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METHOTREXATE-INDUCED TOXICITIES IN CHILDREN WITH MALIGNANCY: A SYSTEMATIC REVIEW AND META-ANALYSIS

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Abstract – *Objective:* *Methotrexate (MTX) is an anticancer and anti-inflammatory drug widely used to treat various diseases. Some patients are more susceptible to toxicity, some show resistance to chemotherapy. The most conducted studies have investigated the effect of gene polymorphism on MTX toxicity, and little data are available about the relationship between MTX dose and type of toxicity.*

Materials and Methods: *In the present meta-analysis, we evaluated 20 studies that focused on the association between MTX dose and MTX-related toxicities.*

Results: *Our pooled analysis showed GI toxicity has higher prevalence (p= 0.51, 95% CI (0.10, 0.91)) compared to other toxicities, follow by hematologic toxicity (p = 0.41, 95% CI (0.25, 0.56)) and mucositis (p = 0.41, 95% CI (0.17, 0.65)). Whereas, renal toxicity (p = 0.32 [95% CI (0.12, 0.52)) showed the lowest prevalence. The results showed a significant prevalence of all toxicity (except the hematologic toxicity) in different dosages. Also, the dose of 12 mg showed higher toxicity than other dosages.*

Conclusions: *To reduce MTX-related adverse events in pediatrics, our findings suggested that more investigations for replacement of MTX are required.*

KEYWORDS: *Malignancy, Pediatric, MTX, Methotrexate, Toxicity, Neoplasm.*

INTRODUCTION

Methotrexate (MTX) is an anticancer and anti-inflammatory drug widely used in treating various diseases including acute lymphoid leukemia (ALL), lymphoma, osteosarcoma, and autoimmune diseases in adults and pediatrics. By disruption in the folate pathway, MTX inhibits purine and pyrimidine precursors for *de novo* synthesis of DNA and RNA and inhibits cell proliferation[1,2]. MTX is one of the most important and widely used anti-rheumatic drugs used in some rheumatic diseases, such as lupus and rheumatoid arthritis (RA)[3]. MTX may cause various adverse events (AEs), including bone marrow suppression, mucosal skin lesions, pneumonitis, and toxicity. High-dose MTX is a useful therapy associated with a low risk of central nervous system (CNS) recurrence in children with malignancy[4,5].

MTX-induced toxicity can lead to discontinuation of the drug in some cases, of which this toxicity possibly is due to the delay in clearance or high dose regimens of MTX[6,7]. Among various types of toxicity, bone marrow suppression and gastrointestinal toxicity (GI) are more serious and cause discontinuation of MTX[1]. In this regard, assessing pediatrics for MTX-related AEs is the most important issue which should be considered.

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Although the disease free-survival rate has increased with MTX, toxicity is a major challenging issue and concerns in MTX use in cancer chemotherapy. Some patients are more susceptible to toxicity, some show resistance to chemotherapy; thus, identifying the susceptible factors can be beneficial in advancing cancer treatment.

MATERIALS AND METHODS

This review was performed in accordance with the PRISMA (Preferred Reporting Items for Systematic reviews and Meta-Analyses) and MOOSE (Meta-analyses Of Observational Studies in Epidemiology) guidelines[8,9].

Search strategy

To evaluate the association between MTX dosage and toxicity in pediatrics with malignancy, we systematically searched the electronic database, including Scopus, Medline/PubMed, EMBASE, Web of sciences (WOS), and Cochrane library using Mesh-standardized keywords: (((malignant* OR neoplasm*) OR "lymphoblastic leukemia" [Mesh] OR "ALL" OR "acute myeloid leukemia" OR" AML" OR "osteosarcoma") AND (("Methotrexate" [Mesh] OR "Methotrexate" OR "Anti-Rheumatic" OR "DMARD" OR "disease modifying anti-rheumatic drug" OR "DMARD" [mesh] OR "Anti-rheumatic Agents" OR "Arthritis, Rheumatoid/drug therapy" AND "pediatric*" OR "paediatric*" OR "children")))) until Jan 2021. There is no restriction for time and language, and the citation lists of selected articles were handsearched for additional papers.

Data extraction

Two reviewers (ME and FR) independently screened titles and abstracts of all initially found articles. Information was extracted from selected studies, including the author's name, year of publication, country, type of study, sample size, gender, mean age type of malignancy, MTX dosage, type of toxicity, and final finding. A third reviewer was consulted to resolve any disagreements between reviewers by discussion until consensus was reached.

Eligible Criteria

To understand the influence of MTX dosage on toxicity development, studies that evaluated specific doses of MTX were selected. Inclusion criteria were considered as following: evaluation of the association between MTX dose and MTX-induced toxicity. All types of studies, including case/control, cohort, cross-sectional, and clinical trial studies, were included. Studies that evaluated MTX-induced toxicity in adults, studies that investigated MTX-induced toxicity in children with other diseases like rheumatoid arthritis, case reports, reviews, letters to editors, and as well as studies reporting cases with incomplete information were excluded.

Quality assessment

Two reviewers (F.R and M.E) individually assessed included studies by NEWCASTLE – OTTAWA checklists using the proper extension for each study design. These checklists consist of multiple questions about selecting subjects, comparability between case and control groups, and outcome assessment[10].

Statistical Analysis

Cochran Chi-square test and I^2 were used to assessing heterogeneity among studies. A fixed-effects model was used when $I^2 < 50\%$, while in the case of $I^2 > 50\%$, a random-effects model was selected. Fixed-model assumes that the population effect sizes are the same for all studies[11]. In contrast, the random-effects model attempted to generalize findings beyond the included studies by assuming that the selected studies are random samples from a larger population[12]. The number of various types of toxicity used to calculate the overall prevalence of side effects and its 95% confidence intervals (CI) were calculated to assess the association between MTX and various types of toxicity in children with different malignancies. According to the heterogeneity test results, either Der Simonian's and Laird's random-effects method or Mantel-Haenszel's fixed-effects method were used to estimate the overall prevalence and 95% confidence intervals[13]. Moreover, subgroup analysis was implemented based on the dosage of MTX and type of malignancies (ALL, acute myeloid leukemia (AML), non-Hodgkin lymphoma (NHL), and Osteosarcoma) as an important variable, which may cause heterogeneity between different toxicities. The Egger's test was used to investigate small study effects due to potential publication bias[14,15]. If there was statistical heterogeneity among the results, a further sensitivity analysis was conducted to determine the source of heterogeneity. After the significant clinical heterogeneity was excluded, the randomized effects model was used for meta-analysis. $p < 0.05$ was considered as statistical significance (2-sided). All data were analyzed using STAT 16 (STATA Corporation, College Station, TX, USA).

RESULTS

Overall, 2022 studies were collected. After removing duplicated studies, 963 studies have remained. During screening titles and abstracts, 92 studies were considered potentially eligible. Subsequently, in full-text screening, 72 studies were excluded, including 8 studies that investigated the MTX toxicity in adult patients, 30 studies evaluated MTX-related toxicity in rheumatoid arthritis, 10 did not provide the MTX dosage, 16 did not report clarified MTX-associated AEs, 5 and 3 were meta-analysis and Letter to the editor, respectively (Figure 1).

Finally, 20 studies were selected for further analyses. Sixteen (80%) of final selected studies were designed as cross-sectional, one (5%) was case-control, and the rest three (15%) were cohort studies (Table 1). The used treatment protocol was described carefully in each study. In all studies, the toxicity assessment was according to Common Terminology Criteria for Adverse Events version 4.0 and graded according to the World Health Organization (WHO) criteria. 11 (55%) of studies used both 2.5 g/m^2 MTX for standard/intermediate risk and 5 g/m^2 MTX for high risk patients[16-26], 5 studies (25%) used 5 g/m^2 dosage (27-30), 3 (12%) of selected studies used 12 g/m^2 dosage (31-33), and only a single study (5%) evaluated the toxicity of 2.5 g/m^2 of MTX (34). For malignancy type, 15 (75%) studies investigated ALL, following by 2 (10%) on osteosarcoma, one (5%) on NHL, one (5%) on ALL/NHL, and one (5%) on ALL/AML (Table 1). MTX-related toxicities are summarized in Table 1.

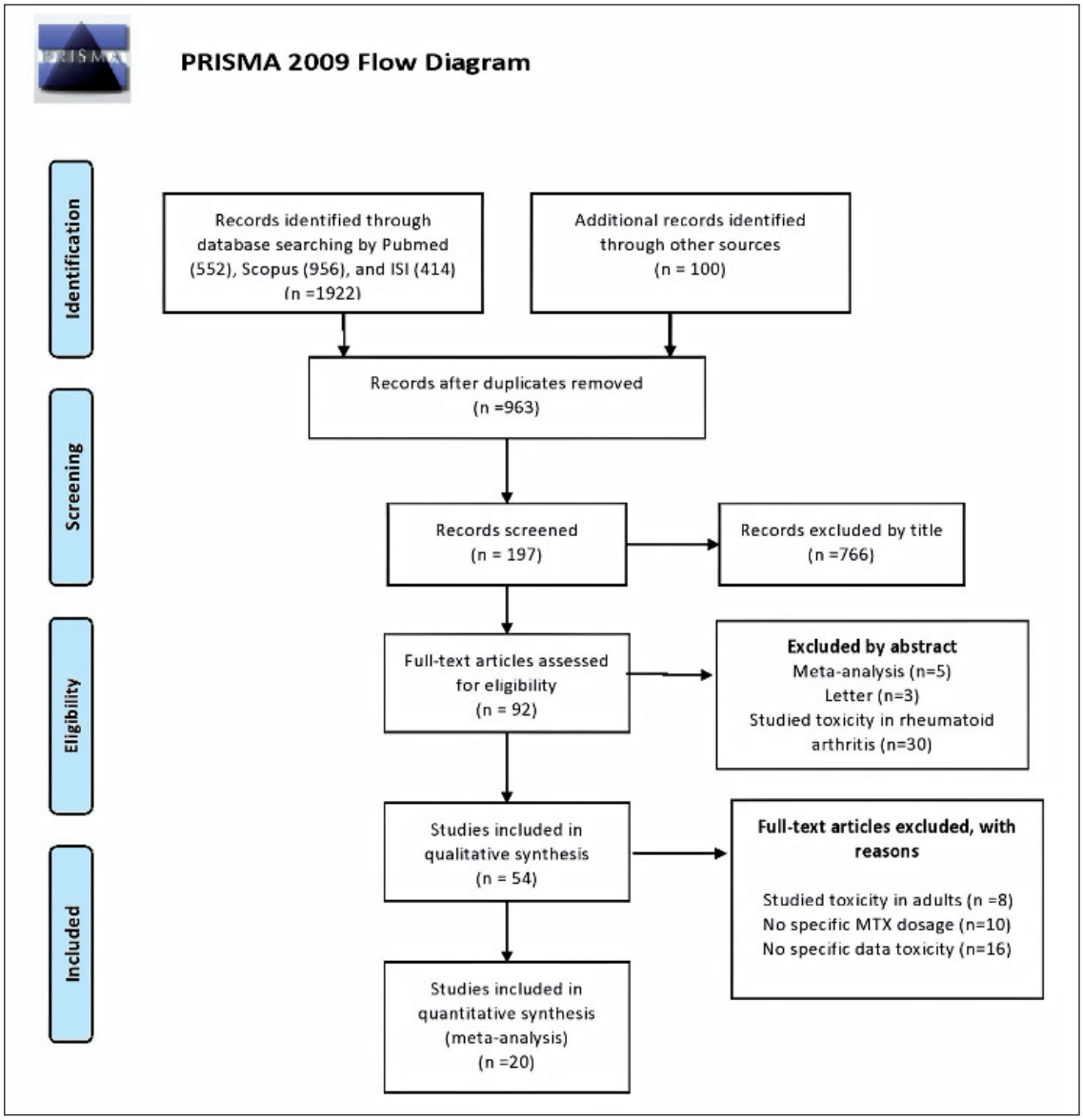

Fig. 1. Flow diagram of the study selection process.

TABLE 1. Characteristics of included studies.

Study ID	Study Design	Country	Malignancy	Mean Age (year)	Population			Toxicity							
					All	Male	Female	Hematologic	Hepatic	Mucositis	Renal	Dermal	Neuro	GI	Combined
D'Angelo et al[16]	Cohort	Italy	ALL	5	151	103	48	44	-	-	-	-	-	-	29
Erčulj et al[27]	Cohort	Slovenia	ALL	6.9	167	80	87	-	13	17	9	20	17	62	-
D' Angelo et al[17]	C.C	Italy	NHL	8.1	95	72	23	37	-	-	-	-	-	-	29
Li et al[28]	C.S	China	ALL	5.8	280	170	110	78	72	-	52	16	-	74	-
Shimasaki et al[34]	C.S	Japan	ALL	6.75	15	6	9	-	7	3	-	-	-	-	-
Kishi et al[19]	C.S	USA	ALL	7.5	53	23	30	-	-	-	-	-	9	-	-
Imanishi et al[20]	C.S	Japan	ALL/ML	6.7	26	16	10	-	5	-	-	-	-	-	-
Liu et al[21]	C.S	China	ALL	5.7	181	115	66	-	21	13	2	12	4	-	-
Park et al[31]	C.S	Korea	Osteosarcoma	11.6	37	21	17	-	35	35	35	-	35	-	-
Razali et al[29]	C.S	Malaysia	ALL	3.8	38	22	16	-	-	1	2	-	-	-	-
Hegyi et al[32]	C.S	Hungary	Osteosarcoma	13.6	59	31	28	59	59	-	-	-	-	-	-
Esmaili et al[22]	C.S	Iran	ALL	6.5	74	46	28	74	74	-	-	-	-	-	-
Kotnik et al[23]	C.S	Slovenia	ALL	5	64	41	59	-	-	19	15	-	-	-	-
Kotnik et al[24]	C.S	Slovenia	ALL/NHL	6.5	88	41	47	-	-	15	-	-	7	-	-
Kotur et al[25]	C.C	Serbia	ALL	7.3	148	94	54	-	130	130	130	130	130	130	
Xue et al[26]	C.S	China	ALL	5	125	71	54	-	41	-	-	-	-	-	-
Lambrecht et al[33]	C.S	Belgium	ALL	13.2	48	23	25	-	48	41	-	-	-	-	-
Liu et al[35]	C.S	China	ALL	6.16	112	59	53	-	15	64	8	-	-	-	-
Yousef et al[18]	C.S	Germany	ALL	7.7	64	31	33	44	-	-	-	-	-	-	-
Yazıcıoğlu et al[30]	C.S	Turkey	ALL	-	106	67	39	-	5	-	12	-	-	-	-

Abbreviations: **C.C**: case control; **C.S**: cross sectional; **GI**: gastrointestinal; **DHFR**: Dihydrofolate reductase; **RCF1**: Reduced folate carrier 1; **ALL**: acute lymphoid leukemia; **MTX**: Methotrexate; **ABCC2**: drug transporter Protein; **ARID5B**: interactive domain-containing protein 5B; Sig: significant; diff: differences; **NHL**: non- Hodgkin lymphoma; **FGR**: folate-related gene; **ML**: malignant lymphoma; **LBL**: lymphoblastic lymphoma; **USA**: united states; **TPMT**: thiopurine methyltransferase.

The most frequent types of toxicity include hepatic toxicity (n=10), mucositis (n=10), and renal toxicity (n=9) (Figure 2); however, combined toxicity was just reported in two studies.

The results of Cochrane Q and I^2 statistics showed significant heterogeneity in all types of toxicity, so estimations of toxicities prevalence were obtained using a random effect model. The summary estimation showed that the significant prevalence of various types of toxicity except the dermatologic toxicity (p=0.28 95% CI (-0.04, 0.60)) in children with different malignancies who treated with MTX ($p < 0.05$) (Figure 3).

Gl toxicity has higher prevalence (p= 0.51, 95% CI (0.10, 0.91)) compared to other toxicities, follow by hematologic toxicity (p= 0.41, 95% CI (0.25, 0.56)) and mucositis (p= 0.41, 95% CI (0.17, 0.65)). Whereas renal toxicity (p= 0.32 [95%CI (0.12, 0.52)) showed the lowest prevalence (Table 2).

Considering the type of malignancies as an essential variable that may cause between studies heterogeneity, the result showed that ALL was the more reported malignancy. Moreover, hepatic toxicity, mucositis, renal toxicity, and neurotoxicity were highly reported in osteosarcoma. However, hematologic toxicity was higher in ALL (p=0.41 [95% CI (0.21, 0.61)]) than other malignancies (Table 3).

Considering the dose of MTX as a sub-group, the results showed a significant prevalence of all toxicity (except the hematologic toxicity) in different dosages. Also, the dose of 12 g/m^2 showed higher toxicity than other dosages (Table 4).

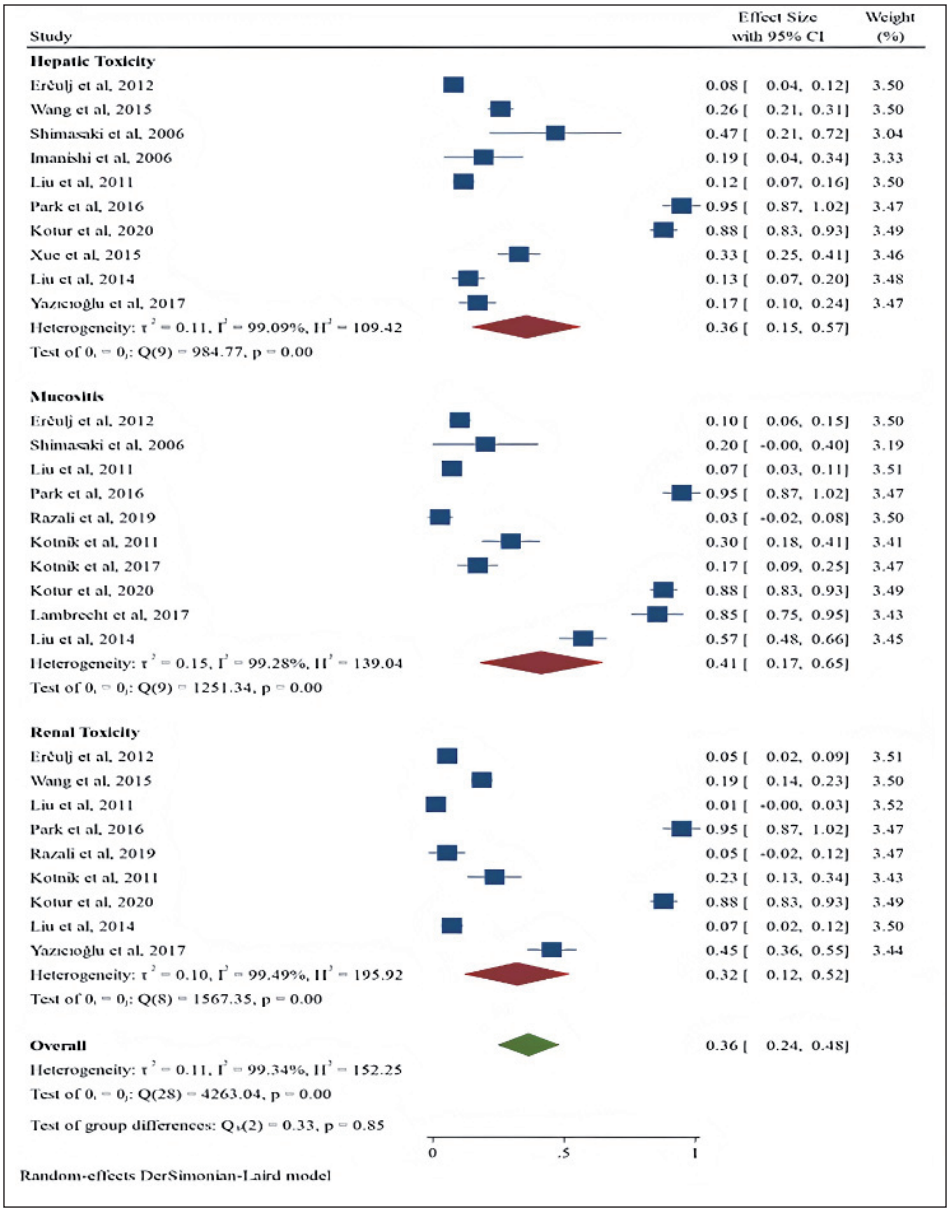

Fig. 2. Forest plot comparing the prevalence of various methotrexate-induced toxicities (most reported toxicities).

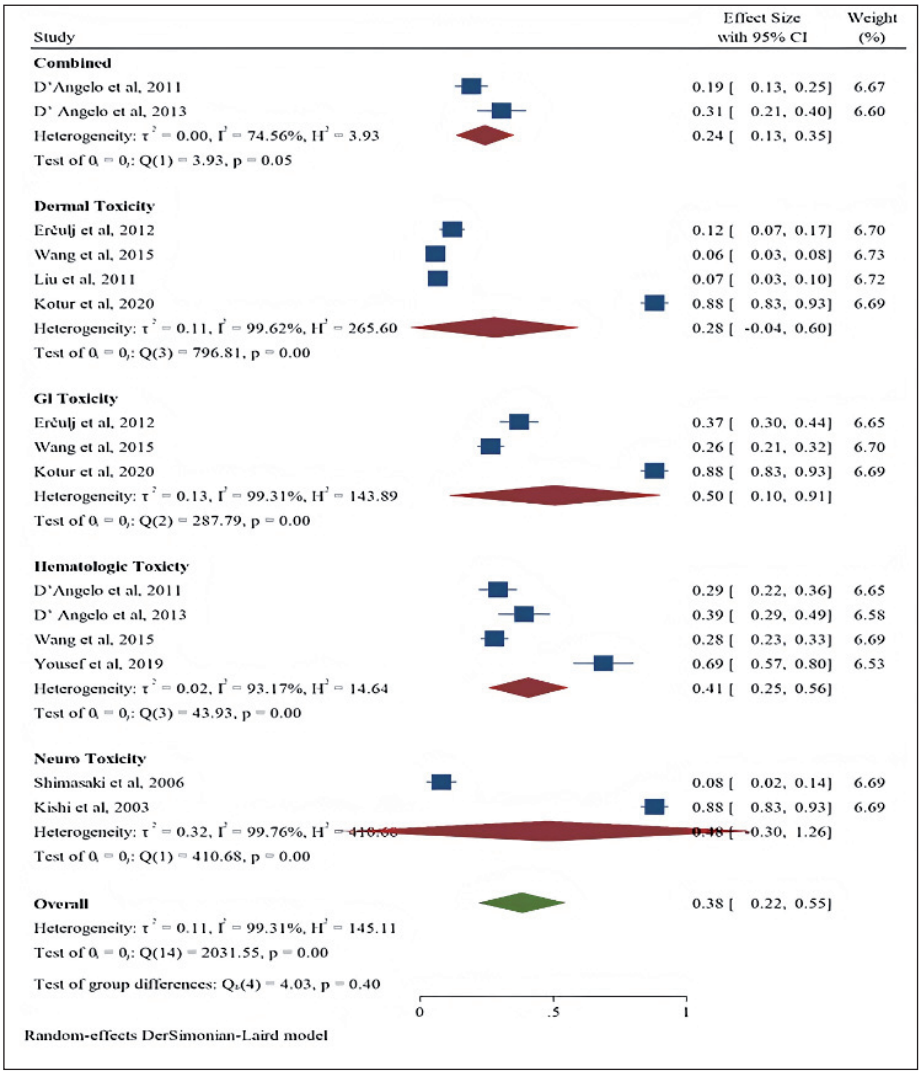

Fig. 3. Forest plot comparing the prevalence of various methotrexate-induced toxicities (less reported toxicities).

TABLE 2. Overall prevalence of various types of toxicity.

Response	*N*	*OR (95% CI)*	*I2*	*Chi-square (p-value)*	*Egger test (p-value)*
Hematologic Toxicity	4	0.41 (0.25, 0.56)	93	43.93 ($p < 0.001$)	2.31 ($p = 0.15$)
Hepatic Toxicity	10	0.36(0.15, 0.57)	99	984.8 ($p < 0.001$)	0.57 ($p = 0.59$)
Mucositis	10	0.41 (0.17, 0.65)	99	125.34 ($p < 0.001$)	1.16 ($p = 0.28$)
Renal Toxicity	9	0.32 (0.12, 0.52)	99	157.35 ($p < 0.001$)	2.16 ($p = 0.068$)
Dermal Toxicity	4	0.28 (-0.04, 0.60)	99	796.81 ($p < 0.001$)	1.38 ($p = 0.30$)
Neuro Toxicity	6	0.37 (0.05, 0.69)	99	133.04 ($p < 0.001$)	1.53 (p = 0.20)
Gl Toxicity	3	0.51 (0.10, 0.91)	99	287.79 ($p < 0.001$)	0.23 ($p = 0.86$)
Combined	2	0.24 (0.13, 0.3	74	3.93 ($p = 0.047$)	

Abbreviations: GI: Gastrointestinal.

TABLE 3. Sub-group analysis based on the type of malignancies.

Response	*Dose*	*N*	*OR (95% CI)*	*I²*	*Chi-square p-value*	*p-value*
Hematologic Toxicity	ALL	3	**0.41 (0.21, 0.61)**	95	42.83 (p < 0.001)	0.84
	NHL	1	**0.39 (0.29, 0.49)**	--	--	
Hepatic Toxicity	ALL	8	**0.3 (0.09, 0.51)**	99	670.06 (p < 0.001)	< 0.001
	ALL/AML	1	**0.19 (0.41, 0.34)**	--	----	
	Osteosarcoma	1	**0.95 (0.87, 1.02)**	--	----	
Mucositis	ALL	8	**0.38 (0.12, 0.63)**	99	932.87 (p < 0.001)	< 0.001
	ALL/ NHL	1	**0.17 (0.09, 0.25)**	--	----	
	Osteosarcoma	1	**0.95 (0.87, 1.02)**	--	----	
Renal Toxicity	ALL	8	**0.24 (0.06, 0.43)**	99	1050 (p < 0.001)	< 0.001
	Osteosarcoma	1	**0.95 (0.87, 1.02)**	--	----	
Dermatologic Toxicity	ALL	4	0.28 (-0.04, 0.60)	99	796.91 (p < 0.001)	----
	ALL	4	0.29 (-0.09, 0.68)	99	84,61 (p < 0.001)	< 0.001
	Osteosarcoma	1	**0.95 (0.87, 1.02)**	--	----	
Gl Toxicity	ALL	3	**0.51 (0.10, 0.91)**	99	287.79 (p < 0.001)	----
Combined	ALL	1	**0.19 (0.13, 0.26)**	--	----	----
	NHL	1	**0.31 (0.21, 0.40)**	--	----	

Abbreviations: **GI**: Gastrointestinal; **ALL**: acute lymphoblastic leukemia; **NHL**: Non-Hodgkin lymphoma; **HL**: Hodgkin lymphoma; **AML**: acute myeloblastic leukemia; **N**: number. *Test of group difference between 4 mg and 8 mg of TCZ.

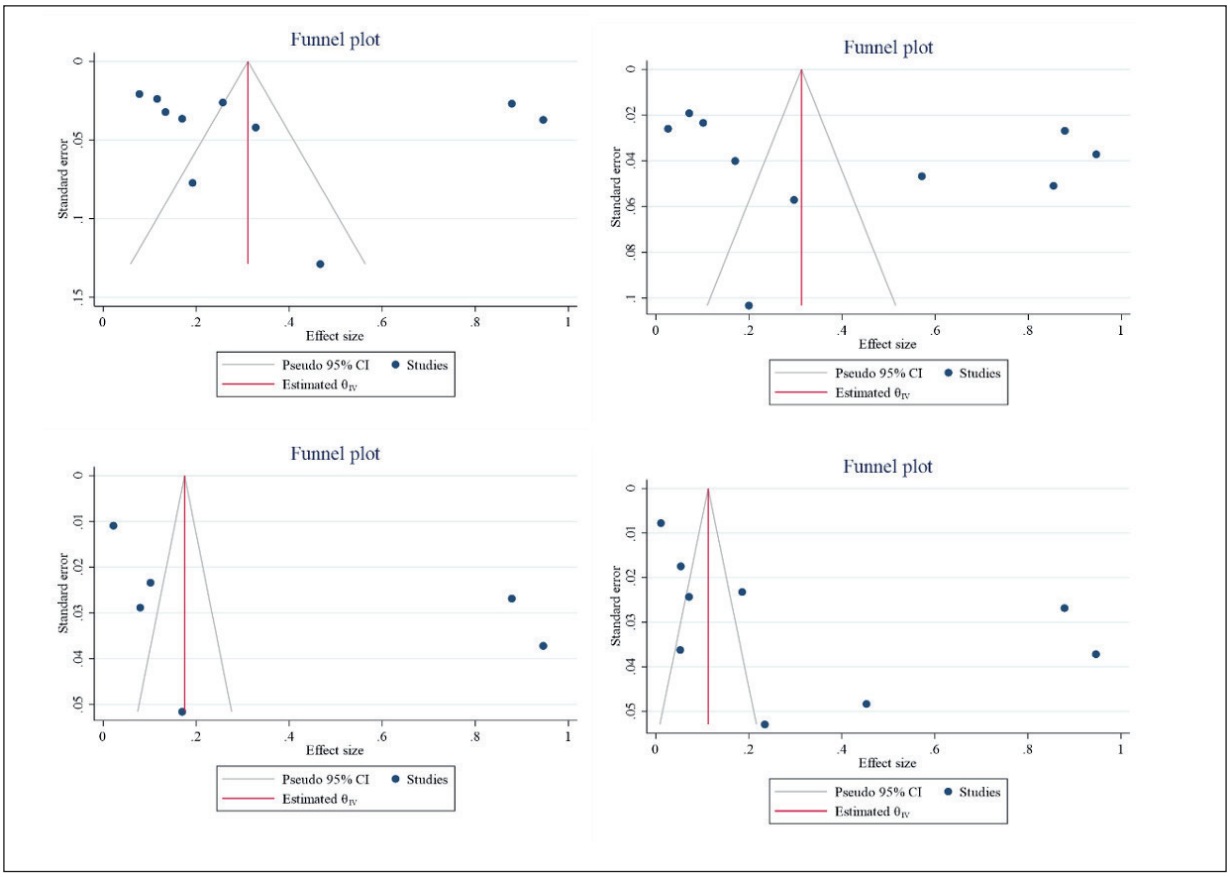

Fig. 4. Funnel plot of the prevalence of metformin hepatic toxicity (**2A**), mucositis (**2B**), neurotoxicity (**2C**), and renal toxicity (**2D**) in children with different types of MTX-induced malignancies.

TABLE 4. Sub-group analysis based on the type of malignancies.

Response	Dose	N	OR (95% CI)	I^2	Chi-square p-value	p-value
Hematologic Toxicity	2.5	--	--	--	--	0.13
	2.5+5	3	**0.45 (0.23, 0.67)**	93	33.29 (p < 0.001)	
	5	1	**0.28 (0.23, 0.33)**	--	--	
	12	--	--	--	--	
Hepatic Toxicity	2.5	1	**0.47 (0.21, 0.72)**	--	----	< 0.001
	2.5+5	4	0.38 (-0.05, 081)	99	467.62 (p < 0.001)	
	5	4	**0.16 (0.07, 0.25)**	89	29.46 (p < 0.001)	
	12	1	**0.95 (0.87, 1.02)**	--	----	
Mucositis	2.5	1	0.20 (-0.0, 0.40)	--	----	< 0.001
	2.5+5	4	**0.38 (-0.08, 0.79)**	99	613.25 (p < 0.001)	
	5	3	0.23 (-0.02, 0.48)	98	29.46 (p < 0.001)	
	12	2	**0.91 (0.82, 1.00)**	52	2.12 (p =0.15)	
Renal Toxicity	2.5	--	--	--	--	< 0.001
	2.5+5	3	0.37 (-0.25, 1.0)	99	9701 (p < 0.001)	
	5	5	**0.16 (0.06, 0.26)**	94	7622 (p < 0.001)	
	12	1	**0.95 (0.87, 1.02)**	--	----	
Dermatologic Toxicity	2.5	--	--	--	--	0.034
	2.5+5	2	0.47 (-0.32, 1.27)	99	619.91 (p < 0.001)	
	5	2	**0.08 (0.02, 0.15)**	79	4.76 (p = 0.03)	
	12	--	--	--	--	
Neurotoxicity	2.5	--	--	--	--	< 0.001
	2.5+5	4	0.29 (-0.13, 0.70)	99	876.4 (p < 0.001)	
	5	1	**0.10 (0.06, 0.15)**	--	----	
	12	1	**0.95 (0.87, 1.02)**	--	----	
Gl Toxicity	2.5	--	--	--	--	< 0.001
	2.5+5	1	**0.88 (0.83, 0.93)**	--	----	
	5	2	0.31 (0.21, 0.42)	81	5.74 (p = 0.02)	
	12	--	--	--	--	
Combined		--	--	--	--	----
	2.5+5	2	**0.24 (0.13, 0.35)**	74	3.93 (p =0.047)	
	5	--	--	--	--	
	12	--	--	--	--	

Abbreviations: GI: gastrointestinal toxicity.

Quality Assessment

For assessing the quality of included studies, the NEW-CASSTLE OTTAWA was used. The maximum scores were 10 for case-control and cross-sectional studies and 9 for cohort studies. The maximum score was 9 in five studies (supplemental 1)[18,22,25,26,33]. In selection criteria, outcome and exposure sections were received an acceptable rating (***Supplementary Figure 1***).

Risk of Bias

The funnel plots give us a visual observation of the publication bias of four comparisons, including hepatic toxicity (Figure 2A), mucositis (Figure 2B), neurotoxicity (Figure 2C), and renal toxicity (Figure 2D). Obviously, five non-symmetrical figures showed that the publication bias is considerable. The funnel plot was useless for other comparisons, because less than 5 studies were included.

DISCUSSION

MTX is used widely in treating malignancies such as ALL, Hodgkin lymphoma (HL), NHL, and rheumatoid arthritis (RA) in both adults and pediatrics. MTX acts as an anti-folate agent, which reduces cell proliferation by disrupting purine and pyrimidine synthesis. To address this, Leucovorin is administered to reduce the toxic consequences of MTX[36,37]. The literature has emerged offering contradictory findings about the influence of Leucovorin in relapse risk and treatment outcome[36,38,39]. In this regard, the investigation of factors that have an impact on toxicity development to increase free-survival rate is essential. Most studies have carried out on the risk factors such as folate-related gene polymor-

phisms that induce the MTX-related toxicities, but much less is known about the association between specific doses of MTX and toxicities. The present systematic review attempts to show the association between toxicity and dose of MTX. Our pooled analysis showed considerable toxicity is associated with MTX in children with different malignancies, of which hepatotoxicity, hematological toxicity, GI toxicity, and mucositis are the most frequent types.

GI toxicity sometimes is restricting treatment, especially in patients with malignancy. In treatment with MTX, the healthy tissues with high turnover, including bone marrow, gastrointestinal tract, and oral mucosa, are indirectly affected[40]. This supports our result that GI toxicity, hematological toxicity, and mucositis were the most MTX-associated toxicities. Additional to the inhibition of cell proliferation, increased level of apoptosis is another proposed mechanism of MTX-associated toxicities. In animal model studies that set out to determine the effect of MTX on the gastrointestinal tract, it was shown that MTX by activating caspase 9, 2 and 3 in intestinal epithelial cells increases the level of apoptosis and subsequently promotes GI toxicity development[41,42].

Also, a pooled analysis showed that dermatologic toxicity was the rare MTX-associated toxicities. There is little published data on the association between MTX and dermatologic toxicity in patients with malignancies. In this context, Kataria *et al*[43] reported a rare case of toxic epidermal necrolysis (TEN) in a young adult with ALL following the use of high-dose MTX. Scheinfeld[44] also reported three cases of toxic skin eruptions associated with MTX. Accordingly, Song *et al*[45] analyzed skin lesions following low-dose long-term treatment with MTX in an adult patient with non-Hodgkin's lymphoma. There was just a little investigation about HD-MTX and dermatologic toxicity in psoriasis patients[46]. Given the mentioned studies, all reported a rare case of dermatologic toxicity in a small number of patients and various cutaneous toxic consequences, particularly when administered in high doses of MTX.

Next to GI toxicity, hematological toxicity is one of the most reported MTX-related toxicity. A possible explanation for this is that MTX, by regulating Wnt/β-catenin pathway signaling, inducing the differentiation of mesenchymal stem to adipocyte[47], causes hematological toxicity. Whereas it was demonstrated MTX-based regimen is accompanied by lower hematological toxicity[48]. Missense mutations in Methylenetetrahydrofolate reductase (MTHFR)(C677T and A1298C) are associated with lower enzyme activity (18,49), following a decrease of enzyme activity. MTX accumulates in the cell and triggers the treatment-related toxicities. It was demonstrated that pediatrics with 677T genotype had an approximately six-fold greater risk for developing hematological toxicity[17].

Further analysis showed that mucositis is also among the frequent MTX-induced toxicity. In this regard, previous researches reported mucositis as a prevalent MTX-induced toxicity due to high epithelial cell division[50]. The pathophysiology of mucositis is complex, but DNA damaging agents such as chemotherapy and ionizing radiation induce the apoptotic transcription factors, including p-53, NF-κB, and Wnt[51], which lead to activating apoptosis and consequently can be accompanied with mucosal toxicity.

Among all toxicities, renal toxicity has the lowest prevalence due to rapid clearance by kidneys[52,53]; additionally, another explanation for this is that most MTX-related renal toxicity reported in patients who received concomitantly other drugs, including cisplatin, penicillin, non-steroidal anti-inflammatory drugs, and sulfonamides which reduce the tubular drug secretion[54]. This is in line with our result that renal toxicity has the lowest prevalence.

It was demonstrated that MTX-induced hepatotoxicity is a dose-dependent issue and 15-50% of patients with elevated aminotransferases who received mild-moderate MTX dose were self-limited with fibrosis and cirrhosis as the more important adverse consequence[49,50]. In this regard, it was shown that the incidence of hepatotoxicity is higher at the beginning of each MTX administration course and reduced after the use of folic acid[51,52]. Our pooled analysis revealed that hepatotoxicity is a common MTX-induced toxicity, inconsistent with other research[22,28,32]. In studies conducted by Kotur *et al* 25 and Esmaili *et al* 22, it was shown that hepatotoxicity is frequent MTX-induced toxicity and suggested that gene mutation in the SLC19A1 gene increases the chance of hepatotoxicity. Mutation in the SLC19A1 gene affects MTX transport, so the drug accumulates within the cell and increases hepatotoxicity[25,55].

One of the most important clinically relevant findings was that mucositis, renal, and neurotoxicity were dose-dependent; thus, these relationships may be proposed as the renal injury with the long-term exposure to MTX and drug-drug interaction following administration of the concomitant drug can cause the development of MTX-related toxicities[39-41].

LIMITATIONS

The major limitation of this study was the high risk of bias. One of the main reasons for selecting all types of studies includes case-control, cross-sectional, and cohort studies.

CONCLUSIONS

This study has shown that even in a standard dose of MTX, serious MTX-related toxicities can limit the treatment and delay the expected response. To address this problem, more investigations to replace the MTX with a new drug are required.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Availability of data and materials

The datasets generated and/or analyzed during the current study are available in the [Pubmed, Web of Science, Scopus, EM Base] repository.

Funding

None.

Authors' contributions

F.R. conceived the manuscript and revised it. S.Gh., A.A, and A.S. did the statistical analysis, wrote the manuscript, and prepared tables and figures.

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Conflict of Interest:

The authors declare no conflict of interest. All procedure performs in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or compare ethical strand.

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