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Original Article

Effect of Methotrexate on Random Blood Sugar Level in Non-Diabetic Patients with Rheumatoid Arthritis

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Abstract

(**Background:** Rheumatoid arthritis (RA) is a chronic inflammatory disease commonly treated with methotrexate (MTX). While MTX effectively controls disease activity, its effect on glucose metabolism, particularly random blood sugar (RBS), remains unclear.
Objectives: To determine whether methotrexate therapy affects random blood sugar (RBS) levels in non-diabetic patients with rheumatoid arthritis.
Methods: Comparative cross-sectional study of 101 consecutively recruited non-diabetic adults with RA (HbA1c < 5.7%) attending a tertiary outpatient clinic (Rizgary Teaching Hospital, Erbil) from October 2024 to April 2025. Patients were classified by methotrexate (MTX) use and glucocorticoid (GC) exposure into four subgroups (MTX±GCs; non-MTX±GCs); MTX users were on a stable dose ≥ 6 months.
Results: Of 101 patients, 52 used MTX and 49 did not. Subgroup sizes were: MTX/GC− (23.76%), MTX/GC+ (27.72%), no MTX/GC− (24.75%), and GCs only (23.76%). RBS was significantly lower in MTX users than non-users (103.4 ± 12.8 vs 121.3 ± 16.6 mg/dL; mean difference −17.9 mg/dL, 95% CI −22.9 to −12.9; $p < 0.001$). By subgroup, mean RBS followed a clear gradient, lowest in MTX/GC−, intermediate in MTX/GC+, and highest in GC-only (overall $p < 0.001$). HbA1c did not differ meaningfully between MTX users and non-users (5.29% vs 5.34%; $p = 0.24$). In multivariable models, MTX exposure remained independently associated with lower RBS (Model A $\beta = -18.55$ mg/dL, 95% CI −25.01 to −12.08; $p < 0.001$; Model B $\beta = -14.80$, 95% CI −23.00 to −6.60; $p = 0.002$).
Conclusions: MTX use was independently associated with lower RBS in non-diabetic RA, including among GC users, while HbA1c remained unchanged. The phrasing “These results suggest a potential short-term glycemic benefit of MTX” is acceptable but a bit strong for cross-sectional data. These findings indicate an association between MTX use and lower RBS, suggesting a possible short-term glycemic benefit that warrants confirmation in longitudinal studies.

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1. Introduction

Rheumatoid arthritis (RA) is a systemic autoimmune disease characterized by chronic, symmetrical, and destructive polyarticular synovitis. Beyond joint inflammation, RA is associated with systemic consequences driven by pro-inflammatory cytokines such as tumor necrosis factor-α (TNF-α), interleukin-6 (IL-

6), and C-reactive protein (CRP), all of which have been implicated in insulin resistance (1, 2).

The interplay between chronic inflammation and glucose metabolism highlights an important clinical concern. impaired glucose regulation has been reported in up to 46% of RA patients within two years of follow-up (3). The prevalence of diabetes mellitus (DM) in RA is estimated at 15–19%, substantially higher than the 4–8% reported in the general middle-aged population (4, 5). Furthermore, RA patients carry a 1.5-fold higher risk of developing DM compared with matched controls (6). These findings underscore the metabolic burden of RA and the need to understand how standard therapies influence glucose homeostasis.

Methotrexate (MTX) is the first-line conventional synthetic disease-modifying antirheumatic drug (csDMARD) for RA. Acting as an antifolate and antimetabolite through inhibition of dihydrofolate reductase (DHFR), MTX is primarily known for its immunosuppressive and anti-inflammatory properties (7). However, accumulating evidence suggests that MTX may exert metabolic effects beyond inflammation control (8, 9).

Mechanistically, MTX inhibits AICAR transformylase, leading to intracellular accumulation of AICAR and subsequent activation of AMP-activated protein kinase (AMPK) (10-12). AMPK is a central regulator of cellular energy metabolism that decreases hepatic glucose production and enhances insulin sensitivity (13). This mechanism parallels the action of metformin (14, 15). Experimental studies further demonstrated that MTX enhances glucose uptake and lipid oxidation in skeletal muscle via AMPK-mediated pathways, providing a biologically plausible explanation for its potential glycemic benefits (16).

Despite these mechanistic insights, clinical evidence remains inconclusive. Some studies reported modest reductions in HbA1c following MTX therapy (17, 18), whereas others failed to show consistent effects (19). A systematic review and meta-analysis suggested that MTX may reduce the risk of developing type 2 DM among RA patients. However, little is known about its effect on random blood sugar (RBS), a marker of short-term glycemic fluctuations that may capture treatment effects not reflected by HbA1c (8).Therefore, this study aimed to examine whether methotrexate (MTX) use is independently associated with lower random blood sugar (RBS) levels in non-diabetic patients with rheumatoid arthritis (RA). Given the frequent co-administration of glucocorticoids (GCs), which are known to induce hyperglycemia, subgroup analyses were conducted according to GC exposure, dose, and duration. We hypothesized that MTX use would be associated with lower RBS, even among GC users, reflecting a potential glucose-modulating effect of MTX beyond its anti-inflammatory action

2 Patients and Methods

2.1 Study Design: A comparative cross-sectional observational study was conducted to evaluate the effect of methotrexate (MTX) on random blood sugar (RBS) levels in patients with rheumatoid arthritis (RA). Patients were stratified according to MTX use and glucocorticoid (GCs) exposure. A cross-sectional design was chosen for this study to examine the association between MTX use and RBS levels at a single point in time. However, it is important to note that this design precludes establishing temporal or causal relationships between MTX exposure and changes in RBS.

2.2 Setting and Duration: The study was conducted in the outpatient rheumatology clinic of Rizgary Teaching Hospital, Erbil, Kurdistan Region, Iraq. Recruitment was carried out consecutively between October 2024 and April 2025, until the target sample size (n = 101) was achieved.

2.3 Participants: A total of 101 RA patients, aged 18–70 years, were enrolled. Eligible patients were consecutively recruited from the outpatient rheumatology clinic to minimize selection bias and ensure representativeness of the clinic population. All fulfilled the 2010 ACR/EULAR classification criteria for RA(20). Participants were categorized into two main groups based on MTX use, and further stratified according to concomitant GCs therapy, yielding four treatment subgroups: MTX without GCs (n=24), MTX with GCs (n=28), No MTX/No GCs (n=25), GCs only (n=24).

2.4 Inclusion Criteria: Age 18–70 years, Diagnosis of RA according to the 2010 ACR/EULAR classification criteria (20), Patients with normal glycemic status (HbA1c <5.7%), For MTX group: stable MTX therapy for ≥6 months, For non-MTX group: not receiving MTX during the study period (patients on alternative DMARDs or biologics were included), Glucocorticoid exposure was defined as any use within the past 3 months, and patients were stratified into groups based on current daily use versus no use of glucocorticoids.

2.5 Exclusion Criteria: Patients with prediabetes (HbA1c 5.7–6.4%) or diabetes (HbA1c ≥6.5%). Current use of insulin or oral hypoglycemic agents, Pregnant or lactating women, Active infection, chronic liver disease, or other systemic autoimmune disorders.

2.6 Data Collection: Data were obtained through structured patient interviews, clinical records, and laboratory investigations. Collected variables included: Demographics: age, sex, BMI. Disease-related: duration of RA, DAS-28 score, RF, Anti-CCP. Treatment-related: MTX use, dose, duration; GCs use, dose, duration; other DMARDs and biologics. Laboratory data: RBS (mg/dL) and HbA1c (%). RBS was obtained from venous samples during routine clinic hours (08:00–14:00). While the timing of the last meal was recorded, no adjustments were made for the potential influence of postprandial glucose levels on the RBS values. HbA1c was measured by standard enzymatic assay, with values <5.7% considered normal, 5.7–6.4% as prediabetes, and ≥6.5% as diabetes, according to the American Diabetes Association (ADA) criteria (21). Comorbidities: hypertension, dyslipidemia, cardiovascular disease, and other chronic illnesses.

The study was conducted in accordance with the principles of the Declaration of Helsinki, and written informed consent was obtained from all participants.

2.7 Ethical Consideration

The study protocol was reviewed and approved by the Scientific and Ethical Committee of the College of Medicine, Hawler Medical University (Approval No. 47; 28-08-2024). All procedures were performed in accordance with the ethical standards of the Declaration of Helsinki. Written informed consent was obtained from all participants before enrollment.

2.8 Statistical analysis

Data were analyzed using IBM SPSS Statistics version 26.0 (IBM Corp., Armonk, NY, USA). Continuous variables were presented as mean ± standard deviation (SD) with 95% confidence intervals (CIs), and categorical variables as frequencies and percentages. Data normality was assessed using the Shapiro–Wilk test and inspection of histograms and Q–Q plots.

Comparisons between two groups used the independent-samples t-test for normally distributed variables and the Mann–Whitney U test for non-normal data. Comparisons among the four MTX/GC exposure subgroups employed one-way ANOVA for parametric data and the Kruskal–Wallis test for nonparametric data, with post hoc analyses when appropriate. Categorical variables were compared using the Pearson χ^2 test.

Patients on alternative DMARDs or biologics were included in the study. We adjusted for the use of other DMARDs and biologics in the multivariable analysis to account for their potential confounding effects on insulin sensitivity

Correlations between continuous variables were assessed using Spearman's rank correlation (ρ). Multivariable linear regression was performed to identify independent predictors of random blood sugar (RBS), including age, sex, BMI, RA duration, RF, anti-CCP, ESR, HbA1c, MTX dose and duration, DAS-28, and GC dose and duration. Model performance was evaluated using R^2, adjusted R^2, and the standard error of estimate, with VIFs and residuals checked for assumption validity.

All tests were two-tailed, and a p-value ≤ 0.05 was considered statistically significant.

3. Results

A total of 101 patients with RA were enrolled. Patients were stratified according to methotrexate (MTX) exposure into MTX users (n = 52) and non-MTX users (n = 49). Based on concomitant glucocorticoid (GC) therapy, they were further divided into four subgroups: MTX without GCs (24 patients, 23.76%), MTX with GCs (28 patients, 27.72%), no MTX and no GCs (25 patients, 24.75%), and GCs only (24 patients, 23.76%), as shown in Table 1.

The mean age of the study population was 50.54 ± 9.09 years (95% CI: 48.75–52.34), with no significant difference among subgroups (p = 0.055). The cohort was predominantly female (91.09%), yielding a female-to-male ratio of approximately 10.2:1. The mean body mass index (BMI) was 30.48 ± 4.49 kg/m² (95% CI: 29.59–31.37), with subgroup means of 29.22 ± 5.15 kg/m² (95% CI: 27.04–31.40) for MTX+/GC−, 31.06 ± 4.80 kg/m² (95% CI: 29.23–32.89) for MTX+/GC+, 31.83 ± 4.10 kg/m² (95% CI: 30.11–33.55) for MTX−/GC+, and 29.72 ± 3.48 kg/m² (95% CI: 28.31–31.13) for MTX−/GC−. No statistically significant difference in BMI was observed among the groups (p = 0.155). As shown in Table 1, MTX users exhibited significantly lower RBS levels compared to non-users (p < 0.001). This difference remained significant after adjusting for potential confounders (Model A β = −18.55 mg/dL, p < 0.001). Key subgroup differences in RBS were observed between MTX/GC−, MTX/GC+, and GC-only users, with the lowest levels in MTX/GC− users

Table 1. Baseline characteristics of the RA patients (n=101).

Variable	MTX+/GC− (n=24)	MTX+/GC+ (n=28)	MTX−/GC+ (n=24)	MTX−/GC− (n=25)	Total (n=101)	p-value
Age/ years						
Mean ± SD	54.50 ± 8.87	50.86 ± 8.36	47.75 ±7.78	49.08 ±10.32	50.54 ± 9.09	0.055 ns
CI 95%	50.75 – 58.25	47.62 – 54.10	44.47 – 51.03	44.82 – 53.34	48.75 – 52.34	
Gender						
Female, n (%)	22 (91.67%)	27 (96.43%)	21 (87.50%)	22 (88.00%)	92 (91.09%)	0.664 ns
Male, n (%)	2 (8.33%)	1 (3.57%)	3 (12.50%)	3 (12.00%)	9 (8.91%)	
F:M ratio	11:1	27:1	7:1	7.33: 1	10.2:1	
BMI (kg/m^2)						
Mean ± SD	29.22 ± 5.15	31.06 ± 4.80	31.83 ± 4.10	29.72 ± 3.48	30.48 ± 4.49	0.155 ns
CI 95%	27.04 – 31.40	29.23 – 32.89	30.11 – 33.55	28.31 – 31.13	29.59 – 31.37	

Abbreviations: MTX, methotrexate; GC(s), glucocorticoid(s); MTX+/GC−, methotrexate (MTX) monotherapy; MTX+/GC+, MTX plus glucocorticoids (GCs); MTX−/GC+, glucocorticoid (GC) monotherapy; MTX−/GC−, neither MTX nor GCs; kg, kilogram; m, meter; BMI, body mass index (kg/m²); SD, standard deviation; CI, 95% confidence interval; n, number of patients; %, percentage; F:M, female-to-male ratio; p-value, probability value; ns, not significant. Statistical tests: Age and BMI were summarized as mean ± SD (with 95% CIs) and compared across groups using one-way ANOVA. Gender (female/male) was summarized as counts and percentages and compared using the Pearson χ^2 test.

Across the MTX/GC exposure strata—MTX+/GC− (n=24), MTX+/GC+ (n=28), MTX−/GC+ (n=24), and MTX−/GC− (n=25)—there was no evidence of between-group heterogeneity for RA duration, RF, anti-CCP, DAS-28, or ESR (all $p \geq 0.093$; Table 2). RA duration was comparable (range of group means 6.94–7.62 years; overall mean (SD) 7.38 (6.62) years; 95% CI 6.03–8.67; p = 0.803). RF values showed substantial dispersion across the cohort (group means ≈38–43 IU/mL; overall 48.29 (90.59) IU/mL; 95% CI 34.39–66.21; p = 0.904), as did anti-CCP (group means ≈65–159 U/mL; overall 93.30 (149.40) U/mL; 95% CI 63.66–122.94; p = 0.093). Disease activity by DAS-28 was similar across groups (means 4.04–4.79; overall 4.44 (1.27); 95% CI 4.18–4.69; p = 0.203), and ESR was likewise comparable (means 26.84–33.29 mm/hr.; overall 30.01 (18.59) mm/hr.; 95% CI 26.59–33.93; p = 0.641).

Table 2. Rheumatoid arthritis duration, autoantibodies, and inflammation across MTX/GC exposure groups.

RA metric / biomarker	MTX+/GC− (n=24)	MTX+/GC+ (n=28)	MTX−/GC+ (n=24)	MTX−/GC− (n=25)	Total (n=101)	p-value
RA duration/ years						
Mean ± SD	7.29 ± 6.82	7.62 ± 6.67	7.62 ± 5.49	6.94 ± 7.65	7.38 ± 6.62	0.803 ns
CI 95%	4.41 -10.17	4.86 – 10.22	5.31 – 9.94	3.78 – 10.10	6.03 – 8.67	
RF (IU/mL)						
Mean ± SD	38.47 ± 30.18	43.40 ± 47.62	38.43 ± 37.54	40.66 ± 53.14	48.29 ± 90.59	0.904 ns
CI 95%	25.73 – 51.22	22.59 – 59.81	22.58 – 54.28	18.73 – 62.60	30.4 – 66.20	
Anti-CCP (U/mL)						
Mean ± SD	158.65 ± 195.62	73.65 ± 132.96	64.85 ± 116.69	79.10 ± 132.19	93.30 ± 149.40	0.093 ns
CI 95%	76.05 – 241.25	21.05 – 126.29	15.58 - 114.13	24.54 – 133.67	63.66 – 122.94	
DAS-28						
Mean ± SD	4.43 ± 1.37	4.79 ± 1.29	4.44 ± 1.23	4.04 ± 1.14	4.44 ± 1.27	0.203 ns
CI 95%	3.86 – 5.01	4.28 – 5.33	3.91 – 4.96	3.57 – 4.51	4.18 – 4.69	
ESR (mm/hr.)						
Mean ± SD	30.62 ± 19.63	33.29 ± 18.61	28.88 ± 17.14	26.84 ± 19.37	30.01 ± 18.59	0.641 ns
CI 95%	22.34 – 38.91	27.17 – 41.50	21.64 – 36.11	18.85 – 34.83	26.59 – 33.93	

Abbreviations: MTX, methotrexate; GC(s), glucocorticoid(s); MTX+/GC−, methotrexate monotherapy; MTX+/GC+, MTX plus glucocorticoids; MTX−/GC+, glucocorticoid monotherapy; MTX−/GC−, neither MTX nor GCs; RA duration, rheumatoid arthritis duration (years); RF, rheumatoid factor (IU/mL); Anti-CCP, anti–cyclic citrullinated peptide (U/mL); DAS-28, Disease Activity Score-28 (units); ESR, erythrocyte sedimentation rate (mm/hr.); SD, standard deviation; CI, confidence interval; n, number of patients; %, percentage; ns, not significant.

Statistical tests: Between-group comparisons used ANOVA for DAS-28 and ESR and Kruskal–Wallis for RA duration, RF, and Anti-CCP.

HbA1c did not differ significantly among MTX+/GC− (5.22 ± 0.23%), MTX+/GC+ (5.35 ± 0.17%), MTX−/GC+ (5.33 ± 0.17%), and MTX−/GC− (5.35 ± 0.26%) (overall mean 5.32 ± 0.21%; Kruskal–Wallis $p = 0.087$). By contrast, RBS varied significantly across groups (Kruskal–Wallis $p < 0.001$): MTX+/GC− had the lowest mean RBS (100.25 ± 9.33 mg/dL), followed by MTX+/GC+ (106.50 ± 15.09 mg/dL), whereas MTX−/GC+ and MTX−/GC− showed higher values (123.83 ± 16.54 and 118.32 ± 17.46 mg/dL, respectively). Group-wise 95% CIs were non-overlapping between MTX-exposed and non–MTX-exposed categories in several comparisons (e.g., MTX+/GC−: 96.31–104.19 vs MTX−/GC+: 116.85–130.82 mg/dL). Table 3 and Figure 1.

Table 3. Glycemic indices—HbA1c (%) and random blood sugar (mg/dL) across methotrexate/glucocorticoid treatment-exposure groups.

Glycemic indices	MTX+/GC− (n=24)	MTX+/GC+ (n=28)	MTX−/GC+ (n=24)	MTX−/GC− (n=25)	Total (n=101)	p-value
HbA1c (%)						
Mean ± SD	5.22 ± 0.23	5.35 ± 0.17	5.33 ± 0.17	5.35 ± 0.26	5.32 ± 0.21	0.087 ns
CI 95%	5.13 – 5.32	5.28 – 5.41	5.26 – 5.41	5.24 – 5.45	5.27 – 5.35	
RBS (mg/dL)						
Mean ± SD	100.25 ± 9.33	106.50 ± 15.09	123.83 ± 16.54	118.32 ± 17.46	112.06 ± 17.40	<0.001 **
CI 95%	96.31 – 104.19	101.52 – 113.15	116.85 – 130.82	111.11 – 125.53	108.61 – 115.49	

Abbreviations: MTX, methotrexate; GC(s), glucocorticoid(s); MTX+/GC−, methotrexate monotherapy; MTX+/GC+, methotrexate plus glucocorticoids; MTX−/GC+, glucocorticoid monotherapy; MTX−/GC−, neither methotrexate nor glucocorticoids; RBS, random blood sugar; SD, standard deviation; CI, confidence interval; n, number of patients. Statistical test: Kruskal–Wallis test for both HbA1c and RBS.

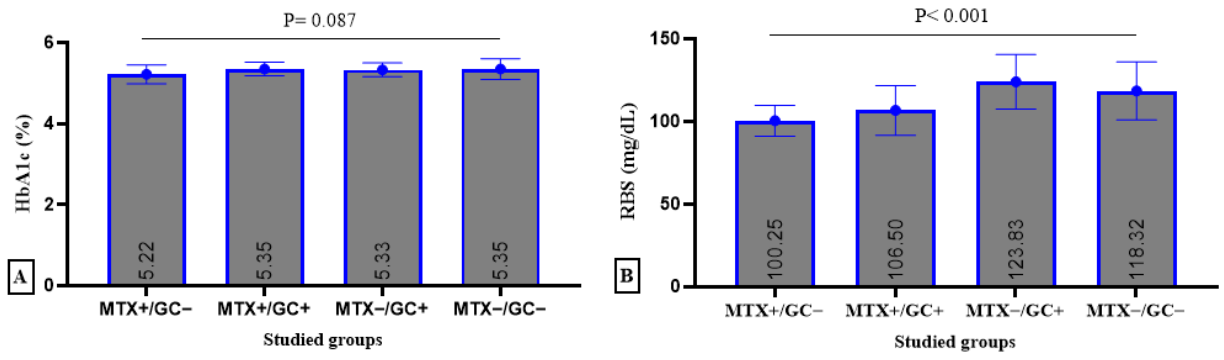

Figure 1. Comparison of mean+ SD HbA1c (A) and RBS (B) levels among the studied rheumatoid arthritis subgroups.

As shown in Table 4, among MTX-exposed patients, MTX dose and MTX duration were comparable between MTX+/GC− and MTX+/GC+ (mean dose 13.02 ± 3.70 vs 13.66 ± 2.40 mg/week; 95% CIs 11.60–14.65 vs 12.83–14.67; p=0.494; mean duration 3.40 ±

3.59 vs 3.94 ± 5.20 years; 95% CIs 1.96–4.96 vs 2.09–6.05; p=0.498). Among GC-exposed patients, GC dose did not differ between MTX+/GC+ and MTX−/GC+ (5.84 ± 2.81 vs 5.42 ± 1.82 mg/day; 95% CIs 4.98–6.73 vs 4.65–6.18; p=0.520). By contrast, GC duration was longer in MTX+/GC+ than MTX−/GC+ (9.00 ± 6.12 vs 6.29 ± 4.71 weeks; 95% CIs 6.63–11.37 vs 4.30–8.28; p=0.042), despite similar doses. Totals across exposed subsets were 13.37 ± 3.05 mg/week for MTX dose (95% CI 12.63–14.29), 3.69 ± 4.50 years for MTX duration (95% CI 2.56–5.02), 5.64 ± 2.07 mg/day for GC dose (95% CI 5.08–6.23), and 7.75 ± 5.63 weeks for GC duration (95% CI 6.18–9.32).

Table 4. Treatment exposure characteristics (dose and duration) by group.

Treatment exposure	MTX+/GC− (n=24)	MTX+/GC+ (n=28)	MTX−/GC+ (n=24)	MTX−/GC− (n=25)	Total (n=101)	p-value
MTX dose (mg/week)						
Mean ± SD	13.02 ± 3.70	13.66 ± 2.40	-	-	13.37 ±3.05	0.494 ns
CI 95%	11.60 - 14.65	12.83 – 14.67	-	-	12.63–14.29	
MTX duration (years)						
Mean ± SD	3.40 ± 3.59	3.94 ± 5.20	-	-	3.69 ± 4.50	0.498 ns
CI 95%	1.96 – 4.96	2.09 – 6.05	-	-	**2.56–5.02**	
GCs dose (mg/day)						
Mean ± SD	-	5.84 ± 2.81	5.42 ± 1.82	-	5.64 ± 2.07	0.520 ns
CI 95%	-	4.98 – 6.73	4.65 – 6.18	-	5.08–6.23	
GCs duration (weeks)						
Mean ± SD	-	9.00 ± 6.12	6.29 ± 4.71	-	7.75 ± 5.63	0.042 *
CI 95%	-	6.63 – 11.37	4.30 – 8.28	-	6.18–9.32	

Abbreviations: MTX, methotrexate; GC(s), glucocorticoid(s); MTX+/GC−, methotrexate monotherapy; MTX+/GC+, methotrexate plus glucocorticoids; MTX−/GC+, glucocorticoid monotherapy; MTX−/GC−, neither methotrexate nor glucocorticoids; SD, standard deviation; CI, confidence interval. Statistical test: Mann–Whitney U test.

In Spearman rank analyses, RBS correlated inversely with MTX exposure, showing moderate negative associations with both MTX dose ($\rho = -0.397$, $p = 0.004$) and MTX duration ($\rho = -0.439$, $p = 0.001$). GC dose correlated positively with RBS ($\rho = 0.282$, $p = 0.043$), whereas GC duration did not ($\rho = 0.058$, $p = 0.682$).

By contrast, HbA1c showed no significant correlations with any exposure variable (MTX dose $\rho = -0.060$, $p = 0.674$; MTX duration $\rho = -0.194$, $p = 0.169$; GC dose $\rho = 0.062$, $p = 0.660$; GC duration $\rho = 0.063$, $p = 0.656$). See Table 5.

Table 5. Spearman correlations between treatment exposures and glycemic indices.

Treatment exposure	Glycemic indices	
	HbA1c	**RBS**
MTX dose (mg/week)		
Spearman's ρ	-0.06 ns	-0.397 **
P value	0.674	0.004
MTX duration		
Spearman's ρ	-0.194 ns	-0.439 **
P value	0.169	0.001
GCs dose (mg/day)		
Spearman's ρ	0.062 ns	0.282 *

P value	0.660	0.043
GCs duration (week)		
Spearman's ρ	0.063 ns	0.058 ns
P value	0.656	0.682

Abbreviations: MTX, methotrexate; GC(s), glucocorticoid (s); RBS, random blood sugar (mg/dL); HbA1c, glycated hemoglobin (%); ρ, Spearman's rank correlation coefficient; p, two-tailed p-value; ns, not significant (p > 0.05); *, p < 0.05; **, p < 0.01.

A simple linear regression evaluated the association between methotrexate (MTX) dose (mg/week) and random blood sugar (RBS). The model was statistically significant, *F* (1,50) =10.09,p=0.003, accounting for 16.8% of the variance in RBS (R=0.410, R^2= 0.168; adjusted R^2=0.151; standard error of estimate = 11.99 mg/dL). MTX dose showed a **negative** association with RBS (unstandardized B=−1.776, SE = 0.559; standardized β=−0.410; t=−3.176, p=0.003), indicating that each additional 1 mg/week of MTX is associated with an average 1.78 mg/dL lower RBS. As shown in Figure 2.

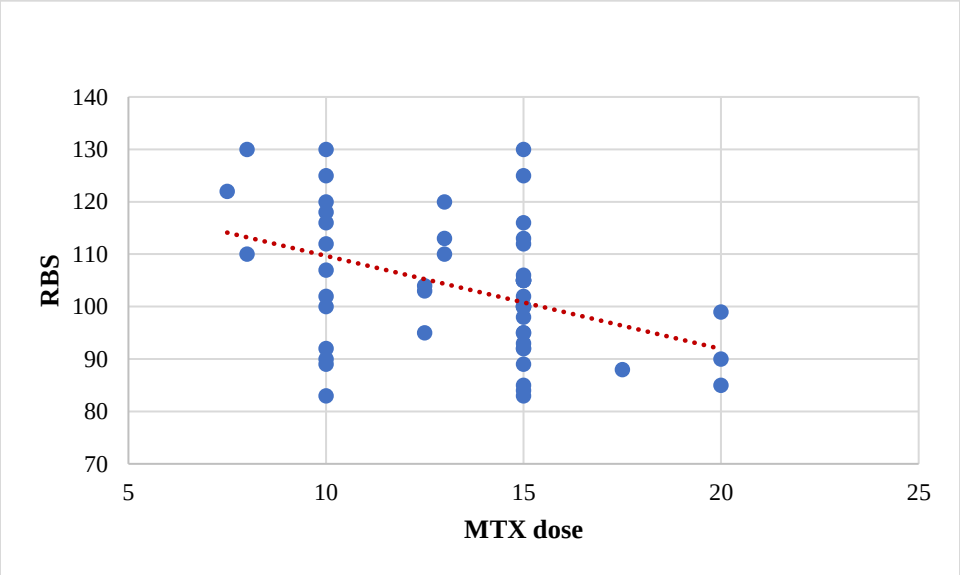

Figure 2. Relationship between methotrexate dose (95% CI= 12.63–14.29) and random blood sugar (95% CI= 108.61 – 115.49). Scatterplot (n=52) with least-squares line shows a clear inverse association between MTX dose (mg/week) and RBS (mg/dL) (B = −1.776 mg/dL per 1 mg/week, SE = 0.559; β = −0.410). The model is significant, *F* (1,50) =10.09, p=0.003, explaining R^2 = 0.168 of variance (adjusted R^2 = 0.151; standard error of estimate 11.99 mg/dL).

A simple linear regression evaluated the association between methotrexate (MTX) duration of use (years) and random blood sugar (RBS)**.** The model was statistically significant, *F* (1,50) =4.29, p=0.043, explaining a small proportion of variance in RBS (R=0.281,R^2=0.079, adjusted R^2=0.061; standard error of estimate = 12.623 mg/dL). MTX duration was inversely associated with RBS (unstandardized B=−0.807, SE = 0.390; standardized β=−0.281; t=−2.072, p=0.043), indicating that each additional year of MTX therapy corresponded to an average 0.81 mg/dL lower RBS. The accompanying scatterplot depicts a shallow downward trend consistent with the modest R^2. As shown in Figure 3.

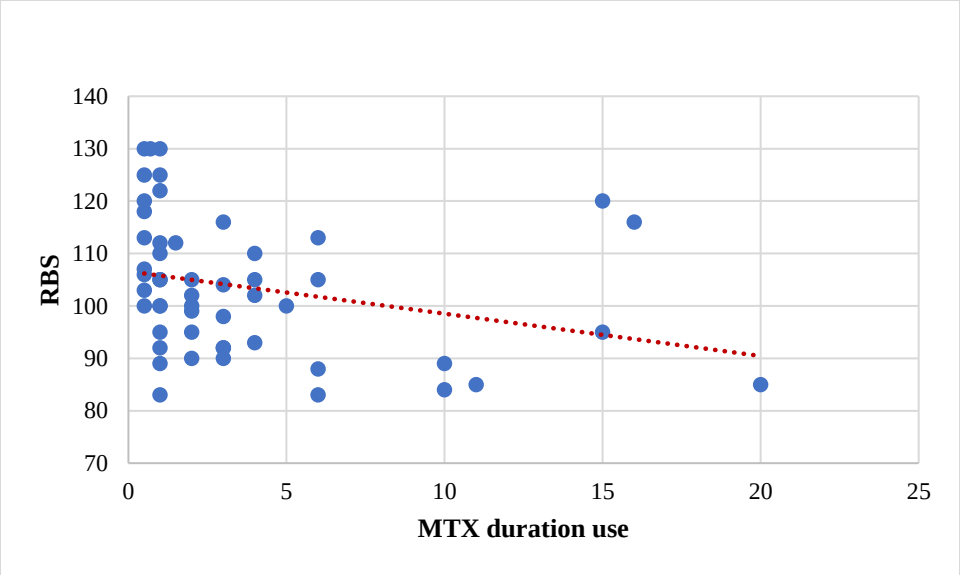

Figure 3. Relationship between methotrexate duration (95% CI= **2.56–5.02) and random blood sugar** (95% CI= 108.61 – 115.49). Scatterplot with least-squares line shows a modest inverse association between MTX duration (years) and RBS (mg/dL) (B = −0.807 mg/dL per year, SE = 0.390; R^2 = 0.079). The regression is statistically significant, *F* (1,50) =4.29, p=0.043, with a standard error of estimate of 12.6 mg/dL.

In the multivariable linear regression for random blood sugar (RBS), age (B = −0.04, 95% CI −0.57 to 0.49, *p*=0.864), gender (B = −6.45, −34.18 to 21.28, *p*=0.624), and BMI (B = −0.01, −1.02 to 1.01, *p*=0.988) were not associated with RBS. RA duration showed a significant inverse association (B = −1.30 mg/dL per year, −2.44 to −0.16, *p*=0.028). RF was positively associated with RBS (B = 0.37 per unit, 0.16 to 0.58, *p*=0.002), whereas Anti-CCP was not (B = −0.03, −0.08 to 0.01, *p*=0.135). ESR was non-significant (B = −0.10, −0.37 to 0.16, *p*=0.416). HbA1c exhibited a strong positive association with RBS (B = 82.59 mg/dL per 1% HbA1c, 40.93 to 124.26, *p*=0.001). Neither MTX duration (B = −0.58, −2.05 to 0.89, *p*=0.408) nor MTX dose (B = −0.39, −2.55 to 1.76, *p*=0.697) nor DAS-28 (B = −2.30, −7.28 to 2.67, *p*=0.336) was significant. Among glucocorticoid exposures, GC dose was positively associated with RBS (B = 3.54 mg/dL per 1 mg/day, 0.46 to 6.61, *p*=0.027), while GC duration was not (B = −0.48, −1.39 to 0.42, *p*=0.267). As shown in Table 6.

Table 6. Multivariable linear regression identifying independent predictors of random blood sugar (RBS) in non-diabetic rheumatoid arthritis patients

Variable	B	SE	95% CI Lower	95% CI Upper	p-value
Age	-0.04	0.25	-0.57	0.49	0.864 ns
Gender	-6.45	12.84	-34.18	21.28	0.624 ns
BMI	-0.01	0.47	-1.02	1.01	0.988 ns
RA duration	-1.30	0.53	-2.44	-0.16	0.028 *
RF	0.37	0.10	0.16	0.58	0.002 *
Anti-CCP	-0.03	0.02	-0.08	0.01	0.135 ns
ESR	-0.10	0.12	-0.37	0.16	0.416 ns

HbA1c	82.59	19.29	40.93	124.26	0.001 *
MTX duration	-0.58	0.68	-2.05	0.89	0.408 ns
MTX dose	-0.39	0.99	-2.55	1.76	0.697 ns
DAS- 28	-2.30	2.30	-7.28	2.67	0.336 ns
GCs dose	3.54	1.42	0.46	6.61	0.027 *
GCs duration	-0.48	0.42	-1.39	0.42	0.267 ns

Abbreviations: MTX, methotrexate; MTX dose, mg/week; MTX duration, years; GC(s), glucocorticoid(s); GCs dose, mg/day; GCs duration, weeks; RA, rheumatoid arthritis; RA duration, years; RF, rheumatoid factor (IU/mL); Anti-CCP, anti–cyclic citrullinated peptide (U/mL); DAS-28, Disease Activity Score-28 (units); ESR, erythrocyte sedimentation rate (mm/hr.); HbA1c, glycated hemoglobin (%), gender (reference: male; female coded as 1, unless stated otherwise). B, unstandardized regression coefficient; SE, standard error; 95% CI, 95% confidence interval; p-value, two-sided significance; ns, not significant; *, $p<0.05$.

4. Discussion

Methotrexate, a cornerstone in the management of rheumatoid arthritis, is primarily known for its immunosuppressive and anti-inflammatory effects. However, its potential influence on metabolic parameters, including blood glucose levels, has sparked interest in recent years. In non-diabetic patients with rheumatoid arthritis, the interplay between systemic inflammation and insulin sensitivity may be modulated by methotrexate therapy. As inflammation subsides, improvements in glucose metabolism could occur, potentially leading to subtle changes in random blood sugar levels (22). Our findings are consistent with previous studies reporting a potential glycemic benefit of MTX beyond its anti-inflammatory effects (23, 24). However, unlike other study (25). we did not observe changes in HbA1c, possibly due to the short duration and normoglycemic baseline in our cohort.

In a consecutively recruited cohort of non-diabetic RA patients, methotrexate (MTX) use was associated with substantially lower random blood sugar (RBS) versus non-use, an effect that persisted after multivariable adjustment for demographic, anthropometric, inflammatory, and treatment covariates. Subgroup analyses showed the lowest RBS in MTX without glucocorticoids (GCs) and the highest in GC-only users, with MTX+GC intermediate supporting a mitigating effect of MTX on GC-related hyperglycemia. Consistent with the cohort's normoglycemia and the short observation window. Our findings are consistent with earlier studies reporting potential metabolic benefits of MTX beyond inflammation control. Tam et al., observed that MTX, alone or with metformin, improved blood glucose in psoriasis patients with metabolic syndrome (24). Russo et al., demonstrated in animal models that low-dose MTX upregulated GLUT4 expression and enhanced metabolic control in diabetic mice (23). Mechanistic data further support these observations: MTX has been shown to enhance insulin receptor signaling and GLUT4 translocation, promoting glucose uptake (26-28).Together, these studies provide biological plausibility for MTX improving glycemic regulation.

However, not all studies confirm this effect. Gisondi et al., reported no significant change in fasting glucose among psoriasis patients treated with MTX (25). Similarly, Wu et al., (29), and Dehpouri et al., (30) found no improvement in HbA1c in inflammatory arthritis cohorts on MTX . These inconsistencies may reflect heterogeneity in baseline glycemic status, study design, or concomitant therapies, including other DMARDs (31).

In our study, HbA1c did not differ between MTX users and non-users and showed no subgroup differences, this finding is consistent

with Wu et al., and Dehpouri et al. (30), who also found no HbA1c changes with MTX, but contrasts with Perdan-Pirkmajer et al., who reported a modest reduction in HbA1c (17). One possible explanation is that our study population excluded diabetics and prediabetics, resulting in relatively normal baseline HbA1c, thus limiting detectable differences. Moreover, HbA1c reflects long-term glycemic control and may be less sensitive to short-term or modest improvements compared to RBS. Concomitant DMARD therapy, which reduces systemic inflammation, may have further masked subtle effects (31).

Our results also reaffirm the diabetogenic properties of glucocorticoids. Patients on GCs alone exhibited the highest RBS values, consistent with prior evidence that GCs impair insulin sensitivity and increase glucose levels (32). Notably, MTX appeared to mitigate these effects, as patients receiving MTX+GCs had significantly lower RBS than GC-only users. This finding highlights the potential role of MTX in reducing the metabolic complications of chronic GC therapy, which remains integral in RA management.

Our results revealed that a higher glucocorticoid dose was independently associated with higher RBS, while longer RA duration was related to lower RBS and rheumatoid factor positivity to higher RBS; HbA1c tracked positively with RBS, supporting internal coherence of glycemic measures.

Consistent with this, bivariate analyses showed an inverse association between RBS and MTX exposure and a positive association with glucocorticoid dose, while glucocorticoid duration showed no association. Notably, MTX dose and duration were not independent predictors after adjustment, suggesting a threshold/class effect of MTX exposure rather than a linear dose–response, whereas age, sex, BMI, anti-CCP, ESR, and DAS-28 were not associated with RBS.

Notably, after adjustment, neither MTX dose nor duration was an independent predictor of RBS. This finding may suggest that the glycemic benefit of MTX acts as a class effect that is achieved upon exposure, rather than following a linear dose-response relationship within the therapeutic range observed in our cohort

The biological plausibility of MTX's metabolic effects is well supported. MTX inhibits AICAR transformylase, leading to intracellular accumulation of AICAR, a potent activator of AMP-activated protein kinase (AMPK) (10, 11). AMPK is a central regulator of energy metabolism, reducing hepatic gluconeogenesis, enhancing insulin sensitivity, and promoting glucose uptake (14, 13, 15). This pathway mirrors the mechanism of metformin, a cornerstone antidiabetic agent (14, 15).

Additionally, MTX has been shown to enhance GLUT4 translocation in skeletal muscle (23, 26-28), thereby improving glucose utilization. Another mechanism may involve the reduction of systemic inflammation and cytokine-driven insulin resistance (19). Taken together, these mechanistic insights reinforce the observed protective association between MTX and RBS in our study.

The current findings expand the therapeutic value of MTX by highlighting its potential metabolic benefits in addition to its established anti-inflammatory effects. By lowering random blood sugar (RBS) and attenuating glucocorticoid-induced hyperglycemia, MTX may contribute to reducing diabetes risk, which is higher among RA patients compared to the general population (6, 3, 33). Moreover, the consistently lower RBS values observed in MTX users suggest that conventional glucose monitoring may underestimate early dysglycemia in this subgroup, which has important diagnostic implications.

Strengths of this study include a well-characterized RA cohort, careful subgroup stratification by treatment regimen, and

parallel evaluation of both RBS and HbA1c.

Conclusions

In this cross-sectional cohort, methotrexate (MTX) use was independently associated with lower random blood sugar (RBS) after adjustment for age, body mass index (BMI), disease activity, and glucocorticoid (GC) exposure. Among GC-treated patients, concurrent MTX therapy was also associated with lower RBS compared with GC treatment alone. HbA1c levels did not differ between MTX-exposed and unexposed groups. These findings suggest a potential short-term glycemic benefit of MTX, warranting prospective studies to establish causality and clinical significance.

Recommendations

MTX may be favored, when appropriate, for RA patients at risk of GC-induced hyperglycemia, pending confirmation from prospective studies, MTX could be considered a preferable DMARD in RA patients with metabolic risk, pending confirmation from longitudinal trials. Clinicians should interpret RBS cautiously in MTX users, as lower values may mask early dysglycemia. complementary tests such as HbA1c or fasting glucose are advised. Routine RBS monitoring is recommended for RA patients receiving GCs without MTX. Future longitudinal studies with larger cohorts and standardized glycemic assessments are needed to clarify MTX's metabolic effects.

Limitations

The study's limitations include its cross-sectional design, which precludes establishing temporal or causal direction between MTX exposure and lower RBS; the non-standardized timing of RBS sampling (fasting vs. postprandial), potentially affecting measurement precision; and the lack of causal inference. Lifestyle factors such as diet, physical activity, and smoking were not controlled, and fasting plasma glucose or oral glucose tolerance tests (OGTT) were not performed. These factors may have introduced residual confounding and influenced the observed associations.

Conflict of Interest Statement: The authors declare that there are no conflicts of interest related to this work.

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References

1. Hotamisligil GS. Inflammation and metabolic disorders. Nature. (2006);444(7121):860-867. https://doi.org/10.1038/nature05485.
2. Ormseth MJ, Swift LL, Fazio S, Linton MF, Chung CP, Raggi P, et al. Free fatty acids are associated with insulin resistance but not coronary artery atherosclerosis in rheumatoid arthritis. Atherosclerosis. (2011);219(2):869-874. https://doi.org/10.1016/j.atherosclerosis.2011.09.005
3. Hoes J, Van Der Goes M, Van Raalte D, Van Der Zijl N, Den Uyl D, Lems W, et al. Glucose tolerance, insulin sensitivity and β-cell function in patients with rheumatoid arthritis treated with or without low-to-medium dose glucocorticoids. Annals of the rheumatic diseases. (2011);70(11):1887-1894. https://doi.org/10.1136/ard.2011.151464.
4. Wild S, Roglic G, Green A, Sicree R, King H. Global prevalence of diabetes: Estimates for the year 2000 and projections for 2030. Diabetes care. (2004);27(5):1047-1053. https://doi.org/10.2337/diacare.27.5.1047
5. Simard JF, Mittleman MA. Prevalent rheumatoid arthritis and diabetes among nhanes iii participants aged 60 and older. The Journal of rheumatology. (2007);34(3):469-473.
6. Solomon DH, Love TJ, Canning C, Schneeweiss S. Risk of diabetes among patients with rheumatoid arthritis, psoriatic arthritis and psoriasis. Annals of the rheumatic diseases. (2010);69(12):2114-2117. https://doi.org/10.1136/ard.2009.125476
7. Banerjee D, Ercikan-Abali E, Waltham M, Schnieders B, Hochhauser D, Li WW, et al. Molecular mechanisms of resistance to antifolates, a review. Acta Biochimica Polonica.

(1995);42(4):457-464. https://doi.org/10.18388/abp.1995_4899.
8. Baghdadi LR, Woodman RJ, Shanahan EM, Wiese MD, Mangoni AA. Genetic polymorphism of the methotrexate transporter abcg2, blood pressure and markers of arterial function in patients with rheumatoid arthritis: Repeated cross-sectional study. Pharmacogenomics and Personalized Medicine. (2018);11(11):205-210. https://doi.org/10.2147/PGPM.S170557.
9. Baghdadi LR. Effect of methotrexate use on the development of type 2 diabetes in rheumatoid arthritis patients: A systematic review and meta-analysis. PLoS One. (2020);15(7):e0235637. https://doi.org/10.1371/journal.pone.0235637.
10. Corton JM, Gillespie JG, Hawley SA, Hardie DG. 5-aminoimidazole-4-carboxamide ribonucleoside: A specific method for activating amp-activated protein kinase in intact cells? European journal of biochemistry. (1995);229(2):558-565. https://doi.org/10.1111/j.1432-1033.1995.tb20498.x.
11. Viollet B, Foretz M, Guigas B, Horman S, Dentin R, Bertrand L, et al. Activation of amp-activated protein kinase in the liver: A new strategy for the management of metabolic hepatic disorders. The Journal of physiology. (2006);574(1):41-53. https://doi.org/10.1113/jphysiol.2006.108506.
12. Hardie DG. Ampk: A target for drugs and natural products with effects on both diabetes and cancer. Diabetes. (2013);62(7):2164-2172. https://doi.org/10.2337/db13-0368.
13. Musi N, Goodyear L. Amp-activated protein kinase and muscle glucose uptake. Acta Physiologica Scandinavica. (2003);178(4):337-345. https://doi.org/10.1046/j.1365-201X.2003.01168.x.
14. Zhou G, Myers R, Li Y, Chen Y, Shen X, Fenyk-Melody J, et al. Role of amp-activated protein kinase in mechanism of metformin action. The Journal of clinical investigation. (2001);108(8):1167-1174. https://doi.org/10.1172/JCI13505.
15. Rena G, Hardie DG, Pearson ER. The mechanisms of action of metformin. Diabetologia. (2017);60(9):1577-1585. https://doi.org/10.1007/s00125-017-4342-z.
16. Pirkmajer S, Kulkarni SS, Tom RZ, Ross FA, Hawley SA, Hardie DG, et al. Methotrexate promotes glucose uptake and lipid oxidation in skeletal muscle via ampk activation. Diabetes. (2015);64(2):360-369. https://doi.org/10.2337/db14-0508
17. Perdan-Pirkmajer K, Pirkmajer S, Thevis M, Thomas A, Praprotnik S, Hočevar A, et al. Methotrexate reduces hba1c concentration but does not produce chronic accumulation of zmp in patients with rheumatoid or psoriatic arthritis. Scandinavian journal of rheumatology. (2016);45(5):347-355. https://doi.org/10.3109/03009742.2015.1105290.
18. Mantravadi S, George M, Brensinger C, Du M, Baker JF, Ogdie A. Impact of tumor necrosis factor inhibitors and methotrexate on diabetes mellitus among patients with inflammatory arthritis. BMC rheumatology. (2020);4(1):39. https://doi.org/10.1186/s41927-2020-00138-3.
19. Baker JF, England BR, George M, Cannon G, Sauer B, Ogdie A, et al. Disease activity, cytokines, chemokines and the risk of incident diabetes in rheumatoid arthritis. Annals of the rheumatic diseases. (2021);80(5):566-572. https://doi.org/10.1136/annrheumdis-2020-219140.
20. Aletaha D, Neogi T, Silman AJ, Funovits J, Felson DT, Bingham Iii CO, et al. 2010 rheumatoid arthritis classification criteria: An american college of rheumatology/european league against rheumatism collaborative initiative. Arthritis & rheumatism. (2010);62(9):2569-2581. https://doi.org/10.1002/art.27584.
21. Elsayed NA, Aleppo G, Aroda VR, Bannuru RR, Brown FM, Bruemmer D, et al. Improving care and promoting health in populations: Standards of care in diabetes—2023. Diabetes care. (2023);46(Supplement_1):S10-S18. https://doi.org/10.2337/dc23-S001.
22. Perdan-Pirkmajer K, Pirkmajer S, Hočevar A, Rotar Ž, Gašperšič N, Praprotnik S, et al. Ab0334 methotrexate reduces hba1c in non-diabetic patients with newly diagnosed rheumatoid arthritis or psoriatic arthritis. Annals of the Rheumatic Diseases. (2014);73:915. https://doi.org/10.1136/annrheumdis-2014-eular.2995.
23. Russo GT, Minutoli L, Bitto A, Altavilla D, Alessi E, Giandalia A, et al. Methotrexate increases skeletal muscle glut4 expression and improves metabolic control in experimental diabetes. Journal of Nutrition and Metabolism. (2012);2012(1):132056. https://doi.org/10.1155/2012/132056.
24. Tam HTX, Thuy LND, Vinh NM, Anh TN, Van BT. The combined use of metformin and methotrexate in psoriasis patients with metabolic syndrome. Dermatology Research and Practice. (2022);2022(1):9838867. https://doi.org/10.1155/2022/9838867.
25. Gisondi P, Cotena C, Tessari G, Girolomoni G. Anti–tumour necrosis factor-α therapy increases body weight in patients with chronic plaque psoriasis: A retrospective cohort study. Journal of the European Academy of Dermatology and Venereology. (2008);22(3):341-344. https://doi.org/10.1111/j.1468-3083.2007.02429.x.
26. Riedinger C, Mendler M, Schlotterer A, Fleming T, Okun J, Hammes H-P, et al. High-glucose toxicity is

mediated by aicar-transformylase/imp cyclohydrolase and mitigated by amp-activated protein kinase in caenorhabditis elegans. Journal of Biological Chemistry. (2018);293(13):4845-4859. https://doi.org/10.1074/jbc.M117.805879.
27. Friedman B, Cronstein B. Methotrexate mechanism in treatment of rheumatoid arthritis. Joint bone spine. (2019);86(3):301-307. https://doi.org/10.1016/j.jbspin.2018.07.004.
28. Yano N, Zhang L, Wei D, Dubielecka PM, Wei L, Zhuang S, et al. Irisin counteracts high glucose and fatty acid-induced cytotoxicity by preserving the ampk-insulin receptor signaling axis in c2c12 myoblasts. American Journal of Physiology-Endocrinology and Metabolism. (2020);318(5):791-805. https://doi.org/10.1152/ajpendo.00219.2019.
29. Wu JJ, Rowan CG, Bebchuk JD, Anthony MS. No association between tnf inhibitor and methotrexate therapy versus methotrexate in changes in hemoglobin a1c and fasting glucose among psoriasis, psoriatic arthritis, and rheumatoid arthritis patients. Journal of drugs in dermatology: JDD. (2015);14(2):159-166.
30. Dehpouri T, Rokni GR, Narenjbon NA, Goldust M, Yamauchi PS, Wollina U, et al. Evaluation of the glycemic effect of methotrexate in psoriatic arthritis patients with metabolic syndrome: A pilot study. Dermatology reports. (2019);11(1):7965. https://doi.org/10.4081/dr.2019.7965.
31. Su Y-J, Chen H-M, Chan T-M, Cheng T-T, Yu S-F, Chen J-F, et al. Disease-modifying anti-rheumatic drugs associated with different diabetes risks in patients with rheumatoid arthritis. RMD open. (2023);9(3):e003045. https://doi.org/10.1136/rmdopen-2023-003045.
32. Dessein PH, Joffe BI, Stanwix AE, Christian BF, Veller M. Glucocorticoids and insulin sensitivity in rheumatoid arthritis. The Journal of Rheumatology. (2004);31(5):867-874.
33. Tian Z, Mclaughlin J, Verma A, Chinoy H, Heald AH. The relationship between rheumatoid arthritis and diabetes mellitus: A systematic review and meta-analysis. Cardiovascular endocrinology & metabolism. (2021);10(2):125-131. https://doi.org/10.1097/XCE.0000000000000244.