Molecular Characterization and Validation of Sex Differences in AD Pathogenesis

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Prevalence of dementia from The Aging, Demographics and Memory Study (ADAMS)

Table 2. National prevalence of dementia, AD and VaD, by age categories

Age	All dementia			AD			VaD		
	combined	male	female	combined	male	female	combined	male	female
71–79 years	4.97 (2.61–7.32)	5.25 (1.25–9.25)	4.76 (1.82–7.70)	2.32 (1.26–3.37)	2.30 (0.80–3.81)	2.33 (0.95–3.70)	0.98 (0.07–1.89)	1.27 (0.00–3.19)	0.76 (0.18–1.35)
80–89 years	24.19 (19.28–29.11)	17.68 (11.66–23.70)	27.84 (20.41–35.28)	18.10 (13.47–22.74)	12.33 (5.82–18.84)	21.34 (14.44–28.24)	4.09 (1.52–6.67)	3.58 (1.37–5.79)	4.38 (0.71–8.05)
≥90 years	37.36 (25.45–49.27)	44.59 (21.70–67.47)	34.69 (23.36–46.02)	29.70 (18.60–40.80)	33.89 (10.00–57.77)	28.15 (17.61–38.69)	6.19 (2.14–10.23)	8.14 (0.0–16.76)	5.46 (1.49–9.44)
Total	13.93 (11.42–16.44)	11.14 (7.78–14.50)	15.74 (12.39–19.08)	9.74 (7.56–11.91)	7.05 (4.25–9.85)	11.48 (8.50–14.46)	2.43 (1.36–3.50)	2.34 (0.74–3.94)	2.48 (1.11–3.86)

Percentages and 95% confidence intervals (in parentheses) provided.

Plassman et al., 2007

Meta-analysis suggests worse cognitive performance of women with AD

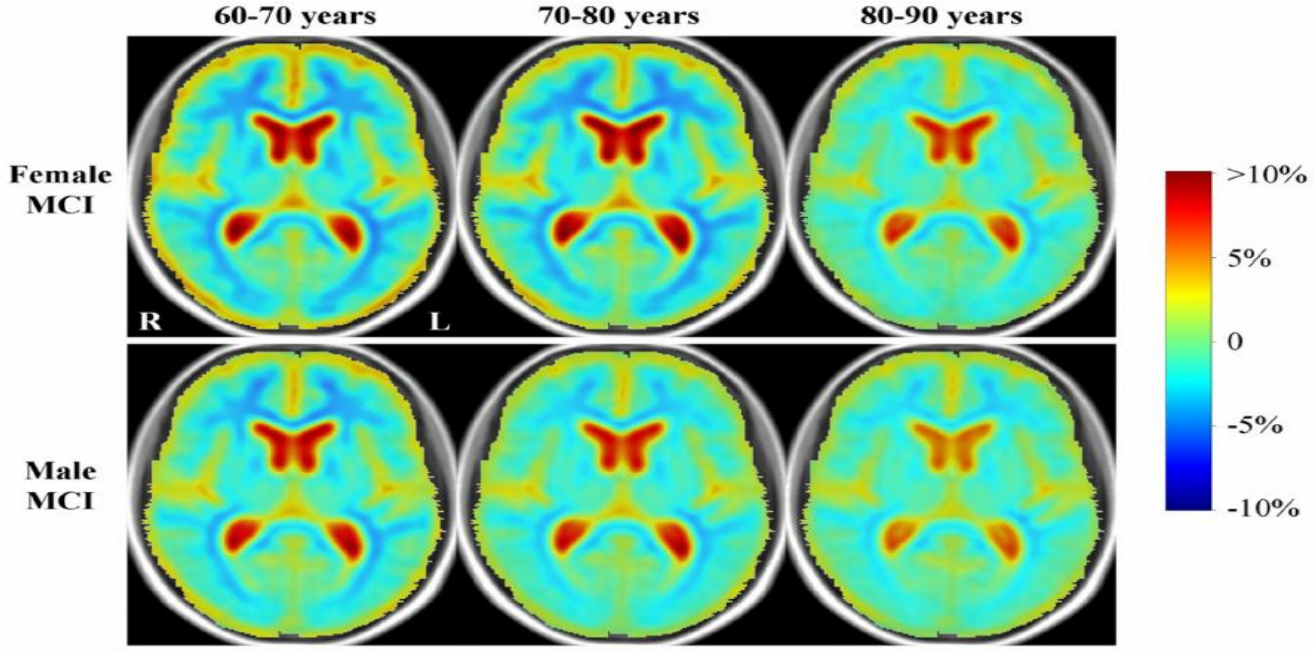
Ref.	Sample size			Semantic	Non-semantic	Verbal	Visual-spatial	Memory
	M	F	Total	*d*	*d*	*d*	*d*	*d*
Marra *et al*[41]	85	168	253	-0.23		-0.23		
Beinhoff *et al*[45]	26	23	49	-0.07	-0.44	-0.22	-0.60	-0.37
Hendersen *et al*[46]	22	24	46	-0.37	-0.37	-0.37	-0.18	
Hendersen *et al*[46]	270	377	647	-0.30	-0.12	-0.30	-0.11	-0.12
Moreno-Martinez *et al*[50]	28	33	61	-0.42		-0.42		
Buckwalter *et al*[55]	72	87	159	-0.46	-0.24	-0.46	-0.24	
McPherson *et al*[71]	23	36	59	-0.24	-0.54	-0.35		-0.71
Ripich *et al*[78]	29	31	60	-0.74		-0.74		
Bayles *et al*[79]	30	33	63	-0.10		-0.10		
Perneczky *et al*[81]	50	43	93	-0.24	-0.12	-0.20	0.02	-0.17
Heun *et al*[101]	17	76	93		-0.18		-0.62	-0.04
Millet *et al*[102]	20	20	40		-0.40	0.08	-0.63	-0.40
Laiacona *et al*[107]	11	15	26	-0.29		-0.29		
Hendersen *et al*[136]	26	27	53	-0.26	-0.22	-0.09	-0.44	-0.15
Hebert *et al*[137]	119	245	364	-0.23	0.04	-0.09		
Total	828	1238	2066	-0.25 (-0.42 to -0.07)	-0.14 (-0.26 to -0.02)	-0.27 (-0.37 to -0.16)	-0.24 (-0.43 to -0.05)	-0.17 (-0.33 to 0.01)

Negative effect sizes favour men and positive effect sizes favour women; numbers in parenthesis are 95%CIs. M: Male; F: Female.

Deficits were not predicted by differences in age, education, dementia severity or other demographic confounds.

Laws et al., 2016

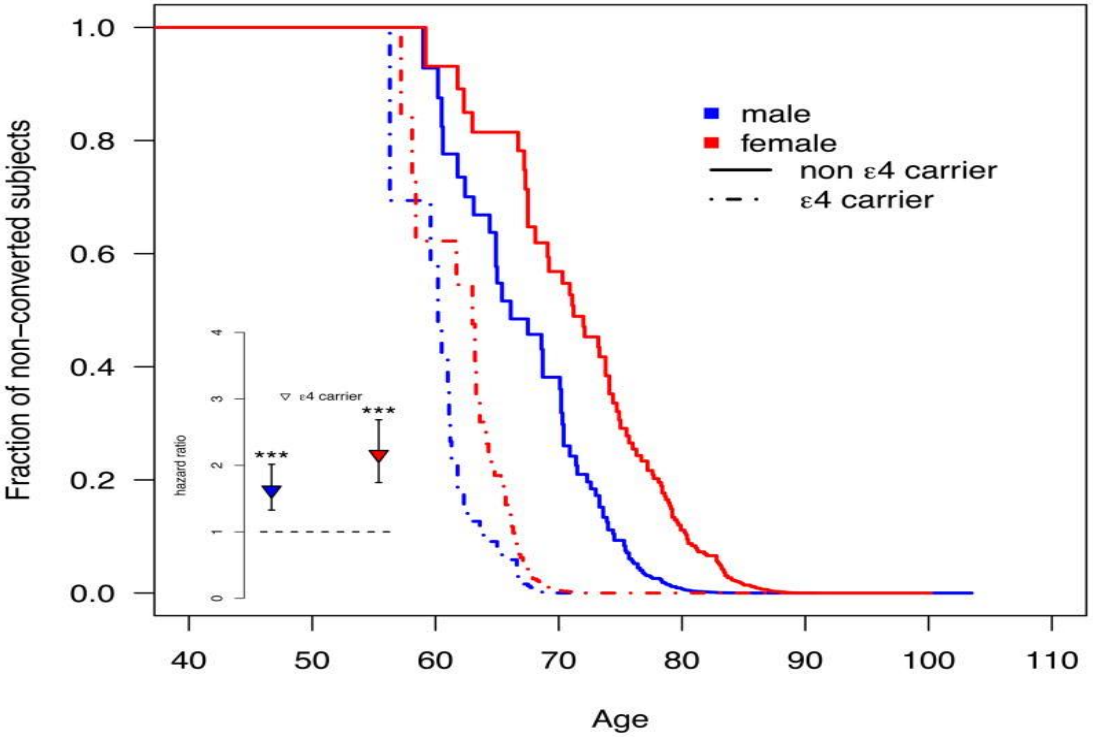
Faster atrophic rates occur in female MCI subjects



an ADNI study with N=1368 MRI scans

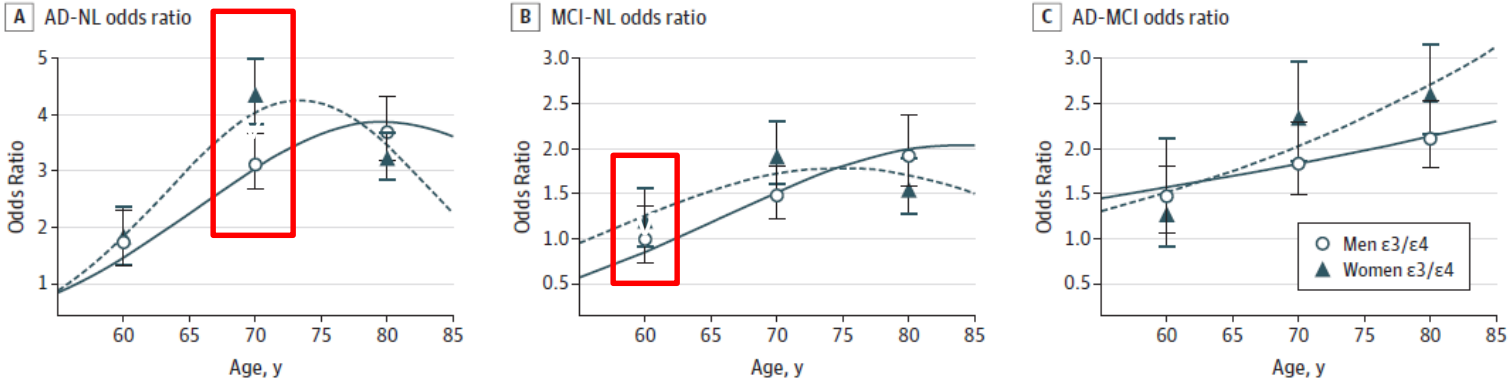
Hua et al., 2010

ApoE4 has divergent effects on gender-specific AD characteristics



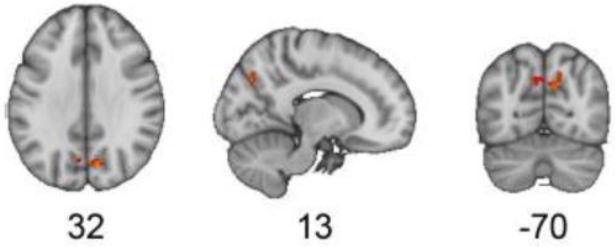
Altmann et al., 2014

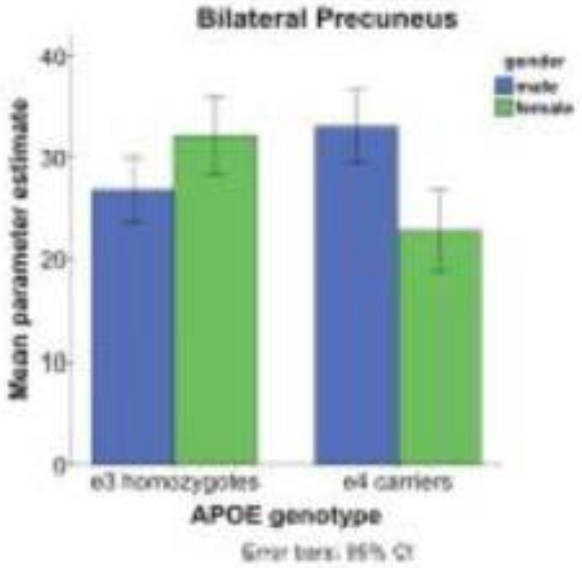
ApoE4 women showed increased risk of AD than ApoE4 man at age 65-75



Neu et al., JAMA 2017

Decrease in functional connectivity in ApoE4 female carriers





Ungar et al., 2014

Project Overview

Aim 1: Assemble, quality control, and perform differential expression on human postmortem gene expression data from female and male AD patients and non-demented controls to identify key sex-specific genes and pathways in AD.

Aim 2: Perform integrative network analysis of the assembled genetic and gene expression data to construct multiscale gene networks and identify key drivers for each AD subgroup.

Aim 3: Characterize the functional roles of identified gender- and ApoE-specific network drivers in AD development and progression using male and female ApoE4 and ApoE3 KI mice without and with 3xTg AD background.

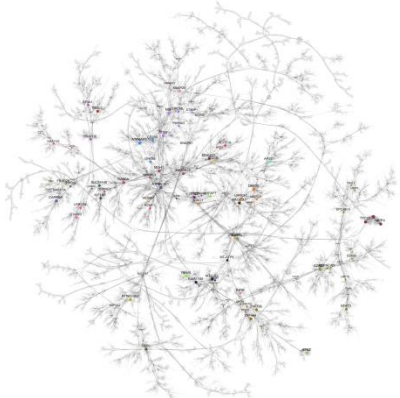
Data

- AD Genetics Data: GERAD, ADGC and CHARGE.
- Curated RNA-sequencing data from over 1448 samples, as well as complete clinical and pathological data from the Mount Sinai Brain Bank (MSBB) and ROSAMP cohorts.

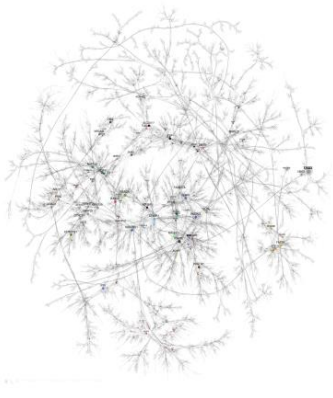
Brain Region	Female (AD)	Female (Normal)	Male (AD)	Male (Normal)	Total
MSBB BA10	133	24	50	27	230
MSBB BA22	120	18	56	21	215
MSBB BA36	98	21	41	24	184
MSBB BA44	64	15	24	17	200
ROSMAP	366	29	190	34	619

Gender- and ApoE4-Specific Coexpression Sub-Networks

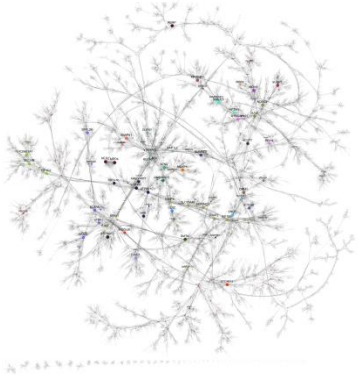
A. Female w/ ApoE4



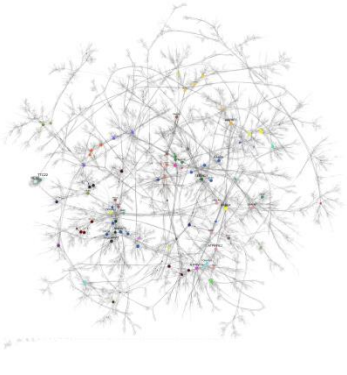
C. Male w/ ApoE4



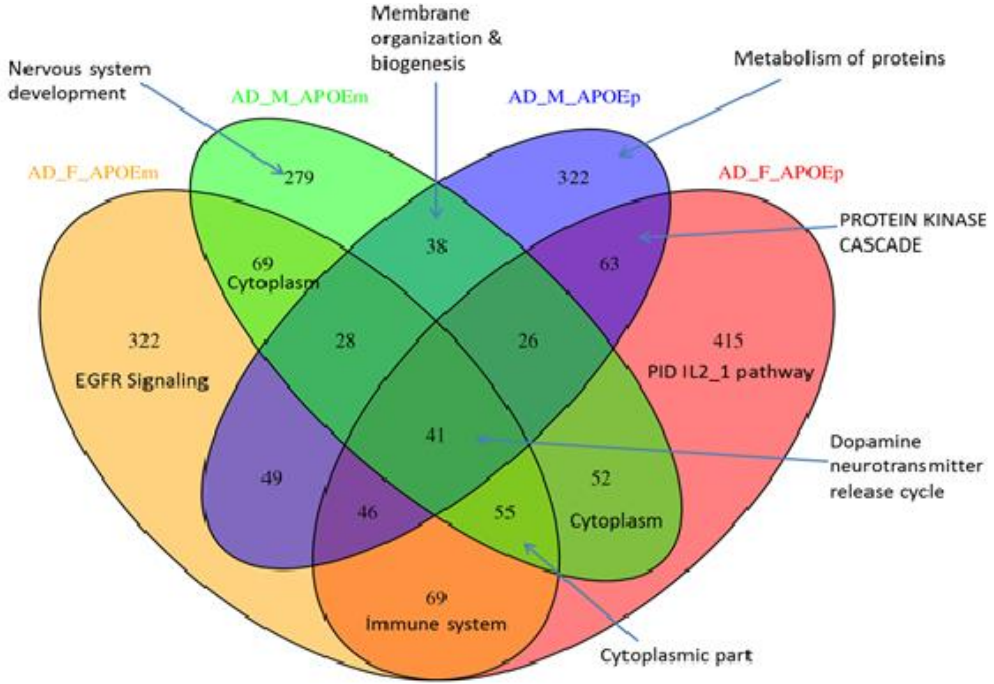
B. Female w/o ApoE4



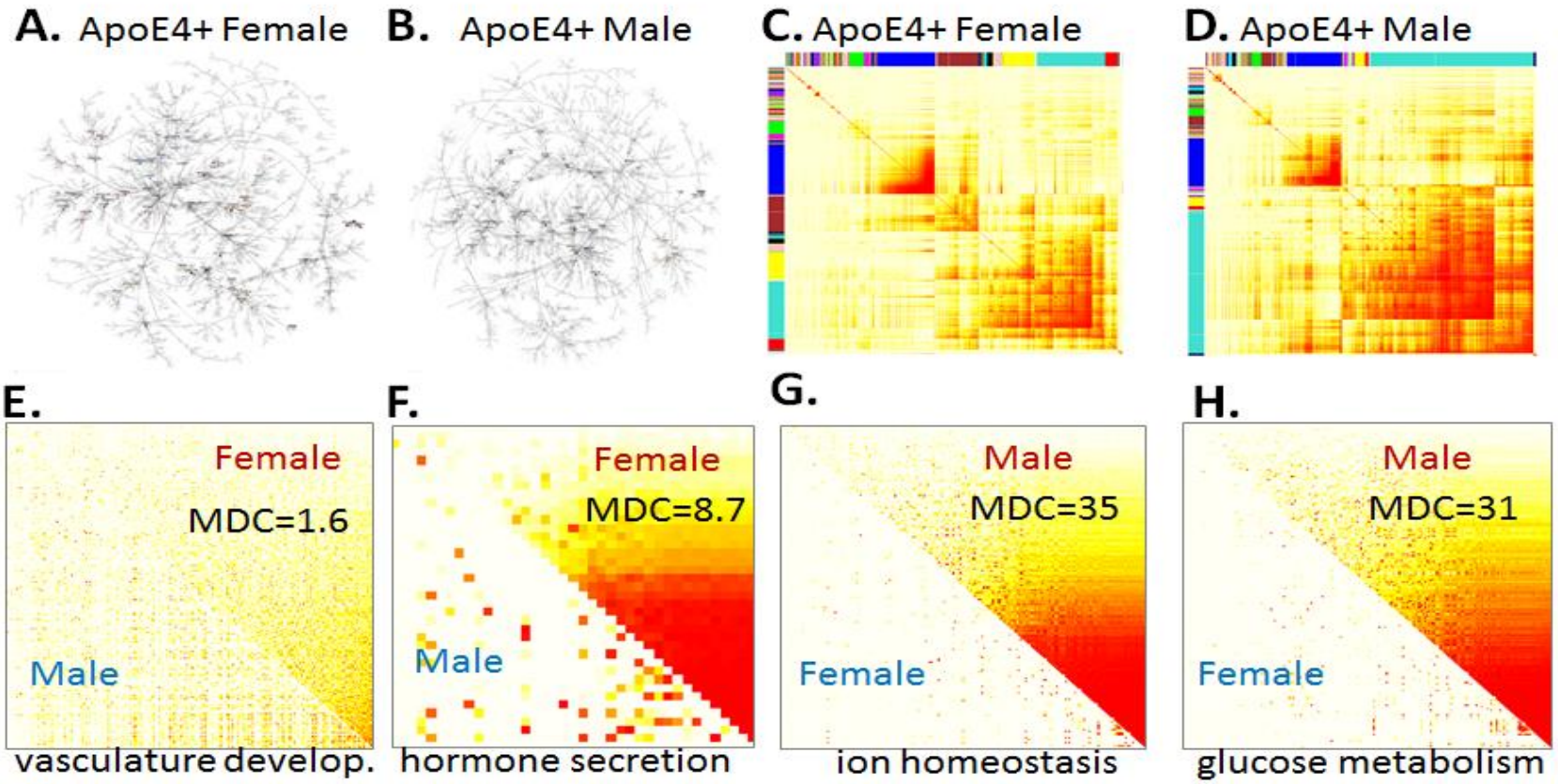
D. Male w/o ApoE4



E. Intersections of driver sets

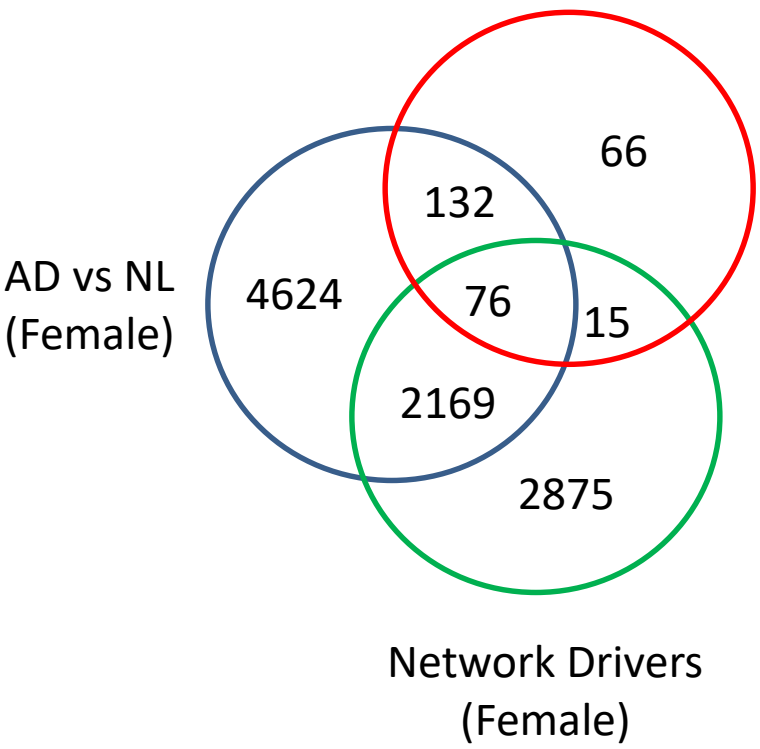


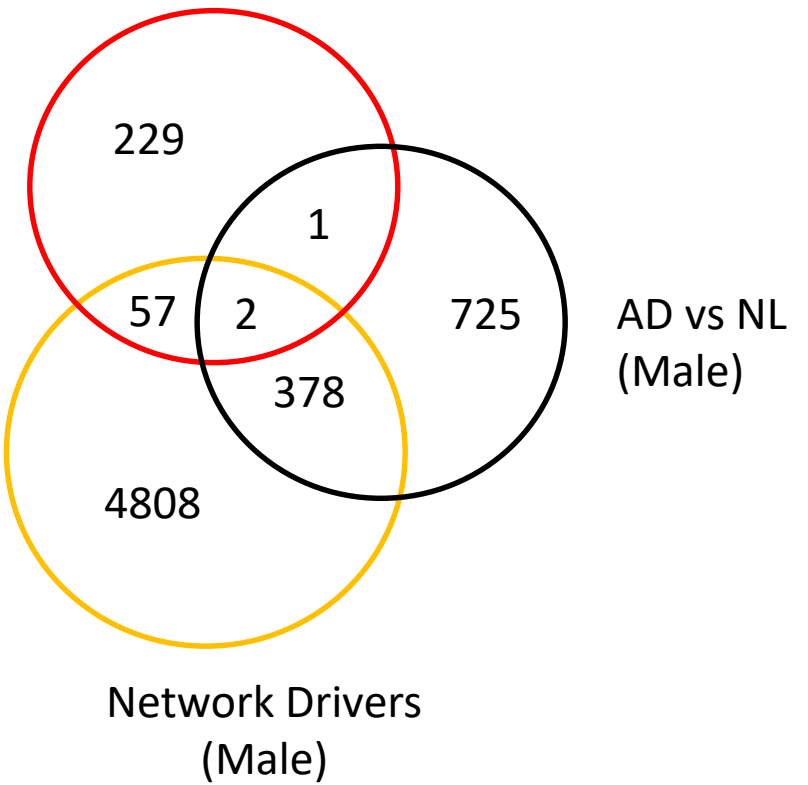
Weighted Coexpression Networks



Identification of Gender Specific Driver Genes in AD

Female vs Male (Differential Expression in AD)





MSBB vs ROSMAP
(Female candidate genes)
(Literature Mining)

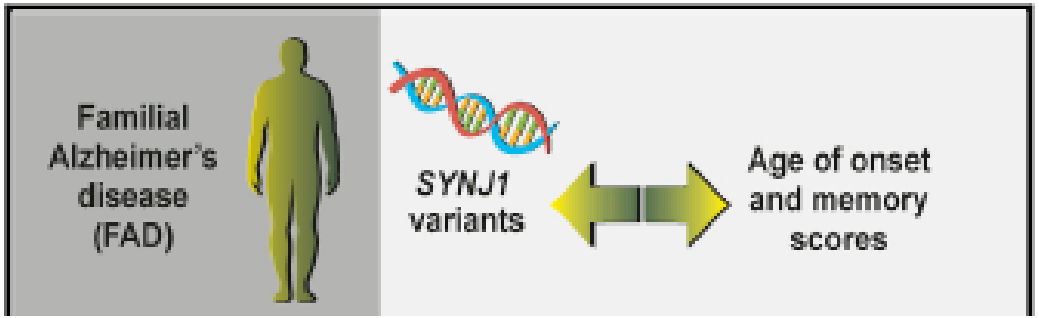
Synaptojanin 1 (synj1): a PIP_2 phosphatase, a rate limiting enzyme in brain PIP_2 metabolism

Article

Cell Reports

Excess Synaptojanin 1 Contributes to Place Cell Dysfunction and Memory Deficits in the Aging Hippocampus in Three Types of Alzheimer's Disease

Graphical Abstract



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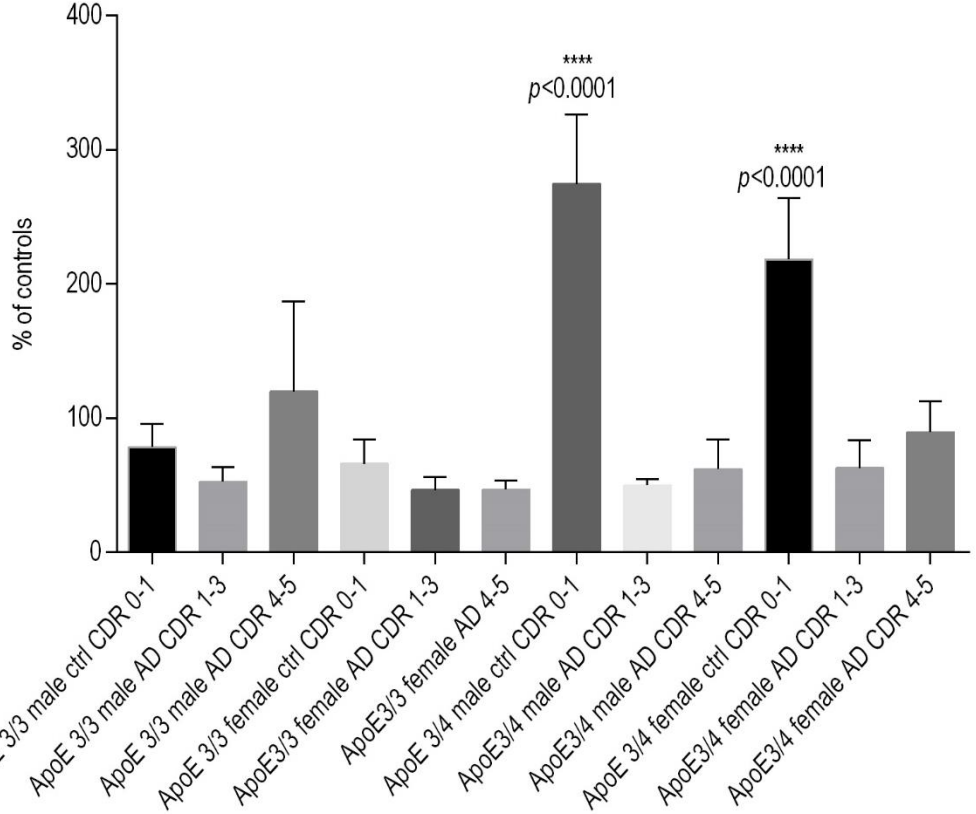
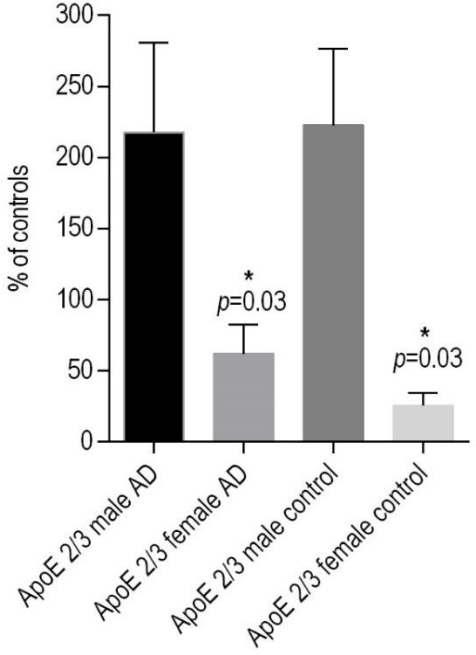
Brain Bank Samples for Validation

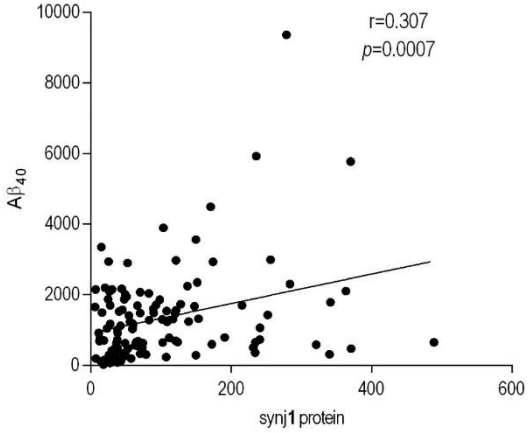
	ApoE 3/3 M AD	ApoE 3/3 F AD	ApoE3/4 or 4/4 M AD	ApoE3/4 or 4/4 F AD	ApoE 3/3 M ctrl	ApoE 3/3 F ctrl	ApoE3/4 or 4/4 M ctrl	ApoE3/4 or 4/4 F ctrl
Age (Y)	81.6± 10.6	89.6± 7.0	80.5± 8.7	86.9± 7.9	64.8± 14.1	77.3± 16.1	74.4± 7.4	69.0± 11.8
N	31	32	26	31	28	21	5	5
PMI (H)	5.46± 3.06	5.41± 3.70	5.44± 3.97	4.71± 2.44	10.43 ±5.48	6.56± 4.49	9.98± 5.04	11.48± 6.00
CDR 0	1	0	0	1	6	7	3	3
CDR 0.5-2	6	12	9	8	6	8	1	1
CDR 3-5	20	14	14	17	1	2	1	0
Maryland BB	4	6	3	5	15	4	0	1

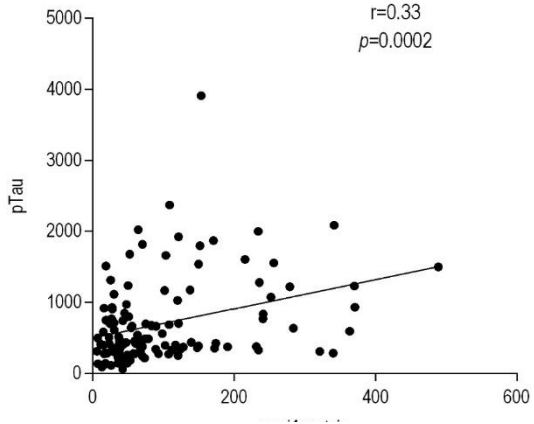
Synj1 expression in AD male and female brain

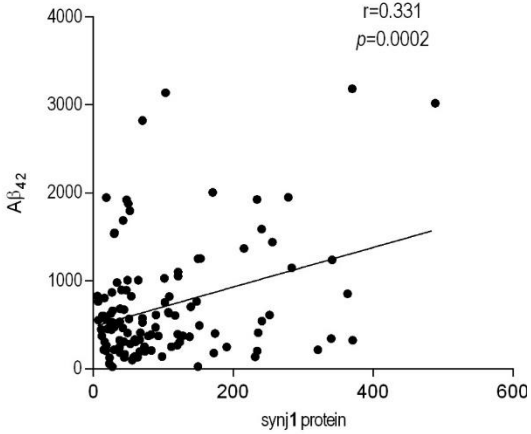


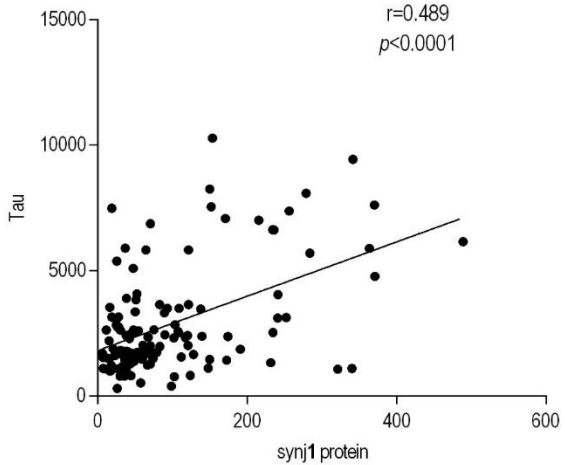


Synj1 expression correlates with AD pathology







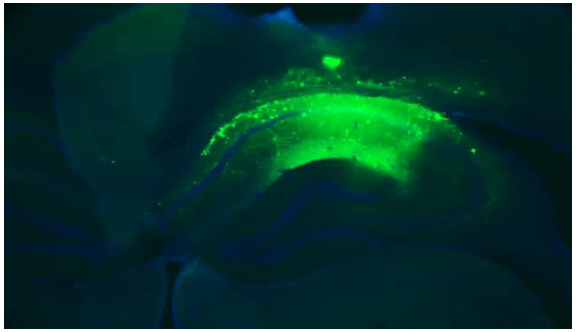


Functional characterization using AD mouse models

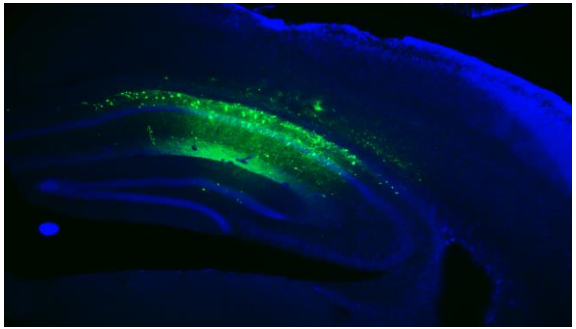
Genotype	Sex	Age	Functional studies
Wildtype (wt)	male female	0-20 mo	Behavioral battery tests, spine morphology, biochemical and IHC studies
APP PS1$^{+/-}$ (Jackson)	male female	0-20 mo	Females showing earlier and more robust defects in cognitive function and increased anxiety than males as measured by Morris water maze (MWM) and inhibitory avoidance (IA) tasks (Clinton et al., 2007)
ApoE3 TR ApoE4 TR (Taconics)	male female	0-20 mo	Age-dependent ApoE4-induced impairments in spatial learning and memory are more profound in female than male mice measured by MWM and Y-maze tests (Bour et al., 2008; Leung et al., 2012)
ApoE3/APP PS1$^{+/-}$ ApoE4/APP PS1$^{+/-}$ (license agreement)	male female	0-20 mo	An earlier and greater degree of spatial learning and memory impairments observed in ApoE/Tg female mice determined by radial arm water maze (RAWM) and novel arm discrimination (NAD) tasks (Hou et al., 2015)
ApoE3/5xFAD ApoE4/5xFAD (La Du's laboratory via MTA)	male female	0-20 mo	E4FAD mice develop pathology and behavioral deficits in ApoE-genotype specific and sex-induced patterns (Tai et al., 2017). In female EFAD mice, cognitive impairment is E4FAD > E3FAD ≥ E2FAD as measured by MWM with the deficits increasing from 2-6 mo of age. In 8mo of age mice, cognitive impairment is E4FAD > E3FAD and females > males as measured by novel object recognition (NOR) and Y-Maze (YM) tasks.

Gene Perturbation Experiments in ApoE4 Mouse Models

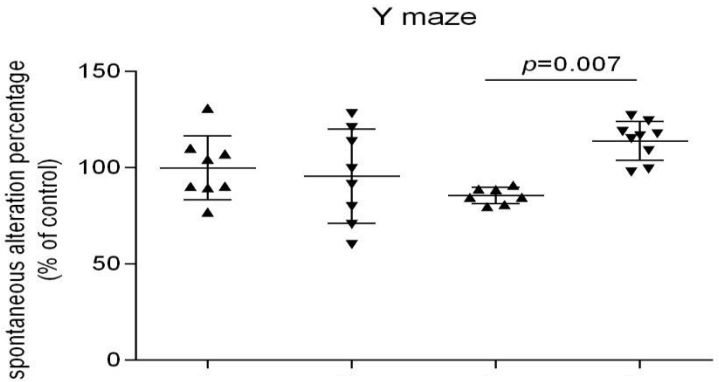
ApoE4 male



ApoE4 female



GFP-AAV shRNA



ApoE4 KI male versus female mice (6 month post injection)

iPSC cell lines for validation and functional analysis

APOE genotype	Family ID	Race	Sex	Dx	Clone type	Available CNS cell type
4/4	FA12-453	Caucasian	F	AD	Polyclonal	NPCs, cortical neurons, astrocytes, microglia
4/4	FA12-481	Caucasian	F	Control	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
4/4	FA12-444	Caucasian	M	AD	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
4/4	iPS 6	Caucasian	M	AD	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
4/4	iPS 20	Caucasian	F	AD	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
4/4	iPS 23	Caucasian	M	AD, CAA	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
3/3	FA12-455	Caucasian	F	Control	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
3/3	iPS 4	Caucasian	M	Control	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
3/3	iPS 5	Caucasian	F	Control	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
3/3	iPS 22	Caucasian	F	AD	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
3/3	iPS 13	Caucasian	F	AD	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
3/3	iPS 14	Caucasian	M	Control	Monoclonal	NPCs, cortical neurons, astrocytes, microglia
3/3	iPS 76	Caucasian	M	Control	Monoclonal	NPCs, cortical neurons, astrocytes, microglia

Summary

- Multiscale network modeling approaches identify novel gender- and ApoE-related pathways and gene modules in AD pathogenesis.
- Several key gender- and ApoE-specific regulators of multiscale networks in AD have been validated in human and mouse brain tissue.
- Functional roles of validated key drivers will be further characterized using various AD mouse models, as well as iPSC cell lines.
- The in-depth mechanistic studies of gender-specific alterations in network and molecular signatures pave a way for discovery of novel therapeutic targets for AD.

Acknowledgement

AMP-AD Consortium

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