

Enhancing pTau217-Based Prediction of Amyloid Pathology in Alzheimer’s Disease with Additional Biomarkers: Insights from a Multi-Cohort Study

¹Wasiu G. Balogun, ²Theresa E. Boots, ²Xuemei Zeng, ²Marissa F. Farinas, ²Michel N. Nafash, ¹Ruyu Shi, ²James E. Emanuel, ³Margaret Bedison, ⁴Cristy Matan, ⁵Neelesh K. Nadkarni, ⁴Alexandra Gogola, ⁶C. Elizabeth Shaaban, ¹Howard J. Aizenstein, ¹Dana L. Tudorascu, ¹Tharick A. Pascoal, ¹Victor L. Villemagne, ⁵Sarah B. Berman, ¹Milos D. Ikonomic, ¹Robert A. Sweet, ⁷Julia Kofler, ⁵Beth E. Snitz, ³M. Ilyas Kamboh, ⁸Anum Saeed, ⁸Steven E. Reis, ¹Ariel Gildengers, ¹Ann D. Cohen, ⁵Oscar I. Lopez, ¹Thomas K. Karikari

¹Department of Psychiatry, University of Pittsburgh; ²Western Psychiatric Institute of Medicine, University of Pittsburgh; ³Department of Genetics, University of Pittsburgh; ⁴Department of Radiology, University of Pittsburgh; ⁵Department of Neurology, University of Pittsburgh; ⁶Department of Health Promotion and Development, University of Pittsburgh; ⁷Department of Pathology, University of Pittsburgh; ⁸Department of Cardiology, University of Pittsburgh

Introduction

- The current method of detecting amyloid-beta (Aβ) using imaging techniques like positron emission tomography (PET) is both invasive and expensive.
- Blood-based biomarkers have shown promise as alternatives to PET due to their greater availability and tolerability for both clinicians and patients.
- Among the various blood-based biomarkers, p-tau217 has emerged as the most reliable and accurate indicator for detecting brain amyloid-beta accumulation^{1,2,3}.
- However, its diagnostic utility may be enhanced when combined with other biomarkers
- This study investigates which biomarkers, when paired with ptau217, can provide a more comprehensive understanding of A β progression and better accuracy of Tau status and cognitive decline

Methodology

- Data from four independent community- and clinical-based cohorts were utilized in this study:
 - Lithium as a Treatment to Prevent Impairment of Cognition in Elders (LATTICE) study began at the University of Pittsburgh in September 2017 as a double-blind, randomized placebo-controlled trial (RCT) study to investigate the potential of neuroprotective potential of lithium in community-dwelling individuals with mild cognitive impairment (MCI). This study included 80 participants, with an Aβ-PET positivity rate of 19%.
 - The Pitt-ADRC study is conducted by the University of Pittsburgh Alzheimer's Disease Research Center (ADRC) in Pittsburgh, Pennsylvania, USA. Participants undergo annual clinical evaluations to monitor their brain health over time and to identify any potential development of cognitive impairment and dementia. This cohort consists of adults with Alzheimer's disease, mild cognitive impairment, and those who are cognitively unimpaired, totaling 1420 participants, with 136 having neuroimaging and Aβ-PET positivity rate of 49%.
 - The Heart Strategies Concentrating on Risk Evaluation Study (Heart SCORE), which involves adults with vascular diseases. This cohort included 147 participants, with an Aβ-PET positivity rate of 17.7%.
 - The Program Project Grant (PPG) Study is an ongoing research initiative at the University of Pittsburgh aimed at examining the pathophysiological mechanisms that contribute to the development of Alzheimer's Disease (AD). The study consists of four sub-projects, with Project 4 (PPG-4) focusing on the analysis of existing banked brain and plasma samples from AD patients to characterize regional differences using the new PET tracer (SMBT-1).
- All participants were recruited from southwestern Pennsylvania.
- Plasma samples were collected and processed at the fluid biomarker lab, utilizing the NULISaseq CNS Disease Panel 120 assay on an Alamar ARGO prototype system at the Department of Psychiatry, University of Pittsburgh, following established protocols^{4,5}
- Aβ PET imaging was performed using 11C-PiB, with global Aβ positivity defined by a threshold of 1.346 SUVR.
- All participants provided written informed consent for all procedures, as approved by the Institutional Review Board of the University of Pittsburgh.
- NULISA Protein Quantification (NPQ) was calculated by dividing the target counts by the internal control counts for each well and then by the median IPC counts, followed by log₂ transformation. Fold changes between groups were determined as 2 raised to the power of the difference in NPQ.
- The average coefficient of variation (CV) for the duplicate measurements of SC across 116 biomarkers was 5.5%.

Statistical Analysis

- Demographic characteristics were presented for each cohort. Continuous variables were reported as median (interquartile range, IQR) and categorical variables were reported as counts (%). Logistic regression models were used to predict amyloid or tau status (positive or negative) and were adjusted for age, sex, race, years of education, APOE ε4 status, and cohort when available
- All NULISA biomarkers were expressed as NPQ values. Bootstrapping with 1000 replicates and stratified sampling by cohort was used to calculate AUC distributions (median AUC presented along with lower 2.5% and upper 97.5% of distribution for 95% confidence interval).

Results

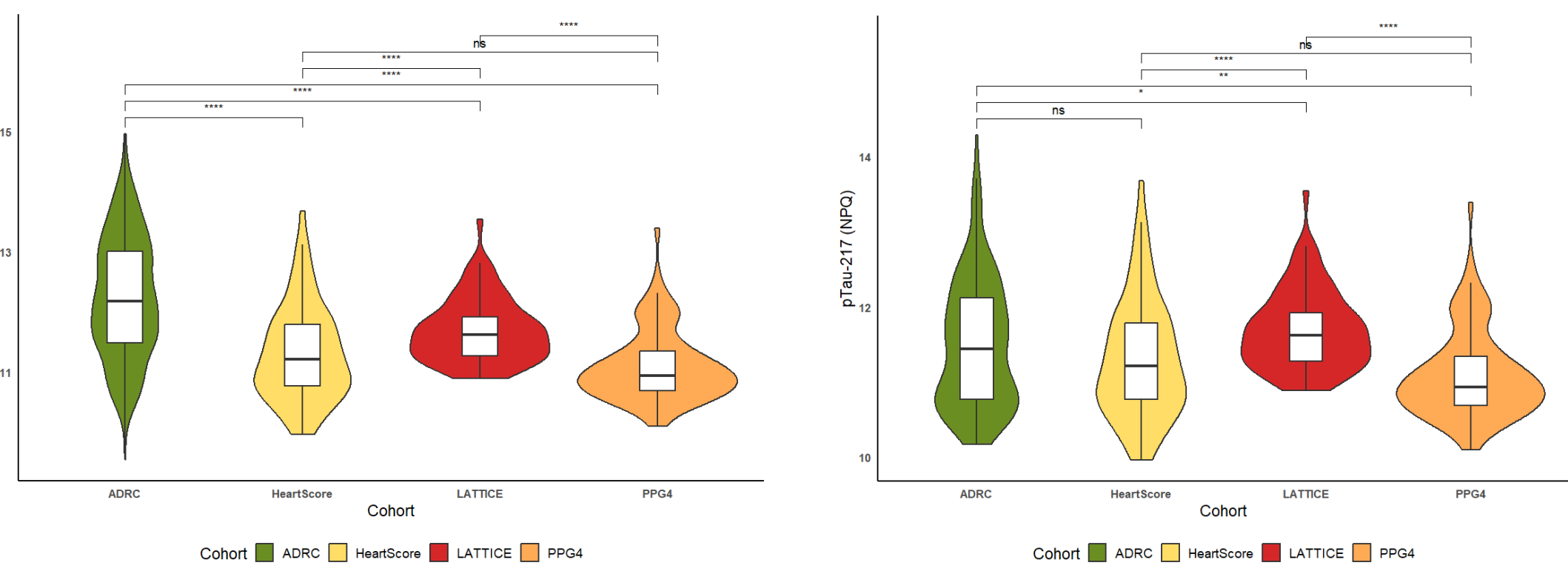


Figure 1: ptau217 to the other cohort. Asterisks indicate significance levels: *, p < 0.05; **, p < 0.01; ***, p < 0.001. **behavior across the cohorts. A showed the ptau217 behavior across the full dataset of the cohort while B showed the behavior of ptau217 in the subset of the full cohort.** The ADRC cohort has higher level of ptau217 due to large number of MCI and Dementia patients included in the cohort compared

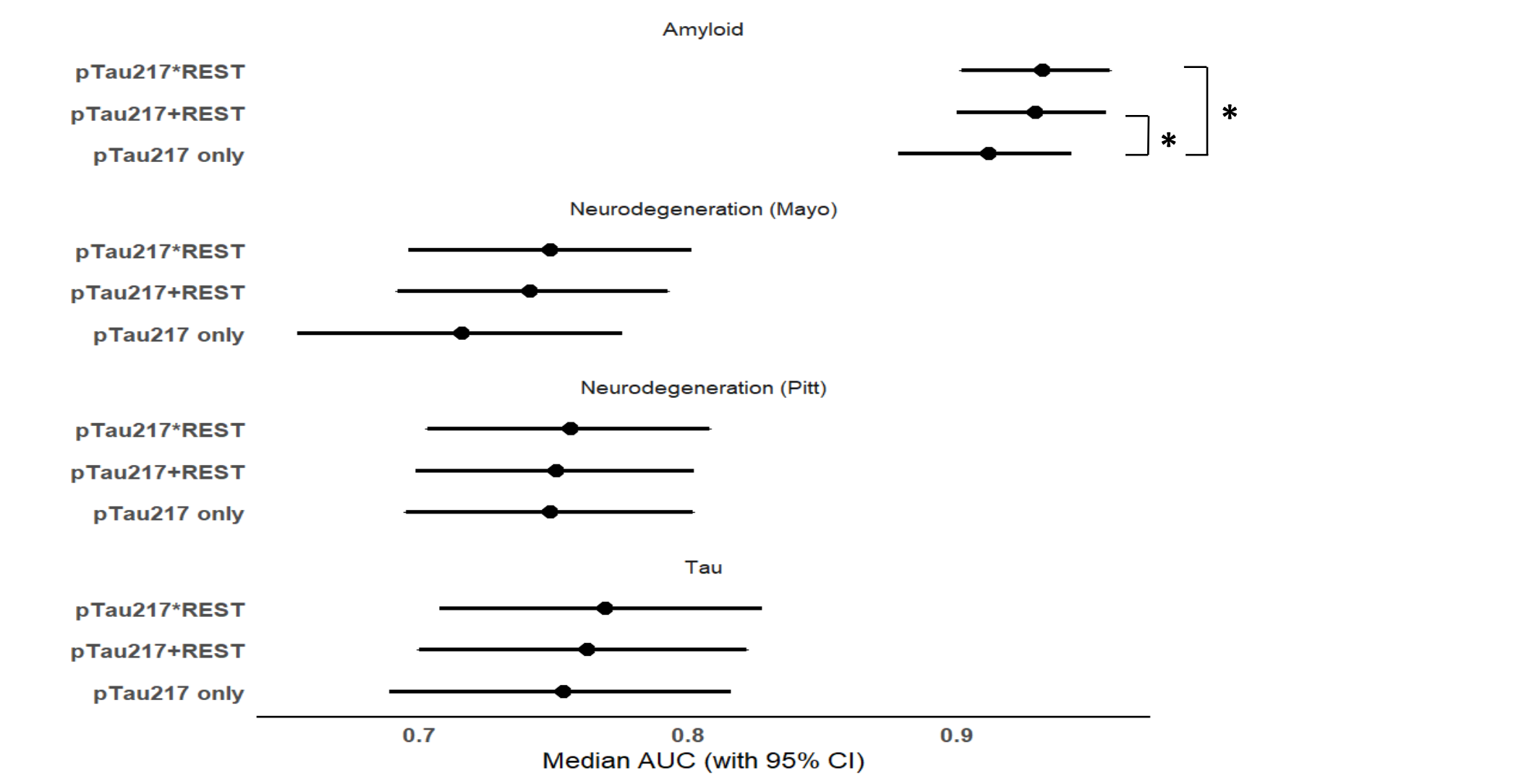


Figure 2: Combination of ptau217 and REST in predicting AD pathology AT(N) when the cohorts were combined. Forest plot of AUC values for ptau217 combined with REST. The combination increased the accuracy of ptau217 in each conditions however, it was only significant in predicting the accuracy of A status. Asterisks indicate significance levels: *, p < 0.05.

Characteristic	ADRC N = 1420	HeartScore N = 139	LATTICE N = 80	PPG4 N = 85	p-value ^a
ADRC					<0.001
Absent	808 (56%)	98 (70%)	0 (0%)	23 (24%)	
Present	592 (42%)	31 (24%)	0 (0%)	6 (25%)	
Unknown	20	10	80	54	
Sex					0.005
Female	745 (53%)	89 (63%)	45 (56%)	44 (52%)	
Male	666 (47%)	40 (29%)	35 (44%)	41 (48%)	
Unknown	9	10	0	0	
Race					<0.001
Asian	7 (0.5%)	0 (0%)	1 (1.3%)	0 (0%)	
Black or African American	111 (7.9%)	40 (29%)	8 (10%)	0 (0.4%)	
Hispanic	0 (0%)	0 (0%)	0 (0%)	1 (1.2%)	
Native Hawaiian or other Pacific Islander	2 (0.1%)	0 (0%)	0 (0%)	0 (0%)	
Other	0 (0%)	0 (0%)	0 (0%)	0 (0.4%)	
White	1,291 (91%)	89 (63%)	71 (88%)	74 (87%)	
Unknown	9	10	0	0	
Age (years at collection)	70.16 (71.64, 77)	74.5 (73.73, 77)	72.8 (71.66, 78)	69.7 (69.62, 73)	<0.001
Unknown	10	10	0	0	
Education	15.22, 13.16, 16.00 (12.00, 16.00)	15.87, 12.72, 16.00 (12.00, 16.00)	15.99, 13.31, 16.00 (14.00, 16.00)	15.91, 2.35, 16.00 (14.00, 16.00)	0.019
Unknown	9	10	0	0	
CDR Score	0.69, 0.62, 0.50 (0.50, 1.00)	0.69, 0.21, 0.00 (0.00, 0.50)	0.50, 0.00, 0.50 (0.50, 0.50)	0.69, 0.16, 0.00 (0.00, 0.50)	<0.001
Unknown	20	25	0	1	

^a Chi-Square Test. Pearson's Chi-Square Test. Pearson's Chi-Square Test with continuity correction. Fisher's Exact Test. Fisher's Exact Test with continuity correction. Fisher's Exact Test with continuity correction. Fisher's Exact Test with continuity correction.

Table 1: Demographic comparison of LATTICE, ADRC, PPG4 and HeartSCORE cohorts. A showed the full cohort of all participants who were enrolled in the study and had their blood taken while B showed subset of the full cohort that had PET imaging and blood taken

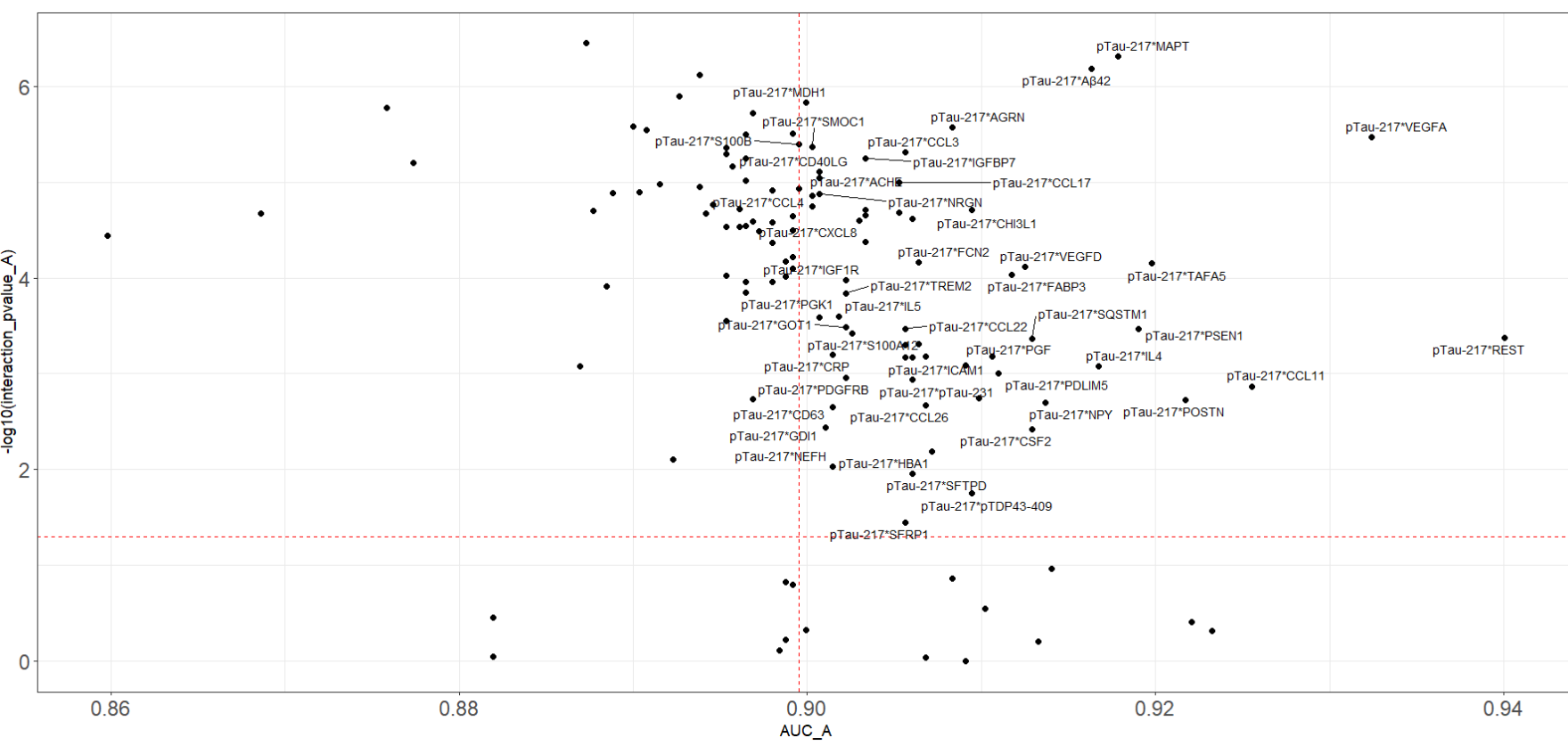


Figure 3: Interaction of full NULISA panel with ptau217 in predicting A status. All models adjusted for Age, Sex, Race, Education, APOE status, and Cohort. Horizontal red line represented p = 0.05 of interaction term in model. Vertical red line represents AUC = 0.8995 (AUC of pTau-217 alone).

Table 2: Longitudinal analysis of ptau217 and REST. Analysis of cognitive decline in ADRC cohort (only individuals that were Cognitively Normal at their first visit; n =322). Cognitive Decline defined as progression in CDR score

pTau-217 Level	REST Level	Hazard Ratio (95% CI)	p-value	Median time to Cognitive Decline
High	High	(baseline)	-	10.375
	Low	1.5121 (1.0166-2.2492)	0.041228	8.496
Low	High	0.4713 (0.3061-0.7256)	0.000635	18.047
	Low	0.6258 (0.4281-0.9146)	0.015483	13.923

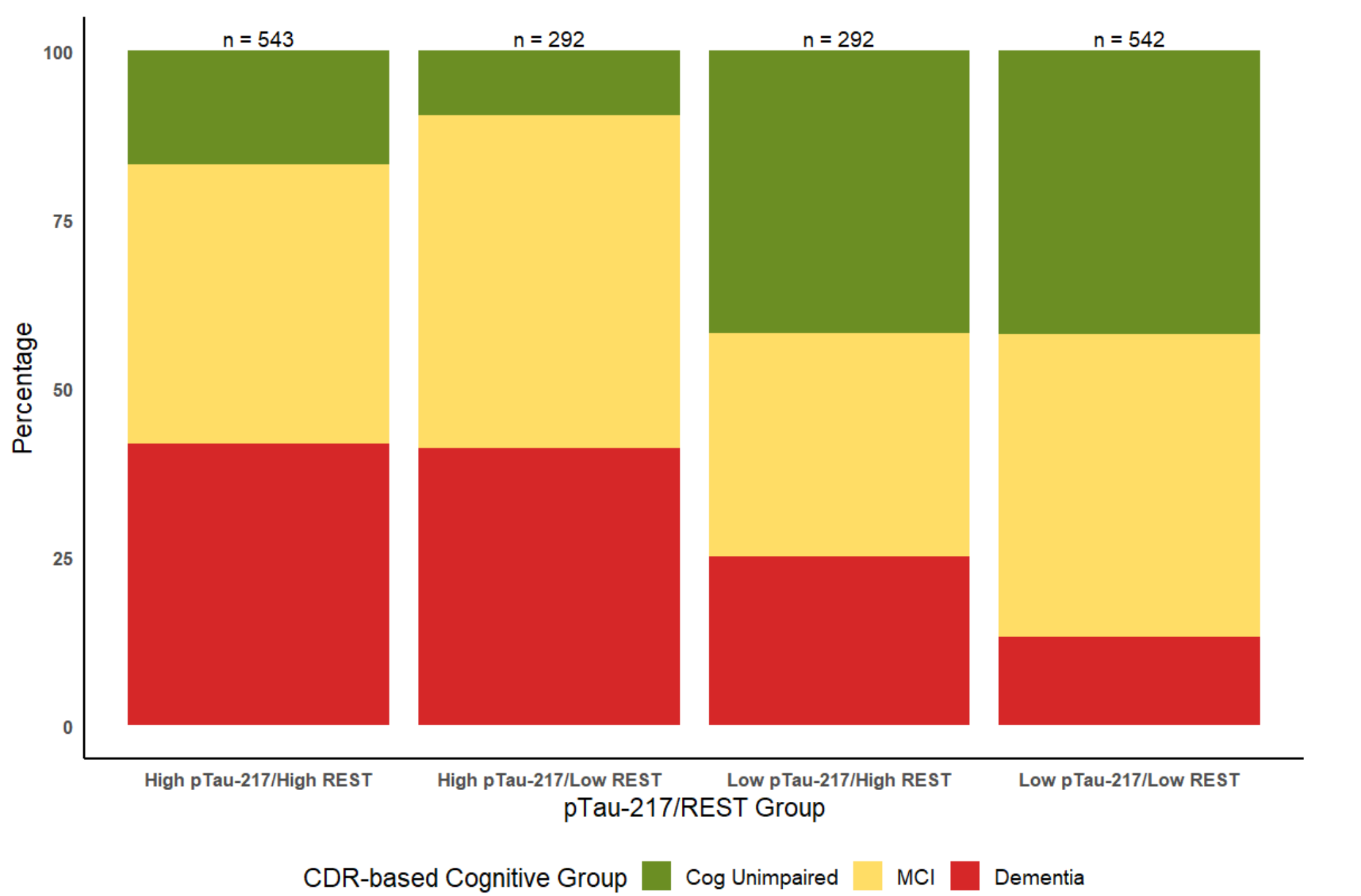


Figure 4: Association of the different levels of ptau217 and REST with cognitive impairments. Distribution of Cognitive group proportions do NOT differ statistically between High/Low REST among High pTau217 groups (X2 = 2.641, DF = 2, p = 0.267), but are just above statistical significance among the Low pTau217 groups (X2 = 5.4464, df = 2, p = 0.06566)

Conclusion

- One key takeaway from this study is that among the more than 120 biomarkers studied, REST protein appears most predicting in predicting amyloid status when combined with ptau217
- The combination also improved the accuracy of tau status and neurodegeneration although it was only significant in predicting amyloid accuracy
- The combination of High pTau-217+Low REST had the lowest survival in predicting cognitive decline, followed by High pTau-217+High REST, Low pTau-217+Low REST, and Low pTau-217+Low REST.
- Our result showed that High level of REST is protective against AD
- Finally, many of these cohorts are part of ongoing studies, and longitudinal follow-up studies will be conducted on them.

References

- Milà-Alomà, M., Ashton, N.J., Shekari, M. *et al. Nat Med* **28**, 1797–1801 (2022).
- Brum WS, Cullen NC, Janelidze S, Ashton NJ, *et al. Nat Aging*. 3(9):1079-1090 (2023).
- Rissman RA, Langford O, Raman R, *et al. Alzheimers Dement*. 20(2):1214-1224 (2024).
- Zeng, X., Lafferty, T. K., Sehrawat, A. et al. Mol neurodegener. 19(1), 68 (2024).
- Feng W, Beer JC, Hao Q, et al. Nat Commun. 14(1):7238 (2023).

Acknowledgement

Funding Sources: The Pitt-ADRC is funded by P30 AG066468. This study used biomarker testing infrastructure established with support from NIH/NIA R01AG083874.