Int J Clin Exp Pathol 2018;11(12):5969-5980
www.ijcep.com /ISSN:1936-2625/IJCEP0086501

Original Article

Drug-naive patients with schizophrenia have metabolic disorders that are not associated with polymorphisms in the LEP (-2548G/A) and 5-HTR2C (-759C/T) genes

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Received October 9, 2018; Accepted October 25, 2018; Epub December 1, 2018; Published December 15, 2018

Abstract: Schizophrenia is a mental disorder that is primarily caused by polygenic mutations. Schizophrenic patients are more likely to suffer from metabolic syndrome (MS), which is usually accompanied by polymorphisms in the leptin (LEP) gene at the -2548 (G/A) locus and the 5-hydroxytryptamine receptor 2C (5-HTR2C) gene at the -759 (C/T) locus. Hence, we hypothesized an association between these polymorphisms and schizophrenia incidence. A total of 148 drug-naive schizophrenic patients and 165 normal controls were enrolled in the study. Blood glucose levels, lipid levels, and other metabolic markers were measured. MALDI-TOFMS was performed to analyse genotypes of LEP and 5-HTR2C at -2548 (G/A) and -759 (C/T) loci, respectively. Patients with first-episode schizophrenia showed higher levels of fasting blood glucose and the 2-h postprandial glucose (2 hPG), as well as higher insulin resistance indices, but showed lower high-density lipoprotein cholesterol (HDL-C) levels compared to those of the controls. The above results were partly observed when the analysis was performed separately in males and females. Schizophrenic and healthy participants showed no significant differences in the genotypes and allele frequencies in the leptin and 5-HTR2C genes. Patients with varying genotypes of -2548 (G/A) in the leptin gene and -759 (C/T) in the 5-HTR2C gene showed no differences in the indices related to the glucose and lipid metabolism. Taken together, drug-naive schizophrenia patients showed some incidence of metabolic disorders, but polymorphisms in the LEP (-2548G/A) and 5-HTR2C (-759C/T) genes were not associated with schizophrenia or metabolic disorders.

Keywords: Schizophrenia, metabolic syndrome, antipsychotic drugs, leptin, 5-HTR2C

Introduction

Schizophrenia is a complex and disabling mental disorder that presents a series of prominent symptoms, such as hallucinations, delusions, impaired cognition, disorganized speech, and behavioural disturbances [1]. The detailed mechanisms underlying the pathogenesis of schizophrenia remain unclear. However, genetic and environmental risk factors are known to contribute to the occurrence of schizophrenia. Although the global morbidity of schizophrenia is about 1% to 2%, its death rate is about twice that of the general population [2]. Studies have revealed that more than 60% of schizophrenic patients die from physical illness [3], especially cardiovascular diseases, which account for 50% of schizophrenia-related mortality. Schizophrenia is closely associated with metabolic syndrome (MS), which is often accompanied by multiple disorders, such as abnormal glucose metabolism, obesity, dyslipidaemia, and hypertension [4, 5]. Surveys in different regions showed that the prevalence of MS in schizophrenia patients was 37% in Finland, 31.7% in Korea, and 35.5% in Shanghai, China [6-8]. Anti-schizophrenic drugs were thought to be the primary cause of MS, because most of the drugs exert sedative effects, thereby inhibiting movement. However, Thakore et al. previously performed abdominal fat deposition scanning on patients with first-onset schizophrenia and consequently showed that the abdominal fat content of schizophrenia patients averaged 3.4

Table 1. PCR amplification primer list

Rsnumber	SNPloci		Primer sequence
7799039	*-2548G/A*	Upstream (Forward primer)	5'-ACGTTGGATGATCTCAGCACTTAGGGAGAC-3'
		Downstream (Reverse primer)	5'-ACGTTGGATGTCCCGTGAGAACTATTCTTC-3'
		Extended primer	TGAGGCGGGAGGATCAG
3813929	*-759C/T*	Upstream (Forward primer)	5'-ACGTTGGATGCCACGTAATGCTGAGTGCTG-3'
		Downstream (Reverse primer)	5'-ACGTTGGATGGAATCTGCACCACGCTCTTG-3'
		Extended primer	ctCTCTTGGCTCCTCCCCTCATCC

Table 2. Basic information of the participants

	Factor	Patients	Control	*P*-value
Metabolic test	Age	25.51 ± 8.27	24.61 ± 3.10	0.275
	Male:Female	53:47	61:57	0.848
Gene polymorphism test	Age	25.88 ± 6.42	25.40 ± 2.88	0.394
	Male:Female	70:67	71:94	0.162

Note: $P < 0.05$ were considered as significant level when patients compared with the normal group.

times higher that of the normal controls, which suggested that schizophrenia patients already presented metabolic disorders prior to taking anti-schizophrenic drugs [9]. An increasing number of studies have revealed the differences in metabolic, hormonal, and stress-related molecules in the sera of drug-naïve schizophrenia patients and healthy controls [10]. These changes likely contribute to the metabolic abnormalities observed in the schizophrenic patients. The cause of the high incidence of MS in schizophrenic patients remains controversial. However, MS has been established to increase the risk of cardiovascular diseases and mortality in schizophrenic patients.

Leptin is encoded by *leptin* gene (LEP) located on chromosome 7q31.3 and is primarily produced by adipose tissue. Leptin plays an important role in the regulation of energy balance by inhibiting appetite and increasing energy consumption through binding to the leptin receptor in the hypothalamus. Disorders in serum leptin levels are associated with diabetes, obesity, and other metabolic diseases. The LEP-2548G/A polymorphism in the promoter region alters leptin transcription levels and is closely related to lipid metabolism [11, 12]. Ferreirajulio et al. investigated the relationship between LEP-2548G>A and lipid metabolism in 136 obese individuals and 76 normal-weight individuals. Their results showed that the LEP (A/A+G/A) genotypes reduce high-density lipoprotein cholesterol (HDL-C) levels and increase triglycerides (TG) levels in schizophrenic patients [13]. In addition, changes in total cholesterol (TC)/HDL ratio indicated that the LEP-2548G/A polymorphism was associated with dyslipidaemia [14]. In a cohort study, the promoter variant of the leptin gene (-2548G/A) showed very strong association with body weight, height, waist circumference (WC), HDLC levels, and serum leptin levels [15, 16]. Given its close association with abnormal lipid metabolism, the LEP (-2548G/A) polymorphism is also likely to be involved in the pathogenesis of schizophrenia, which is therefore an attractive marker for schizophrenia prediction.

5-Serotonin is a neurotransmitter that plays a role in multiple processes involving brain activity, such as mood, energy, and memory [17]. Reduced levels of secreted 5-serotonin and impairment of its receptors lead to various neurogenic diseases, such as schizophrenia, depression, and migraine [18]. Moreover, genetic polymorphisms in genes encoding the 5-serotonin receptors (5-HTRs) are likely to be associated with MS in schizophrenic patients undergoing antipsychotic treatment. The polymorphism in the promoter region of 5-HTR2C (-759C/T) was found to be significantly associated with waist circumference, fasting glucose levels, and triglyceride levels in blood of the female patients after three months of treatment with antipsychotic drugs (olanzapine/risperidone) [19, 20]. Gunes et al. found that polymorphisms in 5-HTR2C and 5-HTR2A significantly affected BMI but did not affect serum insulin, triglyceride, and cholesterol levels in schizophrenic patients treated with olanzapine

Table 3. Difference of related metabolic markers between the two groups ($\bar{x} \pm s$)

Term	Patient	Control	*t*	*p*
BMI	20.73 ± 2.70	20.96 ± 2.25	-0.692	0.49
WAIST (cm)	76.61 ± 10.60	74.67 ± 9.20	1.445	0.15
WHR	0.82 ± 0.06	0.82 ± 0.07	0.009	0.993
FPG (mmol/L)	4.75 ± 0.74	4.18 ± 0.55	6.539	0.000*
2 hPG (mmol/L)	6.50 ± 1.79	4.98 ± 1.31	7.05	0.000*
Ins (uU/mL)	8.57 ± 4.88	6.57 ± 3.77	1.902	0.06
IRI	1.86 ± 1.06	1.25 ± 0.78	2.494	0.014*
TG (mmol/L)	0.94 ± 0.53	0.99 ± 1.138	-0.376	0.707
TC (mmol/L)	4.01 ± 0.83	4.12 ± 0.84	-0.929	0.354
HDL-C (mmol/L)	1.31 ± 0.32	1.50 ± 0.30	-4.556	0.000*
LDL-C (mmol/L)	2.31 ± 0.70	2.16 ± 0.50	1.796	0.074

Note: BMI: body mass index; WAIST: waist circumference; WHR: waist-hip ratio; FPG: fasting blood glucose; 2 hPG: 2-hour postprandial glucose in OGTT test; Ins: Insulin; IRI: insulin resistance index; TC: total cholesterol; TG: triglyceride; HDL-C: high density lipoprotein cholesterol; LDL-C: low density lipoprotein cholesterol. *$P < 0.05$ compared with the normal group.

Table 4. Difference of related metabolic markers between males of two groups ($\bar{x} \pm s$)

Male	Patients	Controls	t	*p*
BMI	21.20 ± 2.32	22.02 ± 2.19	-1.947	0.54
WAIST (cm)	81.66 ± 9.21	80.80 ± 7.26	0.557	0.579
WHR	0.84 ± 0.06	0.85 ± 0.50	-1.756	0.082
FPG (mmol/L)	4.77 ± 0.77	4.25 ± 0.55	4.13	0.000*
2 hPG (mmol/L)	6.40 ± 1.70	5.01 ± 1.29	4.958	0.000*
Ins (uU/mL)	8.15 ± 7.00	6.99 ± 4.30	1.058	0.28
IRI	1.82 ± 1.93	1.35 ± 0.88	1.633	0.107
TG (mmol/L)	0.97 ± 0.59	1.16 ± 1.43	-0.886	0.378
TC (mmol/L)	4.03 ± 0.75	4.14 ± 0.88	-0.742	0.46
HDL-C (mmol/L)	1.25 ± 0.31	1.45 ± 0.30	-3.546	0.001*
LDL-C (mmol/L)	2.42 ± 0.61	2.21 ± 0.55	1.866	0.065

Note: BMI: body mass index; WAIST: waist circumference; WHR: waist-hip ratio; FPG: fasting blood glucose; 2 hPG: 2 hour postprandial glucose in OGTT test; Ins: Insulin; IRI: insulin resistance index; TC: total cholesterol; TG: triglyceride; HDL-C: high density lipoprotein cholesterol; LDL-C: low density lipoprotein cholesterol. *$P < 0.05$ compared with the normal group.

or clozapine [21]. Kang et al. found no association between genetic polymorphisms in the HTR2C gene and MS in schizophrenic patients taking clozapine [22]. To date, it is unclear whether antipsychotic drugs cause the genetic polymorphisms in the 5-HTR2C gene, thereby resulting in MS, or whether antipsychotic drugs and genetic polymorphisms in the 5-HTR2C gene synergistically trigger MS. In the latter case, schizophrenic patients harbor genetic polymorphisms in the 5-HTR2C gene.

Previous studies highlighted the causal effect of antipsychotic drugs on MS in schizophrenic patients but overlooked the implication of MS in schizophrenic patients who did not undergo treatment with antipsychotic drugs. The present study investigated the association between MS and schizophrenia in patients during their first admission to the hospital. Furthermore, we investigated the involvement of genetic polymorphisms in the LEP (-2548G/A) and 5-HTR2C (-759C/T) genes in schizophrenia pathogenesis.

Materials and methods

Participants selection and ethics statement

A total of 148 schizophrenic patients at their first admission to the hospital and 165 matched health controls were included in the study. Patients were diagnosed with schizophrenia according to the fifth edition of the DSM (DSM-5) released by the American Psychiatric Association in 2013. Healthy individuals reported no past or present psychiatric disorder. All participants provided written informed consent. The present study was approved by the local ethics committee of the Second Xiangya Hospital of Central-south University (Changsha, China).

Measurement of metabolic indicators

Basic data on height, weight, waist circumference, hip circumference, blood pressure, were recorded [23]. Fasting blood glucose (FBG) [24], blood lipid [25], and insulin [26] levels were determined as previously described. Body mass index was calculated as follows: BMI = weight value / height value^2 (kg/cm^2). 2 hPG was measured in oral glucose tolerance test (OGTT). Briefly, after fasting for more than 8 h, 5 ml of elbow venous blood was extracted in the morning, and FBG levels were

Table 5. Difference of related metabolic markers between females of two groups ($\bar{x} \pm s$)

Female	Patients	Controls	*t*	*p*
BMI	20.21 ± 3.02	19.83 ± 1.72	0.751	0.455
WAIST (cm)	70.93 ± 9.15	68.12 ± 5.98	1.805	0.075
WHR	0.79 ± 0.06	0.78 ± 0.07	1.408	0.162
FPG (mmol/L)	4.73 ± 0.72	4.10 ± 0.53	5.165	0.000*
2 hPG (mmol/L)	6.61 ± 1.90	4.94 ± 1.33	5.235	0.000*
Ins (uU/mL)	9.03 ± 12.41	6.13 ± 3.08	1.703	0.092
IRI	1.92 ± 0.79	1.14 ± 0.65	2.03	0.045*
TG (mmol/L)	0.91 ± 0.45	0.80 ± 0.66	0.902	0.369
TC (mmol/L)	3.99 ± 0.93	4.09 ± 0.81	-0.573	0.568
HDL-C (mmol/L)	1.38 ± 0.32	1.56 ± 0.29	-2.292	0.004*
LDL-C (mmol/L)	2.20 ± 0.79	2.11 ± 0.44	0.701	0.486

Note: BMI: body mass index; WAIST: waist circumference; WHR: waist-hip ratio; FPG: fasting blood glucose; 2 hPG: 2hour postprandial glucose in OGTT test; Ins: Insulin; IRI: insulin resistance index; TC: total cholesterol; TG: triglyceride; HDL-C: high density lipoprotein cholesterol; LDL-C: low density lipoprotein cholesterol. *$P < 0.05$ compared with the normal group.

Table 6. Difference of related metabolic markers between the male and female patients ($\bar{x} \pm s$)

	The male	The female	*t*	*p*
AGE	25.21 ± 9.79	25.85 ± 6.21	-0.387	0.7
BMI	21.20 ± 2.32	20.21 ± 3.02	1.869	0.066
WAIST (cm)	81.66 ± 9.21	70.93 ± 9.15	5.832	0.000*
WHR	0.84 ± 0.06	0.79 ± 0.06	3.484	0.001*
FPG (mmol/L)	4.78 ± 0.77	4.73 ± 0.72	0.225	0.822
2 hPG (mmol/L)	6.40 ± 1.70	6.61 ± 1.90	-0.576	0.566
Ins (uU/mL)	8.15 ± 7.00	9.03 ± 12.41	-0.444	0.658
IRI	1.82 ± 1.93	1.92 ± 2.79	-0.231	0.831
TG (mmol/L)	0.97 ± 0.59	0.91 ± 0.45	-0.593	0.554
TC (mmol/L)	4.03 ± 0.75	3.99 ± 0.93	-0.214	0.831
HDL-C (mmol/L)	1.25 ± 0.31	1.38 ± 0.32	-2.058	0.042*
LDL-C (mmol/ L)	2.42 ± 0.61	2.20 ± 0.79	1.539	0.127

Note: BMI: body mass index; WAIST: waist circumference; WHR: waist-hip ratio; FPG: fasting blood glucose; 2 hPG: 2 hour postprandial glucose in OGTT test; Ins: Insulin; IRI: insulin resistance index; TC: total cholesterol; TG: triglyceride; HDL-C: high density lipoprotein cholesterol; LDL-C: low density lipoprotein cholesterol. *$P < 0.05$ compared with the normal group.

measured. Afterwards, each patient was asked to drink 250 ml of warm water containing 75 g of glucose within 5 min. After 2 h, elbow venous blood (2 ml) was extracted for the measurement of 2 hPG levels. FBG and 2 hPG are important indicators of impaired fasting glucose (IFG) and impaired glucose tolerance (IGT). According to the diagnostic criteria of diabetes mellitus of WHO in 1999, impaired fasting glucose (IFG) is defined as elevated fasting glucose that does not meet the diagnostic criteria of diabetes mellitus (6.1 mmol/L < FPG < 7.0 mmol/L). Impaired glucose tolerance (IGT) is defined as an increase in blood glucose levels at 2 h after the OGTT test (7.8 mmol/L < 2 hPG < 11.1 mmol/L). Insulin resistance was evaluated using homeostasis model assessment (HOMA) [27, 28]. HOMA insulin resistance index (HOMA IRI) was calculated as follows: HOMA IRI = fasting insulin (μU/ml) * fasting blood glucose (mmol/l)/22.5.

Genomic DNA extraction, primer design, and genotyping

Genomic DNA was extracted from whole blood following the phenol-chloroform method. The primer (**Table 1**) was designed using Sequenom Mass ARRAY Assay Design 3.1 (Sequenom Inc., USA) and synthesized by Invitrogen Inc. (Shanghai, China). Genotyping of the LEP and 5-HTR2C polymorphisms was performed using matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOFMS) according to a previously described method [29].

Statistical analysis

All data were analysed using SPSS 17.0 software. The *t*-test was used to compare the metabolic indicators between patients and control groups. Pearson correlation test was used to analyse the correlation between indices. Genotype testing was performed based on the Hardy-Weinberg principle [30]. The χ^2 test was used to determine significant differences in the genotypes and allele frequencies of leptin (-2548G/A) and 5-HTR2C (-759C/T) between the first-episode patient group and the normal control group. The differences between different genotypes of leptin (-2548G/A) and 5-HTR2C (-759C/T) in metabolic indicators in schizophrenic patients were determined by t-test or one-way analysis of the variance (ANOVA) followed by Dunnett's post hoc test. $P < 0.05$ was considered statistically significant.

Table 7. Correlation analysis between metabolism indicators

	AGE	BMI	WAIST	WHR	TG	TC	HDL-C	LDL-C
r (FPG)	-0.085	0.024	-0.022	0.111	0.086	0.204	0.148	0.119
p (FPG)	0.403	0.816	0.825	0.27	0.393	0.042*	0.14	0.238
r (2 hPG)	-0.103	0.013	-0.116	-0.092	0.156	0.217	0.012	0.186
p (2 hPG)	0.309	0.898	0.249	0.362	0.122	0.030**	0.908	0.05
r (TG)	0.089	0.237	0.243	0.026				
p (TG)	0.38	0.018*	0.015**	0.799				
r (TC)	0.154	0.254	0.151	0.092				
p (TC)	0.126	0.011*	0.133	0.362				
r (HDL-C)	0.097	-0.055	-0.127	-0.066				
p (HDL-C)	0.338	0.59	0.207	0.513				
r (LDL-C)	0.065	0.275	0.189	0.016				
p (LDL-C)	0.52	0.006**	0.06	0.872				

Note: BMI: body mass index; WAIST: waist circumference; WHR: waist-hip ratio; FPG: fasting blood glucose; 2 hPG: 2 hour postprandial glucose in OGTT test; Ins: Insulin; IRI: insulin resistance index; TC: total cholesterol; TG: triglyceride; HDL-C: high density lipoprotein cholesterol; LDL-C: low density lipoprotein cholesterol. *$P < 0.05$, **$P < 0.01$ compared with the normal group.

Results

Data collection

A total of 218 participants completed the metabolic test, including 100 patients and 118 normal volunteers. A total of 302 participants comprising 137 patients and 165 normal volunteers completed the gene polymorphism test. As shown in **Table 2**, the patients and the controls in the metabolic studies and genotyping studies showed no statistically significant differences in gender and age, which ensured the reliability of subsequent experimental analysis.

Differences in metabolic markers between the two groups

We observed no significant differences in body mass index (BMI), waist circumference (WAIST), waist-to-hip ratio (WHR), and fasting insulin, triglyceride, cholesterol, and low-density lipoprotein cholesterol (LDL-C) levels between the first-episode schizophrenia group and the normal control group (**Table 3**). On the other hand, fasting blood glucose ($P < 0.001$) and 2 hPG levels ($P < 0.001$), as well as IRI ($P < 0.05$), were higher in the first-episode schizophrenia patients. The HDL-C levels were lower in first-episode schizophrenia patients ($P < 0.001$).

Furthermore, we performed independent analysis of the differences in related metabolic markers in males and females. Male patients showed higher levels of fasting blood glucose ($P < 0.001$) and 2-h postprandial glucose (2 hPG) ($P < 0.001$) and reduced HDL-C levels ($P < 0.01$) compared to those of the controls. However, the two groups showed no significant differences in insulin resistance index (**Table 4**). Females showed similar patterns with those of the total participants based on the results of metabolic test (**Table 5**). Male patients showed higher waist circumference ($P < 0.001$) and WHR ($P < 0.01$) but lower HDL-C levels ($P < 0.05$) than the female patients (**Table 6**).

Correlation analysis between metabolic indicators

The present study analysed the correlations of age, BMI, WAIST, and WHR with FBG, 2 hPG, TC, TG, HDL-C, and LDL-C levels. Results showed that BMI was positively correlated with TG ($P < 0.05$), TC ($P < 0.05$) and LDL-C levels ($P < 0.01$, **Table 7**). WAIST was correlated with TG only ($P < 0.01$). The correlations of FBG and 2 hPG with TC, TG, HDL-C, and LDL-C levels were further analysed. Only TC was found to be positively correlated with FBG ($P < 0.05$) and 2 hPG levels ($P < 0.01$).

Differences in genotypes and allele frequencies of polymorphisms in the leptin and 5-HTR2C genes between schizophrenic patients and controls

The mass spectrogram and cluster graph of MALDI-TOFMS analysis are present in **Figures 1**, **2**. The genotype distributions (G/G, A/G, and A/A) in leptin -2548 loci were 6.56, 43.80, and 49.64%, respectively, in first-episode schizophrenic patients, as well as 8.48, 36.97 and 54.55%, respectively, in health controls. The genotype (T/T) in 5-HTR2C -759 loci was not observed in first-episode schizophrenic patients and only 1.21% in health controls. The most genotype was C/C that was observed in 75.91%

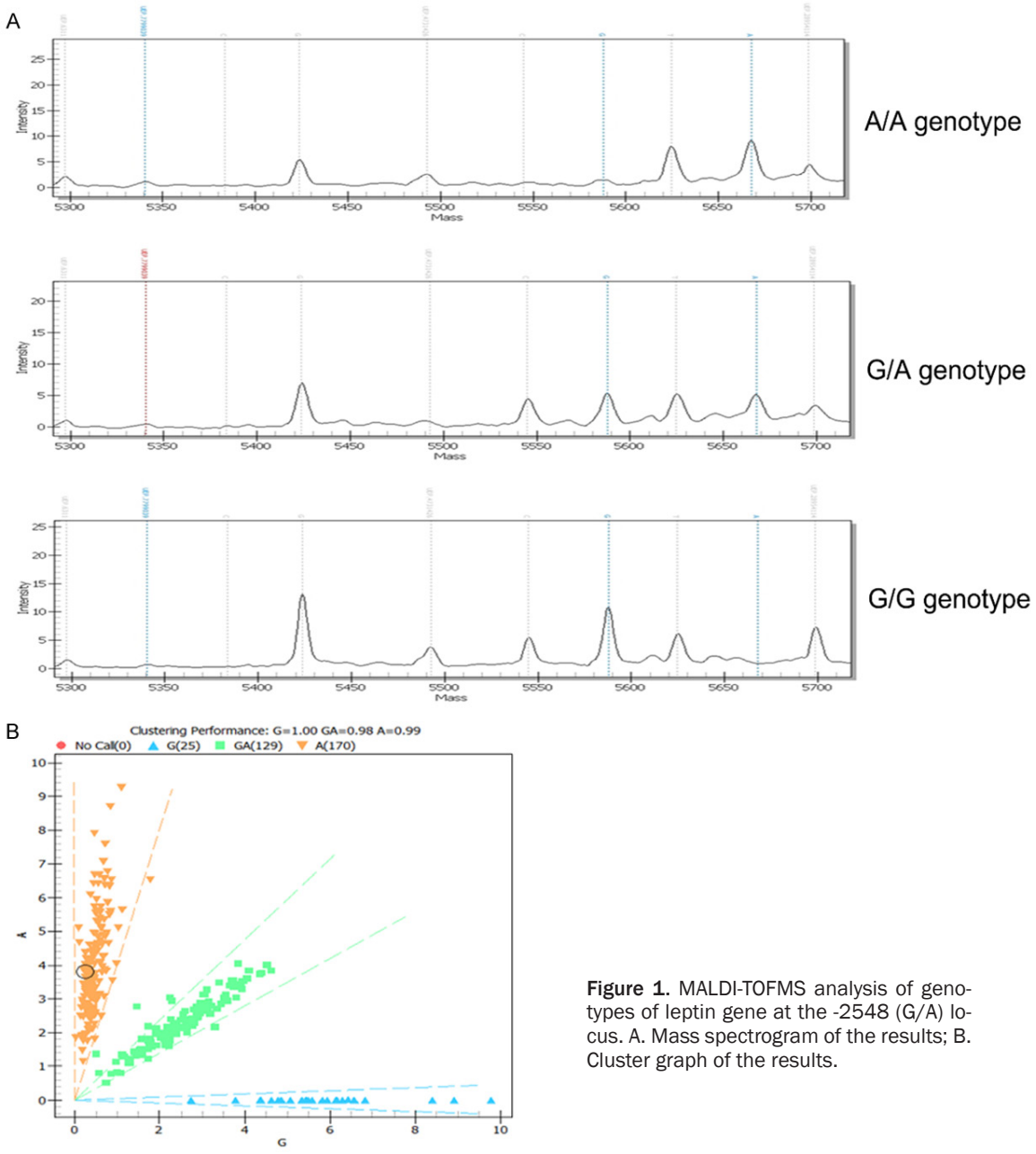

Figure 1. MALDI-TOFMS analysis of genotypes of leptin gene at the -2548 (G/A) locus. A. Mass spectrogram of the results; B. Cluster graph of the results.

of schizophrenic patients and 66.67% of health controls. Results revealed no significant differences in the genotype distributions and allele frequencies in the LEP (-2548G/A) and 5-HTR2C (-759C/T) genes between the first-episode schizophrenic patients and normal controls (**Table 8**).

The levels of metabolic indicators in schizophrenic patients with varying genotypes of leptin and 5-HTR2C genes

As indicated by **Table 9**, patients with varying genotypes of -2548 (G/A) in the leptin gene and -759 (C/T) in the 5-HTR2C gene showed no differences in all these metabolic indices. These suggest that the metabolic changes in schizophrenic patients are not associated with the genotypes of -2548 (G/A) in the leptin gene and -759 (C/T) in the 5-HTR2C gene.

Discussion

Clinical results in the present study showed that FBG and 2 hPG levels were higher in the first-episode schizophrenic patients than in healthy controls. Consistent with our findings, Martina et al. [31] showed that FBG levels (95.8

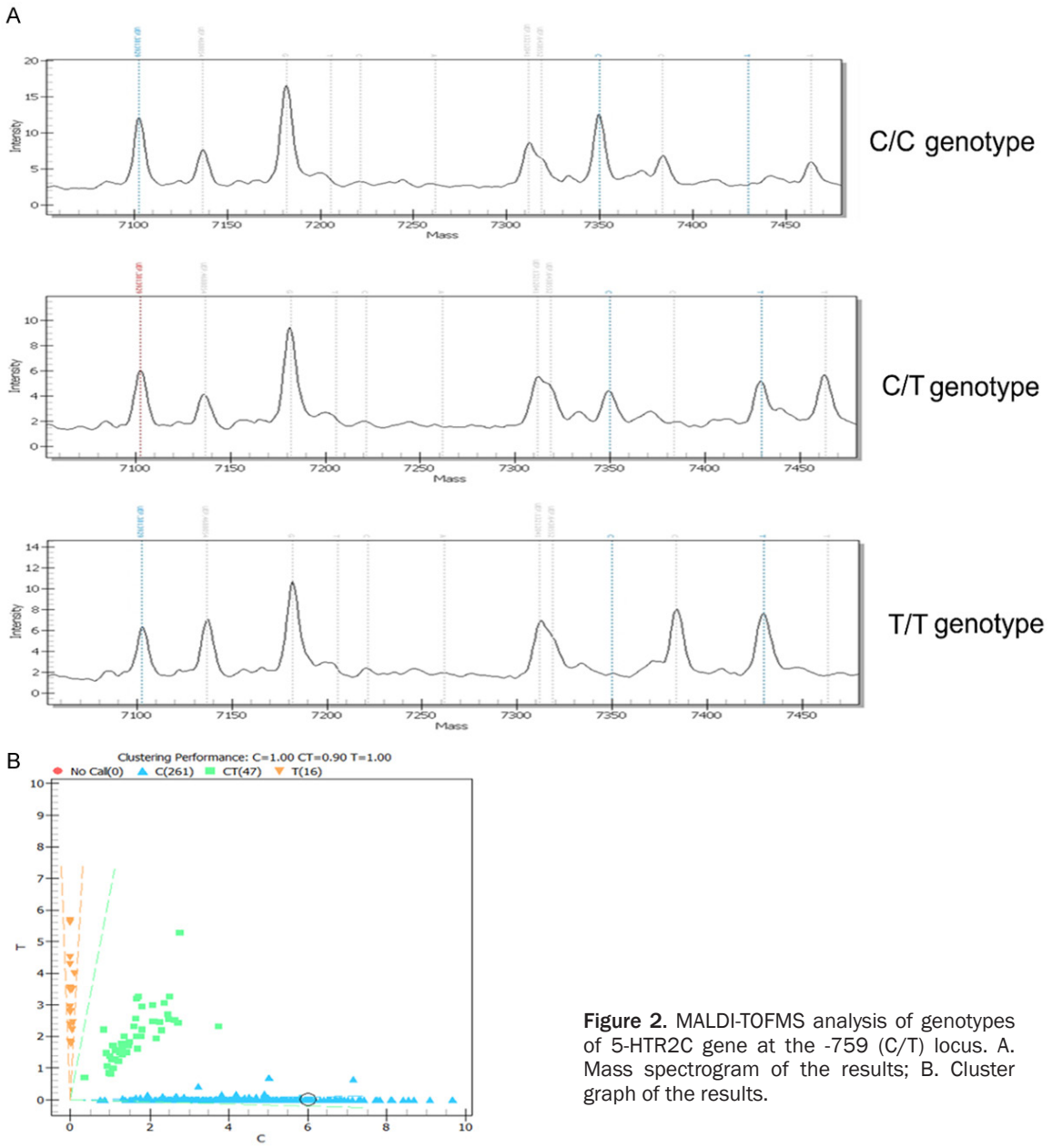

Figure 2. MALDI-TOFMS analysis of genotypes of 5-HTR2C gene at the -759 (C/T) locus. A. Mass spectrogram of the results; B. Cluster graph of the results.

mg/dL) in 26 first-episode, drug-naive schizophrenia patients (average age: 33.6) were elevated relative to those in normal controls (88.2 mg/dL). Spleman et al. [32] showed that 31 drug-naive schizophrenia patients show higher 2 hPG levels and higher incidences of IFG, IGT, and DM (25.8%, 19.3%, and 6.5%, respectively). In the present study, the incidences of IFG, IGT, and DM in schizophrenic patients were 4%, 11%, and 7%, respectively. However, their incidences in healthy controls were only 0%, 2.54%, and 0%, respectively. In addition, the present study was the first to show that IRI was upregulated in the female schizophrenia patients but not in the male schizophrenia patients. Increased IRI is associated with increased risk of type II diabetes mellitus. Taken together, the above findings suggested that glucose metabolism is impaired in schizophrenic patients who did not receive antipsychotic drugs.

A few large-scale studies on lipid metabolism have been conducted in the first-episode schizophrenic patients, although many previous studies focused on the change in lipid

Table 8. Comparison of -2548G/A and-759C/T genotypes and allele frequencies between two groups

Terms			Patient (n = 137)	Control (n = 165)	χ^2	*P*
Genotypes	LEP -2548G/A	*G/G* (%)	9 (6.56)	14 (8.48)	5.878	0.053
		A/G (%)	60 (43.80)	61 (36.97)		
		A/A (%)	68 (49.64)	90 (54.55)		
	5-HTR2C -759C/T	*C/C* (%)	104 (75.91)	110 (66.67)	1.054	0.305
		C/T (%)+*T/T* (%)	33 (24.08) + 0	53 (32.12) + 2 (1.21)		
Allele frequency	LEP -2548G/A	*G* (%)	78 (28.47)	89 (26.97)	0.168	0.682
		A (%)	196 (71.53)	241 (73.03)		
	5-HTR2C -759C/T	*C* (%)	241 (87.96)	273 (82.73)	0.831	0.362
		T (%)	33 (12.04)	57 (17.27)		

Table 9. Levels of metabolic indicators in schizophrenic patients with varying genotypes of leptin and 5-HTR2C genes

	LEP -2548G/A			*P*	5-HTR2C -759C/T		*P*
	G/G	*A/G*	*A/A*		C/C	C/T	
BMI	20.56 ± 1.86	20.82 ± 2.49	20.82 ± 2.94	0.991	20.32 ± 2.47	20.47 ± 4.73	0.849
WAIST (cm)	76.64 ± 8.63	76.44 ± 9.39	77.28 ± 11.84	0.72	70.59 ± 7.38	72.05 ± 12.96	0.663
WHR	0.82 ± 0.02	0.82 ± 0.05	0.81 ± 0.07	0.65	0.78 ± 0.06	0.8 ± 0.07	0.362
FPG (mmol/L)	4.72 ± 0.54	4.67 ± 0.76	4.74 ± 0.75	0.641	4.69 ± 0.7	4.59 ± 0.59	0.708
2 hPG (mmol/L)	6.49 ± 1.2	6.39 ± 1.90	6.43 ± 1.74	0.914	6.74 ± 2.09	5.68 ± 1	0.147
Ins (uU/mL)	6.76 ± 2.62	10.57 ± 14.48	7.49 ± 5.26	0.174	9.79 ± 14.91	7.04 ± 3.64	0.716
IRI	1.98 ± 0.8	2.32 ± 1.44	1.61 ± 1.32	0.227	2.08 ± 3.35	1.64 ± 0.76	0.72
TG (mmol/L)	1.01 ± 0.65	0.87 ± 0.33	0.98 ± 0.65	0.312	0.88 ± 0.43	0.96 ± 0.48	0.645
TC (mmol/L)	3.84 ± 0.88	3.96 ± 0.87	3.99 ± 0.77	0.859	3.92 ± 0.89	4.27 ± 0.89	0.311
HDL-C (mmol/L)	1.28 ± 0.22	1.3 ± 0.32	1.33 ± 0.34	0.671	1.34 ± 0.28	1.52 ± 0.47	0.293
LDL-C (mmol/L)	2.19 ± 0.71	2.23 ± 0.79	2.37 ± 0.64	0.382	2.2 ± 0.83	2.42 ± 0.82	0.504

Note: BMI: body mass index; WAIST: waist circumference; WHR: waist-hip ratio; FPG: fasting blood glucose; 2 hPG: 2 hour postprandial glucose in OGTT test; Ins: Insulin; IRI: insulin resistance index; TC: total cholesterol; TG: triglyceride; HDL-C: high density lipoprotein cholesterol; LDL-C: low density lipoprotein cholesterol. *$P < 0.05$ and **$P < 0.01$ compared with the normal group.

metabolism in patients who received antipsychotic treatment. For example, a clinical antipsychotic trial of intervention effectiveness (CATIE) showed that 64% of the 1,493 patients had varying degrees of hyperlipidemia [33]. Currently, it is unclear whether abnormal lipid metabolism is a side effect of the treatment with drugs or is associated with schizophrenia. In the latter case, abnormal lipid metabolism is likely to result from schizophrenia or abnormal lipid metabolism and, in turn, contributes to the occurrence of schizophrenia. Therefore, determining the correlation between lipid metabolism disorders and schizophrenia is important for understanding the disease pathogenesis. The present study recruited drug-naive schizophrenia patients to eliminate the effects of antipsychotic drugs. Results showed that HDL-C levels in the patient group were significantly lower than those in the control group. This difference remained significant when the male and female patients were analysed separately relative to the male and female controls, respectively. HDL-C is known to have a physiological role in transporting cholesterol from extra hepatic tissues to the liver, thereby preventing the deposition of free cholesterol in extra hepatic tissue cells. Decreased HDL-C level is an independent risk factor for cardiovascular disease and leads to the development of atherosclerosis [34-37]. Therefore, schizophrenic patients are likely to have a higher risk of cardiovascular diseases even without the influence of antipsychotic drugs.

MS is a pathological condition that is characterized by abnormal aggregation of various metabolic components. Patients usually showed

abdominal obesity or overweight, dyslipidaemia (particularly, hypertriglyceridemia), low high-density lipoprotein cholesterol (HDL-C) levels, hypertension, insulin resistance, and/or impaired glucose tolerance [38]. Schizophrenic patients in the present study presented many of the above-mentioned symptoms. According to the International Diabetes Federation (IDF) standard [39], the incidence of MS in the first-episode schizophrenia group was 6% in our study, whereas the corresponding incidence in the controls was 0.08%. Furthermore, our findings showed that the BMI and WAIST of schizophrenia patients were positively correlated with TG, TC, and/or LDL-C levels. In addition, TC was correlated with FBG and 2 hPG levels. The above findings suggested that disorders of both glucose and lipid metabolism could contribute to increased MS incidence.

Leptin plays important role in the regulation of glucose and lipid metabolism [36, 40]. Leptin gene expression is influenced by the polymorphism at the -2548G/A locus in the promoter region [41]. Previous studies have demonstrated that the -2548G/A polymorphism in the leptin gene is significantly associated with weight gain in patients receiving antipsychotic treatments [42, 43]. However, the present study found that the leptin genotypes (G/G+A/G and A/A) at the -2548 locus were not associated with any indicators of glucose and lipid metabolism in the drug-naive patients with schizophrenia. In addition, the patients and normal controls showed no significant differences in genotypes and allele frequencies. However, observed differences in genotype was close to statistical significance ($P < 0.053$). A larger sample of the patients is required in future study to validate the observed differences in genotype.

A few of studies indicated that the -759C/T polymorphism in the 5-HTR2C gene was strongly associated with MS in the schizophrenic patients receiving antipsychotic drugs, such as clozapine, olanzapine, and risperidone [44-47]. However, other studies found no association between -759C/T polymorphism and MS in schizophrenic patients. For example, Theisen found no association between the -759C/T polymorphism in the 5-HTR2C gene and clozapine-induced weight gain among German schizophrenic patients [48]. Furthermore, Kang et al. reported no association between the polymorphism in the 5-HTR2C gene and MS in Korean schizophrenic patients taking clozapine [49]. Taken together, since the above findings involve the influcence of drugs, it is difficult to determine which factors cause a difference in their results. The present study enrolled drug-naive schizophrenia patients and showed that the -759C/T polymorphism in the 5-HTR2C gene was not associated with indicators of glucose and lipid metabolism. Furthermore, we found no significant differences in the genotypes and allele frequencies between the schizophrenic patients and normal controls. Therefore, the association reported in previous studies is likely the result of the effect of the antipsychotic drugs.

The present study provided strong evidence of glucose and lipid metabolic disorders in first-episode, drug-naive schizophrenia patients. Our findings suggested the independent association between MS and the occurrence and development of schizophrenia that is independent of the effects of antipsychotic drugs. However, the -2548G/A polymorphism in the LEP gene and the -759C/T polymorphism in the 5-HTR2C gene showed no association with MS or schizophrenia.

Acknowledgements

This study was funded by the National Natural Science Foundation of China on Evaluation and Related Mechanism of Metabolic Syndrome in Schizophrenic Patients and their First-degree Relatives (NO. 30971051).

Disclosure of conflict of interest

None.

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