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Side effects of methotrexate therapy in patients with rheumatoid arthritis

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Abstract

Introduction: Methotrexate is widely used as the most common disease-modifying anti-rheumatoid drug (DMARD) and is known as the first line treatment for rheumatoid arthritis (RA).
Objectives: To assess the side effects of methotrexate in Iranian patients with RA and to compare them with the known side effects from previous studies.
Patients and Methods: We conducted a cross-sectional study of 300 patients who fulfilled the EULAR 2010 criteria of RA. The following data were recruited from patients' profiles; age, body mass index (BMI), duration of treatment with methotrexate, initiating dose, maximum dose and current dose of methotrexate, history of fatty liver disease or hepatitis B and concomitant use of sulfasalazine, leflunomide or hydroxychloroquine.
Results: In 149 out of 300 patients (49.66%), Methotrexate therapy was stopped or tapered due to side effects including nausea (23%), flu-like symptoms (8%), hepatotoxicity (12%) and hair loss (6%). The patients with hepatotoxicity had a higher duration of treatment with methotrexate (10.35 compared with 5.83; $P<0.001$) and also the higher initiating dose of methotrexate (12.91 compared with 12.17; $P=0.010$). All of the RASS (rheumatoid arthritis severity scale) indexes including disease activity, functional impairment, and physical damage are related to the presence of hepatotoxicity ($P<0.001$).
Conclusion: Methotrexate is an excellent and effective agent for the treatment of RA and its potential side effects during the treatment are dependent on the methotrexate dosage, the level of anti-citrullinated protein antibody (ACPA) and anti-MCV antibodies and concomitant use of other drugs such as leflunomide.

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Introduction

Rheumatoid arthritis (RA) is a different form of non-joint manifestations including fatigue, subcutaneous nodules, lung involvement, pericarditis, peripheral neuropathy, vasculitis, hematologic abnormalities, Sjogren's syndrome and lymphoma (1,2). It affects 0.5%-1% of adults worldwide (3). The history of RA is affected by some factors including gender, age of onset, genotype and phenotype (1,2).

Methotrexate is widely used as the most common disease-modifying anti-rheumatoid drug (DMARD) and is known as the first line treatment for RA (4). Methotrexate is an antifolate metabolite that inhibits dihydrofolate reductase, DNA and RNA synthesis. It also reduces immunogenicity and inflammatory responses (5). The optimal dosage of methotrexate in RA is 15 mg/wk orally, escalating with 5 mg/ methotrexate to 25-30 mg/wk. Despite adjusting the optimal dosage, several studies indicated the broad toxicity spectrum of methotrexate (2). The well-known side effects of methotrexate are nausea, headache, dizziness, rash, cough, diarrhea, pancytopenia, hepatotoxicity, pulmonary fibrosis, infection, neoplasia, infertility, leukopenia, alopecia, gastric irritation, stomatitis, teratogenicity and pneumonitis (6).

One of the greatest concerns about methotrexate is the subsequent hepatotoxicity. It can induce a range of histologic changes such as steatosis, stellate

Key point

In each patient with RA and its potential side effects, methotrexate can be considered as an excellent and effective agent for the treatment.

(Ito) cell hypertrophy, mononucleosis, and hepatic fibrosis (7) and leads to asymptomatic elevation of hepatic enzymes (8) which might induce mild fibrosis and cirrhosis (9).

Objectives

Herein, we aimed to assess the side effects of methotrexate in Iranian patients with RA and to compare them with the known side effects from previous studies.

Patients and Methods

Study design

In this cross-sectional study, we evaluated 300 patients who fulfilled the EULAR 2010 criteria and referred to the outpatient clinic of RA' disease, Rheumatology research center, Beheshti hospital, Qom, Iran, between 2016 and 2019. All RA patients with a history of methotrexate prescription were included in this study.

Demographic data

The following data were recruited from patients' profiles: age, body mass index (BMI), duration of treatment with Methotrexate, initiating dose, maximum dose and current dose of methotrexate, history of fatty liver disease or hepatitis B and concomitant use of sulfasalazine, leflunomide or hydroxychloroquine.

Laboratory data

Blood sampling was prepared at the time of the study. Laboratory kits were used to measure ACPA (anti-citrullinated protein antibody) and anti-MCV antibodies precisely. Up to 300 IU/mL and 1000 IU/mL could be detected by these kits. The lipid profile, fasting blood glucose, hepatic enzymes, HbsAg, and HCV Ab were recorded from the patients' documents that were measured up to one year apart from the study time. All of the measurements were conducted in the laboratory of Beheshti hospital, Qom, Iran.

Side effects

Records were maintained for patients treated with methotrexate, detailing any hepatic or other side-effects leading to drug discontinuation or dose reduction such as hepatotoxicity, nausea, flu-like symptoms, and hair loss. Increased liver function tests in this study were defined as more than 2 times higher from normal ranges for aspartate-aminotransferase (AST) and alanine aminotransferase (ALT).

The severity of RA in every patient was measured by RA severity scale (RASS). The RASS includes three VAS showing disease activity, functional impairment and physical damage. We describe scale end-points as follows; 0; no disease activity (functional impairment or physical damage): 100; worst RA disease activity (functional impairment or physical damage). Disease activity measures joint counts, erythrocyte sedimentation rate (ESR), and assessment of disease activity experienced by the patient. For functional impairment, we considered our assessment of the patient's capacity for self-care, household tasks, social activity, work, and physical dexterity and the level of functional impairment. The physical damage is defined as the X-ray criteria (such as erosions) and anatomic damages. Leukopenia is defined as moderate (3.0-4.0 $\times$ 10^6/μL) or severe (less than 3.0 $\times$ 10^6/μL).

Ethical issues

The research followed the tenets of the Declaration of Helsinki. The institutional ethical committee at Qom University of Medical Sciences approved all study protocols (IR.MUQ.REC.1398.076). Accordingly, written informed consent taken from all participants before any intervention. This study was extracted from M.D thesis of Mostafa Vahedian at this university.

Statistical analysis

We used SPSS software (IBM SPSS Statistics 24; SPSS Incorporation, Chicago, Illinois, USA) for data analysis. The variables were reported using descriptive and inferential statistics. Continuous, ordinal, and categorical variables were reported as mean and standard deviation. For inferential analysis, binary logistic regression was used, where the independent variables were initially included in groups: (A) demographic parameters (age, gender and BMI); (B) clinical conditions (diabetes, fatty liver) (C); medication and dosage of drugs (hydroxychloroquine, sulfasalazine, leflunomide, starting, current and maximum dose of methotrexate). The dependent variables were (A) clinical condition (disease duration, RASS-Disease Activity, RASS-Functional Impairment, RASS-Physical Damage, RASS-Total); (B) biochemical parameters (anti-MCV antibodies, anti-cyclic citrullinated peptides). Univariate association of every variable and hepatotoxicity was evaluated through linear regression analysis. The association of hepatotoxicity and its predictors was evaluated by multivariable linear regression model, adjusted for age, gender, BMI, major symptoms and signs and laboratory findings. The level of significance was defined as $P \leq 0.05$.

Results

A sample of 300 individuals was obtained. The mean age of patients was 49.1 ± 12.7 (years) and the mean BMI was 27.61 ± 5.45 (kg/m^2). There were 65 (21.7%) male patients and 234 (78.3%) female. Methotrexate therapy had side effects including nausea (23%), flu-like symptoms (8%), hepatotoxicity (12%) and hair loss (6%). Table 1 describes demographic data of RA patients with (15 males and 21 females) and without (51 males and 213 females) hepatotoxicity. The mean age at the first visit was 51.47 ± 13.19 and 48.79 ± 12.64 years in patients with and without hepatotoxicity, respectively. BMI was 27.46 ± 5.19 kg/m^2 in patients with hepatotoxicity and 27.63 ± 5.5 kg/m^2 in patients without hepatotoxicity.

Table 1. Demographic data of rheumatoid arthritis patients with and without hepatotoxicity

Characteristics		RA patients		*P* value
		With hepatotoxicity (n=36)	Without hepatotoxicity (n=264)	
Gender	Male	15	51	0.002
	Female	21	213	
Age (y)		51.47	48.79	0.692
BMI (kg/m^2)		27.46	27.63	0.559
Duration of treatment with methotrexate		10.35	5.83	<0.001
Initiating dose of methotrexate		12.91	12.17	0.010
Maximum dose of methotrexate		21.74	18.73	0.193
Concomitant use of hydroxychloroquine		34	230	0.202
Concomitant use of sulfasalazine		10	75	0.927
Concomitant use of leflunomide		12	46	0.024
History of diabetes		5	24	0.365
History of fatty liver disease		7	33	0.254
History of Hepatitis B		1	0	0.120

In addition, the mean initial dose of methotrexate was 12.91 ± 3.4 mg/wk and 12.17 ± 3.6 mg/wk in patients with and without hepatotoxicity, respectively ($P < 0.249$). The mean duration of treatment with methotrexate was 10.35 ± 8.27 years in and 5.83 ± 4.58 years in patients with and without hepatotoxicity, respectively. In this study, hydroxychloroquine was prescribed in 34 patients with hepatotoxicity and 230 patients without hepatotoxicity in different doses according to the severity of the RA disease. Sulfasalazine was used in 10 patients with hepatotoxicity and 75 patients without hepatotoxicity. Leflunomide was prescribed in 12 patients with hepatotoxicity and 46 patients without hepatotoxicity. Five patients with hepatotoxicity and 24 patients without hepatotoxicity had a previous history of diabetes mellitus. Seven patients with hepatotoxicity and 33 patients without hepatotoxicity had a previous history of fatty liver disease and one patient with hepatotoxicity had a history of hepatitis B.

Side effects of methotrexate leading to drug discontinuation or dose change

In 149 out of 300 patients (49.66%), methotrexate therapy was stopped or tapered due to side effects. The side effects which resulted in methotrexate discontinuation are shown in Table 2.

Hepatotoxicity: In 36 patients with the initial dose of 12.92 mg/wk, duration of treatment of 10.35 years and maximum dose (mg/wk) of 21.74 mg/wk developed hepatotoxicity, then the dosage was tapered to 9.79 mg/wk.

Nausea: In 71 patients with the initial dose of 12.21 mg/wk and duration of treatment of 6.31 years and maximum dose of 20.21 mg/wk developed nausea. The dosage was finally tapered to 11.83 mg/wk.

Flu-like symptoms: Around 24 patients with the initial dose of 12.50 mg/wk and duration of treatment of 7.98 years and maximum dose of 21.67 mg/wk developed flu-like symptoms, thereby, the dosage was tapered to 11.56 mg/wk.

Hair loss: About18 patients with the initial dose of 10.28 mg/wk and duration of treatment of 6.72 years and maximum dose of 19.58 mg/wk developed hair loss. The dosage was tapered to 9.58 mg/wk.

Rheumatoid arthritis severity scale in RA patients with and without hepatotoxicity

The range of disease activity was 10-80 and 4-78 in patients with and without hepatotoxicity, respectively. The mean score of disease activity was 41.22 ± 18.23 and 20.81 ± 12.65 in patients with and without hepatotoxicity, respectively ($P < 0.001$). Table 3 describes the RASS in RA patients with and without hepatotoxicity.

The functional impairment range was 20-98 and 3-96 in patients with and without hepatotoxicity with the average of 67.14 ± 22.34 and 29.47 ± 19.33, respectively ($P < 0.001$). The physical damage ranged 4-85 and 1-75 in patients with and without hepatotoxicity with the mean score of

Table 2. Adverse effects of methotrexate leading to drug discontinuation or dose reduction in patients with rheumatoid arthritis

Adverse effects	Number of patients	Initial dose (mg/w)	Duration of treatment (y)	Maximum dose (mg/w)	Final dose (mg/w)
Hepatotoxicity	36	12.92	10.35	21.74	9.79
Nausea	71	12.21	6.31	20.21	11.83
Flu like symptoms	24	12.50	7.98	21.67	11.56
Hair loss	18	10.28	6.72	19.58	9.58

Table 3. Rheumatoid arthritis severity scale in rheumatoid arthritis patients with and without hepatotoxicity

RASS	RA with hepatotoxicity (n=36)			RA without hepatotoxicity (n=264)			*P* value
	Range	Mean	SD	Range	Mean	SD	
Disease activity	10-80	41.22	18.23	4-78	20.81	12.65	<0.001
Functional impairment	20-98	67.14	22.34	3-96	29.47	19.33	<0.001
Physical damage	4-85	37.22	19.61	1-75	17.61	14.10	<0.001

RASS, rheumatoid arthritis severity scale; RA, rheumatoid arthritis.
*Maximum point of each row is 100 points.

37.22 ± 19.61 and 17.61 ± 14.10, respectively ($P < 0.001$).

The association between ACPA, rheumatoid factor and anti-MCV antibodies and side effects of methotrexate in RA patients

We investigated the relationship between ACPA, anti-MCV and rheumatoid factor antibodies and side effects of methotrexate in RA patients such as hepatotoxicity, nausea, flu-like symptoms, and hair loss. The titer of anti-MCV was 614.44 and 73.69 U/mL in the patients with and without hepatotoxicity, respectively ($P < 0.001$). The titer of ACPA was 187.54 and 84.33 U/mL in the patients with and without hepatotoxicity, respectively ($P < 0.001$).

The titer of anti-MCV in patients with nausea was 112.02 U/mL in the positive group and 147.14 U/mL in the negative group ($P = 0.310$). The titer of ACPA in patients with nausea was 100.32 U/mL in the positive group and 95.65 U/mL in the negative group ($P = 0.761$).

The titer of anti-MCV in patients with flu-like symptoms was 174.594 U/mL in the positive and 135.67 U/mL in the negative group ($P = 0.473$). The titer of ACPA in patients with hepatotoxicity was 121.45 U/mL in the positive group and 94.60 U/mL in the negative group ($P = 0.264$).

The titer of anti-MCV in patients with hepatotoxicity was 275.75 U/mL in positive and 130.02 U/mL in the negative group ($P = 0.089$). The titer of ACPA in patients with hepatotoxicity was 124.09 U/mL in the positive group and 95.01 U/mL in the negative group ($P = 0.289$). Table 4 describes the association between ACPA and anti-MCV antibodies and side effects of methotrexate in RA patients.

Univariate and multivariate analysis for hepatotoxicity

The values of gender and anti-MCV Ab were also initially associated with hepatotoxicity through interpretations of only the p-value of the univariate analysis.

However, including these variables mentioned in the multivariate model it was detected that gender values OR = 6.5 (95% CI: 1.6, 26.6) and RASS, functional impairment OR = 1.072 (95% CI: 1.04, 1.1) and anti-MCV Ab OR = 1.006 (95% CI: 1.004, 1.008) were robust predictors of hepatotoxicity. More details are given in Table 5.

Discussion

RA is a chronic autoimmune disease affected by genetic and environmental factors such as smoking, diet, obesity, infections, and microbiota (1,2). Many organs such as skin, lung (3), heart and vessels and especially joints involved by this disease (5). The main current treatment strategy for RA is disease-modifying anti-rheumatic drugs (DMARDs) especially methotrexate (2). Methotrexate makes cells unable to divide to produce proteins (6) and decreases the pain, swelling and the rate of disease progression (8). However, many patients discontinue the use of methotrexate during the treatment period because of side effects (9). They include hepatotoxicity, thrombocytopenia, pancytopenia, renal abnormalities, anemia, mucocutaneous problems (10), nausea (11) and hair loss (12). The risk of side effects may differ within the different dosages of methotrexate (13).

In this research, we analyzed the following data in patients with and without side effects of methotrexate, which caused drug discontinuation or dose reduction: age, BMI, duration of treatment with methotrexate, Initiating dose of methotrexate, maximum dose of methotrexate, concomitant use of hydroxychloroquine, sulfasalazine, and leflunomide, history of diabetes, fatty liver disease, and

Table 4. The association between ACPA and anti-MCV antibodies and side effects of methotrexate in patients with rheumatoid arthritis

Side effects		Anti-MCV	*P* value	ACPA	*P* value
Hepatotoxicity	Positive	614.44	<0.001	187.54	<0.001
	Negative	73.69		84.33	
Nausea	Positive	112.02	0.310	100.32	0.761
	Negative	147.14		95.65	
Flu like symptoms	Positive	174.59	0.473	121.45	0.264
	Negative	135.67		94.60	
Hair loss	Positive	275.75	0.089	124.09	0.289
	Negative	130.02		95.01	

ACPA, Anti- citrullinated protein antibody

Table 5. Univariate and multivariate analysis for hepatotoxicity in patients with rheumatoid arthritis

Predictors	Univariate association		Multivariable model	
	β (95% CI)	*P*	β (95% CI)	*P*
Demographic				
Age (y)	1.00 (0.94-1.06)	0.990		
Gender	5.73 (1.01-32.5)	0.049	6.5 (1.6-26.6)	0.009
BMI (kg/m²)	0.95 (0.81-1.106)	0.518		
Medication				
Hydroxychloroquine	9.50 (0.44-205.15)	0.151		
Sulfasalazine	5.7 (0.4-9.48)	0.99		
leflunomide	0.69 (0.105-4.52)	0.70		
Comorbidity				
Diabetes	3.11 (0.31-30.76)	0.330		
Fatty Liver	1.86 (0.29-11.62)	0.506		
Methotrexate				
Starting dose	0.86 (0.68-1.10)	0.251		
Current dose	0.98 (0.86-1.11)	0.780		
Maximum dose	1.12 (0.88-1.42)	0.350		
Disease duration	1.07 (0.93-1.241)	0.306		
RASS, disease activity	1.23 (1.18-1.38)	0.391		
RASS, functional impairment	1.34 (1.5-1.17)	0.392	1.072 (1.04-1.1)	<0.001
RASS, physical damage	1.22 (1.07-1.38)	0.391		
Rheumatoid arthritis severity scale, total	5.22 (3.5-7.6)	0.391		
Anti-cyclic citrullinated peptides Ab	0.99 (0.9871.003)	0.178		
Anti-MCV Ab	1.07 (1.004-1.010)	<0.001	1.006 (1.004-1.008)	<0.001

hepatitis B. We found that the patients with hepatotoxicity had higher duration of treatment with methotrexate (10.35 compared with 5.83). Similar to our finding Shuji et al reported the positive correlation between the presence of hepatotoxicity and longer duration of treatment with methotrexate (1). Another finding was the association between initiating dose of methotrexate (12.91) and hepatotoxicity. In the present study, we analyzed the relationship between hepatotoxicity and consolidation of methotrexate and the relevant drugs such as hydroxychloroquine, sulfasalazine and leflunomide. The results proved the association of concomitant use of leflunomide and hepatotoxicity and that is compatible with the study by Jeffrey et al (14).

Another finding was the association between the initiating dose of methotrexate and hepatotoxicity (12.91 mg/wk compared with 12.17 mg/wk). In the present study, we analyzed the relationship between hepatotoxicity and concomitant use of methotrexate with hydroxychloroquine, sulfasalazine, and leflunomide. The results proved that the concurrent use of leflunomide predisposes RA patients to develop hepatotoxicity. Our analysis shows that the common side effects which made us to dose reduction or drug discontinuation among our 300 patients were nausea (23%), flu-like symptoms (8%), hepatotoxicity (12%) and hair loss (6%). our finding has some difference with previous studies in the view of most common complications; for example, the research conducted by Ediz et al among 31 patients found that mucositis, fever, infection, and purpura are the most common side effects (4). A possible explanation for this issue could be the different population.

Prevalence of hair loss between our patients (6%) in comparison with the study by Kara et al which shows the 6.6% hair loss in Caucasian race and 14% hair loss in African Americans is compatible and inconsistent due to different races (15). The most serious complication in our study is hepatotoxicity which was diagnosed in 12% of the patients. This finding has some compatibility with the previous finding; for example, the research conducted by Jeffrey et al found that the incidence of hepatic enzyme abnormalities was 14%-31% (14).

Our study shows that the common side effects which caused dose reduction or drug discontinuation among our 300 patients were nausea (23%), flu-like symptoms (8%), hepatotoxicity (12%) and hair loss (6%). The research conducted by Ediz et al among 31 patients found that mucositis, fever, infection, and purpura are the most common side effects (3).

The prevalence of hair loss between our patients was 6% which is compatible with the report by Kara et al which shows 6.6% of hair loss in the Caucasian race and 14% of hair loss in African Americans (15). Around 12 % of our study population was diagnosed with hepatotoxicity which is compatible with the report by the study by Jeffrey et al. They found that the incidence of hepatic enzyme abnormalities was 14-31% (14).

In this study, we analyzed the RASS in RA patients with and without hepatotoxicity. We found a significant relationship between all of the RASS indexes including

disease activity, functional impairment and physical damage with the presence of hepatotoxicity.

In recent years, some factors such as ALT, thrombocyte level, methylenetetrahydrofolate reductase (MTHFR) and MRP8/14 (S100A8/A9) polymorphisms and level of osteopontin are suggested to predict the methotrexate side events (16). Herein, for the first time, we found the association between ACPA and anti-MCV antibodies and side events of methotrexate. Between the four main side impacts which discontinued the treatment with methotrexate, only the hepatotoxicity had a significant relationship with ACPA and anti-MCV antibodies.

Conclusion

Methotrexate is an excellent and effective agent for the treatment of RA and its potential side effects during the treatment are dependent on the methotrexate dosage, the level of ACPA and anti-MCV antibodies and concomitant use of other drugs such as leflunomide.

Limitations of the study

This study has some limitations. Some data are self-reported and subjective. The endoscopic findings of the gastric mucosa were not used to analysis. No liver biopsies were performed to determine the severity of liver failure. As histology based studies were shown the effect of methotrexate on liver fibrosis. In addition, some studies show the discordance of liver enzymes changing and histological findings.

Authors' contribution

MM, SA, MB and MV were the principal investigators of the study. MB and MV were included in preparing the concept and design. MM, AG, JB, SA and SM revisited the manuscript and critically evaluated the intellectual contents. All authors participated in preparing the final draft of the manuscript, revised the manuscript and critically evaluated the intellectual contents. All authors have read and approved the content of the manuscript and confirmed the accuracy or integrity of any part of the work.

Conflicts of interest

The authors declare that they have no competing interests.

Ethical considerations

Ethical issues (including plagiarism, data fabrication, double publication) have been completely observed by the authors.

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