

Plasma Biomarkers for Synaptic Dysfunction in Alzheimer's Disease: Insights from the NULISA Multiplex Assay



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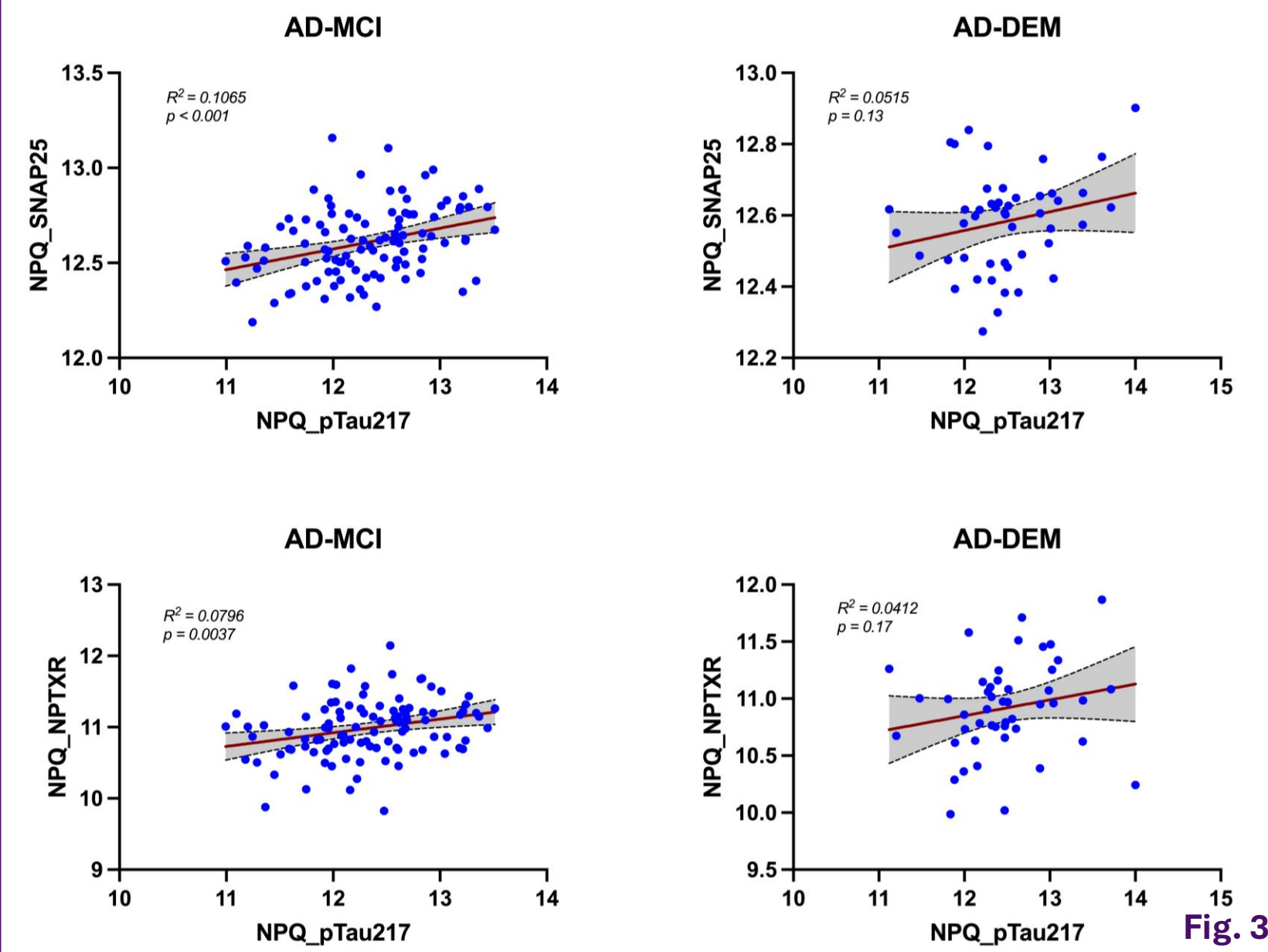
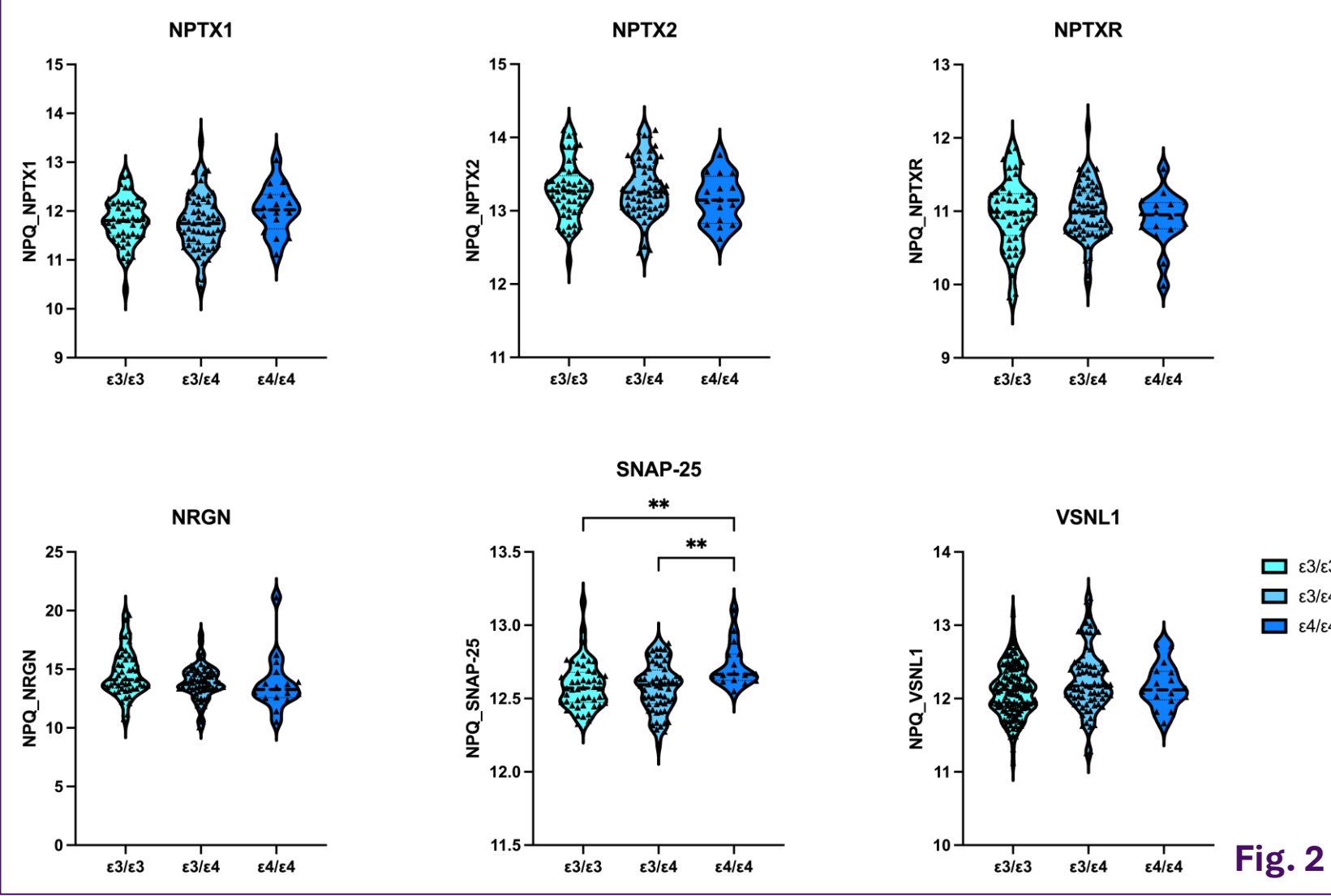
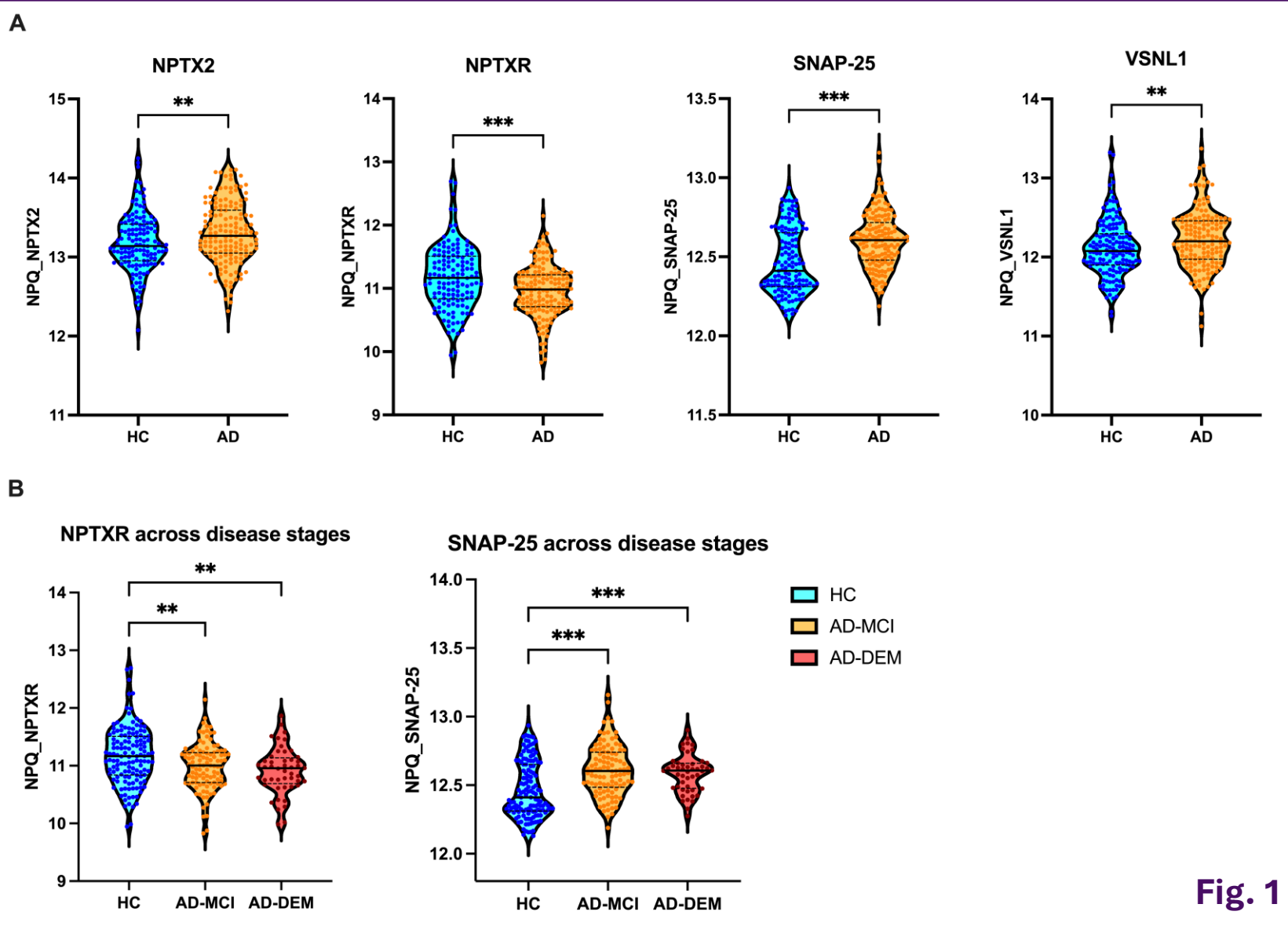
INTRODUCTION

Alzheimer's disease (AD) is characterized by early synaptic dysfunction closely linked to cognitive decline. Synaptic proteins such as SNAP-25, neurogranin, and pentraxins show diagnostic and prognostic value as cerebrospinal fluid (CSF) biomarkers across neurodegenerative diseases¹, but the invasiveness of lumbar puncture limits their clinical use. Blood-based biomarkers offer a scalable alternative, although detecting brain-derived synaptic proteins in plasma is technically challenging. Recent ultra-sensitive immunoassays, including the multiplexed nucleic acid-linked immuno-sandwich assay (NULISA) platform, enable plasma detection of CNS-related proteins², but their utility for assessing synaptic pathology in AD remains to be established.

- The aims of this study were to:
- i. **Assess** plasma detectability and variability of synaptic biomarkers using the NULISA platform
 - ii. **Explore** relationships with *APOE* ε4 status and other AD-related biomarkers
 - iii. **Examine** associations between plasma and CSF synaptic markers

METHODS

The study included 290 subjects, namely 154 CSF biologically confirmed AD patients and 136 controls (HC), who underwent plasma assessment using the NULISA central nervous system (CNS) panel. In a subset of 48 AD patients and 38 HC, the NULISA CNS panel was additionally measured in CSF to evaluate plasma-CSF concordance. From the full panel, only core synaptic proteins detectable in >90% of samples were retained for analysis. Group differences in plasma synaptic biomarker levels were tested with multivariable models adjusted for age, sex, comorbidity burden, and renal function. In the AD subgroup with paired samples, correlations between plasma and CSF biomarker levels were evaluated.



RESULTS

Table 1. Demographic and clinical characteristics of cohort participants.

Demographical and clinical variables	AD (n=154)	HC (n=136)	p-value [#]
Age (years)	72.1 ± 6.9	68.8 ± 6.2	< 0.001
Sex (F) (n, %)	98 (63.6)	75 (55.1)	> 0.93
Disease duration (years)	2.58 ± 1.73	—	
Education (years)	9.0 ± 4.2	11.9 ± 4.0	< 0.001
Cognitive stage (n, %)			
MCI	104 (67.5)		
Dementia	50 (32.5)		
MoCA baseline	16.7 ± 5.9	26.0 ± 3.3	
APOE ε4 carriers (n, %)	77 (60.2)	8 (8.3)	

[#]p values for Mann-Whitney test
Mean ± SD are shown. Significant values are in bold
F female, MoCA Montreal Cognitive Assessment, APOE apolipoprotein E



Of the core synaptic proteins included in the analysis, only NPTX2, NPTXR, SNAP-25, and VSNL1 showed statistically significant differences between AD and HC ($p < 0.05$) (**Fig. 1, panel A**). Notably, only SNAP-25 and NPTXR exhibited consistent differences across disease sub-stages ($p < 0.001$) (**Fig. 1, panel B**).



A significant overall effect of *APOE* genotype was observed for SNAP-25 levels in ε4/ε4 carriers compared with both ε3/ε3 and ε3/ε4 individuals ($p < 0.01$), with no difference between ε3/ε3 and ε3/ε4 (**Fig. 2**). No significant *APOE*-related effects were detected for the other synaptic biomarkers ($p > 0.05$).



Plasma SNAP-25 and NPTXR show significant, stage-dependent associations with pTau217, limited to the MCI stage. Both biomarkers independently predict pTau217 in multivariate models (adjusted $R^2 = 0.109$, $p < 0.01$) (**Fig. 3**).



Plasma – CSF concordance: no significant cross-matrix correlations were observed, supporting matrix-specific biological signatures.

CONCLUSIONS

Automated NULISA-based detection of SNAP-25, NPTXR, and VSNL-1 in plasma offers a promising, minimally invasive approach for monitoring synaptic dysfunction and neuronal injury across the AD spectrum. The APOE ε4-selective modulation of SNAP-25, together with its stage-specific association with pTau217 at the MCI phase, supports a role for these markers in individual disease monitoring and biological subtyping, despite the compartment-specific plasma–CSF dissociation observed. Longitudinal studies are needed to determine their temporal trajectories and clinical utility in tracking disease progression and treatment response.

TAKE-HOME MESSAGES

- ✓ Plasma SNAP-25 is an APOE ε4-sensitive biomarker of synaptic dysfunction in AD.
- ✓ SNAP-25 and NPTXR track pTau217 selectively at the MCI stage, independent of plasma–CSF concordance.
- ✓ NULISA enables scalable, minimally invasive monitoring of synaptic biomarkers across the AD continuum.

FUTURE DIRECTIONS

Future studies should test cross-platform assay reproducibility and whether the MCI-restricted SNAP-25/NPTXR–pTau217 association identifies a window for early therapeutic intervention.

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