No evidence so far of a major role of AKT1 and GSK3B in the pathogenesis of antipsychotic-induced tardive dyskinesia

Levchenko, Anastasia; Vyalova, Natalya; Pozhidaev, Ivan V; Boiko, Anastasiia S; Osmanova, Diana Z; Fedorenko, Olga Yu; Semke, Arkadiy V; Bokhan, Nikolay A; Wilffert, Bob; Loonen, Anton J M

Published in:
Human Psychopharmacology

DOI:
10.1002/hup.2685

IMPORTANT NOTE: You are advised to consult the publisher's version (publisher's PDF) if you wish to cite from it. Please check the document version below.

Document Version
Publisher's PDF, also known as Version of record

Publication date:
2019

Link to publication in University of Groningen/UMCG research database

Citation for published version (APA):

Copyright
Other than for strictly personal use, it is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), unless the work is under an open content license (like Creative Commons).

The publication may also be distributed here under the terms of Article 25fa of the Dutch Copyright Act, indicated by the “Taverne” license. More information can be found on the University of Groningen website: https://www.rug.nl/library/open-access/self-archiving-pure/taverne-amendment.

Take-down policy
If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.
No evidence so far of a major role of AKT1 and GSK3B in the pathogenesis of antipsychotic-induced tardive dyskinesia

Anastasia Levchenko1* | Natalya Vyalova2* | Ivan V. Pozhidaev2 | Anastasiia S. Boiko2 | Diana Z. Osmanova2 | Olga Yu. Fedorenko2,3 | Arkadiy V. Semke2 | Nikolay A. Bokhan2,4 | Bob Wilffert5,6 | Anton J.M. Loonen5,7 | Svetlana A. Ivanova2,3

1 Institute of Translational Biomedicine, Saint Petersburg State University, Saint Petersburg, Russia
2 Tomsk National Research Medical Center of the Russian Academy of Sciences, Mental Health Research Institute, Tomsk, Russia
3 Division for Control and Diagnostics, School of Non-Destructive Testing & Security, National Research Tomsk Polytechnic University, Tomsk, Russia
4 Department of Psychotherapy and Psychological Counseling, National Research Tomsk State University, Tomsk, Russia
5 Unit of PharmacoTherapy, Epidemiology and Economics, Groningen Research Institute of Pharmacy, University of Groningen, Groningen, The Netherlands
6 Department of Clinical Pharmacy and Pharmacology, University Medical Center Groningen, Groningen, University of Groningen, Groningen, The Netherlands
7 GGZ Westelijk Noord-Brabant, Bergen op Zoom, The Netherlands

Correspondence
Anastasia Levchenko, Institute of Translational Biomedicine, Saint Petersburg State University, 7/9, Universitetskaya Embankment, Saint Petersburg 199034, Russia.
Email: a.levchenko@spbu.ru
Natalya Vyalova, Mental Health Research Institute, Tomsk National Research Medical Center of the Russian Academy of Sciences, 4, Aleutskaya Street, Tomsk 634014, Russia.
Email: natarakitina@yandex.ru

Funding information
Siberian Branch, Russian Academy of Sciences, Grant/Award Number: 30

Abstract
Objective: AKT1 and GSK3B take part in one of the intracellular cascades activated by the D2 dopamine receptor (DRD2). This receptor is antagonized by antipsychotics and plays a role in the pathogenesis of antipsychotic-induced tardive dyskinesia (TD). The present study investigated association of several polymorphisms in the two candidate genes, AKT1 and GSK3B, with TD in antipsychotic-treated patients with schizophrenia.

Methods: DNA samples from 449 patients from several Siberian regions (Russia) were genotyped, and the results were analyzed using chi-squared tests and analyses of variance.

Results: Antipsychotic-induced TD was not associated with either of the tested functional polymorphisms (rs334558, rs1130214, and rs3730358).

Conclusions: Despite regulation of AKT1 and GSK3B by DRD2, we found no evidence that these two kinases play a major role in the pathogenesis of antipsychotic-induced TD. These results agree with previously published data and necessitate further exploration of other pathogenic mechanisms, such as neurotoxicity due to excessive dopamine metabolism.

KEYWORDS
AKT1, antipsychotics, GSK3B, pharmacogenetics, schizophrenia, tardive dyskinesia

1 | INTRODUCTION

Iatrogenic tardive dyskinesia (TD) is a debilitating and potentially irreversible neurological side effect of chronic treatment with a number of different types of medication, primarily with antipsychotics (Cornett, Novitch, Kaye, Kata, & Kaye, 2017; Tenback & van Harten, 2011). Typical antipsychotics are associated with higher, 30% incidence of TD, but incidence with atypical antipsychotics is still significant, incapacitating 20% of patients (Cornett et al., 2017; Stegmayer, Walther, & van Harten, 2018). The involuntary movements affect the...
orofacial area and trunk and limbs, defining orofacial and limb-truncal types of TD. There is no consensus on the pathogenic mechanisms behind these features, and different neurotransmitter systems may be involved, but one of the best known hypotheses postulates that the D2 dopamine receptor (DRD2; Beaulieu & Gainetdinov, 2011) plays an important role (Stegmayer et al., 2018; Zai et al., 2018). Furthermore, the two recently FDA-approved drugs to treat TD, valbenazine and deutetrabenazine, decrease the availability of synaptic dopamine by inhibiting the vesicular monoamine transporter type 2 (Stahl, 2018; Zai et al., 2018), which might also support the hypothesis that DRD2 is implicated in the pathogenesis. According to that hypothesis, blockade by antipsychotics of the inhibitory DRD2 of gamma-aminobutyric acid (GABAergic) medium spiny neurons (MSNs) in the striatum results in compensatory upregulation of a supersensitive type of these receptors, which in turn may lead to excessive inhibition of the GABAergic MSNs that take part in the indirect pathway of the extrapyramidal circuit (Stahl, 2018). This inhibition would result in insufficient inhibition of the cortex with the net result of increased uncontrolled movements. Furthermore, the upregulated supersensitive DRD2 may enhance intracellular cascades that lead to suspected neurodegeneration of GABAergic MSNs of the indirect pathway, which would explain irreversible symptoms in some patients.

Apart from the well-known G protein-dependent DRD2 signaling involving inhibition of cAMP (Beaulieu & Gainetdinov, 2011; Greengard, 2001), another pathway is G protein-independent and involves AKT serine/threonine kinase 1 (AKT1) and glycogen synthase kinase 3β (GSK3B; Beaulieu, Del'guidice, Sotnikova, Lemasson, & Gainetdinov, 2011; Beaulieu, Gainetdinov, & Caron, 2009; Freyberg, Ferrando, & Javitch, 2010). In the present study, we looked for evidence that supersensitive DRD2s are involved in the pathogenesis of TD, including suspected neurodegeneration of GABAergic MSNs of the indirect pathway, through activation of the G protein-independent DRD2 signaling. In particular, we evaluated association of functional single nucleotide polymorphisms (SNPs) in AKT1 and GSK3B with TD in schizophrenic patients, treated with typical and atypical antipsychotic drugs. The SNPs rs1130214 and rs3730358 in AKT1 were chosen because of association of the haplotype TC with lower protein levels of AKT1, which suggests impaired mRNA expression or processing (Emamian, Hall, Birnbaum, Karayiorgou, & Gogos, 2004). The variant rs334558, found in the promoter of GSK3B, is also known to be functional, because it determines the expression level of GSK3B, possibly by regulating transcription factor binding to the promoter (Kwok et al., 2005).
were significantly older, and the duration of schizophrenia in these patients was significantly longer than in patients without TD. Additionally, patients with TD were taking higher doses of antipsychotics. In our sample, TD was diagnosed more often in men than in women.

There was no deviation from Hardy–Weinberg equilibrium. Of the three SNPs tested, neither was associated with TD, when genotypes and alleles were compared between the group of patients with TD and the group without it. Table 2 summarizes these results. Association was also lacking when different types of TD, orofacial (AIMS Items 1 to 4) and limb–truncal (AIMS Items 5 to 7), were considered separately (data not shown). Analyses of variance showed that for all genotypes, the variance in the distribution of sums of AIMS scores is equal, indicating that the severity of TD, for both orofacial and limb–truncal types, is not associated with any of the tested SNPs (data not shown).

### DISCUSSION

The G protein-independent DRD2 signaling does not involve cAMP (Beaulieu et al., 2011). In particular, activation of this intracellular cascade results in downregulation of AKT1 that otherwise inhibits GSK3B.

As a result, increased activity of GSK3B is brought by activation of DRD2. These kinases play important roles in many aspects of the life of a cell, such as differentiation, proliferation, metabolism, and survival (Doble & Woodgett, 2003; Dummler & Hemmings, 2007; Emamian, 2012; Frame & Cohen, 2001), including aspects relevant to psychiatric and neurological disorders and response to medication (Beaulieu et al., 2009; Beaulieu et al., 2011; Emamian, 2012; Freyberg et al., 2010; Levchenko et al., 2018). GSK3B is also known to activate apoptosis (Doble & Woodgett, 2003; Frame & Cohen, 2001), which may explain suspected neurodegeneration of GABAergic MSNs of the indirect pathway, resulting in irreversible symptoms in some patients.

Although the studied SNPs are known to alter expression of the DRD2-coupled kinases AKT1 and GSK3B, our study did not find evidence that these variants are associated with antipsychotic-induced TD. In fact, previous studies of AKT1 and GSK3B showed implication of the two kinases in antipsychotic drug action (Beaulieu et al., 2009; Beaulieu et al., 2011; Freyberg et al., 2010), but not in the pathogenesis of TD. In particular, three previous studies that investigated association between two of these SNPs and TD also showed negative results, unless other variables were incorporated into the analyses (Park et al., 2009; Souza et al., 2010; Zai et al., 2008). One of these studies reported association of TD with rs3730358 (AKT1) only when taking into account another SNP rs6275, found in DRD2 (Zai et al., 2008). However, rs6275 was already found to be associated with TD (Zai et al., 2007), so it is unclear to what degree rs3730358 contributes to the reported association. A second study (Park et al., 2009) also reported association with TD only when rs334558 (GSK3B) was analyzed together with the Val66Met polymorphism in the brain-derived neurotrophic factor gene (BDNF; Miura et al., 2014). The third study reported association of the SNP rs6438852, found in high linkage disequilibrium with rs334558 (GSK3B; Kwok et al., 2005), only when age was incorporated as a covariate in the analysis (Souza et al., 2010). Age is the variable known to be robustly associated with TD (Cornett et al., 2017; Tenback & van Harten, 2011; Waln & Jankovic, 2013), so it is unclear to what degree rs6438852 (as a proxy for rs334558) is important in the reported association. As suggested by these studies, AKT1 and GSK3B might play some role in the pathogenesis of antipsychotic-induced TD, possibly via interaction with

### TABLE 1 Demographic and clinical parameters of the studied patient groups

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Patients with TD (n = 121)</th>
<th>Patients without TD (n = 328)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Men, number of</td>
<td>71 (58.7%)</td>
<td>152 (46.3%)</td>
<td>0.020</td>
</tr>
<tr>
<td>Women, number of</td>
<td>50 (41.3%)</td>
<td>176 (53.7%)</td>
<td></td>
</tr>
<tr>
<td>Mean age, years</td>
<td>48 [37.5; 58]</td>
<td>37 [31; 48]</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mean age of onset, years</td>
<td>25 [20; 32]</td>
<td>24 [20; 30]</td>
<td>0.974</td>
</tr>
<tr>
<td>Mean duration of illness, years</td>
<td>20 [12; 29.5]</td>
<td>11 [5; 18]</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mean CPZeq, dose</td>
<td>500 [286.2; 750]</td>
<td>396 [200; 750]</td>
<td>0.021</td>
</tr>
</tbody>
</table>

Note. Range is indicated in square brackets. TD: tardive dyskinesia; CPZeq: chlorpromazine equivalents.

### TABLE 2 Distribution of alleles and genotypes of AKT1 and GSK3B polymorphisms in groups of patients with TD and patients without TD

<table>
<thead>
<tr>
<th>Gene</th>
<th>SNP</th>
<th>Genotypes, alleles</th>
<th>Patients with TD</th>
<th>Patients without TD</th>
<th>OR Value</th>
<th>95% CI</th>
<th>(\chi^2)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>AKT1</td>
<td>rs3730358</td>
<td>GG</td>
<td>87 (71.9%)</td>
<td>231 (70.6%)</td>
<td>1.06</td>
<td>[0.67, 1.69]</td>
<td>1.178</td>
<td>0.555</td>
</tr>
<tr>
<td></td>
<td></td>
<td>GA</td>
<td>29 (24.0%)</td>
<td>88 (26.9%)</td>
<td>0.86</td>
<td>[0.55, 1.39]</td>
<td>0.010</td>
<td>0.940</td>
</tr>
<tr>
<td></td>
<td></td>
<td>AA</td>
<td>5 (4.1%)</td>
<td>8 (2.4%)</td>
<td>1.2</td>
<td>[0.55, 5.36]</td>
<td>3.136</td>
<td>0.208</td>
</tr>
<tr>
<td></td>
<td>rs1130214</td>
<td>G</td>
<td>102 (83.9%)</td>
<td>275 (84.1%)</td>
<td>0.98</td>
<td>[0.66, 1.47]</td>
<td>0.001</td>
<td>0.940</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A</td>
<td>19 (16.1%)</td>
<td>52 (15.9%)</td>
<td>1.02</td>
<td>[0.68, 1.52]</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>GG</td>
<td>53 (43.8%)</td>
<td>146 (44.6%)</td>
<td>0.97</td>
<td>[0.63, 1.47]</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>GT</td>
<td>60 (49.6%)</td>
<td>142 (43.4%)</td>
<td>1.28</td>
<td>[0.84, 1.95]</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>TT</td>
<td>8 (6.6%)</td>
<td>39 (11.9%)</td>
<td>0.52</td>
<td>[0.24, 1.15]</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>G</td>
<td>83 (68.6%)</td>
<td>217 (64.4%)</td>
<td>1.11</td>
<td>[0.81, 1.52]</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>T</td>
<td>38 (31.4%)</td>
<td>110 (33.6%)</td>
<td>0.90</td>
<td>[0.66, 1.24]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>GSK3B</td>
<td>rs334558</td>
<td>GG</td>
<td>25 (21.0%)</td>
<td>62 (19.3%)</td>
<td>1.12</td>
<td>[0.67, 1.89]</td>
<td>0.367</td>
<td>0.832</td>
</tr>
<tr>
<td></td>
<td></td>
<td>GA</td>
<td>59 (49.6%)</td>
<td>158 (48.8%)</td>
<td>1.03</td>
<td>[0.68, 1.57]</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>AA</td>
<td>35 (29.4%)</td>
<td>104 (32.1%)</td>
<td>0.88</td>
<td>[0.56, 1.39]</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>G</td>
<td>55 (45.8%)</td>
<td>141 (43.5%)</td>
<td>1.10</td>
<td>[0.81, 1.48]</td>
<td>0.370</td>
<td>0.540</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A</td>
<td>64 (54.2%)</td>
<td>183 (56.5%)</td>
<td>0.91</td>
<td>[0.68, 1.23]</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Note. SNP: single nucleotide polymorphism; TD: tardive dyskinesia; OR: odds ratio; CI: confidence interval.
DRD2 and BDNF, but this role seems to be minor (Souza et al., 2010). This conclusion could shed some light on the molecular events taking place in GABAergic MSNs of the striatum and indicate that the downstream activation of GSK3β by superactive DRD2 is not a major factor contributing to the suspected degeneration of these neurons.

The results of our study cannot exclude involvement of the G protein-dependent DRD2 signaling leading to cAMP inactivation (Beaulieu & Gainetdinov, 2011; Greengard, 2001) in the pathogenesis of TD, in a similar manner the G protein-dependent DRD1 signaling leading to cAMP activation is involved in levodopa-induced dyskinesia (Loonen & Ivanova, 2013; Santini et al., 2007). To explore this possibility, additional studies are warranted.

As previously suggested, neurotoxicity brought about by excessive dopamine metabolism could be another possible mechanism of the pathogenesis and the main reason of the neurodegenerative processes in TD (Loonen & Ivanova, 2013). This alternative pathogenic mechanism of TD, unrelated to the upregulated supersensitive DRD2s, could be rooted in formation of toxic molecules, such as dopamine quinines and reactive oxygen species, that are by-products of excessive dopamine metabolism brought about by compensatory increased synaptic concentration of dopamine following blockade of DRD2s by antipsychotics (Cho & Lee, 2013; Cyr et al., 2003; Lohr, Kuczenski, & Niculescu, 2003; Loonen & Ivanova, 2013; Waln & Jankovic, 2013). These molecules may be particularly toxic to the GABAergic MSNs of the indirect pathway that may undergo cell death (Loonen & Ivanova, 2013), which would also explain why the symptoms occur late during treatment and in some patients become permanent. Further investigation of genes that respond to oxidative stress, such as manganese superoxide dismutase (SOD2; Al Hadithy et al., 2010; Zai et al., 2018) and phosphatidylinositol-4-phosphate-5-kinase type IIA (PIPK2A; Fedorenko et al., 2014), is therefore warranted.

5 CONCLUSION

This study showed no evidence that AKT1 and GSK3B play a major role in the pathogenesis of antipsychotic-induced TD, despite the important role the two kinases play in the G protein-independent DRD2 signaling. These results support previously reported data and warrant an exploration of other pathogenic mechanisms, such as neurotoxicity due to excessive dopamine metabolism.

ACKNOWLEDGEMENTS

This study was a result of collaboration between the Mental Health Research Institute in Tomsk and the Groningen Research Institute of Pharmacy (GRIP) of the University of Groningen. The part of the study done in Russia was carried out within the framework of the Competitiveness Enhancement Program of Tomsk Polytechnic University; this program did not provide financial support for the study.

FUNDING

This work was supported by the comprehensive program for fundamental scientific research “Interdisciplinary Integrated Studies,” years 2018–2020, of the Siberian Branch, Russian Academy of Sciences (Grant 30).

CONFLICTS OF INTEREST

The authors report no conflicts of interest in this work.

ORCID

Anastasia Levchenko https://orcid.org/0000-0003-3509-1304

REFERENCES


