

Association study of T102C 5-HT_{2A} polymorphism in schizophrenic patients: diagnosis, psychopathology, and suicidal behavior

Humberto Correa, MD, PhD; Luiz De Marco, MD, PhD; Wolfanga Boson, MSc; Rodrigo Nicolato, MD; António L. Teixeira, MD, PhD; Valdir R. Campo, MD; Marco A. Romano-Silva, MD, PhD



The study of the association between schizophrenia and the gene encoding the serotonin (5-HT)_{2A} receptor has been a matter of interest in recent years. The 5-HT_{2A} receptor gene (*5-HTR_{2A}*) has several polymorphisms in the general population, but the T102C polymorphism is probably the most extensively studied. This polymorphism does not result in an amino acid change in the receptor, as both alleles encode a serine at position 34.¹ Nevertheless, there is some evidence showing that the C-allele form can be significantly less functional than the T allele of *5-HTR_{2A}*, both in healthy controls and schizophrenic patients, and this difference is even more prominent in schizophrenic patients.²

The objective of this study was to examine the association between the serotonin (5-HT)_{2A} gene polymorphism (T102C) and suicidal behavior in schizophrenic inpatients. We studied 129 subjects who met the diagnostic criteria for schizophrenia according to a structured clinical interview (MINI-PLUS). Patients underwent a semistructured interview to assess suicide attempt history and its characteristics. In addition, at least one close relative of the patient was interviewed to assess proband and family suicidal behavior. Healthy controls were students and hospital staff members free of psychiatric and medical illness. Genotypes were determined after polymerase chain reaction amplification of the region of 5-HT_{2A}/T102C containing the polymorphic site and digestion with the restriction enzyme HpaI. We found no association between suicidal attempt history and suicide attempt characteristics and genotypic or allele frequencies. Suicidal behavior was also not associated with demographic or psychopathological characteristics. These results suggest that the 5-HT_{2A} gene polymorphism (T102C) is not involved in genetic susceptibility to suicidal behavior, but further studies in a larger sample are needed.

© 2007, LLS SAS

Dialogues Clin Neurosci. 2007;9:97-101.

Keywords: *schizophrenia; suicide; 5-HT_{2A}; polymorphism*

Author affiliations: Department of Mental Health (Humberto Correa); Department of Pharmacology (Luiz De Marco, Wolfanga Boson, Marco A. Romano-Silva); Pharmacogenetics Laboratory (Rodrigo Nicolato, António L. Teixeira, Valdir R. Campo, Marco A. Romano-Silva), Serviço de Psiquiatria, Hospital das Clínicas, Universidade Federal de Minas Gerais (UFMG), Belo Horizonte, Brazil

Address for correspondence: Humberto Correa, MD, PhD, Departamento de Saúde Mental, Faculdade de Medicina, Universidade Federal de Minas Gerais, 30.130-100 Av. Alfredo Balena, 190, Belo Horizonte, Brazil (e-mail: correa@task.com.br)

Additional evidence for a functional role of this mutation comes from a recent meta-analysis showing that, at least in European populations, the C allele is associated with schizophrenia.³

Another point of interest in the study of 5-*HTR*_{2A} is the fact that there are compelling results showing that suicidal behavior is associated with serotonergic dysfunction, and that suicidal behavior is, at least partially, genetically determined.⁴ The 5-HT_{2A} receptor may play a major role in this association.⁵

Since suicidal behavior is a rather frequent event in schizophrenia,⁶ an important question to be answered is whether the T102C 5-HT_{2A} polymorphism could be more closely associated with suicidal behavior than with schizophrenia itself.

Therefore, the aim of our work was to examine the association of the T102C polymorphism of the 5-HT_{2A} gene with schizophrenia diagnosis, psychopathological characteristics, and history of suicidal behavior in a well-characterized sample of Brazilian schizophrenic inpatients.

Material and methods

Subjects

A total of 214 unrelated subjects were enrolled after a full explanation of this study and obtaining of informed consent. This study was approved by the University Ethics Committee.

One hundred and twenty-nine schizophrenic patients, consecutively admitted to Hospital Santa Maria, Belo Horizonte, meeting the *Diagnostic and Statistical Manual of Mental Disorders*, 4th edition (*DSM-IV*) diagnostic criteria based on a structured interview (MINI-PLUS),^{7,8} without any other current comorbid axis I disorders, were enrolled in this study. Healthy controls were students and hospital staff members (n=85), all free of psychiatric and medical illness.

Psychopathology

The psychiatric symptoms of the patients were rated using the Brief Psychiatric Rating Scale (BPRS),⁹ which has been validated in Brazil.¹⁰ The principal component analyses of the symptoms assessed by the BPRS were also performed, and four factors emerged: delusion dimension (DD, items 4 + 12 + 15), hebephrenic dimension (DH, items 3 + 13 + 16), paranoid dimension (DP,

items 10 + 11 + 14) and anxiety-depressive dimension (DA, items 2 + 5 + 9), as previously described.⁵

Suicide history

The suicide history was assessed using a semistructured interview,⁵ a review of medical records, and a supplementary interview with at least one close relative in order to collect information about suicidal behavior in probands. The Lethality Rating Scale (LRS)¹¹ was used to measure the degree of medical damage of the most lethal lifetime suicide attempt. We used an anchored modified version¹² scored 0 to 8 and, as previously described, a lethality score of three or greater as a cutoff point of three in the LRS scores.⁵ The suicide attempt methods, defined by the most lethal lifetime suicide attempt, were classified as nonviolent (drug overdose) or violent (cutting beyond a superficial scratch, jumping from a height, shooting, hanging) as defined elsewhere.¹³

Genotyping

Deoxyribonucleic acid (DNA) was isolated from lymphocytes using routine procedures. Polymerase chain reaction (PCR) amplification of the HT_{2A}/T102C region containing the polymorphic site produced a 342 base-pair (bp) fragment. This was digested with the restriction enzyme HpaII. The uncut product corresponded to the 102T allele. Digested products with 216bp and 126 bp corresponded to the 102C allele.

Statistical analyses

A Chi-square test was used for categorical data and an analysis of variance (ANOVA) for continuous variables. The tests were two-tailed, and *P* was considered significant when *P*<0.05.

Results

Patients and controls were not different regarding demographic characteristics such as ethnicity and gender. Moreover, no differences were observed regarding genotype frequencies across these groups (*Table I*).

We further analyzed the association between the genotype and allele frequencies of the T102C polymorphism and total and factor scores of the BPRS. As shown in *Table II*, no association was found.

The history of a suicide attempt and the suicide attempt characteristics were not associated with genotype, allele frequencies, nor with psychopathological scores (Table III). Furthermore, the demographic characteristics were not statistically different between patients with a suicide attempt history (n=42; [32.5%], 23 were male, 19 were female; 28 were of Caucasian descent, 14 of African descent) and without such a history (n=87; [67.5%], 56 were male, 31 were female; 56 were of Caucasian descent, 31 were of African descent). In both groups the genotype frequencies were in Hardy-Weinberg equilibrium.

Discussion

Two previous meta-analyses had shown an association between the C allele of the T102C polymorphism and schizophrenia.^{3,14} However, these meta-analyses yielded relatively slight odds ratios (ORs) of 1.18 and 1.1, respectively. An important question remains as to whether

some characteristics present in schizophrenic patients, such as suicidality, rather than schizophrenia itself, could be related to the C allele.

There is much evidence to support this hypothesis. First, suicide is the leading cause of premature death in schizophrenic patients^{15,16} and a substantial percentage of patients with schizophrenia also attempt suicide, with estimates of lifetime occurrence ranging from 18% to 55%.¹⁵ Second, a previous study showed an association between the C allele and suicidal thoughts in depressed patients,¹⁷ and we may speculate whether this same finding could be observed in schizophrenia. Third, it is also known that the 5-HT_{2A} receptor is, at least indirectly, involved in suicidal behavior in both depressed and schizophrenic patients.⁶ Finally, it has been shown that the atypical antipsychotic clozapine, which acts on 5-HT_{2A}, could have an antisuicidal action.^{18,19}

To the best of our knowledge, there has been only one study that specifically analyzed the association between

	Genotypes			Sex distribution		Ethnicity	
	TT	TC	CC	Males	Females	Caucasian	African
Schizophrenic patients (n=129)	35 (27%)	62 (48%)	32 (25%)	79 (61%)	50 (39%)	84 (65%)	45 (35%)
Healthy controls (n=85)	22 (25%)	40 (47%)	23 (27%)*	45 (53%)	40 (49%) †	57 (67%)	28 (33%) ‡

Table I. Genotype distribution and demographic characteristics across schizophrenic patients and healthy controls. *: $\chi^2=0.14$, $P=0.93$; †: $\chi^2=1.45$, $P=0.23$; ‡: $\chi^2=0.86$, $P=0.77$

	TT genotype	TC genotype	CC genotype
BPRS Total score	38.7 ± 7.2	36.8 ± 7.2	39.6 ± 7.5 *
Delusion dimension	10.1 ± 3.8	9.5 ± 4.0	10.9 ± 3.9 †
Anxio-depressive dimension	5.4 ± 2.0	5.3 ± 2.2	5.1 ± 2.6 ‡
Hebephrenic dimension	8.4 ± 3.2	8.5 ± 3.0	7.9 ± 3.1 §
Paranoid dimension	5.9 ± 1.9	5.9 ± 1.9	6.3 ± 2.4

Table II. BPRS total scores and factor scores across genotypes in schizophrenic patients. BPRS, Brief Psychiatric Rating Scale. *: $F=1.06$, $P=0.4$; †: $F=11.32$, $P=0.2$; ‡: $F=1.26$, $P=0.28$; §: $F=1.2$, $P=0.3$; ||: $F=0.5$, $P=0.88$

	Suicide attempt history		Lethality		Type of suicide attempt	
	No (n=87)	Yes (n=42)	<3 (n=23)	≥3 (n=19)	Nonviolent (n=22)	Violent (n=20)
TT	24 (27.5%)	11 (26.2%)	7 (30.5%)	4 (21%)	4 (18.2%)	7 (35%)
TC	43 (49.5%)	19 (45.2%)	10 (43.4%)	9 (47.4%)	12 (54.5%)	7 (35%)
CC	20 (23%)	12 (28.6%) *	6 (26.1%)	6 (31.6%) †	6 (27.3%)	6 (30%) ‡
T allele	91 (52.25%)	41 (48.8%)	24 (52.2%)	17 (44.7%)	20 (45.5%)	21 (52.5%)
C allele	83 (47.75%)	43 (51.2%) §	22 (47.8%)	21 (55.3%)	24 (54.5%)	19 (47.5%) ¶
BPRS	38.5 ± 5.6	39.5 ± 5.9 #	38.4 ± 5.7	39.5 ± 6.4 **	38.7 ± 5.8	39.4 ± 6.2 ††

Table III. Distribution of the 5-HT_{2A} receptor gene 102 TC polymorphism in suicidal and nonsuicidal schizophrenic patients. BPRS, Brief Psychiatric Rating Scale. *: $\chi^2=0.48$, $P=0.79$; †: $\chi^2=0.49$, $P=0.78$; ‡: $\chi^2=2.1$, $P=0.36$; §: $\chi^2=0.28$, $P=0.6$; ||: $\chi^2=0.46$, $P=0.5$; ¶: $\chi^2=0.42$, $P=0.52$; #: $F=0.54$, $P=0.6$; **: $F=0.57$, $P=0.57$; ††: $F=0.36$, $P=0.72$

the 102T/C polymorphism of the 5-HT_{2A} gene and suicidality in schizophrenia, showing no positive correlation.²⁰ However, some methodological bias must be taken into consideration, because this study was based on a small sample of 65 schizophrenic and 6 schizoaffective patients. Thus, a type II error could not be reliably excluded. Another point of concern refers to suicide assessment. It has been proposed that suicidal behavior shows a continuum between suicide attempt and suicide completion. In other words, severe suicide attempts are biologically closer to suicide completion, and the seriousness of the suicide attempt might explain differences in serotonergic activity.⁴ Therefore, studying patients with suicidal ideation or suicide plans, as performed in this previous study, can be rather different from a biological point of view than studying patients with suicide attempts or suicide completion.

We were not able to observe any significant association between the T102C polymorphism and suicidal behavior in our sample. We investigated a fairly homogeneous sample of 129 schizophrenic inpatients, as assessed with structured instruments to evaluate diagnosis. Another strength of our study is the fact that suicidal behavior was assessed using a semistructured interview as well as a supplementary interview with at least one close relative, plus a review of medical records. This is rather important, since it has been previously shown that a significant degree of past suicidal behavior was not recorded during routine clinical assessment and, the use of a semistructured screening instrument may improve documentation and detection of lifetime suicidal behavior.¹² Indeed, a phenotypic characterization of suicide attempt, as performed in our study, could be of major interest since some categories of suicidal behavior (ie, more lethal or violent ones) could be more closely associated with a biological marker.²¹

REFERENCES

1. Warren F A, Nordstrom AL, Farde L, Ericksson B. An MspI polymorphism in the human serotonin receptor gene (HTR2): detection by DGGE and RFLP analysis. *Hum Mol Genet.* 1993;2:338.
2. Pollesskaya OO, Sokolov BO. Differential expression of the C and T alleles of the 5-HT_{2A} receptor gene in the temporal cortex of normal individuals and schizophrenics. *J Neurosci Res.* 2002;67:812-822.
3. Abdolmaleky HM, Faraone SV, Glatt SJ, Tsuang MT. Meta-analysis of association between the T102C polymorphism of the 5-HT_{2a} receptor gene and schizophrenia. *Schizophr Res.* 2004;67:53-62.
4. Mann JJ. Neurobiology of suicidal behavior. *Nat Rev Neurosci.* 2003;4:819-827.
5. Corrêa H, Duval F, Mokrani MC, et al. Serotonergic function and suicidal behavior in schizophrenia. *Schizophr Res.* 2002;56:75-85.

We were also not able to observe an association between the T102C polymorphism and schizophrenia diagnosis. This result is in apparent contradiction with a recent meta-analysis.³ However, the authors of this analysis showed that in East Asian countries, there was not a significant association with the C allele or CC homozygosity, indicating strong genetic differences and incompatibility between data from European and East Asian populations. They suggest that data from European and Asian samples should not be pooled when evaluating the involvement of this gene in schizophrenia. Interestingly, the frequency of the T allele was much higher in East Asian patients and controls (59.5% and 57.5%, respectively) than in European patients and controls (40% and 43.5%, respectively). In our sample, the frequency was intermediate between those values, since the T allele frequencies in patients and controls were 51% and 48.5%, respectively. In our sample we performed an ethnic stratification, and we did not observe a significantly different frequency in the studied genotypes between those of Caucasian and those of African descent. However as recently shown, race, as determined by physical evaluation, is a poor predictor of genomic African ancestry in Brazil.²²

In conclusion, as in all case-control psychiatric genetic studies, we must be aware of false-positive or false-negative findings due to ethnic stratification and sample size. We attempted to control this by including a detailed demographic analysis, which demonstrated no significant differences between genotype frequencies and ethnicities between patients with a suicide attempt history and patients without such a history. Despite this, further studies using a larger number of subjects should be carried out to firmly establish the role of the T102C polymorphism of the 5-HT_{2A} gene in suicidal behavior in schizophrenia. □

This work was supported by CNPq.

6. Radomsky ED, Hass GL, Mann JJ, Sweeney JA. Suicidal behavior in patients with schizophrenia and other psychotic disorders. *Am J Psychiatry.* 1999;156:1590-1595.
7. Sheehan DV, Lecrubier Y, Harnett-Sheehan K, et al. The Mini International Neuropsychiatric Interview (M.I.N.I.): the development and validation of a structured diagnostic psychiatric interview. *J Clin Psychiatry.* 1998;59:22-33.
8. Amorim P. Mini International Neuropsychiatric Interview (MINI): validação de entrevista breve para diagnóstico de transtornos mentais. *Rev Bras Psiquiatr.* 2000;22:106-115.
9. Overall J, Gorham D. The brief psychiatric rating scale: a comprehensive review. *Psychol Report.* 1962;10:799-812.
10. Elkis H, Henna JN, Alves TM, Eizeman IB, Campos de Oliveira JR, Feijó de Melo M. Análise fatorial da versão em português do BPRS ancorado (BPRS-A) em pacientes com esquizofrenia refratária. *Rev Psiquiatr Clin.* 1998;25:334-336.

Estudio de asociación del polimorfismo del gen T102C del receptor de serotonina 5-HT_{2A} en pacientes esquizofrénicos: diagnóstico, psicopatología y conducta suicida

El objetivo de este estudio fue examinar la asociación entre el polimorfismo genético (102T/C) del receptor de serotonina (5-HT_{2A}) y la conducta suicida en pacientes esquizofrénicos hospitalizados. Se estudiaron 129 sujetos que cumplieron los criterios diagnósticos para esquizofrenia de acuerdo con una entrevista clínica estructurada (MINI-Plus). A los pacientes se les aplicó una entrevista semiestructurada para evaluar la historia de intentos suicidas y las características de éstos. Además, se entrevistó a lo menos a un pariente cercano del paciente para evaluar la conducta suicida del probando y de familiares. Los controles sanos fueron estudiantes y personal del hospital sin patología médica o psiquiátrica. Los genotipos fueron determinados después de una amplificación de la región 5-HT_{2A}/T102C que contiene el sitio de polimorfismo mediante la reacción en cadena de la polimerasa y de la digestión a través de la enzima de restricción HpaII. No se encontró ninguna asociación entre la historia de intentos suicidas o las características de éstos y el genotipo o frecuencias de los alelos. La conducta suicida tampoco se asoció con características demográficas ni psicopatológicas. Estos resultados sugieren que el polimorfismo del gen (102T/C) para el receptor 5-HT_{2A} no está involucrado en la susceptibilidad genética para la conducta suicida, pero se requiere de futuros estudios en una muestra con más pacientes.

Étude d'association du polymorphisme T102C du gène codant pour le récepteur 5-HT_{2A} chez les patients souffrant de schizophrénie: diagnostic, psychopathologie et comportement suicidaire

Cette étude avait pour but d'examiner l'association entre le polymorphisme (102T/C) du gène codant pour le récepteur sérotoninergique 5-HT_{2A} et le comportement suicidaire chez les schizophréniques. Nous avons étudié 129 sujets ayant rempli les critères diagnostiques de schizophrénie selon un questionnaire clinique structuré (MINI-PLUS). Les patients ont répondu à un questionnaire semi-structuré pour évaluer les antécédents de tentative de suicide et leurs caractéristiques. De plus, au moins un apparenté proche était interrogé pour évaluer les antécédents familiaux pour les comportements suicidaires ainsi que ceux du malade lui-même. Les témoins sains étaient des étudiants et des membres de l'équipe hospitalière sans maladie ni pathologie psychiatrique. Les génotypes étaient déterminés par polymérase chain reaction (PCR) de la région du 5-HT_{2A}/T102C contenant le site polymorphe, et après digestion avec l'enzyme de restriction HpaII. Nous n'avons trouvé aucune association entre les antécédents de tentative de suicide ou les caractéristiques de ces tentatives, et un des génotypes ou des allèles. Le comportement suicidaire n'était pas non plus associé aux caractéristiques démographiques ou psychopathologiques. Ces résultats suggèrent que le polymorphisme du gène 5-HT_{2A} (102T/C) n'est pas impliqué dans la vulnérabilité génétique aux comportements suicidaires. Des études incluant un plus grand nombre de patients sont néanmoins requises.

11. Beck AT, Beck A, Kovacs M. Classification of suicidal behaviors. I. Quantifying intent and medical lethality. *Am J Psychiatry*. 1975;132:285-287.
12. Malone KM, Szanto K, Corbitt EM, Mann JJ. Clinical assessment versus research methods in the assessment of suicidal behavior. *Am J Psychiatry*. 1995;152:1601-1607.
13. Coccaro EF, Sielver LJ, Klar HM, et al. Serotonergic studies in patients with affective and personality disorders correlates with suicidal and impulsive aggressive behavior. *Arch Gen Psychiatry*. 1989;46:587-599.
14. Williams J, McGuffin P, Nothen M, Owen MJ. Meta-analysis of association between the 5-HT_{2A} receptor T102C and schizophrenia. EMAS collaborative Group. European Multicentre Association Study of Schizophrenia. *Lancet*. 1997;349:1221.
15. Roy A. Suicide in schizophrenia, In: Roy A, ed. *Suicide*. Baltimore, Md: Williams & Wilkins; 1996:97-109.
16. Caldwell CB, Gottesman II. Schizophrenics kill themselves too: a review of risk factors for suicide. *Schizophr Bull*. 1990;16:571-589.
17. Du L, Bakish D, Lapierre YD, Ravindran AV, Hrdina PD. Association of polymorphism of serotonin 2A receptor gene with suicidal ideation in major depressive disorder. *Am J Med Genet*. 2000;96:56-60.
18. Meltzer HY, Alphs L, Green AI, et al. International Suicide Prevention Trial Study Group. Clozapine treatment for suicidality in schizophrenia. *Arch Gen Psychiatry*. 2003;60:82-91.
19. Hennen J, Baldessarini RJ. Suicidal risk during treatment with clozapine: a meta-analysis. *Schizophrenia Res*. 2005;73:139-145.
20. Ertugrul A, Kennedy JL, Masellis M, Basile VS, Jayathilake K, Meltzer HY. No association of the T102C polymorphism of the serotonin 2A receptor gene (HTR2A) with suicidality in schizophrenia. *Schizophrenia Res*. 2004;69:301-305.
21. Campi-Azevedo C, De Marco L, Boson W, Romano-Silva MA, Correa H. Association of the serotonin transporter promoter polymorphism with suicidal behavior. *Mol Psychiatry*. 2004;8:899-900.
22. Parra FC, Amado RC, Lambertucci JR, Rocha J, Antunes CM, Pena SD. Color and genomic ancestry in Brazilians. *Proc Natl Acad Sci U S A*. 2003;100:177-182.