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Full Length Research Paper

IL-8 and MCP-1 in children with acute lymphoblastic leukemia and potential correlation with neurotoxicity and thromboembolic phenomena

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Our purpose was to report the safety of methotrexate in children with acute lymphoblastic leukemia treated with the Berlin-Frankfurt-Münster-95 protocol. We also determined whether serial blood assessment of vitamin B12, folate, methotrexate, homocysteine, IL-8 and MCP-1 along with cerebrospinal fluid measurement of IL-8 and MCP-1 had any correlation with neurotoxicity and thromboembolic incidents. Out of 16 patients, 2 exhibited seizures, whereas 1 exhibited central retinal artery occlusion. Due to the small number of patients no robust conclusions could be drawn. More studies are needed so as to determine whether these biological parameters could be used as markers for toxicity.

Key words: acute lymphoblastic leukemia, children, neurotoxicity, thrombosis, methotrexate.

INTRODUCTION

Acute lymphoblastic leukemia (ALL) is the commonest type of cancer during childhood (Pui and Evans, 2006). Long-term event-free survival rates reach nearly 80%, whereas the rate of relapses is approximately 20-25%. From these, approximately one-quarter involves the central nervous system (CNS) (Nguyen et al., 2008).

Currently, most protocols suggest administration of intrathecal methotrexate (MTX) and intravenous infusions of high dose MTX (HDMTX) along with leucovorine (LV) rescue. MTX inhibits enzymes involved in folate homeostasis leading to elevated serum level of homocysteine (Kishi et al., 2003). Hyperhomocysteinemia has been identified as an independent risk factor for arterial ischemic events, such as myocardial infarction, stroke or peripheral vascular disease (Cacciapuoti, 2011).

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Researchers have also reported that increased levels of homocysteine can alter human monocyte function by up-regulating monocyte chemoattractant protein-1 (MCP-1) and interleukin-8 (IL-8); MCP-1 and IL-8 belong to the group of major chemokines responsible for leukocyte trafficking identified in atheromatous plaques (Zeng et al., 2003).

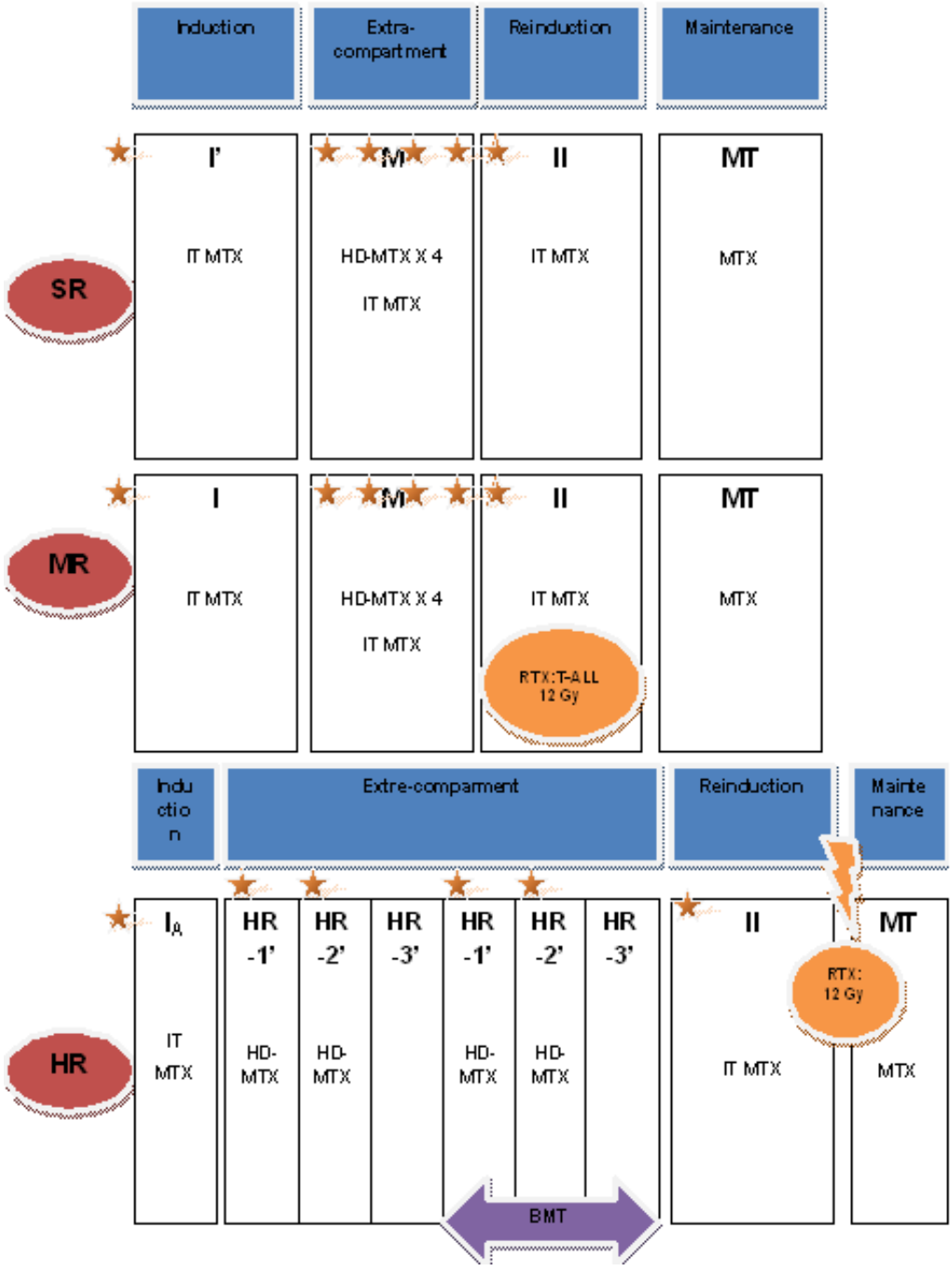
In this small prospective study, our aim was to report our institutional experience regarding the safety of HDMTX, as well as to determine whether vitamin B12, folic acid, MTX levels, homocysteine, IL-8 and MCP-1 levels have any correlation with neurotoxicity and thromboembolic episodes.

PATIENTS AND METHODS

Patients

A small institutional prospective study enrolling sixteen

Figure 1. The ALL-BFM 95 protocol showing with asterisks the time of the biochemical parameters measurements.



SR: standard-risk [prednisone good responder + WBC < 20,000/µl + 1 ≤ age (years) < 6 + non-T + complete marrow remission, day 33 + no t(9;22/BCR/ABL + no t(4;11)/MLLAF4)]; MR: medium-risk [prednisone good responder + complete marrow remission, day 33 + no t(9;22/BCR/ABL) + no t(4;11)/MLLAF4 + at least one of the following WBC ≥ 20,000/µl - age (years) < 1 - age (years) ≥ 6]; HR: high-risk [prednisone poor responder + no complete marrow remission, day 33 + t(9;22/BCR/ABL) + t(4;11)/MLLAF4)]; IT MTX: intrathecal methotrexate; HD-MTX: high-dose methotrexate, BMT: bone marrow transplantation; RTX: CNS preventative irradiation (if CNS positive: 18 Gy for > 2 years, 12 Gy > 1 < 2 years and no RTX for < 1year); I: protocol I; I': protocol I' (only 2 X 30mg/m^2 daunorubicin, days 8 + 15), I_A: protocol I_A (one additional IT MTX dose, day 18, when initial CNS involvement + reduction of prednisone, day 22, discontinued on day 30, + omission of the last 2 doses of asparaginase, days 30 and 33), M: protocol M; HR: HR element; MT: maintenance therapy

Serial blood assessment (10 ml of serum) of vitamin B12, folic acid, MTX levels, homocysteine, IL-8 and MCP-1 along with CSF measurement (5 ml) of IL-8 and MCP-1 levels at diagnosis (before the induction phase), after each 24hour of intravenous MTX infusion, and before the reinduction phase – namely six (6) times for each patient was performed. Levels of serum MTX were measured at 24h, 36h, 42h, 48h and 72h from the HDMTX infusion.

Table 1. Patients' characteristics.

Variables	Patients (n)	Percentage (%)
Sex		
Male	10	62.5
Female	6	37.5
Age		
Less than 1 y	0	0
1 to less than 6 y	12	75
6 to less than 10y	1	6.25
10 to less than 15y	3	18.75
15 y and older	0	0
Initial WBC		
Less than 10 000	6	37.5
10 000 to less than 20 x 10^9/L	3	18.75
20 000 to less than 50 x 10^9/L	1	6.25
50 000 to less than 100 x 10^9/L	2	12.5
100 000 to less than 200 000 x 10^9/L	0	0
200 000 and over	4	25
CNS status		
CNS1	16	100
CNS2	0	0
CNS3	0	0
Immunophenotype		
B	13	81.25
T	3	18.75
Cytogenetics		
Normal karyotype	4	25
Hyperdiploid	4	25
+4.11 (MLL/AF4)	0	0
+9.22 (BCR/ABL)	0	0
+12.21 (TEL/AML1)	0	0
Other	6	50
NCI risk criteria		
Standard risk	5	31.25
Medium risk	9	56.25
High risk	2	12.5
Prednisone response		
Good	14	87.5
Poor	2	12.5
BM day 15		
M1	13	81.25
M2	3	18.75
M3	0	0
Nonremission day 33		
No	0	0
Yes	16	100

children with newly diagnosed acute ALL was conducted in our department between March 2007 and August 2008.

Informed consent was obtained from the parent or guardian of each patient. The study was approved by the local Institutional Research and Ethics Committee.

ALL diagnosis, CNS status, immunophenotyping, cytogenetic and molecular genetic analysis, risk stratification and treatment

ALL diagnosis, CNS status, immune phenotyping, cytogenetic and molecular genetic analysis, risk stratification and chemotherapy were carried out as per the Berlin-Frankfurt-Münster-95 (BFM-95) protocol (Möricke et al., 2008; Bürger et al., 2003) (Figure 1). MTX (Methotrexate, Pfizer Haupt Pharma Wolfratshausen GmbH, Germany) was used.

Toxicity

Neurotoxicity was assessed according to the National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE) v3.

Statistical analysis

We investigated the associations between MTX levels and age, sex or risk group. Of particular importance were the associations among MTX and homocysteine, IL-8, MCP-1, folate levels, vitamin B12 and LV. The differences between 24 and 36 hours (MTX24_36) and the differences between 24 and 42 hours (MTX24_42) were calculated for each individual.

The association of these differences with sex, death, leukemia type, and risk group was tested using the Mann-Whitney test. The association of MTX time differences with risk group was tested using the Kruskal-Wallis test. The association of MTX differences with age was tested using the Pearson correlation coefficient.

The association of IL-8 (serum or CSF), MCP-1 (serum or CSF), folate and vitamin B12 with MTX was tested using mixed-effects models with total MTX as the main effect and time and the corresponding measurement at baseline as control variables.

The same mixed effects model, but with no control variables, were used for testing the effects of sex, risk group, age, death or leukemia type on IL-8, MCP-1, folate, vitamin B12 and LV dose. The statistical program used was SPSS Statistics 17.0

RESULTS - DISCUSSION

Results

Sixteen patients (10 boys and 6 girls), age range: 2-15 years (median: 4.85 years), all Caucasians, were enrolled in the study. The patients' characteristics are summarized in Table 1. Altogether, we analyzed 96 samples (64 treatment cycles).

Fourteen out of 16 children with ALL at their first complete remission, with a median follow up of 36 months, were evaluated. Two children died, 27 and 15 months after the diagnosis during their third and second relapse, respectively. Two children exhibited grade IV myelotoxicity, 2 conjunctivitis, 1 grade II cheilitis-stomatitis and another grade II dermatitis. Two children exhibited grade II seizures, whereas 1 exhibited thrombosis in the nature of central artery occlusion at both eyes that resulted in blindness.

No association was found between the levels of MTX, homocysteine, IL-8, MCP-1, folate and vitamin B12 and toxicity in the 2 children that exhibited seizures and the child that exhibited thrombosis.

No association of MTX differences between MTX24_36 and MTX differences between MTX24_42 or the total amount of MTX with sex, risk group, age, death and type of leukemia were identified. The results of the mixed-effects analysis for IL-8, MCP-1, folate and B12 with MTX, leukemia group, sex, risk group, age and death revealed, also, no association. No association of LV with MTX, sex, risk group, age, death and leukemia group was reported.

We found no statistically significant association between the increase in homeocysteine levels and sex, risk group, leukemia type or death. When folate levels where measured so as to determine whether they influence the MTX levels after therapy, we found that folate levels do not affect MTX levels at 24h from the HDMTX infusion.

Discussion

Despite its clinical success, MTX therapy can lead to hyperhomocysteinemia which is associated with neurotoxicitiy and thromboembolic phenomena (Winick et al., 1992). Since other studies have shown an acute increase of homocysteine plasma levels within 24 hours after HDMTX intravenous administration in patients with ALL (Refsum et al., 1991), the hypothesis that the measurement of homocysteine levels after MTX administration could represent a toxicity indicator came up.

Kishi et al., showed a tendency for a higher plasma homocysteine levels across all time points in ALL patients with seizures treated with HDMTX but not in patients with thrombosis. However, seven days after HDMTX treatment, their plasma concentration returned to baseline (Kishi et al., 2003). In our study, no association between the levels of any biochemical marker in plasma and CSF was revealed when the children that exhibited seizures were compared with the other ALL pediatric patients. Also, no association of MTX24_36, MTX24_42 and the total amount of MTX with sex, risk group, age, death and type of leukemia were identified.

MCP-1 and IL-8 are considered to be a part of the pathophysiological cascade responsible for the formation of atheromatous plaques (Zeng et al., 2003). Our study, revealed no association between the levels of IL-8 and MCP-1 (plasma and CSF), when the child that exhibited thrombosis was compared with the other ALL pediatric patients.

In a more recent study, patients with low folic acid levels before therapy initiation exhibited increasing MTX and homocysteine accumulation contrary to patients with high folic acid levels that exhibited almost no homocysteine accumulation (Vora et al., 2006). Our results showed that folate levels do not affect MTX levels at 24h.

The small number of pediatric patients studied poses serious limitations to the interpretation of our preliminary results. We could not establish that HDMTX was safe, while serial blood assessment of vitamin B12, folic acid, MTX, homocysteine, IL-8 and MCP-1 along with CSF measurement of IL-8 and MCP-1 did not show any correlation with neurotoxicity and thromboembolic side effects.

More clinical studies (with a significant number of patients) need to be undertaken so as to determine whether MTX, homocysteine, IL-8 and MCP-1 or other biological parameters could be used as markers for toxicity. Besides, biological markers in addition to genetic polymorphism should be the focus of future clinical studies so as to research their possible correlation with toxicity.

REFERENCES

Bürger B, Zimmermann M, Mann G, Kühl J, Löning L, Riehm H, Reiter A, Schrappe M (2003). Diagnostic cerebrospinal fluid examination in children with acute lymphoblastic leukemia: significance of low leukocyte counts with blasts or traumatic lumbar puncture. J. Clin. Oncol. (21): 184-8.

Cacciapuoti F (2011). Hyper-homocysteinemia: a novel risk factor or a powerful marker for cardiovascular diseases? Pathogenetic and therapeutical uncertainties. J. Thromb. Thrombolysis (32): 82-8.

Kishi S, Griener J, Cheng C, Das S, Cook EH, Pei D, Hudson M, Rubnitz J, Sandlund JT, Pui CH, Relling MV (2003). Homocysteine, pharmacogenetics, and neurotoxicity in children with leukemia. J. Clin. Oncol. (21): 3084-91.

Möricke A, Reiter A, Zimmermann M, Gadner H, Stanulla M, Dördelmann M, Löning L, Beier R, Ludwig WD, Ratei R, Harbott J, Boos J, Mann G, Niggli F, Feldges A, Henze G, Welte K, Beck JD, Klingebiel T, Niemeyer C, Zintl F, Bode U, Urban C, Wehinger H, Niethammer D, Riehm H, Schrappe M, German-Austrian-Swiss ALL-BFM Study Group (2008). Risk-adjusted therapy of acute lymphoblastic leukemia can decrease treatment burden and improve survival: treatment results of 2169 unselected pediatric and adolescent patients enrolled in the trial ALL-BFM 95. Blood (111): 4477-89.

Nguyen K, Devidas M, Cheng SC, La M, Raetz EA, Carroll WL, Winick NJ, Hunger SP, Gaynon PS, Loh ML, Children's Oncology Group (2008). Factors influencing survival after relapse from acute lymphoblastic leukemia: a Children's Oncology Group study. Leukemia (22): 2142-50.

Pui CH, Evans WE (2006). Treatment of acute lymphoblastic leukemia. N. Engl. J. Med. (354): 166-78.

Refsum H, Wesenberg F, Ueland PM (1991). Plasma homocysteine in children with acute lymphoblastic leukemia: Changes during a chemotherapeutic regimen including methotrexate. Cancer Res. (51): 828-35.

Vora A, Mitchell CD, Lennard L, Eden TO, Kinsey SE, Lilleyman J, Richards SM; Medical Research Council; National Cancer Research Network Childhood Leukaemia Working Party (2006). Toxicity and efficacy of 6-thioguanine versus 6-mercaptopurine in childhood lymphoblastic leukaemia: a randomised trial. Lancet (368): 1339-48.

Winick NJ, Bowman WP, Kamen BA, Roach ES, Rollins N, Jacaruso D, Buchanan GR (1992). Unexpected acute neurologic toxicity in the treatment of children with acute lymphoblastic leukemia. J. Natl. Cancer. Inst. (84): 252-56.

Zeng X, Dai J, Remick DG, Wang X (2003). Homocysteine mediated expression and secretion of monocyte chemoattractant protein-1 and interleukin-8 in human monocytes. Circ. Res. (93): 311-20.