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An Association Between Genetic Variation in the Glutamatergic System and Suicide Attempts in Alcohol-Dependent Individuals

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Background and Objectives: Pathological alterations of glutamatergic systems were observed in neurodegenerative and psychiatric disorders. There is some evidence that this system may be involved in the genetic vulnerability to suicide. The aim of the present study was to analyze possible relationship between the *GRIN2B* polymorphism and suicidal behavior. We hypothesized that this genetic factor may be associated with suicide attempts in alcohol-dependent patients and with death by suicide.
Methods: To analyze the relationship between *GRIN2B* and suicide attempts, the selected rs2268115 polymorphism was genotyped in a sample of 345 alcohol-dependent individuals stratified by the history of suicide attempts. The second part of the study concerning suicide was based on a sample of 510 suicide victims and 450 controls.
Results: The frequency of rs2268115 G allele among alcohol-dependent patients with the history of suicide attempts was significantly higher than among non-suicidal alcohol-dependent individuals (OR = 1.45, $p = .033$). This association was more significant when analyzing alcohol-dependent patients only without co-occurring drug dependence (OR = 1.62, $p = .021$). The analyzed *GRIN2B* polymorphism was associated with a twofold increase in odds of a suicide attempt (OR = 2.01, $p = .004$). No relationships between rs2268115 and death by suicide were identified.
Discussion and Conclusions: Our results suggest that glutamatergic system influence susceptibility to suicide attempts in alcohol-dependent individuals. Suicidal behavior and alcohol dependence may share a common etiology related to the glutamatergic system.
Scientific Significance: The major contribution of the present study is a novel finding of the possible association between *GRIN2B* rs2268115 polymorphism and suicide attempts in alcohol-dependent individuals. (Am J Addict 2017;26:595–601)

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BACKGROUND AND OBJECTIVES

Suicide is a complex behavior with diverse risk factors, which include coexisting psychiatric disorders, addictions, personality traits, somatic comorbidity, and socio-demographics, and demonstrated genetic susceptibility. Alcohol, opioid, and other drugs dependence is consequently considered as an important predictor of suicidal behavior. However, there are a lot of inconsistency when analyzing how this association varies across different types of drugs.[1] Recently, it was reported that risk of suicide attempts is strongest it those with alcohol dependence. Other drug users including opioid, cocaine, stimulants, hallucinogens were less likely to attempt suicide. The lowest likehood of suicidal behavior was linked with cannabis use disorder. This study was performed on a population diagnosed with personality disorders or other psychiatric disorders.[2]

The majority of previous research on the genetic vulnerability to suicidal behavior focused on the serotonergic system, the noradrenergic system and the hypothalamic–pituitary–adrenal axis.[3] The results of the previous studies are often inconsistent and require further clarification. More research is needed to identify, evaluate, and understand the genetic background of suicide behavior.

It seems that the glutamatergic system, which plays an important role in synaptic plasticity and moderates learning, cognition, and memory functions,[4] should be analyzed as another neurosystem influencing vulnerability to suicidal behavior. Glutamate is the major excitatory neurotransmitter in the brain,[5] which operates through metabotropic and ionotropic receptors. The ionotropic receptors are further divided into *N*-methyl-D-aspartate (NMDA) receptors and

non-NMDA receptors.[6] Dysfunctions in the glutamatergic system, especially in NMDA receptors are linked with neurodegenerative disorders, in particular with Alzheimer's disease,[7] Parkinson's disease,[8] Huntington's disease,[9] and epilepsy.[10] There is also growing evidence that the glutamatergic system influences the susceptibility to depression.[4] NMDA receptor is a binding and modulation site for antidepressants.[11] Disrupted function of NMDA receptors was also reported as the possible etiology of schizophrenia,[12] anxiety disorders,[13] neuroticism,[14] antisocial personality disorder,[14] and alcohol dependence.[15]

Pathological alterations of the glutamatergic systems have also been observed in suicide victims.[16] Abnormalities in the gene expression levels of the NMDA receptors had been discovered in patients with depressive disorder who committed suicide.[17]

Recently, Sokolowski et al.[18] performed a family based study with offspring recruited from the emergency department because of severe suicide attempts. They reported the association between the genetic variants in the subset of NMDA receptor (GRIN2B) and suicide attempts. This association was stronger for participants characterized by past year alcohol or drug use disorders, but not by alcohol or drug use at the time of suicide attempt. To the best of our knowledge, this association has not been investigated in other populations.

The purpose of the present study was to further investigate the association between the *GRIN2B* rs2268115 polymorphism and suicidal behavior. We investigated a sample of alcohol-dependent individuals with history of suicide attempt to verify our hypotheses that *GRIN2B* variants might be involved in the vulnerability to suicide attempt, particularly in alcohol or drug-dependent individuals. Additionally, we aimed to analyze the possible relationship between the *GRIN2B* rs2268115 polymorphism and death by suicide.

METHODS

The first part of the study concerning suicide attempts was based on a sample of 345 alcohol-dependent individuals recruited at the abstinence-based, drug-free treatment programs, and in the methadone maintenance treatment clinic in Warsaw, Poland. This cohort consisted of 225 alcohol-dependent patients (with no comorbid drug use disorders) and 120 alcohol and opioid-dependent subjects (with comorbid stimulant use disorder—74% and cannabis use disorder—86%). The diagnosis of alcohol or drug dependence was established after psychiatric examination. In addition, the MINI International Neuropsychiatric Interview was used to diagnose addictions and co-occurring psychiatric disorders.

Lifetime history of suicide attempts as the key dependent variable was evaluated with Suicidality Module of the MINI. The methods of suicide attempts were classified as violent methods: hanging, shooting, jumping from heights, cutting, drowning, other methods, and non-violent suicide method: poisoning (as listed in Chapter X by the ICD-10 classification). Additionally, patients were asked to complete questionnaire that included questions about previously reported risk factors for suicide attempts including demographics, somatic and psychiatric comorbidity, and economic status. To assess the severity of alcohol dependence the Michigan Alcohol Screening Test (MAST) was used. Depressive symptoms were evaluated with the Beck Depression Inventory (BDI). The Barratt Impulsiveness Scale (BIS-11) was used to assess impulsivity. Social support was examined using the Medical Outcomes Study Social Support Survey (MOSSSS). The characteristics of alcohol-dependent participants are presented in Table 1.

To be eligible for the study, participants had to be over 18 years old. The exclusion criteria for the participation were inability to understand the purpose of the study and to give informed consent.

The second part of the study concerning death by suicide was based on a sample of 510 suicide victims autopsied from 2005 to 2013 in the Department of Forensic Medicine at the Medical University of Warsaw, Poland. Suicide as a cause of death was confirmed during post-mortem examinations and finally verified in prosecutor's inquiries. Characteristics of suicide victims, which included basic demographic data, methods of suicide, and blood alcohol concentration (BAC) at the time of death, were derived from post-mortem medical and forensic examination protocols. The majority of cases were males ($n = 440$, 86.3%). The mean age at death was 42.8 ± 16.3 years. In this sample, 44.7% ($n = 228$) of suicide victims had BAC $> .2‰$. Those cases were classified as suicides under the influence of alcohol at the time of death, which is in accordance with Polish law. The average BAC in this group was $1.7 \pm .9‰$. Over 97% of the sample ($n = 496$, 97.3%) committed violent suicide. Hanging was the most frequent suicide method ($n = 416$, 81.6%). These demographic variables of suicide decedents were comparable to Polish national data on suicide.[19]

The comparison group consisted of 450 individuals tested for disputed paternity at the Department of Forensic Medicine as described previously.[20] The male frequency was 52.9% ($n = 238$), mean age was 36.9 ± 10.0 years. This cohort was previously reported as representative for population of Poland.[21]

All participants were Polish Caucasians. Since, according to National Census of Population and Housing, Poland was nearly ethnically homogeneous (97.7% of the population declared Polish ethnonationality), race composition was not additionally analyzed. Alcohol-dependent patients and individuals tested for disputed paternity gave written consent for participation in the study and for the anonymous use of their DNA for research. The study was approved by the Bioethics Committee of the Medical University of Warsaw.

Genotyping

DNA was isolated from blood or saliva samples using standard salting procedures. The selected polymorphism rs2268115 in *GRIN2B* was genotyped by the real-time PCR

TABLE 1. Characteristics of alcohol-dependent participants

Variables	All alcohol-dependent patients, $n = 345$	Alcohol-dependent patients without co-occurring opioid dependence, $n = 225$	Alcohol-dependent patients with co-occurring opioid dependence, $n = 120$	Comparison between alcohol-dependent patients with and without co-occurring opioid dependence, χ^2/U, p-value
Lifetime suicide attempt; *n* (%)	89 (25.8)	63 (28.0)	26 (21.7)	1.64, .20
Violent suicide method; *n* (%)	38 (11.0)	27 (12.0)	11 (9.0)	.25, .62
Gender: male/female (% males)	284/97 (71.9)	161/64 (71.6)	87/33 (72.5)	.035, .85
Age; mean ± SD	40.2 ± 9.8	43.5 ± 9.9	34.2 ± 5.5	5,856, <.001
Severity of alcohol dependence (MAST); median, IQR	29 (16–38)	34 (29–41)	12 (9–19)	2,623, <**.001**
Social support (MOSSSS); median, IQR	64 (49–79)	60 (46–74)	74 (55–82)	9,749, <**.001**
Impulsivity (BIS); median, IQR	69 (64–75)	72 (66–78)	65 (61–70)	7,278, <**.001**
Severe depression (BDI: 29 or more); *n* (%)	62 (18.1)	45 (20.2)	17 (14.3)	1.82, .18

$p < .05$ were boldfaced.

(polymerase chain reaction) using Open Array Technology. Pre-designed primers were obtained from Applied Biosystems (Pre-designed TaqMan SNP Genotyping Assays, 7500 Real Time PCR System, Applied Biosystems, Assay ID C_2682138_1) on an ABI PRISM 9700 platform (Applied Biosystems). The results were analyzed using 7500 System SDS Software (Applied Biosystems).

Among alcohol-dependent patients the genotyping success cell rate was 98.51% (340 samples + 5 undetermined samples), among suicide victims—98.6% (503 samples + 7 undetermined samples), among comparison subjects—99.8% (449 samples + 1 undetermined sample). The study had the power of .8 to detect at $\alpha = .05$ an allelic association conferring OR ~ 1.62.

Statistical Analysis

Genotyping results were tested for Hardy–Weinberg Equilibrium using the Web-Assotest program (http://www.ekstroem.com/assotest/assotest.html). This program was also applied for analyzing genotype distribution in cases and controls assuming dominant, co-dominant and recessive model of inheritance. *p*-Value for model fit (*pfit*) determined the most likely model of inheritance.

Comparison of the genotype and allele distribution after adjusting for age and gender was performed by multiple logistic regression analysis using SPSS software, v. 20.0. Continuous variables were checked for normal distribution by the Kolmogorov–Smirnov test. Differences between analyzed groups with and without a history of suicide attempts were estimated using chi-square tests for categorical variables and the Mann–Whitney *U* tests for continuous non-parametric variables. Logistic regression was applied to examine the adjusted associations between analyzed characteristics and suicide attempts.

RESULTS

The genotype distribution in the analyzed polymorphism rs2268115 in *GRIN2B* was in Hardy–Weinberg equilibrium among suicide victims, controls, and alcohol-dependent individuals (Table 2).

The frequency of G allele among alcohol-dependent individuals with the history of suicide attempts (50.6%) was higher than among non-suicidal alcohol-dependent patients (41.3%, OR = 1.45, CI: 1.03; 2.06, $p = .033$, Table 2).

TABLE 2. Distribution of genotypes of GRIN2B rs2268115 polymorphism and analysis of the association between the analyzed polymorphism and completed suicide or suicide attempt

	Completed suicide		Suicide attempt in alcohol-dependent individuals					
			All alcohol-dependent patients, $n = 340$		Alcohol-dependent patients without co-occurring opioid dependence, $n = 224$		Alcohol-dependent patients with co-occurring opioid dependence, $n = 116$	
Suicide attempt	Suicide victims, $n = 503$	Controls, $n = 449$	Yes, $n = 87$	No, $n = 253$	Yes, $n = 63$	No, $n = 161$	Yes, $n = 24$	No, $n = 92$
HWE p	.647	.558	.454	.634	.402	.382	.889	.703
Genotypes (%)								
G/G	99 (19.7)	86 (19.1)	24 (27.6)	45 (17.8)	20 (31.8)	31 (19.3)	4 (16.7)	14 (15.2)
G/T	254 (50.5)	214 (47.7)	40 (46.0)	119 (47.0)	28 (44.4)	73 (45.3)	12 (50.0)	46 (50.0)
T/T	150 (29.8)	149 (33.2)	23 (26.4)	89 (35.2)	15 (23.8)	57 (35.4)	8 (33.3)	32 (34.8)
Allele (%)								
G	452 (44.9)	386 (43.0)	88 (50.6)	209 (41.3)	68 (54.0)	135 (42.0)	20 (41.7)	74 (40.2)
T	554 (55.1)	512 (57.0)	86 (49.4)	297 (58.7)	58 (46.0)	187 (58.0)	28 (58.3)	110 (59.8)
Allelic comparison OR (CI), p	1.08 (.90; 1.30) .405		1.45 (1.03; 2.06) **.033**		1.62 (1.07; 2.46) **.021**		.94 (.49; 1.78) .855	
Models of inheritance								
Recessive OR (CI)	1.03 (.75; 1.43)		1.78 (1.00; 3.11)		1.75 (.90; 3.41)		1.11 (.33; 3.76)	
p, p fit, GG vs GT/TT	.837, .267		.056, .373		.089, .153		.862, .934	
Co-dominant OR (CI)	1.08 (.90; 1.30)		1.43 (1.02; 2.01)		1.57 (1.05; 2.34)		1.06 (.55; 2.05)	
p, p fit, GG vs TG vs TT	.394, .459		**.038**, .692		**.027**, .812		.853, .959	
Dominant OR (CI)	1.17 (.89; 1.54)		1.51 (.88; 2.60)		1.95 (1.01; 3.77)		1.07 (.41; 2.76)	
p, *p fit*, GT/GG vs TT	.264, .861		.130, .142		.050, .298		.894, .890	

$p < .05$ were boldfaced.

Analysis of genotype distribution in rs2268115 also revealed the association between this polymorphism and suicide attempts in alcohol-dependent patients. Co-dominant model of inheritance had the best fit (OR = 1.43, CI:1.02; 2.01, $p = .038$).

Those relationships were stronger when analyzing only alcohol-dependent patients without co-occurring drug dependence: both in the allelic comparison (OR = 1.62, CI:1.07; 2.46, $p = .021$) and in the analysis of models of inheritance (co-dominant model: OR = 1.57, CI:1.05; 2.34, $p = .027$).

No association between rs2268115 and suicide attempt was found when analyzing only alcohol-dependent patients with co-occurring opioid dependence. To understand those findings, the possible differences between the two groups of alcohol-dependent patients with or without co-occurring opioid dependence were examined. We found that alcohol-dependent only individuals significantly differed from subjects with co-occurring opioid dependence: they were older ($p < .001$, Table 1), had higher scores in MAST ($p < .001$), were more impulsive according to BIS-11 ($p < .001$), and had lower social support as measured by MOSSSS ($p < .001$).

Phenotype-oriented analyses performed in the whole sample of alcohol-dependent individuals revealed correlations between suicide attempts and worse social support ($p < .001$, Table 3), impulsivity ($p < .001$) as well as severe symptoms of depression ($p < .001$). The association between rs2268115 and suicide attempts in alcohol-dependent sample remained significant after controlling for the above-mentioned variables included in the logistic regression model (OR = 1.67, CI:1.14; 2.46, $p = .008$; Table 4).

Given the stronger association between rs2268115 and suicide attempts in alcohol-dependent patients without co-occurring drug dependence, phenotype-oriented analyses were performed within this group to explore this relationship further. Patients who tried to commit suicide in this group were older ($p = .008$, Table 3), more impulsive ($p < .001$), had worse social support ($p < .001$), were more likely to suffer from severe depression ($p < .001$). To analyze risk factors for suicide attempts in alcohol-dependent individuals without

TABLE 3. Comparison between alcohol-dependent patients with and without history of lifetime suicide attempts

Variables	All alcohol-dependent patients, $n = 345$			Alcohol-dependent patients without co-occurring opioid dependence, $n = 225$			Alcohol-dependent patients with co-occurring opioid dependence, $n = 120$		
Lifetime suicide attempt	Yes, $n = 89$	No, $n = 256$	χ^2/U, p value	Yes, $n = 63$	No, $n = 162$	χ^2/U, p value	Yes, $n = 26$	No, $n = 94$	χ^2/U, p-value
Gender: male/female (% males)	62/27 (69.7)	186/70 (72.7)	.75, .39	43/20 (68.3)	118/44 (72.8)	1.14, .29	19/7 (73.1)	68/26 (72.3)	.23, .88
Age; mean ± SD	39.2 ± 9.5	40.6 ± 10	41,680, .29	41.3 ± 10	44.3 ± 10	17,018, **.008**	34.2 ± 7	34.2 ± 5	4,336, .85
Severity of alcohol dependence (MAST); median, IQR	29 (18–39)	30 (15–38)	40,618, .22	34 (28–40)	34 (29–42)	18,768, .33	14 (10–20)	12 (9–18)	3,796, .16
Social support (MOSSSS); median, IQR	54 (41–72)	69 (52–71)	30,128, **<.001**	51 (37–69)	63 (49–76)	14,428, **<.001**	59 (45–73)	77 (59–85)	2,860, **<.001**
Impulsivity (BIS); median, IQR	73 (66–80)	68 (63–73)	31,226, **<.001**	75.5 (69–82)	71 (64–76)	13,466, **<.001**	66.5 (63–70)	65 (61–69)	3,852, .21
Severe depression (BDI: 29 or more); n (%)	35 (39.8)	27 (10.6)	76.5, **<.001**	28 (45.2)	17 (10.6)	65.8, **<.001**	7 (26.9)	10 (10.8)	9.96, **.002**

$p < .05$ were boldfaced.

co-occurring drug dependence logistic regression was performed. The analyzed *GRIN2B* polymorphism was associated with a twofold increase in odds of a suicide attempt (OR = 2.01, CI:1.25; 3.24, $p = .004$, Table 4).

No significant differences in either genotype or allele distributions were found between suicide victims and comparison group (Table 2). Phenotype-oriented analysis performed in these groups also did not reveal any associations between analyzed characteristics including sex, age, method of suicide, or death under the influence of alcohol.

To exclude the possible association between rs2268115 and alcohol dependence, the comparison between the alcohol-dependent group from the first part of the study and the control group from the second part of the study was performed. No association was detected ($p > .05$).

DISCUSSION AND CONCLUSIONS

The major finding of the present study is the association between *GRIN2B* rs2268115 polymorphism and suicide attempts in alcohol-dependent individuals. This relationship was stronger when analyzing alcohol-dependent patients only, without co-occurring opioid dependence. The investigated polymorphism was associated with a twofold increase in odds of suicide attempt in this population.

To the best of our knowledge, prior work did not examine the association between *GRIN2B* rs2268115 polymorphism and suicide attempts in alcohol-dependent patients. Our findings are consistent with previous research,[18] which analyzed relationship between rs2268115 and suicide attempts in general population. Interestingly, in that study the *GRIN2B* rs2268115 association was stronger in a subgroup with diagnosis of alcohol or drug use disorders. Consistently with our work, Sokolowski et al. also reported the minor allele of rs2268115 as a risk factor for suicide attempts.

The glutamatergic dysfunctions were previously reported as involved in psychopathology of suicide. Prior research described the alterations in NMDA receptor complex in the frontal cortex in suicide victims.[22] The differences in cortical NMDA receptors expression were demonstrated in suicide decedents.[16] Higher expression levels of *GRIN2B* were detected in patients with major depressive disorder who committed suicide.[23] Additionally, since NMDA-receptor antagonist ketamine was reported to reduce suicidality in depressed patients, this signaling pathway is considered as a potential part of the pathophysiology of suicidal behavior.[24]

Given above-mentioned results, the second part of our study was aimed to examine the hypothesis that the selected *GRIN2B* polymorphism may be associated with death by suicide. However, our results did not confirm this assumption. Previous genome-wide association studies (GWAS) regarding suicide attempts or suicides also did not report the associations with *GRIN2B*.[25] It seems important to emphasize that, to the best of our knowledge, none of these studies were conducted

TABLE 4. Logistic regression model of factors associated with a history of suicide attempts in alcohol-dependent patients

Variable	Odds ratio (95%CI)	*p*
All alcohol-dependent patients[a]		
GRIN2B rs2268115 polymorphism	1.67 (1.14; 2.46)	**.008**
Impulsiveness (BIS)	1.03 (1.01; 1.05)	**.009**
Social support (MOSSSS)	.98 (.97; .99)	**<.001**
Severe depression (BDI)	4.11 (2.61; 6.47)	**<.001**
Age	.98 (.97;1.00)	.61
Alcohol-dependent patients without co-occurring opioid dependence[b]		
GRIN2B rs2268115 polymorphism	2.01 (1.25; 3.24)	**.004**
Impulsiveness (BIS)	1.04 (1.01; 1.07)	**.009**
Social support (MOSSSS)	.98 (.97; .99)	**.021**
Severe depression (BDI)	4.79 (2.72; 8.47)	**<.001**
Age	.97 (.95; .99)	**.026**

$p < .05$ were boldfaced.

[a]The overall model was statistically significant ($\chi^2 = 100.9$, df = 5, $p < .001$); [b]The overall model was statistically significant ($\chi^2 = 89.5$, df = 5, $p < .0001$).

specifically in alcohol-dependent patients with the history of suicide attempts.

Alterations in the glutamatergic system were described as a possible background not only of suicidal behavior,[18] but also of alcohol dependence.[15,26] Ethanol acts by inhibiting neuronal NMDA receptor function. GRIN2B subunit are particularly sensitive to ethanol-induced inhibition. Chronic ethanol exposure induces an upregulation of various NMDA receptor subunits including GRIN2B and leads to changes in genes expression via methylation. These neuroadaptive changes are considered to play an important role in development of tolerance and alcohol withdrawal symptoms.[27] Previous molecular research reported association between *GRIN2B* polymorphisms and alcohol dependence.[26,28] The results of the present study indicate that *GRIN2B* may be involved in vulnerability to suicide attempts in alcohol-dependent individuals. Increased levels of glutamate have been shown to reduce dopaminergic transmission.[29] Glutamatergic neurotransmission, in particular within the mesolimbic dopamine pathway, plays a crucial role in development of alcohol addiction.[30] We hypothesize that suicidal behavior and alcohol dependence may share a common etiology directly related to the glutamatergic system.

Interestingly, the reported association between rs2268115 and suicide attempts was stronger when analyzing alcohol-dependent patients only, without co-occurring drug addiction. The comparison between alcohol-dependent patients with and without coexisting opioid dependence revealed significant differences. The severity of alcohol dependence and impulsivity levels were much higher in those without opioid dependence. Previously, *GRIN2B* polymorphisms were reported to be associated with risky decision-making and impulsivity.[31] Both the severity of alcohol dependence and impulsivity are well documented risk factors for suicide attempts.[32] More research is required to understand relationships between above-mentioned clinical characteristics and genetic vulnerability to suicidal behavior. In addition, genetic background of suicide attempts may differ in patients with alcohol dependence and those with opioid dependence. Recently, our study demonstrated an association between suicide attempts in opioid-dependent patients and *DISC1* rs2738888 polymorphism, however, no such relationship was found in alcohol-dependent subjects.[33]

The most important risk factor for suicide attempt in alcohol-dependent subjects in the present study was severe depression with almost a fivefold increase in odds of a suicide attempt. Ketamine, the NMDA receptors antagonist, is recently widely discussed as a potential rapid-acting antidepressant and antisuicidal drug.[34] Alcohol-dependent patients with coexisting severe depression and suicidal behaviors might particularly benefit with ketamine treatment.

The interpretation of findings of the present study is difficult because of complexity of the phenotype of suicidal behavior and the wide interactions between the glutamatergic system and other neurotransmitters involved in pathophysiology of suicide. More research is needed to understand the relationships between genetic variants of the glutamatergic system and susceptibility to suicidal behavior. Particularly valuable seems research performed in large populations with phenotypes described in detail.

Limitations

Several limitations of the present study are important to consider. First, the study sample was relatively small, but we were able to analyze the possible association between the selected *GRIN2B* polymorphism and suicidal behavior including suicide attempts and suicides. In addition, the study had sufficient power to conduct preliminary analyses. Moreover, the present study focused only on one genetic alteration in the glutamatergic system. However, we selected this polymorphism on intention, because it was previously reported to be associated to suicide attempts in general population and we aimed to investigate it further.

Scientific Significance

The present findings suggest the possible association between *GRIN2B* rs2268115 polymorphism and suicide attempts in alcohol-dependent individuals. We have not confirmed the hypothesis that the selected *GRIN2B* polymorphism may be involved in genetic background of death by suicide. Alcohol-dependent patient with severe depressive symptoms and history of suicidal behavior may potentially benefit with treatment approach using agents targeting the glutamatergic system.

Declaration of Interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of this paper.

REFERENCES

1. Roy A. Risk factors for attempting suicide in heroin addicts. *Suicide Life Threat Behav*. 2010;40:416–420.
2. Ostergaard ML, Nordentoft M, Hjorthoj C. Associations between substance use disorders and suicide or suicide attempts in people with mental illness: A Danish Nation-wide, prospective, register-based study of patients diagnosed with schizophrenia, bipolar disorder, unipolar depression or personality disorder. *Addiction*. 2017. https://doi.org/10.1111/add.13788 [Epub ahead of print].
3. Mann JJ, Currier DM. Stress, genetics and epigenetic effects on the neurobiology of suicidal behavior and depression. *Eur Psychiatry*. 2010;25:268–271.
4. Reus GZ, Abelaira HM, Tuon T, et al. Glutamatergic NMDA receptor as therapeutic target for depression. *Adv Protein Chem Struct Biol*. 2016;103:169–202.
5. Rojas A, Dingledine R. Ionotropic glutamate receptors: Regulation by G-protein-coupled receptors. *Mol Pharmacol*. 2013;83:746–752.
6. Takahashi T, Feldmeyer D, Suzuki N, et al. Functional correlation of NMDA receptor epsilon subunits expression with the properties of single-channel and synaptic currents in the developing cerebellum. *J Neurosci*. 1996;16:4376–4382.
7. Zhang Y, Li P, Feng J, et al. Dysfunction of NMDA receptors in Alzheimer's disease. *Neurol Sci*. 2016; 37:1039–1047.
8. Sgambato-Faure V, Cenci MA. Glutamatergic mechanisms in the dyskinesias induced by pharmacological dopamine replacement and deep brain stimulation for the treatment of Parkinson's disease. *Prog Neurobiol*. 2012;96:69–86.
9. Beal MF, Kowall NW, Ellison DW, et al. Replication of the neurochemical characteristics of Huntington's disease by quinolinic acid. *Nature*. 1986;321:168–171.
10. Banerjee J, Banerjee Dixit A, Tripathi M, et al. Enhanced endogenous activation of NMDA receptors in pyramidal neurons of hippocampal tissues from patients with mesial temporal lobe epilepsy: A mechanism of hyper excitation. *Epilepsy Res*. 2015;117:11–16.
11. Ghasemi M, Phillips C, Trillo L, et al. The role of NMDA receptors in the pathophysiology and treatment of mood disorders. *Neurosci Biobehav Rev*. 2014;47:336–358.
12. Ju P, Cui D. The involvement of N-methyl-d-aspartate receptor (NMDAR) subunit NR1 in the pathophysiology of schizophrenia. *Acta Biochim Biophys Sin*. 2016;48:209–219.
13. Costi S, Van Dam NT, Murrough JW. Current status of ketamine and related therapies for mood and anxiety disorders. *Curr Behav Neurosci Rep*. 2015;2:216–225.
14. Aragam N, Wang KS, Anderson JL, et al. TMPRSS9 and GRIN2B are associated with neuroticism: A genome-wide association study in a European sample. *J Mol Neurosci*. 2013;50:250–256.
15. Forero DA, Lopez-Leon S, Shin HD, et al. Meta-analysis of six genes (BDNF, DRD1, DRD3, DRD4, GRIN2B and MAOA) involved in neuroplasticity and the risk for alcohol dependence. *Drug Alcohol Depend*. 2015;149:259–263.
16. Dean B, Gibbons AS, Boer S, et al. Changes in cortical N-methyl-d-aspartate receptors and post-synaptic density protein 95 in schizophrenia, mood disorders and suicide. *Aust N Z J Psychiatry*. 2016;50: 275–283.
17. Chandley MJ, Szebeni A, Szebeni K, et al. Elevated gene expression of glutamate receptors in noradrenergic neurons from the locus coeruleus in major depression. *Int J Neuropsychopharmacol*. 2014;17:1569–1578.
18. Sokolowski M, Ben-Efraim YJ, Wasserman J, et al. Glutamatergic GRIN2B and polyaminergic ODC1 genes in suicide attempts: Associations and gene-environment interactions with childhood/adolescent physical assault. *Mol Psychiatry*. 2013;18:985–992.
19. Höfer P, Rockett IRH, Värnik P, et al. Forty years of increasing suicide mortality in Poland: Undercounting amidst a hanging epidemic? *BMC Public Health*. 2012;12:644.
20. Fudalej S, Kopera M, Wolynczyk-Gmaj D, et al. Association between FKBP5 functional polymorphisms and completed suicide. *Neuropsychobiology*. 2015;72:126–131.
21. Ploski R. Homogeneity and distinctiveness of Polish paternal lineages revealed by Y chromosome microsatellite haplotype analysis. *Hum Genet*. 2002;110:592–600.
22. Nowak G, Ordway GA, Paul IA. Alterations in the N-methyl-D-aspartate (NMDA) receptor complex in the frontal cortex of suicide victims. *Brain Res*. 1995;675:157–164.
23. Gray AL, Hyde TM, Deep-Soboslay A, et al. Sex differences in glutamate receptor gene expression in major depression and suicide. *Mol Psychiatry*. 2015;20:1057–1068.
24. Erhardt S, Lim CK, Linderholm KR, et al. Connecting inflammation with glutamate agonism in suicidality. *Neuropsychopharmacology*. 2013;38 :743–752.
25. Sokolowski M, Wasserman J, Wasserman D. Genome-wide association studies of suicidal behaviors: A review. *Eur Neuropsychopharmacol*. 2014;24:1567–1577.
26. Kim JH, Park M, Yang SY, et al. Association study of polymorphisms in N-methyl-D-aspartate receptor 2B subunits (GRIN2B) gene with Korean alcoholism. *Neurosci Res*. 2006;56:220–223.
27. Gass JT, Olive MF. Glutamatergic substrates of drug addiction and alcoholism. *Biochem Pharmacol*. 2008;75:218–265.
28. Wernicke C, Samochowiec J, Schmidt LG, et al. Polymorphisms in the N-methyl-D-aspartate receptor 1 and 2B subunits are associated with alcoholism-related traits. *Biol Psychiatry*. 2003;54:922–928.
29. Seamans JK, Yang CR. The principal features and mechanisms of dopamine modulation in the prefrontal cortex. *Prog Neurobiol*. 2004; 74:1–58.
30. Eisenhardt M, Leixner S, Lujan R, et al. Glutamate receptors within the mesolimbic dopamine system mediate alcohol relapse behavior. *J Neurosci*. 2015;35:15523–15538.
31. Ness V, Arning L, Niesert HE, et al. Variations in the GRIN2B gene are associated with risky decision-making. *Neuropharmacology*. 2011; 61:950–956.
32. Mann JJ, Arango VA, Avenevoli S, et al. Candidate endophenotypes for genetic studies of suicidal behavior. *Biol Psychiatry*. 2009;65: 556–563.
33. Fudalej S, Jakubczyk A, Kopera M, et al. DISC1 as a possible genetic contribution to opioid dependence in a polish sample. *J Stud Alcohol Drugs*. 2016;77:220–226.
34. Wilkinson ST, Sanacora G. Ketamine: A potential rapid-acting antisuicidal agent? *Depress Anxiety*. 2016; 33:711–717.