB, McPherson R, Meisinger C, Melzer D, Meyre D, Mitchell BD, Mukherjee S, Naitza S, Neville MJ, Orru M, Pakyz R, Paolisso G, Pattaro C, Pearson D, Peden JF, Pedersen NL, Pfeiffer AF, Pichler I, Polasek O, Posthuma D, Potter SC, Pouta A, Province MA, Rice K, Ripatti S, Rivadeneira F, Rolandsson O, Sandbaek A, Sandhu M, Sanna S, Sayer AA, Scheet P, Seedorf U, Sharp SJ, Shields B, Sigurethsson G, Sijbrands EJ, Silveira A, Simpson L, Singleton A, Smith NL, Sovio U, Swift A, Syddall H, Syvanen AC, Tonjes A, Uitterlinden AG, van Dijk KW, Varma D, Visvikis-Siest S, Vitart V, Vogelzangs N, Waeber G, Wagner PJ, Walley A, Ward KL, Watkins H, Wild SH, Willemsen G, Witteman JC, Yarnell JW, Zelenika D, Zethelius B, Zhai G, Zhao JH, Zillikens MC, Borecki IB, Meneton P, Magnusson PK, Nathan DM, Williams GH, Silander K, Bornstein SR, Schwarz P, Spranger J, Karpe F, Shuldiner AR, Cooper C, Serrano-Rios M, Lind L, Palmer LJ, Hu FBs, Franks PW, Ebrahim S, Marmot M, Wright AF, Stumvoll M, Hamsten A, Buchanan TA, Valle TT, Rotter JI, Penninx BW, Boomsma DI, Cao A, Scuteri A, Schlessinger D, Uda M, Ruokonen A, Jarvelin MR, Peltonen L, Mooser V, Musunuru K, Smith AV, Edmondson AC, Stylianou IM, Koseki M, Pirruccello JP, Chasman DI, Johansen CT, Fouchier SW, Peloso GM, Barbalic M, Ricketts SL, Bis JC, Feitosa MF, Orho-Melander M, Melander O, Li X, Cho YS, Go MJ, Kim YJ, Lee JY, Park T, Kim K, Sim X, Ong RT, Croteau-Chonka DC, Lange LA, Smith JD, Ziegler A, Zhang W, Zee RY, Whitfield JB, Thompson JR, Surakka I, Smit JH, Sinisalo J, Scott J, Saharinen J, Sabatti C, Rose LM, Roberts R, Rieder M, Parker AN, Pare G,

Oials_e;Donnell CJ, Nieminen MS, Nickerson DA, Montgomery GW, McArdle W, Masson D, Martin NG, Marroni F, Lucas G, Luben R, Lokki ML, Lettre G, Launer LJ, Lakatta EG, Laaksonen R, Konig IR, Khaw KT, Kaplan LM, Johansson A, Janssens AC, Igl W, Hovingh GK, Hengstenberg C, Havulinna AS, Hastie ND, Harris TB, Haritunians T, Hall AS, Groop LC, Gonzalez E, Freimer NB, Erdmann J, Ejebe KG, Doring A, Dominiczak AF, Demissie S, de Faire U, Caulfield MJ, Boekholdt SM, Assimes TL, Quertermous T, Seielstad M, Wong TY, Tai ES, Feranil AB, Kuzawa CW, Taylor HA Jr, Gabriel SB, Holm H, Gudnason V, Krauss RM, Ordovas JM, Munroe PB, Kooner JS, Tall AR, Hegele RA, Kastelein JJ, Schadt EE, Strachan DP, Reilly MP, Samani NJ, Schunkert H, Cupples LA, Sandhu MS, Ridker PM, Rader DJ, Kathiresan S. Novel loci for adiponectin levels and their influence on type 2 diabetes and metabolic traits: a multi-ethnic meta-analysis of 45,891 individuals. PLoS genetics. 2012; 8(3):e1002607.10.1371/journal.pgen.1002607 [PubMed: 22479202]

Debette S, Bis JC, Fornage M, Schmidt H, Ikram MA, Sigurdsson S, Heiss G, Struchalin M, Smith AV, van der Lugt A, DeCarli C, Lumley T, Knopman DS, Enzinger C, Eiriksdottir G, Koudstaal PJ, DeStefano AL, Psaty BM, Dufouil C, Catellier DJ, Fazekas F, Aspelund T, Aulchenko YS, Beiser A, Rotter JI, Tzourio C, Shibata DK, Tscherner M, Harris TB, Rivadeneira F, Atwood LD, Rice K, Gottesman RF, van Buchem MA, Uitterlinden AG, Kelly-Hayes M, Cushman M, Zhu Y, Boerwinkle E, Gudnason V, Hofman A, Romero JR, Lopez O, van Duijn CM, Au R, Heckbert SR, Wolf PA, Mosley TH, Seshadri S, Breteler MM, Schmidt R, Launer LJ, Longstreth WT Jr. Genome-wide association studies of MRI-defined brain infarcts: meta-analysis from the CHARGE Consortium. Stroke. 2010; 41(2):210–7.10.1161/strokeaha.109.569194 [PubMed: 20044523]

Debette S, Markus HS. The clinical importance of white matter hyperintensities on brain magnetic resonance imaging: systematic review and meta-analysis. BMJ. 2010; 341:c3666. [PubMed: 20660506]

den Heijer T, Oudkerk M, Launer LJ, van Duijn CM, Hofman A, Breteler MM. Hippocaxmpal, amygdalar, and global brain atrophy in different apolipoprotein E genotypes. Neurology. 2002; 59(5):746–8. [PubMed: 12221169]

Ferencz B, Laukka EJ, Lovden M, Kalpouzos G, Keller L, Graff C, Wahlund LO, Fratiglioni L, Backman L. The influence of APOE and TOMM40 polymorphisms on hippocampal volume and episodic memory in old age. Front Hum Neurosci. 2013; 7:198.10.3389/fnhum.2013.00198 [PubMed: 23734114]

Fornage M, Debette S, Bis JC, Schmidt H, Ikram MA, Dufouil C, Sigurdsson S, Lumley T, DeStefano AL, Fazekas F, Vrooman HA, Shibata DK, Maillard P, Zijdenbos A, Smith AV, Gudnason H, de Boer R, Cushman M, Mazoyer B, Heiss G, Vernooij MW, Enzinger C, Glazer NL, Beiser A, Knopman DS, Cavalieri M, Niessen WJ, Harris TB, Petrovic K, Lopez OL, Au R, Lambert JC, Hofman A, Gottesman RF, Garcia M, Heckbert SR, Atwood LD, Catellier DJ, Uitterlinden AG, Yang Q, Smith NL, Aspelund T, Romero JR, Rice K, Taylor KD, Nalls MA, Rotter JI, Sharrett R, van Duijn CM, Amouyel P, Wolf PA, Gudnason V, van der Lugt A, Boerwinkle E, Psaty BM, Seshadri S, Tzourio C, Breteler MM, Mosley TH, Schmidt R, Longstreth WT, DeCarli C, Launer LJ. Genome-wide association studies of cerebral white matter lesion burden: the CHARGE consortium. Ann Neurol. 2011; 69(6):928–39.10.1002/ana.22403 [PubMed: 21681796]

Furney SJ, Simmons A, Breen G, Pedroso I, Lunnon K, Proitsi P, Hodges A, Powell J, Wahlund LO, Kloszewska I, Mecocci P, Soininen H, Tsolaki M, Vellas B, Spenger C, Lathrop M, Shen L, Kim S, Saykin AJ, Weiner MW, Lovestone S. Genome-wide association with MRI atrophy measures as a quantitative trait locus for Alzheimer's disease. Molecular psychiatry. 2011; 16(11):1130–8.10.1038/mp.2010.123 [PubMed: 21116278]

Gauderman WJ. Sample size requirements for association studies of gene-gene interaction. Am J Epidemiol. 2002a; 155(5):478–84. [PubMed: 11867360]

Gauderman WJ. Sample size requirements for matched case-control studies of gene-environment interaction. Stat Med. 2002b; 21(1):35–50. [PubMed: 11782049]

Harold D, Abraham R, Hollingworth P, Sims R, Gerrish A, Hamshere ML, Pahwa JS, Moskvina V, Dowzell K, Williams A, Jones N, Thomas C, Stretton A, Morgan AR, Lovestone S, Powell J, Proitsi P, Lupton MK, Brayne C, Rubinsztein DC, Gill M, Lawlor B, Lynch A, Morgan K, Brown KS, Passmore PA, Craig D, McGuinness B, Todd S, Holmes C, Mann D, Smith AD, Love S, Kehoe PG, Hardy J, Mead S, Fox N, Rossor M, Collinge J, Maier W, Jessen F, Schurmann B, van den Bussche H, Heuser I, Kornhuber J, Wiltfang J, Dichgans M, Frolich L, Hampel H, Hull M,

Rujescu D, Goate AM, Kauwe JS, Cruchaga C, Nowotny P, Morris JC, Mayo K, Sleegers K, Bettens K, Engelborghs S, De Deyn PP, Van Broeckhoven C, Livingston G, Bass NJ, Gurling H, McQuillin A, Gwilliam R, Deloukas P, Al-Chalabi A, Shaw CE, Tsolaki M, Singleton AB, Guerreiro R, Muhleisen TW, Nothen MM, Moebus S, Jockel KH, Klopp N, Wichmann HE, Carrasquillo MM, Pankratz VS, Younkin SG, Holmans PA, O'Donovan M, Owen MJ, Williams J. Genome-wide association study identifies variants at CLU and PICALM associated with Alzheimer's disease. Nat Genet. 2009; 41(10):1088–93.10.1038/ng.440 [PubMed: 19734902]

Hollingworth P, Harold D, Sims R, Gerrish A, Lambert JC, Carrasquillo MM, Abraham R, Hamshere ML, Pahwa JS, Moskvina V, Dowzell K, Jones N, Stretton A, Thomas C, Richards A, Ivanov D, Widdowson C, Chapman J, Lovestone S, Powell J, Proitsi P, Lupton MK, Brayne C, Rubinsztein DC, Gill M, Lawlor B, Lynch A, Brown KS, Passmore PA, Craig D, McGuinness B, Todd S, Holmes C, Mann D, Smith AD, Beaumont H, Warden D, Wilcock G, Love S, Kehoe PG, Hooper NM, Vardy ER, Hardy J, Mead S, Fox NC, Rossor M, Collinge J, Maier W, Jessen F, Ruther E, Schurmann B, Heun R, Kolsch H, van den Bussche H, Heuser I, Kornhuber J, Wiltfang J, Dichgans M, Frolich L, Hampel H, Gallacher J, Hull M, Rujescu D, Giegling I, Goate AM, Kauwe JS, Cruchaga C, Nowotny P, Morris JC, Mayo K, Sleegers K, Bettens K, Engelborghs S, De Deyn PP, Van Broeckhoven C, Livingston G, Bass NJ, Gurling H, McQuillin A, Gwilliam R, Deloukas P, Al-Chalabi A, Shaw CE, Tsolaki M, Singleton AB, Guerreiro R, Muhleisen TW, Nothen MM, Moebus S, Jockel KH, Klopp N, Wichmann HE, Pankratz VS, Sando SB, Aasly JO, Barcikowska M, Wszolek ZK, Dickson DW, Graff-Radford NR, Petersen RC, van Duijn CM, Breteler MM, Ikram MA, DeStefano AL, Fitzpatrick AL, Lopez O, Launer LJ, Seshadri S, Berr C, Campion D, Epelbaum J, Dartigues JF, Tzourio C, Alperovitch A, Lathrop M, Feulner TM, Friedrich P, Riehle C, Krawczak M, Schreiber S, Mayhaus M, Nicolhaus S, Wagenpfeil S, Steinberg S, Stefansson H, Stefansson K, Snaedal J, Bjornsson S, Jonsson PV, Chouraki V, Genier-Boley B, Hiltunen M, Soininen H, Combarros O, Zelenika D, Delepine M, Bullido MJ, Pasquier F, Mateo I, Frank-Garcia A, Porcellini E, Hanon O, Coto E, Alvarez V, Bosco P, Siciliano G, Mancuso M, Panza F, Solfrizzi V, Nacmias B, Sorbi S, Bossu P, Piccardi P, Arosio B, Annoni G, Seripa D, Pilotto A, Scarpini E, Galimberti D, Brice A, Hannequin D, Licastro F, Jones L, Holmans PA, Jonsson T, Riemenschneider M, Morgan K, Younkin SG, Owen MJ, O'Donovan M, Amouyel P, Williams J. Common variants at ABCA7, MS4A6A/MS4A4E, EPHA1, CD33 and CD2AP are associated with Alzheimer's disease. Nat Genet. 2011; 43(5):429–35.10.1038/ng.803 [PubMed: 21460840]

Ikram MA, Fornage M, Smith AV, Seshadri S, Schmidt R, Debette S, Vrooman HA, Sigurdsson S, Ropele S, Taal HR, Mook-Kanamori DO, Coker LH, Longstreth WT Jr, Niessen WJ, DeStefano AL, Beiser A, Zijdenbos AP, Struchalin M, Jack CR Jr, Rivadeneira F, Uitterlinden AG, Knopman DS, Hartikainen AL, Pennell CE, Thiering E, Steegers EA, Hakonarson H, Heinrich J, Palmer LJ, Jarvelin MR, McCarthy MI, Grant SF, St Pourcain B, Timpson NJ, Smith GD, Sovio U, Nalls MA, Au R, Hofman A, Gudnason H, van der Lugt A, Harris TB, Meeks WM, Vernooij MW, van Buchem MA, Catellier D, Jaddoe VW, Gudnason V, Windham BG, Wolf PA, van Duijn CM, Mosley TH Jr, Schmidt H, Launer LJ, Breteler MM, DeCarli C. Common variants at 6q22 and 17q21 are associated with intracranial volume. Nat Genet. 2012; 44(5):539–44.10.1038/ng.2245 [PubMed: 22504418]

Jack CR Jr, Knopman DS, Jagust WJ, Petersen RC, Weiner MW, Aisen PS, Shaw LM, Vemuri P, Wiste HJ, Weigand SD, Lesnick TG, Pankratz VS, Donohue MC, Trojanowski JQ. Tracking pathophysiological processes in Alzheimer's disease: an updated hypothetical model of dynamic biomarkers. Lancet Neurol. 2013; 12(2):207–16.10.1016/s1474-4422(12)70291-0 [PubMed: 23332364]

Jack CR Jr, Knopman DS, Jagust WJ, Shaw LM, Aisen PS, Weiner MW, Petersen RC, Trojanowski JQ. Hypothetical model of dynamic biomarkers of the Alzheimer's pathological cascade. Lancet Neurol. 2010; 9(1):119–28.10.1016/s1474-4422(09)70299-6 [PubMed: 20083042]

Jonsson T, Atwal JK, Steinberg S, Snaedal J, Jonsson PV, Bjornsson S, Stefansson H, Sulem P, Gudbjartsson D, Maloney J, Hoyte K, Gustafson A, Liu Y, Lu Y, Bhangale T, Graham RR, Huttenlocher J, Bjornsdottir G, Andreassen OA, Jonsson EG, Palotie A, Behrens TW, Magnusson OT, Kong A, Thorsteinsdottir U, Watts RJ, Stefansson K. A mutation in APP protects against Alzheimer's disease and age-related cognitive decline. Nature. 2012; 488(7409):96–9.10.1038/nature11283 [PubMed: 22801501]

Jonsson T, Stefansson H, Steinberg S, Jonsdottir I, Jonsson PV, Snaedal J, Bjornsson S, Huttenlocher J, Levey AI, Lah JJ, Rujescu D, Hampel H, Giegling I, Andreassen OA, Engedal K, Ulstein I, Djurovic S, Ibrahim-Verbaas C, Hofman A, Ikram MA, van Duijn CM, Thorsteinsdottir U, Kong A, Stefansson K. Variant of TREM2 associated with the risk of Alzheimer's disease. N Engl J Med. 2013; 368(2):107–16.10.1056/NEJMoa1211103 [PubMed: 23150908]

Kaye JA, Swihart T, Howieson D, Dame A, Moore MM, Karnos T, Camicioli R, Ball M, Oken B, Sexton G. Volume loss of the hippocampus and temporal lobe in healthy elderly persons destined to develop dementia. Neurology. 1997; 48(5):1297–304. [PubMed: 9153461]

Khan W, Giampietro V, Ginestet C, Dell'Acqua F, Bouls D, Newhouse S, Dobson R, Banaschewski T, Barker GJ, Bokde AL, Buchel C, Conrod P, Flor H, Frouin V, Garavan H, Gowland P, Heinz A, Ittermann B, Lemaitre H, Nees F, Paus T, Pausova Z, Rietschel M, Smolka MN, Strohle A, Gallinat J, Westman E, Schumann G, Lovestone S, Simmons A. No differences in hippocampal volume between carriers and non-carriers of the ApoE epsilon4 and epsilon2 alleles in young healthy adolescents. J Alzheimers Dis. 2014; 40(1):37–43.10.3233/jad-131841 [PubMed: 24326516]

Lambert JC, Heath S, Even G, Campion D, Sleegers K, Hiltunen M, Combarros O, Zelenika D, Bullido MJ, Tavernier B, Letenneur L, Bettens K, Berr C, Pasquier F, Fievet N, Barberger-Gateau P, Engelborghs S, De Deyn P, Mateo I, Franck A, Helisalmi S, Porcellini E, Hanon O, de Pancorbo MM, Lendon C, Dufouil C, Jaillard C, Leveillard T, Alvarez V, Bosco P, Mancuso M, Panza F, Nacmias B, Bossu P, Piccardi P, Annoni G, Seripa D, Galimberti D, Hannequin D, Licastro F, Soininen H, Ritchie K, Blanche H, Dartigues JF, Tzourio C, Gut I, Van Broeckhoven C, Alperovitch A, Lathrop M, Amouyel P. Genome-wide association study identifies variants at CLU and CR1 associated with Alzheimer's disease. Nat Genet. 2009; 41(10):1094–9.10.1038/ng.439 [PubMed: 19734903]

Lambert JC, Ibrahim-Verbaas CA, Harold D, Naj AC, Sims R, Bellenguez C, Jun G, Destefano AL, Bis JC, Beecham GW, Grenier-Boley B, Russo G, Thornton-Wells TA, Jones N, Smith AV, Chouraki V, Thomas C, Ikram MA, Zelenika D, Vardarajan BN, Kamatani Y, Lin CF, Gerrish A, Schmidt H, Kunkle B, Dunstan ML, Ruiz A, Bihoreau MT, Choi SH, Reitz C, Pasquier F, Hollingworth P, Ramirez A, Hanon O, Fitzpatrick AL, Buxbaum JD, Campion D, Crane PK, Baldwin C, Becker T, Gudnason V, Cruchaga C, Craig D, Amin N, Berr C, Lopez OL, De Jager PL, Deramecourt V, Johnston JA, Evans D, Lovestone S, Letenneur L, Moron FJ, Rubinsztein DC, Eiriksdottir G, Sleegers K, Goate AM, Fievet N, Huentelman MJ, Gill M, Brown K, Kamboh MI, Keller L, Barberger-Gateau P, McGuinness B, Larson EB, Green R, Myers AJ, Dufouil C, Todd S, Wallon D, Love S, Rogaeva E, Gallacher J, St George-Hyslop P, Clarimon J, Lleo A, Bayer A, Tsuang DW, Yu L, Tsolaki M, Bossu P, Spalletta G, Proitsi P, Collinge J, Sorbi S, Sanchez-Garcia F, Fox NC, Hardy J, Naranjo MC, Bosco P, Clarke R, Brayne C, Galimberti D, Mancuso M, Matthews F, Moebus S, Mecocci P, Del Zompo M, Maier W, Hampel H, Pilotto A, Bullido M, Panza F, Caffarra P, Nacmias B, Gilbert JR, Mayhaus M, Lannfelt L, Hakonarson H, Pichler S, Carrasquillo MM, Ingelsson M, Beekly D, Alvarez V, Zou F, Valladares O, Younkin SG, Coto E, Hamilton-Nelson KL, Gu W, Razquin C, Pastor P, Mateo I, Owen MJ, Faber KM, Jonsson PV, Combarros O, O'Donovan MC, Cantwell LB, Soininen H, Blacker D, Mead S, Mosley TH Jr, Bennett DA, Harris TB, Fratiglioni L, Holmes C, de Bruijn RF, Passmore P, Montine TJ, Bettens K, Rotter JI, Brice A, Morgan K, Foroud TM, Kukull WA, Hannequin D, Powell JF, Nalls MA, Ritchie K, Lunetta KL, Kauwe JS, Boerwinkle E, Riemenschneider M, Boada M, Hiltunen M, Martin ER, Schmidt R, Rujescu D, Wang LS, Dartigues JF, Mayeux R, Tzourio C, Hofman A, Nothen MM, Graff C, Psaty BM, Jones L, Haines JL, Holmans PA, Lathrop M, Pericak-Vance MA, Launer LJ, Farrer LA, van Duijn CM, Van Broeckhoven C, Moskvina V, Seshadri S, Williams J, Schellenberg GD, Amouyel P. Meta-analysis of 74,046 individuals identifies 11 new susceptibility loci for Alzheimer's disease. Nature genetics. 2013; 45(12):1452–8.10.1038/ng.2802 [PubMed: 24162737]

Lehtovirta M, Laakso MP, Soininen H, Helisalmi S, Mannermaa A, Helkala EL, Partanen K, Ryynanen M, Vainio P, Hartikainen P, et al. Volumes of hippocampus, amygdala and frontal lobe in Alzheimer patients with different apolipoprotein E genotypes. Neuroscience. 1995; 67(1):65–72. [PubMed: 7477910]

Lehtovirta M, Soininen H, Laakso MP, Partanen K, Helisalmi S, Mannermaa A, Ryynanen M, Kuikka J, Hartikainen P, Riekkinen PJ Sr. SPECT and MRI analysis in Alzheimer's disease: relation to

apolipoprotein E epsilon 4 allele. J Neurol Neurosurg Psychiatry. 1996; 60(6):644–9. [PubMed: 8648331]

Lind J, Larsson A, Persson J, Ingvar M, Nilsson LG, Backman L, Adolfsson R, Cruts M, Sleegers K, Van Broeckhoven C, Nyberg L. Reduced hippocampal volume in non-demented carriers of the apolipoprotein E epsilon4: relation to chronological age and recognition memory. Neurosci Lett. 2006; 396(1):23–7.10.1016/j.neulet.2005.11.070 [PubMed: 16406347]

Liu JZ, McRae AF, Nyholt DR, Medland SE, Wray NR, Brown KM, Hayward NK, Montgomery GW, Visscher PM, Martin NG, Macgregor S. A versatile gene-based test for genome-wide association studies. Am J Hum Genet. 2010; 87(1):139–45.10.1016/j.ajhg.2010.06.009 [PubMed: 20598278]

Liu Y, Yu JT, Wang HF, Han PR, Tan CC, Wang C, Meng XF, Risacher SL, Saykin AJ, Tan L. APOE genotype and neuroimaging markers of Alzheimer's disease: systematic review and meta-analysis. J Neurol Neurosurg Psychiatry. 201410.1136/jnnp-2014-307719

Lu PH, Thompson PM, Leow A, Lee GJ, Lee A, Yanovsky I, Parikshak N, Khoo T, Wu S, Geschwind D, Bartzokis G. Apolipoprotein E genotype is associated with temporal and hippocampal atrophy rates in healthy elderly adults: a tensor-based morphometry study. J Alzheimers Dis. 2011; 23(3): 433–42.10.3233/jad-2010-101398 [PubMed: 21098974]

Morra JH, Tu Z, Apostolova LG, Green AE, Avedissian C, Madsen SK, Parikshak N, Toga AW, Jack CR Jr, Schuff N, Weiner MW, Thompson PM. Automated mapping of hippocampal atrophy in 1-year repeat MRI data from 490 subjects with Alzheimer's disease, mild cognitive impairment, and elderly controls. Neuroimage. 2009; 45(1 Suppl):S3–15.10.1016/j.neuroimage.2008.10.043 [PubMed: 19041724]

Naj AC, Jun G, Beecham GW, Wang LS, Vardarajan BN, Buros J, Gallins PJ, Buxbaum JD, Jarvik GP, Crane PK, Larson EB, Bird TD, Boeve BF, Graff-Radford NR, De Jager PL, Evans D, Schneider JA, Carrasquillo MM, Ertekin-Taner N, Younkin SG, Cruchaga C, Kauwe JS, Nowotny P, Kramer P, Hardy J, Huentelman MJ, Myers AJ, Barmada MM, Demirci FY, Baldwin CT, Green RC, Rogaeva E, St George-Hyslop P, Arnold SE, Barber R, Beach T, Bigio EH, Bowen JD, Boxer A, Burke JR, Cairns NJ, Carlson CS, Carney RM, Carroll SL, Chui HC, Clark DG, Corneveaux J, Cotman CW, Cummings JL, DeCarli C, DeKosky ST, Diaz-Arrastia R, Dick M, Dickson DW, Ellis WG, Faber KM, Fallon KB, Farlow MR, Ferris S, Frosch MP, Galasko DR, Ganguli M, Gearing M, Geschwind DH, Ghetti B, Gilbert JR, Gilman S, Giordani B, Glass JD, Growdon JH, Hamilton RL, Harrell LE, Head E, Honig LS, Hulette CM, Hyman BT, Jicha GA, Jin LW, Johnson N, Karlawish J, Karydas A, Kaye JA, Kim R, Koo EH, Kowall NW, Lah JJ, Levey AI, Lieberman AP, Lopez OL, Mack WJ, Marson DC, Martiniuk F, Mash DC, Masliah E, McCormick WC, McCurry SM, McDavid AN, McKee AC, Mesulam M, Miller BL, Miller CA, Miller JW, Parisi JE, Perl DP, Peskind E, Petersen RC, Poon WW, Quinn JF, Rajbhandary RA, Raskind M, Reisberg B, Ringman JM, Roberson ED, Rosenberg RN, Sano M, Schneider LS, Seeley W, Shelanski ML, Slifer MA, Smith CD, Sonnen JA, Spina S, Stern RA, Tanzi RE, Trojanowski JQ, Troncoso JC, Van Deerlin VM, Vinters HV, Vonsattel JP, Weintraub S, Welsh-Bohmer KA, Williamson J, Woltjer RL, Cantwell LB, Dombroski BA, Beekly D, Lunetta KL, Martin ER, Kamboh MI, Saykin AJ, Reiman EM, Bennett DA, Morris JC, Montine TJ, Goate AM, Blacker D, Tsuang DW, Hakonarson H, Kukull WA, Foroud TM, Haines JL, Mayeux R, Pericak-Vance MA, Farrer LA, Schellenberg GD. Common variants at MS4A4/MS4A6E, CD2AP, CD33 and EPHA1 are associated with late-onset Alzheimer's disease. Nat Genet. 2011; 43(5):436–41.10.1038/ng.801 [PubMed: 21460841]

Negash S, Xie S, Davatzikos C, Clark CM, Trojanowski JQ, Shaw LM, Wolk DA, Arnold SE. Cognitive and functional resilience despite molecular evidence of Alzheimer's disease pathology. Alzheimer's & dementia: the journal of the Alzheimer's Association. 2013; 9(3):e89–95.10.1016/j.jalz.2012.01.009

Nyholt DR. A simple correction for multiple testing for single-nucleotide polymorphisms in linkage disequilibrium with each other. American journal of human genetics. 2004; 74(4):765–9.10.1086/383251 [PubMed: 14997420]

O'Dwyer L, Lamberton F, Matura S, Tanner C, Scheibe M, Miller J, Rujescu D, Prvulovic D, Hampel H. Reduced hippocampal volume in healthy young ApoE4 carriers: an MRI study. PLoS One. 2012; 7(11):e48895.10.1371/journal.pone.0048895 [PubMed: 23152815]

Plassman BL, Welsh-Bohmer KA, Bigler ED, Johnson SC, Anderson CV, Helms MJ, Saunders AM, Breitner JC. Apolipoprotein E epsilon 4 allele and hippocampal volume in twins with normal cognition. Neurology. 1997; 48(4):985–9. [PubMed: 9109888]

Reiman EM, Uecker A, Caselli RJ, Lewis S, Bandy D, de Leon MJ, De Santi S, Convit A, Osborne D, Weaver A, Thibodeau SN. Hippocampal volumes in cognitively normal persons at genetic risk for Alzheimer's disease. Ann Neurol. 1998; 44(2):288–91.10.1002/ana.410440226 [PubMed: 9708558]

Schmidt H, Schmidt R, Fazekas F, Semmler J, Kapeller P, Reinhart B, Kostner GM. Apolipoprotein E e4 allele in the normal elderly: neuropsychologic and brain MRI correlates. Clin Genet. 1996; 50(5):293–9. [PubMed: 9007313]

Schuff N, Woerner N, Boreta L, Kornfield T, Shaw LM, Trojanowski JQ, Thompson PM, Jack CR Jr, Weiner MW. MRI of hippocampal volume loss in early Alzheimer's disease in relation to ApoE genotype and biomarkers. Brain. 2009; 132(Pt 4):1067–77.10.1093/brain/awp007 [PubMed: 19251758]

Seshadri S, Fitzpatrick AL, Ikram MA, DeStefano AL, Gudnason V, Boada M, Bis JC, Smith AV, Carassquillo MM, Lambert JC, Harold D, Schrijvers EM, Ramirez-Lorca R, Debette S, Longstreth WT Jr, Janssens AC, Pankratz VS, Dartigues JF, Hollingworth P, Aspelund T, Hernandez I, Beiser A, Kuller LH, Koudstaal PJ, Dickson DW, Tzourio C, Abraham R, Antunez C, Du Y, Rotter JI, Aulchenko YS, Harris TB, Petersen RC, Berr C, Owen MJ, Lopez-Arrieta J, Varadarajan BN, Becker JT, Rivadeneira F, Nalls MA, Graff-Radford NR, Campion D, Auerbach S, Rice K, Hofman A, Jonsson PV, Schmidt H, Lathrop M, Mosley TH, Au R, Psaty BM, Uitterlinden AG, Farrer LA, Lumley T, Ruiz A, Williams J, Amouyel P, Younkin SG, Wolf PA, Launer LJ, Lopez OL, van Duijn CM, Breteler MM. Genome-wide analysis of genetic loci associated with Alzheimer disease. JAMA. 2010; 303(18):1832–40.10.1001/jama.2010.574 [PubMed: 20460622]

Soininen H, Partanen K, Pitkanen A, Hallikainen M, Hanninen T, Helisalmi S, Mannermaa A, Ryynanen M, Koivisto K, Riekkinen P Sr. Decreased hippocampal volume asymmetry on MRIs in nondemented elderly subjects carrying the apolipoprotein E epsilon 4 allele. Neurology. 1995; 45(2):391–2. [PubMed: 7854548]

Sperling RA, Aisen PS, Beckett LA, Bennett DA, Craft S, Fagan AM, Iwatsubo T, Jack CR Jr, Kaye J, Montine TJ, Park DC, Reiman EM, Rowe CC, Siemers E, Stern Y, Yaffe K, Carrillo MC, Thies B, Morrison-Bogorad M, Wagster MV, Phelps CH. Toward defining the preclinical stages of Alzheimer's disease: recommendations from the National Institute on Aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease. Alzheimers Dement. 2011; 7(3):280–92.10.1016/j.jalz.2011.03.003 [PubMed: 21514248]

Vermeer SE, Longstreth WT Jr, Koudstaal PJ. Silent brain infarcts: a systematic review. Lancet Neurol. 2007; 6(7):611–9.10.1016/s1474-4422(07)70170-9 [PubMed: 17582361]

Highlights

1. It is unknown if novel AD risk loci impact brain structure in non-demented elderly
2. We performed a meta-analysis of genetic association studies in non-demented elderly
3. AD risk variants were associated in aggregate with smaller HV
4. Gene-based tests were significant for *HLA-DRB1* with TBV and *BIN1* with HV
5. Previously debated association of *APOE* risk variant with smaller HV was observed
6. Novel AD risk loci contribute to structural brain aging in older community persons

Table 1

Single-SNP based association of the AD loci with MRI markers of brain aging

Index SNP[a]	Proxy	Closest gene	Chr:position[b]	Distance from Gene[c]	Intra-Cranial Volume (in cm^3)			Total Brain Volume (in % ICV)			Hippocampal Volume (in cm^3)			WMH burden[d]		Brain Infarcts (yes/no)		
					β	SE	p	β	SE	p	β	SE	p	Z-statistics	p	β	SE	p
rs2075650		*APOE*	19:45395619	13.39kb	4.405	2.605	0.091	−0.1	0.072	0.168	−0.042	0.015	0.0054	1.089	0.276	−0.081	0.062	0.195
rs9331896	rs2279590	*CLU*	8:27467686	wg	−3.112	1.795	0.083	−0.104	0.051	0.04	−0.009	0.011	0.416	−1.546	0.122	−0.012	0.043	0.771
rs10792832		*PICALM*	11:85867875	86.95kb	0.763	1.681	0.65	0.064	0.047	0.18	−0.001	0.01	0.863	1.243	0.214	0.003	0.04	0.932
rs6656401		*CR1*	1:207692049	wg	−2.834	2.19	0.196	0.023	0.061	0.713	0.016	0.013	0.211	0.375	0.708	−0.069	0.054	0.197
rs6733839	rs744373	*BIN1*	2:127892810	27.91kb	−1.943	1.862	0.297	−0.07	0.052	0.183	−0.024	0.011	0.027	−0.168	0.867	0.079	0.043	0.064
rs4147929	rs3752246	*ABCA7*	19:1063443	wg	0.103	2.342	0.965	−0.018	0.065	0.786	−0.017	0.014	0.226	NA	NA	0.017	0.058	0.773
rs983392	rs11230161	*MS4A6A*	11:59923508	15.57kb	−3.093	1.675	0.065	−0.059	0.047	0.214	−0.023	0.01	0.021	−1.42	0.156	−0.012	0.043	0.782
rs10948363		*CD2AP*	6:47487762	wg	1.537	1.845	0.405	−0.017	0.052	0.742	0.003	0.011	0.87	1.089	0.276	−0.035	0.044	0.433
rs11771145		*EPHA1*	7:143110762	4.78kb	3.353	1.901	0.078	−0.026	0.053	0.625	0.003	0.011	0.912	NA	NA	−0.023	0.042	0.592
rs3865444		*CD33*	19:51727962	0.36kb	−5.209	1.886	0.0058	0.025	0.053	0.638	−0.019	0.011	0.087	−0.362	0.717	−0.088	0.045	0.048
rs9271192		*HLA-DRB1*[e]	6:32578530	20.92kb	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
rs28834970	rs2322599	*PTK2B*	8:27195121	wg	3.675	1.67	0.028	−0.006	0.047	0.898	−0.003	0.01	0.762	−0.824	0.41	−0.006	0.04	0.89
rs11218343	rs7939826	*SORL1*	11:121435587	wg	4.525	6.239	0.468	−0.165	0.174	0.341	0.011	0.037	0.768	NA	NA	0.316	0.155	0.041
rs10498633		*SLC24A4*	14:92926952	wg	−2.052	2.042	0.315	0.01	0.057	0.858	−0.012	0.012	0.329	0.363	0.717	0.049	0.048	0.304
rs35349669	rs7607736	*INPP5D*	2:234068476	wg	−3.625	1.723	0.035	−0.063	0.048	0.196	−0.01	0.01	0.313	0.856	0.392	−0.003	0.041	0.935
rs190982		*MEF2C*	5:88223420	23.50kb	−1.611	1.918	0.401	0.034	0.054	0.525	0.005	0.011	0.687	0.545	0.586	0.046	0.044	0.3
rs2718058	rs12155159	*NME8*	7:37841534	46.67kb	2.168	1.762	0.218	0	0.05	0.994	0.012	0.01	0.271	−0.438	0.662	0.027	0.042	0.523
rs1476679		*ZCWPW1*	7:100004446	wg	−0.22	1.8	0.903	−0.017	0.051	0.738	−0.01	0.011	0.36	−0.053	0.958	−0.014	0.044	0.754
rs10838725	rs10838726	*CELF1*	11:47557871	wg	−0.433	1.795	0.809	0.085	0.05	0.092	0	0.011	0.992	−1.012	0.312	−0.068	0.043	0.115
rs17125944		*FERMT2*	14:53400629	wg	0.465	2.767	0.867	−0.025	0.078	0.744	0.015	0.016	0.347	−0.574	0.566	0.069	0.071	0.332
rs7274581	rs927174	*CASS4*	20:55018260	wg	−0.435	2.956	0.883	−0.119	0.083	0.152	−0.014	0.017	0.421	0.055	0.956	0.084	0.07	0.228

Key: β, beta (meta-analysis effect estimate) per allele increase of the risk allele; Z-statistic, meta-analysis of Z-statistics (beta/SE) from each study, weighted by effective sample size (product of the sample size and the ratio of the empirically observed dosage variance to the expected binomial dosage variance for imputed SNPs); WMH, white matter hyperintensities; SE, standard error

[a] Index SNP was defined as the SNP with the lowest p at the locus.

[b] Chr:position has been provided for the index SNP as per NCBI build 37 (GRCh37.p10).

[c] Distance from gene start or end (whichever is shortest) is provided in kilo bases (kb) and if within gene, wg notation used.

[d] expressed in cm^3 or on a semi-quantitative 10-point scale in the original study.

[e] Neither the index SNP nor any SNP in LD with index SNP is available in the HapMap based imputed data meta-analysis results $p<0.0025$ ($\alpha=0.05/20$) was considered significant after correcting for number of independent loci tested

Neurobiol Aging. Author manuscript; available in PMC 2016 April 01.

Table 2

Gene-based associations (P<0.05) with MRI markers of brain aging for genes lying within 50kb of AD risk loci

Index-SNP (closest gene)	Gene	Chr	Start	Stop	p (Intra-cranial volume)	p (Total brain volume)	p (Hippocampal volume)	p (WMH burden)	p (brain infarcts)
rs6656401 (CR1)	*CR1*	1	207619472	207865110	0.271	0.0033	0.237	0.069	0.562
rs744373 (BIN1)	*BIN1*	2	127755598	127914903	0.612	0.782	0.00089	0.700	0.072
rs190982 (MEF2C)	*MEF2C*	5	87964057	88249922	0.020	0.815	0.134	0.180	0.033
rs9271192 (HLA-DRB1)	*HLA-DRB1*	6	32496546	32607613	0.467	0.00060	0.170	0.226	0.277
rs9271192 (HLA-DRB1)	*HLA-DQA1*	6	32555182	32661429	0.263	0.0014	0.108	0.059	0.310
rs9271192 (HLA-DRB1)	*HLA-DQB1*	6	32577240	32684466	0.179	0.0057	0.114	0.010	0.208
rs75932628 (TREM2)	*TREML1*	6	41066998	41172087	0.337	0.367	0.048	0.315	0.582
rs12155159 (NME8)	*NME8*	7	37838198	37990002	0.010	0.400	0.138	0.755	0.985
rs1476679 (ZCWPW1)	*PILRB*	7	99905625	100015454	0.0082	0.694	0.162	0.271	0.701
rs1476679 (ZCWPW1)	*PILRA*	7	99921067	100047722	0.0078	0.702	0.183	0.309	0.712
rs1476679 (ZCWPW1)	*ZCWPW1*	7	99948494	100076431	0.0078	0.672	0.196	0.363	0.751
rs1476679 (ZCWPW1)	*MEPCE*	7	99976412	100081749	0.0099	0.626	0.246	0.348	0.739
rs1476679 (ZCWPW1)	*PPP1R35*	7	99982911	100084094	0.0093	0.637	0.291	0.342	0.767
rs1476679 (ZCWPW1)	*C7orf61*	7	100004237	100111894	0.011	0.668	0.320	0.321	0.811
rs11771145 (EPHA1)	*TAS2R60*	7	143090545	143191502	0.931	0.012	0.393	0.086	0.049
rs2279590 (CLU)	*SCARA3*	8	27441576	27584286	0.060	0.031	0.367	0.440	0.651
rs11230161 (MS4A6A)	*MS4A6A*	11	59889079	60002139	0.035	0.375	0.030	0.358	0.804
rs11870474 (ATP5H/KCTD2)	*ICT1*	17	72958779	73067356	0.138	0.047	0.434	0.697	0.195
rs3752246 (ABCA7)	*ABCA7*	19	990101	1115570	0.497	0.589	0.799	0.049	0.301
rs3752246 (ABCA7)	*HMHA1*	19	1015921	1137830	0.337	0.577	0.724	0.046	0.128
rs2075650 (APOE)	*PVRL2*	19	45299392	45442485	0.033	0.470	0.069	0.163	0.056
rs2075650 (APOE)	*TOMM40*	19	45344476	45456946	0.027	0.370	0.084	0.202	0.155
rs2075650 (APOE)	*APOE*	19	45359038	45462650	0.040	0.378	0.118	0.252	0.133
rs2075650 (APOE)	*APOC1*	19	45367920	45472606	0.030	0.488	0.106	0.274	0.163
rs3865444 (CD33)	*CD33*	19	51678334	51793274	0.179	0.046	0.463	0.968	0.100
rs927174 (CASS4)	*AURKA*	20	54894444	55017351	0.262	0.690	0.415	0.771	0.041

Index-SNP (closest gene)	Gene	Chr	Start	Stop	p (Intra-cranial volume)	p (Total brain volume)	p (Hippocampal volume)	p (WMH burden)	p (brain infarcts)
rs927174 (CASS4)	*CSTF1*	20	54917426	55029582	0.400	0.526	0.550	0.823	0.047

Key: WMH, white matter hyperintensities

p<0.0025 (α=0.05/20) was considered significant after correcting for number of independent loci tested; significant p-values after correcting for multiple testing are in bold; Gene-based association analysis was performed for genes within 50kB of index SNP. Only gene-based associations for those genes with p<0.05 with at least one MRI marker is presented. A complete list is presented in Supplementary Table 4.

Table 3

Genetic risk score based association of the AD loci with MRI-markers of brain aging

	With *APOE*			Without *APOE*		
	Beta	**SE**	**p**	**Beta**	**SE**	**p**
Intra-cranial volume (in cm^3)	1.179	2.174	0.59	−6.224	3.945	0.11
Total brain volume (in % ICV)	−0.120	0.061	0.048	−0.166	0.111	0.13
Hippocampal volume (in cm^3)	−0.044	0.013	0.00042	−0.050	0.023	0.029
WMH burden[a]	0.013	0.020	0.52	−0.019	0.038	0.61
Brain infarcts (yes/no)	−0.039	0.052	0.45	0.055	0.094	0.56

Key: Beta, effect estimate, per allele increase of the risk allele; SE, standard error; WMH, white matter hyperintensities

[a] for WMH burden betas and SEs were estimated from the Z-statistics obtained in the WMH burden meta-analysis and do not reflect an interpretable effect size (as the WMH burden was estimated using different scales in participating studies) (Fornage, et al., 2011).