HHS Public Access

Author manuscript

Mol Psychiatry. Author manuscript; available in PMC 2018 March 23.

Published in final edited form as:

Mol Psychiatry. 2018 April ; 23(4): 963–972. doi:10.1038/mp.2017.81.

Genetic Risk for Schizophrenia and Psychosis in Alzheimer Disease

Mary Ann A. DeMichele-Sweet, Ph.D.[a], **Elise A. Weamer, M.P.H.**[b], **Lambertus Klei, Ph.D.**[a], **Dylan T. Vrana**[g], **Deborah J. Hollingshead, M.S.**[d], **Howard J. Seltman, M.D. Ph.D.**[h], **Rebecca Sims, Ph.D.**[k], **Tatiana Foroud, Ph.D.**[l], **Isabel Hernandez, M.D., Ph.D.**[i], **Sonia Moreno-Grau**[i], **Lluís Tárraga**[i], **Mercè Boada, M.D., Ph.D.**[i], **Agustin Ruiz, Ph.D.**[i], **Julie Williams, Ph.D.**[k], **Richard Mayeux, MD**[e], **Oscar L. Lopez, M.D.**[a,b], **Etienne L. Sibille, Ph.D.**[a,j], **M. Ilyas Kamboh, Ph.D.**[c], **Bernie Devlin, Ph.D.**[a], and **Robert A. Sweet, M.D.**[a,b,f]

[a]Department of Psychiatry, University of Pittsburgh, Pittsburgh, PA

[b]Department of Neurology, University of Pittsburgh, Pittsburgh, PA

[c]Department of Human Genetics, University of Pittsburgh, Pittsburgh, PA

[d]Genomics Research Core of the Health Sciences Core Research Facilities, University of Pittsburgh, Pittsburgh, PA

[e]Departments of Neurology, Psychiatry and Epidemiology, Columbia University, New York, NY

[f]VISN 4 Mental Illness Research, Education and Clinical Center (MIRECC) VA Pittsburgh Healthcare System, Pittsburgh, PA

[g]Department of Computational Biology, Carnegie Mellon University, Pittsburgh, PA

[h]Department of Statistics, Carnegie Mellon University, Pittsburgh, PA

[i]Research Center and Memory Clinic of Fundació ACE, Institut Català de Neurociències Aplicades, Barcelona, Spain

[J]Departments of Psychiatry and of Pharmacology and Toxicology, University of Toronto, Toronto, ON, Canada; Campbell Family Mental Health Research Institute of CAMH, Toronto, ON, Canada

[k]Division of Psychological Medicine and Clinical Neuroscience, School of Medicine, Cardiff University, Cardiff, UK

[l]Medical and Molecular Genetics, Indiana University School of Medicine, Indianapolis, IN, USA

Abstract

For questions and correspondence please contact: Robert A. Sweet, M.D., Mail: Biomedical Science Tower, Rm W-1645, 3811 O'Hara Street, Pittsburgh, PA 15213-2593. Express Mail: Biomedical Science Tower, Rm W-1645, Lothrop and Terrace Streets, Pittsburgh, PA 15213-2593. Phone: 412-624-0064, Fax: 412-624-9910, sweetra@upmc.edu, Web: http://www.wpic.pitt.edu/research/sweetlab/.

Conflict of Interest: MAAD-S, EAW, LK, DJH, DTV, DJH, HJS, RS, TF, IH, SM-G, LT, MB, AR, JW, RM, OLL, ELS, MIK, BD, and RAS have no conflicts to report.

Psychotic symptoms, defined as the occurrence of delusions or hallucinations, are frequent in Alzheimer Disease, affecting ~ 40% to 60% of individuals with AD (AD with psychosis, AD+P). In comparison to AD subjects without psychosis, AD+P subjects have more rapid cognitive decline and poor outcomes. Prior studies have estimated the heritability of psychosis in AD at 61%, but the underlying genetic sources of this risk are not known. We evaluated a Discovery Cohort of 2876 AD subjects with (N=1761) or without psychosis (N=1115). All subjects were genotyped using a custom genotyping array designed to evaluate SNPs with evidence of genetic association with AD+P and include SNPs affecting or putatively affecting risk for schizophrenia and Alzheimer disease. Results were replicated in an independent cohort of 2194 AD subjects with (N=734) or without psychosis (N=1460). We found that AD+P is associated with polygenic risk for a set of novel loci and inversely associated with polygenic risk for schizophrenia. Among the biologic pathways identified by the associations of schizophrenia SNPs with AD+P are endosomal trafficking, autophagy, and calcium channel signaling. These findings provide the first clear demonstration that AD+P is associated with common genetic variation. In addition, they provide an unbiased link between polygenic risk for schizophrenia and a lower risk of psychosis in AD. This provides an opportunity to leverage progress made in identifying the biologic effects of schizophrenia alleles to identify novel mechanisms protecting against more rapid cognitive decline and psychosis risk in AD.

Introduction

Psychotic symptoms, defined as the occurrence of delusions or hallucinations, are frequent in Alzheimer Disease (AD+Psychosis, AD+P), affecting ~ 40% to 60% of individuals with AD. In comparison to AD subjects without psychosis, AD+P subjects have more rapid cognitive decline and poor outcomes. Ropacki and Jeste[1] comprehensively reviewed the literature on psychosis in AD from 1990 to 2003, identifying 55 studies comprised of 9,749 subjects. More rapid cognitive decline was the most consistent correlate of AD+P compared to AD without psychosis (AD-P). More recent studies have continued to support the relationship between greater cognitive impairment, more rapid cognitive decline, and AD+P.[2–8] AD+P is further associated with additional psychiatric and behavioral disturbances, the most frequent and troublesome of which are agitation[9] and aggression[10;11]. AD+P leads to greater distress for family and caregivers[12], greater functional impairment,[13] higher institutionalization rates,[14–17] worse health[18] and increased mortality[19] compared to AD-P patients.

Treatment of psychosis in AD patients has been suboptimal due to the limited efficacy of available drugs and their high toxicity in this age group. First line treatments are atypical antipsychotics, which have efficacy similar to conventional antipsychotics for AD+P, with lower rates of motor side effects.[20] However, atypical and conventional antipsychotics have been associated with an increased risk of all-cause mortality after even short-term treatment.[20;21] Other treatments, such as selective serotonin reuptake inhibitors, may have some efficacy[22;23] and improved tolerability.[24] Nevertheless, none of these treatments was derived to prevent or reverse an identified biology of AD+P, and there is no current data to suggest that any of these treatments effectively mitigate against the greater cognitive and functional decline associated with AD+P. It is thus imperative to develop an approach to promote

discovery regarding the biology of AD+P and identify opportunities to intervene to prevent its adverse trajectory.

We initially observed familial aggregation of AD+P,[25] since replicated in two independent cohorts.[4;26] These studies show a remarkable consistency in the estimated 3–4 fold increased odds of psychosis in a family member with AD, given the presence of psychosis in a proband with AD. Similarly, we used two of these cohorts to estimate the heritability of psychosis in AD as 61%.[27;28] Thus, AD+P is likely to be strongly influenced by genetic variation. In keeping with these observations, we recently reported the first Genome-Wide Association Study (GWAS) of AD+P, evaluating 1,299 cases with AD+P and 735 individuals characterized as AD-P. Although no single SNP demonstrated genome-wide significance, likely due to modest sample size, there was suggestive evidence for association with novel loci.

We further found a trend towards association with a group of 11 SNPs that had been identified in initial GWAS studies of schizophrenia and bipolar disorder.[29] That latter finding also provided the biologically intriguing observation that the direction of 7/11 allelic effects on risk for AD+P were opposite that reported in the studies of psychiatric disorder subjects. Since the time of our prior report, genomic studies of schizophrenia risk have identified 128 SNPs in 108 loci that exceed genome-wide significance.[30]

Recently, the use of polygenic risk scores has emerged as an important approach for summarizing genetic effects of a set of SNPs. A polygenic score is a simple, subject-specific summary of the additive effects of alleles on a trait. When computed to predict subjects' risk for a disorder, it is called a polygenic risk score. The score can be obtained from a limited set of SNPs, such as those reaching genome wide significance in association studies, or a larger set based on some other threshold [313233], or the entire genome [34;35]. For example, when alleles at the 108 schizophrenia-associated loci were combined in a polygenic risk score they explained 3.4% of the liability to schizophrenia.[30] For traits in which few or no individual SNPs reach genome wide significance, polygenic risk scores can provide initial evidence for true genetic association of the trait with the SNPs either included within the score or in close linkage disequilibrium [33], providing critical evidence in support of larger scale studies needed to identify the individual affected loci.

Here we follow up on our prior research in an expanded Discovery Cohort of 2876 AD subjects with and without psychosis. All subjects were genotyped using a custom chip designed to evaluate SNPs with evidence of genetic association, most prominently with AD +P, although SNPs affecting or putatively affecting risk for schizophrenia and Alzheimer disease were also assessed. Results were replicated in an independent cohort of 2194 AD subjects with and without psychosis. We found that AD+P is associated with polygenic risk for a set of novel loci and inversely associated with polygenic risk for schizophrenia. These findings provide the first clear demonstration that AD+P is associated with common genetic variation. In addition, they provide an unbiased link between polygenic risk for schizophrenia and a lower risk of psychosis in AD. As efforts to identify the biologic effects of schizophrenia alleles progress, it may be possible to leverage these results to identify novel mechanisms protecting against more rapid cognitive decline and psychosis risk in AD.

Materials and Methods

An overview of the study design and workflow is shown in Figure 1.

Subjects

This study analyzed samples obtained from subjects in two cohorts, an initial Discovery Cohort and an independent Replication Cohort (Table 1). All subjects were diagnosed with possible, probable,[36] or definite[37] AD. Importantly, subjects with a primary diagnosis of Dementia with Lewy bodies[38] were excluded. The above diagnoses resulted from diagnostic evaluations, cognitive testing, and in some cases neuropathologic assessment, conducted during subjects' participation in the following programs as previously described: the University of Pittsburgh Alzheimer Disease Research Center (ADRC),[39;40] the Genetic and Environmental Risk in AD Consortium 1 (UK),[29;41;42] the National Institute on Aging's Late Onset Alzheimer's Disease Family Study (NIA-LOAD),[4;28] the National Institute of Mental Health Genetics Initiative AD Cohort (NIMH),[25] the Fundació ACE Barcelona Alzheimer Treatment and Research Center (ACE),[41;43], the Cardiovascular Health Study (CHS),[3;41] and a consortium of National Institute on Aging Alzheimer Disease Centers (ADC).[44] Collection of clinical data and genetic samples were approved by each sites local Institutional Review Board or Medical Ethics Committee, as appropriate. Additional detail of the individual cohorts and assessment methodology is available in Supplementary Methods and Tables S1–S13.

Characterization of Psychosis

Subjects were characterized for the presence or absence of delusions and hallucinations within the individual studies using the CERAD behavioral rating scale[45] (ADRC and NIA-LOAD), Neuropsychiatric Inventory Questionnaire (NPI-Q,[46] NIA-LOAD, ADC), NPI-Q Spanish Version[47] (ACE), NPI[48] (UK, CHS), and Brief Psychiatric Rating Scale[49] (NIMH). Each of these instruments has established reliability in AD,[4;50] and we have previously used all successfully in analyses of psychosis in AD subjects.[3;4;6;27;39] Details of the application of these assessments for each cohort are provided in the Supplementary Methods. AD+P was defined by the presence of persistent hallucinations or delusions occurring during the course of the dementia, AD-P was defined by the absence of all symptoms at all assessments. Because psychotic symptoms typically emerge in the mild to moderate stages of AD[4] individuals without psychosis but who were still in the early stages of disease at their last assessment (Clinical Dementia Rating[51] score <1, mini-mental state examination score[52] >20) were considered to be at substantial risk of developing AD+P later in their course. Thus, these individuals were excluded from the analysis. We have previously used these approaches to characterizing and defining AD+P and AD-P to demonstrate familial aggregation,[4;25] heritability,[27;28] genetic linkage,[28;53] and suggestive genome-wide association[29] with the AD+P phenotype.

Genotyping

DNA Preparation—Samples from outside sources were shipped on dry ice, stored, and processed by the Genomic Core Lab at the University of Pittsburgh. ACE samples were supplied as whole blood and genomic DNA was extracted using the Qiamp Blood Mini kit

(Qiagen, Valencia, CA). All other centers provided genomic DNA (ADRC, NIA-LOAD, NIMH, UK, ADC) or whole genome amplified DNA (CHS).

Custom Chip for Discovery Cohort—The Genomic Core Lab quantitated all samples by Pico Green (Thermo Fisher, Pittsburgh, PA) and diluted the DNA to 23ng/ul and shipped the plates on dry ice to Affymetrix (Los Angeles, CA) for genotyping. Plates also contained randomized duplicates. Affymetrix confirmed all DNA concentrations by Pico Green assay prior to genotyping. Genotyping used a custom designed Axiom® chip (see SNP selection below), and was performed using the Affymetrix GeneTitan® system as described in the axiom user manual[54] with resultant genotype calls provided for QC and analysis.

iPlex Assay for Genotyping SCZ risk score SNPs and Replication Cohort Testing

iPlex Chemistry: Assays were designed with Assay Designer 4.0 (Agena) and analysis performed using iPlex Gold Genotyping Reagent Set (Agena, San Diego, CA) according to manufacturer's instructions. Target loci were amplified within the samples by multiplex PCR in 1X PCR buffer containing 3.5 mM $MgCl_2$, 25 mM dNTPs, 500 nM each of forward and reverse amplification primer within the multiplex pool and 2.5 U HotStar Taq. dNTPs and primers were removed by incubation with 0.5 U shrimp alkaline phosphotase (SAP) at 37 °C for 40 minutes. SAP was inactivated by incubation at 87 °C for 5 minutes. Single base extension was carried out in 0.2X iPLEX buffer plus, 1X termination mix (containing mass modified termination nucleotides), 1X iPLEX enzyme and primers at 0.84 μM, 1.04 μM and 1.25 μM as appropriate to the relative mass of each primer. Following thermocycling, clean resin and water was added to the MassExtend reaction products. Samples were incubated in clean resin at room temperature with mixing for 5 minutes and centrifuged at 3200 × g for 5 minutes.

Samples were then dispensed to a SpectraChip using the MassArray Nanodispenser according to manufacturer's instructions. Spectra chips were loaded into the MassArray analyzer and spectra acquired for each sample. Genotype calls were made using Typer 4.0 (Agena) by mass identification of extended primer peaks.

SNP Selection

Development of Custom Array for Discovery Cohort—The process of selecting SNPs for the genotyping array involved two principal stages. First SNPs were amalgamated based on genetic signal for association to a small set of phenotypes (Table S14). The bulk of the SNPs were included on the basis of association results from four contrasts reported in three genome-wide studies: a contrast of AD+P versus AD-P,[29] AD+P versus controls,[29] AD versus controls [55;56], and SCZ versus controls [32;57]. An additional unpublished data set (described in[58;59]) of cis-eQTLs affecting gene expression and cis-eQTLs associated with age-related changes in gene expression was also used. For the first four genome-wide association studies (GWAS), SNPs with p-value less than a threshold of 0.01 were selected; for the eQTLs, the threshold was 0.001 and for the 'aging' eQTLs it was 0.05. Note that when a SNP was represented in more than one study, the minimum p-value in any of the 6 datasets was taken as representative for the SNP. To interrogate copy number regions shown

to be associated with schizophrenia, 1574 SNPs were included (1q21.1, 3q29, 15q11.2–15q13.3, 16p13.1, 16p11.2 and 22q11.2, recently reviewed in [60]; and 7q11.23[61]). Finally a small fraction of SNPs were chosen to cover four genes of interest regarding psychotic disorders (SCZ target genes: *NRXN1*,[60] *ERBB4*,[62] *PAK2*,[63] *CHRNA7*[64]) or were nominated from unpublished AD studies (UK SNPs).

Second, SNPs were retained for genotyping by a winnowing process. This process involved removing redundant SNPs, those that could not be genotyped on the Axiom platform, or SNPs not present in 1000 genomes. Of the SNPs passing this step, all SNPs with a minimum p-value < 0.0001 for any study were retained. For the remainder, by using a LD clumping process, we removed SNPs in LD with the retained SNPs ($r^2 > 0.9$) and retained additional SNPs with the smallest p-value in "independent" clumps ($r^2 < 0.9$) by pruning SNPs with Plink (maximum distance for pruning was 5 kb, window width was 25 SNPs, sliding step was 5 SNPs).

SNP Selection for SCZ Risk Score Testing and Follow-up Genotyping in Replication Cohort—For SCZ risk score testing in each cohort we targeted the 128 GWA significant SNPs reported in [30], although not all could be genotyped. Follow-up genotyping in the Replication Cohort also selected SNPs from our custom array that passed Quality Control and with P < 0.0001 for the contrast of AD+P versus AD-P. For the replication cohort we selected Ancestry Informative Markers (AIMs) for European Ancestry based on the results in [65] Specifically, based on results found in with Supplementary Table 1 of Kosoy et al. [65], we selected their "Top 96" European AIMs, of which 82 could be genotyped on the Sequenom platform and 79 passed Quality Control.

Quality Control

QC was performed at the individual level first, then at the SNP level conditional on individual-level data passing QC and individuals of European ancestry. Details of QC are given in Supplementary Material. In brief, genetic data for samples were retained if their nominal sex agreed with genetically determined sex; heterozygosity rate, per subject, revealed no evidence of contamination by other samples; genetic data for subjects expected to be unrelated suggested this were true; and call rate of SNPs > 96.5% per sample. Next ancestry of subjects in the Discovery Cohort was determined using dacGem in GemTools based on 5712 autosomal markers with non-call rate ≤ 0.001, minor allele frequency (MAF) ≥ 0.05, and $r^2 \leq 0.20$. The samples were separated into 5 clusters based on 3 significant ancestry dimensions, four of which likely represent European ancestry and two of these contain the bulk of the subjects (≈ 66%). SNP QC was performed on data from these two clusters. SNPs passed QC if their call rate was > 95%, MAF was ≥ 0.01 and the exact Hardy Weinberg Equilibrium p-value was > 0.005. Ancestry of samples in the Replication Cohort was determined using GemTools based on 79 autosomal Ancestry Informative Markers. The samples separated into 3 clusters based on 2 significant ancestry dimensions (Figure S3).

Statistical Analysis

Association between diagnosis and minor allele count for each SNP was assessed using logistic regression. For the Discovery Cohort, the model also accounted for first 5 ancestry

dimensions whereas for the Replication Cohort it accounted for two. Because some subjects in the Discovery Cohort were related as siblings, inference relied on the generalized estimating equation (gee) approach implemented in the statistical software R, assuming full siblings were correlated at 0.5 (i.e., twice the kinship coefficient for full siblings).

To predict affection status using polygenic risk scores, either unweighted or weighted risk scores could be computed: the unweighted score for a subject is the sum of the count of risk alleles over all genotypes for that subject; a weighted score uses the same principle, but the count of risk alleles per SNP is adjusted by a function of the estimated effect of the SNP on risk (log odds ratio). Both unweighted and weighted scores for AD+P risk were calculated. Scores were derived from results from the Discovery Cohort and then used to predict AD+P status based on genotypes for each subject in the Replication Cohort. For the SCZ-risk score, only a subset of the 128 GWA significant SNPs could be genotyped. For genotyped SNPs, an unweighted score for each AD subject was estimated.

Results

Association of AD+P with novel common variants

Not all of the subjects genotyped on the Axiom array were independent of our previously reported GWAS meta-analysis.[29] Of the 2876 Discovery Cohort subjects described in Table 1a, 1157 of these subjects were in our prior GWAS and the remainder, 1799 subjects (969 AD+P and 750 AD-P), were independent. We, therefore, evaluated association both as a joint analysis of the sample sets (mega-analysis) and by analysis of the independent subjects. For the former we use the traditional threshold for GWAS significance, 5×10^{-8}; for the latter we used a somewhat more lenient threshold based sample sizes and the number of SNPs tested on both samples (5.6×10^{-8}).

For neither the joint analysis (Fig 2a) nor the independent analysis threshold (results not shown) was any SNP significantly associated with risk for AD+P. For the joint analysis there were 67 SNPs with $P < 5 \times 10^{-4}$ (Fig 2a). To test these SNPs we empaneled a Replication Cohort (Table 1b). We successfully designed and assayed either the SNP or a proxy in perfect LD for 60/67 target SNPs.

We next used the results from the Discovery Cohort to assign the “risk allele” at each of the 60 SNPs. Then, by counting the number of risk alleles carried by subjects in the Replication Cohort, we formed an unweighted risk score for each subject. This score significantly predicted AD+P status in the Replication Cohort, showing clear evidence for association (Table 2a). The same is true for a weighted score (Table 2a).

Moreover, although no single SNP was significantly associated with risk for AD+P within or across stages, and only three SNPs approached individual significance when combining stages (Fig 2b and Table 3), 41 out of the 60 SNPs had the same risk allele for both the Discovery and Replication Cohorts (Fig 2b; Fisher Exact Test, p = 0.0062). The 3 SNPs that approached significance are in *RP11-541P9.3* (an antisense transcript) located 5′ to Cyclin G1 (*CCNG1*).

Association of AD+P with polygenic variation associated with schizophrenia

We previously described a significant association between AD+P and a summary statistic from a small number of putative schizophrenia and bipolar disorder risk alleles. Curiously, however, the direction of risk for most alleles was opposite in AD+P.[29] Recently, 128 genome-wide significant SNPs at 108 independent loci have been identified in schizophrenia.[30] When these loci were combined into a polygenic risk score they explained ~3.4% of the variance in schizophrenia risk.[30] We successfully genotyped 101 of these SNPs in the Discovery Cohort. We found that the corresponding unweighted risk score was significantly associated with AD+P (Nagelkerke's pseudo R^2 = 0.32%, P = 0.006). We then genotyped the schizophrenia SNPs in the Replication Cohort. For this analysis, 94 SNPs remained after QC. Results clearly replicated, with close agreement between the two cohorts (Table 2b).

Of note, as in our earlier report,[29] increasing schizophrenia polygenic risk score was associated with reduced risk of psychosis in AD. Consider, for example, its relationship within the Discovery Cohort. To better illustrate this relationship, we calculated an AD+P aligned risk score. For each of the SNPs comprising the schizophrenia polygenic risk score, the allele that increased the risk of developing AD+P was determined and a weighted sum of risks was then computed for each of the samples. The correlation between the AD+P aligned risk scores and the schizophrenia risk scores was −0.159 (p = 5.5e-18, Fig 3).

It should be noted that despite the overall protective effects of schizophrenia polygenic risk score on AD+P risk, a smaller number of individual schizophrenia risk SNPs were associated with increased risk of AD+P. Table 4a details the 20 SNPs that most consistently (as defined by the minimum of the sum of their individual SNP regression coefficients from the analysis of the Discovery and Replication cohorts) were associated with reduced AD+P risk. Table 4b provides similar information for the 10 SNPs that most consistently were associated with increased AD+P risk in the two cohorts.

Discussion

Psychosis occurs in approximately half of individuals affected by AD, serving to identify a subgroup with more rapid decline and poor outcomes. We, and others, have hypothesized that common genetic variation may contribute to the risk of psychosis in AD, based in part on evidence that AD+P aggregates in families, with an estimated heritability of 61%. However, prior studies of the association of common genetic variation with AD+P have been inconclusive.[66;67] We now provide the first clear evidence in support of an association of AD +P with both a unique set of common variants, and with a set of common variants associated with risk for schizophrenia.

Several potential methodologic issues in determining the psychosis phenotype are important to consider in evaluating our findings. First, the need to aggregate multiple cohorts so as to have sufficient power for detection of association with common genetic variation meant that we included sites in which different rating scales were used for ascertainment of psychosis, which could have contributed noise to our phenotypic classifications. Such a limitation, if present, would have reduced our power to detect differences between groups. Thus, it speaks

to the robustness of our findings that despite this potential limitation, we were able to replicate associations across two independent, somewhat heterogeneous cohorts. Perhaps this result is not surprising, in that significant familial aggregation of AD+P was previously identified in three separate family cohorts, each of which relied primarily on a different behavioral rating scale.[4;25;26] Second, we chose to consider delusions and hallucinations together as a psychosis syndrome rather than evaluate them individually. The best approach to this issue likely depends on the question being asked. For example, when evaluating functional neuroanatomy separation of these symptoms could make most sense. However, for genetic studies, there is substantial support for grouping these symptoms. Specifically, studies demonstrating familial aggregation of AD+P[4;25;26] have all used this joint definition, establishing it as suitable for genetic investigation. Similar data in support of individual psychotic symptoms does not exist. Finally, the relationships between clinical Dementia with Lewy Body diagnoses (DLB), Lewy Body neuropathology, and AD+P is complex.[67;68] In brief, DLB pathology may contribute to some proportion, but clearly not account for most, of the occurrence of AD+P. Nevertheless, all sites in the current study used standard diagnostic criteria to identify individuals with probable Lewy Body Dementia and exclude them from analysis (the one exception being the NIMH family study which predated the generation of DLB criteria, but did exclude individuals with parkinsonism or prominent early behavioral disturbance.[69] As practical evidence that these diagnostic approaches are sufficiently rigorous to identify AD separately from DLB, the sites included in the current study have successfully contributed to discovery of common variants for AD risk.[42]

We identified a set of SNPs with suggestive association with AD+P in our Discovery Cohort, confirming this polygenic association with AD+P in an independent Replication Cohort. Although no individual SNP reached genome-wide significance in the meta-analysis, the strongest associations were seen with three SNPs, rs300215, rs6859958 and rs999581, within a single locus. The function of the antisense transcript, *RP11-541P9.3*, is not known. However, it is located 5′ to *CCNG1* and is therefore likely to regulate *CCNG1* expression. In support of this interpretation, rs6859958 and rs999581 have been shown to be eQTLs for *CCNG1* in some tissues.[70] Most cyclins activate cyclin dependent kinases, including *CDK5*, a Tau kinase that promotes phospho-Tau aggregation.[71] In contrast, *CCNG1* has been proposed to competitively inhibit the activation of cyclin dependent kinases by other cyclins.[72] Whether increased levels of Cyclin G1, the protein product of *CCNG1*, is therefore protective against pathological phosphorylation of Tau by CDK5 is not established. Nonetheless, in neocortex of AD subjects Cyclin G1 levels are increased in pyramidal neurons lacking Tau aggregates and are undetectable in those pyramidal neurons containing aggregated phospho-Tau.[73] Because the strongest brain correlate of psychosis in AD is excess phosphorylation of tau (in comparison to the degree of Tau phosphorylation in AD subjects without psychosis),[67] *CCNG1* is thus also a strong functional candidate for AD+P risk.

We also identified and confirmed an association of polygenic risk for schizophrenia with a reduced risk of AD+P. At present, there is little convergent data from family studies to inform on the relationship of schizophrenia risk to AD+P.[9;74] The inverse nature of the association between schizophrenia risk score and AD+P may seem counterintuitive at first. In fact, it was the counterintuitive nature of our findings that motivated us to attempt to

independently replicate them, finding a nearly identical association in a second large cohort. In contrast to our findings, schizophrenia has been shown to share polygenic risk with a number of complex disorders, such as autism and bipolar illness, which may include psychotic symptoms as part of the expressed phenotype.[75] Unlike AD+P, these are disorders of early, or late, neurodevelopment and thus do not occur in the context of neurodegeneration.

Possible genetic mechanisms underlying how the 108 schizophrenia-associated loci confer an increased risk of schizophrenia are just now emerging.[76–78] How these loci may lead to reduced AD+P risk cannot be asserted, but a few exemplars are worth discussion. First, a locus may alter expression of a single gene that has effects during neurodevelopment which increase schizophrenia risk, but when the same altered expression occurs in a brain with an active AD neurodegenerative process, it is protective. For example, rs75968099 is an eQTL for *LRRFIP2* (Table 4), the gene encoding Leucine-rich Repeat Flightless-interacting Protein 2, a regulator of Toll-like receptor 4-mediated signaling in response to inflammatory stimuli. Toll-like receptor 4 signaling helps activate microglia to clear toxic amyloid β from the brain of an AD patient in early disease stages,[79] whereas microglial activation may contribute to excess synaptic elimination in development, increasing risk for schizophrenia.[76] Second, a locus may regulate the expression of gene transcription differently during early neurodevelopment than in the adult brain. Such an effect has recently been described for the schizophrenia risk locus defined by rs55833108, and may similarly be present at loci that confer opposing risks for schizophrenia and AD+P.[78] A third scenario might result from the observation that a SNP and/or locus may be an eQTL for more than one gene. For example, the locus on chromosome 17 defined by rs8082590 was recently reported to show consistent disease and eQTL associations for two genes, *TOM1L2* and *DRG2*,[77] encoding Target Of Myb1 Like 2 Membrane Trafficking Protein and Developmentally Regulated GTP Binding Protein 2, respectively. Developmentally Regulated GTP Binding Protein 2 deactivates the early endosome regulator, Ras-related protein Rab-5A.[80] Thus it is strongly positioned to impact glutamate neurotransmission, a process implicated in the pathogenesis of schizophrenia, via effects on neurotransmitter release[81] and on AMPA receptor internalization.[82] In contrast, Target Of Myb1 Like 2 Membrane Trafficking Protein is necessary for delivery of endosome cargo to autophagosomes, which target protein aggregates and damaged organelles to lysosomes for degradation.[83] The autophagy pathway is strongly implicated in the pathogenesis of AD[84] and, more recently, of schizophrenia,[85] and is also downstream of Toll-like receptor 4 signaling.[86] Finally, we note that the above examples are not comprehensive. Other mechanisms may also contribute to different impacts of loci on risk for schizophrenia and AD+P.

As indicated in Table 4b, we also identified SNPs that showed the same direction of effect for schizophrenia and AD+P risk, Notable among these were two intronic SNPs located in *CACNA1C*, the gene encoding the voltage-dependent L-type calcium channel subunit alpha-1C. Although the genetic mechanism underlying these associations remains an area of active inquiry,[87] convergent data suggests that schizophrenia is associated with reduced voltage-dependent calcium channel function.[88–90] How reductions in voltage-dependent calcium channel function may further increase AD+P risk is not known, however,

impairments of intracellular Ca^{2+} homeostasis are present in AD, and can contribute to synaptic dysfunction and cognitive impairments.[91]

We recently estimated the annual incidence of psychosis in AD at 10%.[40] Thus there is an opportunity to intervene prior to psychosis onset if individual predictors can be identified. Although currently no treatments are established for prevention of AD+P, selective serotonin reuptake inhibitors have some efficacy for treating it,[22;23] and they have acceptable tolerability. Non-pharmacologic treatments may also offer benefit for treating AD+P (reviewed in[92 and40]) and could be adapted for prevention. It is thus worth considering whether genetic variants that associate with psychosis may serve as biomarkers to predict AD+P risk and the associated more rapidly declining cognitive trajectory. Because individual SNP relative risks are typically small, polygenic risk scores have greater predictive power.[31;93] We observed a very modest explanatory power of both the 60 SNP and the schizophrenia polygenic risk scores, each accounting for less than 1% of the AD+P risk. None of these effects is large enough to yield meaningful clinical prediction at present. Still we note that these polygenic scores could have a different magnitude of effect on prediction of a related, clinically relevant construct, such as time to onset of psychosis. However, the development of predictive approaches would clearly benefit from the identification of additional risk loci. Nevertheless, the current findings are a step forward in the development of prevention for psychosis in AD.

In that regard it is noteworthy that our custom array, used to evaluate the Discovery Cohort, was derived, in part, from the one existing GWAS of AD+P. That earlier GWAS was underpowered and limited in the number of loci interrogated.[29] As a consequence it is likely that many SNPs and loci that contribute meaningfully to AD+P risk were not tested in the current study. Despite this limitation, the current study provides confirmation of the hypothesis that AD+P is associated with common genetic variation. As such, it provides strong support for unbiased genome-wide scans of larger cohorts of AD+P and AD-P subjects, which will surely identify individual AD+P risk loci and develop more strongly predictive polygenic risk scores.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

This study was supported by the following federal grants: AG027224 (RAS), AG005133 (RAS), BX000452 (RAS), MH057881 (BD), AG030653 (MIK), AG041718(MIK), and MH093723 (ELS). This project used the University of Pittsburgh HSCRF Genomics Research Core iPlex and Specimen Processing services. Cardiff University was supported by the Wellcome Trust, Medical Research Council (MRC) and/or Alzheimer's Research UK (ARUK) and the Welsh Assembly Government (RS & JW).

The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health, the Department of Veterans Affairs, or the United States Government.

Reference List

1. Ropacki SA, Jeste DV. Epidemiology of and risk factors for psychosis of Alzheimer's disease: a review of 55 studies published from 1990 to 2003. Am J Psychiatry. 2005; 162:2022–30. [PubMed: 16263838]
2. Weamer EA, Emanuel JE, Varon D, Miyahara S, Wilkosz PA, Lopez OL, et al. The relationship of excess cognitive impairment in MCI and early Alzheimer's disease to the subsequent emergence of psychosis. Int Psychogeriatr. 2009; 21:78–85. [PubMed: 18814807]
3. Emanuel JE, Lopez OL, Houck PR, Becker JT, Weamer EA, DeMichele-Sweet MA, et al. Trajectory of cognitive decline as a predictor of psychosis in early Alzheimer disease in the cardiovascular health study. Am J Geriatr Psychiatry. 2011; 19:160–68. [PubMed: 20808116]
4. Sweet RA, Bennett DA, Graff-Radford NR, Mayeux R. Assessment and familial aggregation of psychosis in Alzheimer's disease from the National Institute on Aging Late Onset Alzheimer's Disease Family Study. Brain. 2010; 133:1155–62. [PubMed: 20147454]
5. Seltman HJ, Mitchell S, Sweet RA. A Bayesian model of psychosis symptom trajectory in Alzheimer's disease. Int J Geriatr Psychiatry. 2016; 31:204–10. [PubMed: 26216660]
6. Sweet RA, Seltman H, Emanuel JE, Lopez OL, Becker JT, Bis JC, et al. Effect of Alzheimer's disease risk genes on trajectories of cognitive function in the Cardiovascular Health Study. Am J Psychiatry. 2012; 169:954–62. [PubMed: 22952074]
7. Koppel J, Sunday S, Goldberg TE, Davies P, Christen E, Greenwald BS. Psychosis in Alzheimer's disease is associated with frontal metabolic impairment and accelerated decline in working memory: findings from the Alzheimer's Disease Neuroimaging Initiative. Am J Geriatr Psychiatry. 2014; 22:698–707. [PubMed: 23672944]
8. Koppel J, Goldberg TE, Gordon ML, Huey E, Davies P, Keehlisen L, et al. Relationships between behavioral syndromes and cognitive domains in Alzheimer disease: the impact of mood and psychosis. Am J Geriatr Psychiatry. 2012; 20:994–1000. [PubMed: 22048323]
9. Gilley DW, Whalen ME, Wilson RS, Bennett DA. Hallucinations and associated factors in Alzheimer's disease. J Neuropsychiatry. 1991; 3:371–76.
10. Gilley DW, Wilson RS, Beckett LA, Evans DA. Psychotic symptoms and physically aggressive behavior in Alzheimer's disease. J Am Geriatr Soc. 1997; 45:1074–79. [PubMed: 9288014]
11. Sweet RA, Pollock BG, Sukonick DL, Mulsant BH, Rosen J, Klunk WE, et al. The 5-HTTPR polymorphism confers liability to a combined phenotype of psychotic and aggressive behavior in Alzheimer's disease. Int Psychogeriatr. 2001; 13:401–09. [PubMed: 12003247]
12. Kaufer DI, Cummings JL, Christine D, Bray T, Castellon S, Masterman D, et al. Assessing the impact of neuropsychiatric symptoms in Alzheimer's disease: the Neuropsychiatric Inventory Caregiver Distress Scale. J Am Geriatr Soc. 1998; 46:210–15. [PubMed: 9475452]
13. Scarmeas N, Brandt J, Albert M, Hadjigeorgiou G, Papadimitriou A, Dubois B, et al. Delusions and hallucinations are associated with worse outcome in Alzheimer disease. Arch Neurol. 2005; 62:1601–08. [PubMed: 16216946]
14. Rabins PV, Mace NL, Lucas MJ. The impact of dementia on the family. JAMA. 1982; 248:333–35. [PubMed: 7087127]
15. Lopez OL, Wisniewski SR, Becker JT, Boller F, DeKosky ST. Psychiatric medication and abnormal behavior as predictors of progression in probable Alzheimer disease. Arch Neurol. 1999; 56:1266–72. [PubMed: 10520944]
16. Magni E, Binetti G, Bianchetti A, Trabucchi M. Risk of mortality and institutionalization in demented patients with delusions. J Geriatr Psychiatry Neurol. 1996; 9:123–26. [PubMed: 8873875]
17. Cummings JL, Diaz C, Levy M, Binetti G, Litvan II. Neuropsychiatric Syndromes in Neurodegenerative Disease: Frequency and Signficance. Semin Clin Neuropsychiatry. 1996; 1:241–47. [PubMed: 10320427]
18. Bassiony MM, Steinberg M, Rosenblatt A, Baker A, Lyketsos CG. Delusions and hallucinations in Alzheimer's disease: Prevalence and clinical correlates. Int J Geriatr Psychiatry. 2000; 15:99–107. [PubMed: 10679840]

19. Wilson RS, Tang Y, Aggarwal NT, Gilley DW, Mccann JJ, Bienias JL, et al. Hallucinations, cognitive decline, and death in Alzheimer's disease. Neuroepidemiology. 2006; 26:68–75. [PubMed: 16352909]
20. Schneider LS, Dagerman K, Insel PS. Efficacy and adverse effects of atypical antipsychotics for dementia: meta-analysis of randomized, placebo-controlled trials. Am J Geriatr Psychiatry. 2006; 14:191–210. [PubMed: 16505124]
21. Huybrechts KF, Gerhard T, Crystal S, Olfson M, Avorn J, Levin R, et al. Differential risk of death in older residents in nursing homes prescribed specific antipsychotic drugs: population based cohort study. BMJ. 2012; 344:e977. [PubMed: 22362541]
22. Pollock BG, Mulsant BH, Rosen J, Sweet RA, Mazumdar S, Bharucha A, et al. A randomized, double-blind, placebo-controlled comparison of citalopram and perphenazine for the acute treatment of psychosis and behavioral disturbances associated with dementia. Am J Psychiatry. 2002; 159:460–65. [PubMed: 11870012]
23. Leonpacher AK, Peters ME, Drye LT, Makino KM, Newell JA, Devanand DP, et al. Effects of Citalopram on Neuropsychiatric Symptoms in Alzheimer's Dementia: Evidence From the CitAD Study. Am J Psychiatry. 2016; 173:473–80. [PubMed: 27032628]
24. Kales HC, Valenstein M, Kim HM, McCarthy JF, Ganoczy D, Cunningham F, et al. Mortality risk in patients with dementia treated with antipsychotics versus other psychiatric medications. Am J Psychiatry. 2007; 164:1568–76. [PubMed: 17898349]
25. Sweet RA, Nimgaonkar VL, Devlin B, Lopez OL, DeKosky ST. Increased familial risk of the psychotic phenotype of Alzheimer disease. Neurology. 2002; 58:907–11. [PubMed: 11914406]
26. Hollingworth P, Hamshere ML, Holmans PA, O'Donovan MC, Sims R, Powell J, et al. Increased familial risk and genomewide significant linkage for Alzheimer's disease with psychosis. Am J Med Genet B Neuropsychiatr Genet. 2007; 144B:841–48. [PubMed: 17492769]
27. Bacanu SA, Devlin B, Chowdari KV, DeKosky ST, Nimgaonkar VL, Sweet RA. Heritability of psychosis in Alzheimer disease. Am J Geriatr Psychiatry. 2005; 13:624–27. [PubMed: 16009739]
28. Barral S, Vardarajan BN, Reyes-Dumeyer D, Faber KM, Bird TD, Tsuang D, et al. Genetic variants associated with susceptibility to psychosis in late-onset Alzheimer's disease families. Neurobiol Aging. 2015; 36:3116.
29. Hollingworth P, Sweet R, Sims R, Harold D, Russo G, Abraham R, et al. Genome-wide association study of Alzheimer's disease with psychotic symptoms. Mol Psychiatry. 2012; 17:1316–27. [PubMed: 22005930]
30. Biological insights from 108 schizophrenia-associated genetic loci. Nature. 2014; 511:421–27. [PubMed: 25056061]
31. Purcell SM, Wray NR, Stone JL, Visscher PM, O'Donovan MC, Sullivan PF, et al. Common polygenic variation contributes to risk of schizophrenia and bipolar disorder. Nature. 2009; 460:748–52. [PubMed: 19571811]
32. Genome-wide association study identifies five new schizophrenia loci. Nat Genet. 2011; 43:969–76. [PubMed: 21926974]
33. Anney R, Klei L, Pinto D, Almeida J, Bacchelli E, Baird G, et al. Individual common variants exert weak effects on the risk for autism spectrum disorders. Hum Mol Genet. 2012; 21:4781–92. [PubMed: 22843504]
34. de Los CG, Vazquez AI, Fernando R, Klimentidis YC, Sorensen D. Prediction of complex human traits using the genomic best linear unbiased predictor. PLoS Genet. 2013; 9:e1003608. [PubMed: 23874214]
35. Vazquez AI, Klimentidis YC, Dhurandhar EJ, Veturi YC, Paerez-Rodriguez P. Assessment of whole-genome regression for type II diabetes. PLoS One. 2015; 10:e0123818. [PubMed: 25885636]
36. McKhann G, Drachman D, Folstein M, Katzman R, Price D, Stadlan EM. 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. 1984; 34:939–44. [PubMed: 6610841]
37. Mirra SS, Heyman A, McKeel D, Sumi SM, Crain BJ, Brownlee LM, et al. The consortium to establish a registry for Alzheimer's disease (CERAD). Part II Standardization of the

neuropathologic assessment of Alzheimer's disease. Neurology. 1991; 41:479–86. [PubMed: 2011243]

38. McKeith IG, Dickson DW, Lowe J, Emre M, O'Brien JT, Feldman H, et al. Diagnosis and management of dementia with Lewy bodies: third report of the DLB Consortium. Neurology. 2005; 65:1863–72. [PubMed: 16237129]
39. DeMichele-Sweet MA, Klei L, Devlin B, Ferrell RE, Weamer EA, Emanuel JE, et al. No association of psychosis in Alzheimer disease with neurodegenerative pathway genes. Neurobiol Aging. 2011; 32:555–11.
40. Weamer EA, DeMichele-Sweet MA, Cloonan YK, Lopez OL, Sweet RA. Incident Psychosis in Subjects With Mild Cognitive Impairment or Alzheimer's Disease. J Clin Psychiatry. 2016; 77:e1564–e1569. [PubMed: 28086011]
41. Seshadri S, Fitzpatrick AL, Ikram MA, DeStefano AL, Gudnason V, Boada M, et al. Genome-wide analysis of genetic loci associated with Alzheimer disease. JAMA. 2010; 303:1832–40. [PubMed: 20460622]
42. Lambert JC, Ibrahim-Verbaas CA, Harold D, Naj AC, Sims R, Bellenguez C, et al. Meta-analysis of 74,046 individuals identifies 11 new susceptibility loci for Alzheimer's disease. Nat Genet. 2013; 45:1452–58. [PubMed: 24162737]
43. Lambert MJ, Hatch DR, Kingston MD, Edwards BC. Zung, Beck, and Hamilton Rating Scales as measures of treatment outcome: A meta-analytic comparison. J Consult Clin Psychol. 1986; 54:54–59. [PubMed: 3958302]
44. DeMichele-Sweet MA, Lopez OL, Sweet RA. Psychosis in Alzheimer's disease in the national Alzheimer's disease coordinating center uniform data set: clinical correlates and association with apolipoprotein e. Int J Alzheimers Dis. 2011
45. Tariot PN, Mack JL, Patterson MB, Edland SD, Weiner MF, Fillenbaum G, et al. The behavior rating scale for dementia of the Consortium to Establish a Registry for Alzheimer's Disease. Am J Psychiatry. 1995; 152:1349–57. [PubMed: 7653692]
46. Kaufer DI, Cummings JL, Ketchel P, Smith V, MacMillan A, Shelley T, et al. Validation of the NPI-Q, a brief clinical form of the Neuropsychiatric Inventory. J Neuropsychiatry Clin Neurosci. 2000; 12:233–39. [PubMed: 11001602]
47. Boada M, Cejudo JC, Tarraga L, Lopez OL, Kaufer D. Neuropsychiatric Inventory Questionnaire (NPI-Q): Spanish validation of a brief clinical form of the Neuropsychiatric inventory (NPI). Neurologia. 2002; 17:317–23. [PubMed: 12084358]
48. Cummings JL, Mega M, Gray K, Rosenberg-Thompson S, Carusi DA, Gornbein J. The neuropsychiatric inventory: comprehensive assessment of psychopathology in dementia. Neurology. 1994; 44:2308–14. [PubMed: 7991117]
49. Overall JE, Gorham DR. The brief psychiatric rating scale. Psychol Rep. 1962; 10:799–812.
50. Zubenko GS, Rosen J, Sweet RA, Mulsant BH, Rifai AH. Impact of psychiatric hospitalization on behavioral complications of Alzheimer's disease. Am J Psychiatry. 1992; 149:1484–91. [PubMed: 1357991]
51. Hughes CP, Berg L, Danziger WL, Coben LA, Martin RL. A new clinical scale for the staging of dementia. Br J Psychiatry. 1982; 140:566–72. [PubMed: 7104545]
52. Folstein MF, Folstein SE, McHugh PR. "Mini-mental state" A practical method for grading the cognitive state of patients for the clinician. J Psychiatr" Res. 1975; 12:189–98. [PubMed: 1202204]
53. Bacanu SA, Devlin B, Chowdari KV, DeKosky ST, Nimgaonkar VL, Sweet RA. Linkage analysis of Alzheimer disease with psychosis. Neurology. 2002; 59:118–20. [PubMed: 12105318]
54. Affymetrix axiom assay user manual. 2016
55. Naj AC, Jun G, Beecham GW, Wang LS, Vardarajan BN, Buros J, et al. Common variants at MS4A4/MS4A6E, CD2AP, CD33 and EPHA1 are associated with late-onset Alzheimer's disease. Nat Genet. 2011; 43:436–41. [PubMed: 21460841]
56. 2016. https://www.niagads.org/dataset/ng00027
57. SCZ1 Dataset. 2016

58. Lin CW, Chang LC, Tseng GC, Kirkwood CM, Sibille EL, Sweet RA. VSNL1 Co-Expression Networks in Aging Include Calcium Signaling, Synaptic Plasticity, and Alzheimer's Disease Pathways. Front Psychiatry. 2015; 6:30. [PubMed: 25806004]
59. Seney ML, Chang LC, Oh H, Wang X, Tseng GC, Lewis DA, et al. The Role of Genetic Sex in Affect Regulation and Expression of GABA-Related Genes Across Species. Front Psychiatry. 2013; 4:104. [PubMed: 24062698]
60. Kirov G. CNVs in neuropsychiatric disorders. Hum Mol Genet. 2015; 24:R45–R49. [PubMed: 26130694]
61. Mulle JG, Pulver AE, McGrath JA, Wolyniec PS, Dodd AF, Cutler DJ, et al. Reciprocal duplication of the Williams-Beuren syndrome deletion on chromosome 7q11. 23 is associated with schizophrenia. Biol Psychiatry. 2014; 75:371–77. [PubMed: 23871472]
62. Banerjee A, MacDonald ML, Borgmann-Winter KE, Hahn CG. Neuregulin 1-erbB4 pathway in schizophrenia: From genes to an interactome. Brain Res Bull. 2010; 83:132–39. [PubMed: 20433909]
63. Mulle JG, Dodd AF, McGrath JA, Wolyniec PS, Mitchell AA, Shetty AC, et al. Microdeletions of 3q29 confer high risk for schizophrenia. Am J Hum Genet. 2010; 87:229–36. [PubMed: 20691406]
64. Freedman R, Leonard S, Gault JM, Hopkins J, Cloninger CR, Kaufmann CA, et al. Linkage disequilibrium for schizophrenia at the chromosome 15q13-14 locus of the alpha7-nicotinic acetylcholine receptor subunit gene (CHRNA7). Am J Med Genet. 2001; 105:20–22. [PubMed: 11424985]
65. Kosoy R, Nassir R, Tian C, White PA, Butler LM, Silva G, et al. Ancestry informative marker sets for determining continental origin and admixture proportions in common populations in America. Hum Mutat. 2009; 30:69–78. [PubMed: 18683858]
66. DeMichele-Sweet MA, Sweet RA. Genetics of psychosis in Alzheimer's disease: a review. J Alzheimers Dis. 2010; 19:761–80. [PubMed: 20157235]
67. Murray PS, Kumar S, DeMichele-Sweet MA, Sweet RA. Psychosis in Alzheimer's Disease. Biol Psychiatry. 2014; 75:542–52. [PubMed: 24103379]
68. Sweet RA, Nimgaonkar VL, Devlin B, Jeste DV. Psychotic symptoms in Alzheimer disease: evidence for a distinct phenotype. Mol Psychiatry. 2003; 8:383–92. [PubMed: 12740595]
69. Blacker D, Albert MS, Bassett SS, Go RC, Harrell LE, Folstein MF. Reliability and validity of NINCDS-ADRDA criteria for Alzheimer's disease. The National Institute of Mental Health Genetics Initiative. Arch Neurol. 1994; 51:1198–204. [PubMed: 7986174]
70. Westra HJ, Peters MJ, Esko T, Yaghootkar H, Schurmann C, Kettunen J, et al. Systematic identification of trans eQTLs as putative drivers of known disease associations. Nat Genet. 2013; 45:1238–43. [PubMed: 24013639]
71. Wilkaniec A, Czapski GA, Adamczyk A. Cdk5 at crossroads of protein oligomerization in neurodegenerative diseases: facts and hypotheses. J Neurochem. 2016; 136:222–33. [PubMed: 26376455]
72. Okamoto K, Beach D. Cyclin G is a transcriptional target of the p53 tumor suppressor protein. EMBO J. 1994; 13:4816–22. [PubMed: 7957050]
73. Jordan-Sciutto KL, Morgan K, Bowser R. Increased Cyclin G1 Immunoreactivity During Alzheimer's Disease. J Alzheimers Dis. 1999; 1:409–17. [PubMed: 12214116]
74. Kotrla KJ, Chacko RC, Harper RG, Doody R. Clinical variables associated with psychosis in Alzheimer's disease. Am J Psychiatry. 1995; 152:1377–79. [PubMed: 7653698]
75. Smoller JW, Ripke S, Lee SH. Identification of risk loci with shared effects on five major psychiatric disorders: a genome-wide analysis. Lancet. 2013; 381:1371–79. [PubMed: 23453885]
76. Sekar A, Bialas AR, de Rivera H, Davis A, Hammond TR, Kamitaki N, et al. Schizophrenia risk from complex variation of complement component 4. Nature. 2016; 530:177–83. [PubMed: 26814963]
77. Fromer M, Roussos P, Sieberts SK, Johnson JS, Kavanagh DH, Perumal TM, et al. Gene expression elucidates functional impact of polygenic risk for schizophrenia. Nat Neurosci. 2016; 19:1442–53. [PubMed: 27668389]

78. Li M, Jaffe AE, Straub RE, Tao R, Shin JH, Wang Y, et al. A human-specific AS3MT isoform and BORCS7 are molecular risk factors in the 10q24. 2 schizophrenia-associated locus. Nat Med. 2016; 22:649–56. [PubMed: 27158905]

79. Gambuzza ME, Sofo V, Salmeri FM, Soraci L, Marino S, Bramanti P. Toll-like receptors in Alzheimer's disease: a therapeutic perspective. CNS Neurol Disord Drug Targets. 2014; 13:1542–58. [PubMed: 25106635]

80. Mani M, Lee UH, Yoon NA, Kim HJ, Ko MS, Seol W, et al. Developmentally regulated GTP-binding protein 2 coordinates Rab5 activity and transferrin recycling. Mol Biol Cell. 2016; 27:334–48. [PubMed: 26582392]

81. Wucherpfennig T, Wilsch-Brauninger M, Gonzalez-Gaitan M. Role of Drosophila Rab5 during endosomal trafficking at the synapse and evoked neurotransmitter release. J Cell Biol. 2003; 161:609–24. [PubMed: 12743108]

82. Brown TC, Tran IC, Backos DS, Esteban JA. NMDA receptor-dependent activation of the small GTPase Rab5 drives the removal of synaptic AMPA receptors during hippocampal LTD. Neuron. 2005; 45:81–94. [PubMed: 15629704]

83. Tumbarello DA, Waxse BJ, Arden SD, Bright NA, Kendrick-Jones J, Buss F. Autophagy receptors link myosin VI to autophagosomes to mediate Tom1-dependent autophagosome maturation and fusion with the lysosome. Nat Cell Biol. 2012; 14:1024–35. [PubMed: 23023224]

84. Li L, Zhang X, Le W. Autophagy dysfunction in Alzheimer's disease. Neurodegener Dis. 2010; 7:265–71. [PubMed: 20551691]

85. Merenlender-Wagner A, Malishkevich A, Shemer Z, Udawela M, Gibbons A, Scarr E, et al. Autophagy has a key role in the pathophysiology of schizophrenia. Mol Psychiatry. 2015; 20:126–32. [PubMed: 24365867]

86. Xu Y, Jagannath C, Liu XD, Sharafkhaneh A, Kolodziejska KE, Eissa NT. Toll-like receptor 4 is a sensor for autophagy associated with innate immunity. Immunity. 2007; 27:135–44. [PubMed: 17658277]

87. Eckart N, Song Q, Yang R, Wang R, Zhu H, McCallion AS, et al. Functional Characterization of Schizophrenia-Associated Variation in CACNA1C. PLoS One. 2016; 11:e0157086. [PubMed: 27276213]

88. Purcell SM, Moran JL, Fromer M, Ruderfer D, Solovieff N, Roussos P, et al. A polygenic burden of rare disruptive mutations in schizophrenia. Nature. 2014; 506:185–90. [PubMed: 24463508]

89. Andrade A, Hope J, Allen A, Yorgan V, Lipscombe D, Pan JQ. A rare schizophrenia risk variant of CACNA1I disrupts CaV3 channel activity. Sci Rep. 2016; 6:34233. [PubMed: 27756899]

90. Macdonald ML, Alhassan J, Newman JT, Richard M, Gu H, Kelley RM, Sampson AR, Fish KN, Penzes P, Wills ZP, Lewis DA, Sweet RA. Selective Loss of Smaller Spines in Schizophrenia. Am J Psychiatry. 2017

91. Briggs CA, Chakroborty S, Stutzmann GE. Emerging pathways driving early synaptic pathology in Alzheimer's disease. Biochem Biophys Res Commun. 2017; 483:988–97. [PubMed: 27659710]

92. Geda YE, Schneider LS, Gitlin LN, Miller DS, Smith GS, Bell J, et al. Neuropsychiatric symptoms in Alzheimer's disease: past progress and anticipation of the future. Alzheimers Dement. 2013; 9:602–08. [PubMed: 23562430]

93. Corvin A, Craddock N, Sullivan PF. Genome-wide association studies: a primer. Psychol Med. 2010; 40:1063–77. [PubMed: 19895722]

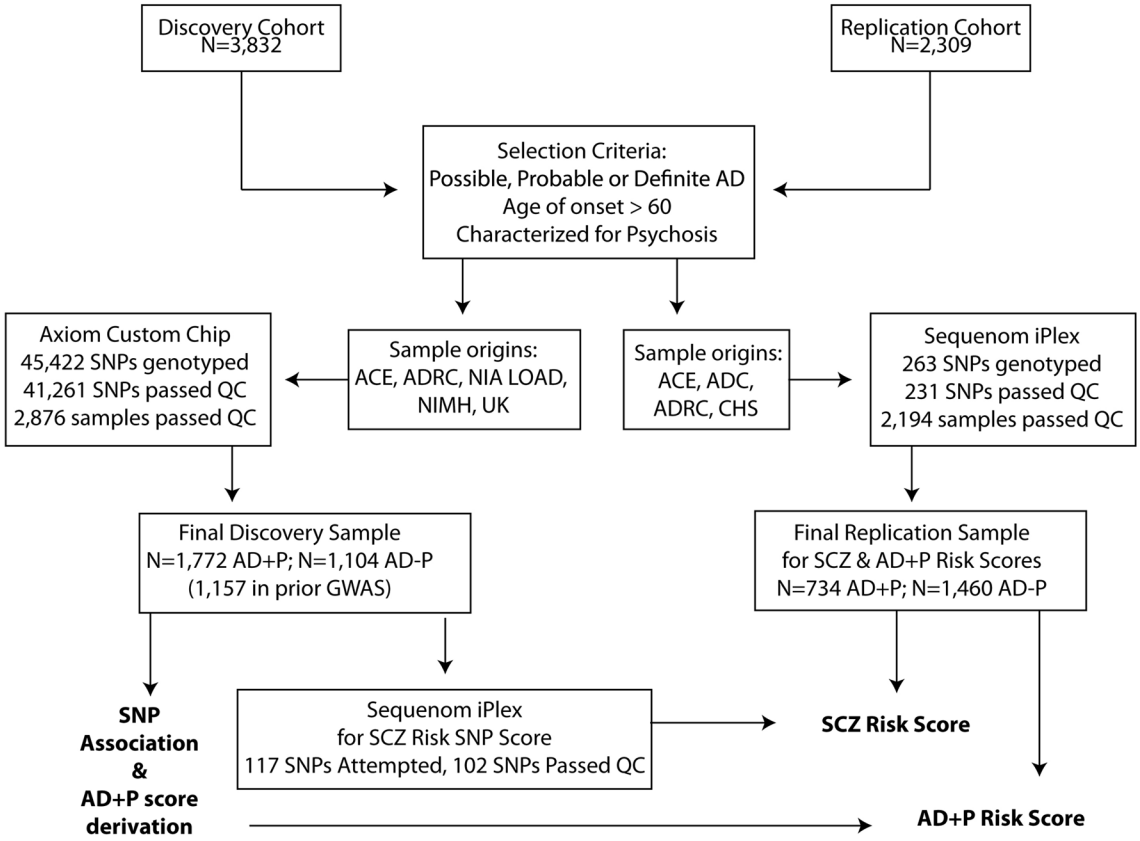

Figure 1. Diagram of the study design and workflow

Abbreviations used: ACE: Fundació ACE Barcelona Alzheimer Treatment and Research Center; ADRC: University of Pittsburgh Alzheimer Disease Research Center; NIA LOAD: National Institute on Aging's Late Onset Alzheimer's Disease Family Study; NIMH: National Institute of Mental Health Genetics Initiative AD Cohort; UK: Genetic and Environmental Risk in AD Consortium 1; ADC: consortium of National Institute on Aging Alzheimer Disease Centers; CHS: Cardiovascular Health Study; AD-P: Alzheimer disease without psychosis; AD+P: Alzheimer disease with psychosis; SCZ: schizophrenia; GWAS: Genome-wide association study

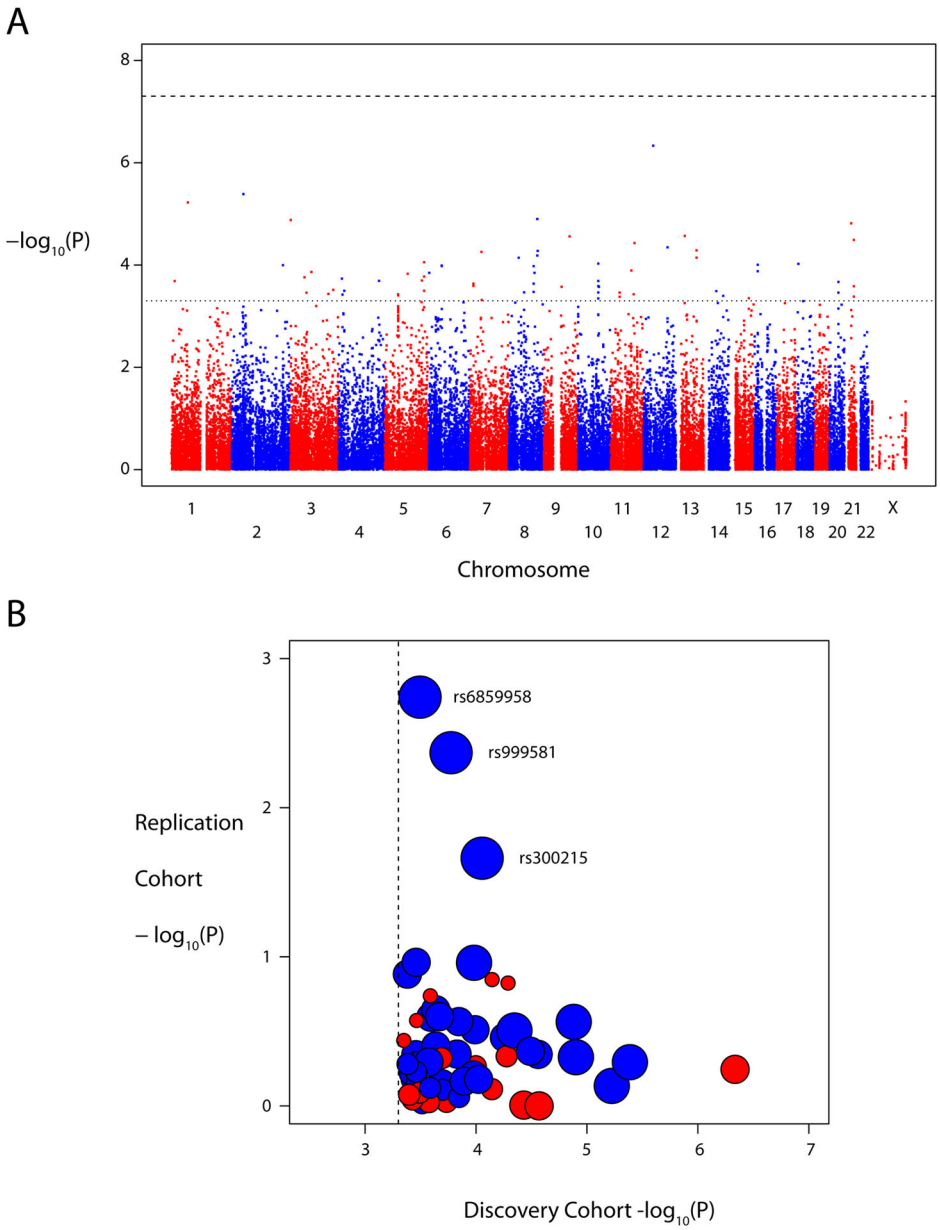

Figure 2. Discovery (a) and replication (b) analysis of AD+P risk SNPs

A. 67 SNPs reached p < 5e-4 in Stage 2 samples (dashed line). **B.** Stage 3 examined 60 of the 67 SNPS, three of which (top blue circles, Table 3) approached significance in meta-analysis ($P = 1.61\times10^{-6}$). In blue, SNPs showing same risk allele in Stages 2 and 3; red, Stage 2 versus 3 results differ in sign (risk allele); size of circle reflects meta-analysis $-\log_{10}(P)$.

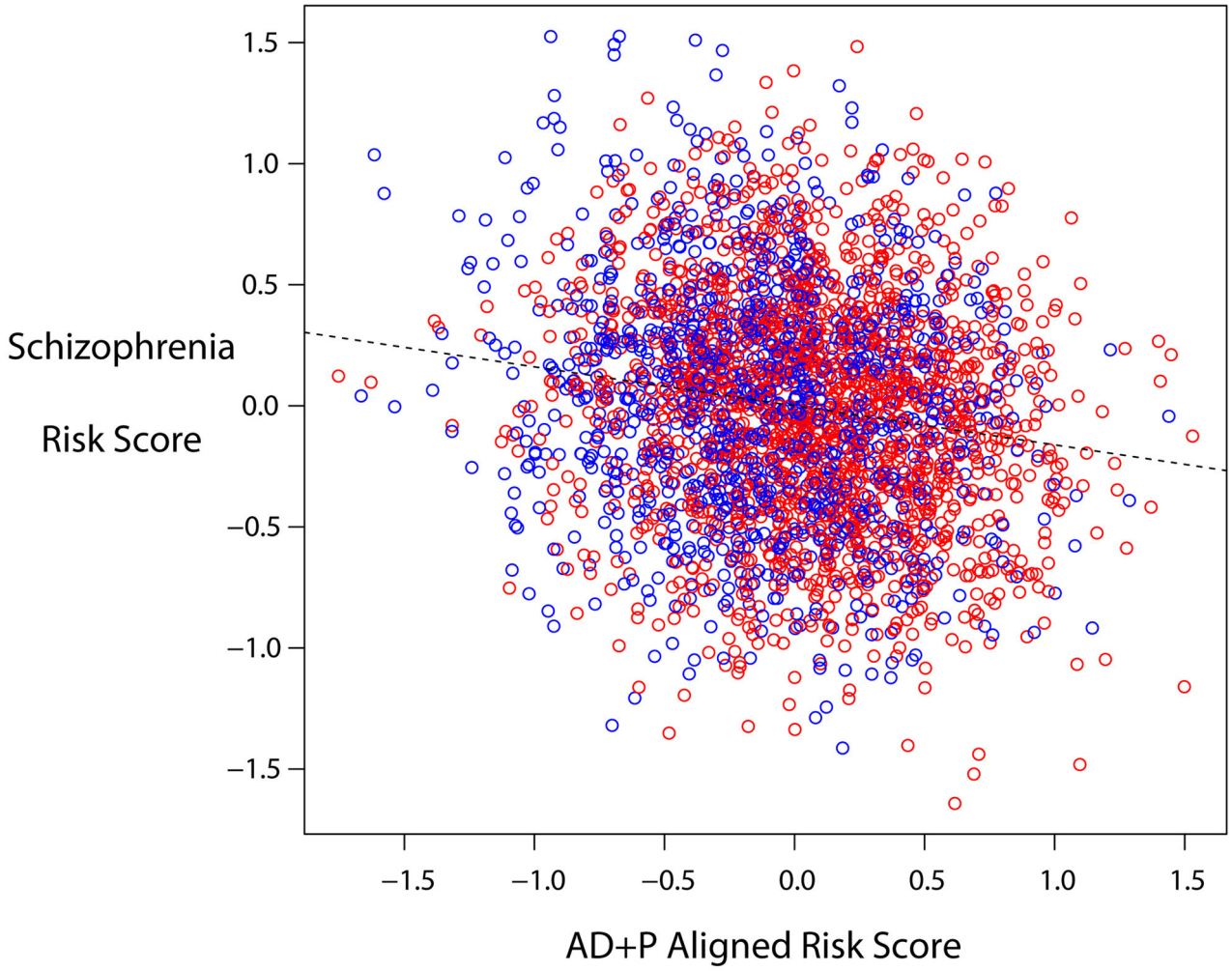

Fig. 3. Relationship between schizophrenia risk score and risk of psychosis in AD

Displayed are the risk scores for each subject, the score for schizophrenia uses the allele found to confer risk in [30], whereas the AD+P aligned score uses the same SNPs but assigns risk according to the AD+P association results. Red and blue circles indicate AD+P and AD-P subjects, respectively.

Table 1

Cohort subject characteristics.

	AD-P N(%) or Mean (SD)	AD+P N(%) or Mean (SD)	Total N (%) or Mean (SD)
A. Discovery Cohort			
N	1115 (38.8)	1761 (61.2)	2876 (100)
Female	722 (64.8)	1262 (71.7)	1984 (69.0)
AOO	75.5 (7.8)	74.1 (7.3)	74.6 (7.5)
Age	80.6 (7.3)	81.0 (6.8)	80.8 (7.0)
Last MMSE	14.6 (6.7)	12.0 (7.2)	13.2 (7.1)
B. Replication Cohort			
N	1460 (66.5)	734 (33.5)	2194 (100)
Female	905 (62.0)	496 (67.6)	1401 (63.8)
AOO	74.6 (8.2)	74.7 (7.4)	74.6 (8.0)
Age	80.5 (7.8)	81.2 (7.1)	80.7 (7.6)
Last MMSE	16.6 (6.6)	14.3 (7.4)	15.9 (6.9)

AD-P: Alzheimer disease without psychosis; AD+P: Alzheimer disease with psychosis; AOO: age of onset; MMSE: Mini mental state exam

Table 2

Risk score analysis: (A) Replication analysis of AD+P risk score; and (B) Schizophrenia risk score in two independent cohorts

(A) AD+P risk score using all 60 SNPs with $p < 5 \times 10^{-}4$ in the Discovery Cohort analysis that were genotyped in the independent Replication Cohort. Models included effects for weighted or unweighted risk score and ancestry eigenvectors. **(B)** Unweighted schizophrenia risk scores were calculated as standardized to number of SNPs available in each of the cohorts.

	b	P	OR	95%CI-OR	R^2(%)[2]
A. AD+P Risk Score					
Weighted Score	0.101	0.014[1]	1.107	1.01–1.21	0.30
Unweighted Score	0.107	0.010[1]	1.113	1.02–1.22	0.34
B. Schizophrenia Risk Score					
Discovery Cohort	−0.098	0.006	0.91	0.84–0.97	0.32
Replication Cohort	−0.091	0.030	0.91	0.83–1.00	0.25

AD+P: Alzheimer disease with psychosis. b: coefficient of the logistic regression (natural log of the odds ratio).

[1] One-sided, prior hypothesis

[2] Nagelkerke pseudo R^2, this value is the R^2 attributed to the risk score

Table 3

Meta-analysis of AD+P risk SNPs

The top three SNPs were located within *RP11-541P9.3* (an antisense transcript) 5′ to Cyclin G1 (*CCNG1*).

	Discovery		Replication		Meta-Analysis	
	Risk Allele	**Z**	**Risk Allele**	**Z**	**Z**	**P**
rs300215	A	3.921	A	2.933	4.530	5.91e-6
rs6859958	C	3.600	C	3.119	4.734	2.21e-6
rs999581	T	3.763	T	2.857	4.725	2.36e-6

AD+P: Alzheimer disease with psychosis

Table 4

(A) SNPs that most consistently were associated with reduced AD+P risk. (B) SNPs that most consistently were associated with increased AD +P risk in the two cohorts

eQTL indicates genes whose brain expression was found to associate with the identified schizophrenia risk SNP (or its assay proxy) with a false discovery rate < 0.05 in the CommonMind Consortium data set.[77] Sherlock indicates additional results of analyses of the CommonMind Consortium data set conducted using a Bayesian approach to prioritize consistency between the disease and eQTL associations in each of the schizophrenia GWAS loci.[87] This approach is motivated by recognition that the presence of an eQTL is not sufficient to indicate disease causality and that many SNPs within a locus are highly correlated, thus examination of the joint association of SNP and eQTL is more likely to identify genes contributing to disease risk. Genes identified by Sherlock with a corrected $p < 0.05$ are shown.

A. Top 20 SNPs Associated with Reduced AD+P Risk

Chr	Position (hg38)	Variant	Ref	Alt	Nearest Genecode Gene	Sum of b	eQTL	Sherlock
1	243391917	rs10803138	A	G	SDCCAG8	−0.365	*SDCCAG8*	None
5	109700365	rs4388249	C	T	MAN2A1	−0.277	*MAN2A1*	None
2	57911057	rs75575209	A	T	VRK2	−0.230	None	None
7	131882504	rs7801375	A	G	15kb 5′ of AC009518.4	−0.206	None	None
1	150059494	rs140505938	C	T	7.9kb 5′ of VPS45	−0.200	None	*CHTOP*
8	88576397	rs7819570	G	T	RP11-586K2.1	−0.198	*ENSG00000253553*	*ENSG00000253553*
11	109507345	rs12421382	C	T	RP11-708B6.2	−0.174	None	None
5	140764079	rs111896713	C	12-mer	22kb 5′ of PCDHA1	−0.171	*PCDHA7, PCDHA8, PCDHA13, PCDHA10, ZMAT2, PCDHA2, SRA1, NDUFA2, ANKHD1-EIF4EBP3, PCDHB3, PCDHAC2, WDR55*	None
1	97368969	rs76869799	C	G	DPYD	−0.162	None	*ENSG00000259946*
2	57760458	rs11682175	T	C	147kb 5′ of VRK2	−0.161	*FANCL*	None
12	110285440	rs4766428	C	T	ATP2A2	−0.155	*ANAPC7*	None
17	18055088	rs8082590	G	A	C17orf39	−0.146	*TOM1L2, DRG2, ATPAF2*	*TOM1L2, DRG2*
X	21362148	rs1378559	T	C	12kb 5′ of CNKSR2	−0.143	None	None
5	154301187	rs11740474	A	T	GALNT10	−0.140	None	None
4	175940150	rs1106568	G	A	GPM6A	−0.131	None	None
3	36817092	rs75968099	C	T	9.7kb 3′ of TRANK1	−0.128	*MLH1, DCLK3, LRRFIP2*	None
2	145678654	rs56807175	T	TC	75kb 5′ of AC079163.1	−0.123	None	None
11	133952674	rs75059851	A	G	IGSF9B	−0.122	None	None
22	41944840	rs1023500	T	C	CENPM	−0.120	*NAGA, CSDC2, WBP2NL, POLR3H*	*NAGA*

A. Top 20 SNPs Associated with Reduced AD+P Risk

Chr	Position (hg38)	Variant	Ref	Alt	Nearest Genecode Gene	Sum of b	eQTL	Sherlock
16	9852462	rs9922678	G	A	GRIN2A	−0.117	None	None

B. Top 10 SNPs Associated with Increased AD+P Risk

Chr	Position (hg38)	Variant	Ref	Alt	Nearest Genecode Gene	Sum of b	eQTL	
18	55865958	rs72934570	C	T	16kb 3′ of RP11-214L13.1	0.297	None	None
4	102225733	rs35518360	A	T	25kb 3′ of SLC39A8	0.284	None	None
20	38824551	rs6065094	A	G	PPP1R16B	0.257	None	None
18	55396445	rs78322266	G	T	TCF4	0.251	None	None
10	102981826	rs55833108	G	T	CNNM2	0.192	*BORCS7, AS3MT, INA*	None
12	2402665	rs2239063	A	C	CACNA1C	0.179	None	None
16	68155437	rs8044995	G	A	NFATC3	0.128	*PRMT7, ATP6V0D1*	None
3	136569563	rs7432375	G	A	STAG1	0.116	*PCCB*	*PCCB*
12	2235794	rs2007044	A	G	CACNA1C	0.111	None	None
5	138515503	rs3849046	C	T	ETF1	0.102	None	None

AD+P: Alzheimer disease with psychosis; Chr: Chromosome; hg38: Human Reference Genome Build 38; Ref: Reference Allele; Alt: Alternate Allele; Sum of b: sum of the SNP's coefficients from the logistic regressions conducted in the two cohorts