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Association study of 21 circadian genes with bipolar I disorder, schizoaffective disorder, and schizophrenia

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Abstract

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Objective—Published studies suggest associations between circadian gene polymorphisms and bipolar I disorder (BPI), as well as schizoaffective disorder (SZA) and schizophrenia (SZ). The results are plausible, based on prior studies of circadian abnormalities. As replications have not been attempted uniformly, we evaluated representative, common polymorphisms in all three disorders.

Methods—We assayed 276 publicly available 'tag' single nucleotide polymorphisms (SNPs) at 21 circadian genes among 523 patients with BPI, 527 patients with SZ/SZA, and 477 screened adult controls. Detected associations were evaluated in relation to two published genome-wide association studies (GWAS).

Results—Using gene-based tests, suggestive associations were noted between *EGR3* and BPI ($p = 0.017$), and between *NPAS2* and SZ/SZA ($p = 0.034$). Three SNPs were associated with both sets of disorders (*NPAS2*: rs13025524 and rs11123857; *RORB:* rs10491929; $p < 0.05$). None of the associations remained significant following corrections for multiple comparisons. Approximately 15% of the analyzed SNPs overlapped with an independent study that conducted GWAS for BPI; suggestive overlap between the GWAS analyses and ours was noted at *ARNTL*.

Conclusions—Several suggestive, novel associations were detected with circadian genes and BPI and SZ/SZA, but the present analyses do not support associations with common polymorphisms that confer risk with odds ratios greater than 1.5. Additional analyses using adequately powered samples are warranted to further evaluate these results.

Keywords

association; bipolar disorder; circadian; gene; schizoaffective disorder; schizophrenia

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Family, twin, and adoption studies suggest heritability estimates of 60–80% for bipolar I disorder (BPI), schizophrenia (SZ), and schizoaffective disorder (SZA) (1–3). The mode of inheritance of BPI has been difficult to define, but current analyses suggest multiple interacting loci contributing to BPI liability (4). This complexity may explain the inconsistencies that led to prior gene-mapping efforts. The traditional approach, called linkage analysis, involves the analysis of shared genomic regions among members of multiply affected families. Identification of such shared (linked) regions can help localize diseased genes. Though several genome-wide scans have now been completed, they differ not only with respect to ascertainment criteria, but also marker density, sample sizes, and method of analysis. Nevertheless, a number of groups have reported linkage signals in overlapping regions. Recent meta-analyses have identified several plausible regions for BPI, including chromosomes 13q, 22q, 9p22.3-21.1, 10q11.21-22.1, and 14q24.1-32.12 (5, 6). These linked regions have not been identified in all studies. The failure to replicate these results may reflect insufficient power, differing designs, the presence of genetic heterogeneity, or the absence of a susceptibility locus in the region.

Another complementary approach for gene mapping, called association analysis, might arguably hold greater promise (7). Association studies involve comparison of genetic polymorphisms among cases and suitable controls. Case–control differences in the frequency of alleles (variants) of particular polymorphisms may help pinpoint etiological factors. Historically, case–control association studies have focused on relatively small genomic regions in contrast to linkage studies. To improve the chances of detecting meaningful associations, researchers have selected particular genes whose products have been implicated in other avenues of bipolar research. This approach is called the 'candidate gene' strategy. Among dozens of candidate genes that have been investigated, consistent associations have been difficult to detect (8). These inconsistent results may be related to variation in ascertainment, phenotype definition, and control selection, limited power

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(studies have typically included fewer than 200 cases or families), and possible confounding by population substructure. Another reason could be that the initial report was simply a false positive.

Among different types of polymorphisms used for gene-mapping studies, bi-allelic single nucleotide polymorphisms (SNPs) have come to the fore. The SNPs have been utilized to evaluate associations across the genome simultaneously. Such genome-wide association studies (GWAS) have helped identify replicable associations for several common multifactorial diseases, including type I and type II diabetes mellitus, Crohn's disease, age-related macular degeneration, and several types of cancers (9). In view of the emerging success in other diseases, GWAS in SZ and BPI are in progress (9–15).

Several lines of evidence point to circadian dysfunction in BPI pathogenesis (16, 17). Patients with BPI display disrupted circadian function, including changes in sleep, appetite, and hormonal secretions, especially during relapse (18, 19). Disruption in circadian and social rhythms may precede the onset of manic episodes (20, 21). Clinical stabilization of bipolar symptoms is usually associated with normalization of the disrupted circadian function. Indeed, mood stabilizers may exert therapeutic effects by modulating circadian function (22–26).

A pathogenic role for circadian dysfunction in BPI can be investigated using genetic association studies. Several circadian gene polymorphisms have been investigated, with mixed results (27–31). Haplotype-based analyses suggest associations at *Bmal I* (*ARNTL*) and Period 3 (*PER3*) (32), as well as *PER3* (31), with nonsignificant associations at *PER2* and *Cry1* (32, 33). Another recent study that analyzed 10 circadian genes reported interactions between SNPs at *BHLHB2, CSNK1E,* and *CLOCK* (34). The glycogen synthase kinase 3-β gene (GSK3-β) has also been extensively investigated in relation to BPI because it regulates the molecular clock in the suprachiasmatic nucleus (SCN) (35, 36). A number of association studies of GSK3 and bipolar disorder have been published with inconsistent results. While some studies have found associations (27, 28), others did not (29, 30, 37). Circadian-rhythm abnormalities in schizophrenia were also reported. Martin et al. (38) showed that elderly schizophrenia patients had disrupted sleep and circadian rhythms compared with age- and gender-matched normal comparison subjects, while Wirz-Justice et al. (39) attributed circadian dysfunction to medication effects. Genetic associations between circadian genes and SZ/SZA have been examined less frequently. We have reported suggestive associations at *ARNTL*, *PER3*, and *Timeless* (*TIMELESS*), based on initial analyses of SNPs from eight circadian genes among patients with BPI and SZ/SZA and among controls, comprising over 1,450 individuals (40).

Since most published studies investigated disparate groups of SNPs in selected circadian genes, it is uncertain whether the reported results are robust. Here, we report on a systematic genetic association study. An international effort called the HapMap project has set out to identify and catalogue common SNPs across the human genome (http://www.hapmap.org). This effort has helped identify SNPs that are correlated extensively at the population level, i.e., SNPs that are in linkage disequilibrium (LD). Dueto the extensive correlations, it is not necessary to genotype all SNPs across a particular genomic region for an association study. Instead, representative SNPs, or 'tag SNPs', can be selected and genotyped. We used this approach. Common tag SNPs in an extended set of circadian genes were assayed in a large, national sample of BPI cases. A sample of SZ/SZA cases was also included. The SZ/SZA samples were intended not only to identify associations with these disorders, but also to provide comparisons with the BPI associations. Several studies have recently shown overlapping association between BPI and SZ/SZA, which may indicate shared genetic etiology (41–44).

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Methods

Clinical

BPI cases—Genomic DNA samples were obtained from BPI patients recruited through the Systematic Treatment Enhancement Program for Bipolar Disorder (STEP-BD) (45). STEP-BD used a network of 18 US treatment centers for standardized evaluation and treatment of patients, including interview schedules based on the Structured Clinical Interview for DSM-IV (SCID) (46), as well as the Mini-International Neuropsychiatric Interview (MINI) (47).

The selected samples represent a subset of the entire STEP-BD dataset. They were pruned by the following factors: consent for genetic studies, diagnosis of BPI, reported Caucasian ancestry, and samples released by the National Institute of Mental Health Genetics Research Initiative Repository (NIMH-GRI) at the time of the study.

SZ/SZA cases—Unrelated patients with SZ or SZA (according to DSM-IV criteria) were recruited at Western Psychiatric Institute and Clinic, Pittsburgh, PA, USA, and surrounding regions (N = 527; n = 314 SZ and n = 213 SZA). Details of ascertainment and diagnosis are described elsewhere (48, 49). The Diagnostic Interview for Genetic Studies (DIGS), a structured diagnostic interview schedule (50), was used to interview the patients. Additional clinical information was obtained from clinical records and from relatives, as required. Consensus diagnoses were assigned following synthesis of the detailed data.

Control samples—Two sets of adult controls screened for absence of psychosis or BPI were included: a Pittsburgh-based sample and a national sample available from the NIMH-GRI. Only control subjects reporting European-American ancestry were selected, in order to keep the control samples consistent with the case samples and to reduce spurious associations due to population substructure. The controls thus reflected the demographic distribution of the BPI patients, who were recruited at sites across the US, with a substantial proportion being recruited at Pittsburgh. The Pittsburgh-based controls were also suitable for comparison with the SZ/SZA cases. We wished to balance the control sample size to the case samples. The NIMH samples were released by the NIMH-GRI following a request for genomic DNA from individuals reporting Caucasian ancestry.

Pittsburgh controls (n = 168): Members of the local community were recruited through the University Center for Social and Urban Research (UCSUR), notices at a University of Pittsburgh Web site, or through advertisements in the local media. UCSUR is an independent center at the University of Pittsburgh experienced in performing telephone recruitment and screening services. UCSUR staff employed random digit-dialing sampling to ascertain potential control individuals. None of the subjects reported history of substance use disorder within the past month, serious medical or neurological illnesses, or mental retardation as defined in the DSM-IV. In addition to the above inclusion/exclusion criteria, individuals who reported a first-degree relative with psychoses or bipolar disorder were excluded.

NIMH-GRI controls (n = 309): Adults were accessed through a representative database maintained by a commercial firm. Consenting participants were assessed using a Web-based instrument regarding medical history, ancestral background, and previous diagnosis/treatment for SZ, SZA, bipolar disorder, auditory hallucinations, or delusions (51–53). Further details are available on the study Web site (http://www.ncbi.nlm.nih.gov/projects/gap/cgi-bin/study.cgi?study_id=phs000021.v2.p1).

All participants reported Caucasian ethnicity. They provided written informed consent, according to the guidelines of the University of Pittsburgh Institutional Review Board.

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Laboratory

Gene selection—The PubMed database was searched using the key words '*circadian*, *clock*, *genes'*. The search yielded 21 genes in December 2006 (Table 1).

SNP selection—We selected 276 'tag SNPs'; i.e., SNPs that would represent common variants across the selected genes. The regions represented included 5 kilobases (kb) upstream and 5 kb downstream of the coding sequence, from the International HapMap Project Web site (http://www.hapmap.org/). All tag SNPs had minor allele frequencies (MAF) greater than 5% in Caucasians, with correlation between SNPs below a predetermined threshold ($r^2 < 0.8$).

Genotype assays—Genomic DNA was extracted from venous blood samples using the phenol chloroform method or using Puregene DNA extraction kits (http://www1.qiagen.com/Products/GenomicDnaStabilizationPurification/GentraPuregeneBloodKit.aspx). Of the identified tag SNPs (n = 276), 268 could be genotyped successfully using the multiplexed, hybridization-based Illumina Golden-Gate Assay (http://www.illumina.com) (54).

Quality control for genotype assays: Seventeen individuals from the Centre d'Etude du Polymorphism Humain (CEPH) genotyped in the HapMap project were used as 'positive controls', as their genotypes for particular SNPs is known from the HapMap database. In addition, 34 other samples (two/assay plate) were used by Illumina, Inc., as internal controls. Three SNPs from the X chromosome were used for gender checks (rs2040962, rs1108444, and rs988457). Out of 276 SNPs genotyped, 268 SNPS were evaluated further for association (excluding the X chromosome SNPs). Eight SNPs were dropped from analysis: three SNPs because of discordant genotypes with CEPH samples (having two or more discordant), four SNPs failing 100% of genotypes, and rs895683 at *ARNTL* gene having a genotype failure rate over 2.5%. For the remaining 268 SNPs, concordance between our genotype calls and HapMap calls was 100%. The mean genotype call rate was 99.96%. Deviations from Hardy-Weinberg Equilibrium (HWE) were evaluated for each SNP using a global significance threshold of $p > 0.005$.

Statistical analysis

LD was analyzed using Hclust software to select tag SNPs (55). Gene-based tests were conducted initially to test for associations, followed by tests with individual SNPs. For gene-based analyses, we used the (T-max) test of association fitting additive, dominant, and recessive models for each gene (56). SNP-based association analyses for the diagnostic phenotype were conducted using the Cochran-Armitage trend test (57).

Tests of population substructure—We used a variation of the genomic control (GC) method (58–60). Samples in the present report were also genotyped for other studies. In sum, 711 SNPs were genotyped from 64 different genes, i.e., 64 generally independent genomic regions. We performed 10,000 iterations of randomly drawing a single SNP from each of the 64 genes to compare between cases and controls, obtaining a distribution of median chi-square tests. The mean value of this distribution was calculated and was divided by the expected median of a chi-square distribution with one degree of freedom (0.456). Since control samples were obtained from two different geographic regions, we conducted identical analyses between the two control groups to assess within-group substructure. Based on our genomic control analysis (10,000 iterations comparing SNPs from 64 different genomic regions we analyzed), we found no differences between BPI cases and controls ($\lambda = 0.85$). If population substructure leads to 'spurious' case–control differences, an inflation in the chi square value would be expected for the SNPs used for the genomic control

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analyses. We found a relatively small inflation in the test statistics between SZ/SZA cases and controls (λ = 1.08). Since our controls were obtained from two different sources, we also performed similar genomic control analyses between controls from Pittsburgh and the NIMH-GRI controls, and found the inflation factor to be smaller than for the SZ/SZA case/control comparisons (λ = 1.04).

Power analysis—We evaluated the power of our sample to detect an odds ratio (OR) of 1.5 under a dominant model. We tested the assumptions that the risk allele was actually analyzed in our sample or that a surrogate was genotyped at r^2 of 0.8 with the risk loci (similar to our primary analyses). We assessed power using risk allele frequencies of 0.15 and 0.5 in the population, and set the type I error threshold to 0.05.

Results

Demographic details and overlap

The sample included 523 patients with BPI (n = 225 men, 298 women; mean age ± SD = 43.11 ± 12.71), 527 with SZ/SZA (n = 340 men, 187 women; age = 37.31 ± 10.12), and 477 control individuals (n = 229 men, 248 women; age = 46.44 ± 16.72). The BPI cases but not the controls reported here are also part of a published GWAS study (see details below) (15). The SZ/SZA sample included 291 patients reported in earlier analysis (40), but the BPI and control samples are independent of that sample.

Gene-based tests

Nominally significant associations were noted at *EGR3* with the BPI sample under two models. The best fits were a recessive model (χ^2 = 7.248, p = 0.014) and additive model (χ^2 = 7.659, p = 0.02). Nominally significant associations were also noted with the SZ/SZA sample at *NPAS2* under a recessive model (χ^2 = 11.321, p = 0.034). Using a dominant model, a trend for association was noted at *RORB* with the SZ/SZA sample (χ^2 = 9.945, p = 0.056). All these associations are uncorrected for multiple comparisons. (See Supplementary Data Tables S1 and S2).

Single SNP-based analyses

Bipolar I disorder—Nominally significant associations (p = 0.05 or better) were detected at 10 SNPs in six genes when genotype distributions were contrasted at individual SNPs between BPI cases and control individuals using the Trend's test [*ARNTL* SNPs rs7126303 (p = 0.040); *Cry2*: rs1554338 (p = 0.031); *CSNK1E*: rs1997644 (p = 0.024); *EGR3*: rs1996147 (p = 0.006); *NPAS2*: rs17025005 (p = 0.009), rs13025524 (p = 0.016), rs11123857 (p = 0.036); and *RORB*: rs17691363 (p = 0.035), rs10217594 (p = 0.026), rs10491929 (p = 0.023); see Table 2].

Schizophrenia/schizoaffective disorder—Nominally significant associations were detected at eight SNPs across four genes: *NPAS2* SNPs rs13025524 (p = 0.025), rs11123857 (p = 0.038), rs6543004 (p = 0.024); *PER2* SNP rs2304674 (p = 0.033), rs2306472 (p = 0.048); *PER3* SNP rs10462021 (p = 0.036), rs2640909 (p = 0.031); and *RORB* SNP rs10491929 (p = 0.023).

SNPs rs13025524 and rs11123857 (*NPAS2*); and rs10491929 (*RORB*) were nominally associated with both BPI and SZ/SZA (p < 0.05). All SNPs with nominally significant associations were localized to introns, except rs2304672 (*PER2*) and rs10462021 and rs2640909 (*PER3*) (all these SNPs are localized to exons; see Table 2).

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Exploratory analyses

Among the BPI cases, associations between SNPs and body mass index, age at onset, gender, and presence or absence of psychosis were tested. Nominally significant associations were detected across 14 genes (See Supplementary Data Table S3).

None of the above associations remained significant following Bonferoni corrections for multiple comparisons.

Analyses in relation to GWAS

The results from the STEP-BD (BPI) cases and controls were analyzed in relation to two independent GWAS reports that have been published recently (13, 15). The present samples overlapped with the Sklar et al. (15) study, so the present associations were evaluated primarily in relation to the Wellcome Trust Case Control Consortium (WTCCC) study (13). Both GWAS reported on identical Affymetrix GeneChip based assays but reported on slightly different sets of SNPs. Across the genomic regions analyzed here, the Sklar et al. study reported on 173 SNPs and the WTCCC analyzed 217 SNPs. Of the 268 SNPs analyzed here, 40 were reported in the Sklar et al. study (14.9%), and 42 SNPs were analyzed in the WTCCC (15.6%). SNP rs8031897 at *FLJ20516* showed nominal evidence for association with BPI in our study (p = 0.05) and a trend for association in the WTCCC study (p = 0.07). SNP rs7126303 (*ARNTL*) showed nominally significant association in our study (p = 0.040). This SNP is in LD with two other SNPs that were associated with BPI at nominal levels in the WTCCC study [rs3816360 (r^2 = 0.73, p = 0.047), rs7947951 (r^2 = 0.536, p = 0.036)]. Similar associations were not observed in the Sklar et al. (15) study. However, SNP rs1554338 at *CRY2* showed suggestive association with BPI in both our study (p = 0.026) and the Sklar et al. (15) study (p = 0.0124).

Power analysis

Under a dominant model, the sample had 84.6% power to detect a nominally significant association (OR 1.5) for a risk allele with 15% MAF in the population, using a type I error threshold of 5%. For a risk allele with MAF 50%, power declined to 74.7% under the same assumptions.

Discussion

We have systematically analyzed tag SNPs to enable a more comprehensive evaluation of circadian genes than has been reported. To our knowledge, this is the most extensive analysis of circadian gene polymorphisms to date, enabling us to evaluate a number of prior published associations simultaneously. We analyzed BPI cases, as well as SZ/SZA cases, using the same set of controls to evaluate associations with these disorders simultaneously. For associations with the diagnostic groups, we focused on gene-based tests. Such tests take account of correlations between SNPs due to LD and thus enable tests of overall associations in discrete genomic regions. Some novel, nominally significant associations were detected. For example, the gene-based tests indicated associations with BPI sample at *EGR3*, a gene that is involved in the transcriptional regulation of other genes mediating biological rhythms (61). Suggestive associations were also detected with SZ/SZA sample at *NPAS2* and *RORB* genes. Several associations at individual SNPs were also detected at *NPAS2*, which is localized to the frontal cortex, a region that may have a critical role in human circadian function (62). Prior reports have also suggested associations at *NPAS2*, but the patterns of associations noted in the present sample are different, so additional investigations are necessary. Despite the suggestive findings, since no association was significant after correcting for multiple testing, our gene-based and SNP based analyses indicate that individual common polymorphisms of the genes investigated are unlikely to

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confer substantial risk for BPI or SZ/SZA, i.e., ORs greater than 1.5. Replicate analyses using the two independent GWAS BPI samples did show direct replication of the specific SNPs that were suggestively associated with BPI, but the comparisons were limited by the modest overlap of the SNPs analyzed in both studies, although *ARNTL* and *CRY2* genes each had SNPs with suggestive associations ($p < 0.05$) in both the current study and at least one of the GWAS studies. Since our samples overlapped with the GWAS study by Sklar et al. (15), we evaluated our results primarily in relation to the WTCCC study (13).

The circadian gene hypothesis could be investigated through additional analyses. First, the relatively small effect sizes noted here indicate that it may be necessary to evaluate larger samples. Rare polymorphisms or copy number variations were not analyzed. We also did not evaluate haplotypes or epistatic interactions, in order to limit the number of comparisons. Risks due to haplotypes at *BHLHB2*, *CSNK1E*, and C*LOCK* genes have been suggested recently (34). We could not test this association, because only one SNP out of the three composing the relevant haplotype was genotyped in our study. It is also possible that other genes not analyzed in the present study also impact circadian function and are associated with BPI. Though a limited number of quantitative traits related to BPI were analyzed in the present study, it remains possible that there are undetected associations with other circadian traits that may underlie BPI pathogenesis. For example, we and others have replicably found that BPI patients are likely to differ from controls with regard to morningness/eveningness, a measure of preference for a stable, quantifiable measure that reflects preferred circadian phase (64).

In conclusion, we report on systematic analyses of selected circadian gene polymorphisms with BPI and SZ/SZA. Our data do not suggest substantial associations (OR > 1.5) with common polymorphisms of the genes analyzed here. Further studies using related circadian variables may be informative.

Supplementary Material

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Table 1

Details of circadian genes analyzed

Gene	Symbol	SNPs analyzed	Associations with gene-based test[a]		No. of associations with individual SNPs[a]	
			BPI	SZ/SZA	BPI	SZ/SZA
Arylalkylamine N-acetyltransferase	*AANAT*	4	−	−	−	−
Aryl hydrocarbon receptor nuclear translocator-like	BmaL1 *(ARNTL)*	22	−	−	1	−
Aryl hydrocarbon receptor nuclear translocator-like 2	*ARNTL2*	25	−	−	−	−
Basic helix-loop-helix domain containing, Class B, 3	*BHLHB3*	4	−	−	−	−
Circadian locomotor output cycles kaput protein	*CLOCK*	10	−	−	−	−
Cannabinoid receptor 1	*CNR1*	17	−	−	−	−
Cryptochrome 2	*CRY2*	7	−	−	1	−
Casein kinase 1 Delta	*CSNK1D*	1	−	−	−	−
Casein kinase 1 Epsilon	*CSNK1E*	9	−	−	1	−
Casein kinase 2	*CSNK2A1*	7	−	−	−	−
D site of albumin promoter (albumin D-box) binding protein	*DBP*	2	−	−	−	−
Early growth response 3	*EGR3*	3	+	−	1	−
Timeless-interacting protein	*FLJ20516*	5	−	−	−	−
Nuclear factor, interleukin 3 regulated	*NFIL3*	9	−	−	−	−
Neuronal PAS domain protein 2	*NPAS2*	60	−	+	3	3
Nuclear receptor subfamily 1, Group D, Member 1	*NR1D1*	6	−	−	−	−
Period 1	*PER1*	2	−	−	−	−
Period 2	*PER2*	9	−	−	−	2
Period 3	*PER3*	14	−	−		2
RAR-related orphan receptor B	*RORB*	46	−	+	3	1
Timeless	*TIMELESS*	6	−	−	−	−

SNP = single nucleotide polymorphism; BPI = bipolar I disorder; SZ = schizophrenia; SZA = schizoaffective disorder.

'−' = significant association not detected at the $p < 0.05$ level.

'+'= significant association detected.

[a] $p < 0.05$, not corrected for multiple comparisons.

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Table 2

Nominal associations with individual single nucleotide polymorphisms (SNPs)

					Trends test p value	
Gene	**Chromosome**	**Location**	**Minor allele frequency**	**SNP**	**BPI**	**SZ**
ARNTL	11p15	Intronic (A/G)	0.37	rs7126303	0.040	0.738
Cry2	11p11.2	Intronic (A/G)	0.07	rs1554338	0.031	0.411
CSNK1E	22q13.1	Intronic (A/G)	0.50	rs1997644	0.024	0.363
EGR3	8p23-p21	Intronic (A/G)	0.37	rs1996147	0.006	0.193
NPAS2	2q11.2	Intronic (A/G)	0.17	rs17025005	0.009	0.085
		Intronic (A/G)	0.29	rs13025524	0.016	0.025
		Intronic (A/G)	0.32	rs11123857	0.036	0.038
		Intronic (A/T)	0.16	rs6543004	0.139	0.024
PER2	2q37.3	Intronic (A/G)	0.24	rs2304674	0.051	0.033
		mrna-utr (C111G)	0.07	rs2304672	0.342	0.048
PER3	1p36.23	Coding-nonsynonymous[a]	0.17	rs10462021	0.334	0.036
		Coding-nonsynonymous	0.28	rs2640909	0.421	0.031
RORB	9q22	Intronic (A/G)	0.05	rs17691363	0.035	0.983
		Intronic (A/G)	0.26	rs10217594	0.026	0.313
		Intronic (A/G)	0.44	rs10491929	0.023	0.023

BPI = bipolar I disorder; SZ = schizophrenia.

Only individual SNPs with associations at the $p < 0.05$ level using the Trends test are listed.

[a]This SNP is in strong linkage disequilibrium with the SNP rs10462020 (647 Gly/Val) (63).