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Value of intravenous 6-mercaptopurine during continuation treatment in childhood acute lymphoblastic leukemia and non-Hodgkin's lymphoma: final results of a randomized phase III trial (58881) of the EORTC CLG

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Between November 1990 and November 1996, EORTC Children Leukemia Group conducted a randomized trial in *de novo* acute lymphoblastic leukemia and lymphoblastic non-Hodgkin's lymphoma patients using a Berlin–Frankfurt–Munster protocol to evaluate the monthly addition of intravenous 6-mercaptopurine (i.v. 6-MP) (1 g/m^2) to conventional continuation therapy comprising per oral MTX weekly and 6-MP daily. Only during the first 18 months of the randomization period, 6-MP p.o. was interrupted for 1 week after each i.v. 6-MP. A total of 877 patients was randomized to either no i.v. 6-MP (Arm A) or additional i.v. 6-MP (Arm B). A total of 217 relapses (91 in Group A *vs* 128 in Group B) and 13 deaths in CR (5 *vs* 8) were reported; a total of 134 patients (55 *vs* 79) died. The median follow-up was 7.6 years. At 8 years, the disease-free survival rate was lower ($P = 0.005$) in Arm B (69.1% (s.e. = 2.2%)) than in Arm A (77.9% (s.e. = 2.0%)), and the hazard ratio was 1.45 (95% CI 1.12–1.89). In conclusion, as delivered in this study, i.v. 6-MP was detrimental to event-free survival.
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Introduction

Over the last 30 years, the results of the treatment of children with acute lymphoblastic leukemia (ALL) and lymphoblastic non-Hodgkin's lymphoma (NHL) have greatly improved. After the implementation of the total therapy concept,[1] survival rates reached approximately 50%. Further modifications of this total therapy regimen have led to current survival rates of approximately 80%.[2] Most of these modifications were introduced in the initial phases of the treatment (induction, consolidation and early or late intensification), whereas continuation therapy remained almost unchanged. The latter combines daily per oral 6-mercaptopurine (6-MP) and weekly, usually per oral, methotrexate (MTX).[3] These two drugs inhibit the *de novo* synthesis of purines and have a synergistic effect. After oral administration, presystemic metabolism of 6-MP causes an important first-pass effect and results in reduced bioavailability and variable plasma concentrations.[4] The inactive prodrug 6-MP is further metabolized intracellularly in cytotoxic and noncytotoxic products (reviewed by Estlin *et al*).[5]

In a preliminary study by Camitta *et al*,[6] it appeared that intravenous 6-MP during postremission therapy of children with ALL was of marked benefit when compared to historical controls. Several reasons might explain a beneficial effect of the intravenous route:[6,7] by short-cutting the first-pass effect and getting around the high variability of intestinal absorption, parenteral administration should lead to higher tissue levels and greater efficacy.[8] As a result of higher plasma concentration, intravenous (i.v.) administration of high doses of 6-MP enhances its penetration into the cerebrospinal fluid and might reduce the incidence of CNS relapses.[9–11] Moreover, parenteral administration guarantees better compliance.

For these reasons, the EORTC Children Leukemia Group (CLG) decided to address the value of i.v. 6-MP given in addition to conventional continuation therapy in a prospective randomized phase III (58881) trial for patients with ALL or lymphoblastic NHL. In the same trial, all or part of the patients had been previously randomized for two other questions: at the start of induction, the first randomization addressed the value of Erwiniase as compared to *Escherichia coli* asparaginase.[12] The second question addressed the value of moderately high-dose cytarabine when combined with high-dose methotrexate during postinduction-consolidation therapy of increased-risk patients.[13] The scope of this report is restricted to the evaluation of i.v. 6-MP during continuation therapy.

Patients and methods

Patients

The trial was started in July 1989 for patients under 18 years of age with previously untreated ALL or lymphoblastic NHL. Patients with mature B-cell ALL (Burkitt-like) or with non-lymphoblastic NHL were excluded. The ALL patients were stratified in a low, an increased and a very high-risk group as previously defined.[12–14] NHL patients were stratified according to Murphy's clinical stage:[15] in the low-risk group if stage I or II and in the increased-risk group if stage III or IV. NHL patients

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who failed induction therapy were switched to the very high-risk group.[12,13] Patients who relapsed were treated according to an adapted Henze's protocol.[16] For registration of a child in the study, an informed consent by his parents or his legal guardian was required. The 58881 protocol has been approved by the EORTC Protocol Review Committee and by the respective ethical committees in each participating country.

Definitions and evaluations

When bone marrow involvement was observed, patients with less than 25% of blasts in the bone marrow were considered as having NHL. All cases were studied for specific lymphoblastic cell characteristics, including morphology, immunophenotype and cytogenetics. Morphologic classification of ALL was according to the French–American–British classification.[17] Classification of lymphomas was according to the working formulation and the revised European–American Lymphoma Classification.[18] Frozen specimens or cell suspensions were evaluated for B- and T-lineage-associated antigens with standard techniques.[19] Cytogenetic analysis of the lymphoblastic cells was performed by R or G banding and chromosomes were classified according to the International system for Human Cytogenetic Nomenclature.[20]

For ALL, complete remission was defined as less than 5% blasts in the bone marrow, recovery of normal hematopoiesis and no evidence of disease at any other site. For NHL patients, CR was defined by the disappearance of all clinical, imaging and cytologic signs of lymphoma.

Central nervous system (CNS) leukemia was diagnosed when neurologic abnormalities related to blastic infiltration of the CNS were observed and/or when blasts were identified on cytocentrifuge examination of CSF in which white blood cell (WBC) count was greater than 5×10^9 cells/l. Infectious, renal, hepatic and neurologic toxicities were evaluated and graded according to the World Health Organization grading system.[21]

Treatment

A Berlin–Frankfurt–Munster (BFM)-based protocol with slight modifications was used.[22] The therapy regimen was recently described.[12,13] During the course of the study, the randomization period for the first question (Erwinia *vs E. coli* asparaginase) did not coincide with that of the third question (addition or not of i.v. 6-MP). The registration for asparaginase started 1 year later than the registration for 6-MP: all patients registered during the first year of the EORTC 58881 trial received *E. coli* asparaginase from Bayer. In a second period, all patients were randomized at diagnosis to receive either *E. coli* asparaginase from MEDAC or Erwiniase. In a third period, after this randomization was stopped because the inferiority of Erwiniase had become evident, all patients received MEDAC *E. coli* asparaginase as first-line treatment.[12] Increased-risk patients only were randomized to receive or not high-dose cytarabine (HD Ara-C) during interval therapy.[13]

All patients were eligible for the third randomization if they remained in first complete remission at the start of continuation therapy: they were randomized to receive either conventional treatment with only oral 6-MP and MTX, or to the same treatment with the addition of 4-weekly i.v. 6-MP.

In the conventional arm, continuation therapy consisted of oral 6-MP (50 mg/m^2/day) and oral MTX (20 mg/m^2/week). In the experimental arm, i.v. 6-MP (1 g/m^2 given over 8 h every 4 weeks) was added to the treatment. During the first 18 months of the trial (November 1990–May 1992), daily oral 6-MP (but not MTX) was interrupted for 1 week after each administration of i.v. 6-MP. After the first 18 months of the trial, the treatment protocol was amended and henceforth called for uninterrupted oral 6-MP treatment in all patients. In both arms, doses of per oral drugs were adjusted in order to maintain the number of leucocytes between 2 and 3×10^9/l.

Statistical analysis

Randomization for the 6-MP question was performed centrally (EORTC Data Center, Brussels)[23] before starting continuation therapy. Patients were stratified according to center, risk-group and previous randomization arms. The primary end point was the disease-free survival (DFS) from the date of randomization to the date of first relapse or to death in CR. The secondary end points were the time from randomization to isolated CNS relapse, with patients with other types of event (relapse, death in CR) being censored at the time of event, and the duration of survival (the time from randomization to death of any cause). The actuarial curves were computed using the Kaplan–Meier technique, and the standard errors (s.e.) of the estimates were obtained via the Greenwood formula.[24] The differences between curves were tested for statistical significance using the two-tailed log-rank test or the log-rank test stratified by a categorical factor.[24] To summarize the overall treatment difference, the hazard ratio of having an event per time in arm B *vs* in arm A was estimated via the Cox proportional hazards model.[25] For the overall comparison, the 95% confidence interval (CI) of the hazard ratio was computed, and for two subgroup analyses, the 97.5% CI was used, due to Bonferonni correction for multiplicity. A total of 820 patients was intended to be randomized in order to detect a significant difference in terms of DFS at 5 years (70 *vs* 80%), corresponding to a hazard ratio of 0.63 (alpha = 5%, beta = 20%). All analyses were performed according to the intent-to-treat principle.

Results

Patients

Between November 1990 and November 1996, a total of 877 patients were randomized: 439 in the no i.v. 6-MP arm, 438 in the i.v. 6-MP arm. The characteristics of both arms are listed in Table 1. Among the 877 patients, 820 were diagnosed with ALL and 57 with NHL: stage I (2 *vs* 2), stage II (4 *vs* 1), stage III (19 *vs* 15) and stage IV (4 *vs* 10). Patients' characteristics were generally well balanced in the two arms. In arm B, the male:female ratio was slightly higher (1.39 *vs* 1.31) as compared with arm A, and the incidence of high WBC ($\geqslant 100 \times 10^9$/l) was also higher (10.9 *vs* 8.0%); the infants represented 1.4% in arm B *vs* 1.8% in arm A, and ALL patients with VHR features represented approximately 6% in both arms. The type of L-asparaginase administered during the induction and intensification phases as well as the type of interval therapy (with or without high-dose cytarabine) were well balanced between the two treatment groups.

Arm B was further divided into two groups: patients registered before 1 January 1992 (randomized during the first 18 months of the protocol), for whom 6-MP per oral was interrupted for 1 week after each administration of i.v. 6-MP, and those registered later who had to receive uninterrupted per oral 6-MP. During

Table 1 Patients' characteristics

	Arm A N = *439 (%)*	*Arm B* N = *438 (%)*
Disease		
ALL	410 (93.4)	410 (93.6)
NHL	29 (6.6)	28 (6.4)
Age at diagnosis (years)		
<1	8 (1.8)	6 (1.4)
1–<10	362 (82.5)	357 (81.5)
⩾10	69 (15.7)	75 (17.1)
Gender		
Male:female	249:190	255:183
Initial WBC (x 10^9/l)		
<25	316 (72.0)	302 (68.9)
25–<50	50 (11.4)	48 (11.0)
50–<100	38 (8.7)	40 (9.1)
100–<250	20 (4.6)	32 (7.3)
⩾250	15 (3.4)	16 (3.7)
Immunophenotype		
B-lineage	364 (82.9)	362 (82.6)
T-lineage	75 (17.1)	76 (17.4)
Chromosomes in ALL pts		
Hyperdiploid	100 (24.4)	88 (21.5)
Diploid	110 (26.8)	102 (24.9)
Pseudodiploid	65 (15.9)	69 (16.8)
Hypodiploid	16 (3.9)	18 (4.4)
Other	41 (10.0)	44 (11.0)
Unknown	78 (19.0)	89 (21.7)
Initial Risk Factor in ALL patients16		
Unknown	1 (0.2)	0 (0)
<0.8	154 (37.6)	153 (37.3)
0.8–<1.2	135 (32.9)	134 (32.7)
1.2–<1.7	105 (25.6)	101 (24.6)
⩾1.7	15 (3.7)	22 (5.4)
Initial CNS involvement in ALL pts		
Unknown	0 (0.0)	3 (0.7)
Not involved	405 (98.8)	400 (97.6)
Involved/clinical signs	5 (1.2)	7 (1.7)
VHR features in ALL patients		
Absence	380 (92.7)	384 (93.7)
Presence	30 (7.3)	26 (6.3)
Randomization to asparaginase		
Not randomized (Bayer, *E. coli*)	32 (7.3)	34 (7.8)
Randomized to Medac, *E. coli*	118 (26.9)	116 (26.5)
Randomized to Erwiniase	111 (25.3)	110 (25.1)
Postrandomization (Medac, *E. coli*)	178 (40.5)	178 (40.6)
Year of registration and type of Asparaginase		
<1992 and non-Medac asparaginase	79 (18.0)	76 (17.4)
<1992 and Medac asparaginase	48 (10.9)	48 (11.0)
⩾1992 and Non-Medac asparaginase	64 (14.6)	68 (15.5)
⩾1992 and Medac asparaginase	248 (56.5)	246 (56.2)
Randomization to HD Ara-C		
Not randomized	57 (13.0)	53 (12.1)
Randomized to no HD Ara-C	96 (21.9)	95 (21.7)
Randomized to HD AraC	103 (23.5)	104 (23.7)
Low risk (not eligible)	183 (41.7)	186 (42.5)

the first period, 127 *vs* 124 patients were randomized in Arms A and B, respectively, the majority of whom (79 *vs* 76) received non-Medac asparaginase. During the second period, 312 *vs* 314 were randomized in the two arms, respectively, out of whom 594 (248 *vs* 246) received Medac asparaginase. Patients' characteristics were well balanced between Arms A and B in the first as well as the second periods.

Type of event by treatment group

At the time of analysis, the median follow-up was 7.6 years (4–12 years). There were 230 events, including 217 relapses and 13 deaths without relapse. The crude rates of different sites of relapse by treatment arm, in ALL and NHL patients are listed in Table 2. There were more relapses in Arm B than in Arm A: 91 (20.7%) in Arm A *vs* 126 (28.8%) in Arm B. This difference was mostly due to an increase of isolated bone marrow relapses (46 (10.5%) patients in Arm A *vs* 60 (13.7%) patients in Arm B) and of combined CNS relapse (17 (3.9%) *vs* 27 (6.2%)). Isolated CNS relapses (15 (3.4%) *vs* 17 (3.9%)) were evenly distributed. The 8-year isolated CNS relapse rates were comparable as well: 3.6% (Arm A) *vs* 4.3% (Arm B), hazard ratio = 1.16. A total of 12 patients died in CR: 5 (1.1%) in Arm A and 8 (1.8%) in Arm B.

DFS by treatment group

The DFS rate at 8 years was 69.1% (s.e. = 2.2%) for the patients in Arm B and 77.9% (s.e. = 2.0%) for those in Arm A (Figure 1) (logrank $P = 0.005$). The estimated hazard ratio was 1.45 (95% CI 1.12–1.89). In ALL patients the estimated hazard ratio was 1.40 (97.5% CI 1.03–1.90) and in NHL patients is was 3.06 (97.5% CI 0.67–13.95). In the latter group, the adjustment for Murphy stage let to a hazard ratio of 2.87 (97.5% CI 0.62–13.38).

Multivariate analysis based on the Cox model indicated that the difference remained highly significant ($P = 0.006$) between the two groups (hazard ratio = 1.44, 95% CI 1.11–1.88) after adjustment for the following factors: initial WBC count ($P < 0.0001$), type of asparaginase ($P = 0.0001$), age ($P = 0.0003$) and sex ($P = 0.01$) (Table 3). Period of recruitment was of prognostic importance only in univariate analysis ($P = 0.03$). In multivariate analysis, it was not ($P = 0.78$) and did not influence the magnitude of the treatment difference ($P = 0.64$). Moreover, in multivariate setting, after adjustment for the same prognostic factors, no significant interaction could be determined between the type of asparaginase (Medac *vs* non-Medac) and the randomized group ($P = 0.22$). In patients who had received Medac *E. coli* asparaginase during induction and intensification, the estimated hazard ratio of the comparison Arm B *vs* Arm A was 1.29 (97.5% CI 0.87–1.91) in multivariate setting, and in those who had received one of the other, less potent asparaginases (*E. coli* asparaginase from Bayer or Erwiniase) it was 1.73 (97.5% CI 1.09–2.75). Using a Cox model, a prognostic score was defined based on the initial WBC, the type of asparaginase, age and sex. Treatment remained significant once adjusted for this score, but the interaction between this score and the treatment arm was not significant ($P = 0.38$), indicating that the treatment difference was quite uniform among the risk groups defined by the above-mentioned factors. Previous randomized treatment during interval therapy, high-dose or no high-dose Ara-C, had absolutely no influence on the treatment difference.

Duration of survival by treatment group

A total of 134 patients died: 55 in Arm A and 79 in Arm B. The 8-year survival rate was lower for the patients randomized in Arm B than for those randomized in Arm A: 81.3% (s.e. = 2.0%) *vs* 87.2% (s.e. = 1.7%) (Figure 2) (logrank $P = 0.03$). The estimated hazard ratio was 1.46 (95% CI 1.04–2.07). In ALL patients, the 8-year survival rates were 81.5% (s.e. = 2.0%) *vs* 86.5% (s.e. = 1.7%), and the hazard ratio was 1.36 (97.5% CI 0.91–2.0.4). In NHL patients the 8-year survival rates were 77.7% (s.e. = 8.1%) *vs* 96.6% (s.e. = 1.8%), and the hazard ratio

was 6.78 (97.5% CI 0.60–95.8). After adjustment for Murphy's stage, it was 5.61 (97.5% CI 1.88–5.61).

Multivariate analysis based on the Cox model led to the same conclusions as for DFS (Table 3). Period of recruitment was not of prognostic importance whether in univariate or multivariate analysis ($P=0.88$). No interaction between recruitment period and treatment difference was observed ($P=0.82$). The results in Arm B were consistently worse than in Arm A independently ($P=0.55$) of the type of asparaginase received. After adjustment for the prognostic score defined above, the difference remained significant. The interaction between this score and treatment arm was far from being significant ($P=0.79$), indicating the independence of the deleterious effect of i.v. 6-MP from this score.

Toxicity and protocol compliance

Generally, toxic side effects were observed in both arms with comparable frequency. Moderate and major infectious complications were evenly distributed between the two groups: 38.7% in Arm A *vs* 36.4% in Arm B. The incidence of liver biologic toxicities was higher in Arm B (22.0%) than in Arm A (16.6%).

The average daily dose/m^2 of oral 6-MP actually given throughout continuation therapy was lower ($P<0.0001$) in Arm B as compared with Arm A: the median (mean) was 55.7 (57.0) mg/m^2 in Arm A *vs* 47.9 (48.3) mg/m^2 in Arm B. Patients randomized in the first period of the trial received a median

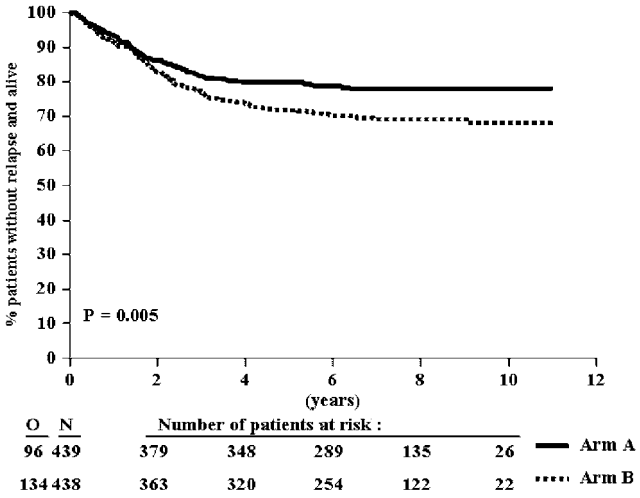

Figure 1 DFS from randomization according to the randomized arm. N = number of patients; O = observed number of events (relapses or deaths in first CR). *P*-value was given by the logrank test.

Table 2 Type of first event in ALL and NHL patients, according to the randomized arm

	ALL pts		*NHL pts*		*ALL+NHL pts*	
	Arm A	*Arm B*	*Arm A*	*Arm B*	*Arm A*	*Arm B*
Continuous CR	317 (77.3)	284 (69.3)	26 (89.7)	20 (71.4)	343 (78.1)	304 (69.4)
Death in CR	5 (1.2)	7 (1.7)	0 (0.0)	1 (3.6)	5 (1.1)	8 (1.8)
Relapse	88 (21.5)	119 (29.0)	3 (10.3)	7 (25.0)	91 (20.7)	126 (28.8)
Bone marrow, isolated	46 (11.2)	57 (13.9)	0 (0.0)	3 (10.7)	46 (10.5)	60 (13.7)
CNS, isolated	15 (3.7)	17 (4.1)	0 (0.0)	0 (0.0)	15 (3.4)	17 (3.9)
CNS, combined	17 (4.1)	26 (6.3)	0 (0.0)	1 (3.6)	17 (3.9)	27 (6.2)
Gonads, isolated	2 (0.5)	5 (1.2)	0 (0.0)	0 (0.0)	2 (0.5)	5 (1.1)
Gonads+BM	6 (1.5)	6 (1.5)	0 (0.0)	0 (0.0)	6 (1.4)	6 (1.4)
Lymph nodes, isolated	0 (0.0)	0 (0.0)	1 (3.4)	1 (3.6)	1 (0.2)	1 (0.2)
Other	2 (0.5)	8 (1.9)	2 (6.8)	2 (7.1)	4 (0.9)	10 (2.3)
Total	410 (100)	410 (100)	29 (100)	28 (100)	439 (100)	438 (100)

Values are expressed as numbers and percent in parentheses.

Table 3 Results of the Cox proportional hazards model regarding the DFS and survival

End point	*Variable*	*Hazard ratio*[a]	*(95% CI)*	P-*value*[b]
DFS				
	Randomized group: Arm B *vs* Arm A	1.44	(1.11, 1.88)	0.006
	Initial WBC[c]	1.30	(1.17, 1.44)	<0.0001
	Sex (male *vs* female)	1.42	(1.09, 1.87)	0.01
	Asparaginase (Non-Medac *vs* Medac)	1.66	(1.28, 2.16)	0.0001
	Age (<1 year *vs* ⩾1 year)	3.41	(1.74, 6.68)	0.0003
Survival				
	Randomized group: Arm B *vs* Arm A	1.42	(1.00, 2.00)	0.047
	Initial WBC[c]	1.27	(1.12, 1.45)	0.0003
	Sex (male *vs* female)	1.67	(1.16, 2.42)	0.006
	Asparaginase (non-Medac *vs* Medac)	1.68	(1.19, 2.37)	0.003
	Age (<1 year *vs* ⩾1 year)	4.01	(1.86, 8.66)	0.0004

[a]A value>1 indicates that the outcome is worse in the given category as compared with the baseline.
[b]*P*-value was determined by the Wald test.
[c]Initial white blood cell (WBC) count ($\times 10^9$/l) categorized as: 1 = <25, 2 = 25 to <50, 3 = 50 to <100, 4 = 100 to <250, 5 = ⩾250.

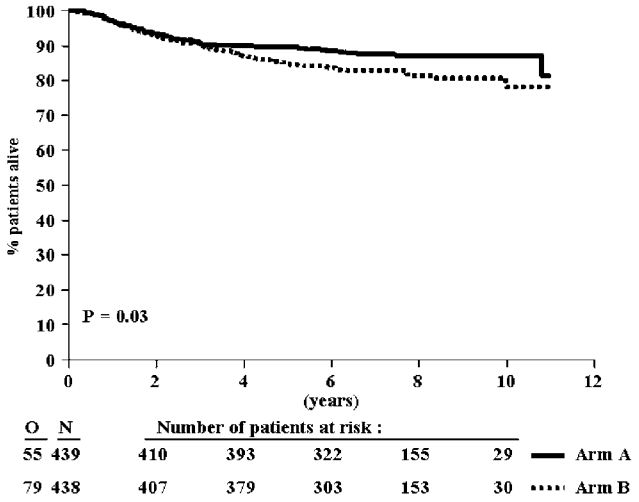

Figure 2 Duration of survival from randomization according to the randomized arm. *N* = number of patients; O = observed number of deaths. *P*-value was given by the logrank test.

(mean) daily dose of oral 6-MP of 53.2 (53.6) *vs* 48.1 (45.6) mg/m^2 in Arms A and B, respectively. In the second period, this was 56.0 (57.9) *vs* 47.9 (49.2) mg/m^2 in Arms A and B, respectively. In Arm B, the median (mean) 4-weekly dose of i.v. 6-MP was 0.83 (0.73) g/m^2; it was 0.90 (0.79) and 0.81 (0.70) g/m^2 in the first and second periods respectively.

Discussion

In contrast to what was expected from previous findings,[6] randomization to i.v. 6-MP during continuation therapy has led to significantly worse outcome. At 8 years, the DFS and survival rates were approximately 9% (69.1 *vs* 77.9%) and 6% (81.3 *vs* 87.2%) lower, respectively, in Arm B.

Other studies have evaluated the efficacy of intermittent i.v. 6-MP as compared to protracted daily per oral 6-MP during post-remission therapy either for all ALL patients[26] or for some risk-groups of patients.[27–29] None of these studies showed any benefit but no detrimental effect either from the use of i.v. 6-MP. These studies differed from ours in at least two respects: they evaluated i.v. 6-MP given instead of and not in combination with per oral 6-MP. Moreover in two of these studies,[27,29] i.v. 6-MP was started early during postremission therapy, concomitantly with and before further intensive multiagent chemotherapy or continuation therapy. Any possible detrimental effect of i.v. 6-MP could thus have been erased by the efficacy of the concomitant and subsequent treatment courses. By contrast, in our study the addition of i.v. 6-MP jeopardized the efficiency of the sole two drugs or at least of one of the two used in order to eradicate any minimal residual disease still present.

As to why the addition of i.v. 6-MP was detrimental, two possible reasons not necessarily mutually exclusive, may be considered. The patients assigned to the i.v. 6-MP arm received a moderately lower cumulative per oral dose. This difference resulted, during the first 18 months of the trial, from the mandatory interruption of per oral 6-MP during every fourth week of the treatment and, throughout the whole of the study, from more downward adjustments of the per oral 6-MP dosing. However, for each period of the study, the deleterious effect of i.v. 6-MP, adjusted for the type of asparaginase previously administered and for other confounding factors remained approximately the same: the increase of the event (relapse or death in CR) rate per time unit in Arm B *vs* Arm A was 55% in the first period and 39% in the second. Overall, the excess of the event rate was 45% (hazard ratio = 1.45). It has already been suggested that the total dose of 6-MP administered is positively correlated with the DFS.[30] The inferiority of Arm B could also result from an inhibition by the i.v. 6-MP of the cytotoxic activity of per oral 6-MP. This activity is mainly related to the intracellular accumulation of 6-thioguanine nucleotides, which are incorporated into DNA and lead to its damage.[31] Alternatively, 6-MP is catabolized into inactive thiouric acid by xanthine oxidase and methylmercaptopurine by thiopurine methyltransferase. Unfortunately, we have no pharmacological data that would have allowed us to verify the hypothesis that i.v. 6-MP could have modified the ratio of activating *vs* inactivating enzymatic activities and thereby have led to a lower steady-state intracellular concentration of thioguanine nucleotides.

Alternatively, Schmiegelow *et al*[32] reported that if the doses of 6-MP given during continuation chemotherapy were tailored in order to sustain high 6-TG levels, the incidence of relapse increased after cessation of therapy in girls with ALL. These authors speculated that above a hypothetical level of 6-TG, residual leukemic cells escaped apoptosis by being maintained in the G0 phase of the cell cycle and allowed to proliferate again after chemotherapy had been stopped. In this regard, it is noteworthy that in our study as well, the divergence of the two DFS curves approximately started from completion of the treatment (Figure 1).

In conclusion, whatever the mechanism involved, addition of 6-MP to continuation therapy was detrimental. Our results also show that, notwithstanding the intensity of the initial induction, consolidation and intensification courses of the BFM protocol, small changes of the continuation therapy may lead to important changes in outcome, which underlines the persisting importance of this treatment phase.

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