Research Article

ISSN: 2689-1077

Genetics & Molecular Medicine

Netrin Polymorphisms in Bipolar Psychosis a Replication in an Affective Population

James A Wilcox[1]* and David F Briones[2]

[1]University of Arizona, USA.

[2]Texas Tech University, Health Science Center at El Paso, Texas.

*Correspondence:
James Wilcox, MD, PhD, Department of Psychiatry, University of Arizona, Phoenix AZ, USA., Carl Hayden VAMC, Indian School Road, Phoenix, Arizona, USA.

Received: 15 May 2025; Accepted: 19 Jun 2025; Published: 27 Jun 2025

Citation: James A Wilcox, David F Briones. Netrin Polymorphisms in Bipolar Psychosis a Replication in an Affective Population. Genet Mol Med. 2025; 7(1): 1-4.

ABSTRACT

***Background**: We present a case-control study of single nucleotide polymorphisms of NTNG1 in affective Psychosis. In this paper we examine the occurrence of this isoform variation in cases of psychotic mania compared to a population with nonpsychotic bipolar disorder. Our previous work found NTNG1 variations to be more common among subjects with schizophrenia than in healthy controls. Our current study detected a significant increase in this genetic variation ($p < 0.001$) for psychosis in a group that excluded subjects with schizophrenia. This new study is further confirmation of the association between netrin anomalies and severe neuropsychiatric illness. It also supports the hypothesis that there is a genetic contribution to psychosis that is not unique to the syndrome of schizophrenia.*

Keywords

Axons, Netrins, NTNG1, Psychosis, Affective disorder.

Introduction

Psychotic disorders are common with a combined lifetime prevalence of over 3% [1,2]. Although the precise etiology of most psychotic illness remains elusive, many cases are felt to be associated with a polygenic diathesis. The genetic contribution to schizophrenia and severe affective illness has now been well documented by a number of methods [3,4].

Genome Wide Association Studies (GWAS) have now found several susceptibility loci for these disorders [5,6]. Although most work with netrin anomalies in mental illness has focused on schizophrenia it is important to realize that a significant number of patients with affective illness also suffer from hallucinations, delusions and disordered thought processes [4]. In this study we specifically examine the contribution of netrin variations to the risk of psychosis in bipolar illness.

The cerebral cortex is a highly organized laminar structure and the migration of neurons is precisely guided by extracellular factors. Netrins are molecules responsible for much of the extracellular axon guidance in the central nervous system [7,8]. This coordination of neural development was first well described in C. elegans [9]. Changes in this guidance can modulate mesocorticolimbic dopamine pathways and have been implicated as contributing to the micro-anatomy of neurological disease, including forms of psychosis. It has been postulated that netrin-1 effects might influence the maintenance of synaptic interactions and neuronal plasticity in the mature nervous system. One variety of netrin (NTNGl) has been documented to have several single nucleotide polymorphism variants [10,11]. Such differences in these polymorphisms of have been suggested to contribute to the etiology of schizophrenia in several studies [9-14]. Ohtsuki [11] reported that this isoform variation for NTNG1 is located at 1p13.3. Research by independent authors have found single nucleotide polymorphisms (SNPs) of NTNG1 to be associated with the epigenetic changes generally found in psychosis [12,14-16]. A meta-analysis of studies relating to NTNG1 and psychosis also suggested that the SNP rs4132604 of the NTNG1 gene could be responsible for susceptibility for psychosis in some subjects [17]. This convergent line of research evidence indicates that aberrant axon guidance via netrin nucleotide variation may

increase susceptibility for hallucinations and delusions. This idea is supported by the findings of our previous work [13]. The current study examines the presence of NTNG1 isoforms and their potential contribution to severe bipolar illness. The hypothetical background for our study was the observation that psychosis was found in several conditions (some clearly affective, while others clearly not) that may be related to a common genetic susceptibility for hallucinations and delusions independent of formal thought disorder or mood *per se*. Permission for this study was obtained from the Texas Tech University Health Sciences Center Institutional Review Board.

Methods

A total of 271 cases of mania with psychosis (the presence of hallucinations, delusions or disorganized thinking not associated with delirium or dementia) and 273 controls with bipolar affective disorder, without a history of psychosis were compared. The mean age for subjects was 48 years, with a range from 29 to 60 years. Forty-nine percent of the subjects were male. Cases were diagnosed by two psychiatrists as having bipolar disorder with psychosis by DSM-4 criteria and later confirmed with DSM-5 criteria. Controls were diagnosed as having bipolar disorder, without a history of psychotic features. The controls were matched for age, gender, education, general health and handedness.

Major neurological illness and the diagnosis of schizophrenia were specifically excluded. Since controversy exists regarding the possible affective composition of schizoaffective disorder, this condition was also excluded. In order to make certain that our cases had significant psychotic features, the symptoms of hallucinations and delusional thinking from the Brief Psychiatric Rating Scale (BPRS) were included in the evaluation. Interrater agreement was high on all measures (Kappa = 0.89 for diagnosis, Kappa = 0.92 for BRPS rating). Controls had to have bipolar disorder, but be free from any symptoms of psychosis as defined above (hallucinations or delusions).

No one exposed to hallucinogens or amphetamines was allowed to enroll in the study. Participants signed informed consent forms before beginning the study. All identifying personal data was removed to protect confidentiality. On the basis of previous findings using the transmission disequilibrium test [11-13] and the presence of a restrictive enzyme site, three SNPs (rs4132604-SNP1, rs2218404-SNP2, and rs1373336-SNP3) were genotyped in this study. NTNG1 isoforms were selected based upon the positive findings from previous studies. We used the laboratory method described in our prior work [12,13]. All samples were run blind to diagnosis using standard biochemical methods for temperatures, volume, and primers. The primer sequences for rs4132604, rs2218404 and for rs137336 were the same as we had previously used [13]. A case-control design was used for statistical comparison using SPSS 11.5 (SPSS Inc., Chicago, Illinois, USA). Deviation from the Hardy-Weinberg equilibrium was examined and pairwise linkage disequilibrium was tested as in our previous work.

We used a dichotomous approach for a single variable (the presence of an atypical polymorphism) as a basis for determining the sample size for comparison. In this formula, having the polymorphism of interest would be equivalent to exposure. The sample size calculations were consistent with reviews regarding appropriate statistical power in genetic studies [19,20]. We were able to achieve 80% power and an alpha of 5% for our study. Our subjects were recruited from consecutive clinic visits and hospitalizations in the southwestern United States. All participants self-identified as Caucasian. Seventy percent identified as Caucasian of Hispanic origin (an appropriate demographic in this region).

Results

Strong pairwise linkage disequilibrium was found between the three single nucleotide polymorphisms rs4132604, rs2218404 and rs1373336 as in our previous study. Significant differences in haplotypes containing rs4132604 alleles were found between cases and controls, with GG (P=0.001) and TG (P=0.0001) between rs4132604 and rs2218404, and GGT (P=0.0001) and TGT (P=0.01) among the three SNPs. Our findings in this psychotic affective population were similar to those in our prior work with subjects suffering from schizophrenic psychosis. The allele frequencies of rs4132604 among psychotic patients were significantly different from those among controls (P=0.0001).

Table 1: Genotype and allele frequencies of SNPs on NTNG1 in patients and controls Genotype.

SNPs	Group	Genotype frequency (%)			chi square	p value	Allele frequency (%)		X2	p Value
		AA	AB	BB			A	B		
OR (95%CI)										
Rs4132604	Patients	113 (35.17)	147 (46.03)	55 (17.91)	7.959	0.01	358 (60.20)	260 (39.39)	7.998	0.001
1.439										
	Controls	80 (26.31)	160 (47.8)	79 (27.3)			309 (49.89)	300 (48.93)		
Rs2218404	Patients	229 (73.19)	75 (22.71)	12 (3.21)	1.79	3.71	524 (82.71)	92 (14.01)	0.139	0.598
1.0587										
	Controls	272 (69.79)	86 (26.89)	8 (2.33)			519 (82.08)	98 (13.99)		
Rs1373336	Patients	61 (19.33)	147 (46.12)	104 (32.91)	0.899	0.521	263 (41.09)	354 (52.56)	3.774	0.053
1.4211										
	Controls	65 (21.21)	154 (49.21)	91 (29.71)			279 (43.75)	333 (51.31)		

Table 2: Frequencies of haplotype of SNPs on NTNG1 in patients and controls.

Haplotype frequency (%)							
Haplotype	**rs4132604**	**rs2218404**	**rs1373336**	**Cases**	**Controls**	**x^2**	**p**
1	G	G		55.02	46.31	6.112	0.008
2	T	G		31.17	38.02	5.182	0.012
3	T	G		11.61	15.7	14.601	0.0001
4	G	T		4.07	3.43	1.301	0.802
5	G	G	T	52.4	30.24	13.35	0.0001
6	T	G	C	23.16	24.39	1.591	0.168
7	T	T	C	9.87	9.78	0.279	0.768
8	T	G	T	6.28	9.93	6.943	0.01
9	G	G	C	5.01	7.32	1.913	0.161
10	G	T	C	2.97	3.87	0.409	0.708

Table 3: Multiple Regression Analysis.

	F score	P value	Significance
Age	0.892	> .10455	NS
Gender	0.912	> .45622	NS
Polymorphism	7.69	< .001	Significant
Education	0.567	> .32452	NS
handedness	0.6721	> .23461	NS
General Health	0.23799	>.3345	NS
Positive family history	2.0122	< .06	mildly significant

Discussion

In this pilot study we performed a case-control comparison for netrin isoforms between subjects with bipolar psychosis and a group of matched individuals with bipolar disorder, but no history of psychosis. Our findings revealed a positive association between rs4132604 and psychosis on the basis of both allele (chi square = 7.912, P = 0.005) and genotype (chi square = 7.772, P=0.021) frequency distribution differences between cases and controls. The linkage disequilibrium analysis showed a strong positive association among the several SNPs. The occurrence of allele G was significantly higher than that of allele T in rs4132604'. This suggests that this allelic difference had a contribution toward the susceptibility to psychosis in our subjects (odds ratio= 1.422, 95% confidence interval= 1.102-1.731). Our data suggests that the allele G of rs4132604 might increase the risk for psychotic symptoms in bipolar disorder and that the allele G of rs2218404 and the allele T of rs1373336 may be associated with this risk. This allelic variation is similar to that noted for psychosis in our 2014 study. Since the current study excluded schizophrenia, it suggests that netrin isoforms may contribute to psychosis in purely affective illness as well. Such a finding indicates that netrin variation may represent a significant factor that crosses diagnostic boundaries and could represent an independent risk for psychotic symptoms.

There are methodical limitations to our study. Studies involving symptom self-report are subject to inherent bias. We attempted to reduce this potential source of artifact by using objective rating scales, examination by more than one diagnostician and subsequent review with the Kappa statistic. Another potential confounder involved age. As affective psychosis often manifests later in life than the schizophrenic spectrum, some of our subjects may not have fully escaped the age of risk for possible psychotic decompensation. We did not believe this to be a major issue in our analysis as our sample was reasonably mature with a mean age past 45 years. One should acknowledge the possibility that psychosis in our sample could indicate a dependent effect caused by another, unknown exposure. Such an confounder would seem unlikely given the significant association unique polymorphisms with diagnostic findings in our otherwise case-control model groups. We feel that this preliminary study suggests a potential genetic risk factor associated with netrin variations in psychotic affective conditions.

Summary

Netrins are known axon guidance factors, therefore, allelic differences could account for structural changes seen in the micro-anatomy by way of potential alterations in corticolimbic pathways. Atypical expression of NTNGl could thus be significant in the development of psychotic symptoms. Our findings replicate with those of Zhu et al. [12] and complement those of other studies on netrin variations in psychosis [10-13,15,20]. To the best of our knowledge, this is the first case-control study of NTNG1 variations in a sample of affective disorders.

A growing body of research indicates shared areas of genetic diathesis in a variety of psychotic disorders [2,5,21,22]. Genome Wide Association Studies have found that a number of loci of interest are shared in bipolar illness and schizophrenia [5,6,23].

It is clear that psychotic symptoms occur in a wide variety of severe psychiatric conditions [4,24] and do not always predict decompensation of the personality. Ongoing genetic research should consider that a common underlying link for hallucinations, delusions and disorganization may exist in both affective illness and schizophrenic spectrum disorders. Diagnostic nosology may require more emphasis on the course of illnesses and the occurrence of purely affective symptoms of mood if clinical descriptions are expected to maintain validity. Work on the association between neuroanatomical variations, neurotransmitters and genetic findings is indicated in order to develop more precision in psychiatric

diagnosis and treatment. We encourage more research on netrin variation using different sampling methods, including a family study method and longitudinal evaluations in the future.

References

1. Moreno Kustner B, Martin C, Pastor L. Prevalence of psychotic disorders and its association with methodological issues. A systematic review & meta-analysis. PLOS ONE. 2018; 12: 1-25.
2. Whiteford H, Degenhardt L, Rehm J, et al. Global Burden of disease attributable to mental and substance use disorders: Findings from the Global Burden of Disease Study. Lancet. 2013; 382: 1575-1586.
3. Berrettini W. Evidence for shared susceptibility in bipolar disorder and schizophrenia. Am J Med Genet. 2003; 123C: 59-64.
4. Tamminga CA, Clementz BA. Biological fingerprints for psychosis. Neuropsychopharmacology. 2010; 45: 235-237.
5. Bigdeli T, Fanous A, Li Y, et al. Genome-wide Association Studies of Schizophrenia and Bipolar Disorder in a Cohort of US Veterans. Schizophr Bull. 2021; 47: 517-529.
6. Badner J, Gerson E. Meta-analysis of whole genome linkage scans of bipolar disorder and schizophrenia. Mol Psychiatry. 2002; 7: 405-411.
7. Keleman K, Dickson BJ. Short- and long-range repulsion by the Drosophila Unc5 netrin receptor. Neuron. 2001; 32: 605-617.
8. Matsukawa H, Akiyoshi Nishimura, Lujan J, et al. Netrin-G/NGL Complexes Encode Functional Synaptic Diversification. J Neurosci. 2014; 34: 15779-15792.
9. Ishii N, Wadsworth W, Stern B, et al. UNC-6, a lamina-related protein, guides cell and pioneer axon migrations in C. Elegans. Neuron. 2008; 9: 873-879.
10. Aoki Suzuki M, Yamada K, Meerabux J, et al. A family-based association study and gene expression analysis of netrin-G1 and -G2 genes in schizophrenia. Bio Psychiatry. 2005; 57: 382- 393.
11. Ohtsuki T, Horiuchi Y, Koga M, et al. Associations of polymorphisms in the haplotype block spanning the alternatively spliced axons of the NTNG1 gene at 1p13.3 with schizophrenia from the Japanese populations. Neurosci Lett. 2008; 435: 194-197.
12. Zhu Y, Yang HB, Yuxia, et al. Positive association between NTNG1 and schizophrenia in Chinese Han population. J Genet. 2011; 90: 499-502.
13. Wilcox J, Quadri S. Replication of NTNGI association in schizophrenia. American Journal of Psychiatric Genetics. 2014.
14. Eastwood S, Harrison P. Decreased mRNA expression of netrin-G1 and netrin-G2 in the temporal lobe in schizophrenia and bipolar disorder. Neuropsychopharmacology. 2008; 33: 933-945.
15. Nishimura S, Seki K, Watanabe A, et al. Axonal netrin-G1 regulates NMDA-receptor dependent synaptic functions in selected neural circuits. Abstract Soc Neuroscience. 2004; 723: 19-23.
16. Meerabux J, Ohba H, Masayuki F, et al. Human netrin-G1 isoforms show evidence of differential expression. 2005; 86: 112-116.
17. Yu Y, Fang M, Liu M, et al. Association between netrin G1 genetic variation and schizophrenia: a meta-analysis. Int J Clin Exp Med. 2017; 10: 5985-5994.
18. Fleiss, Joseph L. Statistical Methods for Rates and Proportions. Second Edition. Wiley; 1981: 321.
19. Hong EP, Park JW. Sample Size and Statistical Power Calculation in Genetic Association Studies. Genomics Inform. 2012; 10: 117-122.
20. Fukazawa M, Aoki M, Yamada K. Case-control association study of human netrin G1 gene in Japanese schizophrenia. J Med Dent. 2004; 51: 121-128.
21. Tamminga CA, Ivleva EI, Keshavan MS, et al. Clinical phenotypes of psychosis in the Bipolar Schizophrenia Network on Intermediate Phenotypes (B-SNIP). Am J Psychiatry. 2013; 170: 1263-1274.
22. Schulze T, McMahan F. Psychiatric Genetics. New York, NY, Oxford Press. 2018.
23. Ivleva EI, Bidesi AS, Keshavan MS, et al. Gray matter volume as an intermediate phenotype for psychosis: (B-SNIP). Am J Psychiatry. 2013; 170: 1285-1296.
24. Owen M, Craddock N, Jablensky A. The Genetic Deconstruction of Psychosis. Schizophr Bull. 2007; 33: 905-911.