A Phase I–II Trial of Escalating Doses of Mitoxantrone with Fixed Doses of Cytarabine plus Fludarabine as Salvage Therapy for Patients with Acute Leukemia and the Blastic Phase of Chronic Myelogenous Leukemia

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BACKGROUND. Cytarabine is an essential drug for inducing remission of acute myelogenous leukemia, and it is also one the most effective drugs used as salvage therapy for patients with all types of relapsed acute leukemia. Nevertheless, there is considerable room for improvement in the treatment of patients with relapsed leukemia in terms of both the reinduction rate and the duration of response. Fludarabine has been shown to augment responses to cytarabine, possibly by increasing the intracellular concentrations of the active metabolite cytarabine triphosphate. Higher-than-standard doses of mitoxantrone have been shown to augment responses to cytarabine, possibly by increasing the DNA strand breaks induced by topoisomerase II; these strand breaks cannot be effectively repaired in the presence of cytarabine triphosphate. This preliminary study was designed to determine whether moderately high doses of mitoxantrone could be added to the combination of fludarabine and cytarabine in an attempt to improve the combination's antileukemic efficacy.

METHODS. Fifty-five adults with relapsed or refractory acute leukemia or the blastic phase of chronic myelogenous leukemia (CML) received salvage therapy with the FLAM regimen, which consisted of fludarabine, cytarabine, and increasing doses of mitoxantrone.

RESULTS. Even with doses of mitoxantrone escalated to as much as 60 mg/m^2 over 4 days, dose-limiting toxicity was not observed. Overall, the complete response rate was 27.3% (15 of 55 patients, including 4 of 17 with acute myelogenous leukemia [AML], 4 of 12 with acute lymphocytic leukemia [ALL], and 7 of 26 with the blastic phase of CML). The median time to complete response was 42 days. Toxicity other than myelosuppression was manifested primarily as hyperbilirubinemia, which was reversible in all cases. Poor performance status and undifferentiated blastic phase of CML were poor prognostic factors for response to FLAM.

CONCLUSIONS. The FLAM regimen is an active salvage regimen and should be formally evaluated in Phase II studies of patients with AML, ALL, and the myeloid and lymphoid blastic phases of CML. ***Cancer* 1999;86:2246–51.**
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KEYWORDS: fludarabine, cytarabine, mitoxantrone, acute lymphoblastic leukemia, acute myelogenous leukemia, blastic phase of chronic myelogenous leukemia, reinduction regimens.

Despite the achievement of an initial complete remission (CR) in the majority of patients with acute leukemia, survival rates for adults are only about 30% at 2 years and 20% at 5 years.[1] Patients who

fail to achieve a CR or who relapse quickly after attaining a remission are unlikely to be cured with chemotherapy alone.[2–4] However, improvements in response and survival in salvage patient populations should translate into an increased cure fraction in previously untreated patients. High doses of cytarabine (ara-C) have been utilized to overcome resistance in leukemic cells, albeit with substantial toxicity.[5] The combination of intermediate doses of ara-C along with fludarabine has also been associated with an increase in CR rate in refractory and relapsed acute myelogenous leukemia (AML), from about 18% to 36%,[6] presumably because fludarabine can significantly increase the intracellular accumulation of the active metabolite ara-C triphosphate (ara-CTP).[7] In addition to optimizing the accumulation of ara-CTP in the leukemic cells, other studies indicate that doses of mitoxantrone, a topoisomerase II–directed drug, can be escalated far beyond the conventional dose schedule, with clinical benefit to patients with acute leukemia when combined with ara-C.[8,9] The rationale for the administration of high dose mitoxantrone comes from in vitro studies demonstrating a uniquely steep dose-response curve in clonogenic assays with ovarian carcinoma and leukemic cells.[10,11] Clinical studies indicate that the tolerance of high dose mitoxantrone is acceptable, even in an elderly patient population.[8] In this study, we report the results of a Phase I–II trial of fixed doses of ara-C plus fludarabine with escalating doses of mitoxantrone (FLAM) in patients with relapsed acute leukemia and the blastic phase (BP) of chronic myelogenous leukemia (CML). Because responses to this regimen were observed in relapsed patients with AML, acute lymphocytic leukemia (ALL), and the myeloid and lymphoid BPs of chronic myelogenous leukemia (CML), we concluded this study before reaching the maximum tolerated dose (MTD) for mitoxantrone in combination with fludarabine plus cytarabine after we reached a dose of mitoxantrone that was about two-thirds of the published single-agent MTD for mitoxantrone. We suggest that formal Phase II studies could be performed with AML, ALL, and both the myeloid and lymphoid (BPs) of CML to define properly the activity of the FLAM regimen.

PATIENTS AND METHODS

Eligibility Requirements

Patients age >15 years with refractory or relapsed AML, ALL, or the BP of CML were eligible for treatment. The leukemia was considered refractory if the patient did not achieve remission after two courses of standard induction therapy or one course of induction therapy containing high dose cytarabine. The leukemia was considered relapsed if the patient had previously obtained remission by the criteria described below. All patients had greater than 30% blast cells in the bone marrow aspirate prior to beginning therapy on this protocol. Lineage was determined using French–American–British (FAB) criteria.[12] Briefly, myeloid leukemias required >3% myeloperoxidase (MPO) positivity in blast cells of the bone marrow aspirate; lymphoid leukemias had >40% terminal deoxynucleotidyl transferase (TdT) positivity in the absence of MPO positivity in the blast cells of the bone marrow aspirate. Undifferentiated blastic crisis of CML was both MPO and TdT negative. Adequate hepatic and renal function, i.e., bilirubin and creatinine <2.0 mg/dL, were required, as well as a left ventricular ejection fraction (LVEF) of ≥50% measured by gated cardiac pool scan and/or echocardiogram. The treatment protocol and consent form were approved by the Surveillance (Human Use) Committee of the M. D. Anderson Cancer Center. Written informed consent was obtained from all patients.

Treatment Schedule

Groups of patients were sequentially enrolled at the assigned dose levels of mitoxantrone, which was given over 15 minutes in 4 equally divided doses starting at Hours 0, 26, 50, and 74. All patients received fludarabine 30 mg/m^2 over 30 minutes starting at Hours 20, 44, and 68, and ara-C 1 g/m^2 over 2 hours starting at Hours 24, 48, and 72. The rationale for the sequence is that fludarabine given before ara-C will enhance ara-CTP cellular accumulation and retention, and both fludarabine and ara-C will prevent the repair of DNA damage induced by mitoxantrone.[6,7,13] Allopurinol 300 mg was given orally every day for 7 days. Supportive care, in terms of transfusions of blood, platelets, antibiotics, and growth factors, was left to the discretion of the treating physician. Typically, patients would be transfused with packed red blood cells if the hemoglobin was <80 gm/L, or with random-donor or single-donor platelets for a platelet count of $<10 \times 10^9$/L or if there were signs of bleeding. Patients were given oral prophylactic antibiotics with either trimethoprim/sulfamethoxasole or levofloxacin, intravenous antibiotics if they were febrile >38.3 °C or symptomatic, and granulocyte-colony stimulating factor at a dose of 5 μg/kg from Day 15 after treatment began until the absolute neutrophil count was higher than 1000 cells/mm^3.

Evaluation and Response Criteria

Bone marrow aspirates and/or biopsies were obtained 14 days after treatment to assess the effectiveness of therapy in clearing the bone marrow of leukemia. Patients who relapsed without clearing the bone mar-

row of leukemia were classified as having "primary resistance." Patients who relapsed after clearing the bone marrow of leukemia were classified as having "secondary resistance." Patients who died while aplastic experienced "aplastic death," whereas patients who died with more than 20% blasts in the bone marrow experienced "death with greater than 20% leukemic infiltrate." Bone marrow aspirates and/or biopsies were obtained weekly after Day 14 to assess remission status.

CR was defined as the achievement of a normocellular bone marrow (hematopoietic cells occupying at least 30% of the cross-sectional area) with fewer than 5% blasts, a granulocyte count of at least 1×10^9/L, and a platelet count of at least 100×10^9/L. The time to CR reflected the interval between Day 1 of therapy and attainment of CR. In patients with CML, complete hematologic remission was further assessed by cytogenetic response. Other responses were as previously defined.[14]

Patients who obtained CR received consolidation consisting of fludarabine 30 mg/m^2 daily $\times$ 3, ara-C 1g/m^2 daily $\times$ 3, and mitoxantrone 10 mg/m^2 daily $\times$ 3. Toxicity was assessed using the National Cancer Institute (NCI) common toxicity criteria.

RESULTS

Patient Characteristics

From April 1992 to May 1993, a total of 55 adults with relapsed or refractory acute leukemia or the BP of CML were enrolled in the study (Table 1). Groups of patients were treated sequentially at the three separate dose levels of mitoxantrone. There were 17 cases of AML, 12 cases of ALL, and 26 cases of the BP of CML, of which 11 were myeloid, 10 were lymphoid, and 5 were undifferentiated. The median age of patients entered was 45 years (range, 18–81 years). A Zubrod performance status of 2 or greater was recorded for 17 of the 55 patients treated. Thirty-two patients were treated on this protocol as their first salvage attempt, 14 as their second, and 9 as their third or beyond. Of the acute leukemia patients, 6 of 29 were primary refractory to initial induction therapy, 9 had an initial CR of 6 months or less, 5 had initial CRs of more than 1 year, and the remainder had initial CRs of 6–12 months.

TABLE 1
Patient Characteristics

		Mitoxantrone dose, mg/m^2		
	Total	40	50	60
Entered	55	24	20	11
Age (yrs)				
≤45	28	11	13	4
>45	27	13	7	7
Gender				
Male	27	15	9	3
Female	28	9	11	8
Performance status				
0–1	38	16	13	9
2–4	17	8	7	2
Diagnosis				
ALL	12	4	5	3
AML	17	8	4	5
CML Blastic Phase	26			
Myeloid	11	5	4	2
Lymphoid	10	5	5	0
Undifferentiated	5	2	2	1
Salvage no.				
1	32	15	12	5
2	14	8	5	1
>2	9	1	3	5
Salvage status				
Primary refractory	6	3	3	0
First CR ≤6 mos	9	5	2	2
First CR >6 mos	14	4	4	6

ALL: acute lymphocytic leukemia; AML: acute myelogenous leukemia; CML: chronic myelogenous leukemia; CR: complete remission.

TABLE 2
Response to Treatment[a]

		Mitoxantrone dose, mg/m^2		
	Total	40	50	60
Complete response	8	4	1	3
Return to chronic phase	7	5	1	1
Early death	1	0	1	0
Aplastic death	13	3	7	3
Death >20% leukemic infiltrate	4	3	1	0
Primary resistance	3	2	1	0
Secondary resistance	19	7	8	4
Total	55	24	20	11

[a] Refer to "Evaluation and Response Criteria" under "Patients and Methods" for definitions of terms.

Induction Response and Toxicity

Responses

All patients were evaluable for response and toxicity. Response is shown in Table 2. Overall, 15 patients (27%) achieved CR with the regimen, including 8 patients with acute leukemia and 7 patients in the BP of CML who returned to a second chronic phase. Thirteen patients (24%) died during remission induction with hypoplastic bone marrows, and 26 (47%) had resistant disease. Three of the 15 CR patients required 2 courses to attain remission; all 3 had shown considerable improvement after their first course but did not attain criteria for remission. The median time to CR was 42 days (range, 21–113 days), with no difference in

TABLE 3
Response by Salvage Attempt

Diagnosis	First salvage	Second salvage	Third salvage	≥Fourth salvage
AML	2/7	1/4	1/4	0/2
ALL	2/5	1/2	1/2	0/3
CML-myeloid blastic phase	3/11			
CML-lymphoid blastic phase	4/10			
CML-undifferentiated blastic phase	0/5			
Overall	11/38	2/6	2/6	0/5

ALL: acute lymphocytic leukemia; AML: acute myelogenous leukemia; CML: chronic myelogenous leukemia.

TABLE 4
Response by Total Mitoxantrone Dose in mg/m^2

Diagnosis	40	50	60
AML	2/8	0/4	2/5
ALL	2/4	1/5	1/3
CML blastic phase			
Myeloid	2/5	0/4	1/2
Lymphoid	3/5	1/5	0/0
Undifferentiated	0/2	0/2	0/1
Overall	9/24	2/20	4/11

ALL: acute lymphocytic leukemia; AML: acute myelogenous leukemia; CML: chronic myelogenous leukemia.

the time to CR between patients who received 40 mg/m^2 as opposed to 60 mg/m^2 of mitoxantrone during induction. Table 3 shows the response by salvage attempt for various diagnoses. Responses in first salvage as well as in multiply relapsed AML and ALL patients were seen. Complete hematologic responses were seen in the myeloid and lymphoid BPs but not in the undifferentiated BP of CML. One patient in lymphoid BP had 19 of 20 diploid metaphases at the time of remission, but none of the other patients in BP had a cytogenetic response. Response rates by dose of mitoxantrone are listed in Table 4. There was no obvious trend toward better response with the higher dose of mitoxantrone. Not unexpectedly, none of the patients who had performance status greater than 2 attained remission on this regimen (Table 5).

Toxicity

As expected, significant myelosuppression was observed in all patients. The median number of days to recovery of granulocyte count $>0.5 \times 10^9$ L was 32 days in the mitoxantrone 40 mg/m^2 group (range, 19–44 days) and 38 days in the mitoxantrone 60 mg/m^2 group (range, 17–42 days). The median number of days to recovery of platelet count $>30 \times 10^9$ L

TABLE 5
Response by Performance Status

	No. of patients (%)		
PS	CR	Died	Resistant
0–1	13 (33)	11 (28)	15 (38)
2	2 (20)	2 (20)	6 (60)
3–4	0	5 (71)	2 (29)

PS: Zubrod performance status; CR: complete remission.

TABLE 6
Infections by Mitoxantrone Level

	No. of patients (%)		
Infection	40 mg/m^2	50 mg/m^2	60 mg/m^2
FUO/sepsis	13 (52)	4 (24)	5 (38)
Pneumonia	6 (24)	9 (53)	4 (31)
Aspergillus/mold	0	3 (18)	1 (8)
Herpes zoster	2 (8)	0	0

FUO: fever of unknown origin.

TABLE 7
Hepatic Toxicity by Mitoxantrone Dose Level

		Toxicity	
Mitoxantrone (mg/m^2)	No. of patients	Grade 3 (%)	Grade 4 (%)
40	24	6 (25)	7 (29)
50	20	6 (30)	5 (25)
60	11	1 (9)	3 (27)

was 31 days in the mitoxantrone 40 mg/m^2 group (range, 20–50+ days) and 47 days in the mitoxantrone 60 mg/m^2 group (range, 25–80+ days). The incidence of serious infectious complications was high (Table 6). However, there was no clear trend for a higher incidence of serious infectious complications for patients who were treated at the higher level of mitoxantrone.

Apart from myelosuppression, hepatic dysfunction, manifested primarily as hyperbilirubinemia, was the most significant dose-related toxicity observed (Table 7). Overall, 60% of the patients treated developed an increase in serum bilirubin level. A bilirubin increase >1.5–3 times normal (Grade 3 toxicity) occurred in 24% of patients. Grade 4 hyperbilirubinemia (>3 times normal) developed in 27% of all patients but was not particularly associated with dose level at the levels studied. Hepatic toxicity was reversible and no deaths due to hepatic dysfunction occurred.

Mucositis was seen infrequently, and only 1 patient had Grade 3 mucositis. Grade 3 renal toxicity

(creatinine 1.5–3 times normal) was observed in 8 patients and Grade 4 renal toxicity (creatinine >3 times normal) in 2 patients. Two of these cases were associated with tumor lysis syndrome and the remainder were associated with amphotericin administration. Grade 3 or 4 nausea and vomiting were not seen, presumably due to effective antiemetic medication. No clinical cardiac dysfunction was observed. Conjunctivitis and cerebellar toxicity were not observed.

Surprisingly, there was a tendency for improved survival among patients older than 45 years (the median age). Further analysis, however, indicated that the younger group contained more patients who were either primarily refractory to therapy or who had already experienced failure with one or more salvage regimens, both of which are known to be very poor prognostic factors.

There were two patients, both in the highest dose mitoxantrone group, who survived for longer than 2 years after being treated with FLAM. One was an AML patient with multiple relapses and diploid cytogenetics who had had an initial CR of about 2 years. She again relapsed approximately 18 months after FLAM followed by 3 consolidation courses of FLAM and achieved a CR on another regimen. The other patient received treatment for the myeloid BP of CML and reentered the chronic phase. She showed signs of disease acceleration after about 18 months and responded to splenectomy, but reentered the BP approximately 2 years after FLAM.

DISCUSSION

Published experience with high doses of mitoxantrone in the treatment of patients with acute leukemia suggested that up to 80 mg/m^2 could be combined with ara-C with acceptable toxicity.[8] Other studies indicated that the combination of fludarabine and ara-C increased the efficacy of cytarabine by increasing intracellular accumulation of ara-CTP.[6–7] The goal of this study was to evaluate the safety and efficacy of high dose mitoxantrone in combination with an established combination of both fludarabine and high dose cytarabine in the treatment of patients with relapsed acute leukemia.

The data reported from this trial have demonstrated that high doses of mitoxantrone can be combined with both fludarabine and high dose cytarabine and administered to relapsed acute leukemia patients without incurring a substantial increase in morbidity and mortality. The nonhematologic toxicity associated with this regimen was no worse that observed in patients treated with a similar regimen without the mitoxantrone[13,15] or treated with different regimens.[2–4] Similarly, the median times to CR were not different, suggesting that there was no increase in the toxicity to normal hematopoietic cells.

The CR rate among first salvage patients with ALL, AML, and the lymphoid BP of CML was comparable to previous published studies of salvage treatment of acute leukemias.[4,6,8,16,17] The differences between higher doses and intermediate doses of mitoxantrone in this study in toxicity, on the one hand, and CR rate, disease free survival, and overall survival, on the other hand, were not significant. However, even with 55 patients treated in this study, we realized that we could not compare the results of the FLAM regimen to either published studies or our own fludarabine-plus-cytarabine studies, because of the small number of patients in each treatment group and the wide divergence of known prognostic factors within each of these groups. Therefore, formal Phase II studies of FLAM in a first salvage setting for patients with ALL, AML, and myeloid and lymphoid BPs of CML are needed to define more precisely the regimen's efficacy. We would suggest that the dose of mitoxantrone at 15 mg/m^2/day for 4 days along with fludarabine 30 mg/m^2/day for 4 days and cytarabine 1000 mg/m^2/day for 4 days is a reasonable Phase II schedule. Other investigators may consider increasing the dosage of mitoxantrone even more.

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