Genome-wide association study of Borderline Personality Disorder reveals genetic overlap with Bipolar Disorder, Major Depression and Schizophrenia

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Abstract

Borderline personality disorder (BOR) is determined by environmental and genetic factors, and characterized by affective instability and impulsivity, diagnostic symptoms also observed in manic phases of Bipolar Disorder (BIP). Up to 20% of BIP patients show comorbidity with BOR. This report describes the first case-control genome-wide association study (GWAS) of BOR, performed in one of the largest BOR patient samples worldwide. The focus of our analysis was: (i) to detect genes and gene-sets involved in BOR; and (ii) to investigate the genetic overlap with BIP. As there is considerable genetic overlap between BIP, Major Depression (MDD) and Schizophrenia (SCZ) and a high comorbidity of BOR and MDD, we also analyzed the genetic overlap of BOR with SCZ and MDD. GWAS, gene-based tests, and gene-set-analyses were performed in 998 BOR patients and 1,545 controls. LD score regression was used to detect genetic overlap between BOR and these disorders. Single marker analysis revealed no significant association after correction for multiple testing. Gene-based analysis yielded two significant genes: *DPYD* ($p=4.42x10^{-7}$) and *PKP4* ($p=8.67x10^{-7}$); and gene-set-analysis yielded a significant finding for exocytosis (GO:0006887, $p_{FDR}=0.019$). Prior studies have implicated *DPYD*, *PKP4* and exocytosis in BIP and SCZ. The most notable finding of the present study was the genetic overlap of BOR with BIP ($r_g=0.28$ [$p=2.99x10^{-3}$]), SCZ ($r_g=0.34$ [$p=4.37x10^{-5}$]), and MDD ($r_g=0.57$ [$p=1.04x10^{-3}$]). Our study is the first to demonstrate that BOR overlaps with BIP, MDD and SCZ on the genetic level. Whether this is confined to transdiagnostic clinical symptoms should be examined in future studies.

Introduction

Borderline Personality Disorder (BOR[1]) is a complex neuropsychiatric disorder with a lifetime prevalence of around 3% (1). Untreated cases often have a chronic and severely debilitating clinical course (1). BOR affects up to 20% of all psychiatric inpatients, and is associated with high healthcare utilization. BOR therefore represents a substantial socio-economic burden (2, (3).

BOR is characterized by affective instability, emotional dysregulation, and poor interpersonal functioning (3). Suicide rates in BOR range between 6% and 8%, and up to 90% of patients engage in non-suicidal self-injurious behavior (4). Other prototypical features include high-risk behaviors and impulsive aggression. Current theories view dysfunctions in emotion processing, social interaction, and impulsivity as core psychological mechanisms of BOR (5).

To date, genetic research into BOR has been limited. Available genetic studies have involved small samples and focused on candidate genes, while no genome-wide association study (GWAS) of BOR patients has yet been performed (6). However, Lubke et al. (2014) conducted a GWAS of borderline personality features using data from three cohorts comprising n=5,802, n=1,332 and n=1,301 participants, respectively (7). Using the borderline subscale of the Personality Assessment Inventory (PAI-BOR), four Borderline personality features (affect instability, identity problems, negative relations, and self-harm) were assessed. The most promising signal in the combined analysis of two samples was for seven SNPs in the gene *SERINC5*, which encodes a protein involved in myelination. Two of the SNPs could be replicated in the third sample. Interestingly, here, the effect was highest for the affect instability items, i.e., features that are key characteristics of manic phases of Bipolar Disorder (BIP).

Understanding of the pathogenesis of BOR remains limited. Both environmental and genetic factors are known to play a role in BOR etiology. Familial aggregation has been demonstrated (8, (9), and heritability estimates from twin studies range from 35% to 65%, with higher heritability estimates being obtained with self-ratings (10, (11, (12).

[1] For the sake of readability, we have decided to use the rather unconventional abbreviation “BOR” for Borderline Personality Disorder and the abbreviation “BIP” for Bipolar Disorder.

The potential comorbidity between BOR and BIP is part of an ongoing debate. For example, Fornaro *et al.* (2016) report substantial comorbidity of ~20% with BIP (13), whereas Tsanas *et al.* (2016) find clear symptomatic differences between these two diagnostic groups. BOR displays an overlap of some symptoms with BIP, such as affective instability (14). In contrast, features such as dissociative symptoms, a feeling of chronic emptiness, and identity disturbances are specific to BOR (15). To date, no twin or family study has generated conclusive results concerning a genetic overlap between the two disorders (16, (17). However, a twin study (18) and a large population based study using polygenic risk score analyses (19) indicate a genetic overlap between Borderline personality features and neuroticism, an established risk factor for BIP and other psychiatric disorders (20).

The present study represents the first case-control GWAS in BOR, and was performed in one of the largest BOR patient samples worldwide. Given the limited heritability and the expected complex genetic architecture of BOR, the sample is too small to generate significant results for single markers. Instead, the main aim of the investigation was to detect: (i) genes and gene-sets with a potential involvement in BOR; and (ii) potential genetic overlap with BIP. As a substantial overlap of common risk variants exists between BIP and Schizophrenia (SCZ), and to a lesser extent between BIP and Major Depressive Disorder (MDD), and as there is also a high comorbidity of BOR and MDD, a further aim of the study was to determine whether any observed genetic overlap between BOR and BIP, MDD and SCZ was driven by disorder-specific genetic factors using LD-score regression and polygenic risk scores (PRS).

Materials and Methods

Participants

The present sample comprised 1,075 BOR patients and 1,675 controls (21). All participants provided written informed consent prior to inclusion. The study was approved by the respective local ethics committees.

Patients were recruited at the following German academic institutions: Department of Psychosomatic Medicine, Central Institute of Mental Health, Mannheim (n=350); Department of Psychiatry and Psychotherapy, University Medical Center Mainz (n=231); and the Department of Psychiatry, Charité, Campus Benjamin Franklin, Berlin (n=494). Inclusion criteria for patients were: age 16 to 65 years; Central European ancestry; and a lifetime DSM-IV diagnosis of BOR. The control sample comprised 1,583 unscreened blood donors from Mannheim, and 92 subjects recruited by the University Medical Center Mainz.

Clinical assessment

Diagnoses of BOR were assigned according to DSM-IV criteria and on the basis of structured clinical interviews. The diagnostic criteria for BOR were assessed using the German version of the IPDE (22) or the SKID-II (23). All diagnostic interviews were conducted by trained and experienced raters. BOR patients with a comorbid diagnosis of BIP or SCZ assessed with SKID-I (23) were excluded.

Genotyping

Automated genomic DNA extraction was performed using the chemagic Magnetic Separation Module I (Chemagen Biopolymer-Technologie AG; Baesweiler; Germany). Genotyping was performed using the Infinium PsychArray-24 Bead Chip (Illumina, San Diego, USA).

Quality control and Imputation

A detailed description of the quality control and imputation procedures is provided elsewhere (24).

Briefly, quality control parameters for the exclusion of subjects and single nucleotide polymorphisms (SNPs) were: subject missingness>0.02; autosomal heterozygosity deviation (|Fhet|>0.2); SNP missingness>0.02; difference in SNP missingness between cases and controls>0.02; and SNP Hardy-Weinberg equilibrium ($p<10^{-6}$ in controls; $p<10^{-10}$ in cases).

Genotype imputation was performed using the pre-phasing/imputation stepwise approach in IMPUTE2/SHAPEIT (default parameters and a chunk size of 3 Mb) (25, (26), using the 1000 Genomes Project reference panel (release "v3.macGT1") (27).

Relatedness testing and population structure analysis were performed using a SNP subset that fulfilled strict quality criteria (INFO>0.8, missingness<1%, minor allele frequency (MAF)>0.05), and which had been subjected to linkage disequilibrium (LD) pruning (r^2>0.02). This subset comprised 63,854 SNPs. In cryptically related subjects, one member of each pair (-hat>0.2) was removed at random following the preferential retention of cases over controls. Principal components (PCs) were estimated from genotype data (see Supplementary Figure 1-6), and phenotype association was tested using logistic regression. The impact of the PCs on genome-wide test statistics was assessed using λ.

Association Analysis

Including the first four PCs as covariates, an additive logistic regression model was used to test single marker associations, as implemented in PLINK (28). The p-value threshold for genome-wide significance was set at $5x10^{-8}$.

Gene-based-analysis

To determine whether genes harbored an excess of variants with small p-values, a gene-based test was performed with MAGMA Version 1.04 (http://ctg.cncr.nl/software/magma) (29) using genotyped markers only, filtered with a MAF>1% (n=284,220). This test uses summary data, and takes LD between variants into account. SNPs within +/-10 kb of the gene boundary were assigned to each gene. Obtained p-values were Bonferroni-corrected for the number of tested genes (n=17,755, $p=2.8x10^{-6}$).

Gene-set-analysis

Gene-set-based analysis was implemented using genotyped markers only, filtered as above. As in the gene-based-analysis, SNPs within +/-10 kb of the gene boundary were assigned to

each gene. Gene-set-analyses were carried out using Gene Ontology (GO, http://software.broadinstitute.org/gsea/msigdb/) terms.

Discovery gene-set-based-analysis was carried out using i-GSEA4GWASv2 (http://gsea4gwas-v2.psych.ac.cn/) (30). The size of the gene-sets was restricted to 20-200 genes, and the major histocompatibility complex (MHC) region was excluded. In total 674 gene-sets were tested. Results were adjusted for multiple testing using false discovery rate (FDR). To validate the significant finding, the respective gene-set was investigated with i.) GSA-SNP, using the p-value of the second-best SNP in each gene (https://gsa.muldas.org) (31), and ii.) MAGMA using summary data and a nominal p-value threshold of $p<0.05$.

LD-Score Regression

To investigate a possible genetic overlap between BOR and SCZ, BIP and MDD, LD-score regression was performed (32). Genetic correlations between BOR and (i) BIP, (ii) SCZ, and (iii) MDD were calculated (33) using the result files of the Psychiatric Genomics Consortium (PGC) metaanalyses for SCZ (33,640 cases & 43,456 controls) (34), BIP (20,352 cases & 31,358 controls) (35) and MDD (16,823 cases & 25,632 controls) (35). There was no overlap in cases or controls of the present BOR GWAS sample with the PGC samples.

Polygenic Risk Score

To determine the impact of polygenic risk on BOR and subgroups (i.e., BOR with and without MDD), polygenic risk scores (PRS) were calculated for each subject based on the above-mentioned PGC datasets.

To obtain a highly informative SNP set with minimal statistical noise, the following were excluded: low frequency SNPs (MAF<0.1); low-quality variants (imputation INFO<0.9) and indels. Subsequently, these SNPs were clumped discarding markers within 500 kb of, and in high LD ($r2\geq0.1$) with, another more significant marker. From the MHC region, only one variant with the strongest significance was retained. PRS were calculated as described elsewhere (36). This involved p-value thresholds $5x10^{-8}$, $1x10^{-6}$, $1x10^{-4}$, 0.001, 0.01, 0.05,

0.1, 0.2, 0.5, and 1.0, and multiplication of the natural logarithm of the odds ratio of each variant by the imputation probability for the risk allele. The resulting values were then totaled. For each subject, this resulted in one PRS for SCZ, MDD, and BIP for each p-value threshold.

In a first step, the association of the PRS for BIP, SCZ, and MDD with BOR case control status was analyzed using standard logistic regression and by including the four PCs as covariates. For each p-value threshold, the proportion of variance explained (Nagelkerke's R^2) in BOR case-control status was computed by comparison of a full model (covariates + PRS) score to a reduced model (covariates only).

For further exploratory analysis, the $p<0.05$ PRS for each disorder was selected (i.e. including all markers that reached nominal significance in the training samples). To determine whether the different scores contribute independently to the case-control status, a regression including the PRS for MDD, SCZ, and BIP and the four PCs was computed. In a secondary analysis, two further models were computed. These included the PRS for BIP and the PRS of either MDD or SCZ, while controlling for the four PCs.

Furthermore, PRS were analyzed by differentiating between controls, and patients with or without comorbid MDD. For each PRS, a linear model was computed using the PRS as a dependent variable, disease state as an independent variable, and the four PCs as covariates. Differences between groups were assessed using post-hoc tests (Bonferroni-corrected).

Results

Sample characteristics

Genetic quality control led to the exclusion of 207 subjects. Reasons for exclusion were: (i) insufficient data quality (low call rate), n=6; (ii) relatedness, n= 63; and (iii) population outlier status, n= 138. After quality control, the sample comprised 998 BOR cases (914 female / 84 male) and 1,545 controls (868 female / 677 male). Mean age for cases was 29.58 years (range: 18–65 years, standard deviation (SD=8.64)). Mean age for controls was 44.19 years

(range: 18–72 years, SD=13.24) (details see Supplementary Table 1). Of the 998 cases, 666 had co-morbid life-time MDD, and 262 did not (data missing for 40 cases).

Single marker analysis

A total of 1,0736,316 single markers were included in the analysis. As expected for GWAS on a complex psychiatric disorder with the current sample size, the single marker analysis revealed no significant hit after correction for multiple testing (see Figure 1&2). The most significant marker was rs113507694 in *DPPA3* on chromosome 12 ($p=2.01x10^{-07}$; OR=0.35, MAF=0.03, INFO=0.59). Single markers with $p<1x10^{-5}$ are listed in Supplementary Table 2.

Gene-based-analysis

In the gene-based-analysis, a total of 17,755 genes were tested. Two genes showed significant association with BOR after correction for multiple testing: the gene coding for Plakophilin-4 on chromosome 2 (*PKP4*; $p=8.24x10^{-7}$); and the gene coding for dihydropyrimidine dehydrogenase on chromosome 1 (*DPYD*, $p=1.20x10^{-6}$). The most significant genes ($p<5x10^{-4}$) are listed in Table 1. The top hit of the previous GWAS of Borderline personality features, *SERINC5,* achieved nominal significance in the present study ($p_{uncorrected}=0.016$).

Gene-set-analysis

Gene-set-analysis with i-GSEA4GWASv2 revealed one significant gene-set: exocytosis (GO: 0006887; $p_{FDR}=0.019$). Of 25 genes in this gene-set, 22 were mapped with variants and 15 showed nominally significant associations. Details on significant and non-significant genes in this gene-set are provided in Supplementary Table 3. All gene-sets with $p_{uncorrected}<0.01$ are shown in Table 2. A technical replication analysis with GSA-SNP and MAGMA confirmed the gene-set exocytosis (GSA-SNP: $p_{uncorrected}=2.32x10^{-4}$; MAGMA: $p_{uncorrected} = 0.056$).

LD-Score regression

Significant genetic correlations with BOR were found for BIP (r_g=0.28; SE=0.094; p=2.99x10^{-3}), MDD (r_g=0.57; SE=0.18; p=1.04x10^{-3}), and SCZ (r_g=0.34; SE=0.082; p=4.37x10^{-5}); A meta-analytic comparison revealed no significant differences between the correlations (all p > 0.13).

Polygenic Risk Score

PRS analysis revealed significant associations with BOR for the PRS of BIP, MDD, and SCZ. SCZ PRS were significant for all investigated thresholds. BIP and MDD scores were significant for all PRS that included SNPs with p-values higher than 0.0001 and 0.001 respectively (see Supplementary Table 4). The share of variance explained in BOR case-control status (Nagelkerke's R^2) by the respective PRS was up to 0.86% for BIP; up to 3.1% for SCZ; and up to 2.1% for MDD (see Figure 3 and Supplementary Table 4).

Simultaneous addition of the PRS for SCZ, BIP, and MDD (threshold p<0.05) to the regression model explained 4.4% of the variance (Nagelkerke's R^2) in BOR case-control status. The PRS for SCZ and the PRS for MDD were significant predictors (p=9.78x10^{-9} and p=1.9x10^{-7}, respectively). The PRS for BIP was not a significant predictor in this model (p=0.28).

A secondary analysis was then performed including: (i) BIP PRS with MDD PRS; and (ii) BIP PRS with SCZ PRS. Here, BIP PRS explained variance independently of MDD PRS (p=0.0067), but not of SCZ PRS (p = 0.11).

Differentiation between cases with and without comorbid MDD and controls revealed significant effects of BOR diagnosis on PRS for BIP, SCZ, and MDD (all p<0.001, see Figure 4). Post-hoc analyses revealed no differences in PRS for the BIP, SCZ, or MDD PRS of the BOR subgroup with comorbid MDD compared to the BOR subgroup without MDD (all p>0.5). Compared to controls, PRS for SCZ and MDD were significantly increased in the BOR subgroups with and without comorbid MDD (all p<0.001). The PRS for BIP only showed a

significant difference to controls in the BOR subgroup with comorbid MDD ($p<0.001$, see Figure 4).

Discussion

The present study is the first case-control GWAS of BOR. As expected, no genome-wide significant association was found for any single marker. In the gene-based test, however, two genes achieved genome-wide significance: *Dihydropyrimidine Dehydrogenase* (*DPYD*) and *Plakophilin4* (*PKP4*). *DPYD* encodes a pyrimidine catabolic enzyme, which is the initial and rate-limiting factor in the pathway of uracil and thymidine catabolism. Genetic deficiency of this enzyme results in an error in pyrimidine metabolism (37). This is associated with thymine-uraciluria and an increased risk of toxicity in cancer patients receiving 5-fluorouracil chemotherapy (http://www.ncbi.nlm.nih.gov/gene/1806). Recent Psychiatric Genomics Consortium (PGC) metaanalyses revealed an association between *DPYD* and SCZ and BIP (34, (38, (39). *DPYD* contains a binding site for the micro-RNA miR-137, which has previously been associated with schizophrenia (40), and a previous exome sequencing study reported two putative functional de novo variants in *DPYD* in cases with SCZ (41). *PKP4* is involved in the regulation of cell adhesion and cytoskeletal organization (42). In pathway analyses of PGC GWAS data, cell adhesion was associated with BIP (43), and SCZ (44), whereas cell junction was implicated in MDD, as well as in an integrative pathway analysis of all three disorders (45).

SERINC5, which was the top hit of the previous GWAS of Borderline personality features (7), achieved nominal significance in the present study. The protein SERINC5 incorporates serine into newly forming membrane lipids, and is enriched in myelin in the brain (46). Previous research suggests that decreased myelination is associated with a reduced capacity for social interaction (7, (47).

The gene-set analyses yielded significant results for exocytosis. In neuronal synapses, exocytosis is triggered by an influx of calcium and critically underlies synaptic signaling.

Dysregulated neuronal signaling and exocytosis are core features of neurodevelopmental psychiatric disorders such as the autism spectrum disorders and intellectual disability (48, (49). Moreover, recent findings from large metaanalyses have implicated dysregulated neuronal signaling and exocytosis in the molecular mechanisms of BIP, SCZ, and MDD (48, (50, (51). These processes may now represent promising starting points for further research into BOR.

The most interesting finding of this study is that BOR showed a genetic overlap with BIP, SCZ, and MDD. Notably, BIP did not show a higher correlation with BOR (r_g=0.28) than SCZ (r_g=0.34) or MDD (r_g=0.57). In view of the present sample size, these values must be viewed with caution. A more accurate estimation of these correlations will require calculations in larger cohorts.

Although comorbid BIP was excluded in the present BOR patients, the possibility that the observed genetic overlap between BOR and BIP was at least partly attributable to misdiagnosis cannot be excluded. However, an alternative explanation appears more likely, i.e. that disorders currently categorized as BOR and BIP share a common genetic background, and they also do so with SCZ and MDD. This hypothesis is supported by the present observation of a genetic overlap between BOR and SCZ, two disorders that are rarely misdiagnosed by psychiatrists, despite the presence of common psychotic symptoms.

An explanation could also be that the genetic commonality between BOR and BIP, SCZ, and MDD might be due to a common effect of MDD. Prior to the introduction of DSM-IV, a history of MDD was required for a diagnosis of BIP, and MDD has a high prevalence in patients with SCZ (25-85%) (52, (53). Therefore, the MDD genetic risk variants that are common to BOR, BIP, and SCZ may be responsible for the observed overlap. For this reason, we conducted two further analyses. First, we compared PRS of BIP, SCZ, and MDD in subsamples of BOR patients with (~60%) and without comorbid MDD. Here, no differences in any of the PRS were found. Second, we performed a joint analysis of PRS of BIP, SCZ, and MDD in a logistic regression analysis in BOR patients vs. controls. Here, no differences were found in any of the PRS. Second, we performed a joint analysis of the PRS of BIP, SCZ, and MDD in

a logistic regression analysis in BOR patients vs. controls. Here, both the SCZ and the MDD risk score explained variance in BOR case-control status independently. Secondary analysis revealed that the BIP risk score explained variance independently of the MDD risk score but not of the SCZ risk score. These results indicate that comorbidity with MDD does not explain the genetic overlap between BOR and BIP, SCZ, and MDD. However, the training sets differ in terms of their power to detect underlying risk variants, and therefore the derived PRS differ in terms of the variance they can explain.

It must be noted, that in the PGC-BIP, -SCZ and -MDD samples, controls are partly overlapping. However, it is unlikely that this drives the genetic correlation of BOR with those disorders as the overlap of controls in these samples is rather small (under 10%) (54). Also, the joint logistic regression analysis demonstrated that polygenic risk for SCZ and MDD contributed independently to the BOR risk (see above).

The present study had several limitations. First, despite being one of the largest BOR samples available worldwide, the sample size was small in terms of the estimation of heritability. Replication of the present results is warranted in larger, independent cohorts. This should include the investigation of non-European samples. Second, no information was available on the presence of common clinical features such as psychotic symptoms and affect instability. This precluded detailed analysis of the identified genetic overlap. Future studies in larger cohorts should also investigate more detailed phenotypes, including comorbid axis I and axis II disorders, such as addiction and personality disorders, respectively. Third, the observation that psychiatric patients often establish non-random relationships with persons affected by the same or another psychiatric disorder (55), and therefore have offspring with a higher genetic risk for psychiatric disorders, might contribute to the observed genetic correlation of BOR with BIP, SCZ, and MDD. However, the LD score method does not investigate the impact of assortative mating (32). Therefore, assessment of the degree to which this phenomenon may have influenced the genetic correlation estimates was beyond the scope of the present study."

Despite these limitations, the results indicate that neither comorbidity with MDD nor risk variants that are exclusive to MDD explain the genetic overlap between BOR and BIP, SCZ, and MDD. Future investigations of larger data sets for BOR and other psychiatric disorders are warranted to refine the analysis of shared and specific genetic risk.

Future studies are warranted to delineate the communalities and specificities of the respective disorders.

Conclusion

In summary, the present study is the first GWAS of patients diagnosed with BOR. The results suggest promising novel genes and a novel pathway for BOR, and demonstrate that, rather than being a discrete entity, BOR has an etiological overlap with the major psychoses. The genetic overlap with BIP is consistent with the observation that some diagnostic criteria for BOR overlap with those for BIP. The overlap between BOR and SCZ and MDD is consistent with previous observations of genetic overlap of other psychiatric disorders (56). Given that BOR patients display specific clinical symptoms not observed in patients with other psychiatric disorders, knowledge of shared and non-shared genetic and clinical features will be important for the development of personalized treatment approaches.

Supplementary information is available at *Translational Psychiatry*'s website.

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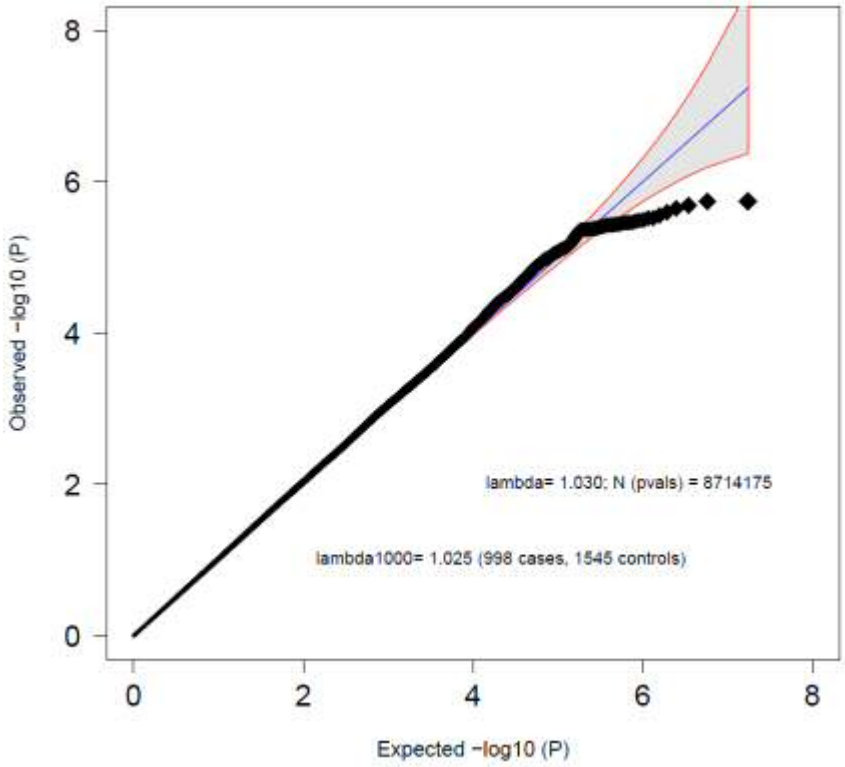
Figure Legends

Figure 1: Quantile-Quantile plot. Quantile-Quantile plot of the case-control analysis (998 cases; 1,545 controls) showing expected and observed –log10 p-values. The shaded region indicates the 95% confidence interval of expected p-values under the null hypothesis.

Figure 2: Manhattan plot showing association results. Manhattan plot of the case-control analysis (998 cases; 1,545 controls). For each SNP, the chromosomal position is shown on the x-axis, and the –log10 p-value on the y-axis. The red line indicates genome-wide significance ($p<5x10^{-8}$) and the blue line indicates suggestive evidence for association ($p<1x10^{-5}$).

Figure 3: Polygenic Risk Score analysis. The proportion of variance explained in case-control status (y axis; Nagelkerke's R^2) by the PRS for BIP, SCZ, and MDD is depicted for the different p-value cutoffs used in the calculation of the PRS. Principal components were included in the models to control for population stratification. PRS = Polygenic risk score; BIP = Bipolar Disorder; MDD = Major Depressive Disorder; SCZ = Schizophrenia; ns = non-significant; 1* $p<0.05$; 2* $p<0.001$; 3* $p<1x10^{-4}$; 4* $p<1x10^{-6}$; 5* $p<1x10^{-8}$; 6* $p<1x10^{-10}$; 7* $p<1x10^{-12}$

Figure 4: Polygenic Risk Score analysis in subgroups. Mean z-standardized PRS and standard error (SE) for BIP, SCZ, and MDD are shown in the control group, all cases, and in cases with and without comorbid MDD. PRS with a p-value threshold of p=0.05 were selected for this comparison and principal components were included in the models to control for population stratification. The numbers at the top of each bar indicate the significance of the difference in the respective PRS in comparison to the control group. BOR = Borderline Personality Disorder; BIP = Bipolar Disorder; MDD = Major Depressive Disorder; SCZ = Schizophrenia; ns = non-significant; 1* $p<0.05$; 2* $p<0.001$; 3* $p<1x10^{-4}$; 4* $p<1x10^{-6}$; 5* $p<1x10^{-8}$; 6* $p<1x10^{-10}$; 7* $p<1x10^{-12}$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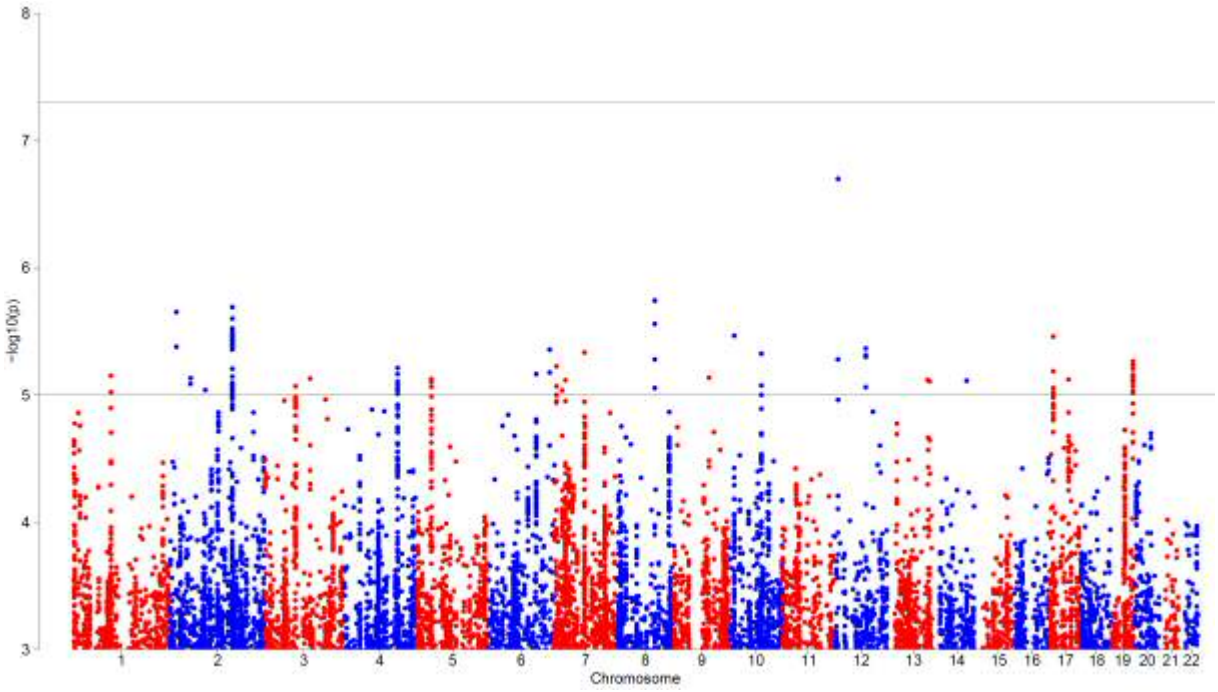
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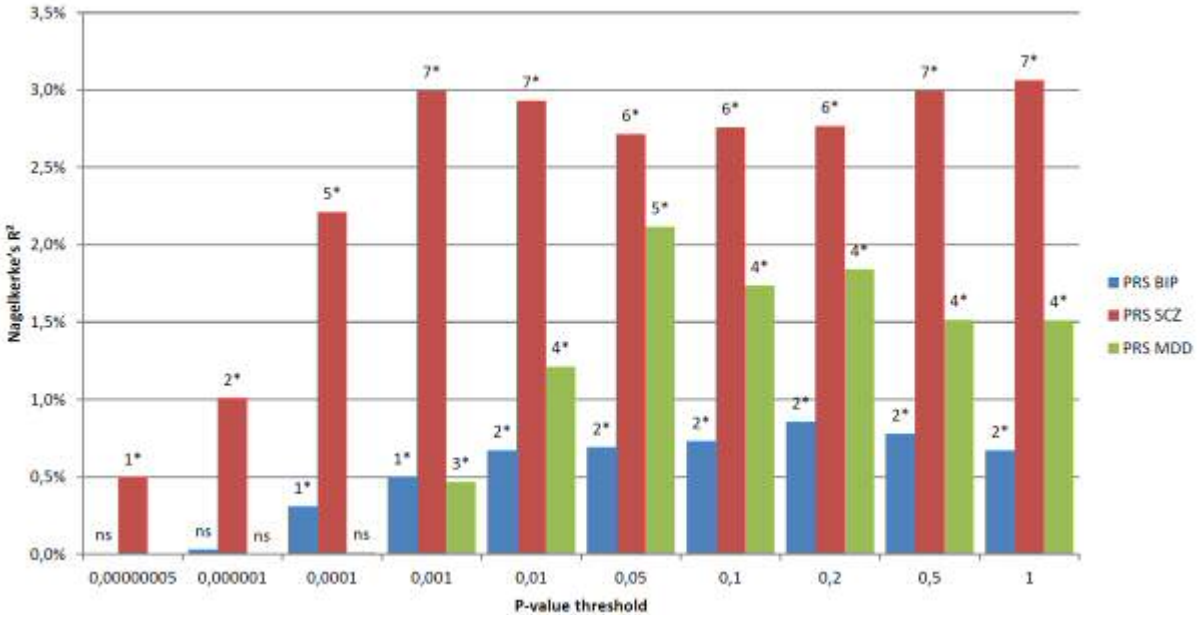
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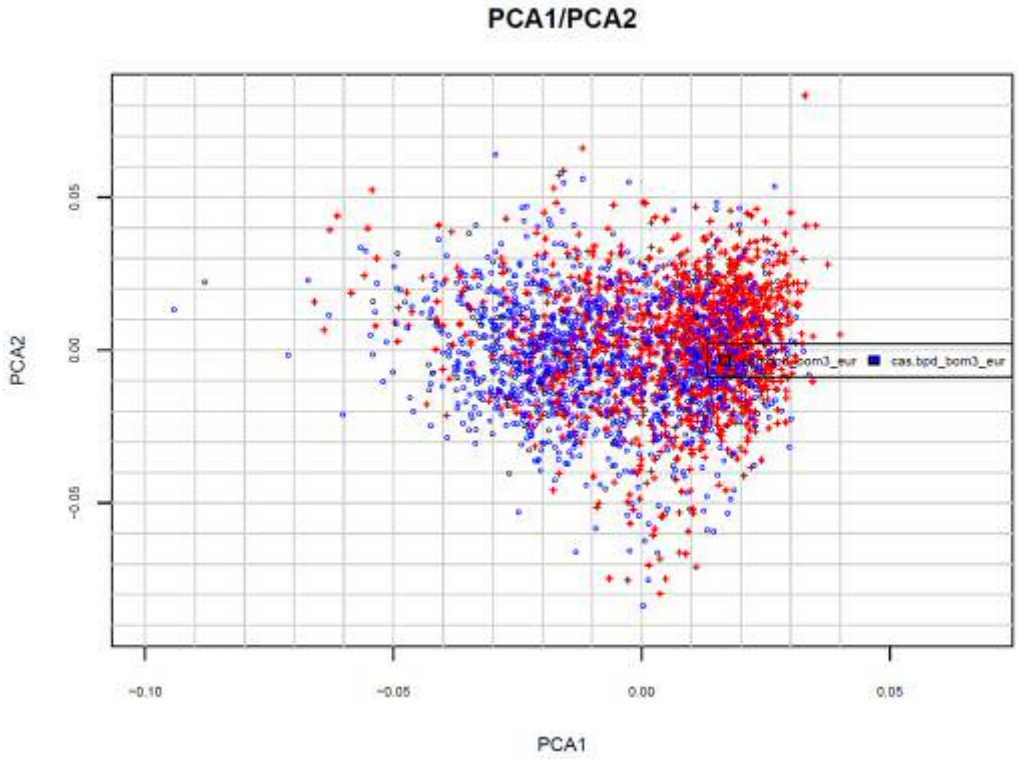
P-value threshold
PRS BIP
PRS SCZ
PRS MDD

Supplementary Information for *Genome-wide association study of Borderline Personality Disorder reveals genetic overlap with the Major Psychoses*

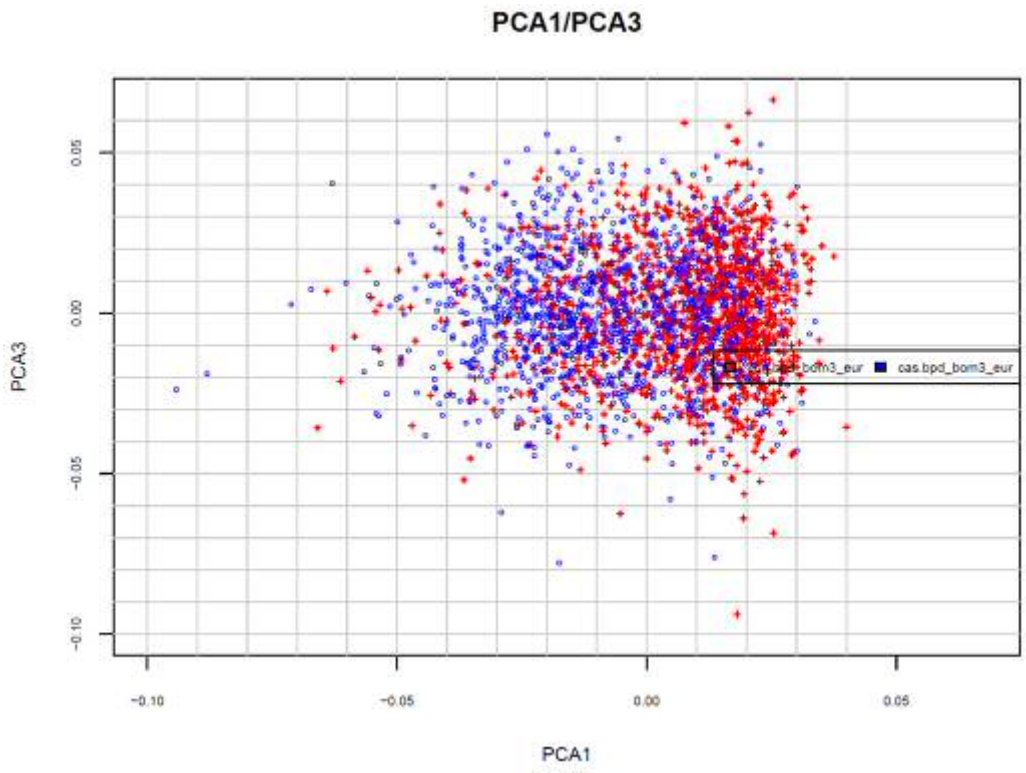
Table of Content

Figures

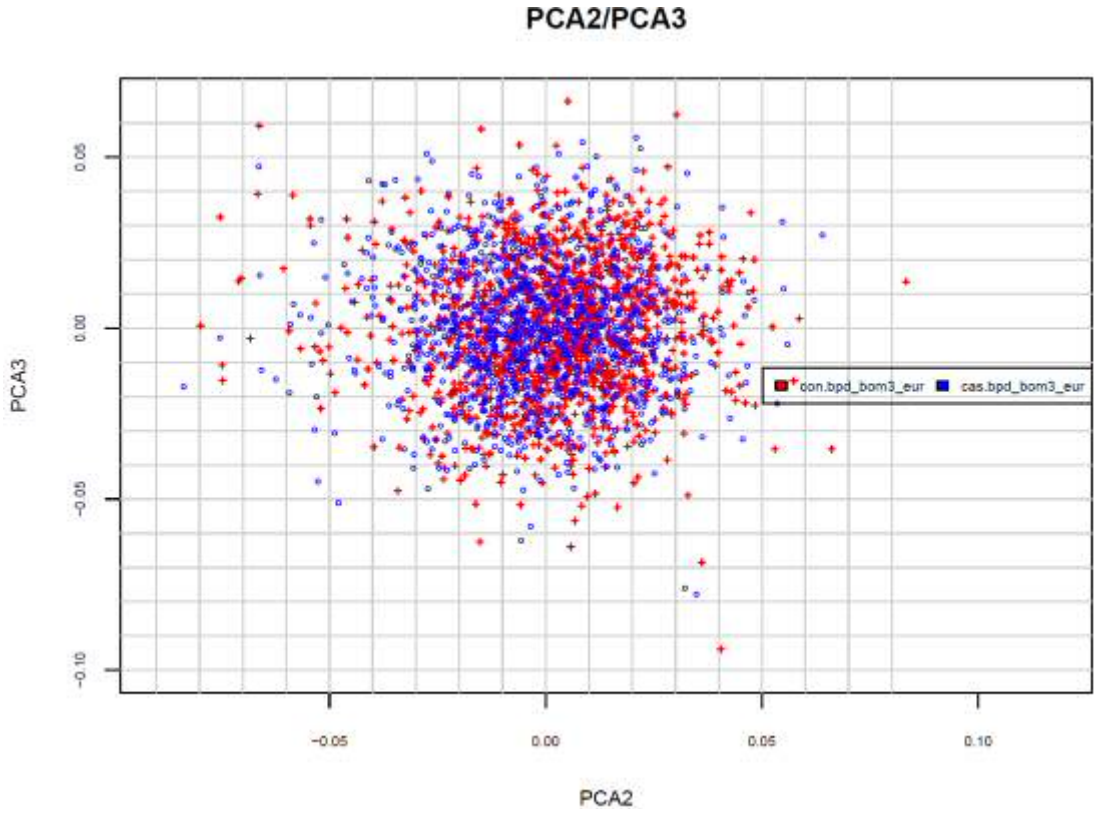
Supplementary Figure 1: Scatter plot of principal components 1 and 2



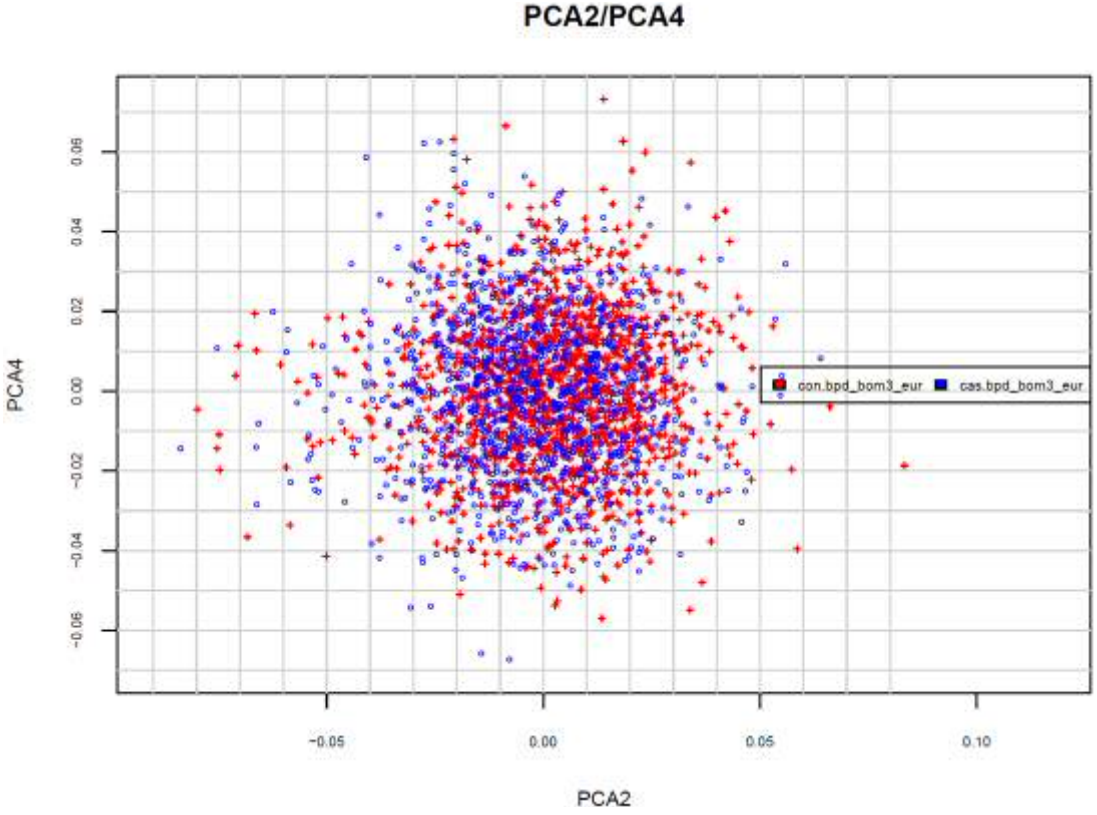
Supplementary Figure 2: Scatter plot of principal components 1 and 3



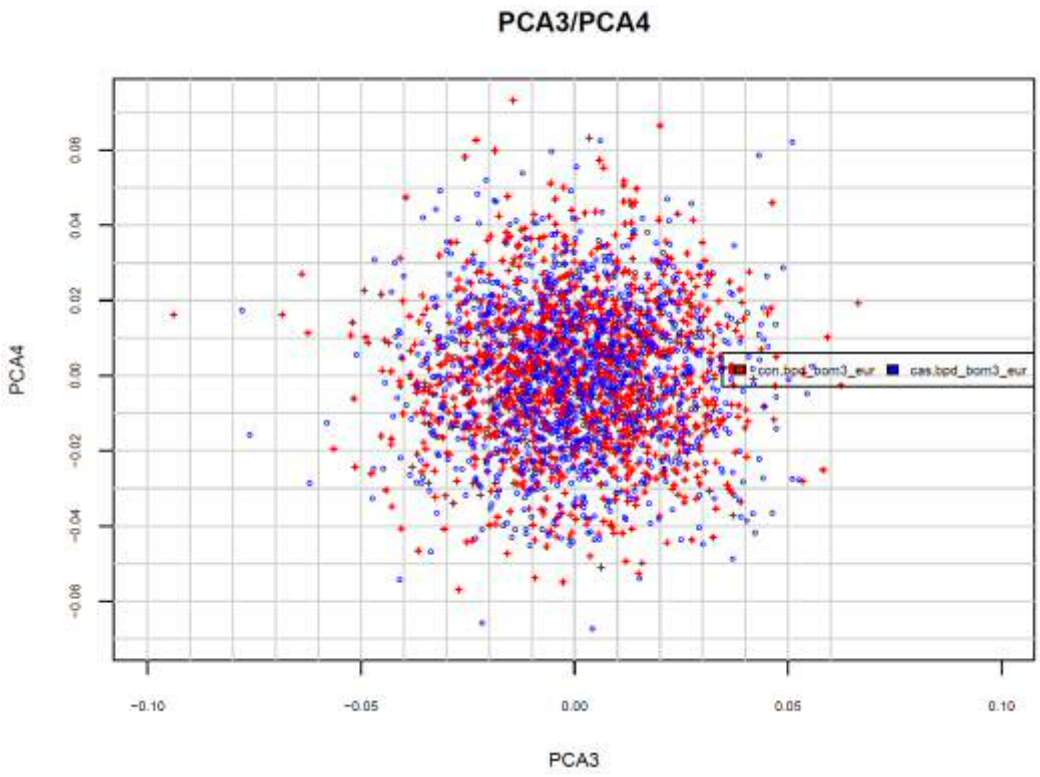
Supplementary Figure 3: Scatter plot of principal components 2 and 3



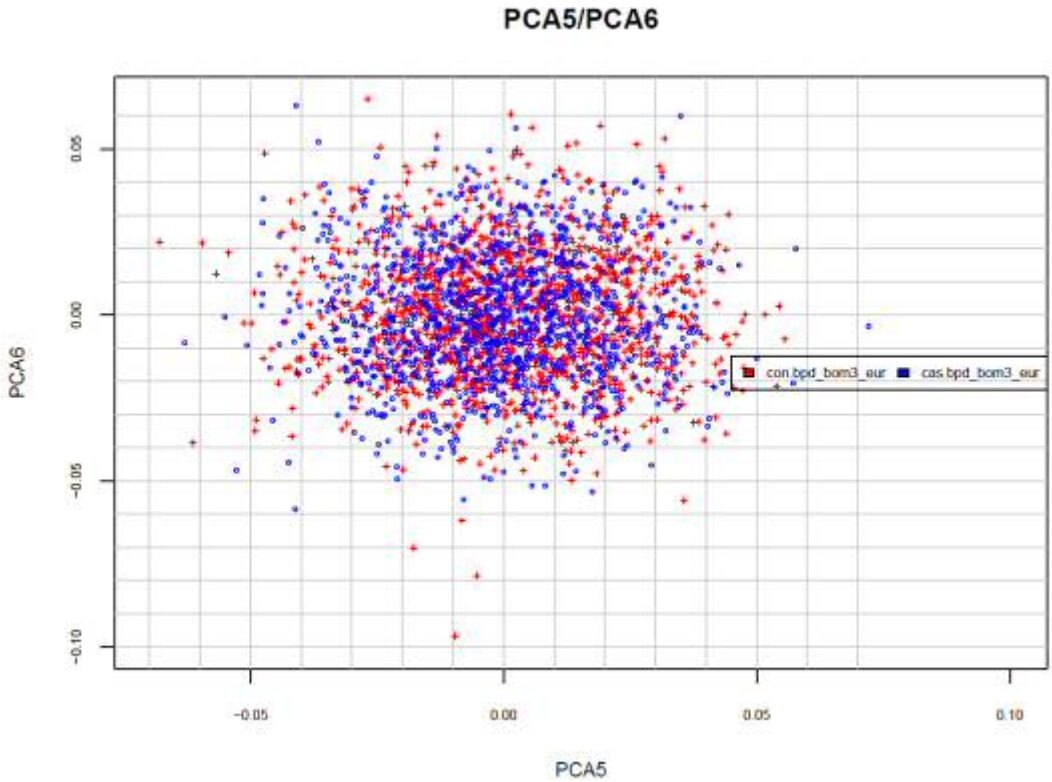
Supplementary Figure 4: Scatter plot of principal components 2 and 4



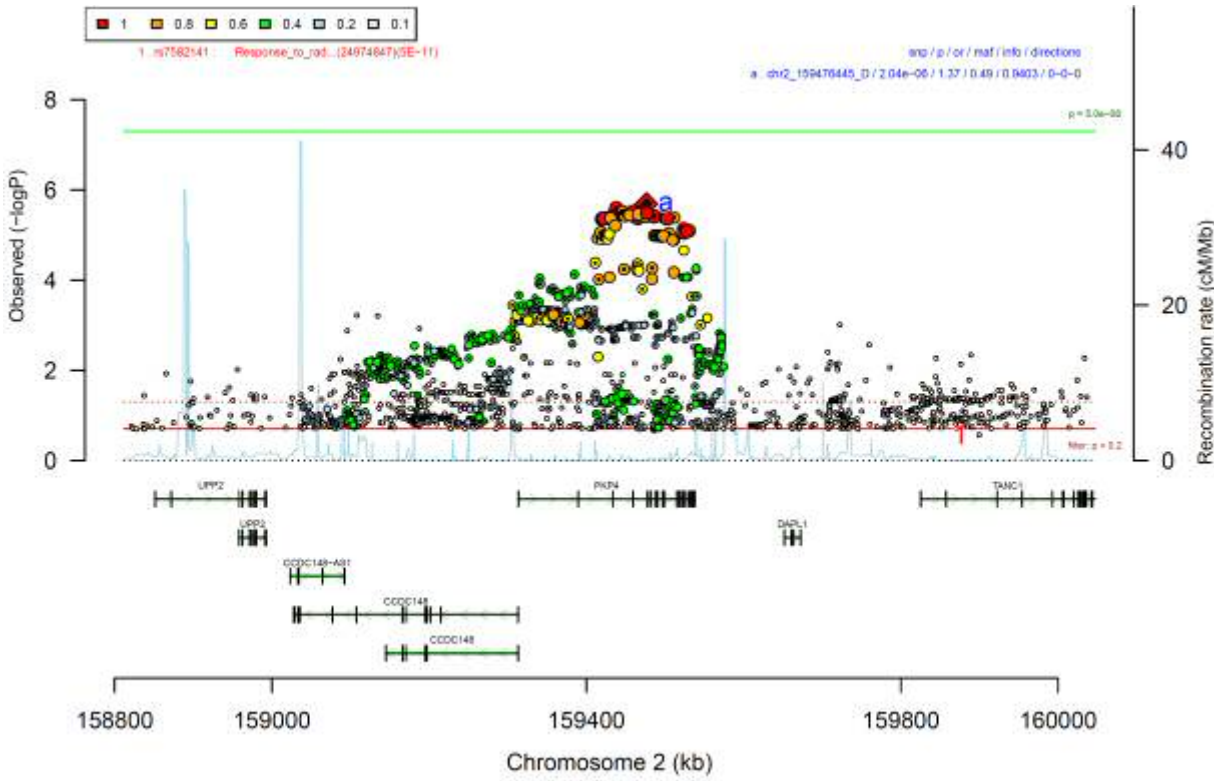
Supplementary Figure 5: Scatter plot of principal components 3 and 4



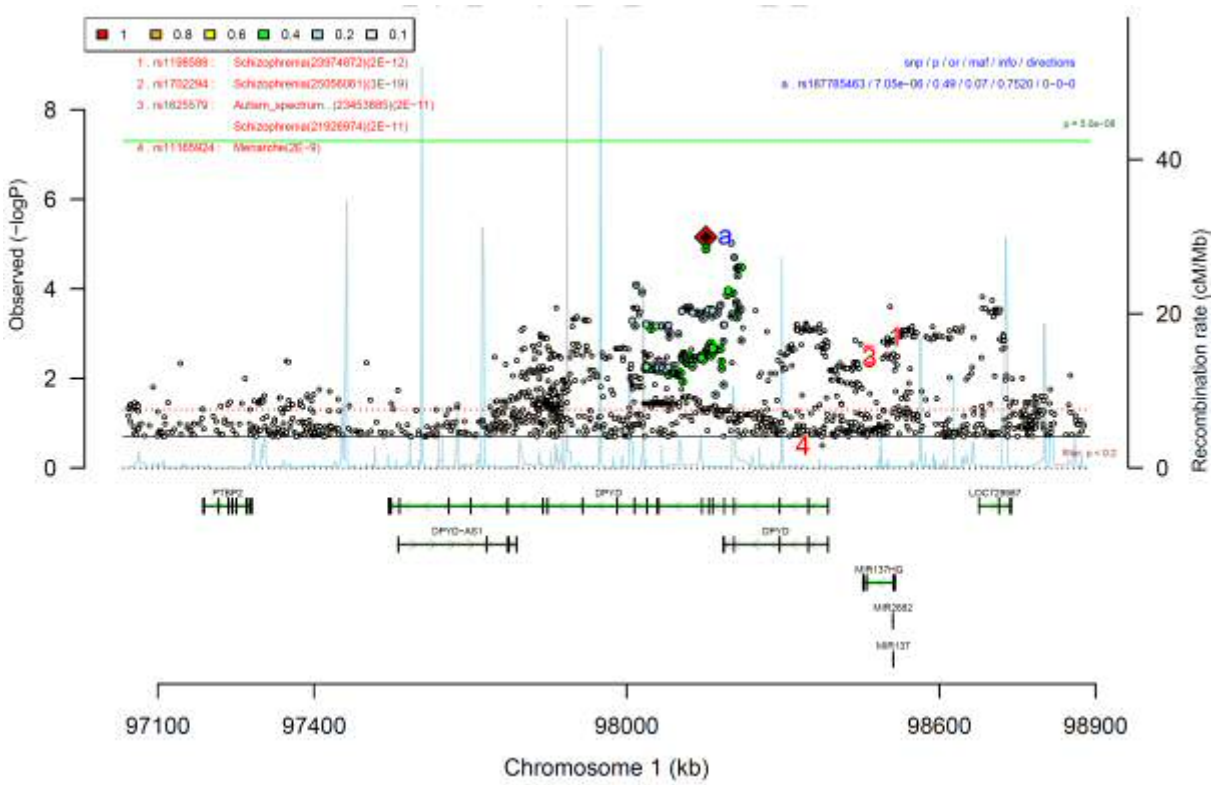
Supplementary Figure 6: Scatter plot of principal components 5 and 6



Supplementary Figure 7: Region plot of PKP4



Supplementary Figure 8: Region plot of DPYD



Tables

Supplementary Table 1: Demographic and clinical information on the final sample

	Cases	Controls
N	998	1,545
Sex (female/male)	914 / 84	868 / 677
Age in years (SD)	29.58 (8.64)	44.19 (13.24)
Comorbidity		
Depression		
(yes / no / missing)	666 / 262 / 40	-
Alcohol dependency		
(yes / no / missing)	163/ 781 / 54	-

Supplementary Table 2: Association results for single markers

Markers are reported with p<1 x10 $^{-5}$, and sorted according to chromosomal position . CHR = chromosome, BP = base pair position, A = allele, FRQ = frequency, SNP = single nuc leotide polymorphism, OR = odds ratio, INFO = imputation info score, SE = standard error, GT = genotyped, IM P = imputed

CHR	SNP	BP	Gene	A1	A2	FRQ cases	FRQ controls	INFO	OR	SE	P	GT
1	rs187785463	98152125	DPYD	A	G	0.0531	0.0739	0.752	0.4935	0.1572	7.05E-06	IMP
1	rs6683957	98200719	DPYD	A	G	0.078	0.1128	1.002	0.6117	0.111	9.46E-06	IMP
2	rs57726666	16203326	GACAT3	A	G	0.9407	0.9125	0.912	1.8855	0.1341	2.24E-06	IMP
2	rs78068563	16208690	GACAT3	A	G	0.9397	0.9127	0.9227	1.8395	0.1324	4.19E-06	IMP
2	rs115689122	52606079	AC087073.1	A	T	0.016	0.0054	0.7809	6.1189	0.4062	8.20E-06	IMP
2	rs62127626	52719291	AC139712.4	A	C	0.0163	0.0057	0.7744	5.9639	0.3984	7.38E-06	IMP
2	rs150592717	89936117		T	C	0.8884	0.869	0.3247	2.2187	0.1796	9.14E-06	IMP
2	rs4664975	159418438	PKP4	A	C	0.4677	0.5152	0.8943	0.7399	0.0678	8.77E-06	IMP
2	rs3771608	159419739	PKP4	A	C	0.5405	0.4847	0.9831	1.345	0.0646	4.42E-06	IMP
2	rs12052933	159420140	PKP4	A	G	0.54	0.4841	0.9852	1.3451	0.0645	4.29E-06	IMP
2	rs10174340	159421412	PKP4	A	T	0.54	0.4841	0.9856	1.3451	0.0645	4.27E-06	IMP
2	rs3771609	159422239	PKP4	T	C	0.46	0.516	0.9859	0.7434	0.0645	4.25E-06	IMP
2	rs3771610	159422317	PKP4	A	G	0.54	0.484	0.9859	1.3451	0.0645	4.25E-06	IMP
2	rs10187426	159425104	PKP4	A	G	0.5399	0.4839	0.9869	1.3451	0.0644	4.21E-06	IMP
2	rs7577672	159429019	PKP4	A	G	0.4748	0.5324	0.9886	0.7523	0.0643	9.56E-06	IMP
2	rs3771614	159430579	PKP4	T	G	0.5318	0.4731	0.9904	1.3443	0.0643	4.26E-06	IMP
2	rs3771616	159430991	PKP4	A	G	0.5318	0.4731	0.9903	1.3444	0.0643	4.23E-06	IMP
2	rs3821291	159431031	PKP4	A	G	0.5318	0.4731	0.9903	1.3444	0.0643	4.23E-06	IMP

2	rs12473797	159431663	PKP4	T	C	0.5319	0.4732	0.9898	1.3449	0.0644	4.14E-06	IMP
2	rs3755408	159431842	PKP4	T	C	0.5318	0.4731	0.9903	1.3445	0.0643	4.21E-06	IMP
2	chr2_159432313_D	159432313	PKP4	I2	D	0.5496	0.4914	0.9516	1.3537	0.0658	4.13E-06	IMP
2	rs2356189	159434500	PKP4	A	G	0.5402	0.4839	0.9869	1.3482	0.0644	3.55E-06	IMP
2	rs3755413	159436702	PKP4	A	G	0.528	0.4703	0.9825	1.3387	0.0646	6.23E-06	IMP
2	rs3771620	159438129	PKP4	T	C	0.4591	0.5157	0.9808	0.7375	0.0647	2.52E-06	IMP
2	rs4664979	159439898	PKP4	T	C	0.4597	0.5161	0.9868	0.7414	0.0644	3.44E-06	IMP
2	rs3771627	159443266	PKP4	T	C	0.4596	0.5161	0.9865	0.741	0.0645	3.31E-06	IMP
2	rs2108215	159446818	PKP4	T	G	0.4674	0.5269	0.9909	0.7404	0.0643	3.01E-06	GT
2	rs11891131	159447509	PKP4	T	C	0.532	0.4731	0.9898	1.347	0.0644	3.71E-06	IMP
2	rs1465236	159448238	PKP4	A	G	0.4682	0.527	0.9894	0.7424	0.0644	3.71E-06	IMP
2	rs999232	159448306	PKP4	C	G	0.468	0.5269	0.9899	0.7422	0.0644	3.62E-06	IMP
2	rs3771631	159448926	PKP4	A	C	0.468	0.5269	0.9898	0.7422	0.0644	3.62E-06	IMP
2	rs3771632	159448946	PKP4	C	G	0.5341	0.4763	0.9806	1.3485	0.0646	3.75E-06	IMP
2	rs2051946	159454789	PKP4	T	G	0.532	0.4732	0.9886	1.3474	0.0644	3.66E-06	IMP
2	rs2051947	159459863	PKP4	A	G	0.5321	0.4733	0.9884	1.3472	0.0644	3.71E-06	IMP
2	rs12694965	159460218	PKP4	T	C	0.4594	0.5159	0.9836	0.7396	0.0646	3.00E-06	IMP
2	rs10191923	159460807	PKP4	T	C	0.5321	0.4735	0.9882	1.3467	0.0644	3.83E-06	IMP
2	rs10191934	159460832	PKP4	T	G	0.5322	0.4736	0.9877	1.3461	0.0644	3.97E-06	IMP
2	rs10191939	159460846	PKP4	A	T	0.4679	0.5267	0.9885	0.7423	0.0644	3.72E-06	IMP
2	chr2_159465312_I	159465312	PKP4	I5	D	0.4498	0.5053	0.9315	0.7371	0.0663	4.28E-06	IMP
2	rs3771643	159466149	PKP4	T	C	0.532	0.4731	0.9889	1.3483	0.0644	3.46E-06	IMP
2	rs3771647	159469420	PKP4	T	C	0.5321	0.4735	0.9893	1.3459	0.0644	3.93E-06	IMP
2	rs7607589	159469920	PKP4	T	C	0.4594	0.5159	0.9845	0.7404	0.0645	3.18E-06	GT
2	chr2_159476445_D	159476445	PKP4	I13	D	0.5489	0.4908	0.9403	1.3685	0.0661	2.04E-06	IMP

Table 1: Results of the gene -based analysis using MAGMA: Most significant genes ($p<5x10^{-4}$) in the gene -based analysis and their chromosomal position. Genes in bold font were significant after correction for multiple testing; CHR = chromosome, NSNPS = number of SNPs, NPARAM = number of parameters used in the model, ZSTAT: z-value of the gene, P = gene p-value

GENE	*CHR*	*START*	*STOP*	*NSNPS*	*NPARAM*	*ZSTAT*	*P*
PKP4	2	159303476	159547941	21	13	4,7924	$8,24x10^{-7}$
DPYD	1	97533299	98396615	105	68	4,7162	$1,20x10^{-6}$
GRAMD1B	11	123315191	123508478	34	28	3,8856	$5,10x10^{-5}$
STX8	17	9143788	9489275	38	33	3,7984	$7,28x10^{-5}$
BMP2	20	6738745	6770910	7	6	3,588	$1,67x10^{-4}$
TRAF3IP1	2	239219185	239319541	11	8	3,5389	$2,01x10^{-4}$
ZP3	7	76016841	76081388	9	7	3,5037	$2,29x10^{-4}$
PINX1	8	10612473	10707394	19	11	3,5034	$2,30x10^{-4}$
GTF3C4	9	135535728	135575471	4	4	3,4851	$2,46x10^{-4}$
DNAH1	3	52340335	52444513	11	8	3,4543	$2,76x10^{-4}$
YKT6	7	44230577	44263893	6	3	3,3841	$3,57x10^{-4}$
CCSER1	4	91038684	92533370	111	78	3,3804	$3,62x10^{-4}$
LRRC59	17	48448594	48484914	8	6	3,3716	$3,74x10^{-4}$
TMEM71	8	133712191	133782914	9	8	3,3668	$3,80x10^{-4}$
BAP1	3	52425020	52454121	3	3	3,345	$4,11x10^{-4}$
AQR	15	35138552	35271995	8	6	3,3299	$4,34x10^{-4}$
FGFR1	8	38258656	38336352	12	10	3,3162	$4,56x10^{-4}$

Table 2: Results of the gene-set analysis: Most significant gene-sets (uncorrected p<0.01) in the gene-set analysis with i-GSEA4GWAS v2 are listed. Gene-sets in bold font were significant after correction for multiple testing, P-value = gene-set p-value, FDR = false discovery rate

Gene-set Name	Number of genes	P-value	FDR P-value
GO: EXOCYTOSIS	**25**	**0.001**	**0.019**
GO: RESPONSE TO ORGANIC SUBSTANCE	30	0.002	0.173
GO: BRAIN DEVELOPMENT	51	0.003	0.888
GO: HORMONE METABOLIC PROCESS	30	0.003	0.511
GO: PROTEIN C TERMINUS BINDING	73	0.003	0.536
GO: LYSOSOME	53	0.007	0.785
GO: LYTIC VACUOLE	53	0.007	0.785
GO: MULTI ORGANISM PROCESS	143	0.007	0.920