High Dose Methotrexate Induced Toxicity in Children with Hematological Malignancy: Incidence and Relation to MTX Level

Sayed HA[1], Ali NHM[1], Shibl A[1]

[1] Pediatric Oncology Department, South Egypt Cancer Institute, Assiut University

Abstract:

Background: Methotrexate is an effective drug for the treatment of malignant hematopoietic neoplasm in children; its antineoplastic activity is enhanced when MTX is used at high dose. Monitoring of patient after high dose methotrexate administration is essential to manage toxicity. Aim of the work: Incidence of HDMTX toxicity in children with hematological malignancies and relation to MTX level.

Patients & Methods: A prospective study conducted on 142 patients with hematological malignancies who received 467 cycles of HDMTX scheduled in their treatment protocols during the period from May 2020 to September 2022, assessment of the patients before HDMTX administration and after the dose to detect any toxicities.

Results: GIT toxicity reported in 434 (92.9%) followed by renal toxicity in form of raised serum creatinine in 114 (24.4%), hepatic toxicity in 51 (10.9%) and myelosuppression in 48 (10.3%). Toxic MTX level at 23h & 66h significantly associated with increased creatinine level. In addition, Toxic MTX level at 23h significantly associated with all systems and GIT toxicities. MTX level at 42h didn't show any significant relations

Conclusion: GIT toxicities are the most common HDMTX toxicities and related to high 23 h MTX level. Both high 23h & 66 h MTX level are good indicators of renal toxicities.

Key words: Hematological malignancies, HDMTX, Pediatrics, Toxicity, MTX level

Received: 3 October 2024
Accepted: 20 November 2024

Authors Information:

Heba Abdel-Razik Sayed
Pediatric Oncology Department, South Egypt Cancer Institute, Assiut University
email: Hebadina2007@yahoo.com

Norhan Hassan Mahmoud Ali
Pediatric Oncology Department, South Egypt Cancer Institute, Assiut University
email: Norhanhassan042@gmail.com

Azza Shibl Sayed
Pediatric Oncology Department, South Egypt Cancer Institute, Assiut University
email: azzashibl@aun.edu.eg

Corresponding Author:
Norhan Hassan Mahmoud Ali
Pediatric Oncology Department, South Egypt Cancer Institute, Assiut University
email: Norhanhassan042@gmail.com

Introduction:

Hematological malignancies are the most frequent cancer in children. Leukemia represents approximately 28%, acute lymphoblastic leukemia (ALL) being the most common type of cancer in children while non-Hodgkin lymphoma (NHL) account for 9% and high dose methotrexate (HDMTX) is an effective drug in treatment of hematological malignancy in children (1).

Methotrexate (MTX) is a synthetic folate antimetabolite and cell cycle specific for S phase which inhibits DNA synthesis, repair, and cellular replication (2).

A methotrexate dose >500 mg/m2 is generally considered a high dose, in the pediatric oncology population, HDMTX is commonly used as a part of treatment protocols among patients with ALL, NHL, and osteosarcoma, with the dose ranging from 1 to 12 g/m2 (3).

HDMTX can cause significant toxicity, including oral mucositis, vomiting, myelosuppression hepatotoxicity, dermatological toxicity, acute kidney injury and other toxicities of HDMTX can lead to significant morbidity (3).

There are many risk factors responsible for high prevalence of toxicity such as methotrexate dose, methotrexate duration, low alkalinization, poor hydration and low rescue dose. Also, drugs that interfere with MTX for renal tubular excretion delay the MTX clearance resulting in renal toxicity (4).

The delay between recognition of toxicity and initiation of treatment can directly contribute to renal and systemic toxicities (5). Some studies concluded that the risks of HDMTX toxicities are associated with serum MTX concentrations (6,7). However, another

study indicated the opposite conclusions (8, 9). Therefore, whether monitoring of the serum concentrations after HDMTX is suitable as the predictor of severe adverse event occurrence is still controversial (10).

Our study aimed to study the incidence of HDMTX induced toxicity in children with hematological malignancies and its relation to serum MTX concentration.

Patients and Methods:

A prospective study conducted at Pediatric Oncology and Haematological Malignancies Department; South Egypt Cancer Institute (SECI) included children with haematological malignancies (142 patients who received 467 cycles of HDMTX infusion scheduled in their treatment protocols during the period from May 2020 to September 2022.

Ethical consideration

The study was approved by the local ethical committee of SECI, Assiut University. An informed written consent was taken from the patients' parents or guardians, before including in the study, after discussing with them the aims and the methods of the study. All other rules advised by the ethical committee were applied.

Study design

All patients aged ≤ 18 years, with haematological malignancies (ALL either de novo or relapsed and NHL) who received at least one HDMTX as part of their treatment protocols were included in the study. Patients with solid tumor and those with hematological malignancies who didn't receive HDMTX or Down syndrome patients with haematological malignancies were excluded.

Methods:

All patients were subjected to Full history taken include: demographic data, diagnosis, immunophenotyping, risk stratification, treatment protocol, treatment phase, HDMTX dose as 1st episode (1st HDMTX) or previous episode (who received previous HDMTX), history of documented risk factors comorbidity as (history of previous toxicity after previous HDMTX, history of chronic hepatic or renal disease and drug intake as salicylates, phenylbutazone, phenytoin and sulphonamide). Full clinical examination is done before each HDMTX administration, Baseline investigations done 24 hours before HDMTX infusion: include complete blood count, liver functions, renal functions, creatinine clearance (CRCL), Abdominal sonar to exclude 3rd space and to check for both kidneys, Chest X ray done ± CT scan is reserved for patients known to have fungal infections to exclude active disease or space effusion. Renal scan done in patients with inadequate CRCL or previous renal toxicity or delayed excretion or in young patient who couldn't collect urine for CRCL.

During HDMTX administration: concurrent nephrotoxic medications was avoided with hyper-hydration before dose on dose 150 ml / m2, Alkalinization of urine was performed by adding sodium bicarbonate (NaHCO3) on a dose of 40 mEq/L , urine pH of not less than 7 should be required before administration of HDMTX and follow up of urine PH with each void. When urine pH of 6.5 is identified, NaHCO3 at a dose of 12.5 mEq/m2 is administered, and for urine pH < 6.5, a dose of 25 mEq/m2 is given till correction of PH of urine.

Dose and rate of HDMTX infusion was administrated according to treatment protocol, patients with ALL treated according to Total XV protocol (11), Total XIII protocol (12) with some modifications at our center (13) ., those with ALL relapse treated according to R16 protocol (14) with some modifications at our center (15) , Patients with NHL treated according to BFM 95 protocols (16) with modifications done to HDMTX from 5gm/m2 to 3gm/m2 in R3&R4 patients and rate of infusions to 8 hours in R4 and 4 hours in R3. Patients are categorized into two groups; ALL group; in which HDMTX is received only with purinthol including ALL, ALL relapse & lymphoblastic lymphoma (LBL) and NHL group; in which HDMTX is usually received among other chemotherapeutic agents. Also, high risk patients of each group are added together as they received more intensive doses when compared with lower/ standard risk patients of each group.

Plasma MTX level was drawn at the end of infusion (at 23h from the start of infusion) in case of 24 hours infusion and at 42 hours from the start of HDMTX for all patients then daily till until MTX level become not detected (ND), the sample are analyzed at drug monitoring laboratory at our SECI.

Measures and recommended Ca leucovorin doses and timing are given according to MTX level and to each protocol shown in supplementary tables (a&b).

Patient monitored clinically and laboratory during and after the dose administration until MTX level become not detected (ND) for the occurrence of any toxicity, Toxicity was graded based on Common Terminology Adverse Effect (CTCAE) version 5 (17). Relation of HDMTX different toxicities with MTX level is analyzed. Each MTX level at 23 hours, 42 hours& 66 hours from the start of HDMTX is considered toxic if reach ≥150 μmol/l, ≥1 μmol/l & > 0.15 respectively (10, 11, 18).

Statistical analysis:

All statistical calculations were done using SPSS (statistical package for the social science; SPSS Inc., Chicago, IL, USA) version 22. Data were statistically described in terms of mean ± standard deviation (±SD), or median and range when not normally distributed, frequencies (number of cases) and relative frequencies (percentages) when appropriate. Comparison of quantitative variables between MTX toxicity groups was done using an independent t-test. For comparing categorical data, Chi square ($\chi 2$) test was performed. P-value is always 2 tailed set significant at 0.05 level.

Results:

This prospective study included 142 patients with hematological malignancies who received (467 cycles) of HDMTX infusion treated at Pediatric Oncology & Hematological Malignancies Department at SECI, Assiut university. Criteria of the study group shown in table 1. The median age of the enrolled patients at diagnosis was 5.5 years. Most of them (57%) were males. Among the ALL group,110 patients were ALL, 4 patients were ALL relapse and only one patient with LBL. History of risk factors was positive in 36 patients: 12 patients with viral hepatitis, 10 patients with previous history of HDMTX toxicity, 13 patients with history of drug intake and one patient with chronic renal disease.

Table 1. Characteristics of the studied patients with hematological malignancies (142 patients) who received HDMTX:

Variable	N. (%)
Age (years)	
(mean ± SD)	7.23 ± 4.25
Median	5.5 (1.7-17)
Age group (years)	
< 10	103 (72.5%)
≥ 10	39 (27.5%)
Sex	
Male	81 (57%)
Female	61 (43%)
M: F ratio	3: 4
Diagnosis	
ALL group (ALL, ALL relapse, LBL)	115 (81%)
NHL group	27 (19%)
IPT	
B -cell	119 (83.8%)
T -cell	23 (16.2%)
Risk stratification	
HR (ALL: HR + NHL: R3 and R4)	53 (37.3%)
LR/SR (ALL:SR and LR + NHL: R2)	89 (62.7%)
Treatment protocol	
ALL	
TOTAL XV	99 (69.7%)
TOTAL XIII	12 (7.7%)
R16	4 (2.8%)
NHL-BFM-95	27 (19%)

Data expressed as mean (SD), frequency (percentage). ALL: acute lymphoblastic leukemia; NHL: non-Hodgkin lymphoma, HR: high risk, LR; low risk, SR; standard risk, BFM; Berlin-Frankfurt-Munster, IPT; immunophenotyping, M: male, F: female, LBL: lymphoblastic lymphoma

Baseline laboratory investigation of studied patients (142) before HDMTX administration (467 cycles)

Mean TLC and neutrophil were 3.52 and 1.74(103/ul) respectively, mean HB is 10.76 mg/dl with range (7.6-15.4), mean platelet count was 241(46-964) (103/ul), mean ALT 53.35 U/L with range (12-455), and mean AST 52.92 U/L with range (6-240), mean serum creatinine is 0.34 mg/dl with range (0.10-0.90).

HDMTX infusion and monitoring criteria in each cycle (n= 467 cycle):

Median number of HDMTX cycles received by patients was 2 with range between 1-9 cycles, with 125 cycles (26.8%) of them were the 1st dose (1st episode).

Although different treatment protocols, most of patients received HDMTX at dose of 5 gm/m2 (309 cycles) (66.2%), in the remaining 158 (33.85) received less than 5 gm/m2. All Patients with NHL received HDMTX in a dose of less than 5 gm/m2 except in 5 cycles and all of them received HDMTX in a rate of less than 24 hours (31 cycles over 4 hours& 42 cycles over 8 hours) except in 3 cycles received over 24h-infusion. Table 2 shows HDMTX infusion criteria in each cycle.

Table 2. High dose methotrexate infusion criteria in each cycle

Criteria	N (467)
Number of cycles	
Median (range)	2 (1-9)
History of risk factor	
Yes	163 (34.9%)
No	304 (65.1%)
Episode	
1st episode	125 (26.8%)
Previous episode	342 (73.2%)
Cumulative dose (gm / m2)	
Mean ± SD	10.48 ± 6.68
Median (range)	10 (1- 45)
HDMTX dose	
< 5gm/m2	158 (33.8%)
5gm/m2	309 (66.2%)
HDMTX infusion	
24 hours	358 (76.7%)
< 24 hours	109 (23.3%)
MTX level at 23-h* (n = 358 cycles) (umol/ L)	
mean ± SD	67.97 ± 37.69
≥ 150 µmol/l (%)	10 (2.8%)
< 150 µmol/l (%)	348 (97.2%)
MTX level at 42-h (umol/ L)	
mean ± SD	1.86 ± 0.28
≥ 1 µmol/l (%)	209 (44.8%)
< 1 µmol/l (%)	258 (55.2%)
MTX level at 66-h (umol/ L) (400 cycles)	
mean ± SD	0.53±3.3
≥ 0.15 µmol/l (%)	223(47.8%)
< 0.15 µmol/l (%)	177(37.9%)
Time of MTX clearance (days)	
Median (range)	3 (2- 7)
≤ 3 days	288 (61.7%)
> 3 days	179 (38.3%)

Data expressed as mean (SD), range, frequency (percentage). MTX: methotrexate
HDMTX: high dose methotrexate hr: hour

HDMTX induced toxicity in each cycle among the study group according CTCAE (n= 467 cycles):

We reported mild to moderate toxicities in 275/ 467 (58.9%) infusions. Within the median time of 3 days (2-7) till non detected MTX level, we reported GIT

toxicities in 434 cycles (92.9%), neutropenia ± fever in 48 cycles (10.3%), hepatic toxicities in 51 cycles (10.9%) and renal toxicities in 114 cycles (24.4%). Significant anemia & thrombocytopenia (G3) was observed in only one cycle each. No other toxicity was observed. No lethal toxicity was reported among our patients. Grades of each system toxicity are shown in table 3.

When studying the frequency of HDMTX induced toxicity as regard the diagnosis groups, we found that GIT toxicities (G2 and G3) were significantly (P > 0.001) higher among NHL group compared with those of ALL group, while G1 were more among ALL group. Although neutropenia was not a prominent toxicity, grade 2 and 3 neutropenia were significantly higher (P > 0.001) in NHL group vs ALL group. No significant difference considering hepatic and renal toxicities were reported between the two groups as shown in table 4.

Relation of methotrexate level with toxicity

We studied the relation of MTX level at 23h, 42h & 66h after the start of HDMTX to G3 or more of all toxicities and of GIT toxicities but due to the small frequencies of neutropenia, renal &hepatic toxicities, we studied the relation of all grades with the MTX level.

MTX level at 23h ≥ 150 umol/l shown to be significantly associated with high incidence of all systems, GIT and renal toxicity (P=< 0.001in each of them). On the other hand, MTX level at 42h ≥ 1 umol/l didn't show any associated toxicities and MTX level at 66 h showed a significant relation to the renal toxicities (P=0.040) as shown in Table 5

Discussion:

Here, we studied 142 patients with hematological malignancies who received 467 cycles of HDMTX prospectively to detect the pattern of HDMTX toxicity in these groups of patients and their relation to MEX level.

We reported mild to moderate toxicities in 275/ 467 (58.9%) infusions with no lethal toxicity reported among our patients. The most common HDMTX induced toxicities in this study was GIT toxicity that reported in 92.9% of cycles. Although that HDMTX having low emetic risk as classified by American Society of Clinical and Oncology guideline for antiemetic therapy (19), emesis was the main toxicity in this study reported in nearly half of the cycles followed by oral mucositis in approximately 40%. Sari et al.,2021 reported mucositis only in 3.4% in their study group (20), Li et al.,2019 reported oral mucositis and vomiting in 85 (21.9%) and 40 (10.3%) cycles (10) and Shan et al.,2020 in another study reported occurrence of mucositis in 56 (18%) infusions with grade 1 (38) and grade II (18) (21). These studies demonstrated the toxicities in ALL patients that may attribute to the lower incidence than our study. Vaishnavi et al., 2018, who studied the toxicities among hematologic malignancies reported similar pattern of toxicity to our results with vomiting and mucositis being the most common toxicity in their patients reported in 53% and 32% respectively (22).

Renal toxicity was the second most common toxicity reported in 24.4% of cycles in our study. In a retrospective study done on mature B/T NHL patients received 3 gm/m2 HDMTX over 3 hours in which delayed clearance and raised creatinine occurred in 71(36.2%) doses and oral mucositis in 17(8.7%) doses associated with delayed clearance (23). A more recently study stated that nephrotoxicity reported in 119 courses (23.2%), including 85 grade 1 (16.5%), 25 grade 2 (4.9%), and 9 grade 3 (1.8%) (19).

Table 3. High dose Methotrexate induced toxicity in the studied patients in each cycle according to CTCAE (15).

		Grade of toxicity			
	Toxicities (%)	G1	G2	G3	G4 and G5
GIT toxicity	434 (92.9%)				
Vomiting	223 (47.8%)	165 (35.3%)	36 (7.7%)	22 (4.7%)	0
Oral mucositis	186 (39.8%)	105 (22.5%)	49 (10.5%)	32 (6.9%)	0
Diarrhea	25 (5.4%)	16 (3.4%)	6 (1.3%)	3 (0.6%)	0
Neutropenia	48 (10.3%)	8 (1.7%)	23 (4.9%)	17 (3.6%)	0
Hepatoxicity	51 (10.9%)				0
Jaundice (increase bilirubin)	2 (0.42%)	1 (0.20%)	1 (0.20%)	0	0
Raised transaminases	49 (10.5%)	31 (6.6%)	9 (1.9%)	9 (1.9%)	0
Renal toxicity (increase creatinine level)	114 (24.4%)	88 (19.1%)	23 (4.9%)	2	0

Table 4. High dose methotrexate induced toxicity among the studied patients regarding the diagnosis

Grades of toxicity	ALL group	NHL group	total	P value
GIT TOXICITY				< 0.001
Grade 0	31 (7.9%)	2 (2.6%)	33 (7.1%)	
Grade I	251 (64.2%)	35 (46.1%)	286 (61.2%)	
Grade II	66 (16.9%)	25 (32.9%)	91 (19.5%)	
Grade III	43 (11%)	14 (18.4%)	57 (12.2%)	
Total	391 (100.0%)	76 (100.0%)	467 (100%)	
Neutropenia				< 0.001
Grade 0	361 (92.3%)	58 (76.3%)	419 (89.7%)	
Grade I	8 (2%)	0(0.0%)	8 (1.7%)	
Grade II	14 (3.6%)	9 (11.8%)	23 (4.9%)	
Grade III	8 (2%)	9 (11.8%)	17 (3.6%)	
Total	391 (100%)	76 (100%)	467 (100%)	
Hepatic toxicity				0.139
Grade 0	354 (90.5%)	62 (81.6%)	416 (89.1%)	
Grade I	23 (5.9%)	9 (11.8%)	32 (62.7%)	
Grade II	7 (1.8%)	3 (3.9%)	10 (19.6%)	
Grade III	7 (1.8%)	2 (2.6%)	9 (17.6%)	
Total	391 (7.9%)	76 (2.9%)	467 (10.9%)	
Renal toxicity				0.092
Grade0	290 (74.2%)	63 (82.9%)	353 (75.6%)	
Grade I	80 (20.5%)	9 (11.8%)	89 (19.1%)	
Grade II	19 (4.9%)	4 (5.3%)	23 (4.9%)	
Grade III	2 (0.5%)	0 (0%)	2 (0.4)	
Total	391 (100%)	76 (2.8%)	467 (100%)	

ALL: acute lymphoblastic leukemia, LBL: lymphoblastic lymphoma, NHL: non-Hodgkin lymphoma

Table 5. Relation between Clinical toxicities and serum methotrexate level

toxicities	MTX level at 23h (n=358)*		P- value	MTX level at 42h (all patients)		P-value	MTX level at 66h (n= 400)**		P-value
	≥ 150umol/l	<150umol/l		≥ 1umol/l	<1umol/l		< 0.15umol/l	≥ 0.15umol/l	
All systems			0.001			0.281			0.065
-yes	5(50%)	46(13.2%)		36(17.2%)	35(13.6%)		20(11.3%)	40 (17.9%)	
-no	5(50%)	302(86.8%)		173(82.8%)	223(86.4%)		157(88.7%)	183(82.1%)	
GIT			<0.001			0.207			0.053
-yes	5(50%)	35(10.1%)		30(14.4%)	27(10.5%)		15(8.5%)	33(14.8%)	
-no	5(50%)	313(89.9%)		179(85.6%)	231(89.5%)		162(91.5%)	190(85.2%)	
Neutropenia			0.190			0.170			0.435
-yes	2(20%)	30(8.1%)		17(8.1%)	31(12%)		19(10.7%)	158(89.3%)	
-no	8(80%)	340(91.9%)		192(91.9%	227(88%)		19(8.5%)	204(91.5%)	
Hepatic			0.995			0.399			0.962
-yes	1(10%)	35(10.1%)		20(9.6%)	31(12%)		18(10.2%)	23(10.3%)	
-no	9(90%)	313(89.9%)		189(90.4%)	227(88%)		159(89.8%)	200(89.7%)	
Renal			<0.001			0.203			0.040
-yes	10(100%)	83((23.9%)		57(27.3%)	57(22.1%)		35(19.8%)	64(28.7%)	
-no	0(0%)	265(76.1%)		152(72.7%)	200(77.5%)		142(80.2%)	159(71.3%)	

Data expressed as frequency (percentage). GIT: gastrointestinal tract; MTX: methotrexate.
*MTX level at 23-h after start of HDMTX in case of 24-hour infusion (358 cycles)
**MTX level at 66-h after start of HDMTX was still detected in 400 cycles.

We reported relatively low incidence of neutropenia (10.3%) of cycles, grade-I in 8 (1.7%), grade-II in 23 (4.9%) and grade-III in 17 (3.6%) cycles. compared to Li et al.,2019 and Sari et al.,2021 (26.54% and 30.7%) respectively (10,20). Shan et al.,2020 reported similar incidence of neutropenia to our results (9%) with myelotoxicity in 12 infusions (4%), with grade II (10) and grade III (2) (21). The low incidence of neutropenia in our study can be related the short follow up of patients till non detected MTX level (within a median of 3 days only).

Hepatotoxicity also reported at low incidence (10.9%) compared to that reported by Sari et al,.2021 (32.2%) (20)

In our study, 23-h plasma MTX level and 66-h MTX level are associated with toxicities. Some studies have demonstrated that the risk of GIT toxicity in form of oral mucositis and hematological toxicity is associated with serum MTX concentrations (6,7). In contrary to our results, Li et al.,2019 in their study to analyze the relationship between MTX level and toxicities (hematological toxicities, transient liver function, vomiting and oral mucositis) they found that neutropenia ≥ 3, vomiting ≥ grade2 were more likely to occur after infusion had MTX level ≥ 1 umol/l at h 44 and other toxicities has no correlation with MTX level (10).Also in contrast to our result, patients with NHL reported severe grade (III/IV) of oral mucositis in 66 (40%) cycles after HDMTX and revealed that clinical and laboratory data and MTX level weren't predictors for these toxicities (48-72 hrs) (24). Also, the study of Sari et al,.2021 found no significant association between the clinical toxicity and MTX levels at the two points (24 and 48 hrs) of measurement (20). This difference may be different studied disease type in which polychemotherapy are received, different risk-intensity of chemotherapy, different protocols and leucovorin rescue.

Conclusion:

GIT toxicities are the most common HDMTX toxicities and related to high 23-h MTX level. Both high 23-h& 66- h MTX level are good indicators of renal toxicities.

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