Investigation of established AD risk SNPs for association with AD in an African American Case control series.

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Abstract

Background: Recent Genome-Wide association studies (GWAS) of Caucasian Late onset Alzheimer's Disease (LOAD) patients and controls have identified 9 novel LOAD susceptibility loci. The relevance of these loci in non-Caucasian populations is unclear. Studies report a higher incidence of Alzheimer's disease in the African-American population when compared to Caucasian populations. Likewise reports suggest that the risk of the *APOE* e4 allele with AD is not as significant in the African-American population as compared with Caucasian populations. Therefore LOAD genetic risk factors identified in Caucasian populations may not be as relevant to the African American population. Consequently it is critical that studies are conducted to elucidate genetic risk factors for this understudied population.

Methods: Subjects were collected at the Mayo Clinic in Jacksonville. All patients were diagnosed by a Mayo Clinic neurologist. DNA was isolated from whole blood using an Autogen instrument. SNPs were genotyped using Applied Biosystems Taqman ® technology. Statistical analysis was carried out using StatsDirect software; SNPs were assessed for association with AD diagnosis using an allelic dosage model with Age at Diagnosis, Gender and number of *APOE* e4 alleles included as covariates.

Results: We tested a total of 11 SNPs and the *APOE* e4 allele for association with AD in our African-American case control series (114 AD, 298 controls). None of the SNPs tested achieved nominally significant association with AD diagnosis. Analysis of *APOE* identified significant increased risk with increasing number of *APOE* e4 alleles (OR=3.92, p<1e-04).

Conclusions: The novel LOAD susceptibility loci identified in recent years through GWAS analysis of Caucasian LOAD patients and controls do not achieve nominally significant association in our African-American case control series. This may be due to the small sample size of our case control series, resulting in the study being underpowered, or may reflect a true fundamental difference in the association of these SNPs with AD in the two populations. Analysis with cognitive quantitative phenotypes collected in these subjects were also carried out. To increase the power of these studies, recruitment/collection of biological samples of AD patients and controls in the African American population is of increasing importance.

Aims

- To determine whether the late-onset Alzheimer's disease (LOAD) susceptibility variants identified using genome-wide association study (GWAS) approaches in Caucasian cohorts associate with LOAD in an African-American case control series.
- To investigate these same loci for influences on memory endophenotypes in an African-American cohort.
- To evaluate this African-American cohort for association with APOE.
- To increase studies of LOAD susceptibility in an understudied population.

Methods

The African-American series (AA) was collected at Mayo Clinic Florida in Jacksonville. All subjects were diagnosed by a Mayo Clinic neurologist and underwent neuropsychometric testing. Controls had a clinical dementia rating (CDR) score of 0 at the time of last testing and LOAD cases had a diagnosis of probable or possible AD according to NINCDS-ADRDA criteria.

Six cognition scores representing two functions of episodic memory were assessed for association with each single nucleotide polymorphism (SNP). Three endophenotypes for logical memory were: Logical Memory Immediate Recall (LMIR), Logical Memory 30 minute delayed recall (LMDR) and Logical Memory Percent Retention scores (LMPR) for recall of two narrative stories. Three endophenotypes for Visual Reproduction were: Visual Reproduction Immediate Recall (VRIR), Visual Reproduction 30 minute Delayed Recall (VRDR) and Visual Reproduction Percent Retention scores (VRPR) for recall of geometric figures. All SNPs were assessed for association with these memory endophenotypes measured at each subject's most recent (last or proximal) visit.

Genotypes were obtained using Taqman® assays.

Analysis were implemented in PLINK where an additive model was employed for each SNP while controlling for covariates detailed in Tables 3 and 4. When both LOADs and controls (All) were analyzed together an additional term for diagnosis was also included (LOAD = 1, control = 0).

Table 1

Case Control Series	Cases	Controls
N	114	298
Male (%)	31 (27%)	69 (23%)
Mean Age at diagnosis (range)	78.05 (61.8 - 97.0)	73.22 (60.1 - 91.6)
APOE ε2/ε2 (%)	1 (0.9%)	4 (1.34%)
APOE ε2/ε3 (%)	4 (3.5%)	61 (20.5%)
APOE ε3/ε3 (%)	39 (34.2%)	135 (45.3%)
APOE ε2/ε4 (%)	6 (5.3%)	9 (3.0%)
APOE ε3/ε4 (%)	45 (39.5%)	81 (27.2%)
APOE ε4/ε4 (%)	19 (16.6%)	8 (2.7%)

Table 1. Demographic information for African American Case Control Series. N = number of subjects. *APOE* ε4 was tested for association with AD in this cohort using an ε4 dosage model with age and gender included in the regression model: OR = 3.73, P=4.94E-11.

Table 2

Cognition Cohort	Cases	Controls
N	44	224
Male (%)	11 (25%)	21 (48%)
APOE ε4+ (%)	33 (75%)	71 (32%)
Mean Age at test (range)	78.9 (52.2 - 91.2)	78.7 (60.5 - 96.4)
Mean Yrs Education (range)	12.8 (4 - 20)	12.6 (2- 20)
Mean Reading Score (range)	90.3 (30 -113)	95.9 (45 - 118)

Table 2. Demographic information for African-American cohort with cognitive testing. Number of subjects (N); number and percentage of male participants; number and percentage of subjects with 1 or 2 copies of the APOE ε4 allele. Mean age at last logical memory test, mean number of years of education and mean reading score are shown. More subjects were tested for logical memory than visual reproduction, therefore the table represents data for the former.

Figure 1

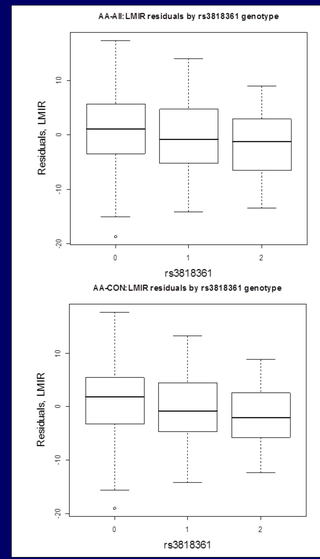

Figure 1. Box plots of logical memory immediate recall (LMIR) scores by CR1 rs3818361 in African-American (AA) control (CON) and combined AD and Control series (ALL).

Table 3

				Previously reported results in Caucasian cohorts					Results in African-American Cohort				
Locus	Chr	SNP (proxy)	r^2 (D')#	Reference	Tested Allele[a]	MAF	OR (95%CI)	P-value	N	Tested Allele	MAF	OR (95%) CI	P-value
ABCA7	19	rs3764650	na	Hollingworth et al	(G)	0.10	1.23 (1.18-1.30)	4.50E-17	394	G	0.27	1.09 (0.74-1.62)	0.670
BIN1	2	rs744373	na	Seshadri et al	G	0.29	1.15 (1.11-1.20)	1.59E-11	403	G	0.49	0.95 (0.68-1.34)	0.773
CD2AP	6	rs9349407 (rs1872505)	1.0 (1)	Naj et al	na	0.27	1.11 (1.07-1.15)	8.60E-09	404	G	0.32	0.89 (0.61-1.3)	0.548
CD33	19	rs3865444 (rs1354106)	0.93 (1)	Naj et al	na	0.30	0.91 (0.88-0.93)	1.60E-09	395	C	0.46	1.12 (0.79-1.61)	0.521
CLU	8	rs11136000	na	Lambert et al	**(A)**	0.38*	0.86 (0.81-0.90)	7.50E-09	410	**C**	0.43	1.18 (0.83-1.67)	0.369
CR1	1	rs3818361	na	Lambert et al	(A)	0.19*	1.19 (1.11-1.26)	8.90E-08	319	A	0.38	0.92 (0.60-1.40)	0.683
CR1	1	rs6656401	na	Lambert et al	(A)	0.19*	1.21 (1.14-1.29)	3.50E-09	342	A	0.04	1.84 (0.82-4.08)	0.136
EPHA1	7	rs11767557	na	Naj et al	C	0.19	0.90 (0.86-0.93)	6.00E-10	398	C	0.17	1.17 (0.76-1.81)	0.479
EXOC3L2	19	rs597668	na	Seshadri et al	C	0.15	1.17 (1.11-1.23)	6.45E-09	363	C	0.40	0.81 (0.56-1.16)	0.241
MS4A	11	rs610932	na	Hollingworth et al	**(C)**	0.42	1.09 (1.06-1.12)	1.80E-14	340	**T**	0.49	0.97 (0.67-1.41)	0.878
PICALM	11	rs3851179	na	Harold et al	(A)	0.37	0.90 (0.82-0.99)	1.30E-09	411	A	0.14	1.34 (0.84-2.15)	0.221

Table 3. Association of Alzheimer's' disease variants, identified in Caucasian GWAS, in an African-America LOAD Cohort. Eleven SNPS were tested for association with LOAD using logistic regression, assuming an additive model and correcting for age at diagnosis, APOEε4 dosage, and gender. # Two SNPs reported by Naj et al could not be tested directly using Taqman® assays and so proxies were identified (SNAP proxy), the r-squared and D' values reported using HapMap build 21 are shown for the pairs of SNPs. a: When the tested allele is not reported or is unclear the minor allele according to the HapMap CEU dataset is shown in parenthesis. *: Minor allele is reported in the control dataset of the respective reference. Alleles highlighted in blue indicate different minor alleles defined in Caucasian and African American populations.

Table 4

				CON			ALL		
Trait	Locus	SNP	Allele	N	Beta (SE)	P-value	N	Beta (SE)	P-value
LMDR	CD33	rs1354106	G	195	1.13 (0.77)	0.142	235	0.94 (0.66)	0.156
	CR1	rs3818361	A	222	-0.86 (0.72)	0.232	260	-0.80 (0.64)	0.211
	EPHA1	rs11767557	C	196	1.99 (1.00)	0.049	236	1.50 (0.86)	0.081
LMIR	CD33	rs1354106	G	195	1.35 (0.69)	0.054	236	1.21 (0.62)	0.053
	CR1	rs3818361	A	222	-1.31 (0.66)	0.047	261	-1.2 (0.60)	**0.005**
	EPHA1	rs11767557	C	196	1.73 (0.92)	0.060	237	1.50 (0.81)	0.066
VRDR	CD2AP	rs1872505	G	53	4.88 (1.28)	**3.90E-04**	86	3.52 (1.02)	**8.68E-04**
VRIR	ABCA7	rs3764650	G	52	-3.08 (1.63)	0.066	85	-2.23 (1.16)	0.060
	CD2AP	rs1872505	G	53	2.98 (1.46)	0.047	86	2.45 (1.15)	0.036
	CD33	rs1354106	G	51	-2.92 (1.72)	0.097	84	-1.61 (1.26)	0.207
VRPR	CD2AP	rs1872505	G	53	13.9 (4.91)	**0.007**	86	7.73 (4.44)	0.086
	CR1	rs3818361	A	57	9.56 (5.01)	0.062	88	10.16 (4.19)	0.018
	PICALM	rs3851179	A	58	12.1 (7.77)	0.126	94	7.59 (6.17)	0.222

Table 4. Association between ten tested SNPs and six measures of memory; Six episodic memory endophenotypes represented by logical memory immediate recall (LMIR), delayed recall (LMDR), percent retention (LMPR), visual reproduction immediate recall (VRIR), delayed recall (VRDR) and percent retention (VRPR) from Wechsler Memory Scale-Revised test were used to assess association with ten SNPs at ten GWAS loci; using multivariable linear regression analysis assuming an additive model, and correcting for age-at-testing, sex, APOE ε4 dosage, education years and reading score. Results are shown where the p-value < 0.25 in both the control (CON) and combined groups (ALL). Nominally significant p-values (<0.05) are highlighted in red.

Conclusions

- We did not find significant association, with LOAD, in our African-American cohort of the recent GWAS variants identified in Caucasians. This may be due to the small sample size of our study or may highlight important differences in genetic risk factors in this population.
- Despite observing no association with risk for LOAD, we did identify nominally significant association of the CR1 risk allele (rs3818361, A) with worse performance in memory tests (LMIR & LMDR).
- We also identified nominally significant associations between some of the memory endophenotypes and variants at the EPHA1, and CD2AP loci.
- The memory endophenotype associations need to be replicated in independent series.
- African-Americans are an understudied population with a high incidence of Alzheimer's Disease; multiple risk factors and a complex genetic etiology that may differ from that of Caucasians. Efforts must be made to increase awareness and study participation in this population.

References

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