A Scan of Chromosome 10 Identifies a Novel Locus Showing Strong Association with Late-Onset Alzheimer Disease

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Strong evidence of linkage to late-onset Alzheimer disease (LOAD) has been observed on chromosome 10, which implicates a wide region and at least one disease-susceptibility locus. Although significant associations with several biological candidate genes on chromosome 10 have been reported, these findings have not been consistently replicated, and they remain controversial. We performed a chromosome 10–specific association study with 1,412 gene-based single-nucleotide polymorphisms (SNPs), to identify susceptibility genes for developing LOAD. The scan included SNPs in 677 of 1,270 known or predicted genes; each gene contained one or more markers, about half (48%) of which represented putative functional mutations. In general, the initial testing was performed in a white case-control sample from the St. Louis area, with 419 LOAD cases and 377 age-matched controls. Markers that showed significant association in the exploratory analysis were followed up in several other white case-control sample sets to confirm the initial association. Of the 1,397 markers tested in the exploratory sample, 69 reached significance ($P < .05$). Five of these markers replicated at $P < .05$ in the validation sample sets. One marker, *rs498055*, located in a gene homologous to *RPS3A* (*LOC439999*), was significantly associated with Alzheimer disease in four of six case-control series, with an allelic *P* value of .0001 for a meta-analysis of all six samples. One of the case-control samples with significant association to *rs498055* was derived from the linkage sample ($P = .0165$). These results indicate that variants in the *RPS3A* homologue are associated with LOAD and implicate this gene, adjacent genes, or other functional variants (e.g., noncoding RNAs) in the pathogenesis of this disorder.

Alzheimer disease (AD [MIM 104300]) is the most significant cause of dementia in developed countries and is clinically characterized by memory loss of subtle onset followed by a slowly progressive dementia that has a course of several years. The risk of AD has a genetic component, as evidenced by an increased risk of AD among first-degree relatives of affected individuals. So far, three genes have been identified that lead to the rare autosomal dominant early-onset form of AD. Mutations in the three genes—β-amyloid precursor protein (*APP* [MIM 104760]) (Goate et al. 1991), presenilin 1 (*PSEN1* [MIM 104311]) (Sherrington et al. 1995), and presenilin 2 (*PSEN2* [MIM 600759]) (Levy-Lahad et al. 1995)—lead to an increase in the production of long amyloid β peptide (Aβ42), the main component in amyloid plaques. The great majority of AD cases are of late onset (age at onset >65 years) and show complex, non-Mendelian patterns of inheritance. Late-onset AD (LOAD [MIM 606626]) probably results from the combined effects of variation in a number of genes as well as from environmental factors. Early genetic studies of LOAD demonstrated that the ε4 variant of *APOE* (MIM 107741) is associated with increased risk of LOAD and with lower age at disease onset in a dose-dependent manner (Corder et al. 1993).

Genomewide linkage screens in patients with LOAD have identified several other chromosomal regions (reviewed by Pastor and Goate [2004]), implying that genetic risk factors other than *APOE* must exist. Putative LOAD-susceptibility loci on chromosomes 9, 10, and 12 have been reported in two or more sample sets by dif-

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ferent groups (Pericak-Vance et al. 1997, 2000; Rogaeva et al. 1998; Kehoe et al. 1999; Myers et al. 2000, 2002; Blacker et al. 2003). Perhaps the most prominent among them is the linkage to chromosome 10, observed in a number of nonoverlapping samples from studies employing distinct approaches, including linkage analysis based on a genomewide screen, a candidate gene–based limited genome screen, and a genome screen that used plasma Aβ levels as a quantitative phenotype (Kehoe et al. 1999; Bertram et al. 2000; Ertekin-Taner et al. 2000; Myers et al. 2000; Blacker et al. 2003; Farrer et al. 2003). Several candidate genes that are under or near the chromosome 10 linkage peaks have been tested for association with LOAD, but none has been consistently replicated (Alzheimer Disease Forum).

To identify the genes and mutations for LOAD, we undertook a screen of putative functional SNPs in 677 genes under the linkage peak, using a powerful set of unrelated cases and controls. A similar approach was used to identify the glyceraldehyde-3-phosphate dehydrogenase gene (*GAPD* [MIM 138400]), located on the short arm of chromosome 12, as a putative LOAD risk gene (Li et al. 2004). Here, we report the findings from this scan of 1,412 SNPs on chromosome 10.

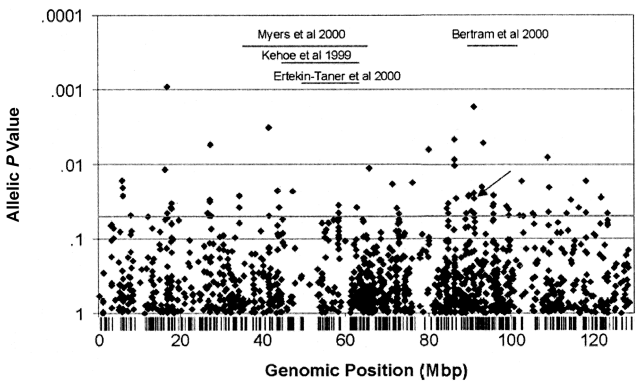

Figure 1 Allelic *P* values of 1,397 exploratory markers from the exploratory sample (*middle*), with a bar graph showing the distribution of annotated genes across chromosome 10 (*bottom*). Marker *rs498055* is noted with an arrow, and a *P* value of .05 is marked with a line. The previously identified linkage peak regions are noted with solid lines and references (*top*). Studies with multipoint LOD scores >2 in white samples were included. Results of single-marker studies were not included.

Material and Methods

Sample-Set Characteristics

Three white clinical case-control series were used in this study: (1) the WU series (422 cases; 382 controls), collected through the Washington University Alzheimer's Disease Research Center (ADRC) patient registry; (2) the UK series (368 cases; 404 controls), collected as part of the Medical Research Council (MRC) Late-Onset AD Genetic Resource, including those from the Cardiff University Wales School of Medicine and from King's College London; and (3) the UCSD series (217 cases; 409 controls), collected through the ADRC of the University of California–San Diego. In total, 1,007 AD cases and 1,195 controls were analyzed. Cases in these series had received a clinical diagnosis of dementia of the Alzheimer type (DAT), with use of criteria equivalent to NINCDS-ADRDA (National Institute of Neurological and Communicative Diseases and Stroke/Alzheimer's Disease and Related Disorders Association) (McKhann et al. 1984) but modified slightly to include AD as a diagnosis for individuals aged >90 years (Berg et al. 1998). The minimum age at onset of DAT was 60 years. Controls were nondemented individuals aged >60 years at assessment who were screened for dementia through use of neuropsychological tests and clinical interviews. Controls were matched with cases for age and sex. These samples all show an expected age and *APOE* ε4–genotype distribution and do not appear to have evidence of population stratification (Li et al. 2004). More-detailed information about these samples can be found elsewhere (Li et al. 2005).

A fourth case-control series was generated by selecting one case per family from our genetic linkage sample (Myers et al. 2002) and matching each of them to an equal number of white, nondemented controls collected in St. Louis (these controls are independent of the controls used in the exploratory sample above). There were 429 cases and 321 controls in this series (mean age at onset for the case series is 73.6 years; mean age at assessment for controls is 75.0 years). The linkage pedigrees from the National Institute of Mental Health (NIMH) series and the NIA series (292 pedigrees; 624 affected individuals) (Myers et al. 2002) were also genotyped for the single SNP significant in all case-control series.

Two small series that consisted of neuropathologically confirmed white cases and controls were derived from the U.S. ADRCs (contributing centers are listed in the Acknowledgments) and from Newcastle upon Tyne, United Kingdom. Of the samples in the U.S. series, 40% were assessed as being at either Braak and Braak stage 5 or 6 (cases) or Braak stage 2 or less (controls). The remaining samples (cases) met neuropathological criteria for AD. Both the controls and cases were selected to be largely free of such complicating pathologies as Lewy bodies and vascular events. The combined series included 360 cases (age range 65–97 years; 220 women) and 252 controls (age range 65–100 years; 123 women).

SNP Selection and Genotyping

Genotyping of all samples was performed with written informed consent/assent from the participating individuals and their caregivers and approval from the participating institutions. Polymorphisms used for genotyping were identified from either the Celera human genome database that includes publicly available SNP data or the Applera Genome Sequencing Initiative database. For this study, we chose gene-based SNPs, with a preference for putative functional mutations, as predicted in the Celera or public SNP databases, with the aim to screen as many predicted genes with at least one variant as possible (table A1 and fig. A1 [online only]). Thus, these SNPs consist of 367 missense/nonsense mutations, 1 donor splice-

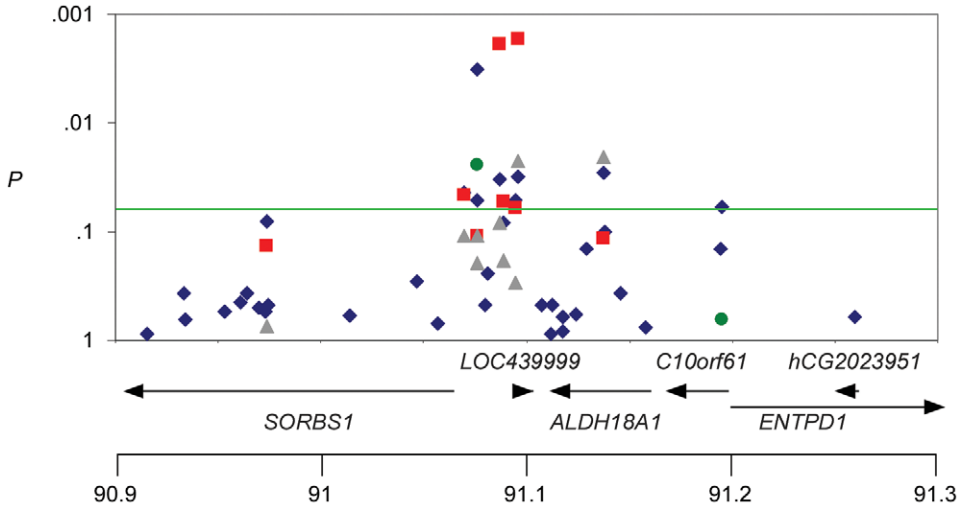

Figure 2 Allelic *P* values of markers around the *RPS3A* homologue region (*LOC43999*) in both exploratory and validation samples, along with a gene map of the region and Celera assembly coordinates (in Mbp). Blue diamonds indicate two-sided explatory sample *P* values; the other symbols indicate one-sided replication sample *P* values for WU (*red squares*), UCSD (*gray triangles*), and UK (*green circle*).

site variant, 172 putative transcription factor binding site mutations, 9 exon-skipping site variants, 109 variants in the UTR, and 739 variants of other types (intronic, silent, and unknown types [SNPs of unknown and silent types were annotated as functional variants in previous genome assemblies]). They cover a total of 677 of 1,270 annotated genes on chromosome 10. All genomic positions for all SNPs and genes are from the Celera Genome Assembly R27. All SNPs had a minor-allele frequency (MAF) of >2% in either cases or controls. The MAF was 2%–5% for 80 exploratory markers and 5%–10% for 165 markers. The remaining markers had MAFs of 10%–50%, with approximately equal numbers of SNPs in each 10% interval.

Genotyping of SNPs was undertaken by allele-specific real-time PCR for individual samples, by use of primers designed and validated in-house (Germer et al. 2000). Cases and controls were always run on the same plate in a blind fashion. Assay quality was scored by an individual who had no access to the sample phenotypes, before the genotyping results were subjected to statistical analysis. Overall, the accuracy of our genotyping was >99%, as determined by internal comparisons of differentially designed assays for the same marker and by comparisons of the same marker across different groups.

Genotyping was performed in stages—markers were first genotyped in one sample set, the exploratory set. Generally, the WU sample set was used as the exploratory sample set. However, when markers were tested for replication in another sample set, we also genotyped that sample set with novel assays that had passed our assay-validation step. Overall, we used the UK sample (105 assays) and the UCSD sample (1 assay) as exploratory sets for <8% of all tested assays. Significant exploratory markers ($P < .05$) were then genotyped in two additional clinical case-control series. After replication in at least one of these other sample sets, additional fine-mapping markers were genotyped near the replicated SNPs. When additional assays for markers near significant exploratory markers were immediately available, they were genotyped in the exploratory sample in parallel with attempting to replicate in the validation samples. Significant markers were followed up as described above. Five SNPs that showed some level of replication in one or both of the additional case-control series were genotyped in a case-control series derived from the families originally used for our genomewide linkage scan. One of these SNPs (*rs498055*) was also genotyped in the case-control series that was composed of neuropathologically confirmed AD cases and controls.

Statistical Analysis

To help exclude assays with possible genotyping errors from the analysis, Hardy-Weinberg equilibrium tests were first performed in both the case and the control samples. Assays with significant deviation from Hardy-Weinberg equilibrium in controls were then examined for genotyping quality ($P < .05$; 63 markers in the exploratory stage). As a result, two assays were dropped from our analysis. One remaining assay with an MAF <10% was significant in the exploratory set but did not validate in the other sample sets and was in Hardy-Weinberg equilibrium.

Pearson's χ^2 test was used to calculate *P* values for the association of an allele with disease status within a single study. This test of association was performed on the basis of the frequency counts of a 2 × 2 contingency table of allele and disease status. Two-sided *P* values are presented for the exploratory study. In the validation stage, one-sided *P* values were calculated if the odds ratios (ORs) were in the direction observed in the exploratory stage. *P* values were not adjusted for multiple comparisons unless otherwise stated. ORs and the 95% CIs for an allelic effect were also estimated. ORs and *P* values for meta-analyses that combine results of multiple sample sets were calculated using the Cochran-Mantel-Haenszel test, and were controlled for the sample set (Agresti 1990). Evidence of

Table 1

Allelic Tests of Replicated Markers and LOAD

Marker, Gene, Position (bp) and Sample	Cases					Controls							Power to Replicate
	No. with Genotype				MAF	No. with Genotype				MAF			
	11	12	22	Total	(%)	11	12	22	Total	(%)	*P*	OR (CI[a])	
rs1057971, PCGF5, 86733401:													
WU[b]	2	54	363	419	6.9	0	33	344	377	4.4	.029	1.62 (1.05–.52)	...
UCSD[c]	2	33	213	248	7.5	2	37	360	399	5.1	.044	1.49 (1.01–.19)	.61
UK[c]	1	39	307	347	5.9	3	32	345	380	5.0	.22	1.19 (.82–.75)	.66
UCSD and UK[c]	3	72	520	595	6.6	5	69	705	779	5.1	.042	1.33 (1.01–.74)	.88
All	5	126	883	1,014	6.7	5	102	1,049	1,156	4.8	.0068	1.43 (1.10–.85)	...
rs498055, LOC439999, 91096111:													
UK[b]	80	175	92	347	48.3	67	194	124	385	42.6	.029	1.26 (1.02–.55)	...
UCSD[c]	64	107	48	219	53.7	85	156	102	343	47.5	.022	1.28 (1.04–.56)	.59
WU[c]	125	175	89	389	54.6	65	200	86	351	47.0	.0017	1.36 (1.14–.61)	.71
UCSD and WU[c]	189	282	137	608	54.3	150	356	188	694	47.3	.00021	1.32 (1.16–.51)	.89
All	269	457	229	955	52.1	217	550	312	1,079	45.6	.00004	1.3 (1.15–.47)	...
rs4417206, ALDH18A1, 91137678:													
UK[b]	36	153	158	347	32.4	61	169	154	384	37.9	.029	.79 (.63–.98)	...
UCSD[c]	17	102	101	220	30.9	50	157	142	349	36.8	.021	.77 (.62–.95)	.59
WU[c]	45	155	190	390	31.4	38	165	148	351	34.3	.12	.88 (.73–.05)	.71
UCSD and WU[c]	62	257	291	610	31.2	88	322	290	700	35.6	.013	.83 (.72–.95)	.90
All	98	410	449	957	31.7	149	491	444	1,084	36.4	.0019	.81 (.71–.93)	...
rs600879, SORCS1, 102662200:													
WU[b]	6	87	325	418	11.8	4	54	319	377	8.2	.017	1.5 (1.07–.09)	...
UCSD[c]	3	42	196	241	10.0	6	53	335	394	8.2	.15	1.23 (.89–.71)	.67
UK[c]	6	66	277	349	11.2	3	63	319	385	9.0	.079	1.28 (.96–.70)	.74
UCSD and UK[c]	9	108	473	590	10.7	9	116	654	779	8.6	.040	1.26 (1.01–.56)	.92
All	15	195	798	1,008	11.2	13	170	973	1,156	8.5	.0043	1.34 (1.10–.65)	...
rs1903908, hCG2039140, 102940843:													
WU[b]	14	97	308	419	14.9	5	77	294	376	11.6	.050	1.34 (1.00–.80)	...
UCSD[c]	6	54	188	248	13.3	2	81	314	397	10.7	.079	1.28 (.96–.71)	.53
UK[c]	11	78	247	336	14.9	10	76	296	382	12.6	.10	1.22 (.94–.57)	.57
UCSD and UK[c]	17	132	435	584	14.2	12	157	610	779	11.6	.029	1.24 (1.03–.50)	.80
All	31	229	743	1,003	14.5	17	234	904	1,155	11.6	.0070	1.28 (1.07–.53)	...

[a] 95% CI for exploratory and total samples; 90% CI for validation samples.

[b] Exploratory sample set.

[c] Validation sample set: one-sided *P* value.

Table 2

Allelic Association in Linkage Case-Control Series

Marker	Location (Mb)	No. of Cases with Genotype			No. of Controls with Genotype				
		11	12	22	11	12	22	P[a]	OR (95% CI)
rs1057971	86.7	1	35	304	0	41	302	.73	.91 (.57–1.43)
rs498055	91.09	110	207	112	58	162	96	.017	1.26 (1.02–1.54)
rs4417206	91.13	45	163	145	59	139	131	.24	.87 (.70–1.09)
rs600879	102.66	285	62	6	275	56	8	1	1.01 (.72–1.43)
rs1903908	102.94	13	87	265	6	84	252	.45	1.12 (.84–1.51)

NOTE.—For cases, one affected sibling was genotyped from each family in the linkage sample (Myers et al. 2002) and was compared with a set of independent controls.

[a] Allelic tests are two sided except for *rs498055*.

replication, rather than multiple testing corrections, was used to evaluate the significance of associated SNPs.

Linkage Analysis

To determine whether *rs498055* contributed to our linkage signal on chromosome 10, we stratified families on the basis of the presence or absence of the risk allele of *rs498055* in the proband of each family. The families used in this analysis were the NIMH and the National Cell Repository for Alzheimer's Disease families from our linkage screen (Myers et al. 2002). The analysis was performed in Mapmaker/SIBS ("All pairs, UNWEIGHTED"). For the "proband" analysis, the (numerically) first individual with the SNP genotype was identified as the proband.

Haplotype Analysis

Several studies have shown that placing individual SNPs into the context of a haplotype increases biological information (Balciuniene et al. 2002; Knoblauch et al. 2002; Van Eerdewegh et al. 2002). Similarly, placing haplotypes into their evolutionary context also increases biological information (Templeton et al. 2005). For the haplotype analysis, we used SNPs that were typed in all three series and were located within ~40 kb of *rs498055*. These criteria resulted in a data set of 11 SNPs in 1,159 controls and 974 cases from the WU, UK, and UCSD case-control samples.

Haplotypes were estimated using the software PHASE (Stephens et al. 2001; Stephens and Donnelly 2003). A set of 95%-plausible haplotype trees was estimated using statistical parsimony in the program TCS (Clement et al. 2000; Templeton et al. 2000).

Association with LOAD was tested by tree scanning (Templeton et al. 2005), which was modified to manage case-control data (Nowotny et al. 2005). A tree scan uses the haplotype network to define tests that are based on each branch of the tree. Each branch represents an a priori defined pooling of haplotypes: haplotypes on one side of the branch are pooled together and define an allele, whereas the haplotypes on the other side are pooled to define a separate allele. This results in a biallelic locus that can be tested for association with the phenotype. A permutation-based analog of the sequential Bonferroni (Westfall and Young 1993) was used to obtain nominal and multiple-test–corrected significance values with the parametric P value used as the test statistic. This permutation method takes into account the correlation structure between tests while correcting for multiple tests.

Results

To identify genetic variation associated with LOAD on chromosome 10, we performed a SNP-based association study with three well-characterized LOAD case-control series. Our strategy was to test markers in one sample set (exploratory sample) and to follow up significant markers in the two remaining sample sets (validation samples). Using this paradigm, we first scanned a relatively large number of gene-based putative functional SNPs across chromosome 10, with the highest SNP den-

Table 3

Linkage Analysis of Pedigrees Stratified by *rs498055*

Sample	No. of Pedigrees	No. Affected	Peak LOD	Location (cM)
All	343	733	3.84	68
Pedigrees with SNPs	292	624	3.18	68
Probands with A	228	488	3.37	68
Probands with G	221	471	2.38	50
Probands without A	64	136	.73	90
Probands without G	71	153	1.18	67

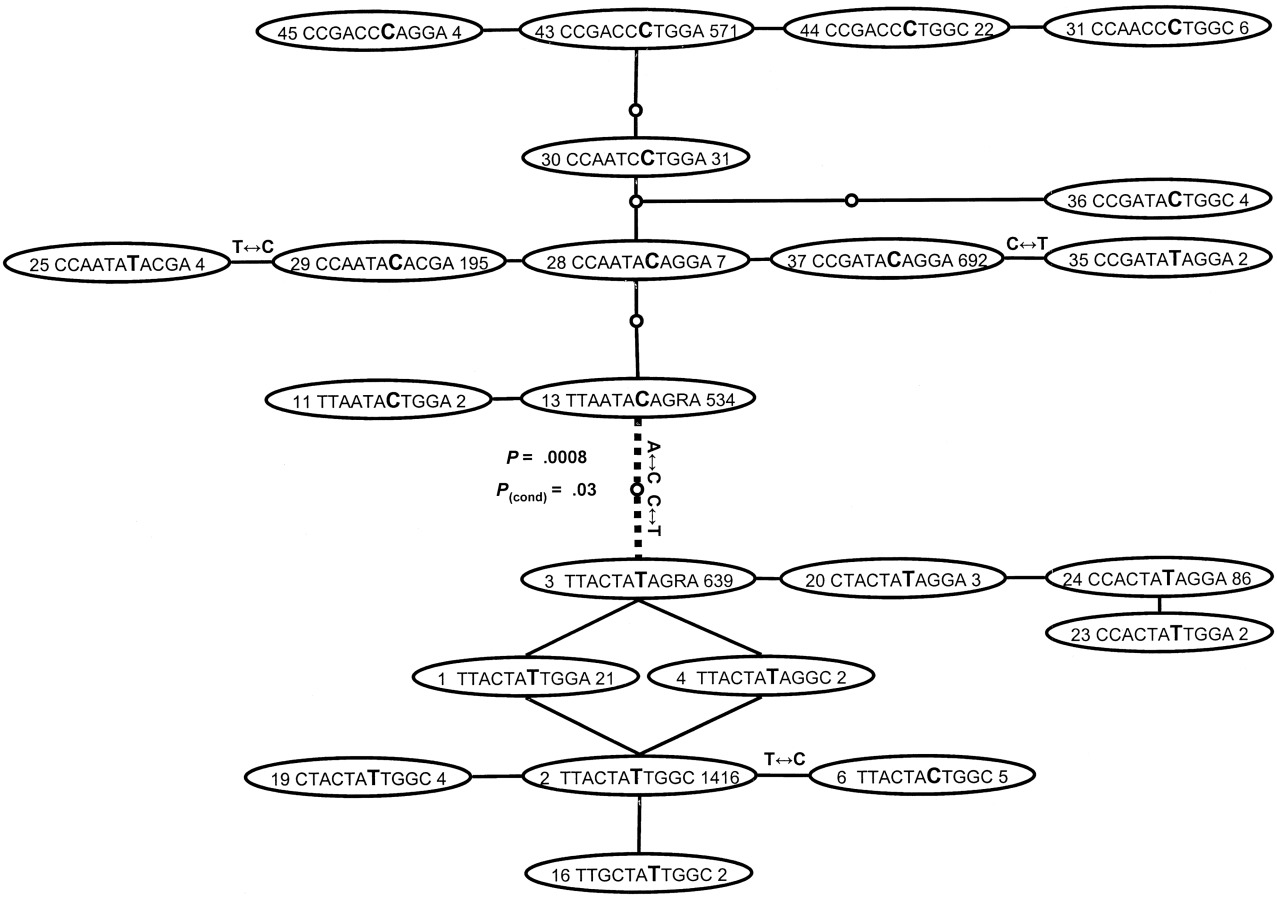

Figure 3 Haplotype networks. Each oval contains the haplotype identification number, the state at each locus, and the number of times it was inferred to occur in this sample set. To simplify the presentation of the network, haplotypes that appear only once in the sample are not shown, and selected haplotypes have been collapsed. The branch that was significant in the tree scan is denoted by the dashed line. *P* values for the original and conditional analyses are also provided. Mutations at *rs498055* are indicated by "T⟷C"; the mutation at *rs495998* is indicated by "A⟷C."

sity in regions directly under the linkage peak reported above. Significant markers were then genotyped in the other two sample sets to attempt replication of the initial association. Regions with markers showing strong association with the exploratory sample and replication in at least one other sample set were then tested with additional markers. Specifically, we genotyped a total of 1,397 SNPs by allele-specific PCR in the exploratory stage (fig. 1), targeting 674 genes. From these, we genotyped 408 genes with 1 marker, 141 with 2 markers, 57 with 3 markers, 47 with 4–7 markers, and the remainder with ⩾8 markers. The majority of exploratory markers (1,291) were tested in the WU sample set. In the UK sample set, 105 markers were genotyped, and 1 marker was genotyped in the UCSD sample set. Of the 1,397 tested SNPs, 69 were significantly associated with LOAD in the exploratory sample ($P < .05$). These markers were scattered across the chromosome, as would be expected because of the high probability of false-positive associations due to the large number of SNPs analyzed (fig. 1). We subsequently genotyped the 69 markers in the two validation sample sets and found 5 that replicated in a meta-analysis combining the two validation sample sets (one-sided $P < .05$) (table 1). One marker, *rs498055*, located in *LOC439999*, a gene with high homology to *RPS3A* (MIM 180478), was significant ($P < .05$) in each of the three sample sets and was the most significant ($P = .00004$) marker in the three-sample meta-analysis (table 1). One other marker, *rs4417206*, located in a neighboring gene *ALDH18A1* (or *PYCS* [MIM 138250]), was also significant in the combined validation study ($P = .013$). Markers *rs4417206* in *ALDH18A1* and *rs498055* in *LOC439999* are ~41 kb apart and are in strong linkage disequilibrium (LD) with one another ($D' = 0.98$; $r^2 = 0.43$).

To determine whether any of the five SNPs that replicated in the meta-analysis (*rs1057971*, *rs498055*, *rs4417206*, *rs600879*, and *rs1903908*) were also asso-

Table 4

Measures of Pairwise D' and r^2 in UK Controls

Marker	Distance	*rs500470*	*rs533383*	*rs533343*	*rs11594687*	*rs7895441*	*rs495998*	*rs17110999*	*rs7906450*	*rs498055*	*rs2296690*
rs500470	.00		1.00	1.00	1.00	1.00	.89	1.00	1.00	.88	.95
rs533383	5.78	.99		.99	1.00	1.00	.89	1.00	1.00	.88	.95
rs533343	.01	.74	.73		1.00	.66	.99	1.00	.92	.98	1.00
rs11594687	4.03	.07	.07	.05		1.00	1.00	1.00	1.00	1.00	1.00
rs7895441	1.40	.37	.37	.22	.03		1.00	1.00	.94	.98	1.00
rs495998	5.76	.54	.54	.48	.11	.25		1.00	1.00	.99	.77
rs17110999	2.17	.26	.27	.36	.02	.72	.18		1.00	1.00	1.00
rs7906450	5.57	.28	.28	.32	.02	.68	.19	.94		1.00	.85
rs498055	1.49	.52	.51	.47	.11	.24	.98	.18	.19		.78
rs2296690	16.03	.09	.09	.07	.69	.04	.10	.03	.02	.10	
rs1804934	.20	.04	.04	.05	.00	.00	.02	.00	.00	.02	.00
rs749049	4.96	.01	.01	.00	.10	.06	.11	.19	.20	.11	.11
hDV68531050	.02	.06	.06	.02	.00	.18	.04	.01	.01	.04	.00
rs2986401	6.55	.03	.03	.04	.05	.19	.00	.33	.30	.00	.07
rs2275272	5.51	.09	.09	.07	.74	.03	.09	.02	.03	.09	.94
rs4417206	8.31	.30	.29	.21	.05	.11	.44	.08	.09	.43	.07
rs11188410	.09	.14	.13	.19	.01	.01	.10	.00	.00	.09	.01
rs10882645	7.81	.03	.03	.03	.05	.17	.00	.31	.33	.00	.06
hDV68531048	12.0	.06	.06	.02	.00	.18	.04	.01	.01	.04	.00
rs11553577	37.1	.14	.13	.19	.01	.00	.09	.00	.00	.08	.02
hCV25943811	.24	.06	.06	.02	.00	.18	.04	.01	.01	.04	.00
rs1418709	65.0	.00	.00	.02	.11	.18	.05	.12	.13	.05	.15

NOTE.—Measures of pairwise D' are shown above the diagonal; r^2 values are shown below the diagonal.

ciated with risk for LOAD in our original linkage study sample (Myers et al. 2002), we genotyped the entire series and performed two analyses. First, we used a case-control approach by selecting one case (proband) per family, and we matched each of them to an equal number of unrelated controls. We chose to use a case-control analysis rather than a discordant–sib-pair analysis because of the greater power in the case-control design and because discordant siblings were available for only a proportion of the cases. A one-sided χ^2 test demonstrated significant evidence of association in the case-control sample with the same allele as in the other case-control series for *rs498055* ($P = .0165$); all other SNPs failed to show any evidence of association (table 2). The ORs observed in the linkage series for *rs498055* were similar to those observed in the other case-control series (OR = 1.26; 95% CI 1.02–1.54).

Marker *rs498055* was also examined in two small series (183 cases/127 controls; 160 cases/106 controls) of neuropathologically confirmed cases and controls. The SNP was not associated with AD risk in these samples ($P = .63$ and $P = .21$, respectively). However, power to replicate our finding in these samples was low (40% and 36%, respectively; 60% power in the combined sample sets).

To further estimate the effect of *rs498055* in the linkage sample, we performed a stratified linkage analysis of the stage II linkage data, on the basis of the genotype of the proband of each pedigree. We performed stratified linkage analyses using the pedigrees in which the proband had a copy of allele A and pedigrees in which the proband had a copy of allele G (table 3). We also considered pedigrees in which the proband was a homozygote for the A allele and in which the proband was a homozygote for the G allele. The results did not show an increase in LOD score in probands with the risk allele. In fact, although the first two groups were roughly the same size, the LOD score was substantially smaller in pedigrees in which the proband had a copy of the risk allele. This suggests that the *rs498055* polymorphism (at 91.1 Mb) may have little direct effect on the linkage findings, which have their peak near *D10S1211* (at 59.9 Mb), and that other loci contributing to disease have yet to be found in this region.

These findings prompted us to focus further follow-up on the region flanking these two genes. A total of 53 markers, covering a 1.49-Mb region, were typed in the exploratory sample, and association of these SNPs was examined. Ten of the markers resulted in a P value <.1 in the exploratory sample, and five were significant at $P < .05$ (fig. 2). After genotyping these markers in the validation samples, *rs498055* remained the only marker that was significantly associated with LOAD in each of the three sample sets.

We examined LD structure in this region, using genotypes from the UK and the WU sample sets. We observed a block of high LD extending from *rs500470* to *rs1418709,* covering at least 190 kb of the genomic region that includes the most-significant markers, *rs498055* and *rs4417206*. Although the D' values among neighboring SNPs were high, the r^2 values were generally low (table 4). The LD structure was comparable between cases and controls. The five significant markers with a P value <.05 (*rs500470, rs533343, rs495998, rs498055,*

rs1804934	*rs749049*	*hDV68531050*	*rs2986401*	*rs2275272*	*rs4417206*	*rs11188410*	*rs10882645*	*hDV68531048*	*rs11553577*	*hCV25943811*	*rs1418709*
1.00	.12	.92	.20	1.00	.98	1.00	.18	.92	.96	.92	.02
1.00	.11	.92	.20	1.00	.98	.96	.19	.92	.93	.92	.03
1.00	.04	1.00	.19	1.00	.98	1.00	.16	1.00	.97	1.00	.20
.68	1.00	.05	1.00	1.00	1.00	1.00	1.00	.05	1.00	.05	1.00
.76	.50	.95	.63	1.00	1.00	.62	.60	.95	.25	.95	.88
1.00	.37	.91	.09	.76	.99	1.00	.09	.91	.94	.91	.23
.16	1.00	1.00	.96	1.00	1.00	.26	.94	1.00	.04	1.00	.86
.27	1.00	1.00	.90	1.00	1.00	.39	.95	1.00	.17	1.00	.87
1.00	.37	.91	.08	.77	.98	.94	.08	.91	.89	.91	.23
1.00	.87	.44	1.00	1.00	1.00	1.00	.86	.44	1.00	.44	1.00
	1.00	1.00	1.00	.99	.98	1.00	1.00	1.00	1.00	1.00	1.00
.02		1.00	.04	1.00	1.00	1.00	.02	1.00	.83	1.00	.41
.00	.04		1.00	.32	1.00	.93	1.00	1.00	.63	1.00	1.00
.01	.00	.02		1.00	1.00	1.00	.99	1.00	1.00	1.00	.94
.00	.13	.00	.07		1.00	1.00	1.00	.32	1.00	.32	1.00
.01	.54	.02	.23	.07		1.00	1.00	1.00	.86	1.00	.91
.00	.08	.00	.04	.01	.04		1.00	.93	1.00	.93	1.00
.01	.00	.02	.95	.07	.23	.04		1.00	1.00	1.00	.94
.00	.04	1.00	.02	.00	.02	.00	.02		.63	1.00	1.00
.00	.06	.00	.05	.01	.03	.95	.05	.00		.63	.94
.00	.04	1.00	.02	.00	.02	.00	.02	1.00	.00		1.00
.02	.16	.05	.42	.14	.41	.09	.42	.05	0	.05	

and *rs4417206*) were all located within this block and exhibited higher r^2 values with *rs498055* than with other neighboring SNPs. (All had $r^2 > 0.43$ with *rs498055*.) Comparison of these results with data in the HapMap project indicates that the block containing *rs498055* extends 419 kb and contains seven genes, *LOC439999, ALDH18A1* (MIM 138250), *C10orf61, ENTPD1* (MIM 601752), *hCG2023951, hCG1781136,* and *C10orf130*.

The tree-scan analysis of 11 SNPs in the region surrounding *rs498055* identified significant results across many branches of the haplotype network. However, the results of the conditional tests suggest that the association observed at these branches is due to their location in the network relative to a single branch. This branch was significant in both the original ($P = .0008$) and the conditional ($P = .03$) analyses (fig. 3). It is marked by mutations creating the SNPs *rs498055* and *rs495998*.

Discussion

Genetic variants in several biological candidate genes under or near the chromosome 10 linkage peaks—including mutations in *CTNNA3* (MIM 607667), *PLAU* (MIM 191840), *IDE* (MIM 146680), and others—have been reported to be associated with LOAD. However, none of the associations in these candidate genes has been consistently replicated (Alzheimer Disease Forum). Indeed, our own studies in the case-control series used in the present study showed no evidence of association with any of these genes (Myers et al. 2004; Nowotny et al. 2005). These findings suggest that the reported association may be false, although it remains possible that the lack of consistent replication may be due to type 1 error, genetic heterogeneity, population stratification, and/or a small genetic effect confounded by sample sizes insufficient to replicate the initial reports. With the technology that was available to us, we performed a broadly scaled and nonbiased genotyping program. This approach would inevitably be burdened by a requirement of multiple-testing corrections to assess potential associations. To mitigate this, we designed a two-step process in which we genotyped ~1,400 SNPs in the exploratory sample set but only 69 markers in the subsequent validation sample sets. This strategy led us to identify five SNPs, located in five genes on chromosome 10, that are associated with LOAD. Although our genotyping scan covers the entire chromosome 10, these significant SNPs are located relatively close to linkage peaks identified in other studies (Bertram et al. 2000). Our analysis included 12 SNPs in *IDE,* 2 SNPs in *PLAU,* and 32 SNPs in *CTNNA3,* but none was significantly associated with LOAD (Busby et al. 2004; Nowotny et al. 2005).

The most consistently associated marker among the five significant SNPs is *rs498055,* which is significant in each of the three initially tested clinical case-control series employed here, with an allelic *P* value of .00004 in the meta-analysis of the three sample sets. The replication *P* value of .00021 is significant even after Bonferroni correction for 69 markers ($P = .014$), and the meta-analysis of these three case-control series used in the screening paradigm is marginally significant even after adjustment for 1,397 SNPs ($P = .051$). The linkage sample–derived case-control series replicates these results, whereas the smaller combined neuropathologically confirmed case-control sample set is not significant. The meta-

analysis of all six sample sets maintains that *rs498055* is significantly associated with AD risk ($P = .0001$).

The tree-scan analysis identified a single branch in the network that is significantly associated with LOAD. This branch is marked by mutations at *rs498055* and *rs495998*. Marker *rs498055* is the most significant SNP in the single-marker association tests (see table 1), and *rs495998* is in high LD with *rs498055* ($r^2 = 0.98$) (table 4). This suggests that the observed effect is a mutation on the background shared and defined by these SNPs. It is also interesting to note that *rs498055* is homoplasious, with mutations inferred on four different haplotypic backgrounds (one major and three minor haplotypes). In some cases, the haplotype structure of a population allows for tests to be conducted at each branch that is marked by a particular SNP, which provides some evidence as to the "causal" nature of the polymorphism. Although no association was detected at the other transitions marked by *rs498055* (a result that suggests that the SNP is not causal), the sample sizes for these tests are too small to provide strong evidence regarding the causality of this SNP. Inclusion of all the associated SNPs in this region in a logistic regression analysis by use of sequential regression (type 1) indicates that the significance derives only from LD with *rs498055*; that is, no other significant association is observed after first including the effect of *rs498055*.

Marker *rs498055* is located in a gene annotated as an *RPS3A* homologue in the Entrez Gene database. Although the function of the *RPS3A* homologue is unknown, it appears that *RPS3A* itself is a strong biological candidate gene for AD. It has been reported that RPS3A mediates the interaction between BCL2 (encoded by *BCL2* [MIM 151430]) and PARP—poly(ADP-ribose) polymerase—(*PARP1* [MIM 173870]) and that BCL2 and RPS3A together prevent apoptosis by inhibiting PARP activity (Hu et al. 2000; Song et al. 2002). Thus, RPS3A is an important player in the early phase of apoptosis, a feature observed in AD-affected brains. However, we have been unable to detect transcripts of the *RPS3A* gene by RT-PCR in RNA from multiple tissues, including brain (data not shown). This may be due to constraints in transcript-specific primer design if a gene has multiple paralogues, as is the case with *RPS3A*. Alternatively, the annotated gene may not be expressed, and this SNP or variants that are in LD are located in a noncoding expressed sequence, such as a microRNA. It is also possible that this SNP, or variants that are in LD, modulate the transcription of neighboring genes. The *SORBS1* (MIM 605264) coding sequence is located 33.7 kb downstream from this SNP and can be considered a strong biological candidate gene. It is involved in insulin signaling and was recently reported to be up-regulated in the hippocampus of AD-affected brains compared with controls (Blalock et al. 2004). *ALDH18A1* (at 91.1 Mb and in tight LD with SNPs in *RPS3A*) encodes a member of the aldehyde dehydrogenase family, which is involved in proline biosynthesis via catalyzing the conversion of L-glutamate to L-glutamate 5-phosphate.

On the basis of the results in the combined validation sample sets, three other markers of interest were also identified, but they are not significant in all three individual samples. The power to replicate the original observation in the exploratory sample for these markers is relatively low in each of the validation samples (table 1). These markers are located in four different genes. *PCGF5* (at 86.7 Mb) encodes polycomb group (PcG) ring finger 5, a component of a multimeric, chromatin-associated PcG protein complex, which is involved in stable repression of gene activity. *SORCS1* (MIM 606283) (at 102.7 Mb) encodes a type 1 receptor containing a Vps10p-domain and a leucine-rich domain that is involved in endocytosis and intracellular sorting. It is most abundantly expressed in the brain (Hermey et al. 1999), and its expression can be differentially affected by neuronal activity (Hermey et al. 2004). *hCG2039140* (at 102.9 Mb) is a predicted gene in the Celera Genome Assembly, encoding a 41-aa polypeptide with no apparent homology to any other known proteins. The potential relevance of these genes with LOAD remains to be examined. Moreover, it is possible that neighboring genes might have a role in AD, since the significant SNPs we identified or variants that are in LD may affect their function.

Although the association with *rs498055* was replicated in the case-control series from the linkage sample, the pedigree analyses suggest that this association did not significantly contribute to the original linkage signal on chromosome 10. Although the power of this analysis is low, it suggests that there may be more than one AD susceptibility gene on chromosome 10.

In our screen, we did not attempt to systematically genotype chromosome 10; rather, we used an opportunistic approach to identify functionally relevant gene-based variants that show significant association with AD in at least two independently collected case-control sample sets. Therefore, we cannot exclude the majority of nonsignificant chromosome 10 genes from those that might contribute to the genetic risk of AD. This would require high-density SNP genotyping incorporating an LD-based approach to SNP selection in the case of the common disease–common variant hypothesis and, ultimately, deep resequencing of all genes, to exclude rare pathogenic variants. However, the results outlined above highlight five SNPs—particularly *rs498055,* which was replicated in four independent case-control series—and corresponding genes as likely AD risk factors on chromosome 10. These findings require functional experiments to validate potential links of the genes and genetic variation to pathways related to disease mechanisms for AD.

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Web Resources

The URLs for data presented herein are as follows:

Alzheimer Disease Forum, http://www.alzforum.org/res/com/gen/alzgene/chromo.asp?c=10

Entrez Gene, http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?db=gene

Online Mendelian Inheritance in Man (OMIM), http://www.ncbi.nlm.nih.gov/Omim/ (for AD, *APP, PSEN1, PSEN2,* LOAD, *APOE, GAPD, RPS3A, PYCS, ALDH18A1, ENTPD1, CTNNA3, PLAU, IDE, BCL2, PARP1, SORBS1,* and *SORCS1*)

References

Agresti A (1990) Categorical data analysis. John Wiley & Sons, New York

Balciuniene J, Emilsson L, Oreland L, Pettersson U, Jazin E (2002) Investigation of the functional effect of monoamine oxidase polymorphisms in human brain. Hum Genet 110:1–7

Berg L, McKeel DW Jr, Miller JP, Storandt M, Rubin EH, Morris JC, Baty J, Coats M, Norton J, Goate AM, Price JL, Gearing M, Mirra SS, Saunders AM (1998) Clinicopathologic studies in cognitively healthy aging and Alzheimer's disease: relation of histologic markers to dementia severity, age, sex, and apolipoprotein E genotype. Arch Neurol 55:326–335

Bertram L, Blacker D, Mullin K, Keeney D, Jones J, Basu S, Yhu S, McInnis MG, Go RC, Vekrellis K, Selkoe DJ, Saunders AJ, Tanzi RE (2000) Evidence for genetic linkage of Alzheimer's disease to chromosome 10q. Science 290:2302–2303

Blacker D, Bertram L, Saunders AJ, Moscarillo TJ, Albert MS, Wiener H, Perry RT, Collins JS, Harrell LE, Go RC, Mahoney A, Beaty T, Fallin MD, Avramopoulos D, Chase GA, Folstein MF, McInnis MG, Bassett SS, Doheny KJ, Pugh EW, Tanzi RE (2003) Results of a high-resolution genome screen of 437 Alzheimer's disease families. Hum Mol Genet 12:23–32

Blalock EM, Geddes JW, Chen KC, Porter NM, Markesbery WR, Landfield PW (2004) Incipient Alzheimer's disease: microarray correlation analyses reveal major transcriptional and tumor suppressor responses. Proc Natl Acad Sci USA 101:2173–2178

Busby V, Goossens S, Nowotny P, Hamilton G, Smemo S, Harold D, Turic D, et al (2004) α-T-catenin is expressed in human brain and interacts with the Wnt signaling pathway but is not responsible for linkage to chromosome 10 in Alzheimer's disease. Neuromolecular Med 5:133–146

Clement M, Posada D, Crandall KA (2000) TCS: a computer program to estimate gene genealogies. Mol Ecol 9:1657–1659

Corder EH, Saunders AM, Strittmatter WJ, Schmechel DE, Gaskell PC, Small GW, Roses AD, Haines JL, Pericak-Vance MA (1993) Gene dose of apolipoprotein E type 4 allele and the risk of Alzheimer's disease in late onset families. Science 261:921–923

Ertekin-Taner N, Graff-Radford N, Younkin LH, Eckman C, Baker M, Adamson J, Ronald J, Blangero J, Hutton M, Younkin SG (2000) Linkage of plasma Aβ42 to a quantitative locus on chromosome 10 in late-onset Alzheimer's disease pedigrees. Science 290:2303–2304

Farrer LA, Bowirrat A, Friedland RP, Waraska K, Korczyn AD, Baldwin CT (2003) Identification of multiple loci for Alzheimer disease in a consanguineous Israeli-Arab community. Hum Mol Genet 12:415–422

Germer S, Holland MJ, Higuchi R (2000) High-throughput SNP allele-frequency determination in pooled DNA samples by kinetic PCR. Genome Res 10:258–266

Goate A, Chartier-Harlin M-C, Mullan M, Brown J, Crawford F, Fidani L, Giuffra L, Haynes A, Irving N, James L, Mant R, Newton P, Rooke K, Roques P, Talbot C, Pericak-Vance M, Roses A, Williamson R, Rossor M, Owen M, Hardy J (1991) Segregation of a

missense mutation in the amyloid precursor protein gene with familial Alzheimer's disease. Nature 349:704–706

Hermey G, Plath N, Hubner CA, Kuhl D, Schaller HC, Hermans-Borgmeyer I (2004) The three Sorcs genes are differentially expressed and regulated by synaptic activity. J Neurochem 88:1470–1476

Hermey G, Riedel IB, Hampe W, Schaller HC, Hermans-Borgmeyer I (1999) Identification and characterization of SorCS, a third member of a novel receptor family. Biochem Biophys Res Commun 266:347–351

Hu ZB, Minden MD, McCulloch EA, Stahl J (2000) Regulation of drug sensitivity by ribosomal protein S3a. Blood 95:1047–1055

Kehoe P, Wavrant-De Vrieze F, Crook R, Wu WS, Holmans P, Fenton I, Spurlock G, Norton N, Williams H, Williams N, Lovestone S, Perez-Tur J, Hutton M, Chartier-Harlin MC, Shears S, Roehl K, Booth J, Van Voorst W, Ramic D, Williams J, Goate A, Hardy J, Owen MJ (1999) A full genome scan for late onset Alzheimer's disease. Hum Mol Genet 8:237–245

Knoblauch H, Bauerfeind A, Krahenbuhl C, Daury A, Rohde K, Bejanin S, Essioux L, Schuster H, Luft FC, Reich JG (2002) Common haplotypes in five genes influence genetic variance of LDL and HDL cholesterol in the general population. Hum Mol Genet 11:1477–1485

Levy-Lahad E, Wasco W, Poorkaj P, Romano DM, Oshima J, Pettingell WH, Yu CE, Jondro PD, Schmidt SD, Wang K, Crowley A, Fu Y-H, Guenette S, Galas D, Nemens E, Wijsman E, Bird T, Schellenberg G, Tanzi R (1995) Candidate gene for the chromosome 1 familial Alzheimer's disease locus. Science 269:973–977

Li Y, Hollingworth P, Moore P, Foy C, Archer N, Powell J, Nowotny P, Holmans P, O'Donovan M, Tacey K, Doil L, van Luchene R, Garcia V, Rowland C, Lau K, Cantanese J, Sninsky J, Hardy J, Thal L, Morris JC, Goate A, Lovestone S, Owen M, Williams J, Grupe A (2005) Genetic association of the APP binding protein 2 gene (APBB2) with late onset Alzheimer disease. Hum Mutat 25:270–277

Li Y, Nowotny P, Holmans P, Smemo S, Kauwe JS, Hinrichs AL, Tacey K, et al (2004) Association of late-onset Alzheimer's disease with genetic variation in multiple members of the GAPD gene family. Proc Natl Acad Sci USA 101:15688–15693

McKhann G, Drachman D, Folstein M, Katzman R, Price D, Stadlan EM (1984) Clinical diagnosis of Alzheimer's disease: report of the NINCDS-ADRDA Work Group under the auspices of Department of Health and Human Services Task Force on Alzheimer's Disease. Neurology 34:939–944

Myers A, Holmans P, Marshall H, Kwon J, Meyer D, Ramic D, Shears S, Booth J, DeVrieze FW, Crook R, Hamshere M, Abraham R, Tunstall N, Rice F, Carty S, Lillystone S, Kehoe P, Rudrasingham V, Jones L, Lovestone S, Perez-Tur J, Williams J, Owen MJ, Hardy J, Goate AM (2000) Susceptibility locus for Alzheimer's disease on chromosome 10. Science 290:2304–2305

Myers A, Wavrant De-Vrieze F, Holmans P, Hamshere M, Crook R, Compton D, Marshall H, et al (2002) Full genome screen for Alzheimer disease: stage II analysis. Am J Med Genet 114:235–244

Myers AJ, Marshall H, Holmans P, Compton D, Crook RJ, Mander AP, Nowotny P, Smemo S, Dunstan M, Jehu L, Wang JC, Hamshere M, Morris JC, Norton J, Chakraventy S, Tunstall N, Lovestone S, Petersen R, O'Donovan M, Jones L, Williams J, Owen MJ, Hardy J, Goate A (2004) Variation in the urokinase-plasminogen activator gene does not explain the chromosome 10 linkage signal for late onset AD. Am J Med Genet B Neuropsychiatr Genet 124:29–37

Nowotny P, Hinrichs AL, Smemo S, Kauwe J, Maxwell T, Holmans P, Hamshere M, et al (2005) Association studies between risk for late-onset Alzheimer's disease and variants in insulin degrading enzyme. Am J Med Genet B Neuropsychiatr Genet 136:62–68

Pastor P, Goate AM (2004) Molecular genetics of Alzheimer's disease. Curr Psychiatry Rep 6:125–133

Pericak-Vance MA, Bass MP, Yamaoka LH, Gaskell PC, Scott WK, Terwedow HA, Menold MM, Conneally PM, Small GW, Vance JM, Saunders AM, Roses AD, Haines JL (1997) Complete genomic screen in late-onset familial Alzheimer disease: evidence for a new locus on chromosome 12. JAMA 278:1237–1241

Pericak-Vance MA, Grubber J, Bailey LR, Hedges D, West S, Santoro L, Kemmerer B, Hall JL, Saunders AM, Roses AD, Small GW, Scott WK, Conneally PM, Vance JM, Haines JL (2000) Identification of novel genes in late-onset Alzheimer's disease. Exp Gerontol 35: 1343–1352

Rogaeva E, Premkumar S, Song Y, Sorbi S, Brindle N, Paterson A, Duara R, Levesque G, Yu G, Nishimura M, Ikeda M, O'Toole C, Kawarai T, Jorge R, Vilarino D, Bruni AC, Farrer LA, St George-Hyslop PH (1998) Evidence for an Alzheimer disease susceptibility locus on chromosome 12 and for further locus heterogeneity. JAMA 280:614–618

Sherrington R, Rogaev EI, Liang Y, Rogaeva EA, Levesque G, Ikeda M, Chi H, et al (1995) Cloning of a gene bearing missense mutations in early-onset familial Alzheimer's disease. Nature 375: 754–760

Song D, Sakamoto S, Taniguchi T (2002) Inhibition of poly(ADP-ribose) polymerase activity by Bcl-2 in association with the ribosomal protein S3a. Biochemistry 41:929–934

Stephens M, Donnelly P (2003) A comparison of Bayesian methods for haplotype reconstruction from population genotype data. Am J Hum Genet 73:1162–1169

Stephens M, Smith NJ, Donnelly P (2001) A new statistical method for haplotype reconstruction from population data. Am J Hum Genet 68:978–989

Templeton AR, Maxwell T, Posada D, Stengard JH, Boerwinkle E, Sing CF (2005) Tree scanning: a method for using haplotype trees in phenotype/genotype association studies. Genetics 169:441–453

Templeton AR, Weiss KM, Nickerson DA, Boerwinkle E, Sing CF (2000) Cladistic structure within the human lipoprotein lipase gene and its implications for phenotypic association studies. Genetics 156: 1259–1275

Van Eerdewegh P, Little RD, Dupuis J, Del Mastro RG, Falls K, Simon J, Torrey D, et al (2002) Association of the *ADAM33* gene with asthma and bronchial hyperresponsiveness. Nature 418:426–430

Westfall P, Young S (1993) Resampling-based multiple testing. John Wiley & Sons, New York