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A Genome-Wide Association Study of Clinical Symptoms of Dissociation in a Trauma-Exposed Sample

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Abstract

Background—Recent work suggests that a subset of individuals with posttraumatic stress disorder (PTSD) exhibit marked dissociative symptoms, as defined by derealization and depersonalization. A dissociative subtype of PTSD was added to the diagnostic criteria listed in the *DSM-5* to capture this presentation of PTSD. This study examined genetic polymorphisms for association with the symptoms that define the dissociative subtype of PTSD using a genome-wide approach.

Methods—The sample was comprised of 484 white, non-Hispanic, trauma-exposed veterans and their partners who were assessed for lifetime PTSD and dissociation using a structured clinical interview. The prevalence of PTSD was 60.5%. Single nucleotide polymorphisms (SNPs) from across the genome were obtained from a 2.5 million SNP array.

Results—Ten SNPs evidenced suggestive association with dissociation ($p < 10^{-5}$). No SNPs met genome-wide significance criteria ($p < 5 \times 10^{-8}$). The peak SNP was rs263232 ($\beta = 1.4$, $p = 6.12 \times 10^{-7}$), located in the adenylyl cyclase 8 (*ADCY8*) gene; a second SNP in the suggestive range was rs71534169 ($\beta = 1.63$, $p = 3.79 \times 10^{-6}$), located in the dipeptidyl-peptidase 6 (*DPP6*) gene.

Conclusions—*ADCY8* is integral for long term potentiation and synaptic plasticity and is implicated in fear-related learning and memory and long term memory consolidation. *DPP6* is critical for synaptic integration and excitation. These genes may exert effects on basic sensory integration and cognitive processes that underlie dissociative phenomena.

Keywords

dissociative subtype; dissociation; posttraumatic stress disorder; genome-wide; gene; ADCY8; DPP6

Introduction

Recent research on the relationship between symptoms of dissociation and posttraumatic stress disorder (PTSD) has identified a distinct subset of individuals with the diagnosis who

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manifest a dissociative subtype of the disorder.[1–6] The subtype is defined primarily by symptoms of derealization (i.e., feeling as if the world is not real) and depersonalization (i.e., feeling as if oneself is not real) and is manifested in approximately 15–30% of individuals with PTSD.[3–6] In some investigations, *DSM-IV* PTSD criteria B3 (flashbacks) and C3 (psychogenic amnesia) have also been associated with the subtype.[3,6] Evidence for the subtype comes from latent profile analyses,[4–6] taxonometric analyses,[7] signal detection analyses,[8] and evaluation of the distribution of dissociative symptoms in large samples,[3,9] and has been replicated in veteran,[5,6] civilian,[4] and cross-cultural samples.[3] As a result of this psychometric and neuroimaging research (reviewed below), the dissociative subtype of PTSD was added to the *Diagnostic and Statistical Manual of Mental Disorders*, Version 5 (*DSM-5*[10]).

Evaluation of the dissociative subtype has important implications for both clinical practice and research. Clinically, dissociation is generally thought to interfere with PTSD treatment and is an important phenomenon to attend to in its own right. From a research standpoint, dissociation represents a source of heterogeneity in the clinical presentation of PTSD that may complicate the search for biomarkers, clinical correlates, and effective treatments if not taken into account. For example, findings of recent functional neuroimaging studies suggest that individuals with the dissociative subtype exhibit a different pattern of brain activation (e.g., hypoactivation of limbic brain regions and increased activation in pre-frontal regions) during exposure to traumatic memories than individuals with PTSD who do not dissociate.[1,2]

Although severity of trauma exposure is a predictor of the dissociative subtype of PTSD,[3,4,6] it is also possible that the capacity for dissociation is influenced by pre-existing biological vulnerabilities. Several studies suggest an association between stress-linked dissociative symptoms and blunted neurobiological reactivity.[11–17] In addition, a handful of small genetic studies have reported associations between candidate genes and broadly defined dissociative symptomatology. Specifically, single nucleotide polymorphisms (SNPs) in FK506 binding protein 5 (*FKBP5*), a gene involved in regulating the hypothalamic-pituitary-adrenal axis, were associated with dissociative symptoms in hospitalized children with medical injuries[18] and in women with a history of childhood trauma.[19] The serotonin transporter gene (*SLC6A4*) has been implicated in dissociation among individuals with obsessive-compulsive disorder[20] and trauma-exposed individuals.[21] Finally, the catechol-O-methyltransferase (*COMT*) gene has been associated with dissociation among children with a history of abuse.[22] These studies suggest the possibility of a genetic contribution to dissociative symptoms, but to date, genetic associations with the dissociative subtype of PTSD have not been evaluated. The aim of this study was to conduct a genome-wide search for polymorphisms associated with the symptoms of dissociation that define the subtype in a sample of trauma-exposed veterans and non-veterans. The phenotype was defined by severity scores on lifetime symptoms of derealization and depersonalization; this dimensional approach was expected to yield enhanced statistical power compared to use of a categorical designation of the phenotype because of the low prevalence of the subtype. The use of a genome-wide approach also allowed for a test of the replicability of the results of prior genetic association studies of dissociation.

Methods

Participants and Procedure

Participants were drawn from a trauma-exposed sample, as described by Logue et al.[23] Briefly, the study included 852 U.S. military veterans and their intimate partners who participated in one of two studies on the genetics of posttraumatic psychopathology and couple conflict behavior. Ancestry was determined using 10,000 randomly chosen

polymorphisms with minor allele frequency (MAF) > .05 using the program STRUCTURE.[24,25] which uses a Bayesian clustering analysis to determine ancestry. The largest subpopulation identified through this procedure (n = 540) self-reported their racial background as white, non-Hispanic. Of these, 491 participants (364 veterans and 127 non-veteran partners) had been exposed to a *DSM-IV* Criterion A[26] trauma, as determined by the Clinician Administered PTSD Scale (CAPS), [27] a validated, semi-structured PTSD interview; 484 had scores on the CAPS interview items assessing derealization and depersonalization and were included in these analyses. In this final sample, 170 were women and 314 were men with a mean age of 52 (range: 21–75); 60.5% of the sample met criteria for a lifetime diagnosis of PTSD (247 veterans and 45 non-veteran partners). All interviews were administered by clinicians with either an MA or PhD in psychology, and were digitally recorded for the purposes of evaluating diagnostic reliability (see below) and maintaining quality control. The Principal Investigator of the study (MWM) oversaw weekly meetings of interviewers to review videotapes, discuss diagnostic concerns, and prevent rater drift. Veterans and their non-veteran partners took part in identical but separate assessment procedures. Partners included in this study had trauma exposure were not necessarily "controls."

Measures

The Clinician Administered PTSD Scale (CAPS)—The CAPS,[27] the gold-standard structured diagnostic interview for PTSD, was used to assess PTSD diagnostic status, PTSD severity, and dissociation severity. Items measure the frequency and intensity of each of the 17 *DSM-IV* PTSD symptoms on a 0–4 scale for a total possible item severity score of 0–8 for each symptom. In addition, the CAPS includes items that assess associated features of PTSD, as listed in the *DSM-IV*. These include two items that assess derealization and depersonalization, which have been used previously to define the dissociative subtype of PTSD.[4–6] For the purposes of this study, the severity scores for each of these two items were combined to form a dimensional dissociation severity score with a possible range of 0–16. Inter-rater agreement (intra-class correlation coefficients) between primary and secondary ratings of ~30% of the diagnostic interview video recordings was high for the dimensional lifetime PTSD severity scores (ICC = .97) and for the dimensional lifetime dissociation severity scores (ICC = .93). Kappa for the lifetime PTSD diagnosis was .87.

Genotyping

DNA was isolated from peripheral blood samples on a Qiagen AutoPure instrument with Qiagen reagents and normalized using PicoGreen assays (Invitrogen, Grand Island, NY, USA). Genotyping was performed on an Illumina OMNI 2.5--8 array and scanned using an Illumina HiScan System (San Diego, CA, USA) according to the manufacturer's specifications. All SNP locations were from the hg19 human-genome assembly (February 2009). Details on call rates, quality control, and elimination of samples are provided in Logue et al.[23] SNPs were eliminated from the analysis if their MAF was < 5% or if they failed a test of Hardy Weinberg equilibrium ($p \leq .000001$) leaving a total of 1,197,702 SNPs for analysis. We also evaluated the possibility of population substructure within the Caucasian sample by using principal components (PC) analysis of 10,000 randomly selected genetic markers (with MAF > .05) in the program EIGENSTRAT.[28] We then took the top 10 PCs from that analysis and entered them into a multiple regression predicting dissociation severity and found no evidence for dissociation-associated population substructure (overall model $F = 1.26$, $p = .25$).

Statistical Analyses

All genetic association analyses were performed using the program PLINK.[29] Quantitative trait analysis was conducted, using the PLINK --linear option, and asymptotic *p*-values were obtained via a Wald test. Genome-wide level of significance was set at $p < 5 \times 10^{-8}$, and suggestive evidence for association was defined by $p < 10^{-5}$. Follow-up analyses that examined the possible moderating role of biological sex were conducted via linear regression and the --interaction command in PLINK. In addition, we followed up on our main results by controlling for the effects of PTSD severity. Finally, to permit comparison with prior work, we examined if SNPs in any genes previously associated with dissociation (*FKBP5*, *SLC6A4*, *COMT*) reached gene-wide level of significance using the max(T) permutation procedure with 5000 replications to calculate *p*-values corrected for the number of SNPs evaluated on each gene (to allow for a more equitable comparison to prior candidate gene studies). Linkage disequilibrium (LD) was evaluated for pairs of SNPs in PLINK.

Results

The mean dissociation severity score was 1.13 (*SD* = 2.59, range: 0 – 15). The majority of participants (78%) evidenced no symptoms of derealization or depersonalization. The number of participants with lifetime PTSD who met presumptive criteria for the dissociative subtype was 19%, as defined by a CAPS frequency score of one or greater and intensity score of two or greater on either the derealization or depersonalization item; these cut-points are frequently used to determine the presence of each PTSD symptom on the CAPS.[30] A chi-square analysis revealed that the prevalence of the subtype did not differ between men (20.3%) and women (18.0%; $p = .55$).

Ten SNPs showed associations with dissociative symptom severity in the suggestive range of $p < 10^{-5}$. No SNPs were genome-wide significant ($p < 5 \times 10^{-8}$). The Manhattan Plot showing the negative log-transformed *p*-values from the genome-wide analysis of dissociation symptom severity is shown in Figure 1; additional details are provided in Table 1.

The SNP with the strongest association with dissociation, rs263232 ($\beta = 1.4$, $p = 6.12 \times 10^{-7}$), is located at 131,808,169 base pair (bp) on chromosome 8 in the adenylyl cyclase 8 (*ADCY8*) gene at 131,792,547 – 132,054,672 bp. In our sample, the MAF of this SNP was 8.9%. Evaluation of the distribution of dissociation severity scores as a function of genotype yielded no evidence of outliers. Approximately 17% of individuals with zero copies of the risk allele had positive scores on dissociation compared to approximately 34% of individuals with one or two copies of the risk allele. The effect of the SNP remained suggestive ($p = 5.66 \times 10^{-7}$) after controlling for PTSD severity and was not significantly modified by sex ($p = .09$). Follow-up evaluation of *ADCY8* revealed four other SNPs (rs4339660, rs7015079, rs263238, rs263234) that were nominally ($p < .05$) associated with dissociation severity (smallest $p = .01$).

A second SNP in the suggestive range was rs71534169 ($\beta = 1.63$, $p = 3.79 \times 10^{-6}$) located at 154,592,866 bp on chromosome 7 in the dipeptidyl-peptidase 6 (*DPP6*) gene located at 153,584,182–154,685,995 bp. In our sample, the MAF of this SNP was 6.5%. Thirty-eight additional SNPs in the gene evidenced nominally significant ($p < .05$) associations with dissociation (details from first author). Approximately 82% of individuals with no copies of the risk allele scored 0 on dissociation severity compared to 67% of individuals with 1 copy of the risk allele (no participants had two copies of the risk allele). The effect of the SNP remained significant ($p = 3.96 \times 10^{-5}$) after controlling for PTSD severity, and there was a

significant SNP X sex interaction (β = −2.34, p = .002). Stratifying by sex showed the effect to be stronger among women (β = 3.26, $p = 6.96 \times 10^{-7}$) than men (β = .93, p = .03).

A number of other SNPs showed suggestive evidence of association with dissociation (see Table 1): three in *chromosome 8 open reading frame 79* (*KIAA1456*), which ranged in their degree of linkage disequlibrium with each other (r^2 range: .32 – .95, *D*' range: .74 – 1.0); one in *K(lysine) acetyltransferase 2B* (*KAT2B*) on chromosome 3; one in *microRNA 3166* (*MIR3166*); and three that were intergenic. In follow-up analyses, the association between these eight SNPs and dissociation severity remained significant after covarying for PTSD severity (with PTSD severity in the model, the largest SNP p-value was p = .0002 for rs77006546 and the smallest SNP p-value was $p = 6.99 \times 10^{-7}$ for rs682457). Four of these SNPs showed suggestive evidence of an association with sex, such that the effects were greater in women compared to men. Specifically, suggestive sex X SNP effects were found for rs9725031 ($\beta_{interaction} = -1.48$, p = .004; $\beta_{men} = 0.57$, p = .07; $\beta_{women} = 2.05$, $p = 1.17 \times 10^{-6}$), and for the three SNPs in *KIAA1456:* rs62488971 ($\beta_{interaction} = -2.44$, p = .0008; $\beta_{men} = 0.70$, p = .11; $\beta_{women} = 3.14$, $p = 2 \times 10^{-7}$); rs2460905 ($\beta_{interaction} = -1.72$, p = .005; $\beta_{men} = .75$, p = .05; $\beta_{women} = 2.46$, $p = 1.44 \times 10^{-6}$); and rs2466273 ($\beta_{interaction} = -1.63$, p = .007; $\beta_{men} = .74$, p = .04; $\beta_{women} = 2.36$, $p = 3.00 \times 10^{-6}$).[1]

Finally, we evaluated SNPs in the three genes that have shown association with dissociation previously. There were 30 SNPs included in the analysis of *FKBP5* and of these, three were nominally significant (smallest uncorrected p-value = .01) but none withstood gene-wide permutation correction. Further, these three did not overlap with *FKBP5* SNPs that have previously shown association with dissociation[18] or PTSD.[32] Thirteen SNPSs in *SLC6A4* were evaluated and none showed significant association with dissociation (all $p > .05$). There were 29 SNPs in *COMT* that were evaluated and one (rs9306235) showed a nominally significant association (p = .04) that did not withstand gene-wide permutation testing (p = .42).

Discussion

We conducted a genome-wide association study to evaluate genetic predictors of derealization and depersonalization symptoms in a trauma-exposed sample with a high prevalence of PTSD. To our knowledge, this is the first genome-wide association test for dissociation symptom severity, as prior work has used a candidate gene approach to evaluate genes commonly examined across psychiatric phenotypes.[18–22] Although no SNPs in this study met standard criteria for genome-wide level of statistical significance, ten SNPs were in the "suggestive" range. Two SNPs were located in genes that have clear potential functional significance for dissociative symptomatology and have been associated with other psychiatric conditions in the past.

The peak SNP, rs263232, was located in *ADCY8* on chromosome 8. *ADCY8* has also been associated with bipolar disorder[33–35] (see also Avramopoulos et al.[36]), and depression comorbid with alcohol dependence in women.[37] *ADCY8* is expressed pre-snyaptically in many areas of the brain, including the cortex, hippocampus, amygdala, thalamus, hypothalamus, and cerebellum.[38] It produces the enzyme adenylyl cyclase (AC8), one of two Ca^{2+}/calmodulin sensitive adenylyl cyclase isoforms that catalyze the conversion of

[1]As dissociation is thought to be related to severe and early childhood trauma, we also examined the main effect of each of the 10 SNPs in the suggestive range of association while controlling for the number of reported instances of childhood sexual trauma occurring before age 13 as assessed by the Traumatic Life Events Questionnaire.[31] The main effect of all SNPs remained suggestive (i.e., $p < 10^{-5}$) with the exception of the three SNPS in *KIAA1456* and the intergenic SNPs rs77006546 and rs9725031. In these cases, the p-value for the SNP main effect fell just outside the cut-off for a suggestive association (p-value ranged from 1.04×10^{-5} to 2.01×10^{-5}).

ATP to cyclic AMP, and thus impacts the same intracellular second messenger system as the pituitary adenylate cyclase-activating polypeptide (*PACAP*) gene recently associated with PTSD in women.[39]

Cyclic AMP is integral to long term potentiation, synaptic plasticity, and hence learning and memory. In the hippocampus, levels of cyclic AMP determine the basal balance between silenced and active synapses; and in AC8 deficient mice, rapid resetting of strongly depolarized presynaptic terminals is prevented. The adenylate cyclase signaling cascade is also critical to central and peripheral hypothalamic-pituitary-adrenal (HPA) axis regulation.

The functional impact of AC8 deficiency in rodents appears to be consistent with these mechanisms (Table 2; also see review by Wang & Zhang [40]). Such rodents are hyper-locomotive and appear to be insensitive to novelty, risk, context, and experience. They also show marked deficits in retention of conditioned fear, reductions in phosphorylated cyclic AMP response element binding protein (pCREB), differences in gene expression at time points relevant to memory consolidation and retention, and dysregulation of the HPA axis.

Failure to encode memories associated with intense emotion or stress has clear relevance to the dissociative subtype of PTSD as the *DSM-IV* PTSD criterion C3, psychogenic amnesia, has been uniquely associated with the subtype.[3,6] Some conceptualize dissociation in PTSD as an unconscious form of emotion avoidance that occurs in response to overwhelming fear, hyperarousal, or anxiety; although speculative, it is also conceivable that dissociation could occur as a neurobiological consequence of hyperarousal in the context of an AC8 deficiency that prevents the encoding and consolidation of contextual information.

Another SNP in the suggestive range of association with dissociation was rs71534169, which is located in the *DPP6* gene on chromosome 7. This effect was stronger in women relative to men, suggesting possible sex-specific pathways in the development and manifestation of dissociative symptoms. *DPP6* has most frequently been studied in relation to multiple sclerosis[41] and amyotrophic lateral sclerosis.[42,43] It has also shown association with autism.[44] The DPP6 protein is an auxillary subunit of voltage-gated potassium-4 channels that influence neuronal excitability and communication of excitability to distal dendrites. DPP6 is involved in regulating the Atype K+ current gradient, which regulates dendritic excitability. Hippocampal recordings from *DPP6* knock-out mice demonstrate a decrease in this gradient and increased dendritic excitability.[45] Given the role of DPP6 in synaptic integration, it is possible that this protein also plays a role in dissociation, a state defined by poor integration of incoming sensory experiences and problems with region-specific cognitive processes that are ordinarily organized dynamically across time.

In addition, three SNPs in *KIAA1456* and one in *KAT2B* showed suggestive evidence of association with dissociation; the effect of the *KIAA1456* SNPs was stronger in women relative to men and was weakened when exposure to childhood sexual abuse was controlled. The *KIAA1456* gene encodes a methyltransferase, thus potentially implicating dysregulation of epigenetic processes in the dissociative subtype of PTSD. In a recent *in situ* hybridization gene expression study, the density of *KIAA1456* was found to be selectively lower in layers I, II, III, and V of Brodman's area 9, but not area 46, of the dorsolateral prefrontal cortex in individuals with schizophrenia compared to controls.[46] *KAT2B* encodes a histone/protein acetylase and has been implicated in DNA repair, replication, and transcription,[47–49] global metabolic regulation,[50] and autophagy,[51] a cellular process by which cells recycle or dispose of internal constituents to maintain normal structure and function under conditions of stress (e.g., nutrient deprivation, hypoxia, and oxidative stress).[52] To our knowledge, *KAT2B* has not previously been associated with a psychiatric phenotype. More extensive

study of both *KIAA1456* and *KAT2B* is needed to confirm their role and specific mechanistic contributions to dissociation following trauma exposure.

The association between these SNPs and dissociation remained strong even after controlling for PTSD severity, suggesting that the association was not driven by the core set of PTSD symptoms. The genes implicated here are also distinct from those identified previously in studies of PTSD. The first genome-wide association study of PTSD[23] (evaluated in this same sample) suggested that the retinoid-related orphan receptor alpha (*RORA*) gene was associated with PTSD at a genome-wide level of significance and a subsequent genome-wide investigation implicated the tolloid-like 1 gene in PTSD.[53] Prior candidate gene work in PTSD in adults has implicated a variety of other genes including steroid 5-α-reductase type 2 (*SRD5A2*[54]), *SLC6A4*,[55–57] pituitary adenylate cyclase-activating polypeptide and its receptor (*ADCYAP1* and *ADCYAP1R1*, respectively[39]), *FKBP5*,[32] and catechol-o-methyltransferase (*COMT*[58]), among others (for a recent review see Cornelis et al.[59]).

Some of these genes implicated in PTSD have also been implicated in candidate gene studies of dissociation. We did not replicate prior findings in smaller samples on the role of *SLC6A4*, *FKBP5*, or *COMT* in dissociation.[18–22] Sample size, age and developmental stage of participants, differences in the measurement of dissociation, differences in trauma exposure history, potential exposure to injury or oxidative stress due to substance abuse or other factors, and use of medications that may compensate or correct for particular genetic contributions to the dissociative phenotype could contribute to discrepancies in findings across studies.

Limitations

The results of this study should be interpreted in light of its limitations. First, the sample size of 484 was relatively small for a genome-wide association study, and there is always the risk that the results obtained here will not replicate in future research. Further, none of the SNPs met the strict level of significance needed to claim genome wide significance; they were all in the “suggestive” range and should be considered as such. Future work will need to evaluate the replicability of these results. In addition, our investigation was limited to white, non-Hispanic, trauma-exposed individuals, and it is unclear if these findings will generalize to other racial or demographic groups. We also were under-powered to fully evaluate potential sex differences in the SNP-dissociation relationships, and these results should be interpreted with caution. We did not examine potential SNP by trauma (i.e., gene by environment) effects given our focus on posttraumatic dissociation in a traumatized sample; it is unknown if these results generalize to dissociative disorders among non-traumatized populations or to trait dissociation. Related to this, it is possible that the assessment of dissociation was subject to recall bias in that participants may have attributed their dissociative symptoms to trauma even if they pre-dated trauma exposure. We focused on a dimensional phenotype, rather than a categorical one, in order to maximize the likelihood of having adequate power to detect small effects. Not all participants met full criteria for a lifetime diagnosis of PTSD, which could obscure findings between those with the dissociative subtype of PTSD from those without. Finally, it is not possible to know from this research whether the SNPs identified in association with the dissociative phenotype increase or decrease function of the genes in which they are embedded. Future basic and clinical studies will be needed to define their precise physiological and clinical impact.

Concerns about the lack of a replication sample are offset by the strengths of the study including our focus on a novel and highly specific psychiatric phenotype that has not been evaluated in genetic investigations to date. In addition, prior work examining the genetics of dissociation has been based on very small sample sizes. The current study represented an improvement upon prior ones as the sample size was larger and the genetic analyses were

genome-wide. An additional strength of the study was its use of structured diagnostic interviews conducted by highly trained interviewers who showed good inter-rater reliability.

Conclusions

In conclusion, this was the first genetic association study of the primary symptoms that define the dissociative subtype of PTSD. Results revealed that SNPs in two genes of interest that are relevant to fear-related memory formation and processing (rs263232 in *ADCY8*), and synaptic integration (rs71534169 in *DPP6*) reached a "suggestive" level of genome-wide significance in their association with symptoms of derealization and depersonalization. These SNPs were still strongly associated with dissociative phenomena after PTSD severity was controlled for, suggesting that the effects were specific to dissociation. Failure to identify individuals with the dissociative subtype of PTSD may hinder efforts to identify the biological and etiological underpinnings of PTSD. Results of this study highlight the importance of using empirically refined phenotypes in genetic association studies.

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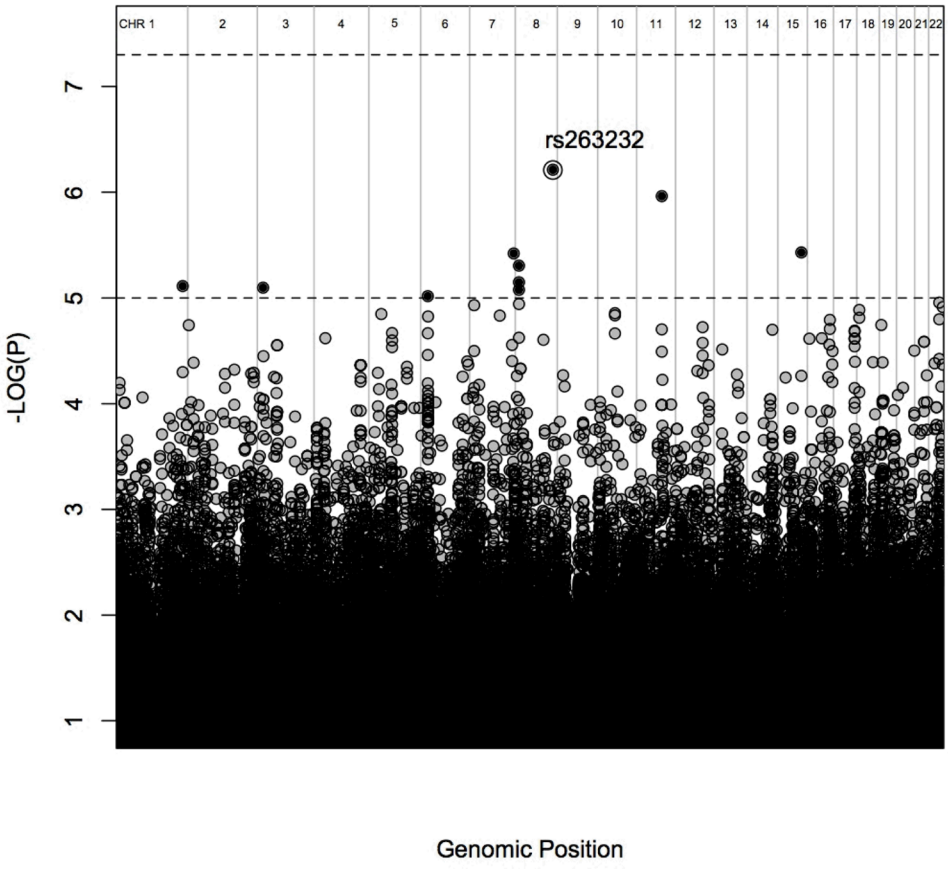

Figure 1.
Manhattan plot of genome-wide association results for dissociation severity. The bottom dashed lined represents suggestive level of statistical significance ($p < 10^{-5}$) and the top dotted lined represents genome-wide level of statistical significance ($p < 5 \times 10^{-8}$). The peak SNP is circled.

Table 1

SNPs showing association with Dissociative Symptom Severity at $p < 10^{-5}$

CHR	SNP	bp	BETA	SE	R^2	*p*	Gene	Other Info
1	rs9725031	232444455	1.13	0.25	0.04	7.72E^{-06}	intergenic	Closest gene is *SIPA1L2*
3	rs1915919	20083881	0.74	0.16	0.04	7.99E^{-06}	*KAT2B*	
6	rs77006546	25219582	1.70	0.38	0.04	9.60E^{-06}	intergenic	Near *CMAHP*
7	rs71534169	154592866	1.63	0.35	0.04	3.79E^{-06}	*DPP6*	
8	rs62488971	12808108	1.58	0.35	0.04	8.37E^{-06}	*KIAA1456*	
8	rs2460905	12834600	1.38	0.30	0.04	4.95E^{-06}	*KIAA1456*	
8	rs2466273	12866004	1.31	0.29	0.04	7.10E^{-06}	*KIAA1456*	
8	rs263232	131808169	1.43	0.28	0.05	6.12E^{-07}	*ADCY8*	
11	rs682457	87949532	1.11	0.22	0.05	1.09E^{-06}	*MIR3166*	microRNA gene
15	rs77963519	82193846	1.56	0.33	0.04	3.71E^{-06}	intergenic	Closest gene is *MEX3B*

Note. SNPs are listed by chromosome and bp. CHR = chromosome, SNP = single nucleotide polymorphism, bp = base pair, SE = standard error, Info = information, *SIPA1L2* = signal-induced proliferation-associated 1 like 2; *KAT2B* = K(lysine) acetyltransferase 2B, *CMAHP* = cytidine monophospho-N-acetylneuraminic acid hydroxylase, pseudogene, *DPP6* = dipeptidyl-peptidase 6, *KIAA1456* = chromosome 8 open reading frame 79, *ADCY8* = adenylate cyclase 8, *MIR3166* = microRNA 3166, *MEX3B* = mex-3 homolog B.

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Table 2

Functional Characteristics ADCY8 Knock-Out Mice

Study	Pertinent Findings
Wong et al., (1999)[60]	ADC8 DKO mice exhibited impaired long-term fear memory.
Schaefer et al., (2000)[61]	ADC8 KO mice showed increased locomotion and more risk taking behavior in a plus maze, even after restraint stress, compared to WT mice. They failed to demonstrate longterm depression (LTD) and exhibited decreased pCREB expression after restraint stress in the CA1 field of the hippocampus. They were lighter than WT mice but consumed a comparable number of calories. After chronic stress, they showed increased basal corticosterone levels and blunted corticosterone responses to a stressor.
Li et al., (2006)[62]	ADC8 KO mice showed diminished morphine tolerance; ADC8/ADC1 DKO mice showed decreased morphine tolerance, withdrawal, hyperlocomotion, conditioned place preference, and pCREB in VTA.
Krishnan et al., (2008)[63]	ADC8 KO mice habituated more quickly to a novel environment, swam longer in a forced swim test, and were more socially interactive with unfamiliar rodents than WT mice. Paradoxically, they showed reduced body weight and decreased sucrose preference and fluid intake. They had lower pERK1/2 in hippocampus, but increased BDNF and neurotrophin signaling in nucleus accumbens.
de Mooij-van Malsen et al., (2009)[34]	A chromosome substitution mouse strain showed increased avoidance behavior linked to chromosome 15 upon which the AC8 gene is located and manifest increased AC8 expression in the hypothalamus and piriform cortex.
Razzoli et al., (2010)[64]	ADC8 KO mice showed increased home box locomotion and climbing during the dark period, were slower to enter a dark compartment after restraint stress, and swam longer in the forced swim test. They also had adrenal hypertrophy, decreased leptin, and otherwise lighter organ weights.
Wieczorek et al., (2010)[65]	ADC8/ADC1 DKO mice showed marked deficits in long-term, but not short term conditioned fear, and marked changes in gene expression at time points relevant to memory consolidation and retention. Selective restoration of forebrain AC8 expression restored memory in DKO animals.
Wieczorek et al., (2012)[66]	ADC8/ADC1 DKO mice evidenced memory deficits that were experience/context independent. They also showed reductions in proteins that are markers for neurotransmission and synaptic plasticity after conditioned fear trials.
Zhang et al., (2011)[67]	ADC8 DKO mice exhibited impaired spatial memory.

Note. ADCY8: Adenylyl cyclase isoform 8 gene; AC8: adenylyl cyclase isoform 8; KO: knock-out; DKO: ADC8/ADC1 double knock-out; WT: wild-type; pCREB: phosphorylated cyclic-AMP response element binding protein; phospoERK1/2: phosphorylated extracellular-signal-regulated kinase type 1/2.