

Supplementary Materials

CDH6 and HAGH protein levels in plasma associate with Alzheimer's disease in *APOE ε4* carriers

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Supplementary Table 1 Results of plasma-based proteome association with Alzheimer's disease in Rotterdam Study Model 2*

Annotation	Uniport ID	Effect size (β)	OR	SE	P-value	FDR-value
Overall population						
CDH6	P55285	0.325	1.384	0.109	3.16E-03	0.288
PRTG	Q2VWP7	0.266	1.304	0.106	1.31E-02	0.471
GZMA	P12544	0.141	1.152	0.060	1.95E-02	0.471
CNTN5	O94779	0.148	1.160	0.064	2.07E-02	0.471
APOE4 stratum						
CDH6	P55285	0.624	1.867	0.174	5.52E-04	0.030
HAGH	Q16775	0.491	1.633	0.139	6.62E-04	0.030
Beta-NGF	P01138	0.491	1.633	0.184	9.27E-03	0.184
JAM-B	P57087	0.365	1.441	0.139	1.03E-02	0.184
MDGA1	Q8NFP4	0.186	1.204	0.073	1.26E-02	0.184
PLXNB3	Q9ULL4	0.416	1.515	0.166	1.39E-02	0.184
CDH3	P22223	0.347	1.415	0.138	1.42E-02	0.184
TN-R	Q92752	0.241	1.272	0.100	1.79E-02	0.203
PDGF-R-alpha	P16234	0.376	1.456	0.167	2.69E-02	0.272
LAIR-2	Q6ISS4	0.112	1.119	0.054	4.01E-02	0.365
LXN	Q9BS40	0.622	1.863	0.307	4.55E-02	0.377
APOE33 stratum						
PRTG	Q2VWP7	0.326	1.386	0.136	1.74E-02	0.884
CNTN5	O94779	0.199	1.220	0.086	2.26E-02	0.884
APOE2 stratum						
SMPD1	P17405	-0.491	0.612	0.206	2.52E-02	0.775
RGMA	Q96B86	0.609	1.839	0.285	4.30E-02	0.775
CLM-1	Q8TDQ1	0.303	1.354	0.143	4.44E-02	0.775
SCARF2	Q96GP6	0.623	1.864	0.300	4.89E-02	0.775

Abbreviations: β , regression coefficient; OR, odds ratio; SE, standard error; *APOE*, apolipoprotein E; FDR, False discovery rate

*Model 2 was adjusted for age, sex, BMI, smoking (current versus non-smokers), educational attainment (high, medium, low) and medication intake (lipid lowering, anti-hypertensive, and anti-inflammatory), N = 300 (AD = 156, Control = 144)

Supplementary Table 2: Results of plasma-based proteome association with Alzheimer's disease in Rotterdam Study adjusting for age, sex and followup time

Uniprot id	Annotation	Effect size (β)	SE	P-value	FDR-value
Overall population					
P55285	CDH6	0.331	0.100	1.05E-03	5.72E-02
Q14793	GDF-8	0.145	0.045	1.26E-03	5.72E-02
Q94779	CNTN5	0.143	0.058	1.45E-02	3.78E-01
Q14108	SCARB2	-0.180	0.075	1.66E-02	3.78E-01
Q9BS40	LXN	0.327	0.152	3.18E-02	3.86E-01
Q9BZZ2	SIGLEC1	-0.124	0.058	3.50E-02	3.86E-01
Q16775	HAGH	0.141	0.069	4.30E-02	3.86E-01
P21757	MSR1	-0.150	0.074	4.38E-02	3.86E-01
Q2MKA7	RSP01	-0.166	0.083	4.63E-02	3.86E-01
Q8NFP4	MDGA1	0.085	0.042	4.64E-02	3.86E-01
Q2VWP7	PRTG	0.193	0.097	4.66E-02	3.86E-01
APOE4 stratum					
P55285	CDH6	0.661	0.167	1.48E-04	1.35E-02
Q16775	HAGH	0.477	0.135	6.47E-04	2.95E-02
Q92752	TN-R	0.256	0.094	7.85E-03	2.38E-01
Q9BS40	LXN	0.746	0.288	1.11E-02	2.52E-01
Q8NFP4	MDGA1	0.165	0.069	1.87E-02	3.40E-01
P41217	CD200	0.349	0.152	2.35E-02	3.57E-01
Q16620	NTRK2	0.608	0.274	2.89E-02	3.69E-01
P57087	JAM-B	0.291	0.136	3.52E-02	3.69E-01
P14384	CPM	0.496	0.234	3.65E-02	3.69E-01
Q96B86	RGMA	0.287	0.138	4.08E-02	3.72E-01
P09919	G-CSF	0.198	0.098	4.72E-02	3.76E-01
APOE33 stratum					
Q14108	SCARB2	-0.241	0.086	5.63E-03	3.15E-01
Q14594	NCAN	0.270	0.099	6.92E-03	3.15E-01
Q94779	CNTN5	0.166	0.073	2.41E-02	4.14E-01
Q9HAN9	NMNAT1	0.113	0.053	3.40E-02	4.14E-01
Q14793	GDF-8	0.134	0.064	3.71E-02	4.14E-01
Q92752	TN-R	0.139	0.066	3.76E-02	4.14E-01
Q96GW7	BCAN	0.171	0.082	3.86E-02	4.14E-01
P37023	SKR3	-0.219	0.110	4.79E-02	4.14E-01
Q9NP84	TNFRSF12A	-0.190	0.096	4.97E-02	4.14E-01
APOE2 stratum					
Q92765	sFRP-3	-0.544	0.218	1.87E-02	9.88E-01
P17405	SMPD1	-0.453	0.204	3.41E-02	9.88E-01
Q16775	HAGH	0.504	0.229	3.58E-02	9.88E-01
Q2TAL6	VWC2	-0.352	0.171	4.83E-02	9.88E-01

Abbreviations: β , regression coefficient; SE, standard error; *APOE*, apolipoprotein E; FDR, False discovery rate

*adjusted for age, sex, follow-up time

Supplementary Table 3: Results of plasma-based proteome association with Alzheimer's disease in Rotterdam Study without imputations

Uniprot id	Annotation	Effect size (β)	SE	P-value	FDR-value
Overall population					
P55285	CDH6	0.334	0.106	1.78E-03	1.60E-01
Q2VWP7	PRTG	0.286	0.100	4.62E-03	2.08E-01
O94779	CNTN5	0.155	0.061	1.24E-02	3.73E-01
O14594	NCAN	0.183	0.088	3.93E-02	7.02E-01
O14793	GDF-8	0.095	0.047	4.42E-02	7.02E-01
P12544	GZMA	0.142	0.071	4.70E-02	7.02E-01
<i>APOE4</i> stratum					
P55285	CDH6	0.638	0.171	3.33E-04	3.00E-02
Q16775	HAGH	0.479	0.147	1.59E-03	7.16E-02
Q92752	TN-R	0.280	0.094	3.72E-03	1.11E-01
P01138	Beta-NGF	0.431	0.182	2.00E-02	3.36E-01
Q8NFP4	MDGA1	0.164	0.070	2.22E-02	3.36E-01
P57087	JAM-B	0.318	0.137	2.24E-02	3.36E-01
P41217	CD200	0.335	0.155	3.31E-02	4.13E-01
Q9BS40	LXN	0.619	0.292	3.67E-02	4.13E-01
P16234	PDGF-R-alpha	0.324	0.159	4.42E-02	4.26E-01
<i>APOE33</i> stratum					
Q2VWP7	PRTG	0.309	0.122	1.22E-02	7.70E-01
O94779	CNTN5	0.193	0.081	1.85E-02	7.70E-01
O14594	NCAN	0.248	0.110	2.57E-02	7.70E-01
<i>APOE2</i> stratum					
P17405	SMPD1	-0.447	0.203	3.51E-02	9.66E-01
Q2TAL6	VWC2	-0.362	0.166	3.69E-02	9.66E-01

Abbreviations: β , regression coefficient; SE, standard error; *APOE*, apolipoprotein E; FDR, False discovery rate

*adjusted for age and sex

Supplementary Table 4: Results of interaction analysis of proteins with *APOE* in Rotterdam Study (*interaction P-value* <0.05)

Uniprot ID	annotation	$\beta_{\text{interaction}}$	SE	<i>P-value</i> _{interaction}
Q16775	HAGH	0.414	0.172	1.70E-02
P09919	G-CSF	0.276	0.125	2.78E-02
O95727	CRTAM	-0.221	0.106	3.77E-02

Abbreviations: β , regression coefficient; SE, standard error

Model: Alzheimer's disease ~ Protein levels * *APOE* ϵ 4 (0/1) + Protein levels + *APOE* ϵ 4 (0/1) + age + sex

Supplementary Table 5: Association of CSF levels of CDH6 levels with AD versus controls in the Amsterdam Dementia Cohort

Stratum*	Effect size (β)	SE	<i>P-value</i>
Overall sample	0.329	0.220	0.136
<i>APOE</i>4 stratum	-0.107	0.476	0.822
<i>APOE</i> 33	0.457	0.363	0.207
<i>APOE</i>2 stratum	1.949	1.233	0.114

Abbreviations: AD, Alzheimer's disease; SE, standard error

*Logistic regression analysis adjusting for age and sex

Supplementary Table 6: Case-Control stratified association of CDH6 protein with CSF biomarkers of AD in the Amsterdam Dementia Cohort

Biomarker*	Effect size (β)	SE	<i>P</i>-value
Controls only			
A β -42	105.387	23.6	$<1 \times 10^{-3}$
<i>P</i> -tau	10.03	1.22	$<1 \times 10^{-3}$
<i>t</i> -tau	76.4	7.6	$<1 \times 10^{-3}$
AD only			
A β -42	12.54	13.602	0.36
<i>P</i> -tau	27.757	4.496	$<1 \times 10^{-3}$
<i>t</i> -tau	254.815	50.7	$<1 \times 10^{-3}$

Abbreviations: AD, Alzheimer's disease; SE, standard error

*Linear regression analysis adjusting for age and sex

Supplementary Table 7: Results of association of protein levels with *APOE* genotypes

	<i>APOE4 vs APOE3[†]</i>			<i>APOE4 vs APOE2[‡]</i>			<i>APOE2 vs APOE3^α</i>		
	β	SE	P-values	β	SE	P-values	β	SE	P-values
Overall									
CDH6	0.051	0.051	0.117	0.163	0.055	3.79X10 ⁻³	-0.085	0.048	0.081
HAGH	-0.025	0.048	0.600	0.073	0.071	0.307	-0.091	0.077	0.237
Cases only									
CDH6	0.079	0.045	0.081	0.143	0.081	0.085	-0.005	0.081	0.945
HAGH	0.059	0.060	0.333	0.005	0.112	0.960	0.094	0.114	0.409
Controls only									
CDH6	-0.060	0.0505	0.235	0.061	0.072	0.406	-0.131	0.057	0.026
HAGH	-0.192	0.0860	0.028	0.002	0.081	0.972	-0.214	0.104	0.042

Abbreviations: β, regression coefficient; SE, standard error; *APOE*, apolipoprotein E

[†]*APOE* 44/34/24 =1 versus *APOE* 33=0; [‡]*APOE* 44/34=1 versus *APOE* 22/23=0; ^α*APOE* 22/23=1 versus *APOE* 33=0

Supplementary Table 8: Genome-wide significant variants ($P < 2.5 \times 10^{-8}$) from GWAS of CDH6 levels in plasma

rsID	Chr.	Position*	Alt.	Locus	Ref.	MAF	β	SE	P-values	R-square	Annotation	CADD
rs111283466	5	31130238	A	5p13.3	G	0.070	1.068	0.178	1.92E-09	0.985	Intergenic	0.292
rs10050733	5	31127060	C	5p13.3	T	0.070	1.061	0.177	2.13E-09	0.990	Intergenic	1.345
rs59671083	5	31126633	A	5p13.3	G	0.070	1.059	0.177	2.21E-09	0.991	Intergenic	8.567
rs13170907	5	31123473	C	5p13.3	T	0.070	1.050	0.176	2.54E-09	0.997	Intergenic	12.90
rs10071438	5	31114078	T	5p13.3	A	0.070	1.049	0.176	2.59E-09	0.997	Intergenic	7.310
rs10044808	5	31134528	T	5p13.3	C	0.061	1.085	0.188	7.97E-09	0.977	Intergenic	0.401
rs10044844	5	31134508	A	5p13.3	G	0.061	1.085	0.188	7.97E-09	0.977	Intergenic	0.729
rs10079279	5	31123143	G	5p13.3	A	0.061	1.053	0.185	1.24E-08	1.000	Intergenic	0.156
rs16900696	5	31113414	G	5p13.3	C	0.061	1.053	0.185	1.25E-08	0.998	Intergenic	6.323
rs28658063	5	31117714	C	5p13.3	G	0.061	1.053	0.185	1.25E-08	0.998	Intergenic	4.499
rs28702563	5	31117764	A	5p13.3	G	0.061	1.053	0.185	1.25E-08	0.998	Intergenic	4.673
rs7732113	5	31114502	A	5p13.3	G	0.061	1.053	0.185	1.25E-08	0.998	Intergenic	1.877
rs13358949	5	31112867	G	5p13.3	A	0.061	1.052	0.185	1.36E-08	0.996	Intergenic	1.647

Abbreviations: Chr., chromosome; MAF, Minor allele frequency; CADD, combined annotation dependent depletion.

* Basepair information is provided as per GRCh37/hg19

Note: variants are sorted as per P-values, β is coded as per alternative allele

Supplementary Table 9: Population characteristics of the BioFINDER study

	AD cases	Controls
N	186	485
Age (SD), years	75.1 (7.2)	70.6 (5.6)
Female (%)	63.90%	46.10%
Body Mass index	24.2	25.6
MMSE (Mean)	21.4	27.6
Aβ42 (pg/mL)	317	533
t-tau (pg/mL)	620	401
p-tau (pg/mL)	117	69
<i>APOE</i> genotype		
<i>APOE</i> 44/34	124	223
<i>APOE</i> 33	58	224
<i>APOE</i> 22/23	4	38

Abbreviations: AD, Alzheimer's disease, SD, Standard deviation, *APOE*, apolipoprotein E gene

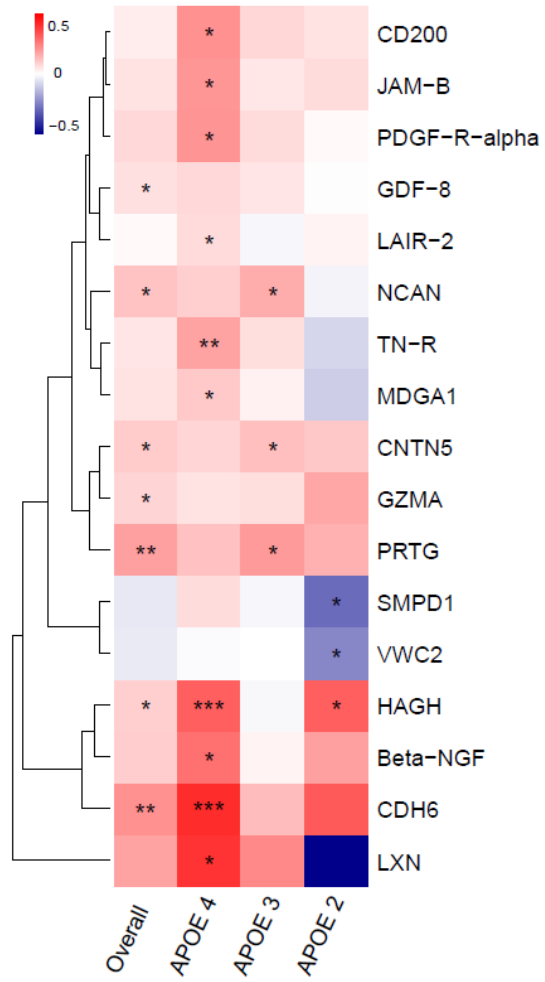
Supplementary Table 10: Population characteristic of Amsterdam dementia Cohort (ADC)

	AD cases	Controls
N (Male/Female)	236 (140/95)	199 (118/81)
Age (Mean±SD)	66 (8)	58 (8)***
MMSE (Mean±SD)	19.7 (5)	28.2 (1.6) ***
Aβ42 (pg/mL)	600.1 (117.3)	1120 (212.3)***
t-tau (pg/mL)	770.5 (415.8)	214 (85.5)***
p-tau (pg/mL)	91 (37.2)	38 (85.5)***
APOE genotyped (N)	219	150
APOE 44/34	128	34
APOE 33	79	93
APOE 22/23	12	25

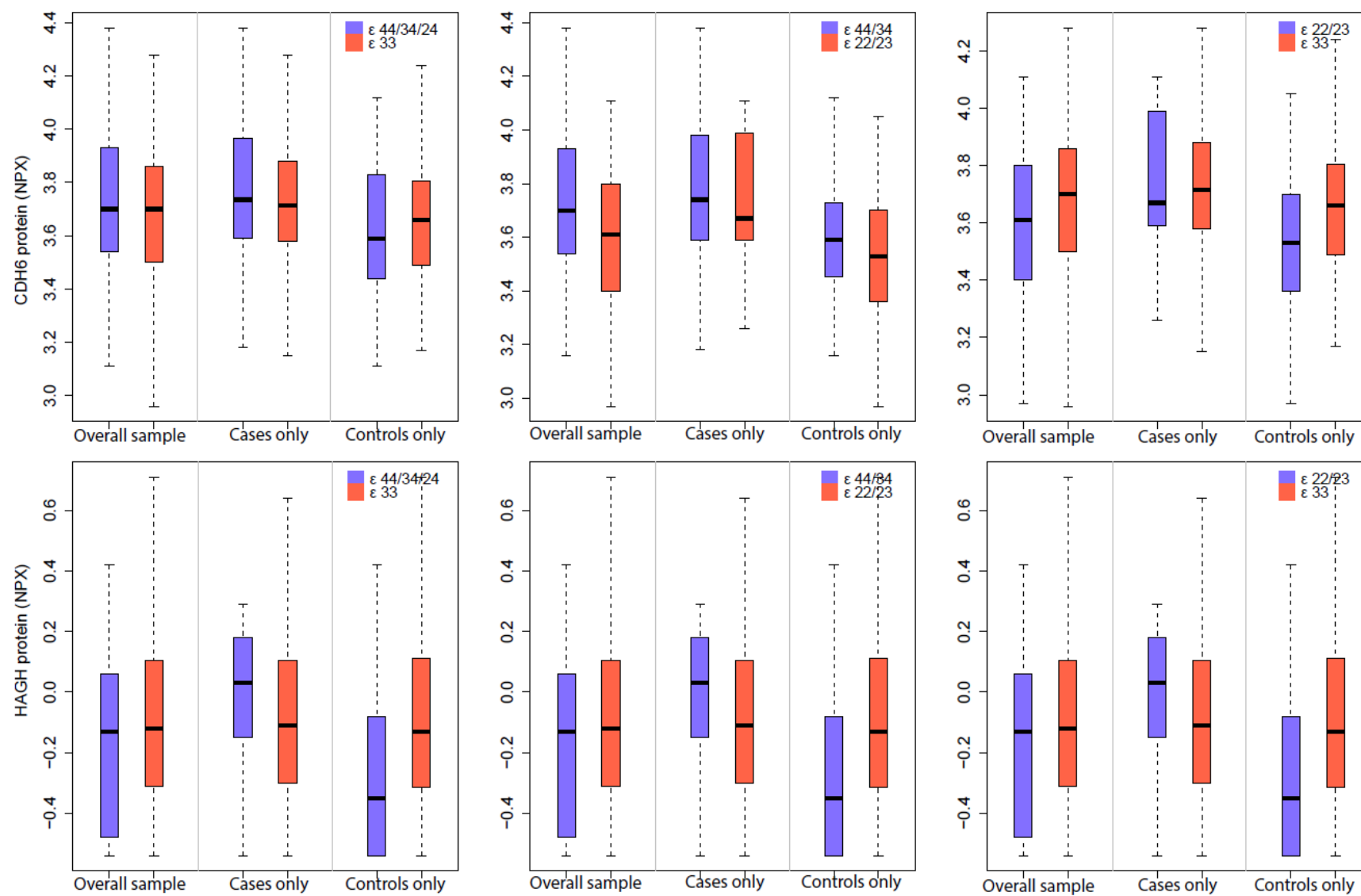
Abbreviations: AD, Alzheimer's disease

Note: Data are reported as medians and interquartile range unless otherwise indicated. *** *P-value* < 0.0001

Supplementary Figure 1:

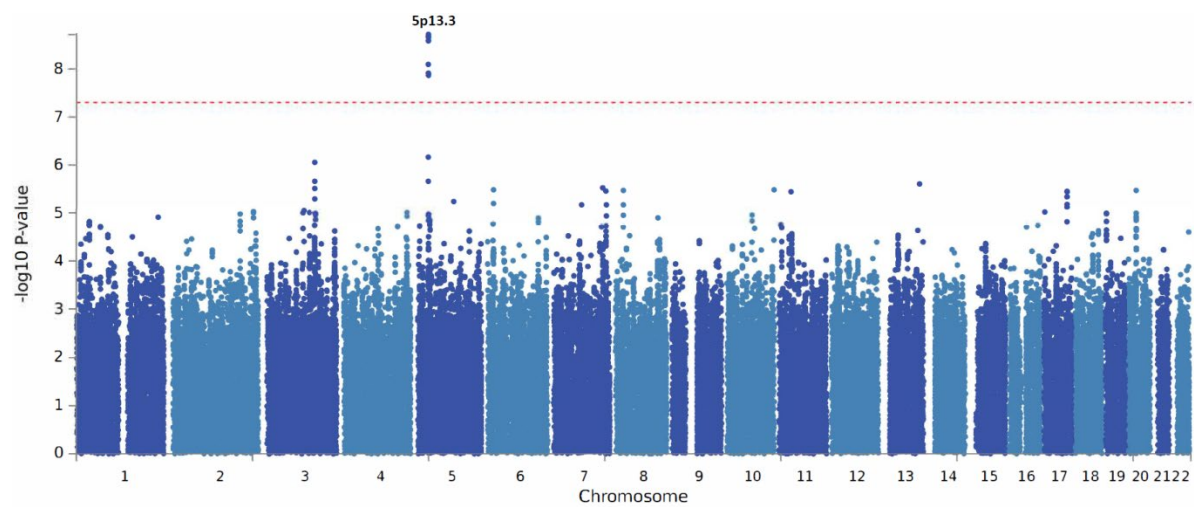


Supplementary Figure 2:

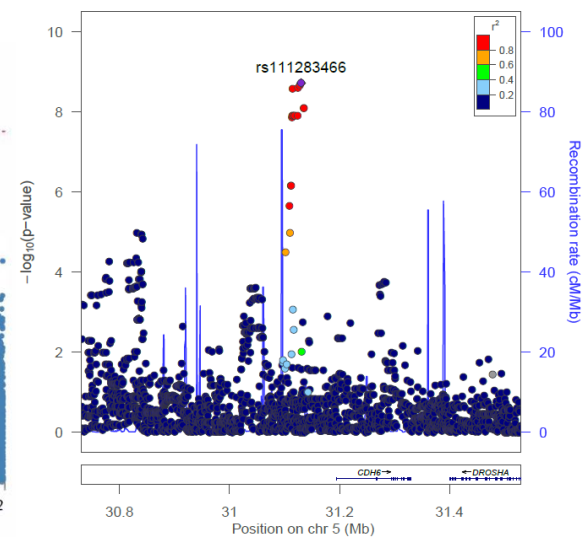


Supplementary Figure 3:

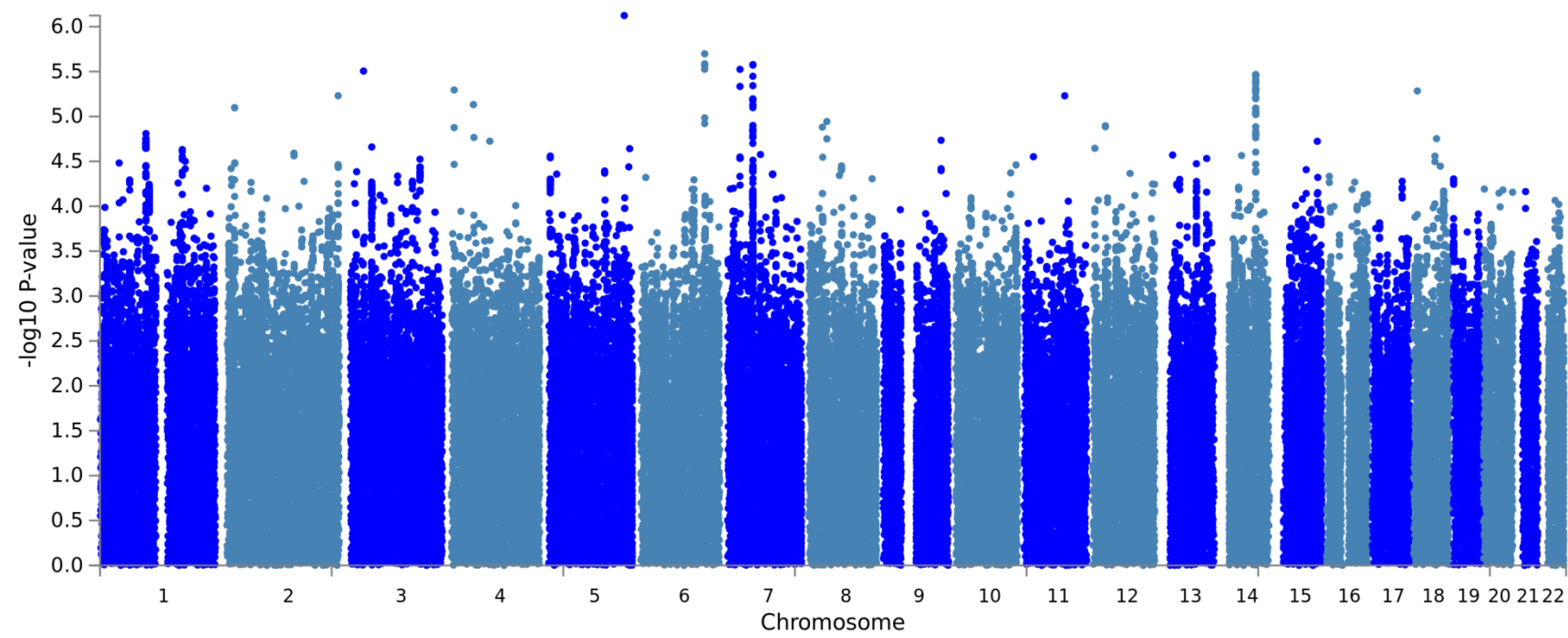
(a)



(b)

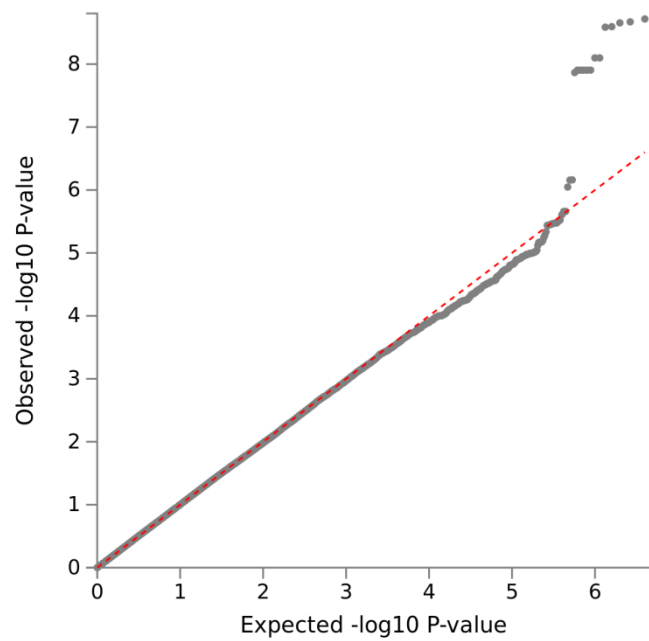


Supplementary Figure 4:

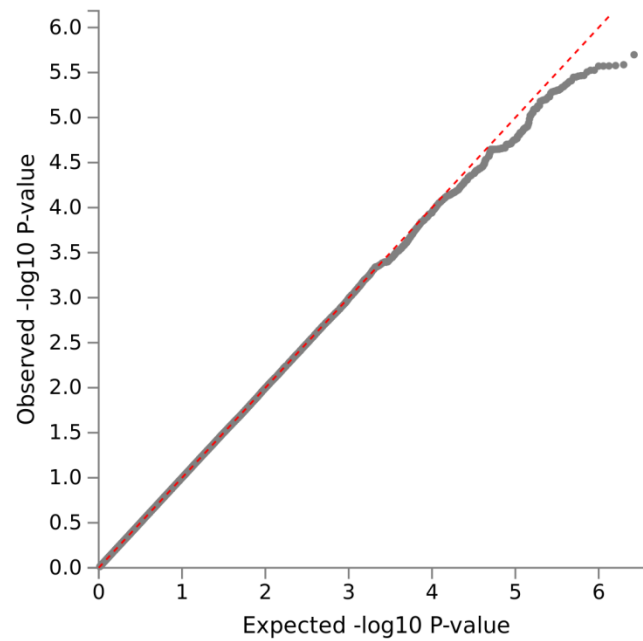


Supplementary Figure 5:

a)



b)



Supplementary Figure 1. Heatmap showing results of association protein levels with Alzheimer's disease (AD) reaching P -value < 0.05 . Color of each box indicates the magnitude of effect size (β) of association and star notations represent the strength of association based on P -values.

Supplementary Figure 2. Boxplots representing the normalized protein expression (NPX) levels of CDH6 and HAGH among $APOE$ 44/34/24 versus $APOE$ 33; $APOE$ 44/34 versus $APOE$ 22/23; $APOE$ 22/23 versus $APOE$ 33 in overall study sample and stratified by case-control status.

Supplementary Figure 3. (a) Manhattan plot for the CDH6 protein levels in plasma. Observed association of all tested single nucleotide polymorphisms (SNPs) on autosomal chromosomes (X -axis) are displayed as $-\log_{10}(P\text{-values})$ on Y -axis. Red line indicates a genome-wide significant association ($P < 2.5 \times 10^{-8}$) with CDH6 levels in plasma. Detected novel genome-wide significant loci 5p13.3 is led by rs11283466 SNP with P value = 1.92×10^{-9} . **(b)** Regional association plot for the association of rs11283466 with CDH6 protein levels in plasma. Plot represent all single nucleotide polymorphisms (SNPs) within 250kb region of lead SNP with height representing the $-\log_{10}(P\text{-values})$. Location of SNPs are based on NCBI build 37 and recombination rates are calculated based on the 1000 Genome project. Color scale represents the r^2 values indicating linkage disequilibrium of SNPs in the region with lead SNP. Arrows on the genes denote the 5'-3' prime orientation.

Supplementary Figure 4. Manhattan plot for the genome-wide association study results of HAGH protein levels in plasma.

Supplementary Figure 5. Quantile-Quantile plots for genome-wide association study results of **(a)** CDH6; **(b)** HAGH protein levels in plasma.