RESEARCH ARTICLE

Schizophrenia Gene Expression Profile Reverted to Normal Levels by Antipsychotics

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Abstract

Background: Despite the widespread use of antipsychotics, little is known of the molecular bases behind the action of antipsychotic drugs. A genome-wide study is needed to characterize the genes that affect the clinical response and their adverse effects.

Methods: Here we show the analysis of the blood transcriptome of 22 schizophrenia patients before and after medication with atypical antipsychotics by next-generation sequencing.

Results: We found that 17 genes, among the 21 495 genes analyzed, have significantly-altered expression after medication (p-value adjusted [Padj] <0.05). Six genes (ADAMTS2, CD177, CNTNAP3, ENTPD2, RFX2, and UNC45B) out of the 17 are among the 200 genes that we characterized with differential expression in a previous study between antipsychotic-naïve schizophrenia patients and controls (Sainz et al., 2013). This number of schizophrenia-altered expression genes is significantly higher than expected by chance (Chi-test, Padj 1.19E-50), suggesting that at least part of the antipsychotic beneficial effects is exerted by modulating the expression of these genes. Interestingly, all six of these genes were overexpressed in patients and reverted to control levels of expression after treatment. We also found a significant enrichment of genes related to obesity and diabetes, known adverse affects of antipsychotics.

Conclusions: These results may facilitate understanding of unknown molecular mechanisms behind schizophrenia symptoms and the molecular mechanisms of antipsychotic drugs.

Keywords: antipsychotics, gene differential expression, next generation sequencing, RNA profile, schizophrenia

Introduction

Schizophrenia is a severe and chronic mental disorder whose onset typically occurs in late adolescence to young adulthood. The lifetime risk of schizophrenia ranges from 0.3% to 0.66% worldwide, and up to 2.3% if other psychotic disorders are considered. The standard of schizophrenia treatment is antipsychotic medications (Insel, 2010). The mechanism of action of these medications tends to block receptors in the brain's dopamine D2 pathways (Jones and Pilowsky, 2002). Antipsychotics only achieve clinical improvement in a percentage of patients and also are highly associated with adverse effects (van Os and Kapur, 2009). Lifetime antipsychotic treatment is frequently required to prevent illness relapses and to maintain functionality. The use of antipsychotics also represents a huge economic burden to society. Studies of clinical response variability have

established the importance of functional mutations detected in CYP metabolic enzymes (Nebert and Dieter, 2000). Other studies focused in genes related to neurotransmitter (dopamine and serotonin) pathways traditionally involved with schizophrenia have not clarified the molecular basis of the clinical response.

Alterations in gene expression could provide relevant information on the molecular basis of the disease and the mechanisms of action of the antipsychotics (Chana et al., 2013). Post-mortem studies of gene expression obtained from brains are affected by the period of time from death to sample collection, by the change in mRNA levels caused by all possible medication given to the individuals, and by the number of genes altered by the agony of death. However, a viable alternative is the study of mRNA levels in lymphocytes from living individuals, which are free of the variability observed in post-mortem studies (Czermak et al., 2004). It has been reported that the expression level of genes encoding neurotransmitter receptors and other proteins is similar in peripheral blood lymphocytes and in the central nervous system (Glatt et al., 2005).

To identify genes targeted by atypical antipsychotics, we investigated the expression profiles altered by these drugs. Here we report the first study, to our knowledge, of gene differential expression in blood between drug-naïve schizophrenia patients and the same patients after treatment with atypical antipsychotics using next-generation sequencing data. We hypothesize that this information could be used to identify biomarkers for the disease, to monitor the effect of the antipsychotics, and for drug repositioning and development. We sequenced total mRNA of a cohort of 22 individuals (Table 1) before treatment and the same individuals 3 months after treatment started. All patients showed improved symptoms after treatment (Table 2). These 22 patients are part of a larger cohort used for a previous case/control study (Sainz et al., 2013).

Methods

Study Setting

The cohort analyzed in this study (Table 1) was obtained from an ongoing epidemiological and three-year longitudinal intervention program of first-episode psychosis (PAFIP) conducted

Table 1. Sociodemographic and Clinical Characteristics of Study Individuals

Characteristics	Total (n = 22) Mean	SD
Age at admission (years)	32.1	10.6
Age at psychosis onset (years)	31.7	10.5
Duration of untreated psychosis (months)	4.5	8.0
Duration of untreated illness (months)	7.8	10.2
	n	%
Sex		
Male	8	36.4
Female	14	63.6
Race		
Caucasians	22	100
Family history of psychosis (yes)	7	31.8
Tobacco use at intake (yes)	13	59.1
Cannabis use at intake (yes)	7	31.8
Alcohol use at intake (yes)	11	50.0

at the outpatient clinic and the inpatient unit at the University Hospital Marques de Valdecilla, Cantabria, Spain (Pelayo-Teran et al., 2008). Conforming to international standards for research ethics, the research in this article was approved by the Cantabria Ethics Institutional Review Board. Patients meeting inclusion criteria and their families provided written informed consent to be included in the PAFIP. The biological samples of patients included in the study were provided by the Valdecilla biobank.

Subjects

All referrals to PAFIP were screened for patients who met the following criteria: (1) 15–60 years; (2) living in the catchment area (Cantabria); (3) experiencing their first episode of psychosis; (4) no prior treatment with antipsychotic medication or, if previously treated, a total lifetime of adequate antipsychotic treatment of less than 6 weeks; and (5) DSM-IV criteria for schizophrenia, schizophreniform disorder, schizoaffective disorder, or brief psychotic disorder. Patients were excluded for any of the following reasons: (1) meeting DSM-IV criteria for drug dependence; (2) meeting DSM-IV criteria for mental retardation; or (3) having a history of neurological disease or head injury. The diagnoses were confirmed using the Structured Clinical Interview for DSM-IV, carried out by an experienced psychiatrist 6 months after the baseline visit. Our operational definition for a first episode of psychosis included individuals with a non-affective psychosis (meeting the inclusion criteria defined above) who have not previously received antipsychotic treatment regardless of the duration of psychosis. Twenty-two individuals, who gave written consent to their participation in the program, fulfilled inclusion criteria at 6 months, and had mRNA samples at baseline and at 3 months, were included in our analyses.

Premorbid and Sociodemographic Variables

Premorbid and sociodemographic information was recorded from patients (Table 1), relatives, and medical records. Age of onset of psychosis was defined as the age when the first continuous (present most of the time) psychotic symptom emerged. Duration of untreated illness was defined as the time from the first unspecific symptoms related to psychosis (for such a symptom to be considered, there could be no return to the previous, stable level of functioning) to initiation of adequate antipsychotic drug treatment. Duration of untreated psychosis was defined as the time from the first continuous (present most of the time) psychotic symptom to initiation of adequate antipsychotic drug treatment. Information about alcohol, cannabis, and tobacco consumption was converted into binary variables and coded for either the presence or absence of use.

Study Design

This is a prospective, randomized, flexible-dose, open-label study. We used a simple randomization procedure. At study intake, all patients were antipsychotic-naïve and were randomized to aripiprazole (n = 13), risperidone (n = 3), olanzapine (n = 1), quetiapine (n = 3), or ziprasidone (n = 2). Dose ranges were 5–30 mg/day aripiprazole, 2–6 mg/day risperidone, 5–20 mg/day olanzapine, 40–160 mg/day ziprasidone, and 100–600 mg/day quetiapine. A rapid titration schedule (5 days) was used until the optimal dose was reached, unless severe side effects occurred. At the treating physician´s discretion, the dose and type of antipsychotic medication could be changed based on clinical efficacy and the profile of side effects during the follow-up period. Antimuscarinic

medication, lormetazepam, and clonazepam were permitted for clinical reasons. No antimuscarinic agents were administered prophylactically. Antidepressants and mood stabilizers were permitted if clinically needed. At the 3-month follow-up, patients were on: aripiprazole (n = 9), risperidone (n = 5), olanzapine (n = 4), quetiapine (n = 1), ziprasidone (n = 2), and risperidone depot (n = 1). The mean daily dose of antipsychotics during follow-up was 181.72 (93.97) mg/day of chlorpormazine-equivalent doses. The mean daily dose of antipsychotics at 3 months was 163.64 (86.47) mg/day of chlorpormazine-equivalent doses. A more detailed description of medication intake during the follow-up period is available upon request.

Clinical Assessment

The severity of the Clinical Global Impression (CGI; 3) scale, the Brief Psychiatric Rating Scale (BPRS; expanded version of 24 items), the Scale for the Assessment of Positive symptoms (SAPS), the Scale for the Assessment of Negative symptoms (SANS), the Calgary Depression Scale for Schizophrenia (CDSS) and the Young Mania Rating Scale (YMRS) were used to evaluate clinical symptomatology. The same trained psychiatrist (Dr Crespo-Facorro) completed all clinical assessments. The severity of symptoms at baseline and at 3 months and the percentage of change at 3 months are reported in Table 2 (a reduction in total scores means clinical improvement).

The analysis of clinical efficacy reveals that there was a significant improvement of general psychopathology (assessed by the BPRS total score change at 3 months) and positive symptoms (hallucinations, delusions, positive formal thought disorders, bizarre behavior; assessed by SAPS total score changes at 3 months; Table 2). The patients were defined as responders to the optimum dose of antipsychotic at 3 months if there was a >50% reduction of the BPRS total score compared to intake. The rate of responders at 3 months was 95.55%.

Laboratory Assessments

Blood samples were assessed for biochemical and hematological parameters. To minimize the effects of diet and technique, blood samples were obtained from fasting subjects from 08:00 to 10:00 hours by the same personnel and in the same setting. A detailed description of methodology followed to assess hematological and biochemical variables (data available upon request). No patient had a chronic inflammation or infection, or was taking medication that might influence the results of blood tests. Smoking status and alcohol use were recorded at baseline. Anthropometric, hematological, and metabolic variables at baseline and 3 months, and changes at 3 months, are described in Table 3.

RNA Extraction

Total RNA was extracted from blood using the Tempus™ Blood RNA Tube and Tempus™ Spin RNA Isolation Kit (Applied Biosystems) using the manufacturer's protocols. To define expression profiles, a key factor is that the RNA is intact. To select

Table 2. Psychopathology at Baseline and 3 Months, and Magnitude of the Change

	Total (n = 22)						
	Baseline		3 months		3-month change from baseline		
Variable	Mean	SD	Mean	SD	Mean	SD	%
CGI total score	6.0	0.8	2.3	1.1	-3.6	1.6	n/a
BPRS total score	53.0	9.1	28.7	3.9	-24.3	10.2	-45.9
SANS total score	5.8	5.7	4.0	3.7	-1.8	6.9	-30.7
SAPS total score	12.0	3.6	0.4	1.0	-11.7	3.9	-97.0
CDSS total score	2.0	3.1	1.4	3.6	-0.6	5.3	-30.2
YMRS total score	10.9	6.7	0.4	1.0	-10.5	6.9	-96.3

BPRS: Brief Psychiatric Rating Scale; CDSS: Calgary Depression Rating Scale for Schizophrenia; CGI: Clinical Global Impression; SANS: Scale for the Assessment of Negative Symptoms; SAPS: Scale for the Assessment of Positive Symptoms; SD: standard deviation; YMRS: Young Mania Rating Scale.

Table 3. Blood and Metabolic Parameters of the 22 Patients at Baseline and at 3 Months

	Total (N=22)						
	Baseline		3 months		3-month change from baseline		
Variable	Mean	SD	Mean	SD	Mean	SD	%
BMI (kg/m2)	22.5	2.7	24.0	2.5	1.4	2.2	6.4
Weight (kg)	62.9	11.0	66.9	10.1	4.0	5.9	6.3
Leukocytes (10^3 μ/L)	7.6	1.8	6.6	1.3	-1.0	2.2	-13.4
Hemoglobin (g/dl)	14.1	1.0	14.0	1.0	-0.1	1.2	-0.5
Glucose (mg/dl)	86.1	12.9	84.4	7.2	-1.7	14.3	-2.0
GGT (U/l)	14.4	7.4	15.8	9.0	1.4	7.7	9.5
Cholesterol (mg/dl)	176.3	30.2	185.6	33.7	9.3	30.2	5.3
Triglycerides (mg/dl)	73.4	26.0	88.9	51.0	15.6	53.7	21.2
HDL cholesterol (mg/dl)	59.4	17.9	58.0	11.8	-1.4	14.9	-2.4
CRP (mg/dl)	0.5	1.1	0.1	0.2	-0.3	1.1	-75.4

BMI: body mass index; CRP: C-reactive protein; GGT: gamma-glutamytransferease; HDL: high density lipoprotein; SD: standard deviation.

only RNA with good quality, the RNA Integrity Number (RIN) was characterized with a Bioanalyzer (Agilent Technologies) and samples with a RIN of at least 7.2 were selected. The selected samples have RINs that range from 7.2 to 9.8 with an average of 8.5. Overall, the rate of samples from patients with a RIN equal to or higher than 7.2 was slightly higher than 89%.

RNA Next Generation Sequencing

The mRNA obtained from blood was sequenced using a Genome Analyzer IIx from Illumina. Total RNA was extracted from peripheral blood of each individual. The mRNA was isolated from the total RNA and, once transformed into cDNA, was fragmented by sonication. Fragments of 300 bp on average were selected to construct the libraries for sequencing. The libraries to sequence were constructed using the reagents according to the indications of the manufacturer (Illumina). Single-end sequences of 35 nucleotides were produced using a Genome Analyzer IIx (Illumina). Alignment of the reads was performed in an Simple Linux Utility for Resource Management High Performance Computer server running Tophat 2.0.6 with default options (Trapnell et al., 2009). Tophat aligns RNA-Seq reads to genomes using the Bowtie 2.0.2 alignment program (Langmead et al., 2009), and then analyzes the mapping results to identify splice junctions between exons.

Differential Expression Statistical Analyses

Bedtools 2.17.0 (multicov option; Quinlan and Hall, 2010) was used to count the amount of reads mapped to each gene. The Reference Sequence gene coordinates were defined using the RefFlat file from the UCSC Genome Bioinformatics Site (retrieved February 28, 2014). DESeq 1.10.1 package (Anders and Huber, 2010), setting up fit-only as fitting method, was used to test for differential expression using gene-count data.

Validation of Expression Data Using quantitative reverse transcription PCR

To validate the expression data obtained by RNA-Seq, we performed quantitative reverse transcription PCR amplification of 10 of the genes that were found to have differential expression between cases and controls (Sainz et al., 2013) on 30 randomly selected samples (15 cases before medication and 15 controls). We used pre-designed Taqman probes (Life technologies) using the manufacturer's instructions for LAMP3 (Hs00180880_m1), RFX2 (Hs01100925_m1), CTAG2 (Hs00535628_m1), GPR128 (Hs00262184_m1), IFI27 (Hs01086373_g1), ADAMTS2 (Hs01029111_m1), BRS3 (Hs00179951_m1), MSLN (Hs00245879_m1), RSAD2 (Hs00369813_m1), and IFI144L (Hs00119115_m1) using GADPH (Hs02758991_g1) as the control. Overall, we obtained a correlation ratio of 87.15% between the two technologies, with gene-individual correlation ratios varying from 75.0% (CTAG2) to 99.9% (MSLN).

Results

We found 17 genes with significant differential expression (Table 4) after adjusting for multiple testing (Padj Value <0.05) in the 21 495 human genes analyzed (Supplementary Table 1). In the previous study (Sainz et al., 2013) we found 200 genes with differential expression between schizophrenia patients and controls (36 schizophrenic, drug-naïve patients and 40 controls). Six of the differentially-expressed genes between untreated/treated patients (ADAMTS2, CD177, CNTNAP3, ENTPD2, RFX2, and UNC45B) also have differential expression between drug-naïve

patients/controls. The 6 schizophrenia-related genes that we found in our previous study case/control (Sainz et al., 2013) represent a large fraction (35.29%) of the 17 genes that we found with differential expression after medication with antipsychotics. This fraction is significantly higher (approximately 39-fold) than the one expected by chance (Chi test, *p*-value = 1.19E-50). All six genes that we previously implicated with schizophrenia have their expression levels significantly higher in patients than in controls and, after medication with atypical antipsychotics, their expression reverted to control levels. Normalized data that reflects this reversion is depicted in Figure 1. These results support the theory that the expression of these six genes could be modulated by the drugs and the possibility that they are implicated in the positive symptoms of the disease (hallucinations, delusions, positive formal thought disorders, bizarre behavior) that are improved by the antipsychotics.

To determine if the 17 genes with altered expression by antipsychotics were enriched for certain biological processes, molecular functions, or cellular localizations, data from the Gene Ontology Repository was analyzed using the tool FatiGO (http://babelomics.bioinfo.cipf.es/). We found enrichment for the cellular components—extracellular matrix and proteinaceous extracellular matrix—which are involved in many functions, such as segregating tissues from one another and regulating intercellular communication. The fraction of genes modulated by antipsychotics in both categories represents 23.53%, while expected fractions are 1.49% and 1.59% respectively (Padj value = 0.015 for both categories). The four genes included in these categories are ENTPD2, MMP8, ADAMTS2, and CRISP3. Gene Set Enrichment Analysis did not show any pathway enriched for the 17 genes with differential expression.

Eight genes out of the 17 (LTF, OLFM4, ADAMTS2, RFX2, ALPL, MMP8, ABCA13, and INSC) appear to associate, either as reported or as mapped gene, with 24 diseases/traits in the Genome Wide
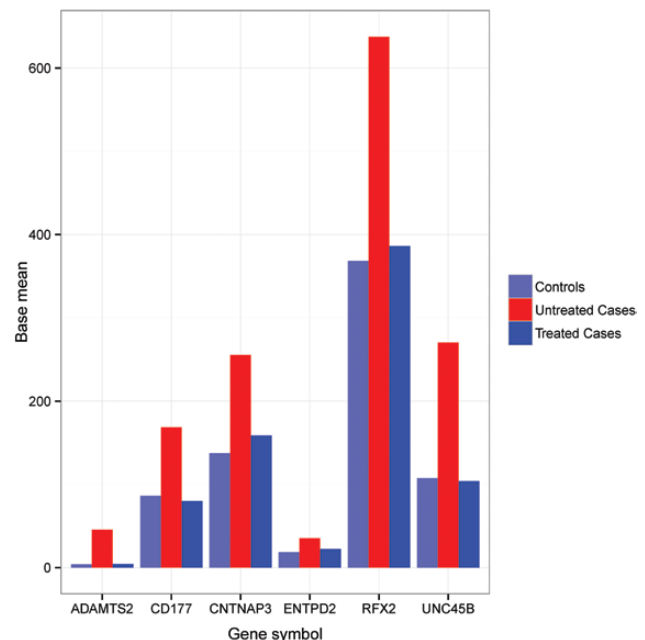

Figure 1. Schizophrenia differential expression genes reverted by antipsychotic medications. Base mean: mean of normalized counts, averaged over all samples for each condition; controls: normalized expression level of controls; untreated cases: normalized expression level of untreated patients; treated cases: normalized expression level from the patients, obtained after 3 months of treatment.

Table 4. Genes with Significant Differential Expression in Schizophrenia Patients Before and After Treatment with Antipsychotics

Gene ID	Gene symbol	Base mean	Base mean untreated	Base mean treated	Fold change	Log2 fold change	Pval	Padj
9509	ADAMTS2	23.86	43.58	4.14	0.10	-3.39	1.93E-45	3.77E-41
57126	CD177	125.21	182.64	67.79	0.37	-1.43	4.16E-15	4.06E-11
146862	UNC45B	228.32	355.59	101.05	0.28	-1.82	9.36E-13	6.09E-09
10321	CRISP3	115.80	67.59	164.01	2.43	1.28	4.87E-09	2.38E-05
85495	RPPH1	26.05	39.65	12.45	0.31	-1.67	3.21E-07	0.00125
249	ALPL	4431.75	5880.32	2983.17	0.51	-0.98	1.49E-06	0.00345
4317	MMP8	183.67	123.53	243.80	1.97	0.98	1.80E-06	0.00345
4973	OLR1	33.13	20.86	45.40	2.18	1.12	1.41E-06	0.00345
10562	OLFM4	137.50	93.08	181.92	1.95	0.97	1.94E-06	0.00345
1088	CEACAM8	187.21	127.35	247.08	1.94	0.96	1.60E-06	0.00345
154664	ABCA13	92.18	61.89	122.47	1.98	0.98	1.37E-06	0.00345
5990	RFX2	520.36	681.17	359.54	0.53	-0.92	7.00E-06	0.01051
4057	LTF	1632.35	1170.32	2094.38	1.79	0.84	6.51E-06	0.01051
2852	GPER1	28.34	36.77	19.90	0.54	-0.89	1.01E-05	0.01408
79937	CNTNAP3	195.23	249.03	141.42	0.57	-0.82	1.68E-05	0.02189
954	ENTPD2	29.04	36.51	21.57	0.59	-0.76	2.52E-05	0.03069
387755	INSC	31.74	41.60	21.87	0.53	-0.93	3.83E-05	0.04395

Base mean: mean normalized counts, averaged over all samples from both conditions; base mean untreated: mean normalized counts from condition A; base mean treated: mean normalized counts from condition B; fold change: fold change from condition A to B; log2 fold change: the logarithm, to basis 2, of the fold change; Pval: *p*-value for the statistical significance of this change; Padj: *p*-value adjusted for multiple testing with the Benjamini-Hochberg procedure, which controls false discovery rate.

Association Studies Catalog (GWAS; retrieved February 28, 2014). The association with diseases includes schizophrenia and bipolar disorder (RFX2; Wang et al., 2010), time to onset of attention deficit hyperactivity disorder (ADAMTS2; Lasky-Su et al., 2008), response to amphetamine (ABCA13; Hart et al., 2012), AIDS progression (LTF; Troyer et al., 2011), metabolic traits (ALPL; Suhre et al., 2011), obesity (OLFM4; Bradfield et al., 2012), obesity-related traits (INSC; Comuzzie et al., 2012), and visceral adipose tissue adjusted for body mass index (ADAMTS2; Fox et al., 2012), among other disease-traits. The three genes associated with obesity represent a fraction (3 out of 8 or 37.5%) larger than the fraction of genes present in the complete GWAS Catalog (15.5%).

According to the scientific literature deposited in the Gene Reference into Function (GeneRIF) repository at the National Center for Biotechnology Information (retrieved February 28, 2014), 15 out of the 17 differential expression genes are annotated for disease or functionality in 719 publications. Among these genes, ABCA13 has been implicated in schizophrenia, bipolar disorder, and depression (Knight et al., 2009). The 15 differential-expression genes annotated are significantly enriched for two known adverse effects caused by the atypical antipsychotics: obesity and diabetes. Five genes—GPER (Haas et al., 2009), LTF (Moreno-Navarrete et al., 2013), MMP8 (Belo et al., 2009), OLR1 (Brinkley et al., 2008), and OLFM4 (Bradfield et al., 2012)—have been implicated with obesity: these genes represent 33.33% of the differential-expression genes, while in the complete GeneRIF repository there are only 3.99% of the human genes annotated for obesity (Fisher; *p* = 0.0002). Four of the genes—ALPL (Kanazawa et al., 2009), LTF (Vengen et al., 2010), MMP8 (van der Zijl et al., 2010), and OLR1 (Tan et al., 2008)—have been implicated with diabetes: these genes represent 26.67% of the differential-expression genes, while 7.88% (Fisher; *p* = 0.026) is expected. Moreover, it is known that all antipsychotics tend to block receptors in the brain's dopamine pathways, and that estrogen increases the concentration of neurotransmitters such as dopamine at neuronal synapses and their receptors, affecting their release, reuptake, and enzymatic inactivation (Mcewen and Alves, 1999). We found a significant enrichment of genes related to estrogen, including an estrogen receptor: GPER (Revankar et al., 2005), LTF (Teng et al., 2002), and OLFM4 (Dassen et al., 2010) represent a fraction of 20.00% of the total, while we expect to find 4.3% (Fisher; *p* = 0.026). Previously, we described an alteration of inflammatory genes in schizophrenia (Sainz et al., 2013): we also find in GeneRIF an enrichment of inflammatory genes among the genes with their expression modulated by the atypical antipsychotics. There are six genes—ALPL (Cheung et al., 2008), GPER (Chakrabarti and Davidge, 2012), LTF (Weinberg, 2001), MMP8 (Leppilahti et al., 2011), OLR1 (Takanabe-Mori et al., 2013) and CRISP3 (Plager et al., 2010)—related to inflammation, representing 40.00%, while only 10.97% is expected (Fisher; *p* = 0.005). These results are encouraging, though we recognize that it is really difficult to assess enrichment for such a small number of genes.

Discussion

We characterized 17 genes with differential expression in schizophrenia patients before and after medication. Previously, we characterized 200 genes with differential expression between non-medicated schizophrenia patients and controls. Six of the genes with expression modulated by the atypical antipsychotic medication also have altered expression in our case/control study of schizophrenia. The six genes with altered expression in both studies are overexpressed in schizophrenia and reverted to normal levels of expression after medication with atypical antipsychotics. The fact that the expression of these six genes is downregulated by antipsychotics and that they are overexpressed in schizophrenia is not proof of causality, but does make them candidates to have a relationship with some of the psychotic symptoms of the disease. It is known that the positive symptoms of the disease, such as hallucinations, delusions, and mood swings, are improved by the antipsychotics, resulting in a lower risk of suicide, better functional capacity, and an improved quality of life (Kapur and Remington, 2001). Given that more than 95% of the individuals had a good response to medication, we can hypothesize that the six genes reported are related functionally to these positive symptoms of psychosis. It is of note that nearly 95% of the patients were categorized as good responders to antipsychotic treatment. It is likely that

the method of calculation we followed to assess the reduction of symptom severity may explain, at least in part, the high response. As has been proposed recently (Leucht et al., 2010; Obermeier et al., 2010), the calculation of responder rates based on percentage reduction of the BPRS total score from baseline has to be carried out after subtracting the 24 minimum points of the total score (24 points means no symptoms). Previous publications, in which the percentage reduction has not been correctly calculated, would have found lower numbers of responders. In addition, the fact that our patients were included in an intensive clinical program for treatment of first-episode individuals may have influenced the high rate of responders. All of the 17 genes characterized with expression significantly altered by the atypical antipsychotics are good candidates to be responsible for some of the adverse effects associated with the drugs: significant weight gain, increased risk of diabetes and stroke, blood clots, sexual dysfunction, and hyperlipidemia (Ucok and Gaebel, 2008). Our analysis of the results, with the caveat that it is based on a small number of genes, supports this hypothesis: according to the scientific literature, there is a significant enrichment of genes related to diabetes and obesity among the differential-expression genes. We also found an enrichment of genes associated to obesity or related traits according to the data deposited in the GWAS Catalog. This suggests that some of the genes with expressions altered by the atypical antipsychotics could be causative of obesity and diabetes. Additionally, the genes characterized with altered expressions are consistent with the known molecular mechanisms of the antipsychotic action: we find that there is a significant enrichment of genes related to estrogen that regulate the levels of dopamine receptors, and it is known that antipsychotics interact with receptors in the brain's dopamine pathways (Seeman, 2002).

We recognize that this study has the limitation of a small sample size and that a larger study is needed. But despite this limitation, we were able to obtain significant results, which could help to predict the effects of the atypical antipsychotics currently used in the clinical practice by monitoring the expression levels of the reported genes. We provide six candidate genes for schizophrenia that could be involved in the positive symptoms improved by the drugs, and a list of 17 genes that could be responsible for some of their adverse effects, such as obesity and diabetes. This information could help us not only to better understand schizophrenia, but also to develop new drugs and to improve the ones used today against psychosis.

Supplementary Material

For supplementary material accompanying this paper, visit http://dx.doi.org/10.1017/S00000000000000

Statement of Interest

The authors declare no conflicts of interest in connection with the article.

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