Supplemental Online Content

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eAppendix

This supplemental material has been provided by the authors to give readers additional information about their work.

1. eAppendix

1. *MRI and PET Acquisition*

For ADNI, structural MRI was acquired on 3T Siemens (SIEMENS Healthineers, Erlangen, Germany) and 3T GE scanners. T1-weighted structural scans were collected using an MPRAGE sequence (TR=2300ms; Voxel size=1x1x1mm; for parameter details see: https://adni.loni.usc.edu/wp-content/uploads/2017/07/ADNI3-MRI-protocols.pdf). PET data was assessed post intravenous injection of ^{18}F-labeled tracers (Flortaucipir: 6x5min time-frames, 75-105min post-injection; Florbetapir: 4x5min time-frames, 50-70min post-injection; Florbetaben: 4x5min time-frames, 90-110min post-injection; for more information see http://adni.loni.usc.edu/methods/pet-analysis-method/pet-analysis/).

In A05, PET data was assessed post intravenous injection of ^{18}F-labeled tracers (^{18}F-flortaucipir: 4x5min time-frames, approx. 80min post-injection; ^{18}F-florbetapir: 2x5min time-frames, approx. 50min post-injection)

2. PET preprocessing

For ADNI, dynamically acquired tau-PET images were realigned and averaged to obtain single Flortaucipir images. Using brain extracted T1-weighted images and ANTs-derived non-linear spatial normalization parameters[68], tau-PET images were affine registered to T1-weighted images, spatially normalized to MNI space and intensity normalized using an inferior cerebellar grey reference[69]. For A05, Tau-PET images were preprocessed by Avid investigators. Specifically, native-space tau-PET images were rigidly co-registered to T1-weighted structural MRI and spatially normalized to MNI standard MNI space using FSL 'fnirt' (https://fsl.fmrib.ox.ac.uk/fsl/fslwiki/FNIRT). SUVRs were obtained by intensity normalization to the inferior cerebellum.

All tau-PET images were then parcelled into 200 Schaefer atlas cortical regions of interest (ROIs) by averaging voxels falling within a given ROI[38]. We specifically chose this atlas because of its

exclusion of areas susceptible to Flortaucipir tracer off-target binding[70,71]. The atlas was masked using a group-specific grey matter mask binarized at 0.3 probability.

3. ***Resting-state fMRI acquisition and preprocessing***

rs-fMRI was obtained using a 3D echo-planar imaging (EPI) sequence with a total of 200 fMRI volumes per subject (TR=3000ms; TE=30ms; flip angle=90°; Voxel size=3.4mm isotropic).

To determine a connectivity template for modelling connectivity-based tau spreading, ten minutes of 3T resting-state fMRI data recorded on Siemens scanners (TR=3s) of 42 CN subjects (age: 72±7.24 years; 26 females) without evidence of AD pathology (i.e. global ^{18}F-Florbetapir SUVR<1.11) were included. The images were slice-time and motion corrected and co-registered to native T1-weighted images. To denoise EPI images, we regressed out nuisance covariates of eroded white matter and cerebrospinal fluid segments plus six motion parameters and their time and dispersion derivatives, followed by detrending and band-pass filtering (0.01-0.08Hz). To further reduce movement artifacts which may compromise connectivity assessment [72] we performed motion scrubbing, removing volumes exceeding a 0.3mm frame-wise displacement threshold, plus one prior and two subsequent volumes. Pre-processed fMRI images were subsequently normalized to MNI space using ANTs.

2. SUPPLEMENTAL TABLES

eTable 1. Demographic and Clinical data stratified by ApoE carriage

	ε4–	ε4+	p-value
	ADNI		
N	130	107	
Sex (F/M)	64/66	61/46	0.29
Age	74.8 (7.03)	72.8 (7.61)	0.04
Clinical Diagnosis (CN/MCI)	87/43	65/42	0.39
Aβ Positivity (-/+)	$90^{b,c}/40^{a,d}$	$37^{a,d}/70^{b,c}$	<0.001
Education (years)	16.6 (2.52)	16.3 (2.44)	0.37
Mean tau-PET follow-up (years)	2.07 (1.09)	1.78 (0.738)	0.02
Mean BL Aβ & tau-PET difference (years)	-0.05 (0.20)	-0.01 (0.25)	0.145
MMSE	29.0 (1.39)	28.1 (1.83)	0.002
	A05		
N	85	45	
Sex	35/50	25/20	0.17
Age	71.6 (9.89)	67.6(9.02)	0.03
Clinical Diagnosis (CN/MCI)	$42^{c,b}/43^{a}$	$10^{a}/35$	0.004
Aβ Positivity (-/+)	$63^{c}/22$	$21^{a,d}/24^{c}$	0.003
Education	15.4 (2.45)	15.8 (2.02)	0.32
Mean tau-PET follow-up (years)	1.45(0.193)	1.37 (0.290)	0.06
MMSE	28.8 (1.46)	28.0 (1.83)	0.01

M male, F female, CN cognitively normal, MCI mildly cognitively impaired, MMSE Mini Mental State Examination, BL Baseline
different from— a CN ε4–, b MCI ε4–, c CN ε4+
different from— a Aβ- ε4–, b Aβ+ ε4-, c Aβ- ε4+, d Aβ- ε4+ (significance level 0.05 Bonferroni corrected)
Mean BL amyloid & tau-PET difference (years) not included for A05; all BL amyloid scans within 30 days of BL tau-PET scans.
Values are presented as mean (SD), Amyloid positivity according to threshold of 1.1 amyloid-PET SUVR.
p-values were derived from ANOVA for continuous measures and from Chi squared tests for categorical measures.
A05 mean BL Aβ & tau -PET differences (year) were not assessed as initial Aβ scans were carried out within 30 days of initial tau-PET scan.
12 A05 subjects did not have available education data.

eTable 2. Regression model ANOVA & Akaike information criterion

	F	p-value	AIC	
			Quadratic	Linear
		Braak Stages		
		ADNI		
Braak III	6.83	0.001	-1270.3	-1260.5
Braak IV	6.58	0.002	-1223.2	-1213.9
Braak V	3.43	0.034	-1157.6	-1154.6
Braak VI	1.43	0.242	-1301.7	-1302.74
		A05		
Braak III	2.58	0.080	-847.1	-845.7
Braak IV	0.86	0.427	-839.3	-841.5
Braak V	2.44	0.091	-710.1	-708.9
Braak VI	0.37	0.690	-743.7	-746.9
		Connectivity Stages		
		ADNI		
Q1	8.14	0.000	-1200.7	-1188.3
Q2	3.37	0.036	-1381.1	-1378.1
Q3	3.37	0.036	-1454.4	-1454.8
Q4	0.96	0.383	-1505.5	-1507.5
		A05		
Q1	1.95	0.147	-703.8	-703.7
Q2	3.74	0.027	-719.9	-716.1
Q3	1.49	0.229	-738.5	-739.3
Q4	0.02	0.981	-819.7	-823.6

AIC Akaike information criterion

The table displays F, and p-values derived from ANOVAs fitted quadratic and linear regression models fitted with interaction effect of ApoE4 risk and Centiloid or Centiloid^2 on the rate of annual tau accumulation in respective Braak stages and connectivity stages in ADNI and A05. The table also displays AICs for the respective quadratic and linear models.

eTable 3. Mediation Results controlled for clinical diagnosis

	B	CI L	CI U	p-value
		ADNI		
Global	0.14	0.03	0.27	0.006
Braak I	0.05	0.17	-0.06	0.38
Braak III	0.19	0.31	0.06	<0.001
Braak IV	0.169	0.29	0.06	0.002
Braak V	0.119	0.24	0.01	0.04
Braak VI	0.10	0.21	-0.02	0.10
		A05		
Global	0.25	0.1	0.44	<0.001
Braak I	-0.01	-0.14	0.12	0.918
Braak III	0.24	0.09	0.43	<0.001
Braak IV	0.20	0.07	0.38	<0.001
Braak V	0.26	0.11	0.45	<0.001
Braak VI	0.21	0. 07	0.39	<0.001

CI L 95% Confidence interval lower, CI U 95% Confidence interval upper.
Values are ACME values derived from mediation analyses with ApoE4 risk as predictor, centiloid as mediator, and the annual tau SUVR rate of change (ROC) in the respective Braak stage as the dependent variable. The table displays beta-estimates and p-values. The mediation models are controlled for age, sex and clinical diagnosis.

eTable 4. Interaction effects estimated by linear regression controlled for clinical diagnosis

	β	t-value	p-value	Par. R^2	Lower cut-point			Upper cut-point		
					mean	CI L	CI U	mean	CI L	CI U
Braak Stages										
ADNI										
Braak III	-0.34	-3.34	<0.001	0.05	16.10	15.43	16.76	75.90	75.05	76.75
Braak IV	-0.36	-3.45	<0.001	0.05	12.44	11.74	13.15	74.20	73.33	75.06
Braak V	-0.27	-2.58	0.010	0.03	12.43	11.43	13.42	79.80	78.76	80.84
Braak VI	-0.18	-1.63	0.105	0.014	–	–	–	–	–	–
A05										
Braak III	-0.26	-2.14	0.035	0.04	15.42	14.14	16.70	60.47	58.32	62.61
Braak IV	-0.15	-1.24	0.219	0.02	–	–	–	–	–	–
Braak V	-0.26	-2.19	0.030	0.04	11.03	9.89	12.17	63.96	61.82	66.0
Braak VI	-0.09	-0.71	0.482	0.03	–	–	–	–	–	–
Connectivity Stages										
ADNI										
Q1	-0.38	-3.72	<0.001	0.06	11.20	10.61	11.80	83.60	82.74	84.45
Q2	-0.26	-2.56	0.011	0.03	13.29	12.39	14.19	80.77	79.72	81.83
Q3	-0.20	-1.86	0.064	0.02	–	–	–	–	–	–
Q4	-0.13	-1.19	0.234	0.01	–	–	–	–	–	–
A05										
Q1	-0.23	-1.95	0.053	0.03	13.31	12.25	14.37	69.70	67.67	71.74
Q2	-0.31	-2.62	0.010	0.07	11.93	10.86	13	56.48	54.38	58.57
Q3	-0.20	-1.65	0.102	0.04	–	–	–	–	–	–
Q4	-0.02	-0.19	0.846	0.02	–	–	–	–	–	–

CI L 95% Confidence interval lower, CI U 95% Confidence interval upper

Values derived from regressions fitted with the interaction effect of ApoE4 risk and Centiloid[2] on the rate of annual tau accumulation in respective Braak stages and connectivity stages in ADNI and A05. Lower and upper cut-points means and CI values estimated through selecting the point of no overlap of 95% and re-overlap of confidence intervals of bootstrapped regressions. The table displays standardized β-estimates, T-values, p-values and partial R squared. The regression models are controlled for age, sex and clinical diagnosis.

eTable 5. Demographic and Clinical data for dementia subjects stratified by ApoE4 carriage

	ε4–	ε4+	p-value
ADNI			
N	15	19	
Sex (F/M)	8/7	5/14	0.21
Age	77.7 (7.70)	75.8 (8.17)	0.485
Aβ Positivity (-/+)	5/10	2/17	0.228
Education (Years)	14.1 (2.53)	16.8 (2.53)	0.005
Mean tau-PET follow-up time (years)	1.76 (0.62)	1.49 (0.59)	0.203
Tau-PET BL amyloid-PET time difference (years)	-0.0493 (0.150)	-0.041 (0.075)	0.839
MMSE	22.2 (5.97)	21.8 (5.59)	0.916
A05			
N	16	19	
Sex	9/7	10/9	1
Age	72.9 (10.7)	75.5 (6.59)	0.383
Aβ Positivity (-/+)	9/7	2[b]/17[a]	0.011
MMSE	21.8 (2.86)	22.3 (4.37)	0.661
Education	15.4 (1.75)	15.5 (2.18)	0.825
Mean tau-PET follow-up time (years)	1.41 (0.256)	1.50 (0)	0.119

M male, F female, MMSE Mini Mental State Examination, BL Baseline

Different from— a ε4+Aβ+, b ε4+Aβ– (significance level 0.05 Bonferroni corrected)

Mean BL amyloid & tau-PET difference (years) not included for A05; all BL amyloid scans within 30 days of BL tau-PET scans.

Values are presented as mean (SD), Amyloid positivity according to threshold of 1.1 amyloid-PET SUVR.

p-values were derived from ANOVA for continuous measures and from Chi squared tests for categorical measures.

A05 mean BL Aβ & tau -PET differences (year) were not assessed as initial Aβ scans were carried out within 30 days of initial tau-PET scan.

2 A05 subjects did not have available education data.

eTable 6. Interaction effects estimated by linear regression in CN, MCI & Dementia subjects

	β	t-value	p-value	Adj. p-value	Par. R^2	Lower cut-point			Upper cut-point		
						mean	CI L	CI U	mean	CI L	CI U
Braak Stages											
ADNI											
Braak III	-0.33	-3.38	0.001*	0.002	0.04	15.40	14.70	16.09	76.67	75.69	77.65
Braak IV	-0.34	-3.58	0.000*	0.002	0.05	11.67	11.01	12.32	80.38	79.61	81.16
Braak V	-0.29	-3.02	0.003*	0.004	0.03	12.05	11.18	12.93	84.07	83.18	84.95
Braak VI	-0.29	-2.88	0.004*	0.004	0.03	12.31	11.40	13.23	79.57	78.60	80.53
A05											
Braak III	-0.25	-2.30	0.023*	0.046	0.05	10.88	9.93	11.83	55.80	54.08	57.52
Braak IV	-0.20	-1.72	0.088	0.118	0.02	–	–	–	–	–	–
Braak V	-0.25	-2.39	0.018*	0.046	0.05	7.27	6.54	8.01	57.12	55.62	58.61
Braak VI	-0.10	-0.93	0.356	0.356	0.04	–	–	–	–	–	–
ADNI											
Q1	-0.38	-3.91	0.000*	0.000	0.05	9.13	8.57	9.70	84.17	83.40	84.94
Q2	-0.33	-3.47	0.001*	0.001	0.04	9.87	9.24	10.50	83.39	82.51	84.27
Q3	-0.24	-2.43	0.016*	0.021	0.02	13.20	12.11	14.29	81.43	80.35	82.51
Q4	-0.15	-1.48	0.141	0.141	0.01	–	–	–	–	–	–
A05											
Q1	-0.24	-2.31	0.022*	0.045	0.04	9.70	9.02	10.37	60.28	58.80	61.77
Q2	-0.30	-2.85	0.005*	0.020	0.07	8.58	7.92	9.24	55.65	54.32	56.98
Q3	-0.18	-1.62	0.108	0.144	0.04	–	–	–	–	–	–
Q4	-0.07	-0.56	0.573	0.573	0.02	–	–	–	–	–	–

CI L 95% Confidence interval lower, CI U 95% Confidence interval upper

Values derived from regressions fitted with the interaction effect of ApoE4 risk and Centiloid^2 on the rate of annual tau accumulation in respective Braak stages and connectivity stages in ADNI and A05 IN CN, MCI and dementia subjects. Lower and upper cut-points means and CI values estimated through selecting the point of no overlap of 95% and re-overlap of confidence intervals of bootstrapped regressions. The table displays standardized β-estimates, T-values, p-values and partial R squared. The regression models are controlled for age and sex. *p-values that fall below an FDR-corrected p-value of 0.05

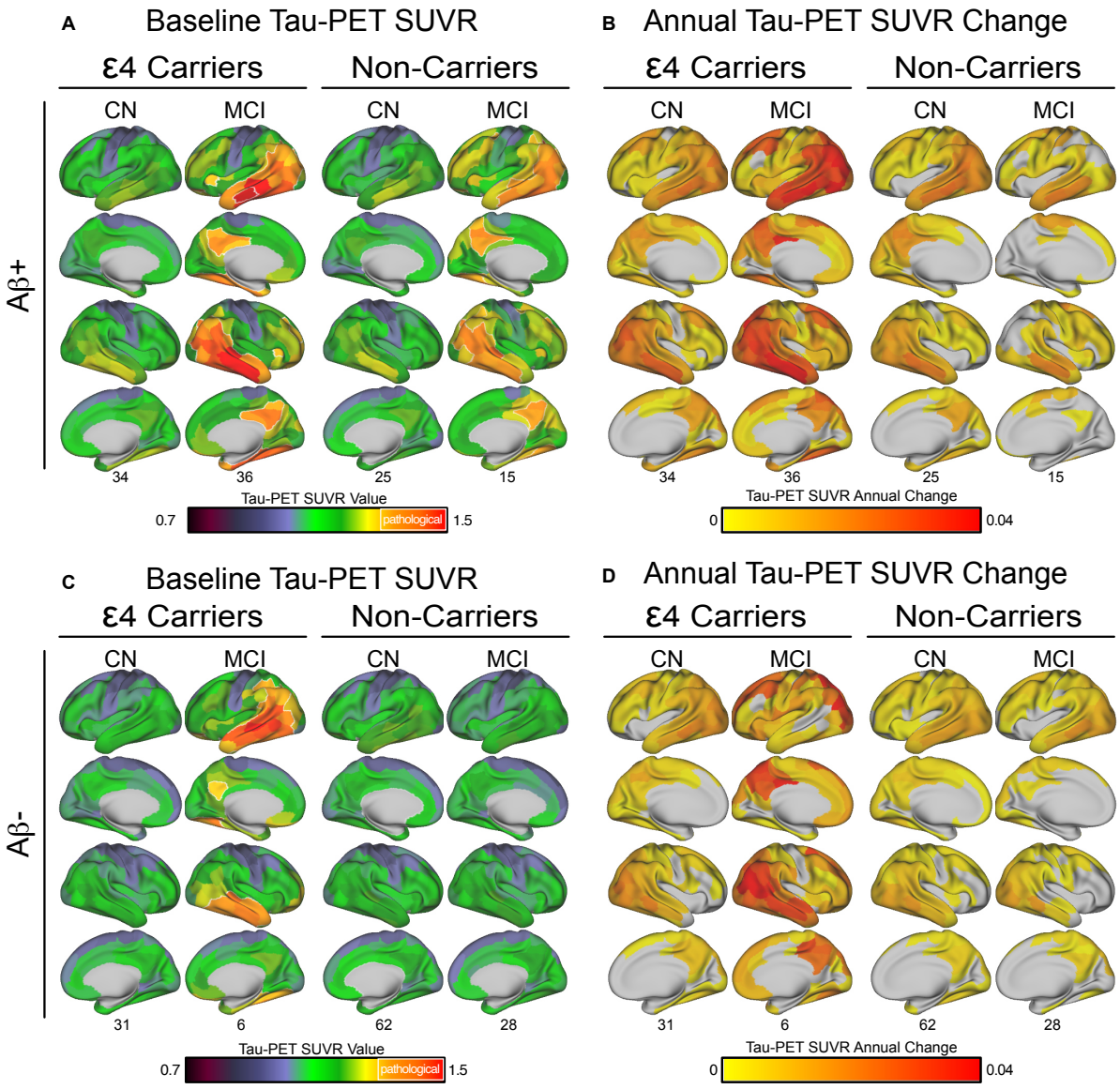
eTable 7. Effect of Annual tau-SUVR change on MMSE change

	β	t-value	p-value	par. R^2
		ADNI		
CN, MCI	-0.32	-5.2	<0.001	0.10
CN, MCI & Dementia	-0.3	-5.12	<0.001	0.09
		A05		
CN, MCI	-0.28	-3.24	0.002	0.08
CN, MCI & Dementia	-0.39	-5.38	<0.001	0.15

Values derived from regressions fitted with the global rate of annual tau accumulation on the annual change in MMSE a sample of CN and MCI subject, and additionally dementia subjects. The table displays standardized β-estimates, T-values, p-values and partial R squared. The regression models are controlled for age and sex.

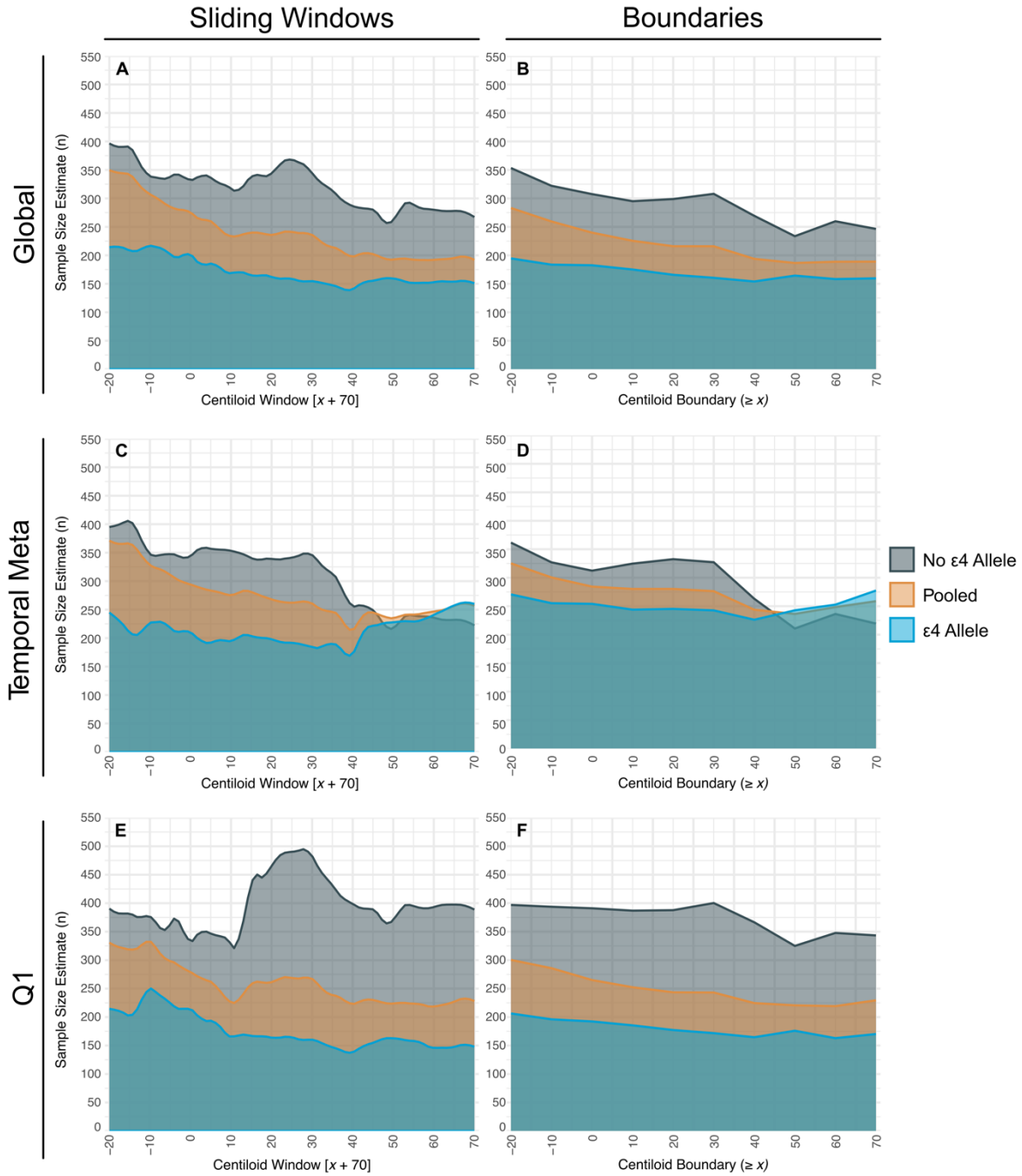
3. SUPPLEMENTAL FIGURES

eFigure 1



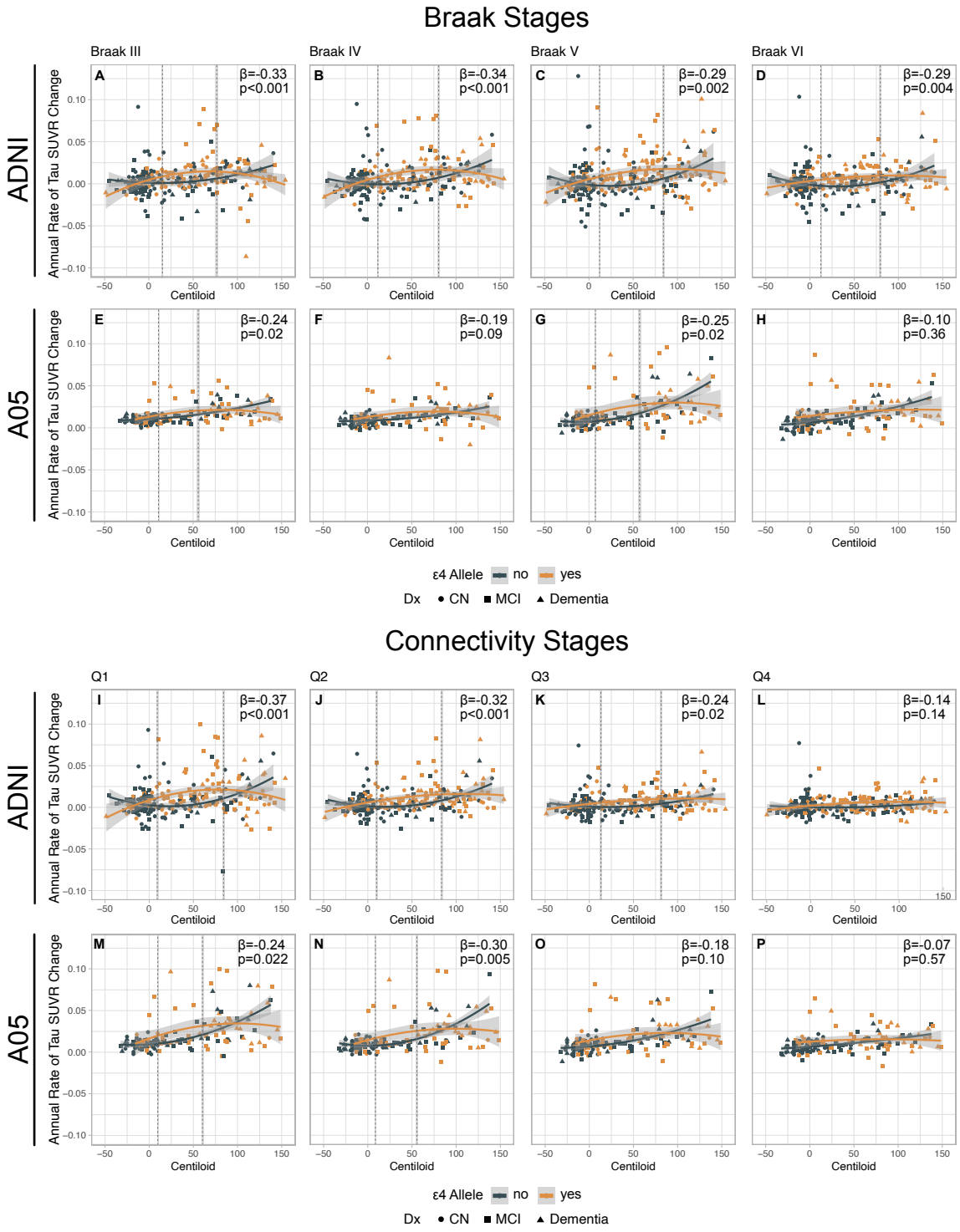
Group-average tau-PET SUVRs in ADNI at baseline stratified by ApoE4 carriership, diagnostic group and Aβ positivity. Tau-PET SUVRs are showed as continuous values, white outlines define areas which surpass a pre-established pathological tau-PET SUVR threshold of 1.3[73] in Aβ+ subjects (A) and Aβ –subjects (C). Number of subjects displayed under each group rendering, Group average tau-SUVR annual change rates defined by linear mixed models, stratified by ApoE4 carriership and diagnostic group in Aβ+ subjects (B) and Aβ – subjects (D).

eFigure 2



Required sample sizes to detect simulated intervention effects with tau change as an endpoint estimated through sliding windows of centiloid starting at [-20;50] moving up in steps of 1 to [100;140] estimated at 30 (A, C, E) and through centiloid boundaries capturing all subjects with a centiloid the same or higher as the boundary starting at -20 increasing in steps of 10 (B, D, F). Sample sizes were estimated according to 3 different tau-PET readout regions: Global (A, B) i.e. average across all 200 cortical Schaefer ROIs, Temporal Meta (C, D) and Q1 (E, F). Results demonstrate that non-e4 carriers may require higher sample sizes to detect intervention effects throughout the centiloid scale, but particularly at earlier centiloid levels.

eFigure 3



Scatterplots illustrating the interaction effect between ApoE4 status and centiloid on the annual rate of tau SUVR change in CN, MCI & Dementia subjects through braak stages III to VI in ADNI (A, B, C, D) and A05 (E, F, G, H) and through connectivity stages Q1 to Q4 in ADNI (I, J, K, L) and A05 (M, N, O, P) showing that ApoE4 carriers show an amyloid-related increase in tau accumulation in early disease stages. Vertical dashed lines represent centiloid threshold of when

groups diverge and converge, estimated according to a non-parametric bootstrapping technique with 1000 iterations identifying the point of where confidence intervals around regression lines diverge and converge, presented with shaded 95%CI threshold.