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eMethods

Postmortem pathological assessments of non-AD brain pathologies.

Transactive response DNA binding protein-43 (TDP-43). Using an antibody against phosphorylated TDP-43, brain sections were examined for the presence of TDP-43 aggregates, which were summarized by a binary variable indicating presence of TDP-43 aggregates in hippocampus, entorhinal cortex, or other brain cortices[1]. The examined brain regions were the amygdala, hippocampus (CA1, subiculum), dentate gyrus, entorhinal cortex, and neocortices (orbital frontal, midfrontal, anterior temporal, and middle temporal cortices).

Hippocampal sclerosis. Coronal sections of mid-hippocampus were examined for hippocampal sclerosis, which was summarized by a binary variable indicating severe neuronal loss and gliosis in CA1 and/or subiculum[2].

Lewy bodies. Immunohistochemical staining using antibodies against α-synuclein were in place for identification of Lewy bodies, which were summarized by a binary variable indicating presence of Lewy bodies in any brain region. The examined brain regions were midfrontal, midtemporal, inferior parietal, anterior cingulate, entorhinal, hippocampus, basal ganglia, and midbrain[3].

Macroinfarcts. Sections of the fixed hemispheres and photos of slabs of frozen hemispheres were examined for the presence of infarcts, which were confirmed by microscopic examination. In the current study, we only included chronic infarcts because we were interested in chronic pathologies that were accumulating over time and were more representative of the pathologies underlying cognitive decline in older adults, as done in our previous publications[4]. Chronic macroinfarcts were pathologically diagnosed as cavities surrounded by gliosis and containing few macrophages, and were summarized by a binary variable indicating presence of one or more macroinfarcts.

Microinfarcts. Brain sections stained by Hematoxylin and Eosin were examined for microinfarcts, which are identifiable by definition only under microscope. Similar to macroinfarcts, only chronic microinfarcts were included in the current study that were summarized by a binary variable indicating presence of one or more microinfarcts. The examined brain regions were six cortical brain regions (frontal, temporal, entorhinal, hippocampal, parietal, and anterior cingulate), two subcortical regions (anterior basal ganglia and thalamus), and the midbrain[4].

Intracranial atherosclerosis. Large arteries of Circle of Willis and their proximal branches were examined for atherosclerosis, which was scored using a semiquantitative scale including none, mild, moderate, severe that was based on the severity of atherosclerosis in each vessel and number of vessels involved. In the current study, we used a binary variable indicating presence of moderate to severe atherosclerosis[5].

Basal ganglia arteriolosclerosis. Small arteries and arterioles in the anterior basal ganglia region (including caudate, putamen, internal capsule, and adjacent globus pallidus) were examined for arterial wall thickening and lumen narrowing, which was scored using a semiquantitative scale

(none, mild, moderate, severe). In the current study, arteriolosclerosis was summarized using a binary variable indicating presence of moderate to severe arteriolosclerosis[6].

Cerebral amyloid angiopathy (CAA). Immunohistochemical staining using antibodies against amyloid-β was in place for visualization of amyloid-β in the walls of parenchymal and meningeal vessels in four regions: midfrontal, midtemporal, angular, and calcarine cortices. Severity of amyloid-β in the vessel walls was scored using a semiquantitative scale, which was summarized in the current study by a binary variable indicating moderate to severe CAA[7].

References.

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Table e-1. Associations of the *APOE ℰ4*-related proteins with amyloid-β (Aβ), tau tangle, and rate of global cognition decline.

Uniprot ID	Gene	Protein name	*APOE ℰ4*	Amyloid-β	Tau tangle	Rate of global cognition decline
			Estimate (SE), P-value			
P21741	*MDK*	Midkine	**0.756 (0.094), <0.001**	**0.545 (0.022), <0.001**	**0.458 (0.040), <0.001**	**-0.024 (0.004), <0.001**
Q9H4F8	*SMOC1*	Sparc-related modular calcium-binding protein 1	**0.736 (0.094), <0.001**	**0.540 (0.022), <0.001**	**0.490 (0.039), <0.001**	**-0.026 (0.004), <0.001**
P05067	*APP*	Amyloid precursor protein	**0.709 (0.095), <0.001**	**0.512 (0.023), <0.001**	**0.463 (0.040), <0.001**	**-0.027 (0.004), <0.001**
O95631	*NTN1*	Netrin-1	**0.697 (0.095), <0.001**	**0.546 (0.022), <0.001**	**0.501 (0.039), <0.001**	**-0.027 (0.004), <0.001**
Q92743	*HTRA1*	Serine protease HTRA1	**0.686 (0.095), <0.001**	**0.282 (0.029), <0.001**	**0.357 (0.041), <0.001**	**-0.024 (0.004), <0.001**
Q8N474	*SFRP1*	Secreted frizzled-related protein 1	**0.776 (0.108), <0.001**	**0.525 (0.028), <0.001**	**0.486 (0.049), <0.001**	**-0.026 (0.005), <0.001**
O94813	*SLIT2*	Slit homolog 2 protein	**0.654 (0.096), <0.001**	**0.461 (0.025), <0.001**	**0.445 (0.041), <0.001**	**-0.026 (0.004), <0.001**
P17948	*FLT1*	Vascular endothelial growth factor receptor 1	**0.593 (0.095), <0.001**	**0.419 (0.026), <0.001**	**0.433 (0.040), <0.001**	**-0.023 (0.004), <0.001**
Q92563	*SPOCK2*	Testican-2	**0.574 (0.097), <0.001**	**0.316 (0.029), <0.001**	**0.339 (0.042), <0.001**	**-0.022 (0.004), <0.001**
P55107	*GDF10*	Growth/differentiation factor 10	**0.700 (0.118), <0.001**	**0.445 (0.031), <0.001**	**0.484 (0.054), <0.001**	**-0.03 (0.006), <0.001**
Q96CG8	*CTHRC1*	Collagen Triple helix repeat-containing protein 1	**0.615 (0.107), <0.001**	**0.506 (0.026), <0.001**	**0.395 (0.042), <0.001**	**-0.024 (0.004), <0.001**
P78333	*GPC5*	Glypican-5	**0.499 (0.097), <0.001**	**0.360 (0.028), <0.001**	**0.308 (0.042), <0.001**	**-0.012 (0.004), 0.002**
Q9ULB1	*NRXN1*	Neurexin1	**0.498 (0.096), <0.001**	**0.340 (0.028), <0.001**	**0.275 (0.042), <0.001**	**-0.022 (0.004), <0.001**
Q92765	*FRZB*	Secreted frizzled-related protein 3	**0.518 (0.101), <0.001**	**0.265 (0.031), <0.001**	**0.255 (0.045), <0.001**	**-0.017 (0.004), <0.001**
O00622	*CCN1*	CCN family member 1	**0.959 (0.185), <0.001**	**0.314 (0.060), <0.001**	**0.436 (0.092), <0.001**	-0.024 (0.013), 0.057
Q9BQ16	*SPOCK3*	Testican-3	**0.484 (0.098), <0.001**	**0.362 (0.028), <0.001**	**0.448 (0.041), <0.001**	**-0.028 (0.004), <0.001**
P02743	*APCS*	Serum amyloid P-component	**0.473 (0.097), <0.001**	**0.211 (0.030), <0.001**	**0.282 (0.042), <0.001**	**-0.024 (0.004), <0.001**
Q9BXS0	*COL25A1*	Collagen alpha-1(XXV) chain	**0.694 (0.147), <0.001**	**0.330 (0.044), <0.001**	**0.271 (0.056), <0.001**	-0.017 (0.006), 0.003

Table e-2. Associations of the *APOE ℰ4*-related proteins with rates of decline in 5 cognitive domains.

Protein	Episodic memory	Semantic memory	Working memory	Processing speed	Visuospatial ability
	Estimate (SE), P-value				
P21741	**-0.029 (0.005), <0.001**	**-0.028 (0.004), <0.001**	**-0.017 (0.004), <0.001**	**-0.018 (0.004), <0.001**	-0.008 (0.004), 0.035
Q9H4F8	**-0.033 (0.005), <0.001**	**-0.028 (0.004), <0.001**	**-0.018 (0.004), <0.001**	**-0.016 (0.004), <0.001**	-0.006 (0.004), 0.105
P05067	**-0.032 (0.005), <0.001**	**-0.03 (0.004), <0.001**	**-0.02 (0.004), <0.001**	**-0.02 (0.004), <0.001**	-0.009 (0.004), 0.016
O95631	**-0.034 (0.005), <0.001**	**-0.031 (0.004), <0.001**	**-0.018 (0.004), <0.001**	**-0.016 (0.004), <0.001**	-0.008 (0.004), 0.037
Q92743	**-0.031 (0.005), <0.001**	**-0.022 (0.004), <0.001**	**-0.018 (0.004), <0.001**	**-0.018 (0.005), <0.001**	**-0.012 (0.004), 0.002**
Q8N474	**-0.032 (0.006), <0.001**	**-0.03 (0.005), <0.001**	**-0.015 (0.005), 0.002**	**-0.018 (0.005), <0.001**	-0.01 (0.005), 0.035
O94813	**-0.032 (0.005), <0.001**	**-0.028 (0.004), <0.001**	**-0.018 (0.004), <0.001**	**-0.016 (0.005), <0.001**	-0.009 (0.004), 0.03
P17948	**-0.029 (0.005), <0.001**	**-0.027 (0.004), <0.001**	**-0.016 (0.004), <0.001**	-0.013 (0.004), 0.003	-0.006 (0.004), 0.105
Q92563	**-0.026 (0.005), <0.001**	**-0.021 (0.004), <0.001**	**-0.018 (0.004), <0.001**	-0.013 (0.005), 0.005	-0.006 (0.004), 0.123
P55107	**-0.036 (0.007), <0.001**	**-0.036 (0.006), <0.001**	**-0.017 (0.005), 0.002**	**-0.022 (0.006), <0.001**	-0.006 (0.005), 0.255
Q96CG8	**-0.03 (0.005), <0.001**	**-0.025 (0.004), <0.001**	**-0.017 (0.004), <0.001**	-0.013 (0.005), 0.01	-0.007 (0.004), 0.086
P78333	**-0.016 (0.005), <0.001**	**-0.019 (0.004), <0.001**	-0.006 (0.004), 0.129	-0.01 (0.004), 0.034	0.003 (0.004), 0.4
Q9ULB1	**-0.024 (0.005), <0.001**	**-0.022 (0.004), <0.001**	**-0.018 (0.004), <0.001**	-0.011 (0.005), 0.013	-0.01 (0.004), 0.011
Q92765	**-0.02 (0.005), <0.001**	**-0.019 (0.005), <0.001**	-0.013 (0.004), 0.004	-0.01 (0.005), 0.041	-0.009 (0.004), 0.046
O00622	**-0.048 (0.012), <0.001**	-0.028 (0.014), 0.05	-0.014 (0.011), 0.207	-0.013 (0.012), 0.263	0.006 (0.015), 0.7
Q9BQ16	**-0.034 (0.005), <0.001**	**-0.026 (0.004), <0.001**	**-0.018 (0.004), <0.001**	**-0.018 (0.004), <0.001**	-0.009 (0.004), 0.016
P02743	**-0.027 (0.005), <0.001**	**-0.023 (0.004), <0.001**	**-0.02 (0.004), <0.001**	-0.013 (0.005), 0.004	-0.009 (0.004), 0.039
Q9BXS0	-0.021 (0.007), 0.003	-0.009 (0.005), 0.107	-0.012 (0.006), 0.042	-0.012 (0.006), 0.052	-0.001 (0.005), 0.869

Bold numbers indicate significant associations after Bonferroni adjustment.

Table e-3. Associations of the *APOE ε4*-related proteins with their bulk tissue RNA expression levels.

Protein	Gene	Spearman correlation coefficient	P-value
P21741	*MDK*	-0.06	0.144
Q9H4F8	*SMOC1*	0.05	0.240
P05067	*APP*	0.07	0.118
O95631	*NTN1*	-0.03	0.516
Q92743	*HTRA1*	**0.15**	**<0.001**
Q8N474	*SFRP1*	0.07	0.173
O94813	*SLIT2*	0.05	0.285
P17948	*FLT1*	**0.17**	**<0.001**
Q92563	*SPOCK2*	0.08	0.052
P55107	*GDF10*	-0.08	0.119
Q96CG8	*CTHRC1*	0.03	0.550
P78333	*GPC5*	0.12	0.006
Q9ULB1	*NRXN1*	0.01	0.821
Q92765	*FRZB*	0.09	0.033
O00622	*CCN1*	0.23	0.013
Q9BQ16	*SPOCK3*	**0.19**	**<0.001**
P02743[a]	*APCS*	**NA**	**NA**
Q9BXS0	*COL25A1*	-0.10	0.126

[a]The RNA expression level of *APCS* did not pass quality control.

Table e-4. Associations of *APOE Ɛ4* with RNA expression levels of the genes of proteins related to *APOE Ɛ4* in participants with valid proteomic data (n=596).

Protein	Gene	Estimate (SE), P-value
P21741	*MDK*	-0.167 (0.073), 0.022
Q9H4F8	*SMOC1*	-0.031 (0.06), 0.604
P05067	*APP*	0.022 (0.034), 0.504
O95631	*NTN1*	-0.038 (0.046), 0.41
Q92743	*HTRA1*	-0.027 (0.043), 0.53
Q8N474	*SFRP1*	-0.075 (0.06), 0.212
O94813	*SLIT2*	0.03 (0.038), 0.426
P17948	*FLT1*	-0.039 (0.069), 0.576
Q92563	*SPOCK2*	-0.003 (0.02), 0.894
P55107	*GDF10*	-0.105 (0.063), 0.097
Q96CG8	*CTHRC1*	0.041 (0.073), 0.572
P78333	*GPC5*	-0.048 (0.058), 0.412
Q9ULB1	*NRXN1*	0.016 (0.029), 0.578
Q92765	*FRZB*	0.007 (0.07), 0.925
O00622	*CCN1*	0.042 (0.081), 0.599
Q9BQ16	*SPOCK3*	-0.001 (0.042), 0.989
P02743[a]	*APCS*	**NA**
Q9BXS0	*COL25A1*	0.038 (0.068), 0.578

The estimates (SE), P-values are derived from separate linear regressions with one of the genes' RNA expression as the outcome, *APOE Ɛ4* as the covariate, controlled for age at death and sex. None of the associations were significant after Bonferroni-correction.

Table e-5. Associations of *APOE Ɛ4* with RNA expression levels of the genes of proteins related to *APOE Ɛ4* in all participants with bulk tissue RNASeq expression data (n=1196).

Protein	Gene	Estimate (SE), P-value
P21741	*MDK*	-0.083 (0.046), 0.072
Q9H4F8	*SMOC1*	0.033 (0.04), 0.409
P05067	*APP*	0.03 (0.023), 0.186
O95631	*NTN1*	-0.026 (0.03), 0.382
Q92743	*HTRA1*	-0.002 (0.028), 0.936
Q8N474	*SFRP1*	0.025 (0.038), 0.517
O94813	*SLIT2*	-0.012 (0.026), 0.639
P17948	*FLT1*	0.036 (0.046), 0.434
Q92563	*SPOCK2*	-0.017 (0.012), 0.156
P55107	*GDF10*	-0.054 (0.043), 0.202
Q96CG8	*CTHRC1*	-0.021 (0.044), 0.634
P78333	*GPC5*	-0.067 (0.037), 0.071
Q9ULB1	*NRXN1*	-0.023 (0.018), 0.205
Q92765	*FRZB*	-0.044 (0.05), 0.375
O00622	*CCN1*	0.021 (0.053), 0.69
Q9BQ16	*SPOCK3*	0.08 (0.027), 0.003
P02743[a]	*APCS*	**NA**
Q9BXS0	*COL25A1*	-0.045 (0.045), 0.311

The estimates (SE), P-values are derived from separate linear regressions with one of the genes' RNA expression as the outcome, *APOE Ɛ4* as the covariate, controlled for age at death and sex. None of the associations were significant after Bonferroni-correction.

Table e-6. Associations of *APOE Ɛ4* with single nuclei RNA expression levels of the genes of proteins related to *APOE Ɛ4* in participants with proteomic data (n=218).

Gene	Astrocytes	Endothelia	Excitatory neurons	Inhibitory neurons	Microglia	Oligodendrocytes	Oligodendrocyte precursor cells
	Estimate (SE), P-value						
MDK	-0.079 (0.112), 0.482	**NA**	-0.092 (0.073), 0.209	-0.007 (0.091), 0.936	**NA**	-0.344 (0.144), 0.018	-0.3 (0.137), 0.029
SMOC1	0.07 (0.125), 0.579	**NA**	0.083 (0.108), 0.446	0.017 (0.071), 0.815	0.105 (0.175), 0.548	0.013 (0.124), 0.916	-0.038 (0.091), 0.678
APP	-0.009 (0.085), 0.919	-0.126 (0.088), 0.155	-0.012 (0.037), 0.734	-0.001 (0.027), 0.965	-0.014 (0.116), 0.902	-0.024 (0.025), 0.337	0.025 (0.038), 0.513
NTN1	0.066 (0.092), 0.469	**NA**	-0.069 (0.143), 0.628	0.03 (0.098), 0.765	**NA**	**NA**	0.041 (0.055), 0.458
HTRA1	-0.069 (0.076), 0.364	-0.01 (0.159), 0.949	0.055 (0.221), 0.804	-0.114 (0.045), 0.011	-0.025 (0.088), 0.772	-0.067 (0.052), 0.2	-0.014 (0.057), 0.805
SFRP1	**NA**	**NA**	**NA**	-0.057 (0.083), 0.493	**NA**	-0.004 (0.091), 0.969	0.194 (0.167), 0.246
SLIT2	0.086 (0.186), 0.644	-0.233 (0.205), 0.258	0.093 (0.062), 0.134	0.103 (0.071), 0.148	-0.065 (0.215), 0.764	-0.008 (0.065), 0.906	0.156 (0.106), 0.144
FLT1	**NA**	-0.073 (0.102), 0.478	**NA**	0.051 (0.07), 0.466	0.353 (0.235), 0.135	**NA**	**NA**
SPOCK2	-0.062 (0.053), 0.247	-0.027 (0.132), 0.839	-0.021 (0.055), 0.702	-0.031 (0.043), 0.474	-0.185 (0.169), 0.274	-0.084 (0.207), 0.685	-0.089 (0.062), 0.153
GDF10	-0.038 (0.147), 0.793	**NA**	0.064 (0.092), 0.489	0.006 (0.083), 0.939	**NA**	**NA**	**NA**
CTHRC1	0.09 (0.152), 0.552	**NA**	-0.041 (0.084), 0.626	0.032 (0.139), 0.818	**NA**	0.099 (0.154), 0.52	-0.15 (0.138), 0.278
GPC5	0.037 (0.058), 0.522	-0.141 (0.2), 0.48	0.169 (0.07), 0.017	0.164 (0.107), 0.126	0.161 (0.127), 0.207	0.003 (0.137), 0.98	0.134 (0.195), 0.492
NRXN1	0.057 (0.066), 0.39	0.172 (0.162), 0.288	0.066 (0.045), 0.146	0.079 (0.051), 0.124	0.133 (0.362), 0.713	0.077 (0.283), 0.786	0.05 (0.061), 0.407
FRZB	-0.107 (0.17), 0.528	-0.158 (0.193), 0.416	-0.022 (0.056), 0.696	-0.052 (0.078), 0.508	**NA**	-0.219 (0.172), 0.203	-0.019 (0.115), 0.871

CCN1	-0.114 (0.132), 0.392	0.211 (0.259), 0.416	-0.023 (0.136), 0.866	**NA**	**NA**	**NA**	**NA**
SPOCK3	0.04 (0.123), 0.745	0.014 (0.197), 0.943	0.18 (0.091), 0.049	0.063 (0.037), 0.091	0.1 (0.121), 0.409	0.004 (0.047), 0.927	0.001 (0.073), 0.992
APCS	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**
COL25A1	-0.131 (0.114), 0.252	**NA**	0.027 (0.062), 0.659	0 (0.069), 0.995	-0.088 (0.161), 0.585	0.199 (0.151), 0.188	-0.17 (0.094), 0.071

The estimates (SE), P-values are derived from separate linear regressions (93 models) with one of the genes' RNA expression in one cell as the outcome, *APOE Ɛ4* as the covariate, controlled for age at death and sex. None of the associations were significant after Bonferroni-correction.

Table e-7. Associations of *APOE Ɛ4* with single nuclei RNA expression levels of the genes of proteins related to *APOE Ɛ4* in all participants with single nuclei RNA expression data (n=424).

Gene	Astrocytes	Endothelia	Excitatory Neurons	Inhibitory Neurons	Microglia	Oligodendroglia	Oligodendrocyte precursor cells
	Estimate (SE), P-value						
MDK	-0.034 (0.076), 0.66	**NA**	-0.058 (0.049), 0.241	-0.028 (0.058), 0.626	**NA**	-0.167 (0.108), 0.12	-0.201 (0.095), 0.035
SMOC1	0.135 (0.082), 0.103	**NA**	0.035 (0.075), 0.639	0.017 (0.045), 0.713	-0.027 (0.117), 0.821	0.105 (0.08), 0.19	-0.03 (0.06), 0.619
APP	-0.008 (0.063), 0.902	-0.063 (0.054), 0.241	-0.005 (0.024), 0.825	-0.003 (0.019), 0.877	0.021 (0.081), 0.798	0.006 (0.017), 0.715	0.032 (0.026), 0.212
NTN1	0.077 (0.063), 0.221	**NA**	0.011 (0.096), 0.908	-0.022 (0.065), 0.735	**NA**	**NA**	0.015 (0.036), 0.672
HTRA1	-0.022 (0.052), 0.671	-0.006 (0.11), 0.957	-0.018 (0.151), 0.905	-0.071 (0.03), 0.018	-0.02 (0.057), 0.72	-0.01 (0.036), 0.783	0.042 (0.039), 0.28
SFRP1	**NA**	**NA**	**NA**	-0.01 (0.056), 0.858	**NA**	-0.083 (0.061), 0.172	0.133 (0.107), 0.215
SLIT2	0.031 (0.125), 0.805	-0.069 (0.14), 0.625	0.106 (0.041), 0.01	0.078 (0.047), 0.097	-0.16 (0.147), 0.278	-0.02 (0.043), 0.648	0.077 (0.071), 0.277
FLT1	**NA**	0.002 (0.069), 0.981	**NA**	-0.061 (0.049), 0.216	**0.61 (0.163), 0.002**	**NA**	**NA**
SPOCK2	-0.009 (0.035), 0.788	-0.078 (0.079), 0.321	-0.027 (0.039), 0.489	-0.012 (0.029), 0.69	-0.14 (0.119), 0.241	-0.112 (0.147), 0.448	-0.023 (0.048), 0.638
GDF10	-0.149 (0.096), 0.121	**NA**	0.044 (0.062), 0.477	-0.04 (0.059), 0.496	**NA**	**NA**	**NA**
CTHRC1	0.047 (0.103), 0.648	**NA**	-0.046 (0.056), 0.415	0.005 (0.088), 0.955	**NA**	-0.018 (0.102), 0.863	-0.06 (0.089), 0.502
GPC5	0.009 (0.039), 0.821	-0.013 (0.136), 0.924	0.12 (0.046), 0.01	0.065 (0.069), 0.347	-0.016 (0.086), 0.854	-0.049 (0.088), 0.581	0.136 (0.128), 0.287
NRXN1	-0.032 (0.047), 0.498	0.029 (0.11), 0.795	0.046 (0.03), 0.132	0.037 (0.034), 0.27	0.032 (0.251), 0.897	-0.039 (0.195), 0.84	0.008 (0.04), 0.837
FRZB	-0.061 (0.107), 0.572	-0.168 (0.134), 0.212	0.026 (0.04), 0.521	0.015 (0.054), 0.787	**NA**	-0.183 (0.115), 0.112	0.056 (0.079), 0.481

CCN1	-0.006 (0.091), 0.945	-0.013 (0.164), 0.937	0.1 (0.096), 0.298	**NA**	**NA**	**NA**	**NA**
SPOCK3	0.003 (0.084), 0.975	-0.027 (0.126), 0.828	0.126 (0.06), 0.037	0.024 (0.025), 0.333	-0.018 (0.078), 0.816	0.048 (0.032), 0.136	-0.006 (0.046), 0.902
APCS	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**
COL25A1	-0.089 (0.075), 0.232	**NA**	0.029 (0.04), 0.462	-0.027 (0.046), 0.564	-0.132 (0.101), 0.191	0.089 (0.102), 0.381	-0.04 (0.061), 0.513

The estimates (SE), P-values are derived from separate linear regressions (93 models) with one of the genes' RNA expression in one cell as the outcome, *APOE Ɛ4* as the covariate, controlled for age at death and sex. The analyses indicated that only the association between *APOE Ɛ4* and single nuclei RNA expression of *FLT1* (Vascular endothelial growth factor receptor 1) in microglia was significant after Bonferroni-correction.

Table e-8. Associations of the 18 *APOE Ɛ4*-related proteins with AD polygenic risk scores that included risk variants except *APOE Ɛ4*.

Protein	Gene	AD risk score (P<5×10^-8)	AD risk score (P<1)
		Estimate (SE), P-value	
P21741	*MDK*	0.106 (0.042), 0.011	0.09 (0.042), 0.031
Q9H4F8	*SMOC1*	0.102 (0.042), 0.015	0.112 (0.042), 0.008
P05067	*APP*	0.098 (0.042), 0.02	0.111 (0.042), 0.009
O95631	*NTN1*	0.117 (0.042), 0.005	0.091 (0.042), 0.031
Q92743	*HTRA1*	0.025 (0.042), 0.557	0.085 (0.042), 0.043
Q8N474	*SFRP1*	0.104 (0.047), 0.026	**0.191 (0.047), <0.001**
O94813	*SLIT2*	0.106 (0.042), 0.012	0.092 (0.042), 0.029
P17948	*FLT1*	0.084 (0.042), 0.045	0.111 (0.042), 0.008
Q92563	*SPOCK2*	**0.168 (0.042), <0.001**	0.05 (0.042), 0.24
P55107	*GDF10*	0.125 (0.056), 0.025	0.151 (0.055), 0.006
Q96CG8	*CTHRC1*	0.049 (0.047), 0.294	0.087 (0.047), 0.064
P78333	*GPC5*	0.031 (0.042), 0.463	0.089 (0.042), 0.033
Q9ULB1	*NRXN1*	**0.131 (0.042), 0.002**	0.022 (0.042), 0.6
Q92765	*FRZB*	0.097 (0.045), 0.031	0.058 (0.045), 0.199
O00622	*CCN1*	0.111 (0.081), 0.173	0.203 (0.087), 0.021
Q9BQ16	*SPOCK3*	0.073 (0.043), 0.089	**0.167 (0.042), <0.001**
P02743[a]	*APCS*	0.054 (0.042), 0.194	0.082 (0.042), 0.05
Q9BXS0	*COL25A1*	0.183 (0.066), 0.006	0.071 (0.068), 0.298

In two series of 18 separate linear regressions the associations of two AD polygenic risk score, derived by applying two p-value thresholds for sequence variants inclusion: <5×10^-8 and <1, were examined with the levels of 18 *APOE Ɛ4*-related proteins. All the models were controlled for age at death and sex.

Table e-9. Protein levels and number of missing of 18 *APOE Ɛ4*-related proteins.

Protein	Gene	Number of missing	Mean	SD
P21741	*MDK*	0	0.0053	0.9988
Q9H4F8	*SMOC1*	0	0.0037	1
P05067	*APP*	8	0.0043	0.9997
O95631	*NTN1*	8	0.0057	0.9983
Q92743	*HTRA1*	0	0.0035	1.0004
Q8N474	*SFRP1*	148	0.0062	0.9957
O94813	*SLIT2*	8	0.0064	0.9973
P17948	*FLT1*	0	0.0056	0.9984
Q92563	*SPOCK2*	8	0.0024	1.0006
P55107	*GDF10*	230	0.0025	0.9988
Q96CG8	*CTHRC1*	112	0.0038	0.9993
P78333	*GPC5*	0	0.0047	0.9992
Q9ULB1	*NRXN1*	0	0.0007	1.0014
Q92765	*FRZB*	61	0.0019	0.9991
O00622	*CCN1*	450	0.0023	1.0058
Q9BQ16	*SPOCK3*	21	0.001	0.9997
P02743	*APCS*	0	0.0006	1.001
Q9BXS0	*COL25A1*	343	0	1

Table e-10. Characteristic of 302 participants who had non-missing levels of 15 *APOE Ɛ4*-related proteins.

Covariates	**Included (n=302)**	**Excluded (n=294)**	**Statistics, p-value**
	Mean (SD) or n (%)		
Age at death baseline, years, Mean (SD)	90.2 (6.4)	89.0 (6.3)	t=-2.14, **p=0.033**
Women, n (%)	218 (72)	192 (65)	χ^2=3.28, 0.070
Education, years, Mean (SD)	15.2 (3.4)	15.6 (3.4)	t=1.45, 0.147
White non-Hispanic, n (%)	295 (98)	281 (96)	χ^2=2.03, 0.154
APOE Ɛ4	70 (23)	63 (22)	χ^2=0.127, 0.722
APOE genotype			χ^2=6.0, 0.308
Ɛ2Ɛ2, n (%)	1 (0.3)	4 (1)	
Ɛ2Ɛ3, n (%)	44 (15)	32 (11)	
Ɛ3Ɛ3, n (%)	187 (62)	188 (66)	
Ɛ2Ɛ4, n (%)	6 (2)	6 (2)	
Ɛ3Ɛ4, n (%)	63 (11)	53 (18)	
Ɛ4Ɛ4, n (%)	1 (0.3)	4 (1)	
Mini Mental State Examination score, Mean (SD)	21.9 (8.4)	22.9 (8.1)	χ^2(KW)=3.89, **0.049**
Global cognition	-0.85 (1.10)	-0.73 (1.05)	t=1.43, 0.155
Alzheimer's dementia, n (%)	109 (36)	91 (31)	χ^2=1.77, 0.184
Pathological diagnosis of Alzheimer's disease, n (%)	115 (38)	110 (37)	χ^2=0.028, 0.867
Immunohistochemical assessments			
Square root of Amyloid-β load, Mean (SD)	1.09 (0.79)	1.04 (0.77)	t=-0.69, 0.493
Square root of Tau tangle density, Mean (SD)	1.51 (1.11)	1.36 (1.09)	t=-1.75, 0.080
Proteins			
P21741	-0.006 (0.945)	0.017 (1.052)	t=0.28, 0.782
Q9H4F8	-0.004 (1.012)	0.011 (0.989)	t=0.18, 0.854
P05067	-0.004 (0.990)	0.013 (1.011)	t=0.2, 0.841
O95631	-0.003 (0.935)	0.015 (1.062)	t=0.22, 0.827
Q92743	-0.003 (1.055)	0.01 (0.943)	t=0.17, 0.866
Q8N474	-0.002 (0.972)	0.023 (1.046)	t=0.25, 0.800
O94813	-0.002 (0.998)	0.015 (0.999)	t=0.2, 0.840
P17948	-0.012 (0.991)	0.024 (1.007)	t=0.44, 0.657

Q92563	-0.002 (0.973)	0.007 (1.03)	t=0.1, 0.920
P55107	-0.010 (0.990)	0.013 (1.008)	t=0.21, 0.832
Q96CG8	-0.003 (0.932)	0.015 (1.105)	t=0.18, 0.857
P78333	0.008 (0.962)	0.001 (1.038)	t=-0.08, 0.933
Q9ULB1	-0.013 (1.041)	0.015 (0.96)	t=0.34, 0.737
Q92765	0.006 (1.038)	-0.003 (0.949)	t=-0.1, 0.924
O00622	-0.045 (1.114)	0.045 (0.904)	t=0.54, 0.592
Q9BQ16	-0.008 (0.965)	0.011 (1.039)	t=0.22, 0.823
P02743	-0.008 (0.974)	0.009 (1.029)	t=0.21, 0.831
Q9BXS0	0 (1.026)	0 (0.961)	t=0, 1

The covariates were compared across the groups by t-test, chi-square, or Kruskal-Wallis tests.

Table e-11. Effect sizes of the 5 and 2 *APOE Ɛ4*-related proteins associated with amyloid-β and tau tangles, respectively.

Protein	Gene	Cohen's f^2	
		Amyloid-β	Tau tangles
Q9H4F8	*SMOC1*	0.013	–
O95631	*NTN1*	0.030	0.098
Q92743	*HTRA1*	0.013	–
Q8N474	*SFRP1*	0.031	–
Q9ULB1	*NRXN1*	0.017	–
Q9BQ16	*SPOCK3*	–	0.033

Table e-12. Associations of *APOE ℰ4* with amyloid-β (Aβ) and tau tangle with and without controlling for *APOE ℰ4*-related proteins.

Model	**Model terms**	**Association of *APOE ℰ4* with**	
		Amyloid-β	**Tau tangle**
		Estimate (SE), P-value	
1	*APOE ℰ4*	**0.550 (0.102), <0.001**	**0.735 (0.141), <0.001**
2	Netrin-1	**0.378 (0.063), <0.001**	**0.409 (0.074), <0.001**
	Secreted frizzled-related protein 1	**0.256 (0.060), <0.001**	NA
	Testican-3	NA	**0.239 (0.072), 0.001**
3	*APOE ℰ4*	0.097 (0.076), 0.198	**0.371 (0.132), 0.005**
	Netrin-1	**0.367 (0.063), <0.001**	**0.347 (0.077), <0.001**
	Secreted frizzled-related protein 1	**0.253 (0.060), <0.001**	NA
	Testican-3	NA	**0.246 (0.071), <0.001**
4	Aβ	NA	**0.636 (0.071), <0.001**
5	*APOE ℰ4*	NA	**0.340 (0.131), 0.010**
	Netrin-1		**0.191 (0.095), 0.045**
	Secreted frizzled-related protein 1		NA
	Testican-3		**0.242 (0.071), <0.001**
	Aβ		**0.270 (0.098), 0.006**

Table e-13. Associations of *APOE ε4*-related proteins with pathological diagnosis of AD and with non-AD brain pathologies.

Gene	AD	TDP-43	Hip. sclerosis	Lewy bodies	Macroinf.	Microinf.	Atheroscl.	Arterioscl.	CAA
	Odds Ratio (95% confidence interval), p-value								
MDK	**7.1 (5.2-9.6), <0.001**	**1.5 (1.3-1.9), <0.001**	1.1 (0.82-1.5), 0.481	1.3 (1.1-1.6), 0.004	1.0 (0.85-1.2), 0.929	1.1 (0.88-1.3), 0.585	0.88 (0.74-1.1), 0.166	0.96 (0.80-1.1), 0.63	**1.8 (1.5-2.3), <0.001**
SMOC1	**6.6 (4.9-8.9), <0.001**	**1.6 (1.3-1.9), <0.001**	1.2 (0.87-1.6), 0.28	**1.4 (1.2-1.7), <0.001**	1.0 (0.85-1.2), 0.894	1.1 (0.94-1.3), 0.204	0.93 (0.78-1.1), 0.392	0.99 (0.83-1.2), 0.886	**1.9 (1.6-2.3), <0.001**
APP	**6.1 (4.5-8.2), <0.001**	**1.4 (1.2-1.7), <0.001**	1.1 (0.85-1.6), 0.372	**1.4 (1.1-1.6), 0.002**	1.0 (0.86-1.2), 0.744	1.2 (0.98-1.4), 0.088	0.97 (0.82-1.2), 0.758	1.0 (0.85-1.2), 0.852	**1.8 (1.5-2.2), <0.001**
NTN1	**6.8 (4.9-9.3), <0.001**	**1.5 (1.3-1.9), <0.001**	1.2 (0.89-1.6), 0.215	**1.4 (1.2-1.7), <0.001**	1.0 (0.84-1.2), 0.976	1.1 (0.91-1.3), 0.365	0.91 (0.76-1.1), 0.268	0.99 (0.83-1.2), 0.911	**1.8 (1.4-2.1), <0.001**
HTRA1	**2.5 (2.0-3.1), <0.001**	**1.4 (1.2-1.7), <0.001**	1.2 (0.91-1.6), 0.174	1.3 (1.1-1.6), 0.005	1.1 (0.93-1.3), 0.258	1.2 (0.99-1.4), 0.065	0.99 (0.83-1.2), 0.867	1.1 (0.91-1.3), 0.384	**1.7 (1.4-2.1), <0.001**
SFRP1	**5.0 (3.7-6.8), <0.001**	**1.4 (1.1-1.7), 0.006**	1.3 (0.93-1.8), 0.126	**1.4 (1.1-1.8), 0.002**	1.1 (0.86-1.3), 0.6	1.2 (0.95-1.4), 0.147	1.0 (0.85-1.3), 0.75	1.0 (0.83-1.2), 0.904	**1.8 (1.4-2.3), <0.001**
SLIT2	**4.0 (3.0-5.2), <0.001**	**1.4 (1.2-1.7), <0.001**	1.1 (0.81-1.5), 0.554	**1.4 (1.2-1.7), <0.001**	0.95 (0.79-1.1), 0.537	1.2 (0.97-1.4), 0.098	0.84 (0.7-1), 0.058	0.97 (0.82-1.2), 0.777	**1.8 (1.5-2.2), <0.001**
FLT1	**3.1 (2.4-3.9), <0.001**	**1.4 (1.2-1.7), <0.001**	1.2 (0.9-1.6), 0.202	1.3 (1-1.5), 0.018	0.98 (0.83-1.2), 0.834	0.99 (0.82-1.2), 0.876	0.94 (0.79-1.1), 0.456	0.90 (0.76-1.1), 0.253	**1.6 (1.3-1.9), <0.001**
SPOCK2	**2.4 (1.9-3.0), <0.001**	**1.4 (1.2-1.7), <0.001**	1.3 (0.93-1.7), 0.129	**1.5 (1.3-1.9), <0.001**	1.1 (0.91-1.3), 0.363	0.98 (0.81-1.2), 0.805	0.85 (0.71-1), 0.063	0.97 (0.81-1.2), 0.700	**1.4 (1.2-1.7), <0.001**
GDF10	**5.7 (3.8-8.6), <0.001**	1.4 (1.1-1.8), 0.009	1.3 (0.87-1.9), 0.207	1.4 (1.1-1.8), 0.006	1.1 (0.88-1.4), 0.419	1.1 (0.84-1.3), 0.669	0.89 (0.72-1.1), 0.328	0.94 (0.75-1.2), 0.563	**1.4 (1.1-1.8), 0.002**
CTHRC1	**4.9 (3.7-6.6), <0.001**	**1.5 (1.2-1.8), <0.001**	1.2 (0.83-1.6), 0.396	**1.4 (1.1-1.7), 0.002**	1.0 (0.86-1.3), 0.686	0.99 (0.81-1.2), 0.897	0.95 (0.78-1.2), 0.604	0.98 (0.80-1.2), 0.805	**1.6 (1.3-2), <0.001**
GPC5	**2.4 (1.9-2.9), <0.001**	1.2 (1.0-1.5), 0.032	1.1 (0.81-1.5), 0.574	1 (0.87-1.3), 0.619	0.93 (0.78-1.1), 0.412	1.1 (0.89-1.3), 0.495	0.86 (0.72-1), 0.082	0.88 (0.73-1), 0.134	**1.4 (1.2-1.7), <0.001**
NRXN1	**2.2 (1.8-2.8), <0.001**	1.2 (1.0-1.5), 0.039	1.2 (0.89-1.6), 0.227	1.1 (0.93-1.4), 0.213	1.0 (0.84-1.2), 0.97	1.1 (0.9-1.3), 0.388	**0.76 (0.64-0.9), 0.002**	0.88 (0.74-1), 0.138	1.2 (1.0-1.4), 0.052
FRZB	**2.0 (1.6-2.5), <0.001**	1.2 (1.0-1.5), 0.035	1.1 (0.81-1.6), 0.489	1.3 (1-1.5), 0.016	1.1 (0.92-1.3), 0.294	1.1 (0.88-1.3), 0.514	1.0 (0.87-1.2), 0.670	1.0 (0.86-1.2), 0.700	**1.7 (1.4-2.1), <0.001**

CCN1	**2.9 (1.6-5.1), <0.001**	1.5 (1.1-2.2), 0.024	1.3 (0.78-2.2), 0.318	1.2 (0.83-1.7), 0.331	1.2 (0.86-1.7), 0.272	1.3 (0.92-1.9), 0.137	0.90 (0.63-1.3), 0.590	0.99 (0.71-1.4), 0.972	**2.6 (1.6-4.1), <0.001**
SPOCK3	**2.8 (2.3-3.6), <0.001**	**1.5 (1.2-1.8), <0.001**	1.1 (0.81-1.5), 0.501	**1.6 (1.3-1.9), <0.001**	1.0 (0.85-1.2), 0.872	1.1 (0.93-1.3), 0.238	0.98 (0.82-1.2), 0.832	1.1 (0.90-1.3), 0.390	**1.6 (1.3-1.9), <0.001**
APCS	**1.8 (1.4-2.2), <0.001**	1.2 (1.0-1.5), 0.036	1.1 (0.81-1.5), 0.535	1.2 (0.99-1.4), 0.057	1.2 (0.97-1.4), 0.111	1.1 (0.92-1.3), 0.268	1.1 (0.94-1.3), 0.217	1.1 (0.92-1.3), 0.308	**1.5 (1.3-1.9), <0.001**
COL25A1	**2.9 (2.0-4.3), <0.001**	1.1 (0.81-1.4), 0.623	1.5 (1.0-2.4), 0.052	1.4 (1.1-1.9), 0.014	1.0 (0.78-1.3), 0.877	0.87 (0.66-1.2), 0.349	0.93 (0.71-1.2), 0.571	0.90 (0.68-1.2), 0.442	1.2 (0.87-1.6), 0.295

In 9 series of logistic regression models, we examined the associations of 18 *APOE ℰ4*-related proteins with pathologic AD and non-AD brain pathologies in separate models. In each model, one of the *APOE ℰ4*-related proteins comprised the model term and the outcome was pathologic AD or one of the non-AD brain pathologies. All the models were controlled for age at death and sex.