High Antileukemic Activity of Sequential High Dose Cytosine Arabinoside and Mitoxantrone in Patients with Refractory Acute Leukemias

Results of a Clinical Phase II Study

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BACKGROUND. The current study was initiated to assess the efficacy and side effects of a timed sequential application of high dose cytosine arabinoside (AraC) in combination with mitoxantrone (S-HAM) in patients with refractory acute myeloid leukemia (AML) or acute lymphoblastic leukemia (ALL).

METHODS. Patients with refractory AML or ALL were eligible for S-HAM salvage therapy, which was comprised of AraC, 1 g/m^2 or 3 g/m^2 every 12 hours, on Days 1, 2, 8, and 9 and mitoxantrone, 10 mg/m^2/day, given on Days 3, 4, 10, and 11.

RESULTS. Of 22 fully evaluable patients, 14 patients (64%) achieved a complete remission whereas 5 patients (23%) succumbed to early death and 3 patients (14%) did not respond. Blood counts recovered at a median of 33.5 days after the start of treatment and complete remission was achieved after a median of 38 days. The median duration of complete remission was 4 months (range, 1–14 months) whereas overall survival time lasted for a median of 4.5 months (range, 1–30+ months). Treatment-associated toxicity was comprised predominantly of infection and diarrhea that reached World Health Organization Grades 3 and 4 in 64% and 32% of patients, respectively. Complementary pharmacokinetic evaluations of plasma AraC and AraU levels revealed no impact of initial AraC administration on the pharmacokinetics of subsequent AraC administrations and failed to demonstrate any evidence of self-potentiation.

CONCLUSIONS. The clinical data show the S-HAM regimen to be a promising approach for the treatment of patients with advanced acute leukemias. However, further evaluation at earlier stages of treatment is needed. ***Cancer* 1997; 79:59–68.**
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KEYWORDS: cytosine arabinoside, acute leukemia, pharmacokinetics, refractory disease.

After the major breakthroughs in the treatment of acute leukemias that were experienced in the 1970s and 1980s and a period of therapeutic stagnation in the late 1980s, new achievements have recently opened new therapeutic options for patients suffering from these disorders. Besides hematopoietic growth factors, which were shown to accelerate the recovery of granulopoiesis and reduce the incidence of severe infectious complications after intensive therapy, and the potential of cytokines to render leukemic cells more sensitive to subsequently administered cell cycle specific agents,[1–6] a better understanding of dosing and timing of antileukemic agents, and cytosine arabinoside (AraC) in particular, have substantially improved the efficacy of currently applied regimens. Hence, it could be shown that

high doses of AraC applied during induction and/or consolidation therapy have a beneficial effect and significantly improve the disease free and overall survival of patients with acute myeloid leukemia (AML).[7–10]

These developments ultimately emerged from the evaluation of high dose AraC in patients with refractory and relapsed acute leukemias as pioneered by Rudnick et al.[11] and Herzig et al.[12] and from increasing insights into the pharmacokinetics and mechanisms of action of this agent. Along these lines, the German AML Cooperative Group introduced the combination of high dose AraC and mitoxantrone (HAM) for the treatment of refractory leukemias, which revealed a high antileukemic activity.[13] The current study attempted to further improve these results by taking advantage of the self-potentiation of high dose AraC through elevated plasma levels of uracil arabinoside (AraU) as proposed by Capizzi et al.[14,15] This group showed that after administration of high dose AraC, high plasma concentrations of AraU are maintained over a prolonged period of time that inhibit the catabolism of the parent compound, resulting in a triexponential plasma decay with a long terminal half-life.[14,15] Furthermore, it was found that AraU may also induce an accumulation of cells in the S-phase and an increase in the activity of the AraC phosphorylating enzyme deoxycytidine kinase, which leads to both an enhancement of AraC incorporation into the DNA and an up to tenfold increase in its cytotoxicity.[16] These findings led to the development of a timed sequential schedule of AraC that is comprised of the application of high dose AraC on Days 1 and 2, which is followed by a second block of AraC 6 days later. Independent from these pharmacokinetic investigations, a timed sequential application of high dose AraC was also developed by Burke et al.,[17,18] who reported on a humoral stimulatory activity that was observed 4 to 8 days after a short term application of high dose AraC. This effect was shown to stimulate the growth and possibly also the recruitment of leukemic blasts into the cell cycle, rendering them more sensitive to an immediately following second series of AraC infusions.[17–19]

Based on these findings, sequentially applied AraC provided the basis for a variety of clinical studies in patients with relapsed and refractory acute leukemias, chronic myeloid leukemia, and malignant lymphomas for which complete response rates of 27–61% were reported.[20–29] Sequential AraC was also successfully applied as first-line therapy for AML.[18,29–32]

These promising reports prompted the German AML Cooperative Group to modify the previously developed HAM combination accordingly into a sequential application (S-HAM) and to evaluate its clinical efficacy in a Phase II study in patients with refractory acute leukemias. This study was complemented by measurements of AraC and AraU plasma levels to also assess the pharmacokinetic background of the sequential AraC schedule.

PATIENTS AND METHODS

Consecutive patients with refractory acute leukemias who were admitted to the Department of Hematology and Oncology of the University of Münster were eligible for the study. The diagnoses of AML and acute lymphoblastic leukemia (ALL) were based on the revised French–American–British Group criteria.[33] Refractoriness against standard chemotherapy was defined according to previously established criteria[34]: 1) primary resistance against 2 cycles of induction therapy in patients with AML or persistent leukemia after both parts of induction therapy in patients with ALL; 2) first early relapse within 6 months of first remission; 3) late relapse after more than 6 months from first remission with nonresponse to retreatment with induction type therapy; and 4) second and subsequent relapse. Patients with primary resistance or early relapse might have received nonsequential HAM for salvage therapy before and received the S-HAM combination at subsequent relapse. Patients with first relapse after a preceding remission duration $>$ 6 months were not treated with S-HAM primarily but only when they did not respond to conventional salvage therapy.

All patients were recruited from the first-line trials of the German AML Cooperative Group and the German ALL Cooperative Group, respectively.[35,36] Patients with AML had received one to two courses of the TAD-9 regimen for remission induction followed by monthly maintenance therapy.[37] First-line therapy for ALL patients was comprised of 8 weeks of induction and reinduction periods followed by long term maintenance with risk-adapted modifications in defined subgroups of patients.[36,38]

Patients older than 18 years meeting the entry criteria were enrolled into the current study and were treated by S-HAM, which was comprised of high dose AraC, 1 g/m^2 or 3 g/m^2 every 12 hours by a 3-hour infusion on Days 1, 2, 8, and 9 and mitoxantrone, 10 mg/m^2/day as a 30-minute infusion on Days 3, 4, 10, and 11, respectively (Fig. 1). To prevent high dose AraC-induced photophobia and conjunctivitis, all patients received glucocorticoid eye drops every 6 hours starting before the first dose and continuing for 24 hours after the last dose of high dose AraC. For selective gut decontamination patients received a combination of oral Co-trimoxazol® (Saarstickstoff-Fatol, Schiffweiler, Germany), colistin sulfate, and amphotericin B suspension (Heyden, Munich, Germany).

Toxicity was evaluated according to the World

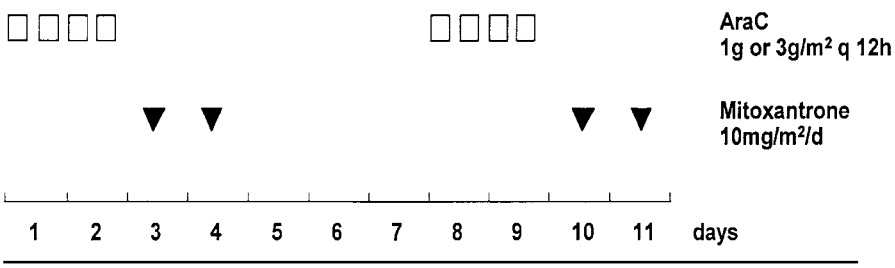

FIGURE 1. Schedule of the sequential high dose cytosine arabinoside and mitoxantrone protocol. AraC: cytosine arabinoside.

Health Organization (WHO) grading system.[39] Response to therapy was assessed according to cancer and leukemia group B (CALGB) criteria.[40] Complete remission was defined by the disappearance of leukemic blasts from the bone marrow and blood as well as from possible extramedullary sites, including the cerebral fluid, and the normalization of peripheral blood counts to thrombocytes $> 100{,}000/\mu L$ and granulocytes $> 1500/\mu L$. Patients showing 5–25% leukemic blasts within an otherwise normal bone marrow and full recovery of peripheral blood counts were considered partial remissions. Patients with $> 25\%$ leukemic blasts in the bone marrow or blood or persistence of extramedullary manifestations were classified as nonresponders, whereas early death was defined as death during therapy or within 6 weeks thereafter.

The duration of critical cytopenia was evaluated by the time for granulocyte recovery to $>500/\mu L$ and thrombocytes to $>20{,}000/\mu L$ from the onset of S-HAM treatment. The time to complete remission was measured from the onset of treatment to the date of documented complete remission and remission duration from the date of documented complete remission to relapse. Survival was measured by the time from the beginning of treatment to death.

Pharmacokinetic Evaluations

For pharmacokinetic evaluations blood samples were collected in vacutainers containing 30 U heparin and 0.1 mM tetrahydrouridine (kindly provided by Pfizer Co., Karlsruhe, Germany). Samples were taken on Days 1 and 8, respectively, before the start of AraC therapy, at 15-minute intervals during the 1st hour, and at 60-minute intervals during the 2nd and 3rd hours of the 3-hour AraC infusions. At the end of the AraC infusions, samples were taken again at 15-minute intervals during the 1st hour and at hourly intervals thereafter for an additional 9 hours.

AraC and AraU plasma levels were measured by reversed-phase high performance liquid chromatography according to a method described in more detail previously.[41] Using a Spherisorb ODS C18 column (125 $\times$ 4.6 mm; Bischoff) with an isocratic eluent comprised of 50 mM phosphate buffer (pH 6.9) with 2% methanol, the lower limit of detection of the AraC assay was 10 ng/mL ($= 0.04\ \mu M$). Measurements were evaluated by using a computerized integration of peak areas (Ramona, Nuclear Interphase, Münster, Germany).

Study Conduct

Prior to therapy, informed consent was obtained from all patients for participation in the current evaluation after they were advised regarding the purpose and investigational nature of the study as well as potential risks. The study design adheres to the declaration of Helsinki and was approved by the local ethics committee prior to its initiation.

RESULTS

Patient Characteristics

Twenty-four patients entered the study, 22 of whom were fully evaluable. Two patients were excluded because of major protocol violations. Of these 22 patients, 18 had AML and 4 had ALL. Their ages ranged from 17 to 66 years (median, 37 years). All patients had received prior chemotherapy for their disease as indicated above.

As depicted in Table 1, 16 patients had early relapses from first-line therapy and 12 of these patients received the original HAM combination as primary salvage therapy. In these patients, S-HAM was applied as second salvage treatment upon recurrence of the disease, which occured after a second complete remission duration of 2 to 4 months. Three additional patients with AML had relapsed after more than 6 months preceding complete remission duration and were refractory to conventional salvage treatment, and another 3 patients with AML had a second relapse (Table 1). All 22 patients received 1 course of S-HAM therapy.

TABLE 1
Patient Characteristics and Disease Status

Characteristics	
No.	22
Age range (yrs)	17–66 (median 37)
Female	11
Male	11
AML	18
ALL	4

Disease Status	AML	ALL
Early relapse, CR duration <6 mos	2	2
Early relapse, CR duration <6 mos and first salvage therapy with HAM	10	2
First relapse, CR duration >6 mos and nonresponse to conventional salvage therapy	3	—
≥Second relapse	3	—

AML: acute myeloid leukemia; ALL: acute lymphoblastic leukemia; CR: complete remission; HAM: high dose cytosine arabinoside and mitoxantrone.

TABLE 2
Complete Remission Rate in Relation to Disease Status and Pretreatment

Disease/pretreatment	No.	CR
AML	18	11 (61%)
ALL	4	3 (75%)
Early relapse, CR duration <6 mos	4	3 (75%)
Early relapse, CR duration <6 mos and first salvage therapy with HAM	12	7 (58%)
Late first relapse, CR duration >6 mos	3	3 (100%)
≥Second relapse	3	1 (33%)

CR: complete remission; AML: acute myeloid leukemia; ALL: acute lymphoblastic leukemia; HAM: high dose cytosine arabinoside and mitoxantrone.

Antileukemic Activity

Fourteen of the 22 evaluable patients (64%) achieved a complete remission whereas 3 patients (14%) were nonresponders. Five patients (23%) were early deaths. Analysis according to the entry criteria revealed no major differences between the four subgroups of refractory leukemias (Table 2). In particular, from the 12 patients who had received prior therapy with nonsequentially administered HAM, 7 (58%) achieved a complete remission after S-HAM treatment (Table 2).

In patients achieving complete remission, recovery of blood counts to thrombocytes $> 20{,}000/\mu$L and granulocytes $> 500/\mu$L occurred at a median of 33.5 days from the start of therapy (range, 26–42 days). Complete remission was verified after a median of 38 days (range, 27–47 days). Prolonged aplasia was not observed in any patient (Table 3). From the 14 patients achieving a complete remission, 3 underwent autologous bone marrow transplantation. Five patients were in ongoing remission at 1^+, 2^+, 2^+, 2^+, and 3^+ months, respectively, whereas 6 patients relapsed after 1, 2.5, 4, 6, 8.5, and 14 months, respectively. The median duration of complete remission was 4 months, and ranged from 1 to 14 months. The median overall survival for all patients was 4.5 months and ranged from 1 to 30^+ months (Table 3).

TABLE 3
Remission Characteristics

	Median	Range
Duration of critical cytopenia	33.5 days	26–42 days
Time to complete remission	38 days	27–47 days
Remission duration	4 mos	1–14 mo
Survival	4.5 mos	$1–30^+$ mo

Side Effects

The side effects that were encountered during S-HAM therapy are summarized in Table 4. The most frequent side effects were nausea/vomiting, infections, diarrhea, and mucositis, which were found in 19 (86%), 18 (82%), 15 (68%), and 10 (45%) courses, respectively. Severe infections according to WHO Grade 3/4 occurred in 14 patients (64%), whereas severe nausea/vomiting, diarrhea, and mucositis was observed in 4 (18%), 7 (32%), and 4 patients (18%), respectively. Major hepatic (1 patient [5%]) toxicity of WHO Grade 3/4 as well as less severe hepatic (6 patients [27%]), renal (2 patients [9%]), cardiac (3 patients [14%]), and skin (4 patients [18%]) toxicity were observed less often (Table 4).

Pharmacokinetic Evaluations

Pharmacokinetic evaluations were performed in 11 patients during S-HAM therapy. As shown in Figures 2 and 3, AraC levels declined after the end of the first infusion in a biphasic manner whereas the decrease of AraU levels occured monophasically. No evidence for a third phase of plasma decay was found for AraC and no measurable AraC concentrations could be detected after 12 hours (i.e., at the beginning of the second AraC infusion). AraC measurements were, therefore, fitted to a two-compartment model and those of AraU to a one-compartment model. The resulting

TABLE 4
Side Effects of S-HAM Therapy

Toxicity	No. (%)	WHO Grade 1/2 (%)	WHO Grade 3/4 (%)
Nausea/vomiting	19 (86)	15 (68)	4 (18)
Infection	18 (82)	4 (18)	14 (64)
Diarrhea	15 (68)	8 (36)	7 (32)
Mucositis	10 (45)	6 (27)	4 (18)
Hepatic	7 (32)	6 (27)	1 (5)
Renal	2 (9)	2 (9)	—
Cardiac	3 (14)	3 (14)	—
Skin	4 (18)	4 (18)	—
CNS	2 (9)	2 (9)	—

S-HAM: sequential application of high dose cytosine arabinoside and mitoxantrone; WHO: World Health Organization; CNS: central nervous system.

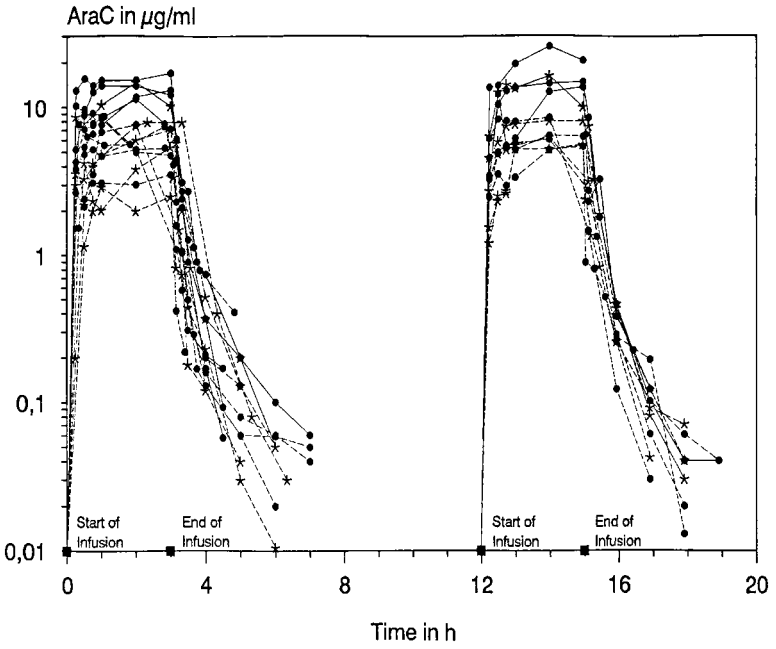

FIGURE 2. Plasma cytosine arabinoside (AraC) kinetics during high dose AraC therapy with 1 g/m^2 or 3 g/m^2. The interrupted and intact lines represent the kinetics after 1 g/m^2 and 3 g/m^2 AraC, respectively. Points and stars show the measurements during Day 1 and Day 8, respectively.

pharmacokinetic parameters are recorded in Table 5. A comparison of the plasma levels of AraC and AraU during the first infusion with those during the following infusions demonstrates uniform patterns of plasma decay curves. No evidence could be found for a nonlinear accumulation of either substance due to a saturation of metabolism or of renal elimination. Accordingly, no significant differences between the pharmacokinetics during the first infusion and the following applications were detected (Table 6) (Fig. 4). The combined evaluation of measurements from all days is summarized in Tables 6 and 7. Further support for the existence of linear pharmacokinetics comes from the great similarity of dose-independent parameters after application of either 1 g/m^2 or 3 g/m^2 AraC, whereas dose-dependent parameters were 3 times higher after 3 g/m^2 AraC.

One patient was an early death due to renal failure, which developed during the first AraC infusion. As would be expected based on the fact that AraU is mainly excreted via the kidneys, the AraU levels declined very slowly, indicating a change from linear to

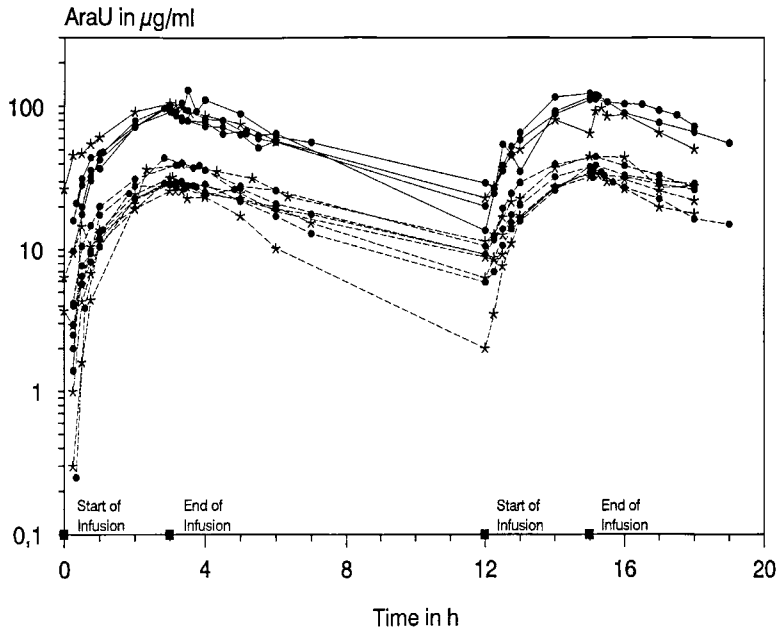

FIGURE 3. Plasma uracil arabinoside (AraU) kinetics during high dose cytosine arabinoside (AraC) therapy with 1 g/m^2 or 3 g/m^2. The interrupted and intact lines represent the kinetics after 1 g/m^2 and 3 g/m^2 AraC, respectively. Points and stars show the measurements during Day 1 and Day 8, respectively.

TABLE 5
Pharmacokinetic Parameters of AraC (A) and AraU (B) during the First AraC Infusion

		AUC in µg*hr/mL			
$t_{1/2\alpha}$ (hr)	$t_{1/2\beta}$ (hr)	(1 g/m^2)	(3 g/m^2)	V_{ss} in L	Cl_{total} in mL/min
0.09	1.07	15	29	36	2183
SD 42%	51%	19%	31%	32%	22%

	C_{max} in µg/mL		AUC in µg*hr/mL			
$t_{1/2}$ (hr)	(1 g/m^2)	(3 g/m^2)	(1 g/m^2)	(3 g/m^2)	V_{SS} in L	Cl_{total} in mL/min
3.77	37	84	253	687	41	131
SD 16%	18%	50%	24%	12%	16%	24%

$t_{1/2\alpha}$: initial half-life; $t_{1/2\beta}$: terminal half-life; $t_{1/2}$: half-life; C_{max}: peak concentration; AUC: area under curve; V_{SS}: volume of distribution during steady state; Cl_{total}: total clearance; Arac: cytosine arabinoside; AraU: uracil arabinoside; SD: standard deviation.

Measurement of plasma levels were performed during and up to 9 hours after the end of the first 3-hour-infusion of 1 g/m^2 and 3 g/m^2 AraC, respectively. Mean values and standard deviations are given.

nonlinear kinetics. In contrast, the AraC levels declined biphasically (terminal half-life = 0.57 hours) and appeared unchanged.

DISCUSSION

Approaches to further improve the efficacy of antileukemic therapy are often based on the introduction of new agents or the application of new dosages. However, they may also take advantage of deeper insights into the pharmacokinetics and metabolism of cytostatic agents, which may translate into a more effective timing and scheduling of antileukemic treatment. Among currently used antileukemic agents, AraC has been the main target drug for such efforts and various schedules have been proposed by different groups.

Capizzi et al. were among the first to suggest a

TABLE 6
Pharmacokinetic Parameters of Cytosine Arabinoside during All Cytosine Arabinoside Infusions and Comparison of Values during the First and Second Infusion

Dose AraC	$t_{1/2\alpha}$ (hr)	$t_{1/2\beta}$ (hr)	AUC in μg*hr/mL	V_{ss} in L	Cl_{total} in mL/min
1 g/m^2	0.11	0.87	16	36	2131
SD	58%	58%	26%	39%	25%
3 g/m^2	0.10	0.78	34	40	2295
SD	44%	45%	24%	25%	21%
Total	0.11	0.83	—	38	2193
SD	54%	55%	—	34%	24%
Values of first over second infusion	1.01	0.93	1.19	1.05	1.07
SD	38%	65%	53%	23%	20%

$t_{1/2\alpha}$: initial half-life; $t_{1/2\beta}$: terminal half-life; $t_{1/2}$: half-life; C_{max}: peak concentration; AUC: area under curve; V_{ss}: volume of distribution during steady state; Cl_{total}: total clearance; AraC: cytosine arabinoside; AraU: uracil arabinoside; SD: standard deviation.
Measurements of plasma levels were performed during and up to 9 hours after the end of the first 3-hour-infusion of 1 g/m^2 and 3 g/m^2 AraC, respectively. Mean values and standard deviations are given.

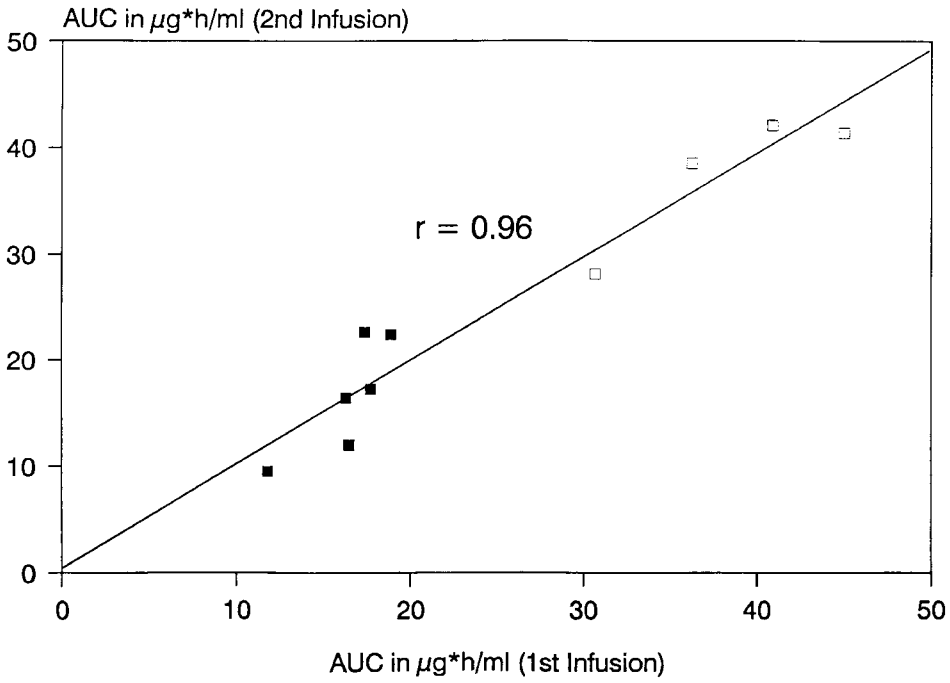

FIGURE 4. Regression analysis of areas under the curve of cytosine arabinoside (AraC) plasma levels for first versus second infusion. Closed and open symbols represent measurements during 1 g/m^2 and 3 g/m^2 AraC, respectively. AUC: area under curve; r: coefficient of correlation.

timed sequential application of AraC, which was found to exert a self-potentiating effect through the interference of high levels of AraU with the metabolism and/or renal elimination of AraC[14,15] and an AraU-induced arrest of cells in S-phase.[16] Independent from this approach, a timed sequential application of AraC was also developed by Burke et al. based on their finding of a humoral stimulatory activity that was observed several days after a first application of high dose AraC.[17,18]

These data prompted the German AML Cooperative Group to modify the previously introduced comination of high dose AraC with mitoxantrone (HAM) accordingly and to assess the efficacy and side effects of

TABLE 7
Pharmacokinetic Parameters of AraU during All AraC Infusions

Dose AraC	$t_{a/2}$ in hr	C_{max} in μg/mL	AUC in μg hr/mL	V_{SS} in L	Cl_{total} in mL/min
1 g/m^2	4.02	37	249	44	134
SD	20%	18%	26%	18%	30%
3 g/m^2	3.82	86	682	43	133
SD	18%	44%	11%	14%	21%
Total	3.95	—	—	44	134
SD	20%	—	—	17%	27%

$t_{1/2\alpha}$: initial half-life; $t_{1/2\beta}$: terminal half-life; $t_{1/2}$: half-life; C_{max}: peak concentration; AUC: area under curve; V_{SS}: volume of distribution during steady state; Cl_{total}: total clearance; AraC: cytosine arabinoside; AraU: uracil arabinoside; SD: standard deviation.
Measurements of plasma levels were performed during and up to 9 hours after the end of the first 3-hour-infusion of 1 g/m^2 and 3 g/m^2 AraC, respectively. Mean values and standard deviations are given.

the sequentially applied S-HAM regimen in a clinical Phase II study in patients with refractory AML and ALL. The high rate of 64% complete remissions achieved in the 22 evaluable patients indicates a high antileukemic activity of the S-HAM combination, especially when taking into account that all patients had refractory disease and were heavily pretreated even with the identical combination of HAM, although it was not given in a timed sequential manner. The latter finding in particular strongly suggests that timed sequential administration may increase the antileukemic activity of high dose AraC, although this conclusion is not based on a prospective randomized comparison. However, the obtained data also compare favorably with other AraC-based salvage regimens, some of which included other combination partners such as etoposide or idarubicin.[13,42–56]

In addition to its use in patients with refractory and relapsed acute leukemias, a timed sequential schedule of antineoplastic chemotherapy was also applied for the treatment of chronic myeloid leukemia and malignant lymphomas, with complete remission rates of 27% to 61%, respectively, achieved.[20–29]

In spite of the apparent increase in antileukemic activity, the S-HAM combination was not hampered by more severe side effects and toxicity. Severe infections, diarrhea, and mucositis comprised the major side effects besides nausea and vomiting, which was experienced by the majority of patients. Time for the recovery of blood counts from critical cytopenia as well as time to complete remission were 33.5 and 38 days, respectively, approximately 5 to 6 days longer than previously reported for the nonsequential HAM combination.[13]

The complementary pharmacokinetic evaluation that was performed in 11 patients showed no evidence of a triexponential plasma decay curve of AraC as previously reported.[14,15] It also revealed no influence of prolonged AraU levels on the subsequent administration of AraC. Hence, the measurements of AraC and AraU plasma kinetics revealed no differences in pharmacokinetic parameters between the AraC infusions on Day 1 compared with Day 8 of the S-HAM regimen. Similarly, a comparison of the terminal half-lives ($t_{1/2\beta}$) and total clearances (Cl_{total}) during the first AraC infusion (AraC: $t_{1/2\beta}$ = 1.07 hours, Cl_{total} = 2183 mL/min; AraU: $t_{1/2\beta}$ = 3.77 hours, Cl_{total} = 131 mL/min) with those calculated for all infusions (AraC: $t_{1/2\beta}$ = 0.83 hours, Cl_{total} = 2193 mL/min; AraU: $t_{1/2}$ = 3.95 hours, Cl_{total} = 134 mL/min) shows almost identical values for either substance. Accordingly, there were no changes in the AUCs as depicted in Tables 5, 6, and 7, respectively. No evidence for a nonlinear accumulation of either substance was observed, not even on Day 8. As further supported by the similarity of dose-independent parameters for both dosages, 1 g/m^2 and 3 g/m^2 AraC, and three times higher values for dose-dependent parameters for 3 g/m^2 AraC, linear pharmacokinetics existed during all days of the S-HAM therapy without any changes in the AraC elimination.

The pharmacokinetic evaluation of the AraC and AraU levels in the patient with renal failure showed a substantially prolonged decline of AraU. Nonetheless, the decline of AraC levels resembled that in other patients and was not influenced in any way by longer lasting AraU levels, indicating that the elimination pathways of AraC are not saturated by AraU, not even during the persistence of high AraU levels for a duration that does not occur normally.

However, the lack of support for the concept of a pharmacokinetically based self-potentiation of AraC should not distract from the high clinical activity of the timed sequential application of S-HAM. This favorable effect may be based on other mechanisms such as the modulation of intracellular pharmacokinetics or the priming effect of the humoral stimulatory plasma activ-

ity as described by Burke et al.[17,18] Although these effects were not investigated in the current study, the clinical results encourage further evaluations and also warrant the assessment of S-HAM in a larger number of patients with relapsed and refractory disease and may even lead to its introduction as first-line therapy for AML.

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