Genetic comparison of symptomatic and asymptomatic persons with Alzheimer disease neuropathology

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Abstract

Objective—To determine whether symptomatic and asymptomatic persons with Alzheimer's disease (AD) neuropathology have different allele counts for single-nucleotide polymorphisms (SNPs) that have been associated with clinical late-onset AD (LOAD).

Methods—Data came from the National Alzheimer's Coordinating Center Uniform Data Set and Neuropathology Data Set, and the Alzheimer's Disease Genetics Consortium (ADGC). Participants had low to high AD neuropathologic change. The 22 known/suspected genes associated with LOAD were considered. "Symptomatic" was defined as Clinical Dementia Rating global score >0.

Results—68 asymptomatic and 521 symptomatic participants met inclusion criteria. SNPs associated with *ABCA7* (odds ratio [OR] = 1.66; 95% confidence interval [CI] = 1.03-2.85) and *MAPT* (OR = 2.18; CI = 1.26-3.77) were associated with symptomatic status. In stratified analyses, loci containing *CD2AP* (OR = 0.35; 95% CI = 0.16-0.74), *ZCWPW1* (OR = 2.98; 95% CI = 1.34-6.86), and *MAPT* (OR = 3.73, 95% CI = 1.30-11.76) were associated with symptomatic status in *APOE* e4 carriers.

Conclusions—These findings potentially explain some of the variation in whether a person with AD neuropathology expresses symptoms. Understanding why some people remain cognitively

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normal despite having AD neuropathology could identify pathways to disease heterogeneity and guide treatment trials.

Introduction

The association between apolipoprotein E (*APOE*) and late-onset AD (LOAD) is well-documented.[1,2] Associations between LOAD and other genetic loci are increasingly recognized: to date,22 loci have been associated with risk of LOAD.[3–5] Most studies documenting these associations compared genetic profiles of clinically-diagnosed cases of dementia or mild cognitive impairment (MCI) with cognitively normal controls.[3,4,6–8] Recently, several studies assessed the association between the above-noted loci and AD neuropathology determined at autopsy, finding associations between 14 of the 22 genes and extent of neuropathologic change.[9–12]

In addition to increasing the extent of neuropathology, other genetic pathways might exist, including varying degrees of expression of the same extent of AD neuropathology. *APOE* (ε4 carrier vs. non-carrier) has been shown to be associated with increased risk of dementia in people with underlying AD neuropathology, even after adjustment for extent of pathology. The presence of the *APOE*ε4 allele might thus be associated with pathologic processes not assessed at autopsy, such as greater levels of toxic Aβ oligomers.[13–15]

Potential relationships between clinical expression of AD neuropathology and other (non-*APOE*) loci associated with LOAD have not been adequately explored. We thus sought to determine whether symptomatic and asymptomatic persons with AD neuropathology have different allele counts for loci that have been associated with clinical AD.

Methods

Study sample

Individuals in this study were research participants assessed with the Uniform Data Set (UDS) evaluation at one of 34 past and present National Institute on Aging/NIH Alzheimer's Disease Centers (NIA ADCs) and whose data were submitted to the National Alzheimer's Coordinating Center (NACC) between 2005 and 2014. The UDS comprises data on demographics, health history, neuropsychological tests, and a complete neurological exam, for participants with normal cognition, MCI, and dementia. Participants with normal cognition are volunteers who are followed with the same study procedures as symptomatic participants.[16] Neuropathology data are available for a subset of UDS participants who consent to autopsy. Genetic data were obtained from the Alzheimer's Disease Genetics Consortium (ADGC), which houses genotype data for UDS participants and other cohorts (https://alios.med.upenn.edu/adgc).

Standard Protocol Approvals, Registrations, and Patient Consents

All participants provided written informed consent. The University of Washington IRB approved research using the NACC database.

Defining neuropathologic AD

A low, medium, or high level of AD neuropathologic change according to modified NIA-Alzheimer's Association (NIA-AA) ABC criteria [17,18] was required of all participants. The ABC score comprises three criteria. Two of these (B score for Braak stage for neurofibrillary tangles[19] and C score for CERAD neuritic plaque frequency[20]) are recorded on all versions of the NACC Neuropathology Form. However, A score (Thal phase[21] for Aβ plaques) was not included on the Form before version 10 (implemented 2014), and thus is not available for most NACC participants. In order to include participants with an early form of Aβ plaque formation, we included those with "diffuse plaque," defined as plaques with no apparent dystrophic neurites, as detected by silver impregnation methods, ubiquitin, tau immunohistochemistry, or Aβ immunohistochemistry.

Participants with sparse, moderate, or frequent diffuse plaques were considered to have a Thal Aβ plaque phase of 1 or higher and thus met inclusion criteria for this study. Similarly, participants with sparse, moderate, or frequent neuritic plaques had a neuritic plaque C score of 1 or higher and met inclusion criteria. Limiting the sample to participants with either neuritic or diffuse plaques approximates to inclusion of all participants who meet NIA-AA criteria for low to high AD neuropathologic change. The resulting study sample included only participants with Aβ plaques, regardless of clinical diagnosis.[13,22]

We excluded participants with a primary neuropathologic diagnosis of dementia with Lewy bodies (DLB). This data element is available in versions 1-9 of the Neuropathology Form. For the few participants assessed with version 10, we performed a conservative exclusion, removing those for whom any Lewy bodies were reported.

Defining asymptomatic AD

Symptoms were defined using the Clinical Dementia Rating global score (CDR), an instrument that grades cognitive and functional abilities.[23] Participants with CDR global score of 0 at their last clinical assessment were defined as having normal cognition, and formed the asymptomatic group. Participants with CDR global score of 0.5 or higher were defined as having clinical characteristics consistent with MCI or dementia and formed the symptomatic group.[13,22] In order to best correlate symptoms and neuropathologic features, we limited the analytic sample to participants who died within one year of the last UDS clinical assessment.

Genetic data

Genetic data were obtained from ADGC. These data were drawn from blood or brain tissue samples sent by individual ADCs. Imputation was used to generate a common set of single-nucleotide polymorphisms (SNPs). Imputations with probability ≥ .90 were included; imputations below this threshold were considered missing data. Uniform stringent quality control measures were applied to remove low-quality and duplicate samples and problematic SNPs. Data were transferred from PLINK format to Excel for additional quality assurance and statistical analysis.

All participants had missing data for no more than four SNPs. Three SNPs (in loci containing *BIN1*, *CLU*, and *MAPT*) had missing data for >10% of participants meaning the SNP was not genotyped and there was no proxy with good quality data to advise imputation. Analyses were performed on available data but interpreted with caution for these SNPs. For the SNP associated with *DSG2*, analysis performed in *APOE* ε4 carriers was not possible due to inadequate variation in allele frequency. Of the 593 participants meeting study inclusion criteria, the 589 of European decent were retained for analysis to decrease potential effects of population stratification.

Data for this study included allelic count for 22 genes known or suspected to be associated with LOAD.[3–5] All SNPs satisfied the Hardy-Weinberg equilibrium at the 0.001 alpha level.

Statistical analysis

Descriptive statistics (frequencies, percentages, Chi-square tests, Fisher's Exact tests) were calculated for demographic and neuropathologic features for asymptomatic and symptomatic participants. For each SNP, the risk allele was that allele (major or minor) associated with higher odds of AD in the literature.[4,5] For robustness to model misspecification, we assume the conventional additive mode of inheritance and use the number of risk alleles (0, 1, or 2) as the genetic predictor variable.[24] We then applied a logistic model where symptomatic AD status was the dependent variable. This model was fit for each SNP individually. In order to account for potential differences between *APOE* ε4 carriers and non-carriers, we performed stratified analyses. All models were adjusted for sex and age at death (continuous). Additional adjustment for years of education returned nearly identical ORs and CIs (results not shown) as those models adjusted for sex and age at death alone. However, including education in the models required dropping six subjects with missing data. Hence the main results presented herein are for the analyses adjusted for sex and age at death.

A risk score was calculated by multiplying the number of risk alleles by the corresponding log-transformed odds ratio from the individual models and summed across all 22 SNPs,[6] across the 21 SNPs with <15% missing data (*MAPT* excluded), and across the 19 SNPs with <10% missing data (*BIN1, CLU*, and *MAPT* excluded). The risk score was calculated for the entire sample and calculated separately for *APOE* ε4 carriers and non-carriers. The risk score was then included in a logistic regression model adjusted for sex and age at death.

Each gene was tested separately for association with symptomatic status and an alpha level of 0.05 was considered as significant in all tests of statistical significance.

All analyses were performed in R version 3.1.1.

Results

At the time of data abstraction, there were 1,127 UDS participants with genotype data and neuropathology data available. Of these, 999 met criteria for AD neuropathologic changes. An additional 77 participants were excluded for Lewy body pathology and 329 were excluded for not having a clinical examination within one year of death. Excluding the four African American and multiracial participants resulted in a sample size of 589 participants,

68 asymptomatic and 521 symptomatic participants. Demographic and neuropathologic features are shown in Table 1 and supplemental e-Table 1. Notably, symptomatic participants were slightly younger than asymptomatic participants and more often had at least one *APOE* ε4 allele. They also had more advanced AD pathology (neuritic plaques and Braak stage for neurofibrillary tangles), as well as more arteriolosclerosis, Lewy bodies, and cerebral amyloid angiopathy.

In the adjusted logistic regression model assuming an additive model of inheritance, the SNP associated with the gene *ABCA7* met statistical significance at the 0.05 alpha level (OR=1.66; 95% CI: 1.03-2.85; *p*=0.049, as did the SNP associated with gene *MAPT* (OR=2.18; CI=1.26-3.77; *p*=0.005) for association with symptomatic status.

In analyses stratified by *APOE* ε4 carrier status, the SNPs linked to *CD2AP, ZCWPWI,* and *MAPT* were associated with odds of symptomatic status in ε4 carriers (OR=0.35, 2.98, and 3.73; 95% CI: 0.16-0.74, 1.34-6.86, and 1.30-11.76; *p*=0.007, 0.008, and 0.017.respectively). No significant findings were observed in non-carriers. Full results for each of the 22 SNPS are presented in Table 2.Each of the SNPs was tested for interactions with APOE. Only two of the SNPS had significant interactions: CD2AP (OR=0.38; 95% CI: 0.16-0.92; *p*=0.03) and ZCWPW1 (OR=2.93; 95% CI: 1.17-7.57; *p*=0.02).

The genetic risk score was associated with increased odds of being symptomatic in all participants (OR=2.61; 95% CI: 1.61-4.36; *p*=. 0002) and among *APOE* ε4 carriers and non-carriers when the score included the 19 SNPs with mostly complete data. When all SNPs, except MAPT, which was missing for approximately 40% of participants, were considered, the risk score remained significant. For both contingencies (19 or 21 SNPs in risk score), the effect size was higher among *APOE* ε4 carriers. When all 22 SNPs (including MAPT) were included, the risk score was still significant in all participants, but sample size was too small for meaningful conclusions on APOE ε4 strata (Table 3).

Sensitivity analysis

The main analysis showed the association with outcome for each SNP, adjusted for age and sex. A sensitivity analysis added adjustment for the following neuropathologic features: vascular disease, Braak stage, Lewy bodies, and cerebral amyloid angiopathy. One of the SNPs that had been significantly associated with symptomatic status (MAPT) remained significantly associated. One of the SNPs (CD2AP) that was significantly associated with symptomatic status only in the *APOE* ε4- positive strata became significant in the entire group. We also undertook a principal component analysis (PCA). Both SNPs that were associated with symptomatic status in the main analysis (ABCA7 and MAPT) remained significant in the additional PCA (supplemental e-Table 2).Finally, we created an additional genetic risk score based on published ORs of developing AD. This was also significantly associated with development of symptoms (supplemental e-Table 3). However, it should be emphasized that these published ORs were derived based on risk of developing AD, whereas the genetic risk score in Table 3 were derived based on risk of expressing symptoms once someone already has AD neuropathology, which is a different concept.

Discussion

We sought to determine whether cognitively symptomatic persons with AD neuropathologic change and asymptomatic persons with AD neuropathologic change have different allele counts for loci that have been previously associated with clinical AD. This potential association has not been well addressed. We found that *ABCA7* and *MAPT* were significantly associated with expression of symptoms. The loci containing *CD2AP* and *ZCWPW1* were significantly associated with symptoms, but only in the *APOE* ε4 positive strata.

The association of *ABCA7* with altered risk of clinical AD has been well documented.[1,2,25] Similarly to *APOE*, *ABCA7* is involved with cholesterol and lipid metabolism.[26] It is also involved with immune function.[2] It is not certain whether *ABCA7*'s effect on altering AD risk is through its effects on immune function, lipid metabolism, or both.[2] *ABCA7* has also been postulated to influence AD risk by clearing Aβ aggregates.[27,28]

For *CD2AP* and *ZCWPW1*, there are multiple genes within the loci that have been associated with increased LOAD risk.[1,2,4,9] Hence, it would be premature to postulate on potential biologic mechanisms of action, except to note that alterations in *CD2AP* have been associated with increased neuritic plaque burden in brains already having AD.[1,9] It has also been postulated to be involved with modulating Aβ clearance and suppression of Aβ toxicity.[2]

MAPT, the gene encoding tau, was known to be associated with Parkinson's disease, progressive supranuclear palsy, and corticobasal degeneration[29] but has only recently been shown to be associated with AD. It is possible that there are different genes at this locus that account for separate effects on AD and Parkinson's disease.[5]

There are several possible pathways by which *ABCA7, CD2AP*, *MAPT*, and *ZCWPW1* might influence risk of expression of symptoms in people with underlying AD neuropathology. These pathways can be considered in the three categories that have been postulated as mechanisms by which LOAD susceptibility alleles might affect the risk of clinical AD: 1) Aβ deposition; 2) downstream pathologic effects such as synaptic loss and neuronal death; and 3) other mechanisms, not related to AD, that contribute to cognitive change.[9]

First, *ABCA7* and *CD2AP* have been associated with increased neuritic plaque burden.[9] In the current study, although all persons met a minimum threshold for AD neuropathology, the symptomatic group had more frequent neuritic plaques. Hence, the effect the loci identified in this study on symptoms might be through known existing pathways of increasing extent of AD neuropathology.

Second, the above-noted clearance functions of *ABCA7* and *CD2AP* might also involve toxic Aβ oligomers. Also, *CD2AP* has been noted to have a direct effect in suppressing Aβ toxicity.[2] Finally, *CD2AP* is involved in synapse formation and thus may have pathologic effects downstream to AD neuropathology.[1,30] These effects of *ABCA7* and *CD2AP* might work through increasing the extent of neurofibrillary tangles and thus the Braak NFT stage.

MAPT has likewise been shown to be involved with several tauopathies and might exert similar effects.[5] Abnormal tau and amyloid deposits may interact synergistically to cause AD,[31] perhaps through the abnormal tau upregulating the neurotoxicity of Aβ or through intensifying the mitochondrial damage caused by Aβ.[32,33]

Third, *ABCA7* is involved with cholesterol and lipid metabolism. The symptomatic group had a higher proportion of cerebrovascular disease, which may have contributed to their higher odds of expressing cognitive symptoms. In addition to *APOE* ε4's effect on AD neuropathology, *APOE* ε4 carriers have increased risk of vascular dementia.[34] While similar effects have not been demonstrated in humans for *ABCA7*, alterations in risk of vascular disease associated with alterations in *ABCA7* function have been noted in mouse models.[35]

Finally, a logical pathway through which genes might influence risk of cognitive symptoms among persons who already have AD neuropathology is through immune response. *ABCA7* does have immune functions in addition to its effect on lipid metabolism.[1,2] *ZCWPW1* has been found to be related to expression of PILRB, a microglia expressed gene tied to neuroinflammation.[36] However, none of the other genes thought to affect AD risk through immune response (*CR1, CD33, MS4A, CLU, EPHA1*)[1] had significant associations in the current study.

Several findings from the current study do not have ready explanations. The loci containing *CD2AP* and *ZCWPW1* have significant effects on expression of symptoms, but only in the *APOE* ε4-positive strata. For *CD2AP* that effect is in the opposite direction of the effect found in the existing literature.[3,4,6–8] It is not surprising that some SNPs might have effects in the opposite direction from that reported in the literature. ORs reported from the literature assess risk of developing AD, whereas the current study assesses a different characteristic, expression of cognitive symptoms once someone already has underlying AD neuropathology. Similarly, the effect of the genetic risk score was more pronounced among people with *APOE* ε4-positive status. One prior study found a stronger association with AD for *CR1* and *CLU* in *APOE* ε4-positive strata and for the *MSA4* gene cluster in *APOE* ε4-negative strata.[37] Another study found an increased risk of clinical AD for carriers of MAPT H1 haplotype, but only in *APOE* ε4-negative strata.[38] The current study found changes in MAPT to be associated with altered risk of cognitive symptoms, primarily driven by altered risk in the *APOE* ε4-positive strata.

These findings can also be compared to the literature on genome-wide association studies (GWAS) of AD neuropathology. In a GWAS meta-analysis of demented participants with moderate to high AD neuropathology vs. non-demented participants with no or low AD neuropathology, the authors found an association with *ABCA7*. The strength of that association (OR=1.24-1.32) was higher than previously reported (OR=1.15). The value from the current study (OR=1.66) is higher but has 95% CIs that encompass both of the prior estimates.[4,12]

Before drawing conclusions, the study limitations must be addressed. First, the sample size was limited, especially in the asymptomatic group. Although significant associations were detected in several loci, other associations might not have been detectable.

Second, 70% of participants were aged 80 years or older. Several loci associated with LOAD have been associated with earlier onset of AD, including *APOE*, *CR1, BIN1*, and *PICALM*.[3] The ability of our study to detect differences might be diminished by the narrow age range studied.

Third, many participants who met clinical and neuropathologic criteria did not have available genetic data. Since both clinical AD cases and controls were selected by ADGC for study, the authors are not aware of a mechanism of selecting participants that would have influenced our results.

Fourth, the sample of subjects used in calculating the risk score was the same as the set used to determine the ORs. While this approach is not ideal, it is sufficient for a priori hypothesis testing. Future work building risk score prediction models should employ a different sample to validate and expand on these findings.

We found differences in several SNPs (rs4147929, rs10948363, rs1476679, and rs393152; corresponding to *ABCA7* and loci containing *CD2AP*, *ZCWPW1*, and *MAPT*, respectively) between symptomatic and asymptomatic persons, all of whom had AD neuropathology. These findings potentially explain some of the variation in whether a person with AD neuropathology expresses symptoms. Understanding why some people remain cognitively normal despite having AD neuropathology could identify pathways to disease heterogeneity and guide treatment trials.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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Table 1

Participant demographics, clinical and neuropathologic characteristics

	Asymptomatic AD (n=68)	Symptomatic AD (n=521)	p-value from Chi-square test[g]
Age at death			0.41
<60	0 (0%)	8 (1%)	
60-69	3 (4%)	53 (10%)	
70-79	14 (21%)	99 (19%)	
80-89	27 (40%)	217 (42%)	
90+	24 (35%)	144 (28%)	
Sex			0.11
Female	30 (44%)	287 (55%)	
Male	38 (56%)	234 (45%)	
Education[a]			0.20
No college	13 (19%)	149 (29%)	
1 - 4 years of college	31 (46%)	221 (43%)	
At least some graduate school	24 (35%)	145 (28%)	
APOE			<0.0001
Non-carrier (0 ε4 alleles)	52 (76%)	245 (47%)	
Heterozygous (1 ε4 allele)	16 (24%)	226 (43%)	
Homozygous (2 ε4 alleles)	0 (0%)	50 (10%)	
Braak stage			<0.0001
0	1 (1%)	4 (1%)	
I-II	34 (50%)	44 (8%)	
III-IV	27 (40%)	126 (24%)	
V-VI	6 (9%)	347 (67%)	
CERAD neuritic plaque frequency			<0.0001
None	12 (18%)	16 (3%)	
Sparse	24 (35%)	69 (13%)	
Moderate	18 (26%)	134 (26%)	
Frequent	14 (21%)	302 (58%)	
Diffuse plaque frequency[b]			<0.0001
None	1 (2%)	10 (2%)	
Sparse	23 (41%)	61 (13%)	
Moderate	13 (23%)	91 (19%)	
Frequent	19 (34%)	318 (66%)	
Infarcts or lacunes[c]			0.91
Not Present	50 (74%)	390 (75%)	
Present	18 (26%)	130 (25%)	
Hemorrhorages and microbleeds			>0.999

	Asymptomatic AD (n=68)	Symptomatic AD (n=521)	p-value from Chi-square test[g]
Not Present	64 (94%)	490 (94%)	
Present	4 (6%)	31 (6%)	
Arteriosclerosis[d]			0.04
None	8 (14%)	66 (15%)	
Mild	31 (55%)	165 (37%)	
Moderate	14 (25%)	141 (31%)	
Severe	3 (6%)	79 (17%)	
Lewy body pathology[e]			0.002
Not Present	63 (93%)	385 (75%)	
Present	5 (7%)	131 (25%)	
Cerebral amyloid angiopathy[f]			0.0003
Not Present	56 (85%)	312 (61%)	
Present	10 (15%)	196 (39%)	

[a] 6 symptomatic participants missing information on years of education

[b] 12 asymptomatic and 41 symptomatic participants were missing information on diffuse plaque frequency

[c] 1 symptomatic participant was missing information on infarcts and lacunes

[d] 12 asymptomatic and 70 symptomatic participants were missing information on arteriosclerosis

[e] 5 symptomatic participants were missing information on Lewy body pathology

[f] 2 asymptomatic and 13 symptomatic participants were missing information on cerebral amyloid angiopathy

[g] Fisher's Exact test performed for age at death, *APOE* ε4, Braak stage, diffuse plaques, hemorrhages and microbleeds, and arteriosclerosis

Table 2

Odds ratio (adjusted for age and sex) for symptomatic AD vs. asymptomatic AD for each SNP

Gene	SNP	Risk allele	OR from IGAP	OR (95% CI)	OR (95% CI) for APOE e4 carriers (1 or 2 e4 alleles)	OR (95% CI) for APOE e4 non-carriers (0 e4 alleles)
ABCA7	rs4147929	A	1.15	**1.66 (1.03,2.85)**	1.25 (0.55,3.34)	1.81 (1.00,3.57)
BIN1	rs6733839	T	1.22	0.96 (0.63,1.47)	1.02 (0.42,2.67)	1.06 (0.65,1.76)
CASS4	rs7274581	T	0.88a	1.12 (0.59,1.97)	1.53 (0.46,3.98)	1.03 (0.46,2.09)
CD2AP	rs10948363	G	1.10	0.73 (0.50,1.09)	**0.35 (0.16,0.74)**	0.99 (0.62,1.62)
CD33	rs3865444	C	0.94a	1.26 (0.86,1.82)	1.28 (0.59,2.69)	1.44 (0.93,2.23)
CELF1	rs10838725	C	1.08	0.99 (0.67,1.47)	0.70 (0.34,1.50)	1.14 (0.71,1.87)
CLU	rs9331896	T	0.86a	0.83 (0.55,1.25)	1.18 (0.47,2.86)	0.73(0.44,1.18)
CR1	rs6656401	A	1.18	1.46 (0.87,2.57)	1.56 (0.59,5.35)	1.44 (0.78,2.83)
DSG2	rs8093731	C	0.73a	0.96 (0.05,5.41)	NA	0.94 (0.05,5.98)
EPHA1	rs11771145	G	0.90a	0.96 (0.65,1.40)	1.29 (0.58,2.75)	0.88 (0.56,1.37)
FERMT2	rs17125944	C	1.14	1.69 (0.81,4.10)	3.05 (0.60,55.84)	1.41 (0.63,3.77)
HLA-DRB5/HLA-DRB1	rs9271192	C	1.11	1.01 (0.68,1.52)	0.93 (0.42,2.19)	1.03 (0.65,1.68)
INPP5D	rs35349669	T	1.08	0.88 (0.61,1.28)	1.07 (0.49,2.28)	0.75 (0.48,1.15)
MAPT	rs393152	A	NA	**2.18 (1.26,3.77)**	**3.73 (1.30,11.76)**	1.77 (0.92,3.42)
MEF2C	rs190982	A	0.93a	1.24 (0.84,1.82)	1.03 (0.44,2.34)	1.15 (0.74,1.81)
MS4A4A	rs983392	A	0.90a	1.29 (0.88,1.90)	1.57 (0.72,3.43)	1.19 (0.75,1.90)
NME8	rs2718058	A	0.93a	0.77 (0.51,1.14)	0.89 (0.38,1.98)	0.74 (0.45,1.18)
PICALM	rs10792832	G	0.87a	1.16 (0.79,1.69)	1.03 (0.45,2.27)	1.15 (0.75,1.77)
PTK2B	rs28834970	C	1.10	1.22 (0.85,1.78)	1.38 (0.68,3.02)	1.10 (0.71,1.72)
SLC24A4/RIN3	rs10498633	G	0.91a	1.24 (0.82,1.86)	1.50 (0.63,3.36)	1.15 (0.70,1.84)
SORL1	rs11218343	T	0.77a	1.72 (0.62,4.08)	2.99 (0.63,10.64)	1.45 (0.32,4.97)
ZCWPW1	rs1476679	T	0.91a	1.31 (0.87,1.95)	**2.98 (1.34,6.86)**	1.04 (0.62,1.69)

[a] ORs for IGAP are for minor allele count. ORs for the current study are for risk allele count. ORs denoted by "a" indicate when the major allele is the risk allele. Hence, ORs denoted by "a" would be expected to be in the opposite direction for this study compared with IGAP.

Note: ORs should be interpreted as the odds of symptomatic vs. asymptomatic for an increase of one risk allele. For example, the odds of symptomatic AD for rs6656401 is 18% higher for a participant with one risk allele vs. no risk alleles. Due to missing values, the number of participants in each cell varies: All participants: max=589, min=473; APOE e4 carriers: max=292, min=234; APOE e4 non-carriers:

max=297, min=239 for all SNPs except the one associated with *MAPT*. For *MAPT*, n=345 for all participants, n=174 for *APOE* ε4 carriers, and n=171 for *APOE* ε4 non-carriers. SNPs in bold were significant at the 0.05 alpha level.

IGAP= International Genomics of Alzheimer's Project; NA= Not applicable; data too sparse for analysis.

Table 3

Genetic risk score for SNP association with symptomatic AD, adjusted for age at death and sex

	19 SNPs with <10% missing data[a]			21 SNPs with <15% missing data[b]			All 22 SNPs		
	n	OR	95% CI	n	OR	95% CI	n	OR	95% CI
All participants	419	2.61	(1.61,4.36)	300	2.15	(1.24,3.88)	184	1.87	(1.02,3.60)
APOE e4 carriers	201	5.01	(1.65,18.54)	143	6.01	(1.20,44.37)	93		NA
APOE e4 non-carriers	218	2.27	(1.30,4.10)	157	1.97	(1.05,3.84)	91	1.77	(0.87,3.82)

[a]The SNPs associated with *BIN1*, *CLU* and *MAPT* were not included.

[b]The SNP associated with *MAPT* was not included.

NA= Not applicable; data too sparse for analysis (one cell in the analysis had < 5 entries).

Note: *APOE* e4 carriers are those subjects with 1 or 2 e4 alleles whereas non-carriers contain 0 e4 alleles. Also note that the 'n' for each model is the overall n for both symptomatic and asymptomatic subjects.