Psychiatry Research 241 (2016) 263–266

Contents lists available at ScienceDirect

Psychiatry Research

journal homepage: www.elsevier.com/locate/psychres

Study of the tetraspanin 18 association with schizophrenia in a Han Chinese population

Lin Wu [a], Fengping Chen [a], Jun Wei [b], Yan Shen [a], Qi Xu [a,*]

[a] National Laboratory of Medical Molecular Biology, Institute of Basic Medical Sciences & Neuroscience Center, Chinese Academy of Medical Sciences and Peking Union Medical College, Tsinghua University, Beijing, 100073, PR China
[b] Division of Health Research, University of the Highlands & Islands, Centre for Health Science, Old Perth Road, Inverness IV2 3JH, UK

ARTICLE INFO

Article history:
Received 5 February 2016
Received in revised form
7 March 2016
Accepted 15 March 2016
Available online 11 May 2016

Keywords:
Tetraspanin 18
Schizophrenia
Heterogeneity

ABSTRACT

A genome-wide association study of Han Chinese samples identified three single-nucleotide polymorphisms in the tetraspanin 18 (TSPAN18) gene to be associated with schizophrenia. However, the replication of the TSPAN18 association was inconsistent across studies. To explore the possible reason for poor replication, we conducted a case-control study to validate the TSPAN18 finding in an independent Chinese sample. The frequency of rs11038167 minor allele (A) was significantly higher only in female patients with thought disorder. Our result suggested that the TSPAN18 gene may be involved in the development of psychotic symptoms and contribute to clinical heterogeneity of schizophrenia.

1. Introduction

Schizophrenia is a chronic, severe and disabling mental disorder that affects approximately 1% of the population worldwide (Lee et al., 2012). A genetic component involved has been indicated by a high heritability of schizophrenia (0.64 in a national family study and 0.81 in a meta-analysis of twin studies) (Lichtenstein et al., 2009; Sullivan et al., 2003) and by the finding from genome-wide association (GWA) study of the disease (Ripke et al., 2013). Schizophrenia is likely to involve a specific cluster of genes that subsequently determine the individual genetic vulnerability (Lang et al., 2007), although its precise genetic mechanisms remain unclear.

Since the first GWA study was published 9 years ago (Lencz et al., 2007), over 30 GWA studies have been conducted and more than 100 susceptibility loci for schizophrenia have been confirmed (Consortium, 2014). However, replication of some susceptibility loci is not ideal across studies even in the same population (Shi et al., 2011; Yue et al., 2011). A recent GWA study of schizophrenia in a Han Chinese population showed strong association signals at three single nucleotide polymorphisms (SNPs rs11038167, rs11038172 and rs835784) within the tetraspanin 18 (TSPAN18) locus (Yue et al., 2011), but the TSPAN18 association failed to be replicated in a Han Chinese sample from the central area of China (Ma et al., 2013), although Yuan et al. replicated the association of the rs835784A allele with risk of schizophrenia (Yuan et al., 2013). Zhang et al. also genotyped 6 SNPs located in TSPAN18 in an independent Chinese Han sample but found no association with the disease (Zhang et al., 2015). Based on the presence of clinical heterogeneity of schizophrenia, the present study thus investigated the association between schizophrenia and the above SNPs in an independent sample from Han Chinese individuals living in the northern region of China in order to confirm if the TSPAN18 gene was associated with different clinical phenotypes such as positive symptoms (hallucination, delusion, thought disorder and disorders of movement and behavior).

2. Methods

2.1. Subjects

The study sample consisted of 3101 patients (1109 females and 1992 males aged 41.6 ± 11.5 years at time of recruitment) and 3038 healthy controls (1836 females and 1202 males aged 43.0 ± 14.4 years at time of recruitment); their demographic characteristics is given in Table S1. All subjects were of Chinese Han origin and geographically came from the northern region of China. Clinical diagnosis was made by at least two senior psychiatrists according to the criteria of the Diagnostic and Statistical Manual of mental disorders, fourth edition (DSM-IV) for schizophrenia (Fenton, 1996). Clinical information was collected to perform symptom-based association analysis. The control subjects were recruited

* Corresponding author.
E-mail address: xuqi@pumc.edu.cn (Q. Xu).

http://dx.doi.org/10.1016/j.psychres.2016.03.057
0165-1781/

from local communities, and those who had history of major psychiatric or neurological disorders and those who had family history of severe forms of psychiatric disorders, were excluded. Written informed consent was obtained from all subjects or their first-degree relatives in accordance with the Helsinki Declaration. This study was approved by the Ethics Committee of the Chinese Academy of Medical Science and Peking Union Medical College.

2.2. Genotyping of SNPs

Genomic DNA used for genetic analysis was extracted from peripheral white blood cells using a standard phenol-chloroform extraction method. SNPs rs11038167 and rs835784 present in the TSPAN18 locus were genotyped by a TaqMan protocol. The sequences of each primer and probe used are given in Table S2.

2.3. Statistical analysis

The pattern of linkage disequilibrium (LD) was evaluated by HaploView using genotype data from HapMap (HapMap Genome Browser release #28) CHB subjects. D' and r-squared (r^2) measurements were used to estimate LD between rs11038167, rs11038172 and rs835784. The Hardy-Weinberg equilibrium (HWE) for genotypic distributions of SNPs were examined by the chi-square (χ^2) goodness-of-fit test. UNPHASED 3.1 software was applied to analyze the genotyping data for allelic and genotypic associations (Dudbridge, 2008). Four schizophrenia symptoms including hallucination, delusion, thought disorder and disorders of movement and behavior were analyzed in this study. To reduce the false positive rate due to multiple testing, we applied 10,000 permutations to work out a global p-value for the null hypothesis that all the odds ratios (ORs) were equal. We also combined our results with other three studies to conduct meta-analysis of rs11038167 and rs835784.

3. Results

Based on the LD map, rs11038172 is in strong LD with rs11038167 and rs835784 that may represent the LD signals in this haplotype block (Fig. S1). The genotypic distributions of both rs11038167 and rs835784 were in HWE ($p > 0.05$) (Table S3). The χ^2 test showed that neither genotypic nor allelic modeling revealed disease association for rs11038167 and rs835784 (Table S4). We also conducted stratified analysis by gender and age and combination analysis with other three studies, but all failed to show genetic association with schizophrenia (Tables S5 and S6).

Symptom-based analysis showed that the frequency of rs11038167A allele was significantly higher in female patients with thought disorder ($\chi^2=5.93$, $p=0.015$, $OR=1.247$, *95%CI* 1.044–1.490), with a global p-value of 0.031 after 10,000 permutations (Table 1). The recessive model also showed that rs11038167 and rs835784 were associated with schizophrenia in female patients but not in male patients. The rs11038167A allele was associated with delusion ($\chi^2=4.03$, $p=0.045$, $OR=0.891$, *95%CI* 0.796–0.997) but such an association did not survive a correction with 10,000 permutations (global $p=0.081$); the dominant model also showed association of rs11038167 and rs835784 with delusion (Table S7). Gender-stratified samples revealed a gender specific effect on genetic association as the frequency of rs11038167A allele was significantly lower in male patients with delusions, but such an association did not survive a correction with 10,000 permutations neither (global $p=0.076$); the dominant model also showed that rs11038167 and rs835784 were associated with delusion in male

Table 1
Association of rs11038167 and rs835784 with thought disorder.

	With thought disorder		Without thought disorder			
Test model SNP: m/M[a]	***n***	***Freq***[b] **(%)**	***n***	***Freq***[b] **(%)**	***p***[c]	***OR (95%CI)***
All patients						
rs11038167 (A; C)						
Allele: A/C	1335/1745	43.3	1215/1649	42.4	0.474	1.038 (0.937–1.151)
Dominant: (AA+AC)/CC	1043/497	67.7	944/488	65.9	0.296	1.085 (0.931–1.264)
Recessive: AA/(AC+CC)	292/1248	19.0	271/1161	18.9	0.980	1.002 (0.834–1.204)
rs835784 (A; G)						
Allele: A/G	887/2059	30.1	746/1778	29.6	0.656	1.027 (0.914–1.153)
Dominant: (AA+AG)/GG	743/730	50.4	640/622	50.7	0.887	0.989 (0.851–1.150)
Recessive: AA/(AG+GG)	144/1329	9.8	106/1156	8.4	0.213	1.182 (0.909–1.537)
Female						
rs11038167 (A; C)						
Allele: A/C	528/702	42.9	325/539	27.6	**0.015**[d]	1.247 (1.044–1.490)
Dominant: (AA+AC)/CC	407/208	66.2	262/170	60.6	0.067	1.270 (0.984–1.639)
Recessive: AA/(AC+CC)	121/494	19.7	63/369	14.6	0.033	1.435 (1.029–1.999)
rs835784 (A; G)						
Allele: A/G	358/832	30.1	214/574	27.2	0.160	1.154 (0.945–1.410)
Dominant: (AA+AG)/GG	295/300	54.8	189/205	48.0	0.620	1.067 (0.827–1.376)
Recessive: AA/(AG+GG)	63/532	10.6	25/369	6.3	0.022	1.748 (1.085–2.816)
Male						
rs11038167 (A; C)						
Allele: A/C	807/1043	43.6	890/1110	44.5	0.583	0.965 (0.850–1.096)
Dominant: (AA+AC)/CC	632/289	68.8	682/318	68.2	0.793	1.026 (0.846–1.244)
Recessive: AA/(AC+CC)	171/754	18.5	208/792	20.8	0.202	0.864 (0.689–1.082)
rs835784 (A; G)						
Allele: A/G	529/1227	30.1	532/1204	30.7	0.738	0.976 (0.845–1.127)
Dominant: (AA+AG)/GG	448/430	51.0	451/417	52.0	0.696	0.963 (0.798–1.162)
Recessive: AA/(AG+GG)	81/797	9.2	81/787	9.3	0.939	0.987 (0.715–1.364)

[a] m/M, indicates minor allele/major allele.
[b] The minor allele frequency for allelic model, 'mm+mM' frequency for dominant model, and 'mm' for recessive model, where 'm' indicates minor allele, 'M' indicates the major allele.
[c] For Allelic/Dominant/Recessive models, p values were calculated by chi-squared test.
[d] Global p-value of 0.031 from 10,000 permutations.