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Influence of *ABCB1 3435C>T* Polymorphism on Methotrexate Safety in Patients with Psoriasis — A Secondary Publication

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Abstract: *Background:* Methotrexate is a highly effective systemic treatment for moderate to severe psoriasis, but drug toxicity may limit its use. Recent evidence suggests that it is necessary to take into account the individual characteristics of methotrexate pharmacokinetics, which are determined by the presence of polymorphisms of genes encoding methotrexate carrier proteins, to predict the risk of methotrexate-induced toxicity. *Aim:* The research aims to assess the associations of *ABCB1* rs1045642 (*3435C>T*) polymorphism with methotrexate safety for patients with moderate and severe psoriasis. *Methods:* The study included 75 psoriasis patients treated with methotrexate with 21 days of follow-up duration. Data on adverse drug reactions (ADR) were collected using a clinically structured questionnaire, and complete and biochemical blood tests, and urinalysis were performed. The severity of ADR was assessed using visual analog scales and the CTCAE toxicity scale. The severity of gastrointestinal ADR was assessed using the GSRS questionnaire. Genotyping was carried out by real-time PCR. *Results:* Gastrointestinal toxicity was detected in 38 patients (50.67%). The mean GSRS score was 7.97 ± 9.18. Analysis of differences in the ADR incidence showed the presence of statistically significant differences in the frequency of ADR in the gastrointestinal tract: the toxic effect of methotrexate was more often observed in carriers of the T allele of the *ABCB1* rs1045642 polymorphism (*3435C>T*), (CC: 2 (14.3%), TC: 18 (52.9%), TT: 18 (66.7%), $P = 0.006$). Binomial regression demonstrated the presence of a statistically significant effect of the rs1045642 single-nucleotide polymorphism of the *ABCB1* gene on the incidence of ADR from the gastrointestinal tract (OR = 8.64, $P = 0.008$). *Conclusion:* An association of *ABCB1* rs1045642 single-nucleotide polymorphism with the safety of methotrexate therapy in patients with moderate and severe psoriasis was revealed. The data obtained can be used to personalize the prescription of methotrexate to psoriasis patients.

Keywords: Pharmacogenetics; Biomarkers; Drug safety; Psoriasis; Methotrexate; Single-nucleotide polymorphism; Adverse reactions

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1. Introduction

Psoriasis is one of the most common skin diseases [1]. Psoriasis is a chronic relapsing disease whose therapeutic goal is to gain control of the disease course and minimize adverse drug reactions (ADRs) by using drugs that maintain long-term remission and are well tolerated by patients [2,3]. Methotrexate (4-deoxy-4-amino-10-methylfolic acid) is a highly effective drug for the systemic treatment of moderate to severe psoriasis, but the toxicity of the drug may limit its use [4]. According to the meta analysis conducted by West *et al.*, methotrexate has been reported in an average of 28.3% of patients. Gastrointestinal tract disorders are the most frequent ADR for methotrexate: 18.2% of patients have nausea and vomiting; 11.1% have ulcerations of the oral mucosa and other mucositis (gingivitis, ulcerative stomatitis, enteritis); 7.5% of patients reported the occurrence of abdominal pain; 6.6% of patients reported the occurrence of functional bowel disorders [5]. The quality of life of patients is severely affected, which in 13–28% of cases leads to forced interruption of treatment [6]. The main mechanism for the development of ADRs during methotrexate therapy is its cytotoxic effect on rapidly dividing cells, namely, inhibition of folate metabolism in tissues with high proliferation of cells with a high need for purines, thymidine, and methionine. Since the epithelium of the gastrointestinal tract is characterized by a high cell population renewal rate, folate deficiency is the main mechanism in the development of this group of ADRs [7]. Patients with psoriasis are often forced to use methotrexate for long periods of time, and there is currently no algorithm that can predict individual patient response to therapy; the study of safety issues of therapies is a highly urgent task. Due to the fact that low doses of methotrexate are used for psoriasis therapy, measuring the drug concentration in the plasma to predict its toxicity has no clinical significance. Recently, much attention has been paid to the role of genetic factors in individual tolerance to psoriasis therapy [8,9], they may be able to predict the development of about half of adverse responses to treatment [10].

The frequency of gastrointestinal tract ADRs may be influenced by genetic factors, namely the presence of polymorphisms in genes encoding methotrexate transporter proteins. According to current research, to predict the risk of methotrexate-induced toxicity, it is necessary to take into account individual characteristics of its pharmacokinetics, which are determined by the presence of polymorphisms in genes encoding proteins that carry methotrexate [7,9,11].

The bioavailability of methotrexate is dependent on ABC family transporters, which transport methotrexate molecules from enterocytes into the lumen of the gastrointestinal tract, as well as from P-glycoprotein (ABCB1 protein), a transporter protein that transports methotrexate molecules from enterocytes into the lumen of the gastrointestinal tract. The *ABCB1* gene, encoding P-glycoprotein, has a significant degree of polymorphism. The *3435C>T* polymorphism, which is a substitution of a cytosine nucleotide for a thymidine nucleotide at position 3435, has the highest clinical significance and prevalence [12]. It is proved that a low level of *ABCB1* expression in the intestine and kidneys leads to a decrease in P-glycoprotein content in these organs and consequently to more complete absorption and slower excretion of its substrates, which include methotrexate. As a result, the concentration of methotrexate in blood plasma is elevated, thus increasing the likelihood of developing ADRs [13].

This paper aims to identify associations of the single-nucleotide polymorphism *ABCB1* rs1045642 (*3435C>T*) with the safety of methotrexate therapy in patients with moderate to severe psoriasis. In particular, it aims to assess the frequency and severity of adverse drug reactions of the gastrointestinal tract and to analyze the relationship between the frequency and severity of adverse drug reactions and patients' genotypes.

2. Materials and methods

2.1. Study design

The prospective study was conducted in two phases: at the first stage, dynamic patients with moderate and

severe psoriasis were monitored, who were being treated in the 24-hour inpatient unit of the dermatology department, to identify adverse drug reactions that may be associated with methotrexate intake, their frequency, and severity.

From the moment of inclusion in the study, clinical, demographic, and laboratory examinations of the patients were carried out, and psoriasis severity was assessed using the Psoriasis Area and Severity Index (PASI) [14]. Data on adverse drug reactions were collected using a specially developed structured questionnaire, and complete and biochemistry blood tests, and urinalysis were performed. We analyzed the causal relationship between methotrexate intake and the development of ADRs using the Naranjo scale and the Liverpool causality assessment tool to assess the cause of adverse drug effects. The severity of adverse drug reactions was assessed using visual analog scales and the CTCAE (Common Toxicity Criteria for Adverse Event) scale.

In the second stage of the study, the frequency of alleles of the single-nucleotide polymorphism of the gene *ABCB1* (rs1045642) in this sample of patients and the association between the presence of genetic polymorphism and gastrointestinal safety parameters of methotrexate therapy were analyzed.

2.2. Eligibility criteria

75 patients diagnosed with psoriasis were included in the study. The patients were hospitalized in the Clinical Dermatology Department of the Russian Ministry of Health and were treated with methotrexate in the recommended therapeutic dosages.

The criteria for inclusion of patients in the study were:

(1) Presence of written informed consent of the patient to participate in the study.

(2) Patients with clinical forms of psoriasis: common psoriasis, pustular psoriasis (von Zumbusch pustular psoriasis, Barber's palms and soles), psoriatic erythroderma, arthropathic psoriasis.

(3) Patients receiving methotrexate during hospitalization.

The criteria for non-inclusion were:

(1) Severe somatic pathology.

(2) Psychotic state or history of severe psychotic illness.

(3) Concomitant use of drugs that affect the pharmacokinetics and/or pharmacodynamics of methotrexate.

The exclusion criteria were:

(1) Refusal of the patient to continue participation in the study.

Inclusion in the study occurred within the first 24 hours after the first methotrexate injection. Informed consent for inclusion in the study was obtained from each patient, and comprehensive information was given about the study, its aims, and results.

2.3. Research conditions

The work was carried out on the basis of the Federal State Budgetary Institution State Scientific Center for Children's Culture of the Ministry of Health of Russia (Director, Academician of the Russian Academy of Sciences, Doctor of Medical Sciences, Professor A.A. Kubanov). Pharmacogenetic studies were carried out at the Research Institute of Molecular and Personalized Medicine of the Ministry of Health.

2.4. Duration of the study

The study was conducted from 2019 through 2021, and the planned duration of follow-up for each patient was 21 days. There were no shifts in the planned time intervals during the study.

2.5. Medical intervention

2.5.1. Clinical research methods

The data on symptomatic adverse drug reactions were identified during the patient interview using a specially developed clinically structured questionnaire [15]. Questions about the tolerability of methotrexate therapy were asked to the patient daily from the moment the patient was included in the study throughout the hospitalization period. The principle of questionnaire development was based on the methodology of targeted detection of adverse drug reactions, the effectiveness of which was proved in the work of Tsvetova [16]: the patient was asked questions regarding the presence of each of the expected symptomatic adverse drug reactions across all organs and systems.

In case of a positive response, the severity of ADR was assessed by the patient independently using the visual-analog numerological evaluation scale graded from 1 to 10, they were asked to mark their perception of symptom severity. In addition, the severity of ADR was assessed using the CTCAE v5.0 Toxicity Severity Scale (Common Toxicity Criteria for Adverse Event, version 5.0, 2017) [17].

The severity of adverse drug reactions in the gastrointestinal tract was assessed using a specialized questionnaire to assess the quality of life of patients with regard to GI symptoms — GSRS (gastrointestinal symptoms rating scale) [18]. The questionnaire included 15 questions assessing patients' discomfort with symptoms of pain, reflux, dyspepsia, diarrhea, and constipation. The patient self-assessed the severity of the symptom constellation using targeted questions with a score from 1 to 7. Based on the sum, we calculated the toxicity in the gastrointestinal tract for each patient.

The Naranjo scale was used to assess the causal relationship between methotrexate intake and ADR [19]. Only those ADRs that had a definite, probable, possible degree of association were considered in the study.

2.5.2. Laboratory research methods

Biological material for genomic DNA extraction was obtained from 4 ml of venous blood collected from all patients after signing informed consent to participate in the clinical trial in the Department of Clinical Dermatology of FGBU GNTCDC Ministry of Health of Russia using a vacuum system VACUETTE (Greiner Bio-One, Austria) into tubes with 0.5 M EDTA. Blood was drawn regardless of food intake and duration of methotrexate therapy. Biological samples of whole blood were frozen at -70°C, then transported in thermocontainers to the Research Institute of Molecular and Personalized Medicine of the Russian Ministry of Health (Moscow) for genetic analysis. DNA extraction from whole venous blood samples was performed using the reagent kit "S-Sorb" (Syntol LLC, Russia) according to the manufacturer's protocol. Determination of allelic variants of *ABCB1* single-nucleotide polymorphism (C3435T, rs1045642) was performed using a commercial reagent kit (Syntol LLC, Russia) by the method of real-time PCR on the CFX96 Touch Real-Time System with CFX Manager software version 3.0 (BioRad, USA). The amplification program included incubation at 95°C for 3 minutes, followed by denaturation at 95°C for 15 seconds, and annealing at 63°C for 40 seconds for 39 cycles. The fluorescence signal was detected by the channel for the fluorophore FAM (carboxyfluorescein, absorption wavelength 492 nm, fluorescence wavelength 520 nm) and a channel for HEX fluorophore (hexachlorofluorescein, absorption wavelength 535 nm, fluorescence wavelength 556 nm).

2.6. Statistical analysis

Statistical analysis was performed using the methods of parametric and non-parametric statistics using the package of applied programs STATISTICA v10.0 (StatSoft Inc., USA). When selecting the method, the normality of sample distribution was assessed using the Shapiro-Wilk *W*-test, homogeneity of variance was assessed using the Fisher's *T*-test (when comparing two samples) and Levene's test (when comparing several samples). Differences were considered statistically significant at $P < 0.05$ (at statistical power > 80%). Student's

t-test or its nonparametric analog was used to compare two samples of quantitative data: Mann-Whitney *U*-test. When comparing quantitative data from several samples simultaneously, parametric single- and multivariate analysis of variance (and their non-parametric analogs) were used: Kruskal-Wallis analysis (ANOVA) and the Jonckheere-Terpstra criterion (when testing the shift hypothesis against ordering alternatives). Qualitative characteristics were compared using Fisher's chi-square test. The effect of one variable on the other was assessed by regression analysis.

2.7. Ethical review

The study was approved by the Local Ethical Committee of the "Russian Medical Academy of Continuing Professional Education" Ministry of Health of the Russian Federation, protocol No. 9 of November 13, 2018. The results of the review of the study protocol were found to be satisfactory, and the results of the study were recommended for publication.

3. Results

3.1. Participants of the study

Of the 75 patients included in the study, there were 47 males (62.7%) and 28 females (37.3%), aged between 19 and 84 years. The mean duration of hospitalization was 24.6 ± 8.2 days.

All patients included in the study had progressive stage of psoriasis. Patients received methotrexate in the form of a solution (trade name: Methotrexate-Ebeve; manufacturer: EbevePharma, Austria) administered intramuscularly once a week. The therapeutic dose of methotrexate was selected in accordance with the clinical guidelines of the 2019 Russian Society of Dermatovenerologists and Cosmetologists. The mean dose of methotrexate was 14.05 ± 3.49 mg. The average number of injections was 3.15 ± 1.00. All patients received methotrexate in combination with folic acid, which was taken at a dosage of 5 mg 12–24 hours after each methotrexate injection.

3.2. Gastrointestinal adverse drug reactions observed in patients with psoriasis during methotrexate therapy

Manifestations of toxic effects of methotrexate developed in 38 patients (50.67%). All patients who experienced one or another gastrointestinal ADRs were questioned using the GSRS Digestive Symptom Severity Questionnaire, which assesses the quality of life of patients with regard to complaints of various GI disorders. The most common intensity of symptoms was rated by patients as 1 (minor discomfort) to 4 points (relatively severe but tolerable discomfort). The maximum total score on the scale was 27, the minimum was 3. The mean value was 7.97 ± 9.18 points.

Analysis of gastrointestinal ADRs was carried out for each category of symptoms individually. Stomatitis was detected in 3 patients (4%). In two manifestations, one patient developed on the fourth to fifth day after the first methotrexate injection, and in one patient on the third day after the second injection of the drug. The average duration of symptoms was 10 days. Severity of manifestations according to the CTCAE scale in all patients was 1 point (mild toxicity, slightly or moderately expressed symptoms not requiring therapeutic intervention), as stomatitis was manifested by ulcers in the oral cavity and moderate soreness, while the patient's eating was not disturbed.

Diarrhea was noted in 17 patients (22.67%). Symptoms appeared, as a rule, a few hours after methotrexate injection, persisted for 2–3 days, and occurred again at the next injection. The severity of manifestations on the CTCAE scale in the majority of patients was 1 point, as the times of diarrhea were not more than 4 times

a day. Three patients had diarrhea up to 6 times a day, and the severity of manifestations on the CTCAE scale corresponded to 2 points.

Complaints of nausea were presented by 29 patients (38.67%). Symptoms occurred an average of 4.5 hours after methotrexate injection and lasted an average of about three days (2.96 days), recurring after repeated injections. Patients rated the severity of nausea in scores using a visual analog scale: The mean score was 6 [6,8]. The maximum score was 10 and the minimum score was 3. In five patients, nausea disrupted the usual eating pattern and was therefore rated as 2 on the CTCAE scale. In the other 24 patients, nausea only led to a decrease in appetite and corresponded to a score of 1. Two patients experienced vomiting while taking methotrexate that did not require special rehydration therapy (CTCAE score = 1).

Two patients noted the occurrence of heartburn 7–12 hours after methotrexate injection, lasting for 4–6 days. Heartburn manifested as an unpleasant burning or stinging sensation in the chest area.

12 patients (16%) complained of abdominal pain of different nature: tingling, aching, pulling. One patient experienced pain in the right hypochondrium. They developed within 1–2 days after injection and were short-lived: the duration of no more than 24 hours. The severity of abdominal pain rated by patients on a visual analog scale was 1.05 ± 1.54 points on average. On the CTCAE scale, the pain was rated as 1 point, as it was moderate.

In addition to the adverse drug reactions described, 33 patients (44%) described other symptoms that were grouped under the category of dyspepsia. Patients complained of abdominal rumbling, a feeling of air in the abdomen, belching, which patients described as the release of air out of the stomach through the mouth, and flatulence. Patients complained of discomfort in the upper abdomen and the stomach area, a feeling of sour or bitter liquid from the stomach flowing down the throat, and a feeling of unpleasant emptiness in the stomach. Some patients complained of a reduced, compared to normal, ability to empty the bowels; others complained of alternating liquid and too hard stools, with liquid stools predominating, and a sudden need to empty the bowels. Dyspeptic symptoms occurred within the first 24 hours after methotrexate injection and persisted for several days. After subsequent injections, the symptoms recurred. GSRS assessment of the gastrointestinal tract also revealed the predominance of dyspeptic syndrome over abdominal pain syndrome, reflux syndrome, and diarrhea syndrome.

The structure and severity of ADRs in the gastrointestinal tract are presented in **Table 1**.

Table 1. Structure and severity of gastrointestinal toxicity of methotrexate during psoriasis therapy

Adverse drug reactions	Number of patients, abs. (relative %)	Method for assessing severity	Degree of severity/ average score*
Dyspepsia	33 (44%)	CTCAE	1–2
Nausea	29 (38, 67%)	CTCAE	1–2
		VAS	2.78 ± 3.55
Vomiting	2 (2, 67%)	CTCAE	1
Diarrhea	17 (22, 67%)	CTCAE	1–2
Abdominal pain	12 (16%)	CTCAE	1
		VAS	1.05 ± 1.54
Pain in the right hypochondrium	4 (5, 33%)	CTCAE	1
Stomatitis	3 (4%)	CTCAE	1
Heartburn	2 (2, 67%)	–	–
All adverse drug reactions in the gastrointestinal tract	38 (50, 67%)	GSRS	7.97 ± 9.18

*The severity was assessed in degrees of severity according to the CTCAE scale. The severity was assessed in points according to the GSRS and visual analogue scales (VAS).

3.3. Frequency analysis of allele and genotype distribution at polymorphic marker *3435C>T* of *ABCB1* gene (rs1045642) in psoriasis patients treated with methotrexate

The distribution of allelic variants of single-nucleotide polymorphism *3435C>T* of the *ABCB1* gene (rs1045642) and their distribution conformity to the Hardy-Weinberg law in patients with psoriasis treated with methotrexate were analyzed.

According to the results of *ABCB1* genotyping by polymorph marker rs1045642 in 75 patients with psoriasis treated with methotrexate, the following results were obtained (**Table 2**).

(1) The number of patients who are carriers of the all-allelic variant of the wild-type genotype *ABCB1* rs1045642 (CC genotype) amounted to 14 patients (18.67%);

(2) The number of patients who are heterozygous carriers of the C3435T polymorphism of the *ABCB1* gene (CT genotype), amounted to 34 patients (45.33%);

(3) The number of patients who were homozygous carriers of C3435T polymorphism of *ABCB1* gene (TT genotype) amounted to 27 patients (36.00%).

The distribution of genotypes followed the Hardy-Weinberg law (Fisher's exact test $\chi^2 = 0.32$; $P = 0.572$).

Table 2. The analysis of differences in the incidence of gastrointestinal toxicity of methotrexate in *ABCB1* rs1045642 CC, CT, and TT patients

Indicator	CC (0), n = 14	CT (1), n = 34	TT (2), n = 27	Reliability differences (*P* value)
Adverse drug reactions in the gastrointestinal tract	2 (14.3%)	18 (52.9%)	18 (66.7%)	0.006
Stomatitis	1 (7.1%)	2 (5.9%)	0 (0.0%)	0.407
Diarrhea	1 (7.1%)	8 (23.5%)	8 (29.6%)	0.261
Nausea	2 (14.3%)	13 (38.2%)	14 (51.9%)	0.064
Vomiting	0 (0.0%)	2 (5.9%)	0 (0.0%)	0.290
Heartburn	0 (0.0%)	0 (0.0%)	2 (7.4%)	0.161
Dyspepsia	1 (7.1%)	17 (50.0%)	15 (55.6%)	0.008
Abdominal pain	1 (7.1%)	11 (32.4%)	14 (51.9%)	0.016
Pain in the right hypochondrium	0 (0.0%)	2 (5.9%)	2 (7.4%)	0.595

3.4. Analysis of the possible association between *ABCB1* rs1045642 polymorphism and the occurrence of adverse drug reactions in the gastrointestinal tract

Results of the analysis of differences in the incidence of adverse drug reactions in the gastrointestinal tract between group of patients with genotypes rs1045642 CC, CT, and TT are presented in **Table 2**.

The comparison between the group of patients with the rs1045642 CC homozygote and the group of patients with the remaining genotypes is presented in **Table 3**. To calculate the statistical significance of the presence of minor allelic variant T rs1045642 with the incidence of ADRs when using methotrexate for psoriasis treatment, the number of T allele in the patient's genotype was denoted by 0, 1 or 2, where 0 is the CC genotype, 1 is the CT genotype, 2 is the TT genotype. The analysis of the frequency of gastrointestinal disorders revealed significant differences between groups of patients with CC, CT, and TT genotypes rs1045642: methotrexate toxicity was more frequent in carriers of CT and TT genotypes ($P = 0.006$).

However, when comparing groups with the CC genotype and other genotypes (CT and TT), the strength of the association remained unchanged ($P = 0.006$). The result of binomial regression showed a statistically significant effect of *ABCB1* gene rs1045642 polymorphism on the incidence of ADR development in the

gastrointestinal tract: estimation –2.16, OR = 8.64, 95% CI OR: 1.78–42,01, *P* = 0.008.

Table 3. The analysis of differences in the incidence of gastrointestinal toxicity of methotrexate in *ABCB1* rs1045642 CC and CT + TT patients

Indicator	CC (n = 14)	CT + TT (n = 61)	Reliability of differences (*P* value)
Adverse drug reactions in the gastrointestinal tract	2 (14.3%)	136 (59.0%)	0.006
Stomatitis	1 (7.1%)	2 (3.3%)	0.467
Diarrhea	1 (7.1%)	16 (26.2%)	0.168
CTCAE-1 diarrhea	0 (0.0%)	14 (23.7%)	0.059
Nausea	2 (14.3%)	27 (44.3%)	0.053
Vomiting	0 (0.0%)	2 (3.3%)	1.100
Heartburn	0 (0.0%)	2 (3.3%)	1.100
Dyspepsia	1 (7.1%)	32 (52.5%)	0.005
Abdominal pain	1 (7.1%)	25 (41.0%)	0.026
Pain in the right hypochondrium	0 (0.0%)	4 (6.6%)	1.000

When analyzing the frequency of occurrence of individual types of ADRs, statistically significant patterns were also identified. Dyspepsia was characteristic for carriers of genotypes CT and TT (*P* = 0.008), nausea was also detected predominantly in carriers of these genotypes (*P* = 0.05).

However, when comparing patients homozygous for the C allele rs1045642, with others, the relationship between carriage of CT and TT genotypes and the occurrence of dyspeptic events became more pronounced (*P* = 0.005), as well as the relationship between carriage of CT and TT genotypes and the occurrence of nausea (*P* = 0.053).

The result of binomial regression demonstrated the presence of statistically significant influence of *ABCB1* gene rs1045642 polymorphism on the frequency of nausea: estimate -1.56, OR = 4.76, 95% CI OR: 0.98–23.13, *P* = 0.05, as well as the development of dyspepsia: estimation -2.66, OR = 14.34, 95% CI OR: 1.76–116.57, *P* = 0.013.

No significant association was found between groups of patients with different genotypes (*P* = 0.261). However, when patients carrying the T allele were combined into the group with the CT and TT genotypes, the reliability of the differences between the frequency of diarrhea of the 1st degree of severity and carriage of the T allele was at a probability level of 0.06 (*P* = 0.059).

Abdominal pain was more frequently reported by carriers of the allele T: CC: 1 (7.1%), CT: 11 (32.4%), TT: 14 (51.9%), *P* = 0.016. Combining carriers of the T allele into one group confirmed the established pattern: CC: 1 (7.1%), CT + TT: 25 (41.0%), *P* = 0.026. The result of binomial regression construction demonstrated a statistically significant effect of alleles of rs1045642 polymorphism of *ABCB1* gene on the occurrence of pain in the abdomen: estimation –2.2, OR = 9.03, 95% CI OR: 1.109–73.5, *P* = 0.04.

Analysis of symptoms such as stomatitis, vomiting, heartburn, and right hypochondrium pain also revealed no statistically significant differences in patients with different genotypes for *ABCB1* rs1045642 polymorphic marker.

4. Discussion

The high efficacy of methotrexate and the possibility of long-term therapy allow methotrexate to be considered as one of the drugs of choice in the treatment of severe psoriasis. However, the development of adverse drug reactions significantly reduces patients' quality of life and adherence to treatment. In this regard, it is relevant to predict the safety of methotrexate therapy, including the use of pharmacogenetic studies.

Results of the study of the influence of the presence of polymorphisms of the methotrexate transporter protein gene demonstrate that carriage of the T allele rs1045642 in psoriasis patients on methotrexate therapy is associated with a higher incidence of adverse drug reactions in the gastrointestinal tract: carriers of the mutant T allele (genotypes CT and TT) more often reported toxic effects of methotrexate.

A possible explanation for this pattern may be the accumulation of methotrexate in the body due to its delayed excretion in carriers of the minor T allele *ABCB1* (rs1045642). Presumably, this is due to the effect of gene polymorphism on the functioning of the transporter protein encoded by the gene. The *ABCB1* gene encodes P-glycoprotein: it is likely that carriers of the minor T allele have reduced activity of this protein, whose substrate is methotrexate. This can cause methotrexate to be eliminated from the body more slowly. As a consequence, methotrexate reaches target receptors in greater quantities, its cytotoxic effect is realized, which manifests itself as an increased risk of adverse reactions.

5. Conclusion

The results of the pharmacogenetic study *3435C>T* polymorphism of the *ABCB1* gene may be taken into account to improve the safety of therapy for psoriasis patients receiving methotrexate, as carriers of the minor T allele have an increased risk of adverse drug reactions, possibly related to genetically determined slowing of methotrexate excretion. The data obtained may form the basis for an algorithm for predicting the safety of methotrexate therapy in patients with moderate to severe psoriasis. However, further research is needed to increase the accuracy and reliability of the prediction to evaluate the different manifestations of methotrexate toxicity and to identify other biomarkers for the safety of methotrexate therapy.

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Disclosure statement

The authors declare no conflict of interest.

Author contributions

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All authors approved of the final version of the article and took responsibility for the integrity of all parts of the

article.

References

[1] Psoriasis. Clinical recommendations, 2020, Approved by the All-Russian Public Organization "Russian Society of Dermatovenerologists and Cosmetologists" January 15, 2020, Approved by the Scientific and Practical Council of the Ministry of Health of the Russian Federation.

[2] Parisi R, Iskandar IYK, Kontopantelis E, 2020, National, Regional, and Worldwide Epidemiology of Psoriasis: Systematic Analysis and Modelling Study. BMJ, 2020(369): 1–15. http://doi.org/10.1136/bmj.m1590

[3] Lebwohl M, 2005, Clinician's Paradigm in the Treatment of Psoriasis. J Am Acad Dermatol, 53(1): S59–69. http://doi.org/10.1016/j.jaad.2005.04.031

[4] Chikin VV, Znamenskaya LF, Minaeva AA, 2014, Pathogenic Aspects of Treatment of Psoriatic Patients. Vestnik Dermatologii Ivenerologii, 2014(5): 86–90.

[5] West J, Ogston S, Foerster J, 2016, Safety and Efficacy of Methotrexate in Psoriasis: A Meta-Analysis of Published Trials. PLoS One, 11(5): e0153740. http://doi.org/10.1371/journal.pone.0153740

[6] Yazici Y, Sokka T, Kautiainen H, et al., 2005, Long Term Safety of Methotrexate in Routine Clinical Care: Discontinuation is Unusual and Rarely the Result of Laboratory Abnormalities. Ann Rheum Dis, 64(2): 207–211. http://doi.org/10.1136/ard.2004.023408

[7] Bedoui Y, Guillot X, Sélambarom J, et al., 2019, Methotrexate an Old Drug with New Tricks. Int J Mol Sci, 20(20): 5023. http://doi.org/10.3390/ijms20205023

[8] Ray-Jones H, Eyre S, Barton A, et al., 2016, One SNP at a Time: Moving Beyond GWAS in Psoriasis. J Invest Dermatol, 136(3): 567–573. http://doi.org/10.1016/j.jid.2015.11.025

[9] Sutherland A, Power RJ, Rahman P, et al., 2016, Pharmacogenetics and Pharmacogenomics in Psoriasis Treatment: Current Challenges and Future Prospects. Expert Opin Drug Metab Toxicol, 12(8): 923–935. http://doi.org/10.1080/17425255.2016.1194394

[10] Sychjov DA, 2011, Recommendations for the Use of Pharmacogenetic Testing in Clinical. Kachestvennaya Klinicheskaya Praktika, 2011(1): 3–10.

[11] Lima A, Bernardes M, Azevedo R, et al., 2014, *SLC19A1*, *SLC46A1* and *SLCO1B1* Polymorphisms as Predictors of Methotrexate-Related Toxicity in Portuguese Rheumatoid Arthritis Patients. Toxicological Sciences, 142(1): 196–209. http://doi.org/10.1093/toxsci/kfu162

[12] Whetstine JR, Gifford AJ, Witt T, et al., 2001, Single Nucleotide Polymorphisms in the Human Reduced Folate Carrier: Characterization of a High-Frequency G/A Variant at Position 80 and Transport Properties of the His(27) and Arg(27) Carriers. Clin Cancer Res, 7(11): 3416–3422.

[13] Marzolini C, Paus E, Buclin T, et al., 2004, Polymorphisms in Human *MDR1* (P-Glycoprotein): recent Advances and Clinical Relevance. Clin Pharmacol Ther, 75(1): 13–33. http://doi.org/10.1016/j.clpt.2003.09.012

[14] Hallas J, Harvald B, Gram LF, et al., 1990, Drug Related Hospital Admissions: The Role of Definitions and Intensity of Data Collection, and the Possibility of Prevention. J Intern Med, 228(2): 83–90. http://doi.org/10.1111/j.1365-2796.1990.tb00199.x

[15] Stockwell DC, Slonim AD, 2006, Quality and Safety in the Intensive Care Unit. J Intensive Care Med, 21(4): 199–210. http://doi.org/10.1177/0885066606287079

[16] Cvetov VM, 2007, Monitoring of Adverse Reactions of Drugs in an Outpatient Clinic at the Present Stage, dissertation, Chelyabinsk, 130.

[17] US Department of Health and Human Services, National Institutes of Health, National Cancer Institute, 2017,

Common Terminology Criteria for Adverse Events (CTCAE) Version 5, viewed January 6, 2022, https://ctep.cancer.gov/protocoldevelopment/electronic_applications/docs/CTCAE_v5_Quick_Reference_8.5x11.pdf

[18] Svedlund J, Sjödin I, Dotevall G, 1988, GSRS-A Clinical Rating Scale for Gastrointestinal Symptoms in Patients with Irritable Bowel Syndrome and Peptic Ulcer Disease. Dig Dis Sci, 33(2): 129–134. http://doi.org/10.1007/BF01535722

[19] Busto U, Naranjo CA, Sellers EM, 1982, Comparison of Two Recently Published Algorithms for Assessing the Probability of Adverse Drug Reactions. Br J Clin Pharmacol, 13(2): 223–227. http://doi.org/10.1111/j.1365-2125.1982.tb01361.x