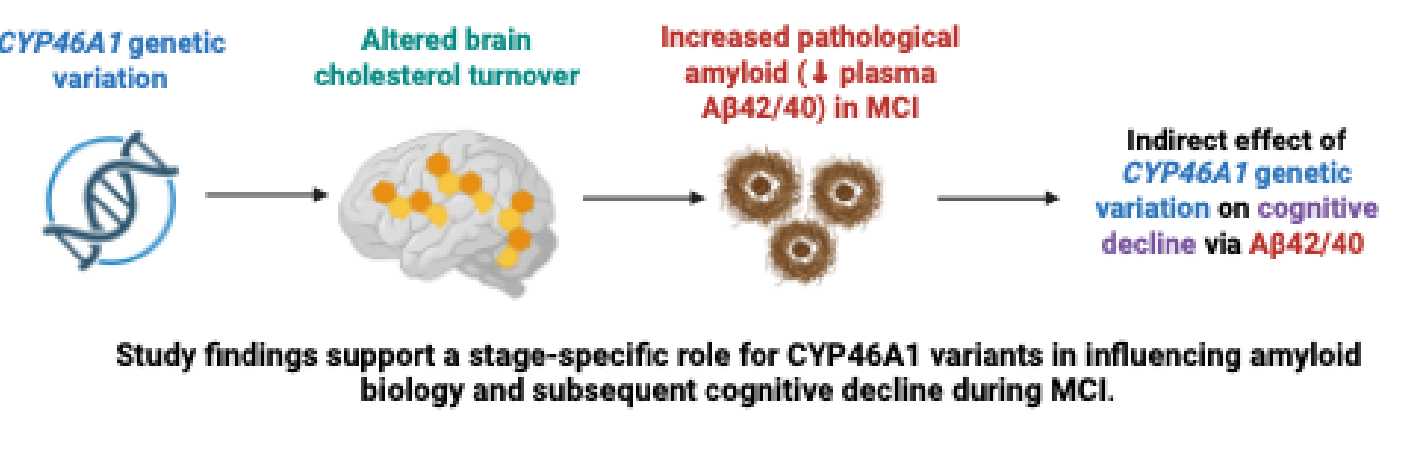


Genetic Variation in CYP46A1 is Associated with Alzheimer's Disease Biomarkers and Cognitive Decline

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Graphical Abstract



Background

- What is known:
- CYP46A1 regulates brain cholesterol homeostasis through cholesterol metabolism
 - Altered cholesterol metabolism has been implicated in Alzheimer's disease
 - Blood biomarkers are transforming AD diagnosis, participants selection and clinical trials

- Knowledge Gap
- Do *CYP46A1* variants influence AD biomarkers?
 - Are these associations stage-specific?
 - Do biomarker changes mediate cognitive decline?

- Objective:
- Determine whether *CYP46A1* variants influence AD biomarkers and mediate subsequent cognitive decline

Methodology

- Study Population
- Alzheimer's Disease Neuroimaging Initiative [ADNI]
 - N = 1,164 participants [N=381 cognitively normal; N=566 MCI; N=217 AD]

- Genetics
- 96 *CYP46A1* SNPs identified using PLINK
 - Tag SNP selection using Haploview

- | Biomarker outcomes | Cognitive outcomes |
|---|---|
| <ul style="list-style-type: none">Blood: Aβ42/40, ptau181, NfLImaging: white matter hyperintensities (WMH) | <ul style="list-style-type: none">Composite scores for memory (ADNI-MEM), executive function (ADNI-EF), language (ADNI-LAN) and visuospatial function (ADNI-VS) |

- Statistical analyses
- Linear regression to investigate association between *CYP46A1* tag SNPs and AD biomarkers in whole group and cognitive subgroups. FDR <.10 applied for multiple comparisons
 - Mediation analysis to investigate associations between weighted gene score and individual CYP46A1 tag SNPs with AD biomarkers from linear regression, and cognitive decline. Cognitive subgroup, and AD biomarkers selected based on a priori linear regression.
- All analyses adjusted for: age, sex, education E4 presence, and ancestry.

Results

Table 1: Study participant demographics						
		Mean (SD) or N(%)				
		Whole group (N=979)	CN (N=393)	MCI (N=457)	AD (N=129)	Between-group comparisons
Age		72.41 (7.18)	72.58 (6.02)	71.80 (7.62)	74.04 (8.45)	F(2)=19.51, p<.001
Female (%)		461 (47%)	225 (57%)	184 (40%)	52 (40%)	X ² (2)=27.22, p<.001
Years of Education		16.30 (2.60)	16.66 (2.43)	16.14 (2.68)	15.76 (2.69)	F(2)=29.28, p<.001
APOE-ε4 presence	0 copies	548 (56%)	270 (69%)	241 (53%)	37 (29%)	X ² (2)=66.77, p<.001
	1 copy	347 (35%)	113 (29%)	168 (37%)	66 (51%)	X ² (2)=21.97, p<.001
	2 copies	84 (9%)	10 (2%)	48 (11%)	26 (20%)	X ² (2)=42.44, p<.001
MMSE total		27.72 (3.49)	28.99 (2.06)	27.55 (2.07)	21.37 (5.16)	F(2)=1322, p<.001
Ab42/40		0.1406 (0.0560)	0.1638 (0.0516)	0.1387 (0.0569)	0.0989 (0.0309)	F(2)=57.85, p<.001
Ptau-181		27.2462 (14.3049)	22.7929 (9.0865)	26.7242 (14.7722)	37.7833 (16.0576)	F(2)=47.89, p<.001
NfL		42.5191 (24.3688)	38.9835 (23.1362)	41.2567 (24.1690)	52.3391 (23.5778)	F(2)=18.28, p<.001
WMH		6.6799 (9.1383)	5.9908 (9.3297)	6.6742 (8.1743)	8.7997 (11.2900)	F(2)=4.622, p=.01

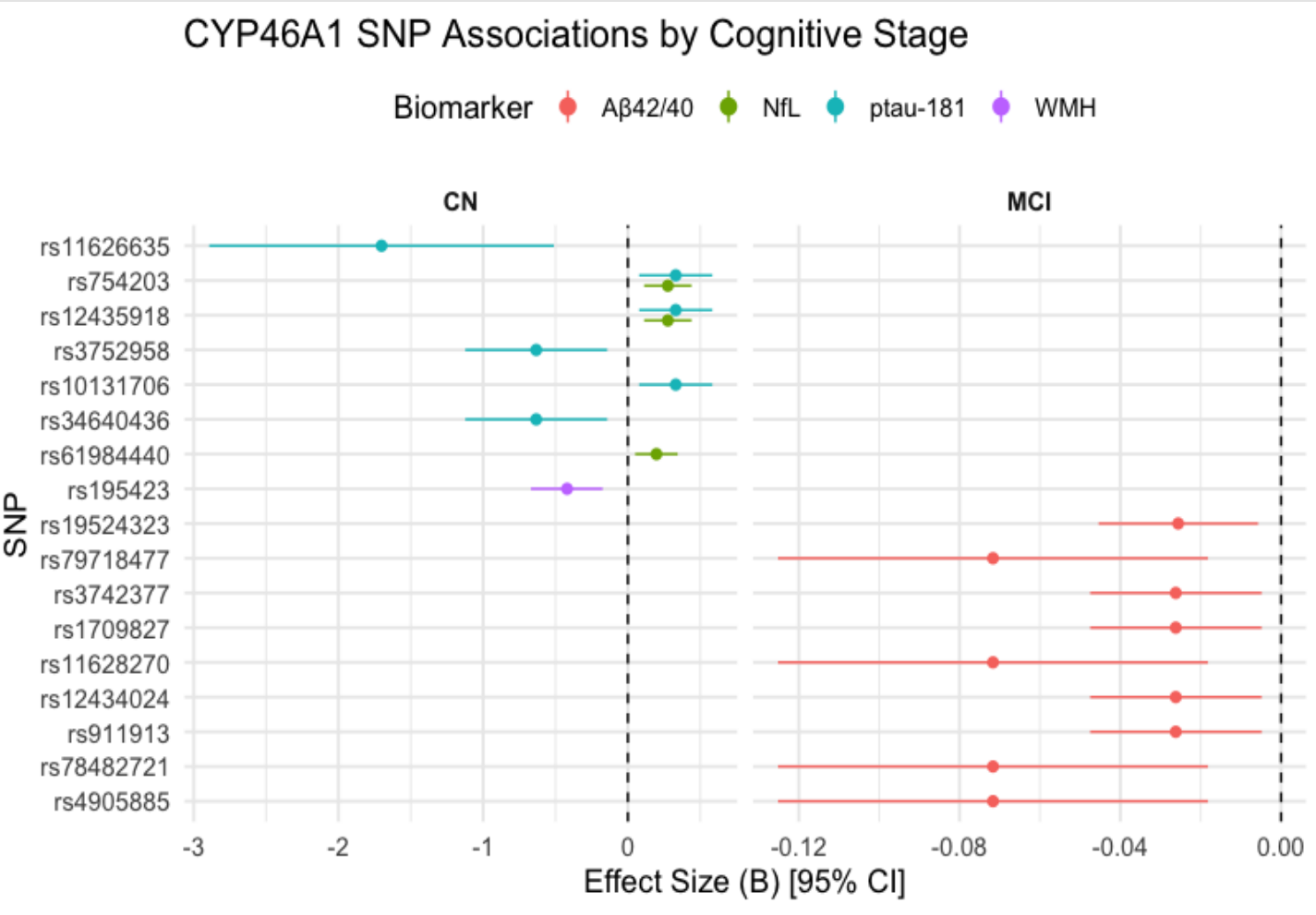


Figure 1: Significant associations between CYP46A1 tag SNPs and biomarkers in CN (left) and MCI (right). In CN (N=381), associations were identified with ptau-181, NfL, and WMH, though these effects were inconsistent. In MCI (N=566), associations were identified with Aβ42/40.

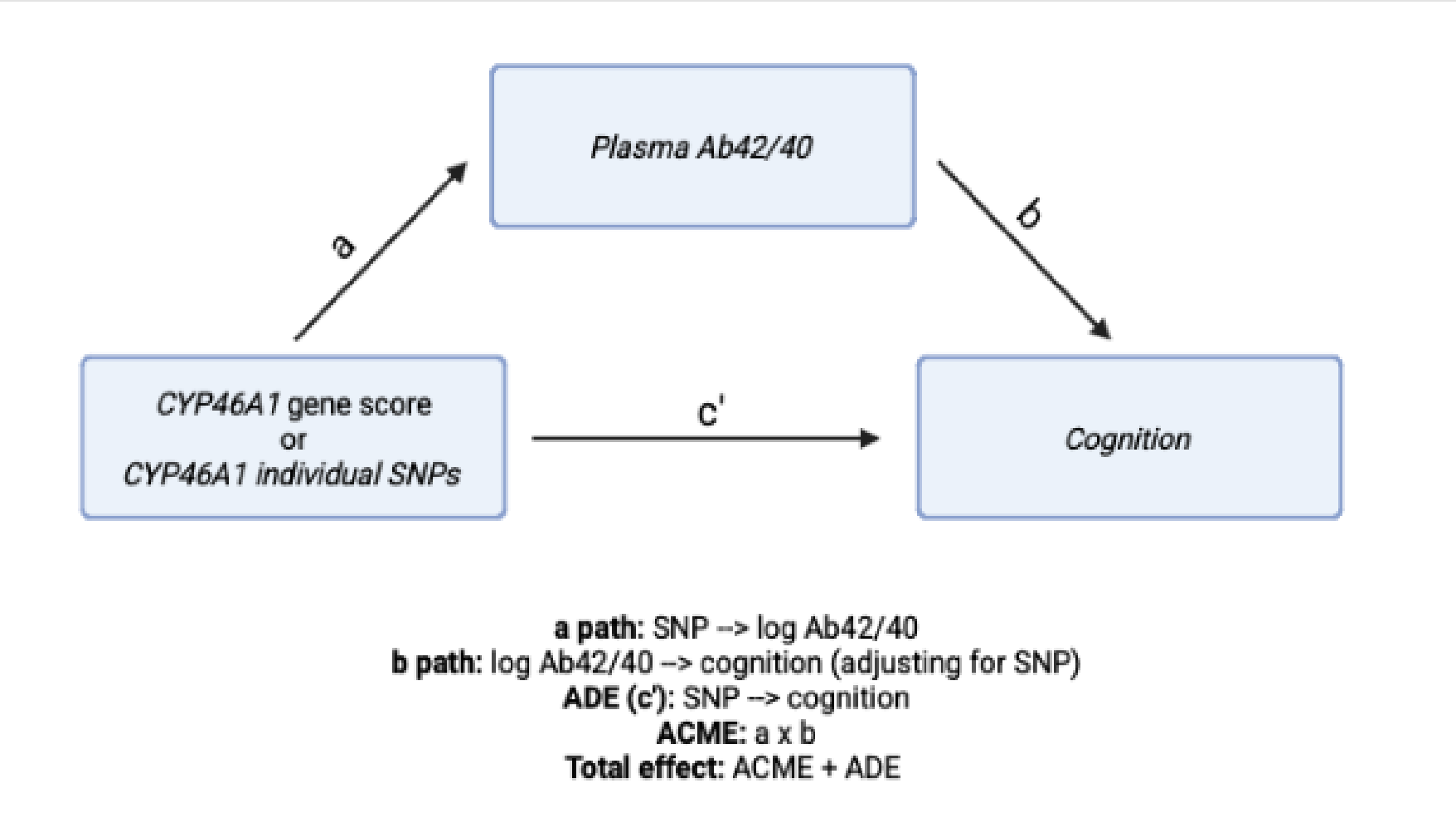


Figure 2: Mediation model testing the indirect effect of CYP46A1 genetic variation on longitudinal cognitive decline through Aβ42/40 in MCI. Plasma Aβ42/40 mediated >40% of the association between CYP46A1 genetic variation and decline in memory, executive function, and language (see Table 2)

Table 2: Indirect effects of CYP46A1 weighted gene score on longitudinal cognitive decline mediated by plasma Aβ42/40				
Cognitive Domain	ADE	ACME	Total effect	% mediated
Memory	-0.00171, p=0.902	-0.00787, p=0.047	-0.00958, p=0.488	82.18%
Executive Function	-0.00703, p=0.533	-0.00505, p=0.047	-0.01208, p=0.313	41.79%
Language	-0.00706, p=0.515	-0.00748, p=0.05	-0.01453, p=0.20	51.47%
Visuospatial function	0.00985, p=0.079	-0.00094, p=0.165	0.00890, p=0.108	-10.56%

Key Findings:

CYP46A1 genetic variation influences plasma Aβ42/40 during MCI, contributing indirectly to subsequent cognitive decline

- Supported by:
- ✓ CYP46A1 variants were associated with plasma Aβ42/40 specifically in MCI.
 - ✓ No consistent associations were observed in the overall cohort or AD.
 - ✓ Plasma Aβ42/40 mediated >40% of the association between CYP46A1 genetic variation and decline in memory, executive function, and language.
 - ✓ Findings support a stage-specific role of cholesterol metabolism in early AD.

Clinical and Research Implications

- Stage-specific effects suggest brain cholesterol metabolism is most relevant during MCI
- Blood-based biomarkers may provide mechanistic insight linking genetics to cognitive decline
- Findings support further evaluation of cholesterol homeostatic pathways via key enzymes (eg: CYP46A1) as potential therapeutic targets

Future Directions

- Independent validation:
- Replicate findings in external cohorts with biomarker-characterized individuals across the AD continuum
 - Confirm stage-specific associations in diverse populations

- Functional characterization:
- Investigate how significant CYP46A1 variants influence gene expression, protein activity, and cholesterol turnover
 - Determine whether variants alter production of 24S-hydroxycholesterol (metabolite of brain cholesterol metabolism), or downstream AD biomarkers

- Expand to other enzymes in the brain cholesterol pathway
- Evaluate additional genes involved in cholesterol synthesis, transport, metabolism, and efflux (eg: APOE, ABCA1, CYP27A1, DHCR24, LRP1)
 - Identify convergent genetic pathways influencing AD biomarkers and cognitive decline

- Clinical translation
- Identify genetically supported therapeutic targets regulating brain cholesterol homeostasis
 - Evaluate whether cholesterol pathway genetics can improve patient stratification or identify individuals most likely to benefit from targeted therapies

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