

MODERN TREATMENT APPROACHES IN PSYCHOSES. PHARMACOGENETIC, NEUROIMAGISTIC AND CLINICAL IMPLICATIONS

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Abstract

We approached the theme of modern pharmacological treatment strategies and the implication of pharmacogenetic testing in the management of child and adolescent psychoses. Our research was conducted in the period 2010 - 2015 on 210 patients, as follows: 150 children and adolescents with psychosis – a group of 75 children-G1, who benefited in choosing the pharmacotherapy from pharmacogenetic testing and 75 without testing-G2 and 60 children with UHR (ultra-high risk) for psychosis - 30 benefited of pharmacotherapy after pharmacogenetic testing and 30 without. The patients were also evaluated through MR Spectroscopy at baseline and after pharmacotherapy. The efficacy of the chosen therapy in correlation with the pharmacogenetic testing was evaluated through the mean change in the PANSS (Positive And Negative Syndrome Scale) total scores of the applied scales and through the change registered for the relevant neurobiological markers and MR (magnetic resonance spectroscopy) metabolites, from baseline till endpoint in different timepoints. The pharmacogenetic testing was done through genotyping the SNP – single nucleotide polymorphisms through RT-PCR (polymerase chain reaction), after DNA extraction. Our results show statistically significant differences of the clinical scores between the studied groups: for the subjects who benefited of pharmacogenetic testing, the PANSS, the global functioning scores proved a higher clinical improvement, a better compliance and also an improvement concerning the MR spectroscopy dosed metabolites values. Our research was a proof, sustaining the use of the pharmacogenetic testing in clinical practice and the value of investigating relevant neurobiological and neuro-imagistic markers for a personalized, tailored therapy in child and adolescent psychosis and for UHR categories, as a fruitful pathway of intervention and care.

Rezumat

Am abordat tema strategiilor moderne de farmacoterapie, a implicării diferitelor strategii de intervenție și a testării farmacogenetice în managementul psihozelor la copil și adolescent. Cercetarea a fost realizată în perioada 2010 - 2015 pe 210 pacienți, astfel: 150 copii și adolescenți cu psihoze - un grup de 75 de copii-G1, care a beneficiat în alegerea farmacoterapiei de testare farmacogenetică prealabilă și 75 fără testare-G2 și 60 de copii UHR – cu înalt grad de risc pentru psihoză - 30 au beneficiat de farmacoterapie după testarea farmacogenetică, 30 fără. De asemenea, pacienții au fost evaluați prin spectroscopie de rezonanță magnetică (SRM) la momentul inițial și după farmacoterapie. Eficacitatea terapiei alese în corelație cu testarea farmacogenetică a fost evaluată prin intermediul modificării scorurilor totale ale scalelor aplicate și prin schimbarea înregistrată pentru markerii neurobiologici relevanți și a metabolitelor SRM, de la valoarea inițială până la punctul final în diferite momente de timp. Testarea farmacogenetică a fost realizată prin genotiparea SNP – polimorfismelor nucleotidice singulare, prin RT-PCR (reacție de polimerizare în lanț), după prelevarea de ADN. Rezultatele noastre arată diferențe semnificative statistic ale scorurilor clinice între grupurile studiate: pentru acei subiecți care au beneficiat de testare farmacogenetică, scorurile PANSS, scorurile globale ale funcționării au dovedit o îmbunătățire clinică mai puternică, o complianță mai bună și, de asemenea, îmbunătățire în ceea ce privește valorile metabolitelor dozați prin SRM. Cercetarea noastră este o dovadă care susține utilizarea testării farmacogenetice în practica clinică și valoarea investigării markerilor neurobiologici și neuroimagingistici relevanți pentru o terapie personalizată, individualizată în psihozele la copil și adolescent și pentru categoriile UHR, drept o cale de succes pentru intervenție și îngrijire.

Keywords: psychoses, pharmacogenetic testing, spectroscopy, neurobiologic markers, neuroimaging markers

Introduction

The pharmacologic management of psychoses imposes a different and integrative approach of each case, initiated as early as possible, in a multidisciplinary team, which should be flexible, coherent and synergic, in a non-restrictive, non-stigmatizing frame, which should promote the preventive strategies [1, 4]. The new discoveries in the field of pharmacogenetics and neuroimaging bring opportunities for different classification systems, which make the connection between symptoms, neuronal circuits and the neurobiological markers [6, 15]. The clinical diagnosis influences the neurobiologic research and the neurobiologic and pharmacogenetic research has impact on the medication response and the clinical evolution [2, 8-11]. Those interdisciplinary inter-relations will contribute to the impact on the risk factors, as well as on the protective ones, putting the basis of specific pharmacologic interventions [7, 12-14]. Globally there are problems concerning the management and treatment of psychoses, especially for the paediatric population [2, 11-13]. The innovative therapies encountered a stagnation and no or few medication has been discovered and studied [3, 5]. The existent pharmacologic therapies, even if we are talking about the new generation of atypical antipsychotics, are not efficient on the negative symptomatology, on the cognitive dimension and concerning the improvement of the life quality of the patients and their families, especially if they didn't benefit of a specific algorithm, in order to be indicated to the patients with psychosis [13, 16].

One of our main objectives was creating a matrix electronic platform in order to multimodally stratify the patients on symptomatic constellations and for an algorithm for calculating and choosing the treatment of election in correlation with the neurobiological, pharmacogenetics, neuroimaging markers and clinical profile of the target patients. The UHR (ultra-high risk) phenotype represents a high risk for the psychosis onset and we need to offer special attention to the fact that the deficient functioning is also correlated with subsequent neurobiological processes. Those aspects must be known, when we choose the suitable pharmacotherapy, which should take the neurobiological, metabolic, neuroimaging and pharmacogenetic markers into account [1, 13]. Through the pharmacogenetic testing, the effects of the genetic variations on the medication response, safety, tolerability and efficacy, those genetic variations - polymorphisms, being responsible for the metabolism type, the pharmacokinetics and pharmacodynamics of the medication, are investigated. So that, extensive, slow, intermediary and ultra-rapid metabolizers are identified [4, 13].

The major part of the psychiatric medication is metabolized at the level of cytochrome P450-

CYP2D6 (85% of the antidepressive medication and 50% of the antipsychotics) and CYP2C19 but also by CYP1A2 - 24% of the antidepressives and 18% of the antipsychotics by CYP3A4 - 38% of the antidepressives and 23% of the antipsychotics. So that, the enzymes of the P450 cytochrome - CYP2D6 metabolize antidepressants - SSRI's (serotonin reuptake inhibitors) - sertraline, paroxetine, fluoxetine, fluvoxamine, all first generation, typical antipsychotics, also atypical antipsychotics - aripiprazole, risperidone, olanzapine; quetiapine and ziprasidone by CYP3A4; clozapine by CYP1A2, CYP2C19, CYP2D6, CYP3A4 and olanzapine by CYP2D6 but also by CYP1A2. Those aspects are very significant, when we choose the medication that should be administered [1, 4, 13].

In our research, we also captured the modern pharmacologic treatment approaches correlated with the evaluation of the neuroimaging markers, especially through magnetic resonance spectroscopy (MR) [15]. The main objectives of our study were: the evaluation of the efficacy of the different pharmacologic interventions in the child and adolescent psychoses and in the UHR (ultra-high risk) for psychosis group and also the evaluation, through neuroimaging - spectroscopy, of the modification of the metabolites in correlation with the chosen pharmacotherapy, after and without pharmacogenetics testing, in correlation with the clinical evolution of the patients [13, 15]. The MR spectroscopy offers the possibility to quantify the chemical components from localized areas of the brain through non-invasive methods [15].

It is a versatile instrument for the clinical evaluation, longitudinal monitoring and for the evaluation of the efficacy of the administered treatment. So, the MR spectroscopy permits the *in vivo* identification and quantification of the biochemical substances [1, 15].

Materials and Methods

The present study is part of an extended research on pharmacological treatment in child and adolescent psychoses performed at the University Hospital for Child and Adolescent Psychiatry and Neurology Timișoara, Romania, between 2010 and 2015 and found especially on neurobiological, neuroimaging and clinical aspects, but also on specific pharmacogenetic correlations.

The study consisted of 210 patients, as follows: 150 patients, children and adolescents with psychosis and 60 patients with ultra-high risk of developing psychosis (UHR - ultra-high risk). The patients included in the study, were aged between 13 and 20 years (median age 15.74 ± 4). We obtained for each patient the informed assent and the informed consent from the parents/legal tutors. Our study is in accordance with the Ethical Committee regulations of the University of Medicine and Pharmacy

“Victor Babeș” Timisoara, Romania, with the ICH-GCP (Good Clinical Practice) regulations and guidelines. Our research populations were divided in 2 groups: from the 150 children with psychosis – 75 received treatment after pharmacogenetic testing and 75 did not; from the 60 UHR for psychosis group – 30 received treatment after pharmacogenetic testing and 30 did not (Figure 1).

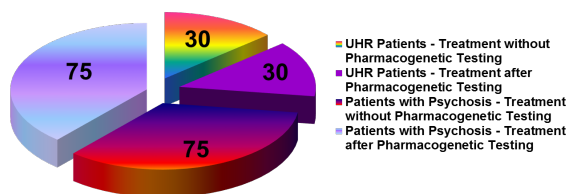


Figure 1.

The study samples of children with psychosis and UHR for psychosis in function of the administered pharmacotherapy

We applied the following scales: PANSS - Positive And Negative Syndromes Scale, CGI-S/I - clinical global impression of severity and improvement, CGAS - clinical global assessment of functioning and UKU – the adverse events scale. The pharmacogenetic testing was performed through the genotyping - SNP – single nucleotide polymorphisms, through RT-PCR, after the DNA prelevation. The genotypes of the allelic variants CYP * have been determined through the specific allelic fluorescence measurement, using the software for allelic discrimination. The identification of the alleles CYP2D6 *3 *4 *5, *41, responsible for the medication metabolizing types, was significant. Also, we identified the polymorphisms of the gene for the serotonergic receptors- 5HT_{2A}/5HT_{2C}. Also, the patients have been evaluated through MR spectroscopy at baseline and after the pharmacotherapy. So, the spectroscopy investigates key aspects of the cerebral function and metabolism. Through the MR spectroscopy we quantified the following metabolites: NAA - N-acetyl aspartat, GABA - gama-aminobutiric acid, Asp - aspartat, CR - creatine, Gln - glutamine, GPC – glicero-phospho-coline, PC - phosphocoline, PCr – phospho-creatine, Tau - taurine, N-MDA - N-metil-D-aspartat, Ser - serine, Gly - glicine, Cho - choline. The efficacy of the chosen therapy in correlation with the pharamacogenetic testing, has been evaluated through the modification of the applied scales total scores and through the change registered for the relevant neurobiologic markers and for the MR Spectroscopy metabolites, from the initial values till endpoint, in each time-point. As well, the efficacy of the chosen pharmaco-therapy in correlation with the pharmacogenetic testing and the variation of the cerebral metabolites quantified

through the MR spectroscopy was evaluated through the change of the mean total scores of the scales (PANSS, CGI-S/I, CGAS, UKU) from baseline till endpoint in different timepoints.

Results and Discussion

We identified the pharmacogenetics polymorphism at the level of CYP450 enzymes and so we observed in our studied samples the WT - wild type or normal type metabolizer, the patients who had SNP, who need in the clinical practice, the adjustment of the doses of the administered pharmacotherapy, as well as careful choosing of the medication and the WT/SNP = mixt type, who encounter also some difficulties in this area. So, the pharmacogenetics, CYP testing permitted us to adjust the medication doses accordingly and to avoid the antidepressive medication and antipsychotics, which are metabolized at the level of the CYP, where we found polymorphisms [1, 4, 13].

Also, we identified the genotypes WT/-759C/T and SNPs -795C/T in some of the patients from studied groups, these having potential of multiple adverse effects as response to SSRI antidepressants, especially relevant weight gain [11].

Concerning the plasmatic concentrations of the administered antipsychotic medication, depending on the pharmacogenetic profile of the patient, we obtained the results presented in Figure 2.

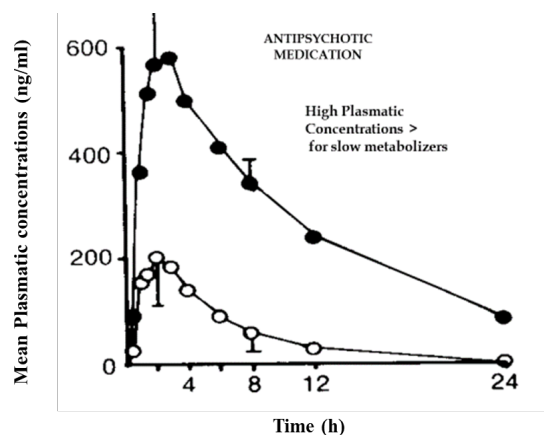


Figure 2.

Plasmatic concentrations of the administered antipsychotic medication for the slow (●) and extensive (○) metabolizers in the studied samples

So, we found high plasmatic concentrations of the antipsychotic medication for the slow metabolizers, meaning that this patient group could encounter adverse effects to the medication [1-4].

In order to correctly understand the clinical implications of this pharmacogenetic testing, when choosing the proper antipsychotic treatment, we

must know the CYP metabolism pathways for the medication, which are captured in Figure 3.

Antipsychotic	CYP2D6	CYP3A4	CYP1A2
Aripiprazole	+++	+	
Clozapine	+	+	+++
Iloperidone	+++	+	
Olanzapinae	+		+++
Paliperidone	+++	+	
Quetiapine		+++	
Risperidone	+++	+	
Ziprasidone		+	

Figure 3.

The CYP450 enzymes responsible for the metabolism of the atypical antipsychotics

So that, according to the clinical implications of the pharmacogenetic testing in psychoses, especially for children and adolescents, we observed that: for the patients with polymorphism, the SNP genotype and in the case of slow metabolizers, we must avoid

prescribing antipsychotics having strong metabolic correlations with CYP2D6 (risperidone, aripiprazole, haloperidol); we should preferably choose quetiapine, ziprasidone, eventually olanzapine or clozapine. From the category of antidepressive medication we should avoid for these patients especially the SSRI's. In the case of ultra-rapid metabolizers we must adjust the medication doses in order to have response to the administered medication, meaning the doses should be increased [1, 13].

It is very important to make the pharmacogenetic testing, in order to identify which patients could be non-responders for specific administered antidepressants and antipsychotics, because generally it takes approximatively 6 weeks until we can expect response to the chosen medication and 40% prove non-response or insufficient response to the medication. We also obtained interesting results concerning the MR spectroscopy quantified metabolites and their variation from baseline till endpoint (Figure 4 and Figure 5).

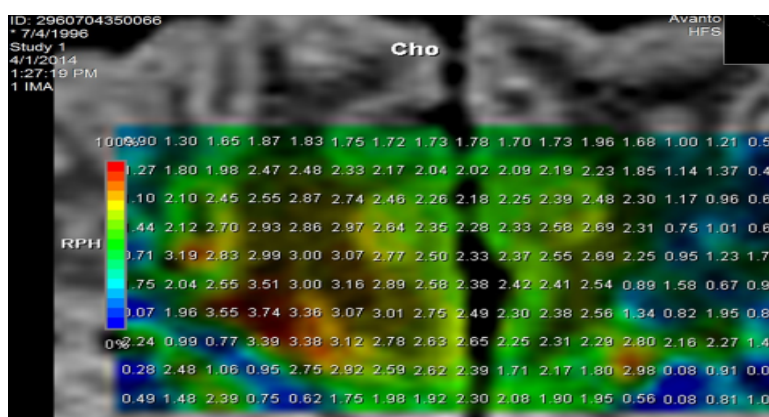


Figure 4.

Results for MR spectroscopy metabolites concentrations

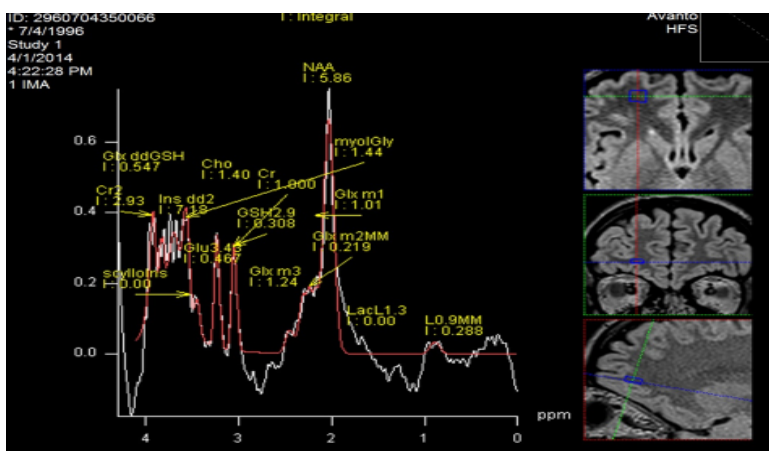


Figure 5.

Results for MR spectroscopy metabolites concentrations and peaks

For the UHR-ultra-high risk patients for psychosis, the pathologic changes of the metabolites are identified

even before the onset of psychosis. So that, the pathologic changed values of the metabolites before

the onset could help the pharmacologic intervention to be early and targeted. We also made some correlations concerning the glutamatergic pathway and the treatment response – the patients who had good clinical response, showed also the normalization of

the metabolites levels identified through the spectroscopy [15].

The results concerning MR spectroscopy metabolites peaks are presented in Figure 6.

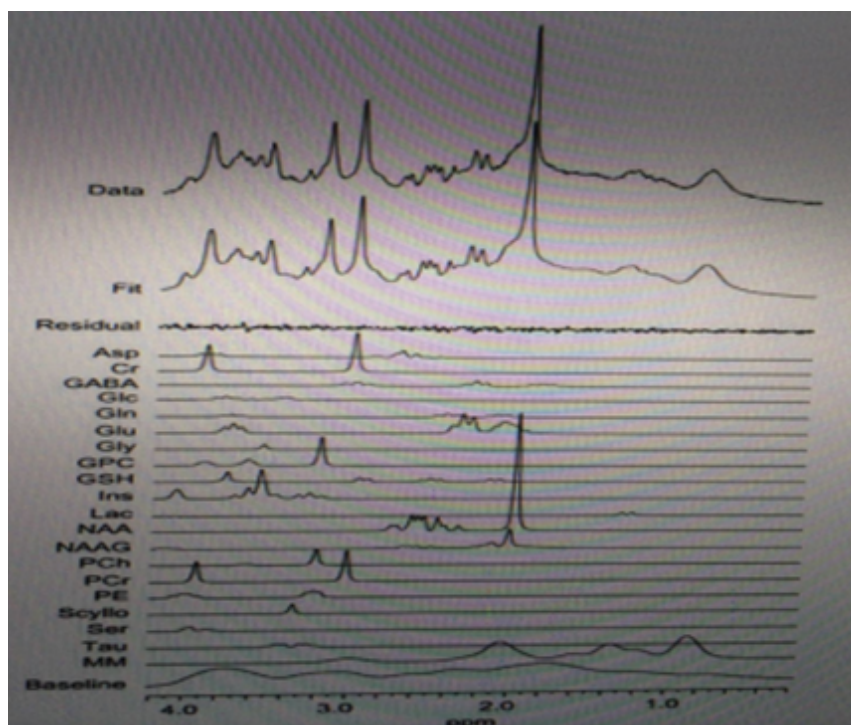


Figure 6.

Results for MR spectroscopy metabolites peaks

Some of the cerebral metabolites' modifications are reversible, if proper, carefully chosen pharmacotherapy, in function of the pharmacogenetic, neuroimaging and clinical profile of the patient, is administered [13, 15].

In both studied groups – with psychosis and UHR – we obtained statistically significant differences of the scores, between the patient group, who benefited of pharmacogenetics testing, when choosing the proper pharmacotherapy and the other group, in each timepoint of the evaluation for all the scales ($p < 0.001$, significance level $\alpha = 0.001$). The PANSS and CGI-S scores registered a statistically significant decrease and the CGAS functioning scores an improvement and the spectroscopy metabolites values improved, being equivalent with a good clinical evolution and medication response in the pharmacogenetically tested group. Through comparing the total PANSS scores and the values of the metabolites in each 2 with 2 different timepoints, through the application of the Wilcoxon signed Ranks nonparametric test,

we obtained statistically significant differences in the group who benefited of pharmacogenetics testing, with choosing the suitable pharmacotherapy, proving a good clinical evolution in time and medication response ($p < 0.001$, significance level $\alpha = 0.001$). The results prove that the patients, who take a pharmacotherapy chosen after the pharmacogenetics testing, registered the improvement of the Spectroscopy metabolites, as a positive response to the chosen pharmacotherapy. In the other group, without pharmacogenetics testing, we could observe non-response, lack of improvement of the metabolites correlated with multiple adverse effects in the UKU scale. Comparing the differences between the 2 groups, concerning the total mean PANSS scores for each analysed moment, applying the Mann-Whitney nonparametric test, we observed the decrease of the PANSS scores, good clinical evolution in the G1 group – with pharmacogenetics testing and poor scores with non-response in G2 (without pharmacogenetic testing) (Table I).

Table I

The comparison of the PANSS scores between G1- study group with pharmacogenetics testing and G2 without, in each timepoint

Time point	Group	PANSS Medium	Std. Deviation	Std. Error Mean
T0	G1	117.2	28.16	3.87
	G2	120.4	35.19	7.04
T1	G1	103.4	23.60	3.24
	G2	122.2	32.62	6.52
T2	G1	90.8	19.40	2.67
	G2	126.4	30.37	6.07
T3	G1	79.8	14.48	1.99
	G2	128.9	30.84	6.17
T4	G1	69.7	12.29	1.69
	G2	138.2	26.14	5.23

Through Spearman correlation test, we obtained statistical significant positive correlations between the improvement of the metabolites in spectroscopy

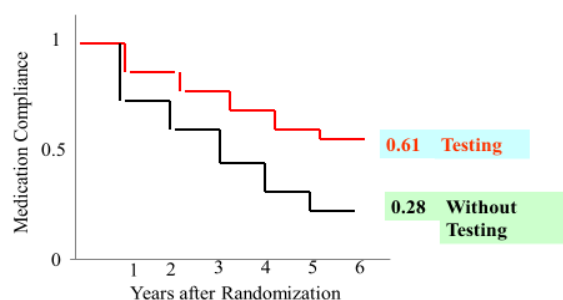
and the pharmaco-genetic testing for choosing the suitable pharmaco-therapy, this group having also good response to the medication (Table II).

Table II

Spearman correlations transformed z, between the scale scores and the metabolites improvement for the studied group with Pharmacogenetic Testing and without

	Patients with Psychosis						UHR for Psychosis					
	with Pharmacogenetic Testing			without Pharmacogenetic Testing			with Pharmacogenetic Testing			without Pharmacogenetic Testing		
Correlations	r*	z**	z-STD**	r*	z**	z-STD**	r*	z**	z-STD**	r*	z**	z-STD**
Lower Total PANSS Scores-Metabolite values Improvement	0.969	0.521	0.373	0.313	0.369	0.363	0.974	0.844	0.698	0.269	0.189	0.939
Low CGI-S Scores-Illness Severity Metabolite values Improvement	0.977	0.355	0.301	0.221	0.311	0.253	0.982	0.185	0.195	0.112	0.223	0.179
High functioning Scores-CGAS Metabolite values improvement	0.983	0.846	0.675	0.683	0.721	0.701	0.989	0.619	0.577	0.378	0.398	0.347

Concerning the medication compliance of the study samples we obtained the results presented in Figure 7.

**Figure 7.**

The prescribed medication compliance for the studied groups with pharmacogenetic testing and without it

Our research data, prove that the emergence of the pharmacogenetic testing, of the neurobiologic and neuroimaging evaluation, as modern approaches, pinpoint a new stage in the clinical psychopharmacology, in which the genotype and the

biomarkers influence the way we choose the therapy, increasing the safety and efficacy of the utilized medication [6, 13, 15].

Our future projects are based on the implementation of testing the relevant parameters – neurobiological, pharmacogenetic and neuro-imaging markers – in the routine clinical practice as golden standard methods.

Conclusions

The medication acts on the neurobiologic modifications and not only on the categorical diagnostic criteria.

This is why, a modern approach focused on the neurobiological, pharmacogenetic and neuro-imaging markers, is needed. We have to find a new treatment approach sensitive and adapted to the existent needs of the patients.

The pharmacogenetic testing is useful in order to avoid in psychosis, the medication that is not suitable for the specific genetic, pharmacologic,

neuroimaging and clinical profile of the patient. The pharmacogenomic information should be incorporated in the electronic files of the patient because it is useful in choosing all the future treatments.

All these correlations between the neurobiologic, pharmacogenetic, neuroimaging and clinical markers and pathways are vital in choosing a proper medication in psychosis, adapted to the needs of the patient, in the frame of a modern, tailored medicine and pharmacotherapy.

The improvements registered concerning the neuroimaging parameters in patients, who had pharmacotherapy prescribed after the pharmacogenetic testing; prove once again the efficacy of this fruitful modern path of care in pharmacogenetics, genomics and pharmacotherapy.

References

1. Cacabelos R., Hashimoto R., Takeda M., Pharmacogenomics of antipsychotics efficacy for schizophrenia. *Psychiatry and Clinical Neurosciences*, 2011; 65: 3-19.
2. Calarge C.A., Miller D.D., Predictors of Risperidone and 9-Hydroxyrisperidone Serum Concentration in Children and Adolescents. *J. Child Adolesc. Psychopharmacol.*, 2011; 21(2): 163-169.
3. Dargani N.V., Malhotra A.K., Safety profile of iloperidone in the treatment of schizophrenia. *Expert Opin. Drug Saf.*, 2014; 13(2): 241-246.
4. Drozda K., Müller D.J., Bishop J.R., Pharmacogenomic Testing for Neuropsychiatric Drugs: Current Status of Drug Labeling, Guidelines for Using Genetic Information, and Test Options. *Pharmacother. J. Hum. Pharmacol. Drug Ther.*, 2014; 34(2): 166-184.
5. Echim G., Vari C.E., Kolcsar M., Cotoi O.S., Osz B.E., Groșan A., Gall Z., Dogaru M., Time and dose-related side effects of risperidone in rats. *Farmacia*, 2015; 63(6): 845-850.
6. Gervasini G., Caballero M.J., Carrillo J.A., Benitez J., Comparative Cytochrome P450 *In Vitro* Inhibition by Atypical Antipsychotic Drugs. *ISRN Pharmacol.*, 2013; 2013: 1-5.
7. Ghibu S., Ilie I., Mureșan A., Mogoșan C., Perspectives in the experimental study of the metabolic syndrome. *Farmacia*, 2015; 63(4): 482-486.
8. Lohoff F.W., Ferraro T.N., Pharmacogenetic considerations in the treatment of psychiatric disorders. *Expert Opin. Pharmacother.*, 2010; 11(3): 423-439.
9. Mackenzie B., Souza R.P., Likhodi O., Tiwari A.K., Zai C.C., Sturgess J., Müller D.J., Pharmacogenetics of antipsychotic treatment response and side effects. *Therapy*, 2010; 7(2): 191-198.
10. Müller D.J., Chowdhury N.I., Zai C.C., The pharmacogenetics of antipsychotic-induced adverse events. *Curr. Opin. Psychiatry*, 2013; 26(2): 144-150.
11. Nussbaum L.A., Dumitrașcu V., Tudor A., Grădinaru R., Andreescu N., Puiu M., Molecular study of weight gain related to atypical anti-psychotics: clinical implications of the CYP2D6 genotype. *Rom. J. Morphol. Embryol.*, 2014; 55(3): 877-884.
12. Nussbaum L., Andreescu N., Nussbaum L.M., Grădinaru R., Puiu M., Ethical Issues Related to Early Intervention in Children and Adolescents with Ultra High Risk for Psychosis: Clinical Implications and Future Perspectives. *Rev. Rom. Bioet.*, 2014; 12(3): 64-81.
13. Schatzberg A.F., Nemeroff C.B., Textbook of Psychopharmacology, Third Edition, American Psychiatric Publishing Inc, 2004.
14. Stagg C.J., Rothman D.L., Magnetic Resonance Spectroscopy, Tools for Neuroscience Research and Emerging Clinical Applications, First edition, Academic Press, 2014.
15. Voiculescu S.E., Le Duc D., Roșca A.E., Zeca V., Chițimșu D.M., Arsene A.L., Drăgoi C.M., Nicolae A.C., Zăgrean L., Schöneberg T., Zăgrean A.M., Behavioral and molecular effects of prenatal continuous light exposure in the adult rat. *Brain Research*, 2016; 1650: 51-59.
16. Zhang J.P., Malhotra A.K., Pharmacogenetics and antipsychotics: therapeutic efficacy and side effects prediction. *Expert Opin. Drug Metab. Toxicol.*, 2011; 7(1): 9-37.