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Association of BDNF Gene Val66Met Polymorphism with Suicide Attempts, Focused Attention and Response Inhibition in Patients with Schizophrenia

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ABSTRACT

Introduction: The relationship between BDNF gene Val/Met polymorphism and clinical symptoms, attention and executive functions in patients with schizophrenia was investigated in this study. Also, BDNF Val66Met gene polymorphism was compared between patients and healthy controls. Thus, genetic factors that may affect both the etiology and cognitive functions in schizophrenia were evaluated.

Methods: BDNF Val66Met gene polymorphism was investigated in 102 patients with schizophrenia and 98 healthy controls. Cognitive functions were evaluated by the Wisconsin Card Sorting Test (WCST) and Stroop Test.

Results: There was no difference in terms of the genotypic or allelic distribution of BDNF Val66Met polymorphism between patients and healthy controls. A significantly higher percentage of suicide attempts were found in the patients having Met allele (Val/Met and Met/Met). Met allele was associated with failure in focused attention and response inhibition in patients with schizophrenia.

Conclusion: The presence of the Met allele could be associated with the risk of suicide attempts in patients with schizophrenia. Impairment in executive function areas such as focused attention and response inhibition appears to be related to the Met allele.

Keywords: Schizophrenia, cognitive functions, BDNF, Val66Met polymorphism

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INTRODUCTION

Impairment of cognitive functions is common in patients with schizophrenia, and this impairment is thought to be one of the main features of the disease. Attention impairment restricts success in many other cognitive functions, as attention is the primary step for higher-level cognitive processes (1). There is a disruption in planning and initiating mental activity and information processing speed in patients with schizophrenia. Dysfunctions in planning, maintaining purposeful behavior and behavioral flexibility in patients with schizophrenia indicate the presence of dorsolateral prefrontal cortex damage (2). Brain-derived neurotrophic factor (BDNF) which is associated with neuronal differentiation, proliferation, and synaptic plasticity is known to be an important regulator of cognitive processes such as learning and memory (3).

The BDNF gene is localized at the 11p13 chromosome near p14 border (4). A single nucleotide polymorphism (rs6265: Val66Met) in the sequence encoding the BDNF gene (196th nucleotide guanine-adenine replacement in the codon) results in the replacement of the valine with methionine in the peptide sequence (Val66Met). It is known that Val66Met polymorphism plays a role in intracellular traffic and packaging of precursor BDNF (proBDNF) but not a significant role in the function of the mature BDNF protein. Changes in proBDNF are important for

Highlights

- **There was no difference in genotype and allele distributions of BDNF Val66Met polymorphism between the patient and the control groups which was interpreted as not being a genetic predictor to show a predisposition to schizophrenia.**
- **The Met allele's presence (Val/Met and Met/Met), which means lower activity in the BDNF system, was found to be associated with the risk of suicide attempts in patients with schizophrenia.**
- **Poor performance in the the Stroop test was determined in those having the Met allele compared to Val allele in patients with schizophrenia.**
- **Having a Met allele may deepen the impairment in frontal region functions such as focused attention and response inhibition in patients with schizophrenia.**
- **BDNF gene Val66Met polymorphism has not been found to play a role in areas such as executive functions, perseveration, working memory, conceptualization, and abstract thinking as measured by Wisconsin Card Sorting Test (WCST).**

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synaptic plasticity. This polymorphism affects the intracellular traffic and activity-dependent secretion of BDNF and affects hippocampal functions (5). The relationship of this polymorphism with clinical findings has been an important research subject. In previous studies, Val66Met was found to reduce the age of onset and modify the clinical phenotype of schizophrenia (6). In the literature, differing results have been reported in studies evaluating the relationship between BDNF Val66Met polymorphism and cognitive functions in patients with schizophrenia. Zhang et al. found that the BDNF Val66Met Polymorphism was related to poor visuospatial/constructional performance in patients with schizophrenia. Also, the BDNF Met variant was found to be specific to attentional decrements in patients with schizophrenia (7). On the other hand, Rybakowski et al. reported that the Val66Met polymorphism of BDNF was associated with cognitive functions on WCST in bipolar disorder but not in schizophrenia (8).

These differences may be due to the heterogeneous nature of schizophrenia. These different results reported have led to the need for new studies in this area, including the clinical features and severity of the disease. For this purpose, in our study, whether the clinical severity and characteristics of the disease or cognitive changes are related to the Val66Met allele were examined. Our hypothesis is that patients with schizophrenia with the Met allele in the Val66Met polymorphism may have an earlier age of onset, more severe disease, and worse performance on the WCST and Stroop test. Moreover, by comparing patients with schizophrenia with healthy controls in terms of BDNF Val66Met gene polymorphism, it is aimed to determine genetic factors that may affect the schizophrenia etiology.

METHODS

Our study is an association study based on comparing the results with statistical methods by examining the frequency of an allele in a gene localisation in patients with schizophrenia and the healthy control group. Ethics committee approval was received from Ankara Numune Training and Research Hospital Scientific Research Evaluation Commission with the registration number 2011/295 on 04/01/2012. Written informed consents were obtained from all individual participants included in the study.

Participants

According to DSM-IV-TR, patients admitted to Ankara Numune Training and Research Hospital Psychiatry Outpatient Clinic and diagnosed with schizophrenia were included in the study. A total of 102 patients with schizophrenia in remission who received stable-dose drug therapy in the last six months were included in the study. In order to apply the scales evaluating cognitive performance, individuals with at least five years of education were included in the patient group. Patients with mental retardation, a history of neurological illness, persistent substance use in the last six months, a history of head trauma to cause loss of consciousness, a history of depression in the past six months, or a history of depressive attacks in the past six months were not included in the study. Patients who scored three or less from all of the Positive and Negative Syndrome Scale (PANSS) sub-scores (delusion, unusual thought content, hallucinatory behavior, conceptual disorganization, mannerism/posturing, blunted affect, social withdrawal, lack of spontaneity) were considered in remission (9). Since these patients were monitored in our clinic, outpatient clinical notes were examined from the system. Patients who had not been hospitalized for six months or more, who did not receive treatment changes, and who met the remission criteria according to the PANSS scores recorded at the time of the interview were included in the study.

The control group consisted of 98 healthy volunteers, who agreed to participate in the study and matched in terms of age, sex, and marital status, who were among the healthcare professionals and their relatives working in the same hospital. In addition to the patient group exclusion criteria, those who previously had a history of psychiatric illness or had a first-degree relative with schizophrenia were not included in the control group.

Procedure

Sociodemographic data were obtained by interviewing patients with schizophrenia included in the study. Both clinical interviews and PANSS were performed, and remission status was evaluated. The Calgary Depression Scale for Schizophrenia was used to exclude depression. In the control group, the Structured Clinical Interview for DSM-IV-TR Axis I Disorders (SCID-I) was applied to exclude psychiatric disorders, and the sociodemographic characteristics were recorded. In order to evaluate the cognitive functions of the patients, two basic measurement tools (the WCST and Stroop Test), which were previously used to evaluate cognitive functions and widely accepted in the literature, were used. A 5 cc peripheral blood sample was taken to determine the BDNF gene polymorphism type. Due to the fact that variables such as fasting and sample collection time did not make any difference in genetic analysis, blood samples were taken during working hours when the patients applied to the outpatient clinic, after obtaining their consent. The sample taken was analysed in Hacettepe Teknokent Research and Development Laboratories.

Data Collection Tools

Sociodemographic Data Form: This form includes demographic information such as age, sex, educational status, marital status, residence status, employment status, and alcohol and substance use history. It also includes information such as the age of onset of the disease, duration of the disease, subtype of the disease, the number of hospitalizations, drug treatments used, and family history for the patient group. Duration of disease and duration of untreated psychosis were recorded according to the statements of the individuals and their relatives.

Structured Clinical Interview for DSM-IV Axis I Disorders (SCID-I): SCID-I is a structured clinical interview scale developed by First et al. to evaluate DSM-IV Axis I disorders (10). Adaptation into Turkish, and the validity and reliability study of the scale has been made (11).

Wisconsin Card Sorting Test (WCST): It is developed by Heaton et al. (12), evaluates frontal lobe functions and measures working memory, executive functions and attention performances. Karakaş did a validity and reliability study in Turkey (13). A total of 13 different points are calculated in WCST: Total number of responses (WCST-1), the total number of errors (WCST-2), the total number of corrects (WCST-3), number of categories completed (WCST-4), number of perseverative responses (WCST-5), number of perseverative errors (WCST-6), number of non-perseverative errors (WCST-7), percentage of perseverative errors (WCST-8), trials to complete first category (WCST-9), number of conceptual level responses (WCST-10), percentage of conceptual level responses (WCST-11), failure to maintain set (WCST -2), and learning to learn (WCST-13).

Stroop Test: It measures perception setup, ability to change according to changing demands under a disturbing effect, suppressing a usual behavior pattern, and performing an unusual behavior and attention process (14). In this test, in which scores such as error and response time are calculated, poor performance is manifested by the inability to resist a habitual (or automated) response such as reading, increase in color-naming time or naming the wrong color (13). It consists of five sections and five separate completion times are calculated for each. Stroop-1: the

time to read the black printed color names, Stroop-2: the time to read the color names printed in different colors, Stroop-3 the time to say the color of the printed circles, Stroop-4: the time to say the color of neutral words printed in color, and Stroop-5: the time to say the color of the color names printed in different colors. In addition, Stroop interference was calculated by subtracting Stroop 2 from Stroop 5 and included in the analysis.

PANSS: It is used to measure the general psychopathology and symptom level in patients with schizophrenia and other psychotic disorders. It was developed in 1987 by Kay et al. (15). The validity and reliability in Turkish were completed in 1999 by Kostakoğlu et al. (16).

Calgary Depression Scale for Schizophrenia (CDSS): This scale is used by the interviewer to evaluate depression in patients with schizophrenia and measure depressive symptoms and changes in severity. It was developed by Addington et al. (17). The cut-off value was accepted as 11 for this test, which was used to exclude depression in patients with schizophrenia. Scores lower than 11 were included in the study (18).

DNA Analysis and Determination of BDNF Gene Polymorphisms

A total of 5cc blood samples were taken to 0.5 M EDTA tubes and were stored at -20°C, from the patient group, and healthy controls who were included in the study. DNA Isolation: Ready spin column was made with a DNA isolation kit (Bioteke, DP1802). In addition, 2mM MgCl2 (Bioron, Germany), 0.15 mM dNTP (each) (Larova, 0100) for polymerase chain reaction (PCR) amplification, 0.5 pmol/µl from each primer (Alpha, Canada), 2.5U/µl Hot Start Taq DNA polymerase enzyme (Bioron, Germany) and PCR buffer was used. Primer 1 (Forward) sequence: TGTTTGCAGCATCTAGGTAAT, Primer 2 (Revers) sequence: ATGGGACTCTGGAGAGCGTG. Amplifications were performed at different minutes and were graduated with a 35-cycle program. To determine the Val66Met genotype, amplification products were incubated for 3.5 hours at 37° C with five units/µl Nla III (5'CATG 3 ') restriction enzymes. Then, 246 bp fragments in the enzyme cut were evaluated as BDNF Val/Val genotype; 246, 168 and 78 bp fragments were evaluated as BDNF Val/Met genotype; and 168 and 78 bp fragments were evaluated as Met/Met mutant allele genotype.

Statistical Analysis

The Hardy-Weinberg equilibrium in schizophrenia and normal controls was tested using the χ^2 test for goodness of fit. Shapiro Wilk test was used to check if the distribution of a continuous variable was normal. Descriptive statistics were shown as mean ± standard deviation for continuous variables, and as numbers and percentages for categorical variables.

The Student t-test assessed the significance of the difference between patient and control groups in terms of means. The significance of the difference in median values between patient and control groups and genotype groups was investigated with the Mann-Whitney U-test.

Binary logistic regression analysis was done using clinical and neuropsychological variables to predict a history of suicide attempts. Statistical significance was accepted as $p<0.05$. The data were analyzed in the SPSS for Windows 22.0 package program.

RESULTS

Sociodemographic Results

A total of 69 (67.6%) of the patient group were male, and 33 (32.4%) were female; 62 (63.3%) of the control group were male, and 36 (36.7%) were female. The mean age was 33.8±8.7. The patient and control groups' sociodemographic characteristics are given in Table 1, and there were no statistical differences between the groups except the duration of education (t=2.648 p=0.009).

While 61 patients (59.8%) had no family history of schizophrenia, 41 (40.2%) had a family history of the disease. Twenty nine patients (28.4%) had a history of illness in 1st-degree relatives, and 12 (11.8%) had a history of the disease in 2nd-degree relatives. When the patients' treatments were evaluated, 57 (55.9%) had single antipsychotic use, and 45 (44.1%) had at least two antipsychotic use. While five (4.9%) of 102 patients had typical antipsychotic use, 97 (95.1%) had atypical antipsychotic use. Chlorpromazine equivalent doses for the antipsychotics used, history of suicide attempts in the past and the PANSS scores of the patients are given in Table 1. The mean CDSS score of the patients was found to be 3.04±3.4 (minimum=0, maximum=10).

Cognitive Functions

In terms of WCST-3 (z=-2.112, p=0.035), WCST-4 (z=-3.185, p=0.001), WCST-10 (z=-3.408, p=0.001), WCST-11 (z=-4.038, p<0.001), the scores of the control group were higher than those of the patient group. WCST-

Table 1. Comparison of sociodemographic/clinical characteristics and BDNF gene Val66Met polymorphisms of patients with schizophrenia and healthy controls

Variables	Patients with schizophrenia (n:102)	Healthy controls (n:98)	Statistical analysis
Age (years), mean±SD	34.9±9.0	32.7±8.2	t=1.185 p=0.071
Sex, n (%)			χ^2=0.425 p=0.515
Female	33 (32.4%)	36 (36.7%)	
Male	69 (67.6%)	62 (63.3%)	
Education (years), mean±SD	9.9±3.1	11.2±3.4	t=2.648 p=0.009
Marital status, n (%)			χ^2=3.952 p=0.139
Married	32 (31.4%)	44 (44.9%)	
Single	53 (52.0%)	42 (42.9%)	
Widow	17 (16.7%)	12 (12.2%)	
Employment status, n (%)			χ^2=2.814 p=0.093
Working in a job	40 (40.2%)	50 (50.1%)	
Unemployment	62 (60.8%)	48 (49.9%)	
BDNF gene Val66Met, n (%)			χ^2=0.015 p=0.902
Val/Val	71 (69.6%)	69 (70.4%)	
Val/Met or Met/Met	31 (30.4%)	29 (29.6%)	
Age of disease onset (years), mean±SD	23.1±6.6		
Duration of untreated psychosis (months) , mean±SD	17.2 ± 29.7		
Duration of disease (months), mean±SD	140.6 ± 107.0		
History of suicide attempts, n (%)	31 (30.4%)		
Chlorpromazine equivalent dose (mg), mean±SD	634.3±436.2		
PANSS score, mean±SD			
Positive symptoms	13.0±4.8		
Negative symptoms	18.0±5.6		
General psychopathology	29.5±7.9		
Total	60.5±16.5		

BDNF: brain-derived neurotrophic factor, Val: Valine; Met: Methionine, PANSS: Positive and Negative Syndrome Scale, SD: Standard deviation

2 (z=-3.899, p<0.001), WCST-5 (z=-3.976, p<0.001), WCST-6 (z=-3.883, p<0.001), and WCST-8 (z=-3.879, p<0.001) were higher in the patient group compared to the control group. Comparative WCST scores of the patient and control groups are given in Table 2.

When the patient and control groups are compared in terms of Stroop test scores, Stroop-1 (z=-5.120, p<0.001), Stroop-2 (z=-2.977, p=0.003), Stroop-3 (z=-5.285, p<0.001), Stroop-4 (z=-6.117, p<0.001), Stroop-5 (z=-4.968, p<0.001), the disruptive effect score (z=-5.120, p<0.001), and the Stroop interference score (z=-4.778, p<0.001) were found to be longer in patients with schizophrenia than in the control group (Table 2).

The Genotype Distributions of BDNF Val66Met Polymorphism

In the distribution of BDNF Val66Met polymorphism, Val/Val was found as 70% (140/200), Val/Met 26.5% (53/200), Met/Met 3.5% (7/200). The following values were determined in the patient group: Val/Val:69.6% (71/102), Val/Met:24.5% (25/102), Met/Met:5.9% (6/102). The following values were determined in the control group: Val/Val:70.4% (69/98), Val/Met:28.6% (28/98), Met/Met:1% (1/98). When the patient and control groups were examined according to genotypes, there was no significant difference between the two groups (χ^2=3.69, p=0.158). In the allele distribution for the study participants, Val was 83.3% (333/400) and Met 16.8% (67/400). The following rates were found in the patient group: Val: 81.9% (167/204), Met was 18.1% (37/204); and the following rates were found in the control group: Val: 84.7% (166/196), and Met: 15.3% (30/196). When the patient and controls were examined based on alleles, there was no significant difference between them (χ^2=0.58, p=0.448).

Table 2. Comparison of WCST and Stroop test scores of patients with schizophrenia and healthy controls

Variables	Patients with schizophrenia (n:102)		Healthy controls (n:98)		Statistical analysis
	Median	IQR	Median	IQR	
WCST -total number of corrects	64	23.5	67.5	14.25	z= -2.112 p=0.035
WCST -total number of errors	63	29.25	54	47.25	z= -3.899 p<0.001
WCST -number of perseverative responses	40.5	39	33	29.5	z= -3.976 p<0.001
WCST-total number of perseverative errors	35.5	28.25	29	27	z= -3.883 p<0.001
WCST-total number of non-perseverative errors	22	17.25	18	18	z= -1.308 p=0.191
WCST -percentage of perseverative errors	27.7	22.7	22.6	18	z= -3.879 p<0.001
WCST-number of completed categories	2.7	1.8	3.8	1.9	z: -3.185 p=0.001
WCST-number of conceptual level response	44	30	57	24.75	z= -3.408 p=0.001
WCST-conceptual level response percentage	34.4	23.4	44.5	41.3	z= -4.038 p<0.001
Stroop-1	10	3.3	8.6	2.2	z= -5.120 p<0.001
Stroop-2	10.2	3.3	9.2	2.5	z= -2.977 p=0.003
Stroop-3	14.7	6.7	11.8	3.3	z= -5.285 p<0.001
Stroop-4	20.6	10.7	14.9	5.6	z= -6.117 p<0.001
Stroop-5	31.3	15.8	22.7	9.5	z= -4.968 p<0.001
Disruptive effect score	11.1	3.3	9.6	2.2	z= -5.120 p<0.001
Stroop interference	21.2	15	13.2	8	z= -4.778 p<0.001

*WCST: Wisconsin Card Sorting Test, IQR: Interquartile range, Stroop: Stroop-1: the time to read the black printed color names, Stroop-2: the time to read the color names printed in different colors, Stroop-3 the time to say the color of the printed circles, Stroop-4: the time to say the color of neutral words printed in color, Stroop-5: the time to say the color of the color names printed in different colors.

Clinical Characteristics and BDNF Val66Met Polymorphism in Patients with Schizophrenia

Patients with schizophrenia were compared by grouping the Val/Val and having the Met allele (Met/Met and Val/Met). The history of suicide attempts was found to be statistically significantly higher in patients with schizophrenia having the Met allele (Val/Met and Met/Met), (χ^2=4.592, p=0.032). No differences were found between the two groups for other clinical features including age of disease onset (z=-0.922, p=0.356), family history of psychiatric illness (χ^2=0.056, p=0.813), smoking (χ^2=0.279, p=0.597), PANSS positive (z=-0.973, p=0.330), negative (z=-1.239, p=0.215), general (z=-0.655, p=0.512), and total scores (z=-1.015, p= 0.310).

There was a history of suicide attempts in 31 (30.4%) of 102 patients with schizophrenia. Clinical characteristics, PANSS scores, Stroop, and WCST scores of patients with and without a history of suicide attempts were compared. No difference was found between the values other than the PANSS positive score, Stroop-1, and Stroop-2 values. PANSS positive scores of patients were higher in those with a history of suicide attempts (z=-2.865, p=0.004). Stroop-1 was lower in those with a history of suicide attempts (z=-2.616, p=0.009) whereas, Stroop-2 was higher in those with a history of suicide attempts (z=-1.979, p=0.048). In binary logistic regression analysis to predict the history of suicide attempts in patients, analysis with p<0.05 (met allele status -Val/Met and Met/Met, PANSS positive score, Stroop-1 and Stroop-2) were included as independent variables. The statistical effect of Stroop-2 value could not be demonstrated in the logistic regression model. However, PANSS positive scores, Stroop 1 scores and having Met allele effectively predicted a history of suicide attempts in patients with schizophrenia (Table 3). The last model explained 23.6% (Nagelkerke R2) of the history of suicide attempt variation.

Relationship Between WCST and Stroop Test Scores and BDNF Val66Met Polymorphism in Patients with Schizophrenia

In patients with schizophrenia, WCST scores and subscale scores were compared by grouping the Val/Val and Met allele (Met/Met and Val/Met) genotypes. In terms of the WCST-3 (z=0.721, p=0.471), WCST-2 (z=0.469,

Table 3. Logistic regression model of history of suicide attempts in patients with schizophrenia

	B	Std. Error	Wald	p	Exp(B)
Variables*					
PANSS positive symptoms score	0.12	0.046	6.749	0.009	1.128
Have a Met allele	1.248	0.515	5.867	0.015	3.482
Stroop-1	-0.316	0.148	4.598	0.032	0.729
Stroop-2	0.107	0.105	1.037	0.309	1.113
Constant	-0.892	1.456	0.375	0.540	0.410

*Those who found statistically significant difference in first step analysis (p<0.05), Stroop: Stroop-1: the time to read the black printed color names, Stroop-2: the time to read the color names printed in different colors.PANSS: Positive and Negative Syndrome Scale, Met: Methionine,

Table 4. Comparison of the relationship between Stroop* test scores and BDNF Val66Met polymorphism in patients with schizophrenia

Variables	Val/Val (n: 71)		Val/Met and Met/Met (n:31)		Statistical analysis
	Median	IQR	Median	IQR	
Stroop-1	10.2	3.4	9.9	3.2	z= 0.760 p=0.447
Stroop-2	10.3	4	10	1.5	z= 0.382 p=0.702
Stroop-3	14.5	5.8	17	10.6	z= 2.234 P=0.026
Stroop-4	19.5	11.2	25	7.7	z= 2.103 p=0.035
Stroop-5	29.4	16.4	36.4	19.3	z= 2.202 p=0.028
Disruptive effect score	11.2	3.4	10.8	3.2	z= 0.760 p=0.447
Stroop interference	26.9	18.4	19.1	13.9	z=-2.584 p=0.010

*Stroop: Stroop-1: the time to read the black printed color names, Stroop-2: the time to read the color names printed in different colors, Stroop-3 the time to say the color of the printed circles, Stroop-4: the time to say the color of neutral words printed in color, Stroop-5: the time to say the color of the color names printed in different colors. BDNF: brain-derived neurotrophic factor, Val: Valine, Met: Methionine, IQR: Interquartile range.

p=0.639), WCST-5 (z=0.222, p=0.824), WCST-6 (z=0.211, p=0.833), WCST-7 (z=1.081, p=0.280), WCST-8 (z=0.346, p=0.730), WCST-9 (z=0.465, p=0.642), WCST-10 (z=0.156, p=0.876), WCST-11 (z=0.022, p=0.983), and WCST-12 (z=0.766, p=0.443) scores, there were no statistically significant differences between the groups.

When the two groups were compared in terms of Stroop test scores, there were no statistically significant differences in terms of Stroop-1 (z=0.760, p=0.447), Stroop-2 (z=0.382, p=0.702) and disruptive effect scores (z=0.760, p=0.447).

In patients with schizophrenia carrying the Met allele (Met/Met and Val/Met), Stroop-3 (z=-2.234, p=0.026), Stroop-4 (z=-2.103, p=0.035), Stroop-5 (z=-2.202, p=0.028), and Stroop interference (z=-2.584, p=0.010) were found to be statistically significantly longer than those carrying the Val/Val genotype. The relationship between Stroop test scores and BDNF Val66Met polymorphism in patients with schizophrenia is shown in Table 4.

DISCUSSION

Cognitive Functions

WCST has been the most frequently used test examining executive functions. Many studies have shown that patients with schizophrenia have moderate-to-severe impairments in various parts of this test, such as category and perseverative errors (19). In line with the literature, the patient group performed worse in terms of the total number of right answers, number of completed categories, number of conceptual level responses, and percentage of conceptual level responses. Also, the total number of errors, the total number of perseverative responses, the total number and percentage of perseverative errors were higher in the patient group.

In our study, patients who were in remission and who used stable doses of antipsychotic therapy for at least six months were included, and the possible effects of positive and negative symptoms on cognitive functions were minimized.

Atypical antipsychotics have been shown to improve impaired cognitive functions in schizophrenia. Studies with the first episode and drug-naive patients exclude the antipsychotic effect of Val66Met polymorphism with cognitive impairment in schizophrenia (20).

In our study, prolonged times in all sub-scales of the Stroop test in the patient group indicated executive dysfunction and perseveration tendency in patients with schizophrenia, which is in accordance with previous studies (21).

Since education level, marital status, and working status are likely to affect cognitive functions, patient and control groups were matched for these parameters. Thus, the WCST and Stroop test results used in our study were evaluated independently from these parameters, which is an important advantage of our study.

BDNF Val66Met Polymorphism

Different results have been reported in studies investigating the relationship between BDNF gene Val66Met polymorphism and schizophrenia. In a study, the BDNF gene has been reported to predispose to schizophrenia (22). A meta-analysis conducted in 2,955 patients with schizophrenia found no relationship between BDNF gene Val66Met polymorphism and schizophrenia (23). There was no difference in genotype and allele distributions between the patient and the control groups in our study, which was interpreted as not being a genetic predictor to show a predisposition to schizophrenia.

It has not yet been determined how the Val or Met allele predicts the risk of schizophrenia, and this polymorphism has been shown to create functional differences (5). These functional differences have been suggested to occur with the effects of Val/Met change on the intracellular traffic and activity-dependent secretion of BDNF (24).

In a study, the BDNF gene Val66Met polymorphism was associated with the age of onset of schizophrenia (25). With the hypothesis that the relationship between BDNF Val66Met genotype and age of onset may differ in different ethnic groups, suggesting that this gene affects the onset of schizophrenia in certain races (26), no relationship was found between the age of onset and the genotypes of the BDNF Val66Met polymorphism in our study. In a meta-analysis, another genetic polymorphism of BDNF, C270T, was also mentioned to be a weak but significant risk for schizophrenia (27). For this reason, it may be more meaningful to look at all the genetic polymorphisms of BDNF in a larger sampling in order to determine the relationship between genetic variants of BDNF and the age of onset of schizophrenia.

A difference in BDNF expression was detected in suicide attempts and completed suicide cases before. A meta-analysis of 12 studies found that the Met allele posed a risk for suicidal behavior (28). In our study, the history of suicide attempts was statistically more significant in the group having the Met allele (Val/Met and Met/Met) following these studies. In a prospective study, the relationship between BDNF concentrations and BDNF Val66Met polymorphism and suicide was evaluated, and it was reported that low concentrations increased the risk of suicide. In this study, depressive symptoms decreased, and BDNF concentrations increased after selective serotonin reuptake inhibitor treatment, but no relation was found with the polymorphism mentioned in our study. The absence of the blood BDNF level measurement in our study caused the inability to interpret the relationship between BDNF and clinical results and polymorphism. Monitoring blood BDNF levels during the onset of the disease and before treatment can provide information about the BDNF system's activity. In future studies, adding measurement of BDNF blood concentrations may be more useful in evaluating the results.

The relationship between BDNF gene Val66Met polymorphism and prefrontal cortex performance was examined in bipolar disorder; a significantly better cognitive performance was found in those with

Val/Val genotype in WCST (29). Concerning the BDNF Val66Met polymorphism, people with the Met allele have performed worse in memory tests than the Val allele (5). Despite these studies, in a study comparing BDNF gene polymorphism and prefrontal cortex activity in patients with schizophrenia, no relationship was found between BDNF Val66Met polymorphism and WCST results (8). In accordance with this study, no significant difference was found between Val/Val and Met allele (Met/Met and Val/Met) genotypes in WCST subscale scores in patients with schizophrenia in our study. These different results of the studies may be due to the different tests used to evaluate cognitive functions.

In our study, poor performance in the Stroop test was determined in those having the Met allele compared to Val allele in the patient group, in addition to poor performance in the patient group compared to healthy controls. Having a Met allele may deepen the impairment in frontal region functions such as focused attention and response inhibition in patients with schizophrenia. In a study in the elderly population, Met allele was found to be associated with lower Stroop interference (30). However, a similar study in patients with schizophrenia has not been found in the literature. Supporting the results obtained in our study with new studies will clarify the effects of this polymorphism on cognitive functions in patients with schizophrenia, and determine which areas play a more prominent role.

Our study should be evaluated with some limitations. First of all, serum BNDF levels could not be measured simultaneously in our study. Examining the relationship between genetic variation and blood parameters can make future studies stronger. Second, the sampling needs to be expanded in terms of genetic associations. Finally, no evaluation was made according to the type comorbidities and duration of antipsychotic treatments used by the patients. Antipsychotic use and comorbid diseases, which may have an effect on cognitive functions, may be planned to be included in the evaluation in future studies to work with a more homogeneous patient group.

In conclusion, the Met allele's presence, which means lower activity in the BDNF system, was found to be associated with the risk of suicide attempts in patients with schizophrenia. Impairment in executive function areas, such as focused attention and response inhibition, appears to be related to the Met allele as shown by the Stroop test. However, this functional genetic polymorphism has not been found to play a role in areas such as executive functions, perseveration, working memory, conceptualization, and abstract thinking as measured by WCST. Identifying the factors that determine cognitive functions in patients with schizophrenia will contribute to pharmacotherapy and rehabilitation programs to increase cognitive function. For this purpose, it would be more enlightening to study this polymorphism with other polymorphisms of the BDNF gene and other gene polymorphisms that are thought to be related. Therefore, we believe that multidisciplinary and multicenter studies with a large population of patients are needed in this field.

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