Published in final edited form as:
Alcohol Clin Exp Res. 2011 February ; 35(2): 376–385. doi:10.1111/j.1530-0277.2010.01353.x.

Limited Associations of Dopamine System Genes With Alcohol Dependence and Related Traits in the Irish Affected Sib Pair Study of Alcohol Dependence (IASPSAD)

Laura M. Hack, **Gursharan Kalsi**, **Fazil Aliev**, **Po-Hsiu Kuo**, **Carol A. Prescott**, **Diana G. Patterson**, **Dermot Walsh**, **Danielle M. Dick**, **Brien P. Riley**, and **Kenneth S. Kendler**
Department of Human and Molecular Genetics (LMH, DMD, BPR, KSK), Virginia Institute for Psychiatric and Behavioral Genetics, Virginia Common-wealth University, Richmond, Virginia; Department of Psychiatry (GK, FA, DMD, BPR, KSK), Virginia Institute for Psychiatric and Behavioral Genetics, Virginia Commonwealth University, Richmond, Virginia; Ankara University (FA), Ankara, Turkey; Department of Public Health (P-HK), National Taiwan University, Taipei, Taiwan; Department of Psychology (CAP), University of Southern California, Los Angeles, California; Shaftesbury Square Hospital (DGP), Belfast, Northern Ireland, United Kingdom; and Health Research Board (DW), Dublin, Ireland

Abstract

Background—Over 50 years of evidence from research has established that the central dopaminergic reward pathway is likely involved in alcohol dependence (AD). Additional evidence supports a role for dopamine (DA) in other disinhibitory psychopathology, which is often comorbid with AD. Family and twin studies demonstrate that a common genetic component accounts for most of the genetic variance in these traits. Thus, DA-related genes represent putative candidates for the genetic risk that underlies not only AD but also behavioral disinhibition. Many linkage and association studies have examined these relationships with inconsistent results, possibly because of low power, poor marker coverage, and/or an inappropriate correction for multiple testing.

Methods—We conducted an association study on the products encoded by 10 DA-related genes (*DRD1-D5, SLC18A2, SLC6A3, DDC, TH, COMT*) using a large, ethnically homogeneous sample with severe AD ($n = 545$) and screened controls ($n = 509$). We collected genotypes from linkage disequilibrium (LD)-tagging single nucleotide polymorphisms (SNPs) and employed a gene-based method of correction. We tested for association with AD diagnosis in cases and controls and with a variety of alcohol-related traits (including age-at-onset, initial sensitivity, tolerance, maximum daily drinks, and a withdrawal factor score), disinhibitory symptoms, and a disinhibitory factor score in cases only. A total of 135 SNPs were genotyped using the Illumina GoldenGate and Taqman Assays-on-Demand protocols.

Reprint requests: Laura M. Hack, BS, Department of Psychiatry, Virginia Institute for Psychiatric and Behavioral Genetics, P.O. Box 980424, Virginia Commonwealth University, Richmond, VA 23298-0424; Tel: +804-828-4120; Fax: +804-828-1471; hacklm@vcu.edu.

SUPPORTING INFORMATION
Additional Supporting Information may be found in the online version of this article:
Table S1. Marker Information and Nominal *p*-Values for AD and Alcohol-Related Traits for All SNPs
Table S2. Marker Information and Nominal *p*-Values for Disinhibitory Symptom Counts and Factor Score for All SNPs
Please note: Wiley-Blackwell is not responsible for the content or functionality of any supporting information supplied by the authors. Any queries (other than missing material) should be directed to the corresponding author for the article.

Results—Of the 101 SNPs entered into standard analysis, 6 independent SNPs from 5 DA genes were associated with AD or a quantitative alcohol-related trait. Two SNPs across 2 genes were associated with a disinhibitory symptom count, while 1 SNP in *DRD5* was positive for association with the general disinhibitory factor score.

Conclusions—Our study provides evidence of modest associations between a small number of DA-related genes and AD as well as a range of alcohol-related traits and measures of behavioral disinhibition. While we did conduct gene-based correction for multiple testing, we did not correct for multiple traits because the traits are correlated. However, false-positive findings remain possible, so our results must be interpreted with caution.

Keywords

Dopamine; Disinhibitory Psychopathology; Genetics; Association Study; Alcohol Dependence

Owing to its involvement in a broad range of functions, alteration in dopamine (DA) activity appears to play a central role in the etiology and/or treatment of many psychiatric disorders. DA's posited role in alcohol dependence (AD) stems from its involvement in the mesocorticolimbic reward pathway, which spans from the ventral tegmental area (VTA) to the nucleus accumbens (NA) and prefrontal cortex (Koob, 1992). DA was first implicated in mediating the effects of reward in Olds and Milner's (1954) classic experiments. Subsequent behavioral studies have generated considerable additional evidence to show that dopaminergic transmission in the mesocorticolimbic pathway is essential to reinforcing reward (Schultz, 1998).

Of all addictive substances, alcohol has created one of the greatest societal burdens (Rehm et al., 2009). AD is a clinically and etiologically heterogeneous condition that is 50 to 60% heritable (Dick et al., 2009; Prescott et al., 2006). Because of its etiological heterogeneity, considering subtypes of individuals with AD may increase power to detect underlying susceptibility variants. A recent latent class analysis of our sample found that cases could be divided into three classes based on comorbidities: a severe (S) class with the highest probabilities of all comorbidities and high novelty seeking (NS); a depressed (D) class with the highest probability of neuroticism and high probability of depression; and a mild (M) class with the lowest probabilities of all comorbidities (Sintov et al., 2010). These classes are consistent with the idea that alternate pathways to the development of AD exist, including negative affect regulation, in which alcohol consumption is a means of relieving negative mood states, and behavioral disinhibition, in which high consumption is part of an overall tendency to behave impulsively and to seek excitement (Sintov et al., 2010). The relatively independent nature of these pathways is supported by evidence that one common factor is largely responsible for the genetic susceptibility to internalizing disorders, while another common genetic factor explains most of the variation in externalizing disorders (Kendler et al., 2003). These common genetic liabilities may help explain why internalizing phenotypes, such as depression and anxiety/neuroticism, and disinhibitory phenotypes, including drug dependence (DD), antisocial behavior, and attention deficit/hyperactivity disorder (ADHD), are highly comorbid in many samples, including our Irish sample (Hasin et al., 2007; Kessler et al., 2006).

As the mesocorticolimbic pathway may be involved in the rewarding aspects of externalizing behavior, DA genes are reasonable candidates for susceptibility to AD as well as other disinhibitory psychopathology. We considered AD, ADHD, antisocial personality disorder (ASPD), conduct disorder (CD), DD, and NS to be part of the externalizing spectrum. Several studies suggest that a disinhibitory personality style, including NS, shares a common genetic influence with disorders in the externalizing spectrum (Jang et al., 2000; Krueger et al., 2002; Young et al., 2000). No twin studies have reported a direct genetic

overlap between ADHD and AD; however, we included this phenotype in the disinhibitory spectrum because childhood/early adolescent studies suggest ADHD shares genetic liability with CD (Dick et al., 2005; Eaves et al., 2000; Knopik et al., 2009; Nadder et al., 2002; Silberg et al., 1996; Tuvblad et al., 2009).

We examined ten DA system genes, including the following: the five receptors, *DRD1-D5*; two transporters, solute carrier family 18 member A2 (*SLC18A2* or vesicular monoamine transporter type 2, *VMAT2*) and solute carrier family 6, member 3 (*SLC6A3* or dopamine active transporter, *DAT* or *DAT1*); and three enzymes, tyrosine hydroxylase (TH), dopa decarboxylase (DDC), and catechol-*O*-methyltransferase (*COMT*). These genes are related to DA binding, biosynthesis, and catabolism, and they cover about 60% of the genes with these Gene Ontology terms. If no studies of association between a particular gene and trait are discussed below, we are unaware of any reports (either positive or negative) with rigorous methodology that have examined these associations.

DRD2

DRD2 (11q22–q23) has been examined most thoroughly in relation to AD and disinhibitory phenotypes. The majority of human studies have focused on the TaqIA restriction fragment length polymorphism (rs1800497), which a recent metaanalysis estimated to be associated with AD at a modest odds ratio (OR) of 1.31 (Le Foll et al., 2009). Neville and colleagues (2004) found this SNP to be within the coding region of the neighboring ankyrin repeat and kinase domain containing 1 (*ANKK1*) gene; therefore, the most parsimonious explanation is that phenotypic associations are because of this nonsynonymous coding change in *ANKK1*. However, this SNP could be tagging a polymorphism in *DRD2*, which contains several SNPs in modest linkage disequilibrium (LD) (approximately 0.7) with rs1800497. Furthermore, Dick and colleagues (2007) found weak associations between variants in *DRD2* and AD. Studies examining association with *DRD2* and aspects of heroin dependence have generally been positive (Le Foll et al., 2009). The meta-analysis of Gizer and colleagues (2009) identified no association of *DRD2* with ADHD.

Other DA Receptors

Rigorous studies of *DRD1* (5q35.1) and AD have typically estimated positive associations with modest effect sizes (Batel et al., 2008; Kim et al., 2007), while reports examining this phenotype and *DRD3* (3q13.3) have been negative (Le Foll et al., 2009). A recent review supports an association between the 48-bp variable number of tandem repeats (VNTR) in *DRD4* (11p15.5) and an intermediate phenotype termed urge for addictive substances, which refers to craving for substances of abuse (McGeary, 2009). We know of no rigorous reports investigating associations between *DRD5* (4p16.1) and AD or related traits.

Le Foll and colleagues' (2009) review notes that certain variants in *DRD3–D5* confer an increased risk of heroin dependence, while research on psychostimulant dependence has either been negative or inconclusive. A recent meta-analysis examining ADHD reported significant associations with variants in both *DRD4* and *DRD5*, whereas there was no association with *DRD3* (Gizer et al., 2009). Additionally, Kim and colleagues (2007) identified variants in *DRD1* that increase scores for the disinhibitory personality trait NS in alcohol-dependent subjects. Of all DA genes, *DRD4* has been the best studied for its role in NS. Munafò and colleagues' (2008) meta-analysis identified association with C521T (rs1800955) but not with *DRD4*'s 48-bp VNTR.

Transporters

SLC18A2 shuttles cytosolic monoamines into synaptic vesicles. Schwab and colleagues (2005) reported an association with variation in *SLC18A2* (10q25) and AD, but we are unaware of any reports of association with other disinhibitory psychopathology. SLC6A3 terminates DA signaling by removing this neurotransmitter from synaptic clefts. Some research has indicated that striatal SLC6A3 density and availability is reduced in alcohol-dependent subjects (Lind et al., 2009). van der Zwaluw and colleagues' (2009) review notes that many investigators have identified association between *SLC6A3*'s (5p15.3) best studied 40-bp VNTR and alcohol-withdrawal symptoms but typically not with AD. Generally, association studies of other drug use phenotypes and *SLC6A3* have been negative (Bousman et al., 2009; Li et al., 2006), although Guindalini and colleagues (2006) did find that alleles in a 30-bp VNTR increased risk of cocaine abuse. Additionally, there have been mixed results of association with *SLC6A3*'s functional 40-bp VNTR in relation to antisocial behavior in adolescence (Burt and Mikolajewski, 2008; Guo et al., 2007; Jorm et al., 2001; Schulz-Heik et al., 2008; Young et al., 2002). The meta-analysis of Gizer and colleagues (2009) found association of variants in *SLC6A3* with ADHD.

Enzymes

TH is the rate-limiting enzyme in DA synthesis. Dahmen and colleagues (2005) showed an increased frequency of the Val allele (Val81Met polymorphism) of *TH* (11p15) in patients with early-onset AD. Association studies of ADHD have been negative (Faraone and Khan, 2006), and no rigorous reports have examined any of the other disinhibitory traits. The final enzyme in the synthesis pathway, DDC, converts L-DOPA to DA. No published studies have reported an association with DDC (7p12.2) and AD or any disinhibitory phenotypes that we studied, including ASPD, ADHD, CD, DD, or NS.

COMT is a degradatory enzyme for catecholamines. The most well-researched polymorphism in *COMT* (22q11.21) is the common G>A transition (rs4680), which results in a valine to methionine substitution (Val158Met) and a decrease in enzyme activity by 3- to 4-folds (Lachman et al., 1996). Many studies have investigated the association between the low-activity allele (Met) and AD with inconsistent results (Köhnke, 2008). Investigators have reported associations between this SNP and methamphetamine abuse (Bousman et al., 2009) and NS (Golimbet et al., 2007; Tsai et al., 2004), while Cheuk and Wong's (2006) meta-analysis estimated no association with ADHD.

Study Goals

The primary goal of the present study was to test for association between AD and several alcohol-related quantitative traits, such as initial sensitivity (ISENS), and SNPs in 10 DA genes in a large, homogeneous sample. Because twin studies have found that a single genetic factor is largely responsible for the genetic susceptibility to AD and several other disinhibitory phenotypes and traits, a second goal of the study was to test for association of these SNPs with relevant symptoms of disinhibitory disorders as well as a disinhibitory factor score. To our knowledge, we are the first group to report results on an association analysis of both quantitative alcohol-related traits and symptoms of disinhibitory disorders within alcohol-dependent cases for a large group of DA-related genes.

MATERIALS AND METHODS

Subjects and Phenotype Measurement

Participants in the Irish Affected Sib Pair Study of Alcohol Dependence (IASPSAD) were recruited in Ireland and Northern Ireland between 1998 and 2002. Further details of the study design, sample ascertainment, and clinical characteristics of this sample are described elsewhere (Prescott et al., 2005). In brief, ascertainment of probands was mainly conducted in community alcoholism treatment facilities and public and private hospitals. Probands were eligible for study inclusion if they met the current DSM-IV criteria (American Psychiatric Association, 1994) for AD and if all four grandparents had been born in Ireland, Northern Ireland, Scotland, Wales, or England. After a prospective family was identified through probands, parents and potentially affected siblings whom the probands provided permission to contact were recruited.

Probands, siblings, and parents were interviewed by clinically trained research interviewers, most of whom had extensive clinical experience with alcoholism. The assessment included demographic characteristics, lifetime history of AD and other comorbid conditions, alcohol-related traits, personality features, and clinical records. All participants provided informed consent. Controls were recruited in the Northern Ireland from volunteers donating at the Northern Ireland Blood Transfusion Service and in the Republic from the Garda Siochana (the national police force) and the Forsa Cosanta Aituil (the army reserve). Controls were screened and their samples excluded if they reported a history of heavy drinking or problem alcohol use. In the present case–control study design, we included 545 independent AD cases and 509 controls with an ample yield of high-quality DNA for genotyping. The selection of cases was random with respect to AD severity and comorbid phenotypes.

In addition to the binary diagnosis of AD, we chose to examine several quantitative alcohol-related traits because examining such traits provides more power than analysis of dichotomous phenotypes; prior research in this sample detected linkage and association signals with these traits (e.g., Kuo et al., 2006). We assessed age-at-onset of AD (ONSET), subjective response to ethanol, maximum drinks in 24 hours (MAX24), and a factor score of withdrawal symptoms (WDSFS). ONSET was defined as the age at which the first criterion for DSM-IV AD was satisfied. Subjective response to ethanol was assessed using the self-rating of the effects of ethanol (SRE, Schuckit et al., 1997) to form two scores, ISENS and tolerance/maximum drinking (TOLMX). The SRE inquires about how many drinks were needed for a subject to experience effects from alcohol consumption at different stages of use. ISENS is based on “the first 5 times you ever drank,” and items contributing to TOLMX concern the “period when you drank the most.” The score of each measure was computed by summing the number of drinks required to produce an effect and dividing by the number of effects endorsed. The SRE has been shown to have good internal consistency and test–retest reliability to successfully identify people who had low response to alcohol in a laboratory challenge test and to be associated with AD diagnosis in several populations. Because of non-normal distributions of the regression residuals, we log transformed values for ISENS and TOLMX. MAX24 refers to the largest number of drinks an individual reported ever having consumed in 24 hours. The withdrawal severity factor score was based on ten symptoms in the SSAGA interview (such as hands trembling, feeling anxious following cessation or reduction of drinking). To account for the possible non-equal contribution of each symptom to withdrawal severity, a factor analysis was conducted (for details see Kuo et al., 2006). A factor score of withdrawal severity for each individual was derived based on the item loadings on one major factor, which accounted for 70% of the variance in these symptoms based on the entire IASPSAD sample.

We also tested the DA genes for association with scores for disinhibitory disorders. Symptom counts for alcohol dependence (ADsx, range 3 to 7) were assessed using the Semi-Structured Assessment of the Genetics of Alcoholism (SSAGA, version 11, Bucholz et al., 1994), modified to reduce assessment time. Counts for illicit substance use drug dependence (DDsx, range 0 to 7), conduct disorder (CDsx, range 0 to 14), and antisocial personality disorder (ASPDsx, range 0 to 9) were collected using adapted versions of the Structured Clinical Interview for the DSM-IV Disorders (SCID, Spitzer and Williams, 1985). Drugs assessed as part of substance use included cannabis, sedatives, stimulants, cocaine, opiates, hallucinogens, and other drugs (e.g., steroids, nitrous oxide). All drugs were considered illicit in this context because the subjects were asked only about nonmedical use (e.g., use without a prescription, use in greater amounts/more often than was prescribed, or use not in the intended manner). Other measures include retrospective reports of childhood attention deficit/hyperactivity disorder (ADHDsx) using items from Wender's Childhood Problem Behavior Checklist (Wender, 1971) and novelty seeking scores (NS) using the 18-item version from Cloninger's Tridimensional Personality Questionnaire (Cloninger et al., 1994). Scores for ADHDsx and NS were rescaled such that their range is 0 to 1. Finally, we tested for association with a factor score based on the item loadings on one major factor for all disinhibitory phenotypes. We modeled this analysis off those previously conducted by investigators in Virginia (Dick et al., 2008; Hettema et al., 2008) and in the Netherlands (Boomsma et al., 2000). Because of non-normal distributions, we log transformed ADsx, ASPDsx, CDsx, and DDsx for the factor score analysis.

Tag SNP Selection and Genotyping

The majority of genotyping was conducted in Dr. David Goldman's laboratory at NIAAA using the Ilumina GoldenGate method. For details of study design, see Hodgkinson and colleagues (2008). In instances where additional SNPs had to be genotyped to complete tagging in our sample, we selected LD-tagging SNPs (tSNPs) with Tagger (de Bakker et al., 2005) as implemented in Haploview 3.2 (Barrett et al., 2005) using the default criteria of $r^2 \geq 0.8$ and minor allele frequency (MAF) ≥0.2. As common variation is generally considered to be ≥0.05, our tagging SNPs capture very common variation. Because SNPs genotyped using the Ilumina GoldenGate platform were chosen based on being African haplotype tagging, some SNPs have a MAF < 0.2 in our Caucasian sample. However, no SNP has an MAF < 0.01, which was our threshold for eliminating SNPs in the overall sample. For genes displaying several isoforms, the longest isoform was chosen for tag selection but to limit genotyping load and cost, 5′ and 3′ regions of the genes and ESTs were not directly tagged.

tSNPs were genotyped in-house as monoplex reactions using Taqman Assays-on-Demand (Applied Biosystems, Foster City, CA). To ensure uniformity and accuracy, all reaction steps were performed using the Eppendorf 5075 automated liquid-handling platform. Stringent evaluation of initial data is important to avoid artifactual effects of genotyping errors; therefore, all genotypes were independently assessed by two raters. Ambiguous calls were discussed and in cases of non-resolution, genotypes were dropped from the analyses. Individual DNA samples with 20% or more missing genotypes across the entire study were also excluded. Individual SNPs were excluded if they showed deviation from Hardy–Weinberg equilibrium (using a regular cut-off p-value of 0.001) in the overall sample and controls alone, had an MAF < 1%, or had a low genotyping call rate (<80%). The present study reports on the results from genes included in the DA functional domain (Hodgkinson et al., 2008), including *DRD1–5, SLC18A2, SLC6A3, DDC, TH*, and *COMT*. Several coding SNPs were genotyped, such as rs155417 and rs5326 in *DRD1*, rs6279 in *DRD2*, rs6347 in *SLC6A3*, rs11575542 and rs11575377 in DDC, and the well-studied nonsynonymous SNP rs4680 in *COMT*. The well-known TaqIA polymorphism in *DRD2* was not included because, as noted previously, it is actually located in a different gene.

Statistical Methods

Single-marker analyses were implemented in PLINK 1.07 (Purcell, http://pngu.mgh.harvard.edu/~purcell/plink/) using logistic regression for the binary trait of AD and linear regression for the quantitative traits in cases only to calculate effect size (either OR or regression coefficient) and significance level. We used sex as a covariate in the logistic regressions and both age and sex as covariates in the linear regressions. To address the possibility of type I error because of multiple testing of several SNPs within individual genes, we permuted each *p*-value 10,000 times using the gene-based set test in PLINK and only reported the empirical *p*-value here if it was significant after this correction. We reasoned that gene-based correction was sufficiently conservative because all selected genes have a priori evidence of association with AD and/or related phenotypes. We used the set-based test in PLINK for multiple test correction because this method allows for identification of independent SNPs determined by a selected threshold. We changed the default threshold for LD from $r^2 = 0.5$ to $r^2 = 0.8$ because our LD-tagging SNPs were selected based on $r^2 \geq 0.8$. Additionally, we did not correct for multiple phenotypes because (i) they reflect reasonable a priori hypotheses and (ii) they are, in some cases, substantially inter-correlated, making it difficult to implement any simple multiple test correction.

We used the FACTOR procedure in SAS version 9.2 (SAS Institute Inc., Cary, NC) to determine the factor structure of ADsx, ASPDsx, ADHDsx, CDsx, DDsx, and NS scores. All phenotypes were entered into a principal component analysis using the default orthogonal rotation method. We then calculated factor scores for each individual with nonmissing data for all disinhibitory phenotypes and tested for association with these scores.

Power estimates for this study were calculated using QUANTO 1.2.4 (http://hydra.usc.edu/gxe) for the dichotomous AD outcome (Table 1) and the continuous traits (Table 2) using a two-sided *t*-test with a significance level of 0.05 and a range of MAF from 0.05 to 0.4. We assumed an additive mode of inheritance. Effect sizes are listed as OR from 1.1 to 1.5 for AD and variation in the traits from 1 to 5% for continuous outcomes. The power for the dichotomous outcome was based on a lifetime population risk for AD of 12.5% (Hasin et al., 2007).

RESULTS

Missingness

Genotyping was completed for 135 SNPs in ten DA system genes, but 10 SNPs were excluded because of a low genotyping rate (<80%) and 24 because of a MAF < 0.01. The average genotyping call rate for the remaining 101 SNPs was 98.7% (90.6 to 100%). All remaining SNPs were in Hardy–Weinberg equilibrium (regular cutoff *p*-value of 0.001). Geno-typing error rate was estimated using duplicates at 0.6%. Among the 1054 genotyped individuals, 26 (8 cases and 18 controls) were excluded because of ≥20% missing genotypes, leaving a total of 1,028 individuals. Of them, 592 individuals were missing no genotypes, 374 were missing 1 to 5, 40 were missing 6 to 10, 15 were missing 11 to 15, and 7 were missing 16 to 20. After QC measures were completed, the following number of SNPs from each gene were entered in standard analysis: 4 SNPs in *DRD1*, 15 in *DRD2*, 13 in *DRD3*, 4 in *DRD4*, 7 in *DRD5*, 11 in *SLC18A2*, 12 in *SLC6A3*, 22 in *DDC*, 3 in *TH*, and 10 in *COMT*.

Principal Component Analysis

Table 3 lists the means and standard deviations for the disinhibitory symptom counts as well as the Pearson correlations among counts included in the factor analysis. By both a scree plot and a traditional eigenvalue criterion, only one factor was evident with an eigenvalue of

2.55 that accounted for 42% of the variance. The eigenvalue difference between the first and second factors was 1.56 and all other factors comprised <17% of the variance. The component loadings for each of the symptom counts were as follows: ADsx, 0.41; ADHDsx, 0.65; ASPDsx, 0.82; CDsx, 0.87, DDsx, 0.56, and NS scores, 0.47. The comparatively low factor loading for ADsx may be explained by the restricted range of AD symptoms. Subjects could only have within the range of 3 to 7 symptoms and over 80% had ≥6 symptoms.

Single-Marker Association

In Table 4, marker information, nominal *p*-values, and effect sizes are provided for the 6 independent SNPs (in bold) from 5 DA genes that were significant after permutation testing with either AD or a quantitative alcohol-related trait. All other SNPs in this table did not pass permutation testing. Effect sizes are listed as OR for AD and as β, the regression coefficient, for the quantitative traits. We used an additive model in which each additional minor allele will increase (+) or decrease (−) the units of measure (symptoms, years, scores, etc.) by the amount of the coefficient. For example, in rs11575542, β = 0.80 for DDsx, which means that each additional minor allele will increase DD symptoms by 0.80 of a symptom. β Values are more difficult to interpret for ISENS, TOLMX, and the factor score because they are log transformed and for ADHDsx and NS because their units have been rescaled. For information on the full list of SNPs, see Table S1. Three SNPs (1 in *DRD4* and 2 in *SLC6A3*) were associated with AD, 2 SNPs (in *DRD5* and *TH*) with WDSFS, and 1 in *DRD3* with ISENS.

Table 5 presents the marker information, nominal *p*-values, and effect sizes for SNPs (in bold) that were significant after permutation with the disinhibitory symptoms and the disinhibitory factor score. All other SNPs in this table did not pass permutation testing. See Table S2 for a complete listing of all SNPs that underwent analysis for disinhibitory phenotypes. Two independent SNPs across 2 genes were associated with one or more disinhibitory symptom counts after permutation testing, including 1 SNP in *SLC6A3* with ADsx and 1 in *DDC* with DDsx. Additionally, 1 SNP in *DRD5* was significantly associated with for the factor score.

DISCUSSION

For greater than a half century, dopaminergic dysregulation has been implicated in AD and other disinhibitory psychopathology. Molecular genetics studies over the past 20 years have attempted to demonstrate associations with DA genes and disinhibitory phenotypes, producing an inconsistent and controversial literature. Meta-analyses, reviews, and reports with rigorous methodology suggest that variation in DA genes does contribute to susceptibility to disinhibitory traits, although not to the extent and effect size originally hypothesized.

We attempted to address some of the problems that have riddled candidate gene studies by using a relatively large, ethnically homogeneous sample with severe AD. We genotyped a sufficient number of tSNPs to cover most of the variation within 10 DA system genes, including *DRD1–D5, SLC18A2, SLC6A3, DDC, TH*, and *COMT*. Our study not only tested for association with the categorical diagnosis of AD but also with quantitative alcohol-related traits, which give more power than dichotomous traits and provide additional clinical information beyond a binary phenotype. Within alcohol-dependent cases only, we tested for association with disinhibitory psychopathology, including symptoms for AD, ASPD, ADHD, CD, DD, and scores for NS. Additionally, we assessed for association with a disinhibitory factor score. While there is no direct genetic connection between childhood ADHD and alcohol abuse/dependence, we included it in our analysis because it loaded onto

a single disinhibitory factor. Finally, we only reported findings that were significant after set-based permutation, which limits the possibility that any of our results are false positives.

Overall, we found evidence for association with modest effect sizes between a small number of DA-related genes and AD, alcohol-related traits, and disinhibitory phenotypes. The minor allele frequencies for several of the positive SNPs are low. The limited number of positive signals suggests that these 10 DA system genes play a minor role in susceptibility to AD and related disinhibitory psychopathology, which is consistent with previous meta-analyses, reviews, and reports with rigorous methodology.

Receptors

In agreement with reports from several other groups (Gorwood et al., 2001; Lee and Ryu, 2002; Wiesbeck et al., 2006), we did not find significant association after permutation between the well-studied *DRD3 BalI* polymorphism (rs6280) and AD. However, we did show that another SNP (rs2654754, $p = 0.0021$, $\beta = 0.24$) in *DRD3* is associated with the quantitative trait of ISENS. Perhaps we obtained these findings when other researchers did not because our sample size is larger, we captured most of the variation in *DRD3* with our 11 tSNPs, and we assessed quantitative alcohol-related traits in addition to the dichotomous phenotype. However, the chance that this is a false positive is greater in light of the fact that the MAF = 2.5%.

Moreover, we identified an association with 1 SNP in *DRD4* (rs12280580, $p = 0.011$, OR = 1.28) and AD. While no other groups have reported associations with this particular SNP and AD or disinhibitory psychopathology, several meta-analyses have noted associations with variants in *DRD4* and urge for addictive substances (McGeary, 2009), ADHD (Gizer et al., 2009), and NS (Munafò et al., 2008). Further-more, we identified associations with the same SNP in *DRD5* and two phenotypes, including withdrawal (rs7655090, $p = 0.0017$, $\beta = -0.37$) and the factor score (rs7655090, $p = 0.0094$, $\beta = -0.34$). The fact that *DRD5* is associated with an alcohol-related trait and the factor score suggests it may contribute to AD through the broader disinhibitory spectrum. However, as only one SNP is associated with the factor score and this polymorphism has a low MAF (4.9%), this finding might represent a false positive.

Perhaps we did not identify strong evidence for association of DA genes with the general disinhibitory factor because our design assessed disinhibitory phenotypes in subjects with AD. Other investigators who have been successful in finding associations with a factor score (of internalizing behavior) included a broader range of subjects (Boomsma et al., 2000; Hettema et al., 2008). Another explanation is that variation in DA genes may contribute more to risk for specific disorders than the liability to a general disinhibitory spectrum of disorders.

Transporters

Two SNPs in *SLC6A3* (rs27048, $p = 0.042$, OR = 0.68; rs10052016, $p = 0.00055$, OR = 1.33) were associated with the dichotomous phenotype of AD. Within the same gene, we also identified an association between the common allele (C) of another SNP (rs6350, $p = 0.0021$, $\beta = -0.36$) and AD symptoms. Lind and colleagues (2009) found association of the same allele in rs6350 with problem drinking in a Finnish population.

Enzymes

We identified association of one SNP in *TH* (rs11564717) with the withdrawal factor score ($p = 0.0094$, $\beta = 0.59$).We did not show association of the Val81Met (rs6356) variant with

any alcohol-related traits, although Dahmen and colleagues (2005) identified association of this polymorphism with early-onset AD.

Furthermore, we did not detect any signal with *COMT*'s well-studied functional polymorphism rs4680, which has been associated with a number of disinhibitory phenotypes, including AD (Köhnke, 2008), methamphetamine abuse (Bousman et al., 2009), and NS (Golimbet et al., 2007; Tsai et al., 2004). One explanation for this may be that none of the potential risk alleles were found in our population. It is noteworthy that the SNP in DDC (rs11575542, $p = 0.0028$, $\beta = 0.80$) that we identified as associated with DD symptoms is a missense coding polymorphism that results in a substitution from Arg to Gln; however, the MAF of this SNP in our sample is only 1.5%, which increases the likelihood that it is a false positive.

Although, in many ways, we improved the design of previous candidate gene studies, our report still has limitations. First, as noted previously, a proper correction for multiple testing has been problematic in these studies. Using too liberal an approach will maximize power but is likely to lead to false positives. A correction method that is too conservative will decrease power to detect true results. We attempted to strike a balance between the two approaches by using gene-based correction. However, we did not correct for testing multiple phenotypes because there is a priori evidence of association of at least some of the 10 DA system genes with each of the phenotypes examined. Furthermore, many of these phenotypes are highly inter-correlated, making an appropriate correction problematic. It can be argued that our approach is still too liberal. The possibility that some proportion of our findings represents false positives is plausible. Therefore, our findings should be considered tentative, pending the outcome of attempted replications. Secondly, quantitative alcohol-related traits and disinhibitory symptoms were measured only among cases but not controls, so the values for these traits are not representative of the full variation in the population. Thus, the most meaningful replication of our study would be in population sample. Thirdly, although the LD patterns of the DA genes are compatible with the Hapmap CEPH population data, it remains possible that we lack complete coverage of common variation in our Irish sample. Additionally, the impact of rare functional polymorphisms was not assessed. Finally, we did not include all genes that affect dopaminergic tone, such as dopamine β-hydroxylase (*DBH*) and the monoamine oxidase (*MAO*) genes.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

This work was supported by National Institutes of Health grant R01-AA-11408 and the Irish Health Research Board for original data collections and data analysis.

REFERENCES

American Psychiatric Association. Diagnostic and Statistical Manual of Mental Disorders. 4th ed. Washington, DC: American Psychiatric Association; 1994.

de Bakker PI, Yelensky R, Pe'er I, Gabriel SB, Daly MJ, Altshuler D. Efficiency and power in genetic association studies. Nat Genet. 2005; 37:1217–1223. [PubMed: 16244653]

Barrett JC, Fry B, Maller J, Daly MJ. Haploview: analysis and visualization of LD and haplotype maps. Bioinformatics. 2005; 21:263–265. [PubMed: 15297300]

Batel P, Houchi H, Daoust M, Ramoz N, Naassila M, Gorwood P. A haplotype of the DRD1 gene is associated with alcohol dependence. Alcohol Clin Exp Res. 2008; 32:567–572. [PubMed: 18341651]

Boomsma DI, Beem AL, van den Berg M, Dolan CV, Koopmans JR, Vink JM, de Geus EJ, Slagboom PE. Netherlands twin family study of anxious depression (NETSAD). Twin Res. 2000; 3:323–324. [PubMed: 11463154]

Bousman CA, Glatt SJ, Everall IP, Tsuang MT. Genetic association studies of methamphetamine use disorders: a systematic review and synthesis. Am J Med Genet B Neuropsychiatr Genet. 2009; 150B:1025–1049. [PubMed: 19219857]

Bucholz KK, Cadoret R, Cloninger CR, Dinwiddie SH, Hesselbrock VM, Nurnberger JIJ, Reich T, Schmidt I, Schuckit MA. A new, semi-structured psychiatric interview for use in genetic linkage studies: a report on the reliability of the SSAGA. J Stud Alcohol. 1994; 55:149–158. [PubMed: 8189735]

Burt SA, Mikolajewski AJ. Preliminary evidence that specific candidate genes are associated with adolescent-onset antisocial behavior. Aggress Behav. 2008; 34:437–445. [PubMed: 18366104]

Cheuk DK, Wong V. Meta-analysis of association between a catechol-O-methyltransferase gene polymorphism and attention deficit hyperactivity disorder. Behav Genet. 2006; 36:651–659. [PubMed: 16676224]

Cloninger, CR.; Przybeck, TR.; Svrakic, DM.; Wetzel, RD. The Temperament and Character Inventory (TCI): A Guide to Its Development and Use. St Louis, MO: Center for Psychobiology of Personality, Washington University; 1994.

Dahmen N, Völp M, Singer P, Hiemke C, Szegedi A. Tyrosine hydroxylase Val-81-Met polymorphism associated with early-onset alcoholism. Psychiatr Genet. 2005; 15:13–16. [PubMed: 15722952]

Dick DM, Aliev F, Wang JC, Grucza RA, Schuckit M, Kuperman S, Kramer J, Hinrichs A, Bertelsen S, Budde JP, Hesselbrock V, Porjesz B, Edenberg HJ, Bierut LJ, Goate A. Using dimensional models of externalizing psychopathology to aid in gene identification. Arch Gen Psychiatry. 2008; 65:310–318. [PubMed: 18316677]

Dick, DM.; Prescott, CP.; McGue, M. The genetics of substance use and substance use disorders. In: Kim, Y-K., editor. Handbook of Behavior Genetics. New York: Springer Science + Business Media, LLC; 2009.

Dick DM, Viken RJ, Kaprio J, Pulkkinen L, Rose RJ. Understanding the covariation among childhood externalizing symptoms: genetic and environmental influences on conduct disorder, attention deficit hyperactivity disorder, and oppositional defiant disorder symptoms. J Abnorm Child Psychol. 2005; 33:219–229. [PubMed: 15839499]

Dick DM, Wang JC, Plunkett J, Aliev F, Hinrichs A, Bertelsen S, Budde JP, Goldstein EL, Kaplan D, Edenberg HJ, Nurnberger JJ, Hesselbrock V, Schuckit M, Kuperman S, Tischfield J, Porjesz B, Begleiter H, Bierut LJ, Goate A. Family-based association analyses of alcohol dependence phenotypes across DRD2 and neighboring gene ANKK1. Alcohol Clin Exp Res. 2007; 31:1645–1653. [PubMed: 17850642]

Eaves L, Rutter M, Silberg JL, Shillady L, Maes H, Pickles A. Genetic and environmental causes of covariation in interview assessments of disruptive behavior in child and adolescent twins. Behav Genet. 2000; 30:321–334. [PubMed: 11206087]

Faraone SV, Khan SA. Candidate gene studies of attention-deficit/hyperactivity disorder. J Clin Psychiatry. 2006; 67:13–20. [PubMed: 16961425]

Gizer IR, Ficks C, Waldman ID. Candidate gene studies of ADHD: a meta-analytic review. Hum genet. 2009; 126:51–90. [PubMed: 19506906]

Golimbet VE, Alfimova MV, Gritsenko IK, Ebstein RP. Relationship between dopamine system genes and extraversion and novelty seeking. Neurosci Behav Physiol. 2007; 37:601–606. [PubMed: 17657431]

Gorwood P, Limosin F, Batel P, Duaux E, Gouya L, Adès J. The genetics of addiction: alcohol-dependence and D3 dopamine receptor gene. Pathol Biol (Paris). 2001; 49:710–717. [PubMed: 11762133]

Guindalini C, Howard M, Haddley K, Laranjeira R, Collier D, Ammar N, Craig I, O'Gara C, Bubb VJ, Greenwood T, Kelsoe J, Asherson P, Murray RM, Castelo A, Quinn JP, Vallada H, Breen G. A dopamine transporter gene functional variant associated with cocaine abuse in a Brazilian sample. Proc Natl Acad Sci U S A. 2006; 103:4552–4557. [PubMed: 16537431]

Guo G, Roettger ME, Shih JC. Contributions of the DAT1 and DRD2 genes to serious and violent delinquency among adolescents and young adults. Hum Genet. 2007; 121:125–136. [PubMed: 17120049]

Hasin DS, Stinson FS, Ogburn E, Grant BF. Prevalence, correlates, disability, and comorbidity of DSM-IV alcohol abuse and dependence in the United States: results from the National Epidemiologic Survey on Alcohol and Related Conditions. Arch Gen Psychiatry. 2007; 64:830–842. [PubMed: 17606817]

Hettema JM, An SS, Bukszar J, van den Oord EJ, Neale MC, Kendler KS, Chen X. Catechol-O-methyltransferase contributes to genetic susceptibility shared among anxiety spectrum phenotypes. Biol Psychiatry. 2008; 64:302–310. [PubMed: 18436194]

Hodgkinson CA, Yuan Q, Xu K, Shen PH, Heinz E, Lobos EA, Binder EB, Cubells J, Ehlers CL, Gelernter J, Mann J, Riley B, Roy A, Tabakoff B, Todd RD, Zhou Z, Goldman D. Addictions biology: haplotype-based analysis for 130 candidate genes on a single array. Alcohol Alcohol. 2008; 43:505–515. [PubMed: 18477577]

Jang KL, Vernon PA, Livesley WJ. Personality disorder traits, family environment, and alcohol misuse: a multivariate behavioral genetic analysis. Addiction. 2000; 95:873–888. [PubMed: 10946437]

Jorm AF, Prior M, Sanson A, Smart D, Zhang Y, Easteal S. Association of a polymorphism of the dopamine transporter gene with externalizing behavior problems and associated temperament traits: a longitudinal study from infancy to the mid-teens. Am J Med Genet. 2001; 105:346–350. [PubMed: 11378848]

Kendler KS, Prescott CA, Myers J, Neale MC. The structure of genetic and environmental risk factors for common psychiatric and substance use disorders in men and women. Arch Gen Psychiatry. 2003; 60:929–937. [PubMed: 12963675]

Kessler RC, Adler L, Barkley R, Biederman J, Conners CK, Demler O, Faraone SV, Greenhill LL, Howes MJ, Secnik K, Spencer T, Ustun TB, Walters EE, Zaslavsky AM. The prevalence and correlates of adult ADHD in the United States: results from the National Comorbidity Survey Replication. Am J Psychiatry. 2006; 163:716–723. [PubMed: 16585449]

Kim DJ, Park BL, Yoon S, Lee HK, Joe KH, Cheon YH, Gwon DH, Cho SN, Lee HW, Namgung S, Shin HD. 5′ UTR polymorphism of dopamine receptor D1 (DRD1) associated with severity and temperament of alcoholism. Biochem Biophys Res Commun. 2007; 357:1135–1141. [PubMed: 17466946]

Knopik VS, Heath AC, Bucholz KK, Madden PA, Waldron M. Genetic and environmental influences on externalizing behavior and alcohol problems in adolescence: a female twin study. Pharmacol Biochem Behav. 2009; 93:313–321. [PubMed: 19341765]

Köhnke MD. Approach to the genetics of alcoholism: a review based on pathophysiology. Biochem Pharmacol. 2008; 75:160–177. [PubMed: 17669369]

Koob GF. Drugs of abuse: anatomy, pharmacology and function of reward pathways. Trends Pharmacol Sci. 1992; 13:177–184. [PubMed: 1604710]

Krueger RF, Hicks BM, Patrick CJ, Carlson SR, Iacono WG, McGue M. Etiologic connections among substance dependence, antisocial behavior, and personality: modeling the externalizing spectrum. J Abnorm Psychol. 2002; 111:411–424. [PubMed: 12150417]

Kuo PH, Neale MC, Riley BP, Webb BT, Sullivan PF, Vittum J, Patterson DG, Thiselton DL, van den Oord EJ, Walsh D, Kendler KS, Prescott CA. Identification of susceptibility loci for alcohol-related traits in the Irish Affected Sib Pair Study of Alcohol Dependence. Alcohol Clin Exp Res. 2006; 30:1807–1816. [PubMed: 17067344]

Lachman HM, Papolos DF, Saito T, Yu YM, Szumlanski CL, Weinshilboum RM. Human catechol-O-methyltransferase pharmacogenetics: description of a functional polymorphism and its potential application to neuropsychiatric disorders. Pharmacogenetics. 1996; 6:243–250. [PubMed: 8807664]

Le Foll B, Gallo A, Le Strat Y, Lu L, Gorwood P. Genetics of dopamine receptors and drug addiction: a comprehensive review. Behav Pharmacol. 2009; 20:1–17. [PubMed: 19179847]

Lee MS, Ryu SH. No association between the dopamine D3 receptor gene and Korean alcohol dependence. Psychiatr Genet. 2002; 12:173–176. [PubMed: 12218663]

Li Y, Shao C, Zhang D, Zhao M, Lin L, Yan P, Xie Y, Jiang K, Jin L. The effect of dopamine D2, D5 receptor and transporter (SLC6A3) polymorphisms on the cue-elicited heroin craving in Chinese. Am J Med Genet B Neuropsychiatr Genet. 2006; 141B:269–273. [PubMed: 16526040]

Lind PA, Eriksson CJ, Wilhelmsen KC. Association between harmful alcohol consumption behavior and dopamine transporter (DAT1) gene polymorphisms in a male Finnish population. Alcohol Clin Exp Res. 2009; 29:417–429.

McGeary J. The DRD4 exon 3 VNTR polymorphism and addiction-related phenotypes: a review. Pharmacol Biochem Behav. 2009; 93:222–229. [PubMed: 19336242]

Munafò MR, Yalcin B, Willis-Owen SA, Flint J. Association of the dopamine D4 receptor (DRD4) gene and approach-related personality traits: meta-analysis and new data. Biol Psychiatry. 2008; 63:197–206. [PubMed: 17574217]

Nadder TS, Rutter M, Silberg JL, Maes HH, Eaves LJ. Genetic effects on the variation and covariation of attention deficit/hyperactivity disorder (ADHD) and oppositional-defiant/conduct disorder (ODD/CD) symptomatologies across informant and occasion of measurement. Psychol Med. 2002; 32:39–53. [PubMed: 11883729]

Neville MJ, Johnstone EC, Walton RT. Identification and characterization of ANKK1: a novel kinase gene closely linked to DRD2 on chromosome band 11q23.1. Hum Mutat. 2004; 23:540–545. [PubMed: 15146457]

Olds J, Milner P. Positive reinforcement produced by electrical stimulation of septal area and other regions of rat brain. J Comp Physiol Psychol. 1954; 47:419–427. [PubMed: 13233369]

Prescott CA, Madden PA, Stallings MC. Challenges in genetic studies of the etiology of substance use and substance use disorders: introduction to the special issue. Behav Genet. 2006; 36:473–482. [PubMed: 16710779]

Prescott CA, Sullivan PF, Myers JM, Patterson DG, Devitt M, Halberstadt LJ, Walsh D, Kendler KS. The Irish Affected Sib Pair Study of Alcohol Dependence: study methodology and validation of diagnosis by interview and family history. Alcohol Clin Exp Res. 2005; 29:417–429. [PubMed: 15770118]

Rehm J, Mathers C, Popova S, Thavorncharoensap M, Teerawattananon Y, Patra J. Global burden of disease and injury and economic cost attributable to alcohol use and alcohol-use disorders. Lancet. 2009; 373:2223–2233. [PubMed: 19560604]

Schuckit MA, Smith TL, Tipp JE. The Self-Rating of the Effects of alcohol (SRE) form as a retrospective measure of the risk for alcoholism. Addiction. 1997; 92:979–988. [PubMed: 9376780]

Schultz W. Predictive reward signal of dopamine neurons. J Neurophysiol. 1998; 80:1–27. [PubMed: 9658025]

Schulz-Heik RJ, Maentz SK, Rhee SH, Gelhorn HL, Young SE, Timberlake DS, Smolen A, Stetler GL, Sakai JT, Crowley TJ. Case-control and within-family tests for an association between conduct disorder and DAT1. Psychiatr Genet. 2008; 18:17–24. [PubMed: 18197081]

Schwab SG, Franke PE, Hoefgen B, Guttenthaler V, Lichtermann D, Trixler M, Knapp M, Maier W, Wildenauer DB. Association of DNA polymorphisms in the synaptic vesicular amine transporter gene (SLC18A2) with alcohol and nicotine dependence. Neuropsychopharmacology. 2005; 30:2263–2268. [PubMed: 15988470]

Silberg J, Rutter M, Meyer J, Maes H, Hewitt J, Simonoff E, Pickles A, Loeber R, Eaves L. Genetic and environmental influences on the covariation between hyperactivity and conduct disturbance in juvenile twins. J Child Psychol Psychiatr. 1996; 37:803–816.

Sintov ND, Kendler KS, Young-Wolff KC, Walsh D, Patterson DG, Prescott CA. Empirically defined subtypes of alcohol dependence in a Irish family sample. Drug Alcohol Depend. 2009; 107:230–236. [PubMed: 20022183]

Spitzer, RL.; Williams, JBW. Structured Clinical Interview for DSM-III-R (SCID). New York: New York State Psychiatric Institute, Biometrics Research Department; 1985.

Tsai SJ, Hong CJ, Yu YW, Chen TJ. Association study of catechol-O-methyltransferase gene and dopamine D4 receptor gene polymorphisms and personality traits in healthy young Chinese females. Neuropsychobiology. 2004; 50:153–156. [PubMed: 15292670]

Tuvblad C, Zheng M, Raine A, Baker LA. A common genetic factor explains the covariation among ADHD, ODD, and CD symptoms in 9–10 year old boys and girls. J Abnorm Child Psychol. 2009; 37:153–167. [PubMed: 19015975]

Wender, PH. Minimal Brain Dysfunction in Children. New York: John Wiley & Sons, Inc.; 1971.

Wiesbeck GA, Dürsteler-Macfarland KM, Wurst FM, Walter M, Petitjean S, Müller S, Wodarz N, Böning J. No association of dopamine receptor sensitivity in vivo with genetic predisposition for alcoholism and DRD2/DRD3 gene polymorphisms in alcohol dependence. Addict Biol. 2006; 11:72–75. [PubMed: 16759339]

Young SE, Smolen A, Corley RP, Krauter KS, Defries JC, Crowley TJ, Hewitt JK. Dopamine transporter polymorphism associated with externalizing behavior in children. Am J Med Genet B Neuropsychiatr Genet. 2002; 114:144–149.

Young SE, Stallings MC, Corley RP, Krauter KS, Hewitt JK. Genetic and environmental influences on behavioral disinhibition. Am J Med Genet. 2000; 96:684–695. [PubMed: 11054778]

van der Zwaluw CS, Engels RC, Buitelaar J, Verkes RJ, Franke B, Scholte RH. Polymorphisms in the dopamine transporter gene (SLC6A3/DAT1) and alcohol dependence in humans: a systematic review. Pharmacogenomics. 2009; 10:853–866. [PubMed: 19450132]

Table 1

Power for Dichotomous Alcohol Dependence (AD) Outcome

MAF	OR	Power
0.05	1.1	0.0765
	1.2	0.1526
	1.3	0.2711
	1.4	0.4163
	1.5	0.5663
0.1	1.1	0.1006
	1.2	0.2459
	1.3	0.4552
	1.4	0.6665
	1.5	0.8271
0.2	1.1	0.1406
	1.2	0.3908
	1.3	0.6836
	1.4	0.8819
	1.5	0.9676
0.3	1.1	0.1690
	1.2	0.4817
	1.3	0.7895
	1.4	0.9447
	1.5	0.9902
0.4	1.1	0.1855
	1.2	0.5288
	1.3	0.8331
	1.4	0.9635
	1.5	0.9948

OR, odds ratio; MAF, minor allele frequency.

Table 2

Power for Continuous Outcomes

MAF	Variance	Power
0.05	0.01	0.6418
	0.02	0.9089
	0.03	0.9814
	0.04	0.9968
	0.05	0.9995
0.1	0.01	0.6418
	0.02	0.9089
	0.03	0.9814
	0.04	0.9968
	0.05	0.9995
0.2	0.05	0.6418
	0.1	0.9089
	0.2	0.9814
	0.3	0.9968
	0.4	0.9995
0.3	0.05	0.6418
	0.1	0.9089
	0.2	0.9814
	0.3	0.9968
	0.4	0.9995
0.4	0.05	0.6418
	0.1	0.9089
	0.2	0.9814
	0.3	0.9968
	0.4	0.9995
0.4	0.05	0.6418
	0.1	0.9089
	0.2	0.9814
	0.3	0.9968
	0.4	0.9995

MAF, minor allele frequency.

Table 3

Means and Standard Deviations for Disinhibitory Symptom Counts and Pearson Correlations Between Counts

	ADsx (6.39, 1.03)	ADHDsx (0.50, 0.20)	ASPDsx (2.02, 2.75)	CDsx (2.86, 2.99)	DDsx (0.82, 1.4)	NS (0.58, 0.22)
ADsx	1.00					
ADHDsx	0.18	1.00				
ASPDsx	0.20	0.36	1.00			
CDsx	0.22	0.45	0.78	1.00		
DDsx	0.13	0.19	0.34	0.38	1.00	
NS	0.18	0.31	0.18	0.22	0.19	1.00

ADsx, alcohol dependence symptoms; ADHDsx, attention deficit/hyperactivity symptoms; ASPDsx, antisocial personality disorder symptoms; CDsx, conduct disorder symptoms; DDsx, drug dependence symptoms; NS, novelty seeking scores. Means and standard deviations are listed in parentheses. ADHDsx and NS scores were rescaled such that their range is 0 to 1.

Alcohol Clin Exp Res. Author manuscript; available in PMC 2012 September 16.

Table 4

Marker Information, Nominal *p*-Values, and Effect Sizes for SNPs Significantly Associated With AD or Alcohol-Related Traits

Gene, Chr, mRNA Accession #	SNP	Alleles	bp	Location	Call rate	MAF	AD (*n* = 1,028)	Freq cases versus controls	ONSET (*n* = 436)	ISENS (*n* = 428)	TOLMX (*n* = 427)	MAX24 (*n* = 436)	WDSFS (*n* = 436)
DRD3 Chr 3 NM_000796	rs2654754	T/C	115338486	intron 5	99.8	0.025	0.30 (0.74)		1.00 (0.01)	**0.0021** * (0.24)	0.07 (0.16)	0.18 (5.34)	0.09 (0.29)
DRD4 Chr 11 NM_000797	rs12280580	C/G	616220	5′ near gene	92.7	0.354	**0.011** * (1.28)	**0.38/0.32**	0.66 (−0.24)	0.59 (0.01)	0.55 (0.02)	0.17 (1.82)	0.76 (−0.02)
DRD5 Chr 4 NM_000798	rs7655090	A/G	9374973	3′ near gene	98.1	0.049	0.60 (1.11)		0.33 (1.10)	0.63 (−0.03)	0.16 (−0.09)	0.15 (−3.96)	**0.0017** ** (−0.37)
SLC6A3 Chr 5 NM_001044	rs27048	C/T	1465645	intron 8	97.1	0.451	**0.042** * (0.68)	**0.57/0.53**	0.68 (−0.21)	0.24 (0.03)	0.61 (0.01)	0.79 (−0.31)	0.24 (0.06)
	rs10052016	A/G	1481111	intron 4	93.9	0.412	**0.00055** * (1.33)	**0.45/0.37**	0.69 (−0.19)	0.19 (0.03)	0.13 (0.04)	0.73 (0.38)	0.30 (−0.05)
TH Chr 11 NM_199292	rs11564717	C/T	2143465	intron 12	99.9	0.408	0.31 (0.68)		0.70 (−0.84)	0.20 (0.14)	0.91 (0.01)	0.05 (10.16)	**0.0094** * (0.59)

AD, alcohol dependence; ONSET, age-at-onset of AD; ISENS, initial sensitivity; TOLMX, tolerance/maximum drinking; MAX24, maximum drinks in 24 hours; WDSFS, withdrawal severity factor score; MAF, minor allele frequency. ISENS and TOLMX were log transformed. Nominal *p*-values significant after permutation are in bold.

* Empirical *p*-value ≤ 0.05;

** empirical *p*-value ≤ 0.01.

Effect sizes for *p*-values significant after permutation are listed in parentheses as odds ratios for AD and as regression coefficients for quantitative traits. An odds ratio >1 or a positive regression coefficient indicates that the minor allele (listed second under “Alleles” column) increases risk of the phenotype.

Table 5

Marker Information, Nominal *p*-Values, and Effects Sizes for SNPs Significantly Associated With Disinhibitory Symptom Counts

Gene, Chr, mRNA Accession #	SNP	Alleles	bp	Location	Call rate	MAF	ADsx (*n* = 436)	ADHDsx (*n* = 426)	ASPDsx (*n* = 436)	CDsx (*n* = 436)	DDsx (*n* = 432)	NS (*n* = 429)	Factor score (*n* = 426)
DRD5 Chr 4 NM_000798	rs7655090	A/G	9374973	3′ near gene	98.1	0.049	0.15 (−0.20)	0.08 (−0.05)	0.17 (−0.55)	0.03 (−0.89)	0.07 (−0.37)	0.38 (−0.03)	**0.0094** * (−0.34)
SLC6A3 Chr 5 NM_001044	rs6350	C/T	1496199	exon 2	99.9	0.076	**0.0021** * (−0.36)	0.94 (0.00)	0.52 (0.22)	0.80 (−0.09)	0.89 (−0.02)	0.31 (0.03)	0.89 (−0.02)
DDC Chr 7 NM_001082971	rs11575542	G/A	50305196	exon 14	99.7	0.015	0.22 (−0.22)	0.48 (−0.03)	0.54 (0.32)	0.63 (−0.26)	**0.0028** * (0.80)	0.86 (0.01)	0.52 (0.11)

ADsx, alcohol dependence symptoms; ASPDsx, antisocial personality disorder symptoms; ADHDsx, attention deficit/hyperactivity symptoms; CDsx, conduct disorder symptoms; DDsx, drug dependence symptoms; NS, novelty seeking scores; MAF, minor allele frequency. Nominal *p*-values significant after permutation are in bold.

* Empirical *p*-value ≤ 0.05.

Effect sizes for *p*-values significant after permutation are listed in parentheses as odds ratios for AD and as regression coefficients for quantitative traits. An odds ratio >1 or a positive regression coefficient indicates that the minor allele (listed second under “Alleles” column) increases risk of the phenotype.