Imaging Phenotypes in Alzheimer's Disease and MCI: Genes, Pathways and GWAS

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ALOIS ALZHEIMER (1864-1915)

- German scientist, initially a professor of Psychology in Breslau. Later in Munich he worked on histopathology. With Franz Nissl, worked to establish the neuropathological basis of mental illness. Alzheimer published on cerebral arteriosclerosis in 1904 and Huntington’s chorea in 1911. In 1907 he published the pathology of AD.
Overview

• Recent progress in understanding, detecting & monitoring Alzheimer’s disease - the role of genetics and biomarkers
  – US and world-wide Alzheimer’s Disease Neuroimaging Initiative (ADNI)
  – Mild cognitive impairment (MCI): Can we detect AD earlier? Early MCI - How early can we detect AD?

• GWAS and pathway based studies by the ADNI Genetics Core and other groups
  – Studies of older adults with cognitive complaints
  – Integrating brain imaging and genomics: challenges and future directions
Biomarkers are needed for early diagnosis, to predict transitions from NCI to MCI to AD and clinical trials of disease modifying therapies

Funded by the National Institute on Aging


Industry Scientific Advisory Board (ISAB) and Site PIs, Study Coordinators, and 821 subjects enrolled in 58 sites in US and Canada
Industry: Precompetitive Collaboration

ADNI Industry Scientific Advisory Board

New members Abbott, Genentech, Roche, Bayer

PIB/PET Supplement: Alzheimer’s Association and GE Healthcare
Cerebrospinal Fluid Extension: Alzheimer’s Association, AstraZeneca, Cure Alzheimer’s Fund, Merck, Pfizer and an anonymous foundation

Genome-Wide Genotyping: Gene Network Sciences, Merck, Pfizer and an anonymous foundation

Genome-Wide Genotyping Genetic Analysis: NIBIB, Merck, Pfizer and an anonymous foundation
GOALS OF ADNI-1
($40 M From NIH, $25 M from ISAB, Foundations & FNIH;
Funded from 10/1/2004 To 9/30/2009
with 1 Year No Cost Extension To 9/30/2010)

• Optimize and standardize biomarkers for clinical trials
• Validate biomarkers as measures of change
• Validate biomarkers as diagnostics or predictors
• Establish world-wide network for clinical AD studies and treatment trials
• Genetics: Only APOE genotyping included for purpose of stratification; DNA & LCLs banked
NIA GRAND OPPORTUNITIES (GO) ARRA GRANT
($24 M From NIH; Funded From 9/30/2009 to 9/30/2011)

• Adds cohort of 200 very mild “early” MCI (EMCI)
• LPs on 100 of new subjects
• Follow ADNI-1 controls/MCI additional yr
• F18 amyloid imaging on ALL existing and new ADNI/GO subjects (AV-45)
• Added Genetics Core
• Complete analysis of all ADNI data
SCOPE OF ADNI-2
($40 M From NIH & $29 M From ISAB, Foundations & FNIH; Funded From 10/1/2010 To 9/30/2015)

• Goal to continue to follow >400 controls and MCI from ADNI-1 for 5 more years and enroll:
  – 100 additional EMCI (supplements 200 from GO)
  – 150 new controls, LMCI, and AD
• MRI at 3, 6, months and annually
• F18 amyloid (AV-45)/FDG baseline and Yr 2
• LP on 100% of subjects at enrollment
• Genetics Core
ADNI-1: Naturalistic Study of AD

Naturalistic study of AD progression

- 200 NORMAL 3 yrs
- 400 MCI 3 yrs
- 200 AD 2 yrs
- Visits every 6 months

- 57 sites
- Clinical, blood, LP
- Cognitive Tests
- 1.5T MRI

Some also have
- 3.0T MRI (25%)
- FDG-PET (50%)
- PiB-PET (approx 100)

All data in public database:
UCLA/LONI/ADNI: No embargo of data
Future ADNI sites

Courtesy of Maria Carillo of the Alzheimer’s Association
ADNI Genetics Core

Genome Wide (620K SNP) Array and Copy Number Variation
Brain-Genome Association Strategies

Candidate Gene/SNP
- Risacher et al 2010

Biological Pathway
- Sloan et al 2010
- Swaminathan et al 2010 PiB ROIs & amyloid pathway
- Reiman et al 2009 Mol Psych schizophrenia study

Genome-wide Analysis
- Potkin et al 2009; Saykin et al 2010
- Shen et al 2010 ROIs;
- Stein et al 2010 voxels

ROI
- Egan et al 2001 COMT

Circuit
- Reiman et al PNAS 2009;
  Also Ho et al 2010 FTO

Whole Brain
- Reiman et al 2008 cholesterol pathway genes
Imaging Phenotypes in ADNI: Automated Cortical Parcellation and High Throughput Computation
Voxel-Based Morphometry (VBM)
Molecular Imaging of MCI/AD: Metabolism and Amyloid Deposition

[18F]FDG

[11C]PIB

IUSM 5/07
Global Grey Matter Density of Patient Groups (AD, MCI-Converter, MCI-Stable) Relative to HC Participants

n=693 (148 AD, 62 MCI-C, 277 MCI-S, 206 HC)

Covaried for age, gender, education, handedness and total intracranial volume (ICV)

HC>AD

HC>MCI-Converters

HC>MCI-Stable

p<0.005 (FDR), k=27

Risacher, Saykin, Shen et al; Current Alzheimer Research 2009; 6(4): 347-361
Relationship of Global Grey Matter Density Among Patient Groups (AD, MCI-Converter, MCI-Stable)

n=693 (148 AD, 62 MCI-C, 277 MCI-S, 206 HC); p<0.005 (FDR), k=27

Covaried for age, gender, education, handedness and total intracranial volume (ICV)

MCI-Stable>AD

MCI-Stable>MCI-Converters

MCI-Converters>AD – No Significantly Different Voxels

Risacher, Saykin, Shen et al; Current Alzheimer Research 2009; 6(4): 347-361
AD Phenotype: MTL Grey Matter Density, Volume, and Cortical Thickness in the ADNI Sample at Baseline
N=693 (148 AD, 62 MCI-C, 277 MCI-S, 206 HC); p<0.005 (FDR)

Risacher, Saykin, Shen et al; Current Alzheimer Research 2009; 6(4): 347-361
Regions Showing the Greatest Effect Sizes when Comparing MCI-Converter and MCI-Stable Participants at Baseline

Effect Size of Imaging Biomarkers for MCI-Converters vs. MCI-Stable

- Volume
- Cortical Thickness
- Grey Matter Density
- Volume (mod. VBM)

Major Genes: EOAD & LOAD

LOAD: genetic factors account for ~60-80% of risk (Gatz et al 2006); APOE accounts for up to 50% (Ashford & Mortimer 2002); so up to 30% remains to be found.
Many More Candidates
AlzGene Database:
Meta-Analysis of Top Candidate Genes for AD

http://www.alzforum.org/res/com/gen/alzgene/default.asp

“Top 40”: Oct 12, 2010
Genome-Wide Association Studies (GWAS)

“Gene Chip” - Illumina Human 610-Quad

- Mitochondrial SNPs
- Non-synonymous SNPs
- MHC markers
- Y chromosome SNPs

- 620,901 markers (~90% genomic coverage, CEU)
- Single nucleotide polymorphisms (SNPs)
- Copy number variation (CNVs) probes
Genome-wide association study identifies variants at CLU and PICALM associated with Alzheimer’s disease

Denise Harold, Michael Prentki, L. J. De Stasio, K. M. L. J. De Stasio, and A. De Stasio

Alzheimer’s disease is the most common form of dementia and is highly prevalent (with heritability of up to 70%) but genetically complex. Neurobiological disorders in the disease are characterized by extracellular amyloid plaques and intracellular neurofibrillary tangles containing phosphorylated tau protein. These four genes have been definitively implicated in the etiology of Alzheimer’s disease. Mutations in the genes encoding amyloid precursor protein (APP) and presenilin 1 and 2 (PSEN1, PSEN2) cause rare, mendelian forms of the disease, usually with an early onset. However, in the more common form of the disease, only APOE has been established unequivocally as a susceptibility gene. Attempting to identify other Alzheimer’s disease loci, several genome-wide association studies (GWAS) have been previously conducted. All have identified strong linkage disequilibrium between APOE and Alzheimer’s disease risk, raising the possibility that APOE may have a role in the disease. Here, we report the results of a large GWAS of Alzheimer’s disease cases and controls. We performed a GWAS of 5,000 Alzheimer’s disease cases and 4,500 controls. The results were replicated in an independent cohort of 4,000 Alzheimer’s disease cases and 5,000 controls. We identified two loci associated with Alzheimer’s disease: one near CLU and one near PICALM. These findings suggest that CLU and PICALM may play a role in the pathogenesis of Alzheimer’s disease.
Seshadri et al 2010 Largest AD GWAS

Genome-wide Analysis of Genetic Loci Associated With Alzheimer Disease

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Context  Genome-wide association studies (GWAS) have recently identified CLU, PICALM, and CR17 as novel genes for late-onset Alzheimer disease (AD).

Objectives  To identify and strengthen additional loci associated with AD and confirm these in an independent sample and to examine the contribution of recently identified genes to AD risk prediction in a 3-stage analysis of new and previously published GWAS on more than 35,000 persons (8371 AD cases).

Design, Setting, and Participants  In stage 1, we identified strong genetic associations ($p < 10^{-5}$) in a sample of 3006 AD cases and 14,642 controls by combining new data from the population-based Cohorts for Heart and Aging Research in Gencomic Epidemiology consortium (1367 AD cases [1973 incident]) with previously reported results from the Translational Genomics Research Institute and the Mayo AD GWAS. We identified 2708 single-nucleotide polymorphisms (SNPs) with $p < 10^{-6}$. In stage 2, we pooled results for these SNPs with the European AD Initiative (2032 cases and 5328 controls) to identify 38 SNPs (10 loci) with $p < 10^{-8}$. In stage 3, we combined data for these 10 loci with data from the Genetic and Environmental Risk in AD consortium (3133 cases and 6995 controls) to identify 4 SNPs with $p < 10^{-9}$. These 4 SNPs were replicated in an independent Spanish sample (1140 AD cases and 1209 controls). Genome-wide association analyses were completed in 2007-2008 and the meta-analyses and replication in 2009.

Main Outcome Measure  Presence of Alzheimer disease.

Results  Two loci were identified to have genome-wide significance for the first time: rs744373 near BIN1 (odds ratio [OR], 1.13; 95% confidence interval [CI], 1.06-1.21 per copy of the minor allele; $p = 1.59 \times 10^{-11}$) and rs597668 near EXOC3L2/BLOC15S/ MARK4 (OR, 1.18; 95% CI, 1.07-1.29; $p = 6.45 \times 10^{-9}$). Associations of these 2 loci plus the previously identified loci CLU and PICALM with AD were confirmed in the Spanish sample ($p < .05$). However, although CLU and PICALM were confirmed to be associated with AD in this independent sample, they did not improve the ability of a model that included age, sex, and APOE to predict incident AD (improvement in area under the receiver operating characteristic curve from 0.847 to 0.849 in the Rotterdam Study and 0.702 to 0.705 in the Cardiovascular Health Study).

Conclusions  Two genetic loci for AD were found for the first time to reach genome-wide statistical significance. These findings were replicated in an independent population. Two recently reported associations were also confirmed. These loci did not improve AD risk prediction. While not clinically useful, they may implicate biological pathways useful for future research.

Seshadri et al. JAMA 2010;303(18):1832-40, 5/12/10
Meta-analysis Confirms CR1, CLU, and PICALM as Alzheimer Disease Risk Loci and Reveals Interactions With APOE Genotypes

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Objectives: To determine whether genotypes at CLU, PICALM, and CR1 confer risk for Alzheimer disease (AD) and whether risk for AD associated with these genes is influenced by apolipoprotein E (APOE) genotypes.

Design: Association study of AD and CLU, PICALM, CR1, and APOE genotypes.

Setting: Academic research institutions in the United States, Canada, and Israel.

Participants: Seven thousand seventy cases with AD, 3055 with autopsies, and 8169 elderly cognitively normal controls, 1092 with autopsies, from 12 different studies, including white, African American, Israeli-Arab, and Caribbean Hispanic individuals.

Results: Unadjusted, CLU (odds ratio [OR], 0.91; 95% confidence interval [CI], 0.85-0.96 for single-nucleotide polymorphism [SNP] rs1136000), CR1 (OR, 1.14; 95% CI, 1.07-1.22; SNP rs3818361), and PICALM (OR, 0.89; 95% CI, 0.84-0.94, SNP rs3851179) were associated with AD in white individuals. None were significantly associated with AD in the other ethnic groups. APOE ε4 was significantly associated with AD (ORs, 1.80-9.05) in all but 1 small white cohort and in the Arab cohort. Adjusting for age, sex, and the presence of at least 1 APOE ε4 allele greatly reduced evidence for association with PICALM but not CR1 or CLU. Models with the main SNP effect, presence or absence of APOE ε4, and an interaction term showed significant interaction between presence or absence of APOE ε4 and PICALM.

Conclusions: We confirm in a completely independent data set that CR1, CLU, and PICALM are AD susceptibility loci in European ancestry populations. Genotypes at PICALM confer risk predominantly in APOE ε4-positive subjects. Thus, APOE and PICALM synergistically interact.

Arch Neurol. Published online August 9, 2010. doi:10.1001/archneurol.2010.201
Pathways:
Ab (pink)
Neurofibrillary tangles (blue)
Inflammation (green)
Atherosclerosis (yellow)
Synaptic dysfunction (purple)
Others (orange)
Alzheimer’s Disease Neuroimaging Initiative biomarkers as quantitative phenotypes: Genetics core aims, progress, and plans

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Publications using ADNI GWAS data (partial)

Hippocampal Atrophy as a Quantitative Trait in a Genome-Wide Association Study Identifying Novel Susceptibility Genes for Alzheimer’s Disease

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Abstract

**Background:** With the exception of APOE ε4 allele, the common genetic risk factors for sporadic Alzheimer’s Disease (AD) are unknown.

**Methods and Findings:** We completed a genome-wide association study on 381 participants in the ADNI (Alzheimer’s Disease Neuroimaging Initiative) study. Samples were genotyped using the Illumina Human610-Quad BeadChip. 516,645 unique Single Nucleotide Polymorphisms (SNPs) were included in the analysis following quality control measures. The genotype data and raw genetic data are freely available for download (LONI, http://www.loni.ucla.edu/ADNI/Dataset). Two analyses were completed: a standard case-control analysis, and a novel approach using hippocampal atrophy measured on MRI as an objectively defined, quantitative phenotype. A General Linear Model was applied to identify SNPs for which there was an interaction between the genotype and diagnosis on the quantitative trait. The case-control analysis identified APOE and the new risk gene, TOMM40 (translocase of outer mitochondrial membrane 40), at a genome-wide significance level of <10^-6 (10^-11 for a haplotype). TOMM40 risk alleles were approximately twice as frequent in AD subjects as controls. The quantitative trait analysis identified 21 genes or chromosomal areas with at least one SNP with a p-value <10^-6, which can be considered potential “new” candidate loci to explore in the etiology of sporadic AD. These candidates included EFNA5, CAND1, MAGI2, ARSB, and PRUNE2, genes involved in the regulation of protein degradation, apoptosis, neuronal loss and neurodevelopment. Thus, we identified common genetic variants associated with the increased risk of developing AD in the ADNI cohort, and present publicly available genome-wide data. Supportive evidence based on case-control studies and biological plausibility by gene annotation is provided. Currently no available sample with both imaging and genetic data is available for replication.

**Conclusions:** Using hippocampal atrophy as a quantitative phenotype in a genome-wide scan, we have identified candidate risk genes for sporadic Alzheimer’s disease that merit further investigation.

A TOMM40 variable-length polymorphism predicts the age of late-onset Alzheimer's disease

The e4 allele of the apolipoprotein E (APOE) gene is currently the strongest and most highly replicated genetic factor for risk and age of onset of late-onset Alzheimer's disease (LOAD). Using phylogenetic analysis, we have identified a polymorphic poly-T variant, rs10524523, in the translocase of outer mitochondrial membrane 40 homolog (TOMM40) gene that provides greatly increased precision in the estimation of age of LOAD onset for APOE e3 carriers. In two independent clinical cohorts, longer lengths of rs10524523 are associated with a higher risk for LOAD. For APOE e3/4 patients who developed LOAD after 60 years of age, individuals with long poly-T repeats linked to APOE e3 develop LOAD on an average of 7 years earlier than individuals with shorter poly-T repeats linked to APOE e3 (70.5 ± 1.2 years versus 77.6 ± 2.1 years, P = 0.02, n = 34). Independent mutation events at rs10524523 that occurred during Caucasian evolution have given rise to multiple categories of poly-T length variants at this locus. On replication, these results will have clinical utility for predictive risk estimates for LOAD and for enabling clinical disease prevention studies. In addition, these results show the effective use of a phylogenetic approach for analysis of haplotypes of polymorphisms, including structural polymorphisms, which contribute to complex diseases.

Keywords: AD genetics; phylogenetic analysis; TOMM40; APOE; poly-T variants
Whole genome association study of brain-wide imaging phenotypes for identifying quantitative trait loci in MCI and AD: A study of the ADNI cohort

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ABSTRACT

A genome-wide, whole brain approach to investigate genetic effects on neuroimaging phenotypes for identifying quantitative trait loci is described. The Alzheimer’s Disease Neuroimaging Initiative 1.5 T MRI and genetic dataset was investigated using voxel-based morphometry (VBM) and Freesurfer parcellation followed by genome-wide association studies (GWAS). One hundred forty-two measures of grey matter (GM) density, volume, and cortical thickness were extracted from baseline scans. GWAS, using PLINK, were performed on each phenotype using quality-controlled genotype and scan data including 530,992 of 620,903 single nucleotide polymorphisms (SNPs) and 733 of 818 participants (175 AD, 354 amnestic mild cognitive impairment, MCI, and 204 healthy controls, HC). Hierarchical clustering and heat maps were used to analyze the GWAS results and associations are reported at two significance thresholds (p<10^-7 and p<10^-5). As expected, SNPs in the APOE and TOMM40 genes were confirmed as markers strongly associated with multiple brain regions. Other top SNPs were proximal to the EPHA4, TPS3 and NXP1 genes. Detailed image analyses of rs6403843 (flanking NXP1) revealed reduced global and regional GM density across diagnostic groups in T1 relative to CG homozygotes. Interaction analysis indicated that AD patients homozygous for the T allele showed differential vulnerability to right hippocampal GM density loss. NXP1 codes for a protein implicated in promotion of adhesion between dendrites and axons, a key factor in synaptic integrity, the loss of which is a hallmark of AD. A genome-wide, whole brain search strategy has the potential to reveal novel candidate genes and loci warranting further investigation and replication.
Methodological Overview

Shen et al 2010 [Epub], NeuroImage

Baseline MRI Scans

- FreeSurfer: 56 volume or cortical thickness measures
- VBM: 86 GM density measures

QC’ed genotyping data

142 QTs

530,992 SNPs

GWAS of Imaging Phenotypes

Strong associations represented by heat maps

GWAS of candidate QT

VBM of candidate SNP

Refined modeling of candidate association
GWAS of Mean Grey Matter Density: Right Hippocampus (Manhattan Plot)

Shen et al 2010  NeuroImage
NXPH1: Novel Candidate Gene for AD that codes for neurexophilin-1 protein

Expressed in Brain

Neuroexphilin-1’s likely role in synaptogenesis:

Protein forms a very tight complex with alpha neurexins, a group of proteins that promote adhesion between dendrites and axons (EntrezGene).
Voxelwise GWAS: Ran genome-wide association for a quarter of a million points across 700 subjects - new gene discovery method; many new SNPs; power calculations for replication (Stein et al, NeuroImage, 2010a)

GRIN2b, a common glutamate receptor genetic variant, is associated with greater temporal lobe atrophy and with AD; NMDA-receptor is a target for memantine therapy (Stein et al, NeuroImage, 2010b)

FTO, an obesity risk gene carried by 46% of Europeans, is associated with 10-15% frontal and occipital atrophy, and with a ~1.7kg weight gain, on average (April Ho et al, PNAS, 2010)
Genome-wide association study of CSF biomarkers $A\beta_{1-42}$, t-tau, and p-tau$_{181p}$ in the ADNI cohort

**ABSTRACT**

**Objectives:** CSF levels of $A\beta_{1-42}$, t-tau, and p-tau$_{181p}$ are potential early diagnostic markers for probable Alzheimer disease (AD). The influence of genetic variation on these markers has been investigated for candidate genes but not on a genome-wide basis. We report a genome-wide association study (GWAS) of CSF biomarkers ($A\beta_{1-42}$, t-tau, p-tau$_{181p}$, p-tau$_{181p}$/$A\beta_{1-42}$, and t-tau/$A\beta_{1-42}$).

**Methods:** A total of 374 non-Hispanic Caucasian participants in the Alzheimer's Disease Neuroimaging Initiative cohort with quality-controlled CSF and genotype data were included in this analysis. The main effect of single nucleotide polymorphisms (SNPs) under an additive genetic model was assessed on each of 5 CSF biomarkers. $p$ Values of all SNPs for each CSF biomarker were adjusted for multiple comparisons by the Bonferroni method. We focused on SNPs with corrected $p < 0.01$ (uncorrected $p < 3.10 \times 10^{-8}$) and secondarily examined SNPs with uncorrected $p$ values less than $10^{-5}$ to identify potential candidates.

**Results:** Four SNPs in the regions of the APOE, LOC100129500, TOMM40, and EPC2 genes reached genome-wide significance for associations with one or more CSF biomarkers. SNPs in CCDC134, ABCG2, SREBF2, and NFATC4, although not reaching genome-wide significance, were identified as potential candidates.

**Conclusions:** In addition to known candidate genes, APOE and TOMM40 and one hypothetical gene LOC100129500 partially overlapping APOE; one novel gene, EPC2; and several other interesting genes were associated with CSF biomarkers that are related to AD. These findings, especially the new EPC2 results, require replication in independent cohorts.
Baseline Abeta1-42 CSF Level by Diagnosis Group and TOMM40 (rs2075650) Genotype

N = 377
AD+MCIc: 60 AA, 62 AG, 14 GG
MCI-S: 75 AA, 55 AG, 12 GG
HC: 77 AA, 22 AG, 1 GG

Kim, Swaminathan, et al Neurology (Jan 4 2011)
CSF Biomarkers: Total Tau

Figure 1

Manhattan plot (A) and quantile-quantile plot (B) of total tau

3.1 x 10^{-8} corrected p < .01

EPC2 (rs4499362)

Kim, Swaminathan, et al. *Neurology* (Jan 4 2011)
New finding from CSF t-tau GWAS: Enhancer of polycomb homolog 2 (EPC2)

Kim, Swaminathan, et al *Neurology* (Jan 4 2011)
Enhancer of polycomb homolog 2 (EPC2)

- Multiple SNPs were associated with t-tau at $p<10^{-6}$
- Involved in formation of heterochromatin (Doyon et al. 2004) & Microdeletion syndrome of 2q23.1 (mental retardation, short stature & epilepsy (van Bon et al. 2010))

![Raw Expression Level of EPC2 in Hippocampus](http://human.brain-map.org/mri_viewer.html?probes=1045018,1045019)
Genome Wide Association Study (GWAS) on Annual Percent Change of 1.5T MRI: Initial Data

• 818 ADNI Subjects
  – 589 cases (MCI or AD), 229 controls
  – 476 males, 342 females

• 620901 +2 Markers
  – 620901 from Illumina 610 Quad array
  – 2 APOE SNPs

• Extensive QC protocol

Saykin et al (ICAD 2009)
Annual Percent Change in Hippocampal Volume and Grey Matter Density (ADNI Cohort)

Covaried for baseline age, sex, education, handedness & ICV

Role of APOE in Rate of Change

Main Effect versus Interaction

Adjusted Annual Percent Change in Hippocampal Volume

APOE (Chr 19): rs429358 is the epsilon 4 allele marker
TOMM40 (Chr 19): translocase of outer mitochondrial membrane 40 homolog (LD with APOE)
CADH8 (Chr 16): cadherin 8, type 2; synaptic adhesion, axonal growth/guidance (no data in AD)

Saykin et al Alzheimers’s & Dementia (2010); 6:265–273
Adjusted Annual Percent Change in Hippocampal Gray Matter Density

APOE (Chr 19): rs429358 is the epsilon 4 allele marker / TOMM40 in LD with APOE
MAD2L2 (Chr 1) mitotic arrest deficient-like 2 (mitotic spindle assembly)
LOC728574 (Chr 22): similar to retinitis pigmentosa GTPase regulator isoform C

Saykin et al Alzheimer’s & Dementia (2010); 6:265–273
Earlier Detection than MCI?
“Pre-MCI” Imaging & Clinical Status

Older adults with cognitive complaints show brain atrophy similar to that of amnestic MCI

A.J. Saykin, PsyD; H.A. Wishart, PhD; L.A. Rabin, PhD; R.B. Santulli, MD; L.A. Flashman, PhD;
J.D. West, MS; T.L. McHugh, MA; and A.C. Mamourian, MD

Abstract—Objective: To examine the neural basis of cognitive complaints in healthy older adults in the absence of memory impairment and to determine whether there are medial temporal lobe (MTL) gray matter (GM) changes as reported in Alzheimer disease (AD) and amnestic mild cognitive impairment (MCI). Methods: Participants were 40 euthymic individuals with cognitive complaints (CCs) who had normal neuropsychological test performance. The authors compared their structural brain MRI scans to those of 40 patients with amnestic MCI and 40 healthy controls (HCs) using voxel-based morphometry and hippocampal volume analysis. Results: The CC and MCI groups showed similar patterns of decreased GM relative to the HC group on whole brain analysis, with differences evident in the MTL, frontotemporal, and other neocortical regions. The degree of GM loss was associated with extent of both memory complaints and performance deficits. Manually segmented hippocampal volumes, adjusted for age and intracranial volume, were significantly reduced only in the MCI group, with the CC group showing an intermediate level. Conclusions: Cognitive complaints in older adults may indicate underlying neurodegenerative changes even when unaccompanied by deficits on formal testing. The cognitive complaint group may represent a pre–mild cognitive impairment stage and may provide an earlier therapeutic opportunity than mild cognitive impairment. MRI analysis approaches incorporating signal intensity may have greater sensitivity in early preclinical stages than volumetric methods.

NEUROLOGY 2006;67:834–842
Baseline Hippocampal Gray Matter Density in MCI & Cognitive Complaints

*N=40,40,40

Saykin et al, NEUROLOGY 2006;67:834-842

\[ \text{Gray Matter Density} \]

Left Hippocampal GM Density

Right Hippocampal GM Density

\[ \text{Gray Matter Density} \]

- * MCI < HC, \( p < .001 \)
- * CC < HC, \( p < .005 \)
3.3K SNP Targeted Neurogenetics Array

Ensembl v38 - Apr 2006  http://www.ensembl.org/Homo_sapiens/karyoview
Genetic Pathway-Based Hierarchical Clustering Analysis of Older Adults With Cognitive Complaints and Amnestic Mild Cognitive Impairment Using Clinical and Neuroimaging Phenotypes

Chantel D. Sloan,1 Li Shen,2,3 John D. West,2 Heather A. Wishart,4 Laura A. Flashman,4 Laura A. Rabin,4 Robert B. Santulli,4 Stephen J. Guerin,4 C. Harker Rhodes,5 Gregory J. Tsongalis,5 Thomas W. McAllister,4 Tim A. Ahles,6 Stephen L. Lee,7 Jason H. Moore,1,8,9,10 and Andrew J. Saykin2,4,11, *
# Cluster Results: Role of Genes

## TABLE II. The Hypothesized Role of Each Gene Found in One of the Three Clusters on AD and MCI Based on a Literature Search With Sample References

<table>
<thead>
<tr>
<th>Gene (alias)</th>
<th>Suggested role in AD/MCI</th>
</tr>
</thead>
<tbody>
<tr>
<td>ABCB1</td>
<td>Regulates beta-amyloid levels [Lam et al., 2001; Kuhnke et al., 2007]</td>
</tr>
<tr>
<td>APBA1 [MINT1, X11]</td>
<td>Binds APP and affects cleavage and translocation [Miller et al., 2006; Ho et al., 2008; Saito et al., 2008] [also known as MINT1, X11]</td>
</tr>
<tr>
<td>BACE1</td>
<td>Cleaves APP [Haass, 2004; McConlogue et al., 2007; Willem et al., 2009]</td>
</tr>
<tr>
<td>BACE2</td>
<td>Cleaves APP, a BACE1 homolog [Stockley and O’Neill, 2007], increases IL-1R2 secretion [Kuhn et al., 2007]</td>
</tr>
<tr>
<td>BCL2</td>
<td>Induces apoptosis [Lu et al., 2005]</td>
</tr>
<tr>
<td>BCL2L1 [BCL-X]</td>
<td>Anti-apoptotic signaling [Lukiw and Bazan, 2006; Shimohama, 2009]</td>
</tr>
<tr>
<td>CASP7</td>
<td>Apoptosis regulator, neuron loss in AD [Pompl et al., 2003; Matsui et al., 2006]</td>
</tr>
<tr>
<td>CHAT</td>
<td>Synthesizes acetylcholine, which is depleted in AD [Burgess et al., 2009], ChAT fibers increasingly immunoreactive in AD, and MCI [Cuello et al., 2007]</td>
</tr>
<tr>
<td>CST3</td>
<td>Studies show mixed results, colocalizes with beta-amyloid [Lin et al., 2003; Monastero et al., 2005; Nacmias et al., 2006]</td>
</tr>
<tr>
<td>DRD3</td>
<td>Associated with depression symptoms that co-occur with AD associated [Serretti et al., 2007]</td>
</tr>
<tr>
<td>DRD5</td>
<td>Connection to AD uncertain, normally functions as dopaminergic receptor [Cosentino et al., 2009]</td>
</tr>
<tr>
<td>IL6</td>
<td>Inflammatory response, tau phosphorylation [Papassotiropoulos et al., 2001; Quintanilla et al., 2004]</td>
</tr>
<tr>
<td>LRP1</td>
<td>Involved in APP processing and trafficking [Waldron et al., 2008; Yamada et al., 2008]</td>
</tr>
<tr>
<td>NAT1</td>
<td>Folate metabolism [Johnson et al., 2004]</td>
</tr>
<tr>
<td>PSEN2</td>
<td>Gamma-secretase complex formation with PSEN1, well-established AD susceptibility gene [Bertram and Tanzi, 2008; Bertram, 2009; Marcon et al., 2009]</td>
</tr>
</tbody>
</table>

Amyloid Pathway PET Study: [11C]PiB

Gene and SNP selection

Search “amyloid” on Gene Ontology

Genes in the AlzGene database

Common genes

SNPs located in these genes

Quality control:
1. Sample & SNP call rate > 0.90
2. Minor allele frequency > 0.20

274 SNPs in 15 genes

Gene-based association analysis

Dominant model; LD $r^2 = 0.5; p = 0.05$

Whole-brain voxel-wise analysis

Quantitative Phenotype

Standardized uptake value ratio (SUVR)

1. Anterior cingulate
2. Frontal cortex
3. Parietal cortex
4. Precuneus

Average SUVR

Covariates:
1. Age
2. Gender
3. Diagnosis at scan
4. APOE ε4 status

Swaminathan et al, ASHG, Wash DC, Nov 2010
Amyloid Gene Pathway-PiB: Preliminary Results

- **Gene-based association analysis:**
  - *DHCR24* significantly associated with AVG ($p=0.012$)
  - One SNP (rs7551288) in gene found to be significant
  - Dominant effect of minor allele

- **Whole-brain voxel-wise-analysis:**
  - Increased PiB uptake in frontal regions
  - Frontal cortex known to have increased PiB uptake in AD patients

- **DHCR24 gene:**
  - 24-dehydrocholesterol reductase - enzyme that synthesizes cholesterol from desmosterol (Peri et al. *J Mol Endocrinol*, 2008)
  - AKA: *seladin-1* or *selective AD indicator-1*
  - Reduced expression in temporal cortex in AD
  - Neuroprotective role
    - Confers resistance against Aβ and oxidative stress-induced apoptosis
    - Possible mediator of neuroprotective effects of estrogens/SERMs.

*Swaminathan et al, ASHG, Wash DC, Nov 2010*
Future Directions: ADNI Phenotype Pathways

GWAS or Candidate Signals as of mid 2010

Adapted from Saykin et al Alzheimer’s & Dementia (2010); 6:265-273
Biomarkers are needed for early diagnosis, to predict transitions from NCI to MCI to AD and clinical trials of disease modifying therapies

Shaw LM, Korecka M, Clark CM, Lee VM.-Y, Trojanowski JQ. 
ADNI Genetics: Next Steps

• New DNA, RNA, cell line sample collection
• ADNI-1 data analysis is the major focus:
  – Baseline and rate of change
  – Genotype and copy number variation
  – Genome wide and candidate approaches
  – Pathway based analyses
  – Associations with other variables & biomarkers
  – Collaborative projects & other cohorts

• Future:
  – Targeted DNA and RNA resequencing – identify key regions for intensive scrutiny
  – Epistasis, Transcriptomics/expression, microRNA
  – Epigenomics (DNA methylation, etc)
ADNI Genotyping Working Group

Indiana University
• Imaging Genomics Lab
  – Andrew J. Saykin
  – Li Shen
  – Mark Inlow
  – Sungeun Kim
  – Kwangsik Nho
  – Shannon Risacher
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• Jason Moore (Dartmouth)
• Paul Thompson (UCLA)
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• Foundation for the NIH
  – Anonymous Foundation (Challenge Grant)
  – Gene Network Sciences
  – Merck
  – Pfizer (DNA extraction)
• Alzheimer’s Association
• Indiana Economic Development Corporation (IEDC)
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