

Multi-omics Characterization of Middle Onset Alzheimer’s Disease (MOAD)

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Background

Alzheimer’s disease (AD) is characterized by accumulation of β-amyloid plaques and phosphorylated tau. Age at onset varies widely and is associated with distinct genetic architectures.

- Early-onset AD (EOAD; typically defined <65 years with onset usually much earlier) is rare and often linked to high-penetrance variants in APP, PSEN1, and PSEN2, though these explain only a small proportion of cases.
- Late-onset AD (LOAD; typically defined >65 years) is largely polygenic and strongly associated with APOE4.
- Less is known about AD patients with symptom onset between 45–60 years and don’t carry the genetic associations of MOAD and LOAD, defined here as middle-onset Alzheimer’s disease (MOAD). We hypothesized that MOAD has a distinct genetic architecture..

Methods

Autopsy-confirmed cases from the Mesulam Institute at Northwestern University included MOAD (onset ≤60 years, n=109), LOAD (rigorously defined with onset ≥70 years, n=109), and controls without dementia (death >70, n=17). After quality control, whole-genome sequencing was performed on MOAD (n=84), LOAD (n=79), and controls (n=14).

- Candidate gene analyses focused on 309 AD-associated genes identified through familial studies and genome-wide association studies (GWAS).
- Rare variants were prioritized based on ACMG pathogenicity classification, predicted deleteriousness (REVEL ≥0.75), and ClinVar annotations. P-values calculated using Fisher’s exact test.
- Pathway analyses were conducted using the Reactome database using likely implicated rare variants in candidate genes in MOAD vs LOAD.
- Short tandem repeats (STR) analyses were completed using ExpansionHunter bioinformatics tool comparing MOAD vs LOAD.

Demographics

Percentage of females: MOAD 48%, LOAD 57%, controls 64%. p =0.335

Atypical clinical AD phenotypes (behavioral variant/dysexecutive/ PPA/PCA) among MOAD vs LOAD

- MOAD: 44 cases with atypical clinical presentation (52%)
- LOAD: 17 cases with atypical clinical presentation (22%)

MOAD vs LOAD **p = 0.000047**

Kruskal–Wallis APOE4 dosage analyses across all three groups: **P=0.031** for Control < MOAD < LOAD

Group	ε4 dosage = 0	ε4 dosage = 1	ε4 dosage = 2	N	Mean ε4 dosage
Controls	11	3	0	14	0.214
MOAD	49	27	8	84	0.512
LOAD	35	35	9	79	0.671

Table 1. APP, PSEN1, PSEN2 analyses

case	phenotype	ch	position	ref	alt	gene	impact	Amino Acids	polyPhen Prediction	siftPrediction	Gnomad exome all Af	Gnomad exome all An	revel	Family history
2157	LOAD*	chr14	73219194	A	G	PSEN1	MODERATE	I/V	benign	deleterious	3e-06	1461528	0.554	no data
1521	MOAD	chr14	73192786	G	A	PSEN1	MODERATE	A/T	probably damaging	deleterious	.	.	0.964	father with AD, AAO 46
1679	MOAD	chr14	73192712	G	C	PSEN1	MODERATE	G/A	probably damaging	deleterious	6e-06	1461862	0.921	father with AD, AAO 55
1802	MOAD	chr14	73192748	C	T	PSEN1	MODERATE	P/L	probably damaging	deleterious	3e-06	1461676	0.981	no known dementia
1857	MOAD	chr14	73219177	C	A	PSEN1	MODERATE	A/E	probably damaging	deleterious	.	.	0.946	father with AD, AAO 40
1952	MOAD	chr14	73217182	G	A	PSEN1	MODERATE	A/T	probably damaging	deleterious	.	.	0.928	father with LBD (autopsy confirmed), AAO 52
886	MOAD	chr14	73170945	C	T	PSEN1	MODERATE	A/V	probably damaging	deleterious	7e-06	1461884	0.958	no data
1640	MOAD	chr1	226883774	C	T	PSEN2	MODERATE	R/W	benign	deleterious	0.003697	1461874	0.61	father with AD, AAO 80
428	MOAD	chr1	226883774	C	T	PSEN2	MODERATE	R/W	benign	deleterious	0.003697	1461874	0.61	no data
1343	MOAD	chr21	26051070	A	G	APP	MODERATE	S/P	possibly damaging	deleterious	0.000311	1461882	0.547	no known dementia
1104	MOAD	chr21	25891784	C	T	APP	MODERATE	V/I	probably damaging	tolerated	1e-06	1461620	0.78	mother with dementia, AAO 48
1470	MOAD	chr21	25891784	C	T	APP	MODERATE	V/I	probably damaging	tolerated	1e-06	1461620	0.78	mother with dementia, AAO 45

- 1 LOAD (1.3%) case with likely pathogenic rare variant in PSEN1 while 11 MOAD (13.1%) cases carried mostly pathogenic/ Likely pathogenic rare variants in APP (3 cases), PSEN1 (6 cases), and PSEN2 (2 cases). MOAD vs LOAD **p= 0.005**
- Of those 11 MOAD cases there are 6 cases (55%) with parental family history of early onset dementia. Those could arguably be labeled as typical familial EOAD

Table 2. APOE rare variants

case	phenotype	ch	position	ref	alt	gene	impact	Amino Acids	polyPhenPrediction	siftPrediction	Gnomad exome all Af	Gnomad exome all An	revel	Family history
1322	MOAD+	chr19	44909101	C	G	APOE	MODERATE	R/G	possibly damaging	deleterious	0.00069	1435384	0.581	Mother with dementia AAO 90
1386	MOAD	chr19	44908783	C	T	APOE	MODERATE	R/C	probably damaging	deleterious	0.000684	1405748	0.695	Mother with AD AAO 65
1524	MOAD+	chr19	44909101	C	G	APOE	MODERATE	R/G	possibly damaging	deleterious	0.00069	1435384	0.581	Unknown

- These 3 cases all carry APOE4 so there might be interaction, those rare APOE variants might lead to earlier onset among APOE4 carriers

Table 3. variants classified as pathogenic / likely pathogenic

case	phenotype	chr	position	ref	alt	gene	genotype	consequence	impact	amino acids	disease	mode of inheritance reported	revel	Family history
1832	LOAD	chr19	1058155	G	T	ABCA7	hete	stop_gained	HIGH	E/*	Alzheimer's	Autosomal dominant (risk)	0.013	Mother AD 65
424	MOAD	chr3	155116689	AC	-	MME	hete	Frameshift_variant	HIGH	P/X	Charcot-Marie-Tooth disease axonal type 2T	A-R or A-D	.	No data
853	MOAD	chr3	155144383	C	T	MME	hete	stop_gained	HIGH	R/*	Spinocerebellarbarataxia 43	A-R or A-D	.	No data

Discussion

- **APOE4** in MOAD is more common than controls and less common than LOAD
- MOAD cases could be triggered by **rare variants in APOE** gene which could influence age at onset (interaction with APOE4?)
- (13.1%) of cases in the middle onset AD group carried pathogenic rare variants in known familial AD genes (**APP, PSEN1, and PSEN2**), and >50% have strong family history, those are classified traditionally as familial EOAD
- Rare variants in **MME** (which encodes neprilysin, an amyloid-β (Aβ) degrading enzymes) are specifically implicated in MOAD
- Single rare variant in **WDR81** (which is involved in endo-lysosomal trafficking, neuronal growth and adult hippocampal neurogenesis) is specifically implicated in LOAD.
- STR in **CACNA1A** is implicated in MOAD, while STR in **SORL1** is implicated in LOAD
- **The immune pathway** results suggests that MOAD might be triggered by infections and dysregulated immune inhibition while LOAD is associated with dysregulated microglial clearance of misfolded proteins

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Table 4. rare variants shared by >3 cases

Total cases	MOAD n	LOAD n	Cont n	Chr	position	ref	alt	gene	Impact	amin o Acids	Tpoly Phen Prediction	sift Prediction	Var gnomad exome_ al l Af	Var gnomad exome_ all An	revel	P *
5	3	1	1	chr2	162504648	T	A	KCNH7	MODERATE	N/I	possibly damaging	deleterious	0.00597	1454462	0.747	0.721
5	3	1	1	chr22	26027319	C	T	MYO18B	MODERATE	R/W	probably damaging	deleterious	0.005728	1461692	0.632	0.721
5	0	5	0	chr17	1733880	G	A	WDR81^	MODERATE	G/R	probably damaging	deleterious	0.004972	1460512	0.584	0.025
5	3	2	0	chr1	201076930	C	T	CACNA1S	MODERATE	S/N	possibly damaging	deleterious	0.009758	1461840	0.521	0.721
4	2	2	0	chr5	177504605	G	T	DOK3	MODERATE	A/D	probably damaging	tolerated	0.001661	1458410	0.545	0.683

Figure 1: immune pathway analyses

