



Investigating *cis*-SNP/mRNA level associations of candidate Alzheimer's disease genes in different brain regions of patients and controls

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ABSTRACT: Background: It has been shown that mRNA levels of many human transcripts are under genetic control. We postulate that some Alzheimer's disease (AD) susceptibility variants confer disease risk via influencing gene expression in the brain. If correct, such variants will associate with both mRNA levels in the brain and disease risk. **Objective:** To investigate *cis*-SNP/mRNA level associations of 13 AD candidate genes in the cerebellum and temporal cortex of pathologically-confirmed AD patients and controls with other neurologic diseases. **Methods:** Using quantitative PCR, we measured mRNA levels of 13 AD candidate genes in the cerebella of 197 AD patients vs. 177 non-AD controls, and in the temporal cortex of 184 ADs vs. 185 non-ADs. We obtained *cis*-SNP genotypes that reside within the genes ± 1 cM flanking regions from the genome-wide association study of these subjects performed on the Illumina Hap300 platform (Carrasquillo et al., 2009). The *cis*-SNP/mRNA associations for each brain region and diagnostic group were obtained via multivariate linear regression analyses within PLINK, using age, gender and ApoE genotypes as covariates. When cases and controls were analyzed together, diagnosis was also included as an additional covariate. **Results:** There were 258 *cis*-SNP/mRNA associations detected in the cerebellum of ADs, 198 in non-ADs and 247 in the combined group with nominal p values of 0.05 to 3.05E-10. There were 164 *cis*-SNP/mRNA associations detected in the temporal cortex of ADs, 178 in non-ADs and 190 in the combined group with nominal p values of 0.05 to 2.14E-05. There were 7 genes with *cis*-SNP/mRNA associations in the ADs with nominal p<0.05 in both the cerebellum and the temporal cortex, and consistent direction of association with the minor allele. There were 3 such genes detected in the controls and five in the combined groups. Comparison of *cis*-SNP/mRNA associations in the ADs vs. non-ADs vs. combined ADs+non-ADs, revealed consistent associations in 4 genes in the cerebellum, where *IDE* *cis*-SNP/mRNA associations were the strongest and reached genome-wide significance in the ADs and combined subjects. Our findings on *IDE* *cis*-SNP/mRNA associations in the cerebellum are published (Zou et al., 2010). In the temporal cortex, two genes showed consistent and nominally significant associations in ADs, non-ADs and combined ADs+non-ADs. Although not reaching study-wide significance after stringent Bonferroni correction, the strongest *cis*-SNP/mRNA association in the temporal cortex occurred for *GSTO2*, previously implicated in AD as a candidate gene (Li et al., 2003) that may harbor e-SNPs with influence on its expression in the brain (Webster et al., 2009). **Conclusions:** These results suggest that there may exist variants in AD candidate genes that influence brain gene expression. Some of these variants appear to have brain-region and disease-specific effects whereas others may be more ubiquitous. Our top *cis*-SNP/mRNA hit in the temporal cortex, *GSTO2*, merits additional follow-up studies based on our data and previous publications to further delineate the role of *GSTO2* e-SNPs in brain gene expression and AD risk. (*Equal contribution).

Table 1. Genes and SNPs tested

Genes	Genic	Flanking	Total
APOE	0	207	207
CTNNA1	6	105	111
CTNNA3	228	204	432
CTSD	2	202	204
GAB2	14	222	236
GSTO2*	4	216	220
IDE	7	205	212
LRKK2	44	260	304
LRRM3	25	229	254
MAPT	14	119	133
SORCS2	117	314	431
SORT1	4	201	205
WWC1	34	291	325
TOTAL	499	2775	3274

Table 1. Brain expression levels of 13 genes were measured by quantitative PCR in the temporal cortex and cerebellum of ADs and non-ADs. *GSTO2 levels could not be detected in the cerebellum. Number of SNPs tested within (genic) or ± 1 cM flanking regions of each gene are shown.

Table 2. Autopsy subjects analyzed.

	TCX		CER	
N	184	185	197	177
Female	54%	41%	51%	36%
Mean age (±SD)	73.6 (5.4)	71.8 (5.4)	73.6 (5.6)	71.7 (5.5)

Figure 1. P values of *cis*-SNP/mRNA associations in CER of ADs+non-ADs

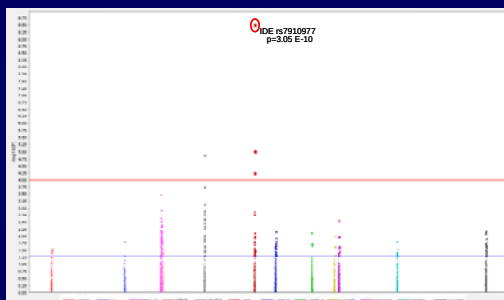


Figure 2. P values of *cis*-SNP/mRNA associations in TCX of ADs+non-ADs

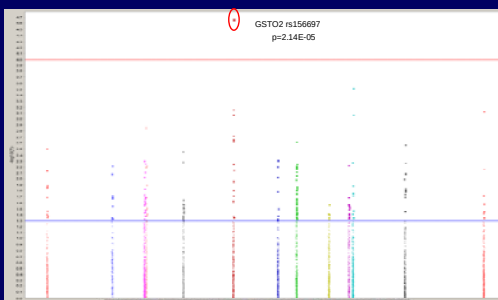


Table 3. Concordant and nominally significant *cis*-SNP/mRNA associations in the AD and non-AD CER.

CHR	SNP	BP	GENE	AD				CON				AD+CON			
				Beta	L95	U95	P	Beta	L95	U95	P	Beta	L95	U95	P
10	rs7910977	94199856	IDE	-1.00	-1.33	-0.67	1.25E-08	-0.35	-0.58	-0.12	0.003	-0.66	-0.86	-0.46	3.05E-10
10	rs1999763	94309972	IDE	-0.99	-1.43	-0.55	1.92E-05	-0.27	-0.54	-0.002	0.050	-0.58	-0.83	-0.33	9.09E-06
11	rs230669	77553034	GAB2	0.27	0.12	0.42	4.17E-04	0.24	0.09	0.40	0.003	0.25	0.14	0.36	1.26E-05
10	rs3737225	94239962	IDE	-1.10	-1.72	-0.47	7.05E-04	-0.49	-0.93	-0.05	0.032	-0.80	-1.19	-0.42	5.55E-05
11	rs2512081	78496771	GAB2	-0.19	-0.34	-0.04	0.017	-0.24	-0.40	-0.08	0.004	-0.21	-0.32	-0.10	1.69E-04
10	rs2619656	68509720	CTNNA3	-0.15	-0.30	-0.01	0.036	-0.14	-0.27	-0.004	0.045	-0.17	-0.26	-0.07	0.001
11	rs9667214	78495953	GAB2	-0.22	-0.42	-0.02	0.029	-0.26	-0.47	-0.05	0.015	-0.24	-0.38	-0.09	0.001
10	rs11187060	94294112	IDE	-0.29	-0.53	-0.06	0.016	-0.19	-0.35	-0.02	0.027	-0.24	-0.38	-0.09	0.001
10	rs7068134	67450904	CTNNA3	-0.20	-0.37	-0.04	0.015	-0.19	-0.36	-0.02	0.032	-0.19	-0.31	-0.07	0.002
10	rs7068134	67450904	LRRM3	-0.13	-0.25	-0.01	0.034	-0.19	-0.36	-0.02	0.032	-0.19	-0.31	-0.07	0.002

Table 4. Concordant and nominally significant *cis*-SNP/mRNA associations in the AD and non-AD TCX.

CHR	SNP	BP	GENE	AD				CON				AD+CON			
				Beta	L95	U95	P	Beta	L95	U95	P	Beta	L95	U95	P
10	rs156697	106029175	GSTO2	-0.24	-0.36	-0.11	2.53E-04	-0.18	-0.30	-0.06	0.004	-0.19	-0.28	-0.10	2.14E-05
10	rs157080	106045953	GSTO2	-0.22	-0.35	-0.08	0.002	-0.14	-0.27	-0.02	0.03	-0.16	-0.25	-0.07	8.88E-04
10	rs156699	106043631	GSTO2	-0.21	-0.35	-0.08	0.003	-0.14	-0.27	-0.02	0.03	-0.16	-0.25	-0.07	8.47E-04
4	rs1878519	8161682	SORCS2	0.20	0.05	0.35	0.009	0.26	0.07	0.45	0.01	0.21	0.09	0.34	8.59E-04
4	rs1281110	8238342	SORCS2	-0.17	-0.33	0.00	0.046	-0.25	-0.44	-0.06	0.01	-0.24	-0.36	-0.11	2.79E-04

Table 5. Concordant and nominally significant *cis*-SNP/mRNA associations in the AD CER and TCX.

CHR	SNP	BP	GENE	SNP-TYPE	Temporal Cortex				Cerebellum			
					Beta	L95	U95	P	Beta	L95	U95	P
4	rs4689673	7323995	SORCS2	GENIC	0.21	0.06	0.37	0.008	0.29	0.13	0.46	6.44E-04
10	rs7076847	68683472	CTNNA3	GENIC	-0.38	-0.71	-0.06	0.023	-0.38	-0.60	-0.16	6.93E-04
17	rs4793171	40487816	MAPT	FLANKING	-0.41	-0.65	-0.18	6.29E-04	-0.32	-0.55	-0.09	0.008
10	rs7004732	68881735	CTNNA3	GENIC	-0.29	-0.58	-0.01	0.047	-0.32	-0.52	-0.13	0.001
11	rs569072	77155604	GAB2	FLANKING	0.29	0.08	0.50	0.007	0.31	0.12	0.49	0.001
11	rs585513	77168498	GAB2	FLANKING	0.30	0.09	0.51	0.005	0.31	0.12	0.49	0.001
10	rs7083570	68806813	CTNNA3	GENIC	-0.31	-0.59	-0.03	0.031	-0.31	-0.50	-0.12	0.001
4	rs3845438	7327442	SORCS2	GENIC	0.35	0.11	0.59	0.004	0.40	0.15	0.65	0.002
10	rs2133696	68869124	CTNNA3	GENIC	-0.36	-0.67	-0.05	0.023	-0.32	-0.52	-0.12	0.002
10	rs6480263	68951300	CTNNA3	GENIC	-0.37	-0.67	-0.06	0.022	-0.32	-0.52	-0.12	0.002
10	rs6480274	68988527	CTNNA3	GENIC	-0.37	-0.67	-0.06	0.022	-0.32	-0.52	-0.12	0.002
10	rs4990414	68112054	LRRM3	FLANKING	-0.14	-0.27	-0.01	0.033	-0.18	-0.32	-0.05	0.007
1	rs53531	110630940	SORT1	FLANKING	0.16	0.03	0.29	0.010	0.16	0.03	0.28	0.013
10	rs3125309	69152443	CTNNA3	FLANKING	-0.20	-0.40	-0.01	0.043	-0.17	-0.31	-0.04	0.014
10	rs10509252	67491726	LRRM3	FLANKING	0.35	0.11	0.59	0.005	0.33	0.07	0.60	0.016
5	rs7101998	16842719	WWC1	FLANKING	0.34	0.01	0.66	0.045	0.35	0.07	0.63	0.016
17	rs3744474	40455596	MAPT	FLANKING	-0.19	-0.34	-0.03	0.024	-0.16	-0.32	-0.003	0.047

Table 6. Concordant and nominally significant *cis*-SNP/mRNA associations in the non-AD CER and TCX.

CHR	SNP	BP	GENE	SNP-TYPE	Temporal Cortex				Cerebellum			
					Beta	L95	U95	P	Beta	L95	U95	P
17	rs1908342	40659391	MAPT	FLANKING	0.28	0.12	0.44	8.89E-04	0.18	0.03	0.29	0.015
10	rs7912066	67698726	CTNNA3	GENIC	-0.30	-0.57	-0.02	0.0386	-0.27	-0.40	-0.13	1.80E-04
17	rs9116564	40660914	MAPT	FLANKING	0.33	0.13	0.54	0.001	0.18	0.02	0.33	0.024
17	rs9998256	40406871	MAPT	FLANKING	0.23	0.08	0.38	0.009	0.14	0.02	0.26	0.025
12	rs8004709	27023544	LRRK2	FLANKING	0.19	0.06	0.32	0.005168	0.12	0.01	0.23	0.028
10	rs10998638	67457843	CTNNA3	GENIC	0.48	0.13	0.83	0.007528	0.19	0.02	0.37	0.032
12	rs7976837	38651603	LRRK2	FLANKING	0.13	0.01	0.26	0.03403	0.13	0.03	0.23	0.011

Table 7. Concordant and nominally significant *cis*-SNP/mRNA associations in the AD+non-AD CER and TCX.

CHR	SNP	BP	GENE	SNP-TYPE	Temporal Cortex				Cerebellum			
					Beta	L95	U95	P	Beta	L95	U95	P
11	rs1942885	78490701	GAB2	FLANKING	-0.17	-0.33	-0.02	0.030	-0.19	-0.31	-0.07	0.002
10	rs7076847	68683472	CTNNA3	GENIC	-0.39	-0.67	-0.10	0.008	-0.26	-0.43	-0.09	0.003
10	rs2133696	68869124	CTNNA3	GENIC	-0.29	-0.56	-0.02	0.036	-0.23	-0.39	-0.08	0.004
10	rs10823057	68956367	CTNNA3	GENIC	0.27	0.01	0.54	0.041	0.21	0.05	0.38	0.011
10	rs7897267	94138780	IDE	FLANKING	0.15	0.01	0.29	0.042	-0.27	-0.47	-0.06	0.012
10	rs10998638	67457843	CTNNA3	GENIC	0.34	0.10	0.57	0.005	0.18	0.04	0.32	0.012
10	rs4587651	94113637	IDE	FLANKING	0.20	0.06	0.34	0.005	-0.26	-0.46	-0.05	0.015
5	rs10515498	137614229	CTNNA1	FLANKING	-0.17	-0.33	-0.02	0.028	0.13	0.02	0.24	0.016
10	rs7083570	68806813	CTNNA3	GENIC	-0.29	-0.54	-0.04	0.025	-0.18	-0.33	-0.03	0.019
10	rs1904611	68899313	CTNNA3	GENIC	-0.30	-0.56	-0.05	0.021	-0.18	-0.34	-0.03	0.020
17	rs2165846	42296365	MAPT	FLANKING	-0.11	-0.21	0.01	0.030	-0.10	-0.19	-0.01	0.023
10	rs7350385	68868513	CTNNA3	GENIC	-0.30	-0.56	-0.05	0.021	-0.17	-0.33	-0.02	0.032
11	rs569072	77155604	GAB2	FLANKING	0.25	0.08	0.42	0.004	0.14	0.01	0.27	0.034
10	rs1904607	68891190	GAB2	FLANKING	-0.28	-0.55	-0.02	0.034	-0.17	-0.33	-0.01	0.042
11	rs585513	77168498	GAB2	FLANKING	0.24	0.07	0.41	0.005	0.13	0.00	0.27	0.045
11	rs230656	77568377	GAB2	FLANKING	0.15	0.00	0.30	0.050	-0.11	-0.23	0.00	0.048