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Phase I trial of gemtuzumab ozogamicin in intensive combination chemotherapy for relapsed or refractory adult acute myeloid leukemia (AML): Japan Adult Leukemia Study Group (JALSG)-AML206 study

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In order to investigate better molecular-target therapy for acute myeloid leukemia (AML), we conducted a phase I trial of a combination of gemtuzumab ozogamicin (GO) with conventional chemotherapy. Between January 2007 and December 2009, a total of 19 adult Japanese patients with relapsed or refractory CD33-positive AML (excluding acute promyelocytic leukemia) were enrolled. All registered patients received a standard dose of cytarabine (Ara-C) (100 mg/m^2 × 7 days), combined with either idarubicin (IDR) (10–12 mg/m^2 × 3 days) or daunorubicin (DNR) (50 mg/m^2 × 3–5 days), and then GO (3–5 mg/m^2), which was administered 1 day after the last infusion of IDR (IAG regimen) or DNR (DAG regimen). While doses of both GO and IDR and the administration period of only DNR were increased, the dose-limiting toxicity (DLT) was assessed. Among 19 patients (nine in the IAG regimen, 10 in the DAG regimen), the median age was 59 years (range 33–64), and the relapsed/refractory ratio was 13/6. In the therapy using 3 mg/m^2 GO in the IAG or DAG regimen, grade 3/4 leukopenia and neutropenia were observed in all patients, but none had grade 3/4 non-hematological toxicities, except febrile neutropenia. Three patients in the IAG regimen who were administered 5 mg/m^2 GO showed DLT. No patients had veno-occlusive disease or sinusoidal obstructive syndrome. In conclusion, 3 mg/m^2 GO combined with Ara-C and IDR or DNR can be safely administered, and phase II trials should be conducted to investigate the clinical efficacy of the combination therapy. (*Cancer Sci* 2011; 102: 1358–1365)

Current standard induction treatment for acute myeloid leukemia (AML) involves drug regimens with two or more agents that include an anthracycline or anthraquinone and cytarabine (Ara-C).[1–6] A recent clinical trial of the Japan Adult Leukemia Study Group (JALSG) for younger adult patients (16–64 years of age) with newly diagnosed AML showed a 77.9% complete remission (CR) rate.[4] Remission rates achieved by us and others range approximately 55–90% in adult patients, depending on the composition of the population treated.[1–6] However, these high CR rates did not always translate into improved outcomes for patients, mainly because approximately 40–50% eventually relapsed. Although there are various clinical trials for patients with relapsed or refractory AML, the probability of a second CR is approximately 50% in younger patients, but the duration of CR is nearly always much shorter than the first CR. No standard chemotherapy regimen provides a high rate and durable CR for patients with relapsed/refractory AML, and all such patients should be considered eligible for clinical trials if available.[7]

Among newer antileukemia agents being examined for the treatment of AML, an antibody to CD33 antigen is one of the most promising drugs. The CD33 antigen is expressed on 80–90% of AML blasts and acts as a target for antibody-mediated destruction. Gemtuzumab ozogamicin (GO) is a recombinant humanized anti-CD33 monoclonal antibody conjugated to calicheamicin (a cytotoxin), which is 1000 times as potent as doxorubicin.[8,9] This conjugated antibody is rapidly internalized and causes subsequent apoptosis.[10] GO was shown to be effective in patients with relapsed AML in nonrandomized studies and gained regulatory approval in the United States (the US Food and Drug Administration [FDA]) for relapsed older patients (older than 60) with AML.[11,12] GO was also approved by the Japanese government in 2005 for use in patients with relapsed/refractory AML, but only for monotherapy based on a phase I/II study for Japanese patients.[13] GO does not cause alopecia or mucositis, even though it causes myelosuppression, an infusional syndrome, and liver damage such as hyperbilirubinemia and/or hepatic transaminitis (or elevation of transaminase). Several studies have indicated that GO combined with conventional chemotherapy would provide a more potent antileukemia effect than GO monotherapy.[14–19] We considered that addition of GO to conventional chemotherapy in induction therapy would improve the clinical outcome of AML patients of all ages. To find the optimal usage of GO in combination with conventional chemotherapy for relapsed or refractory AML, we conducted a phase I study. Here we report the results of this JALSG-AML206 trial in adult patients with relapsed or refractory AML, younger than age 65, in which the dosage of GO, combined with our two types of standard remission induction therapy for de novo AML, were tested.[4]

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This study was registered at UMIN Clinical Trials Registry (http://www.umin.ac.jp/ctr/index-j.htm) as UMIN000001141 and UMIN000001142.

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Materials and Methods

Patient eligibility. Between January 2007 and December 2009, 19 eligible patients with relapsed and refractory AML were enrolled in the present study. The inclusion criteria were as follows: (i) diagnosed as $CD33^+$AML (excluding acute promyelocytic leukemia); (ii) relapsed ≥6 months after the first CR (CR1) or were refractory to initial standard induction therapy; (iii) age: 20–64 years old; (iv) 0–2 by the Eastern Cooperative Oncology Group (ECOG) performance status; (v) no active double cancer; (vi) adequate cardiac, renal and hepatic function with left ventricular ejection fraction ≥50%, creatinine ≤2.0 mg/dL, bilirubin ≤1.5 mg/dL; (vii) no uncontrolled infection; and (viii) no human immunodeficiency virus (HIV) infection. Patients who received more than 500 mg/m^2 of daunorubicin (DNR) in a prior therapy were ineligible to DNR-including protocol. Cytogenetic abnormalities were grouped by standard criteria and classified according to the UK Medical Research Council (MRC) classification.[20]

Study design. The study was conducted by six designated institutions among JALSG members, and consisted of two parts: idarubicin (IDR), Ara-C plus GO (IAG regimen), and DNR, Ara-C plus GO (DAG regimen). The treatment schedules of both regimens are shown in Figure 1.

IAG regimen. The starting doses (level 1) consisted of IDR 10 mg/m^2 administered intravenously (d.i.v.) over 30 min daily for three consecutive days (days 1–3), Ara-C 100 mg/m^2 as a continuous intravenous infusion (c.i.v.) for seven consecutive days (days 1–7) and GO 3 mg/m^2 for 2 h d.i.v. on day 4. While the dose and schedule of Ara-C were fixed, doses of IDR and GO were increased in levels 2 and 3 as shown in Figure 1.

DAG regimen. The starting doses (level 1) consisted of DNR 50 mg/m^2 administered d.i.v. over 30 min daily for three consecutive days (days 1–3), Ara-C 100 mg/m^2 c.i.v. (days 1–7) and GO 3 mg/m^2 for 2 h d.i.v. on day 4. While the dose and schedule of Ara-C were fixed, doses of DNR and GO were scheduled to increase in levels 2, 3 and 4 (Fig. 1).

All patients were hospitalized during therapy and received optimal supportive care. For prophylaxis of GO infusion reaction, antihistamines and corticosteroids were given 1 h before the infusion. Granulocytopenic patients were placed in single rooms with conventional isolation or in laminar airflow rooms. Broad-spectrum antibiotics were given for fever higher than 38°C in the presence of granulocytopenia, and were continued until defervescence and recovery of granulocyte counts above 0.5×10^9/L. Random donor platelet concentrates were administered to maintain a platelet count above 20×10^9/L. Packed red blood cell (RBC) transfusions were performed to maintain hemoglobin above 7.0 g/dL.

Response criteria. Responses were evaluated according to the recommendations of the International Working Group.[21] A CR was defined as disappearance of all clinical and/or radiological evidence of disease with ≤5% marrow blasts, neutrophil (ANC) count $\geq 1 \times 10^9$/L and platelet (PLT) count $\geq 100 \times 10^9$/L. A CR without PLT recovery (CRp) had identical marrow results and ANC recovery as for CR, but with PLT $<100 \times 10^9$/L and $\geq 20 \times 10^9$/L. Partial remission consisted of a peripheral blood recovery as for CR, but with a decrease in marrow blasts of ≥50% compared with baseline before therapy, and not more than 6–25% blasts in the marrow. All other responses were considered failures. After the IAG or DAG treatment, patients received the most appropriate AML therapy determined by their individual physicians.

Adverse events/toxicities. During the entire period of induction, blood cell counts were performed daily and liver and renal

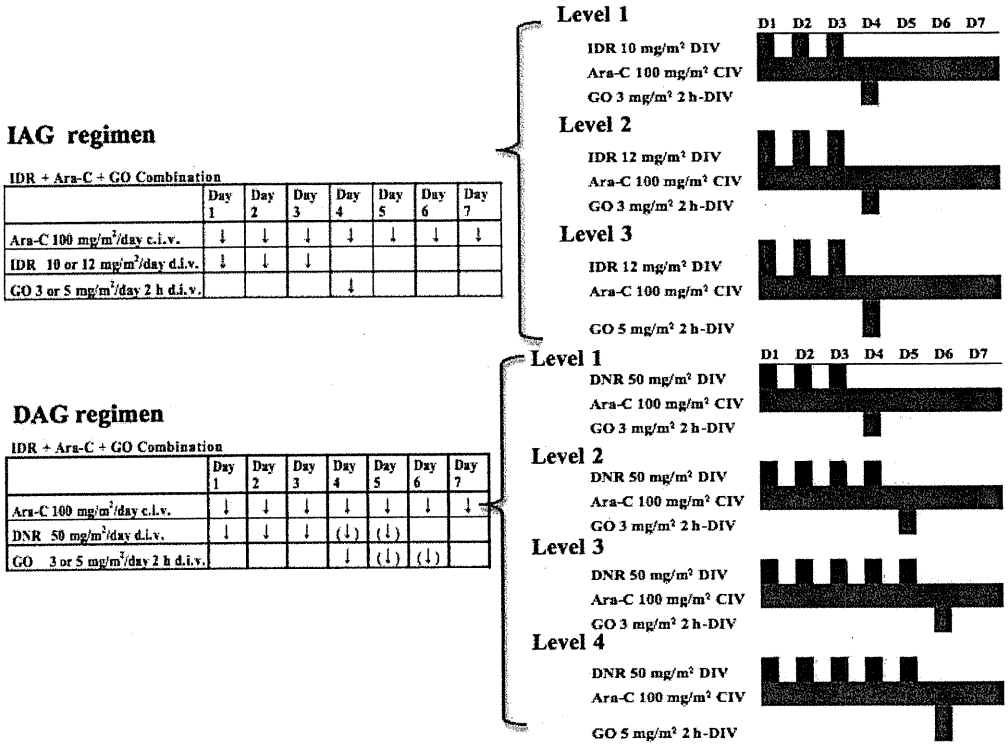

Fig. 1. Treatment schedule of the Japan Adult Leukemia Study Group (JALSG)-AML206 study. Ara-C, cytarabine; CIV and c.i.v., continuous intravenous infusion; DIV and d.i.v., drip intravenous infusion; DNR, daunorubicin; GO, gemtuzumab ozogamicin; IDR, idarubicin.

Table 1. Patient characteristics

	Overall (*n* = 19)	IAG regimen (*n* = 9)	DAG regimen (*n* = 10)
Male:Female	9:10	4:5	5:5
Age (years)†	59 (33–64)	61 (52–64)	58 (33–62)
≤60	10	3	7
>60	9	6	3
WBC ($\times 10^9$/L)†	3.0 (1.0–39.2)	3.7 (2.6–39.2)	2.05 (1.0–25.3)
Blast (%)†	42.8 (7.9–96.8)	56.4 (17.3–88.0)	29.9 (7.9–96.8)
CD33 positivity in blast (%)†	89.4 (39.0–100)	92.9 (62.8–100)	80.6 (39.0–96.9)
Disease status			
Relapsed/Refractory	13/6	7/2	6/4
FAB type (no. patients)			
M0	1		1
M1	3	2	1
M2	8	3	5
M4	6	3	3
M5	1	1	
Cytogenetic group (no. patients)			
Favorable	2	1	1
Intermediate	11	6	5
Adverse	6	2	4
Performance status (no. patients)			
0	1	0	1
1	18	9	9

†Median value and range in parentheses. FAB, French-American-British Classification; WBC, white blood cells.

blood tests three times weekly. Electrocardiography (ECG) was also performed once a week.

Hematological and non-hematological toxicity was graded according to the Common Terminology Criteria for Adverse Events (CTCAE) ver 3.0, National Institutes of Health.

Statistical analysis. The primary objective of the study was to determine the maximum tolerated dose (MTD) and dose-limiting toxicity (DLT) of GO in combination with standard chemotherapy in Japanese patients. Dose escalation of anthracycline and GO in the IAG or DAG treatment followed a standard 3 + 3 phase I design in which cohorts of three patients at a time were treated at a dose and schedule level. If no DLT was observed, the next cohort was escalated to the next level. If one or two of the first three patients experienced a DLT, up to a total of six patients were enrolled at the same dose level. The next cohort was escalated only if a total of less than two patients presented with a DLT. If three of the first three patients experienced a DLT, the dose-escalation was stopped and the prior dose level was considered the MTD.

All ≥grade 3 drug-related nonhematological toxicities that occurred after treatment were considered DLT, with the exception of nausea and vomiting (if manageable with supportive care), alopecia, drug-related fevers, asymptomatic abnormalities of lactate dehydrogenase, alkaline phosphatase, disturbances of electrolytes and febrile neutropenia (FN) as these are common events in patients with relapsed AML.

Myelosuppression was not considered a DLT except for prolonged bone marrow aplasia longer than 6 weeks (or 42 days). Secondary objectives were to evaluate the efficacy of these treatment regimens.

The study was approved by the Institutional Review Board at each participating institution. Written informed consent was obtained from all patients before registration in accordance with the Declaration of Helsinki. The study was registered at the University Hospital Medical Information Network (UMIN) Clinical Trials Registry (http://www.umin.ac.jp/ctr/) as UMIN000001141 and UMIN000001142.

Results

Patient characteristics. A total of 19 patients with relapsed or refractory $CD33^+$AML were enrolled and evaluated (Table 1). The median age of patients was 59 years (range 33–64), the male/female ratio was 9/10, and the relapsed/refractory ratio was 13/6. The median value of blasts in the bone marrow before treatment was 42.8% (range 7.9–96.8%), and the median expression of CD33 antigen was 89.4% (range 39–100%). Patient characteristics in the IAG and DAG groups were similar, with the exception of age. Patients older than 60 years were more frequently enrolled in the IAG regimen. Among adverse cytogenetic groups, four patients had complex karyotypes (two in each group), one had t(6:9) in the DAG group and one had inv(5)del(7) in the DAG group.

Safety. *In the IAG regimen.* Hematological toxicities were commonly observed as expected for re-induction therapy (Table 2). Levels of white blood cells (WBC) at the time of GO administration tended to be lower than 3.0×10^9/L and those of ANC were $<1.5 \times 10^9$/L. Grade 4 leukopenia and neutropenia

Table 2. IAG regimen: hematological toxicities

	Level 1 (*n* = 3) (IPt-1/IPt-2/IPt-3)	Level 2 (*n* = 3) (IPt-4/IPt-5/IPt-6)	Level 3 (*n* = 3) (IPt-7/IPt-8/IPt-9)
WBC ($\times 10^9$/L) at GO administration	2.4/1.1/5.4	1.3/0.4/2.3	0.8/1.2/3.0
WBC (grade 3/4)	0/3	0/3	0/3
Days to nadir after GO administration	4/6/13	10/5/10	6/5/7
ANC ($\times 10^9$/L) at GO administration	1.7/1.5/4.4	0.5/0/1.0	0.3/0.2/0.4
ANC (grade 3/4)	0/3	0/3	0/3
Days to nadir after GO administration	11/6/10	7/5/7	6/13/7
Days toward ANC recovery	31/35/26	24/34/35	42/38/24
PLT ($\times 10^9$/L) at GO administration	62/64/146	24/51/159	87/23/44
PLT (grade 3/4)	3/0	2/1	2/1
Days to nadir after GO administration	8/8/14	10/10/14	11/5/14
PLT transfusion (units)	90/130/100	130/130/50	70/220/70
Days toward PLT recovery	31/NA/NA	NA/43/35	25/87/31
Hemoglobin (grade 0/1/2/3/4)	0/1/2/0/0	0/1/2/0/0	1/1/1/0/0
RBC transfusion (units)	4/4/12	4/6/2	8/16/4

ANC, neutrophils; GO, gemtuzumab ozogamicin; NA, data was not available because the next treatment proceeded before platelet recovery due to disease progression; PLT, platelets; RBC, red blood cells; WBC, white blood cells.

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Table 3. IAG regimen: non-hematological toxicities

	Level 1 (n = 3)	Level 2 (n = 3)	Level 3 (n = 3)
Febrile neutropenia (grade 0/3/4)	0/3/0	1/2/0	0/2/1
Sepsis (grade 4)	0	0	1
Cerebral abscess (grade 4)	0	0	1
Hepatic toxicity (grade 0/1/2/3)	2/0/1/0	2/0/1/0	1/0/1/1
Nausea and vomiting (grade 0/1/2/3)	3/0/0/0	3/0/0/0	1/0/2/0
Diarrhea (grade 0/1/2/3)	3/0/0/0	3/0/0/0	2/1/0/0
Edema (grade 0/1/2/3)	3/0/0/0	3/0/0/0	2/1/0/0
Skin rash (grade 0/1/2/3)	3/0/0/0	3/0/0/0	2/0/1/0
VOD/SOS	0	0	0

SOS, sinusoidal obstructive syndrome; VOD, veno-occlusive disease.

was observed in all patients. Days to nadir of ANC after GO administration were 5–13 days, and days toward ANC-recovery were 24–42 days. As one patient in level 3 (IPt-7) did not recover from neutropenia for 42 days (6 weeks), we regarded this prolongation of neutropenia as a DLT.

All patients had grade 4 thrombocytopenia and required plenty of PLT transfusion. Some patients took more than 30 days to recover to at least the initial level of PLT. As one patient in level 3 (IPt-8) required 220 units of PLT transfusion and took 87 days for recovery without disease progression, we regarded this prolongation of thrombocytopenia as a DLT.

Among non-hematological toxicities (Table 3), febrile neutropenia (FN) was common and severe. One patient in level 3 (IPt-9), although eventually recovered and attained CR, suffered from grade 4 neutropenia, sepsis and brain abscess. We regarded this FN with an infectious episode as a DLT.

Most non-hematological toxicities other than FN were clinically manageable and none of the patients had grade 4 hepatic toxicity, veno-occlusive disease (VOD) or sinusoidal obstructive syndrome (SOS).

In the DAG regimen. Grade 4 leukopenia and neutropenia was observed in all patients (Table 4). All except one patient in level 2 (DPt-6) recovered within 5 weeks. Grade 3/4 of thrombocytopenia was also observed in all patients, and plenty of PLT transfusion was required. The majority of patients recovered from thrombocytopenia within 5 weeks except one patient (DPt-6) who died of central nervous system (CNS) bleeding due to progression of leukemia within 30 days. The patient, DPt-6, was a 60-year-old man who was refractory to initial induction therapy. His leukemic blasts were reduced 47% in his bone marrow (BM) on day 15 of DAG level 2 (10 days after GO) and 4% in his peripheral blood (PB) on day 19. However, the duration of his response was short as his blasts rapidly increased to 85.2% in the BM on day 23 and 57% in PB on day 26. He suffered from disseminated intravascular coagulation (DIC) and eventually CNS bleeding on day 26, although the platelet count was maintained at >40 × 10^9/L. Autopsy confirmed that progression of leukemia was the cause of his death without any clinical effect of the chemotherapy.

Among the non-hematological toxicities (Table 5), although FN was common and severe, none of the patients developed fatal infection, or had VOD or SOS. None of the grade 4 non-hematological toxicities developed either. As all patients in level 3 of the IAG regimen had DLT as mentioned above, the safety review board (SRB) recommended that level 4 of the DAG should be cancelled, because 5 mg/m^2 GO would be too toxic in combination with chemotherapy. Our previous study[4] indicated that the dose and schedule of DNR of level 3 of the DAG is equally effective and intensive as those of IDR of levels 2 and 3 in the IAG. Therefore, we considered that adding 5 mg/m^2 of GO to DNR + Ara-C (level 4 of the DAG) would be as toxic as level 3 of IAG, and accepted the recommendation of the SRB.

Antileukemic activity. A CR was achieved in nine of 19 patients and one attained a CRp, making the overall response rate 52.6%. In addition, two patients obtained partial remission, and four patients showed blast clearance, but three patients were resistant to therapy (Table 6). CR/CRp was observed in all levels of IAG and DAG. A CR was obtained in two patients with adverse karyotypes such as t(6:9) and complex. The rate of

Table 4. DAG regimen: hematological toxicities

	Level 1 (n = 3) (DPt-1/DPt-2/DPt-3)	Level 2 (n = 4) (DPt-4/DPt-5/DPt-6/DPt-7)	Level 3 (n = 3) (DPt-8/DPt-9/DPt-10)
WBC (×10^9/L) at GO administration	1.1/1.2/0.7	2.4/1.7/0.5/0.3	0.6/1.7/2.0
WBC (grade 3/4)	0/3	0/4	0/3
Days to nadir after GO administration	7/10/7	7/11/3/8	3/5/7
ANC (×10^9/L) at GO administration	0.4/0.6/0.2	0.2/1.3/0.2/0.0	0.4/1.0/1.2
ANC (grade 3/4)	0/3	0/4	0/3
Days to nadir after GO administration	7/8/7	11/13/5/8	7/7/12
Days toward ANC recovery	26/29/33	23/18/NA/28	34/24/26
PLT (×10^9/L) at GO administration	361/47/122	71/199/53/3	32/147/193
PLT (grade 3/4)	2/1	3/1	3/0
Days to nadir after GO administration	11/11/17	13/13/17	14/8/17
PLT transfusion (units)	150/60/90	50/70/110/170	170/60/40
Days toward PLT recovery	22/39/31	32/20/NA/26	28/29/21
Hemoglobin (grade 0/1/2/3/4)	0/3/0/0/0	2/1/1/0/0	0/1/2/0/0
RBC transfusion (units)	18/0/6	0/6/10/10	6/6/8

ANC, neutrophils; NA, data was not available because of central nervous system bleeding due to disease progression before ANC and PLT recovery; PLT, platelets; RBC, red blood cells; WBC, leukocytes.

Table 5. DAG regimen: non-hematological toxicities

Toxicity	Level 1 (n = 3)	Level 2 (n = 4)†	Level 3 (n = 3)
Febrile neutropenia (grade 0/3/4)	0/2/1	1/3/0	1/2/0
Hepatic toxicity (grade 0/1/2/3)	2/0/1/0	3/0/1/0	1/1/1/0
Nausea and vomiting (grade 0/1/2/3)	2/0/1/0	4/0/0/0	2/0/1/0
Colitis (grade 0/1/2/3)	2/0/1/0	4/0/0/0	3/0/0/0
Diarrhea (grade 0/1/2/3)	3/0/0/0	4/0/0/0	2/0/1/0
Cardiac (grade 0/1/2/3)	3/0/0/0	3/0/1/0	2/0/1/0
VOD/SOS	0	0	0

†One patient in level 2 died of CNS bleeding due to disease progression. SOS, sinusoidal obstructive syndrome; VOD, veno-occlusive disease.

Table 6. Response

	IAG regimen			DAG regimen			Overall	
	Level 1 (n = 3)	Level 2 (n = 3)	Level 3 (n = 3)	Level 1 (n = 3)	Level 2 (n = 4)	Level 3 (n = 3)	(n = 19)	
CR	1	2	1	1	3	1	9	52.6%
CRp			1				1	
PR				1		1	2	
Blast clearance	1		1	1		1	4	
Resistant disease	1	1			1		3	

CR, complete remission; CRp, CR without platelet recovery; PR, partial remission.

Table 7. Response according to patient characteristics

Overall response (CR + CRp)	10/19 (52.6%)
Disease status	
Relapsed	8/13 (61.5%)
Refractory	2/6 (33.3%)
Cytogenetic group	
Favorable	1/2 (50.0%)
Intermediate	7/11 (53.6%)
Adverse	2/5 (40.0%)

CR, complete remission; CRp, CR without platelet recovery.

response tended to be higher in relapsed patients (61.4%) than in patients refractory to initial therapy (33.3%) (Table 7).

Discussion

As Kell *et al.*[19] suggested, the development of antibody-directed chemotherapy with more specificity against leukemic blasts has been one of the goals of cancer treatments for several years. CD33 antigen has emerged as a favored target epitope because it is expressed in over 80–90% of AML blasts.[22] Although unconjugated humanized anti-CD33 monoclonal antibodies has met with little success in relapsed disease, the antigen–antibody complex is rapidly internalized, suggesting that this would be a convenient drug delivery system to leukemia cells. GO is a humanized anti-CD33 monoclonal antibody conjugated to the extremely potent (toxic) antitumor drug calicheamicin. In the final report of a phase II trial in the USA and Europe, 277 patients were treated with standard doses of GO (9 mg/m^2, 2 h d.i.v. on days 1 and 15).[23] The response rate of younger patients was 27% (CR, 13%; CRp, 14%). Other clinical trials reported similar results with an approximate response rate of 26% (CR, 13%; CRp, 13%),[8,11,24] and the phase II part of the clinical trials in Japan resulted in a response rate of 30% (CR, 25%; CRp, 5%).[13]

As clinical efficacy of GO monotherapy for patients with relapsed or refractory AML has been limited, clinical studies are required for exploration of the role of GO in combination therapy with conventional chemotherapy. Even though several groups in the USA and Europe have been evaluating the potential of GO already in different situations in the treatment of AML, the optimal usage of GO in combination therapy is still unknown, especially for Japanese patients. For this reason, we conducted the present study, starting from phase I, in order to evaluate the safety of GO-combined therapy.

As the final goal of our study is to investigate whether GO-combined therapy is meaningful for de novo adult AML (younger than age 65 years), we selected standard induction therapies, which are IDR 12 mg/m^2 on days 1–3 plus Ara-C 100 mg/m^2 on days 1–7, and DNR 50 mg/m^2 on days 1–5 plus Ara-C 100 mg/m^2 on days 1–7, as partner chemotherapeutic regimens.[4]

In the present study for relapsed or refractory AML, GO was administered on the next day after the final administration of anthracycline (IDR or DNR) with continuing administration of Ara-C.

As expected, grade 3/4 hematological toxicities and febrile neutropenia was observed in most patients, but those toxicities were clinically manageable. None of the patients died of adverse events, although one patient died of disease progression. The DLT (prolongation of neutropenia and thrombocytopenia, and serious infection [i.e. cerebral abscess]) were observed in all patients in level 3 of the IAG regimen (a dose of 5 mg/m^2 GO), but none in level 2 of the IAG regimen or level 3 of the DAG regimen. Therefore, the MTD of the IAG regimen was determined as level 2 (i.e. 3 mg/m^2 GO, 12 mg/m^2 IDR and 100 mg/m^2 Ara-C), and that of the DAG regimen as level 3 (i.e. 3 mg/m^2 GO, 50 mg/m^2 DNR and 100 mg/m^2 Ara-C).

Several attempts that combined the approved dosage of GO (9 mg/m^2, administered twice) with chemotherapy resulted in excess toxicity such as infection and liver toxicity, including increased risk of VOD/SOS.[25] The Cancer and Leukemia Group B (CALGB) 19902 study indicated that the dose schedule of 9 mg/m^2 GO on day 7 and 4.5 mg/m^2 GO on day 14 with high-dose Ara-C (3 g/m^2 per day for 5 days) caused a high rate of treatment-related death (four of the first seven patients, 57%).[18] In the present study, severe hepatotoxicity or VOD/

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Table 8. Selected phase II trials of gemtuzumab ozogamicin (GO)-combining therapy for relapsed or refractory adult acute myeloid leukemia (AML)

Authors (name of regimen)	Institutes	No. patients	Median age (range) (years)	Combination of drugs	Dose and schedule of GO	% Response (CR/CRp)	Median OS (months)	Grade 3/4 non-hematological toxicity
Tsimberidou *et al.*[14] 2003 (MFAC)	MDACC	32	53 (18–78)	FLD: 15 mg/m^2 i.v. q12 h/day, days 2–4 Aa-C: 500 mg/m^2 2 h d.i.v. q12 h/day, days 2–4 CSA: 6 mg/kg 2 h d.i.v. + 16 mg/kg c.i.v., days 1, 2	4.5 mg/m^2 2 h d.i.v., day 1	34 (28/6)	5.3	Hyperbilirubinemia (18%), hepatic transaminitis (9%), VOD (3%)
Alvarado *et al.*[15] 2003 (MIA)	MDACC	14	61 (34–74)	IDR: 12 mg/m^2/day i.v., days 2–4 Ara-C: 1.5 g/m^2/day, days 2–5	6 mg/m^2 2 h d.i.v., days 1, 15	42 (21/21)	2	Sepsis (71%), liver damage, VOD (14%)
Chevallier *et al.*[16] 2008 (MIDAM)	France	62	56 (16–71)	Ara-C: 1.5 g/m^2 2 h d.i.v. q12 h/day, days 1–5 MIT: 12 mg/m^2/day i.v., days 1–3	9 mg/m^2 2 h d.i.v., day 4	63 (50/13)	9.5	Hyperbilirubinemia (16%), VOD (3%), early toxic death (6%)
Fianchi *et al.*[17] 2008 (G-Ara-My)	Italy	53	M	G-CSF: 5 μg/kg/day s.c., days 1–8 Ara-C: 100 mg/m^2/day c.i.v., days 2–8 or 4–8	6 mg/m^2 2 h d.i.v., day 9	45 (43/2)	9	Infection (36%), infusion reaction (5.5%), VOD (2%)
Stone *et al.*[18] 2010 (CALGB 19902)	CALGB	37	64 (55–70)	Ara-C: 3 g/m^2 3 h d.i.v./day, days 1–5	9 mg/m^2 2 h d.i.v., day 7	35 (32/3)	8.9	Hepatic transaminitis (29%), hyperbilirubinemia (27%), infection (92%), death of tox (8.1%)

Ara-C, cytarabine; CALGB, Cancer and Leukemia Group B; c.i.v., continuous venous infusion; CR, complete remission; CRp, CR without platelet recovery; CSA, cyclosporin A; d.i.v., drip venous infusion; FLD, fludarabine; G-CSF, granulocyte colony stimulating factor; IDR, idarubicin; iv, venous infusion; MDACC, MD Anderson Cancer Center; MIT, mitoxantrone; q12 h, every 12 h; OS, overall survival; VOD, veno-occlusive disease.

SOS was not observed in either of the IAG or DAG regimens, because we selected an initial dose of GO at 3 mg/m^2.

The MRC group already indicated in the AML15 prelude trial that a combination of 3 mg/m^2 but not 6 mg/m^2 of GO with intensive chemotherapy was safe and feasible for a multicenter trial in induction and consolidation therapy.[19] Our study confirmed a safe dose of GO as 3 mg/m^2, even though the timing of administration was different.

Although the present study was not designed to assess efficacy, it was of note that CR and CRp were achieved in nine (47.4%) and one (5.2%), respectively, out of 19 patients with relapsed or refractory AML. This overall rate of response, 52.6%, was comparable to the results of previous phase II trials for relapsed or refractory AML[14–18] (Table 8).

Clinical efficacy of the combination of GO with IDR + Ara-C (named MIA) was already evaluated by the MD Anderson Cancer Center.[15] Compared with our IAG regimen, the response rate of MIA (42%; CR, 21%; CRp, 21%) was quite similar, but their incidence of severe non-hematological toxicity was higher. Despite the fact that the doses of Ara-C and GO were lower in our IAG regimen, this combination will be feasible as an induction therapy for relapsed or refractory AML.

The MRC AML15 prelude trial[19] investigated safety and efficacy of GO in combination with DNR + Ara-C, in which DNR (50 mg/m^2 for 3 days) and Ara-C were combined with 3 mg/m^2 GO on day 1. Hematolopoietic recovery was satisfactory, and although two of eight enrolled patients developed grade 3 toxicity, all patients achieved CR and tolerated subsequent chemotherapy. In levels 2 and 3 of our DAG regimen, although the dose of DNR was higher than that of the MRC trial, the recovery from myelosuppression was satisfactory without excess of unexpected non-hematological toxicity.

During this phase I trial of GO in combination with chemotherapy for relapsed or refractory AML, several multicenter trials to investigate the role of GO combination for *de novo* AML have been completed in the USA and Europe. Burnett *et al.*[26] presented the results of the MRC AML15 trial, in which 1113 mostly younger, newly diagnosed patients with AML (except acute promyelocytic leukemia) were randomly assigned to one of three conventional induction therapies with or without 3 mg/m^2 GO on day 1. After achieving CR, 978 patients were randomly assigned to GO in combination with chemotherapy in course 3 of the consolidation therapy. The addition of GO was well tolerated with no significant increase in toxicity. Although there was no overall difference in response or survival, a predefined analysis by cytogenetic risk groups showed a significant survival benefit for patients with favorable risk and a trend for those with intermediate risk disease.

A similar study conducted by the Southwest Oncology Group (SWOG) was reported in abstract format.[27] In this SWOG 106 study, 627 patients with untreated AML (age 18–60 years) were randomly assigned to receive induction therapy either with Ara-C (100 mg/m^2 × 7 days) + DNR (60 mg/m^2 × 3 days) or with Ara-C (100 mg/m^2 × 7 days) + DNR (45 mg/m^2 × 3 days) + GO (6 mg/m^2). An interim analysis showed a CR rate of 66% in the GO-combined arm and 69% in the chemotherapy-alone arm (control arm), ruling out the originally hypothesized increase in CR of 12% by the addition of GO. There was no difference in disease-free survival (DFS) either, and the rate of fatal adverse events was higher in the GO-combined arm compared with the control arm (5.8% *vs* 0.8%). Based on these negative findings of the GO-combined arm, the FDA recommended to withdraw GO from the market in the USA.

However, as Burnett *et al.*[26] suggested, the SWOG 106 study is confounded, as the dose of DNR was lower in patients given GO, which might have masked any benefit of GO. In addition, the induction death rate in the GO arm was similar to what had been reported in other AML induction trials, but the mortality rate of the control arm was unexpectedly low. Nevertheless, in the SWOG study the benefit in the favorable subtype of AML was similarly observed in the MRC study.

Another smaller phase II study reported a high molecular response rate and DFS by GO in combination with high-dose Ara-C for core binding factor (CBF) leukemias.[28]

In conclusion, the present study demonstrated that 3 mg/m^2 of GO with IDR + Ara-C or DNR + Ara-C can be administered safely in younger adult patients with relapsed or refractory AML. As three clinical studies of GO-combined chemotherapy for newly diagnosed adult AML have indicated, there are subsets of AML, such as CBF leukemias, that could benefit from the addition of GO to conventional therapy. Intensive induction chemotherapy followed by a modest dose of GO like in our study protocol will be safely provided for salvage therapy regardless of cytogenetic risk groups. Fortunately, GO is still commercially available in Japan, therefore there is a need for confirmatory studies that investigate the efficacy of GO-combined chemotherapy for patients with AML as both initial and salvage therapy.

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Disclosure Statement

The authors have no conflict of interest.

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A randomized comparison of 4 courses of standard-dose multiagent chemotherapy versus 3 courses of high-dose cytarabine alone in postremission therapy for acute myeloid leukemia in adults: the JALSG AML201 Study

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A randomized comparison of 4 courses of standard-dose multiagent chemotherapy versus 3 courses of high-dose cytarabine alone in postremission therapy for acute myeloid leukemia in adults: the JALSG AML201 Study

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We conducted a prospective randomized study to assess the optimal postremission therapy for adult acute myeloid leukemia in patients younger than 65 years in the first complete remission. A total of 781 patients in complete remission were randomly assigned to receive consolidation chemotherapy of either 3 courses of high-dose cytarabine (HiDAC, 2 g/m² twice daily for 5 days) alone or 4 courses of conventional standard-dose multiagent chemotherapy (CT) established in the previous JALSG AML97 study. Five-year disease-free survival was 43% for the HiDAC group and 39% for the multiagent CT group ($P = .724$), and 5-year overall survival was 58% and 56%, respectively ($P = .954$). Among the favorable cytogenetic risk group (n = 218), 5-year disease-free survival was 57% for HiDAC and 39% for multiagent CT ($P = .050$), and 5-year overall survival was 75% and 66%, respectively ($P = .174$). In the HiDAC group, the nadir of leukocyte counts was lower, and the duration of leukocyte less than 1.0×10^9/L longer, and the frequency of documented infections higher. The present study demonstrated that the multiagent CT regimen is as effective as our HiDAC regimen for consolidation. Our HiDAC regimen resulted in a beneficial effect on disease-free survival only in the favorable cytogenetic leukemia group. This trial was registered at www.umin.ac.jp/ctr/ as #C000000157. (*Blood.* 2011;117(8): 2366-2372)

Introduction

Approximately 70% to 80% of the newly diagnosed younger adult patients with acute myeloid leukemia (AML) achieve complete remission (CR) when treated with an anthracycline, usually daunorubicin (DNR) or idarubicin (IDR), and cytarabine (Ara-C); however, only approximately one-third of these patients remain free of disease for more than 5 years.[1-5] If CR patients are left untreated, almost all of them will relapse and die.[6] Therefore, postremission therapy is indispensable. Postremission therapy is divided into consolidation and maintenance therapy. In the previous studies of Japan Adult Leukemia Study Group (JALSG) for adult AML (AML87, 89, 92, and 95),[1-3,5] we administered 3 courses of consolidation therapy and 6 courses of intensified maintenance therapy. In the AML97 study,[7] we conducted a randomized study to compare the conventional 3-course consolidation and 6-course maintenance therapies with 4 courses of intensive consolidation therapy without maintenance and demonstrated no difference in overall survival (OS) and disease-free survival (DFS). Therefore, the 4 courses of conventional standard-dose multiagent chemotherapy (CT) became the standard regimen in Japan. On the other hand, multiple cycles of high-dose cytarabine (HiDAC) have been commonly used as consolidation therapy in the United States and other countries. However, our national medical insurance system did not allow us to use HiDAC until 2001, and thus we could not use HiDAC in the previous treatment regimens for leukemia. We therefore conducted this prospective, multicenter cooperative

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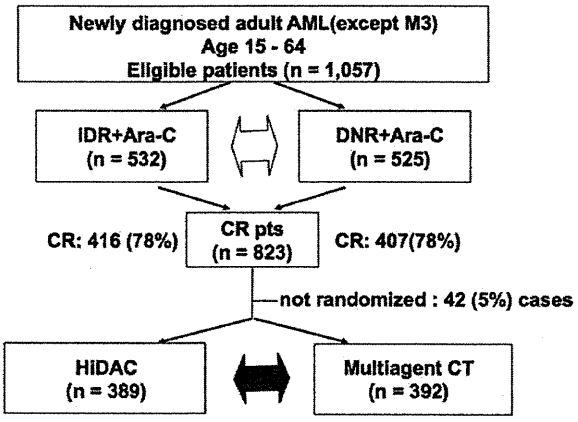

Figure 1. CONSORT diagram.

study to compare 4 courses of multiagent CT with 3 courses of HiDAC therapy after its approval in April 2001.

Methods

Patients

From December 2001 to December 2005, 1064 newly diagnosed adult patients 15 to 64 years of age with de novo AML were consecutively registered from 129 participating institutions. AML was first diagnosed by the French-American-British classification at each institution. Peripheral blood and bone marrow smears of registered patients were reevaluated by the central review committee. French-American-British M3 was not registered. Eligibility criteria included adequate function of liver (serum bilirubin < 2.0 mg/dL), kidney (serum creatinine < 2.0 mg/dL), heart and lung, and an Eastern Cooperative Oncology Group performance status between 0 and 3. Patients were not eligible if they had prediagnosed myelodysplastic syndrome or prior chemotherapy for other disorders. Cytogenetic abnormalities were grouped by standard criteria and classified according to the Medical Research Council classification.[8] The study was approved by institutional review boards at each participating institution. Written informed consent was obtained from all patients before registration in accordance with the Declaration of Helsinki.

Induction therapy consisted of Ara-C 100 mg/m^2 for 7 days and either IDR (12 mg/m^2 for 3 days) or DNR (50 mg/m^2 for 5 days). If patients did not achieve remission after the first course, the same therapy was administered once more. The outcome of induction therapy was reported to the JALSG Statistical Center before the consolidation therapy started. All CR patients were stratified according to induction regimen, number of courses of induction, age and karyotype, and randomized to receive either 4 courses of multiagent CT or 3 courses of HiDAC therapy. The first course of multiagent CT consisted of mitoxantrone (7 mg/m^2 by 30-minute infusion for 3 days) and Ara-C (200 mg/m^2 by 24-hour continuous infusion for 5 days). The second consisted of DNR (50 mg/m^2 by 30-minute infusion for 3 days) and Ara-C (200 mg/m^2 by 24-hour continuous infusion for 5 days). The third consisted of aclarubicin (20 mg/m^2 by 30-minute infusion for 5 days) and Ara-C (200 mg/m^2 by 24-hour continuous infusion for 5 days). The fourth consisted of Ara-C (200 mg/m^2 by 24-hour continuous infusion for 5 days), etoposide (100 mg/m^2 by 1-hour infusion for 5 days), vincristine (0.8 mg/m^2 by bolus injection on day 8), and vindesine (2 mg/m^2 by bolus injection on day 10). Each consolidation was started as soon as possible after neutrophils, white blood cells (WBCs), and platelets recovered to more than 1.5 × 10^9/L, 3.0 × 10^9/L, and 100.0 × 10^9/L, respectively. In the HiDAC group, 3 courses of Ara-C 2.0 g/m^2 by 3-hour infusion every 12 hours for 5 days were given. Each course was started 1 week after neutrophils, WBCs, and platelets recovered to the aforementioned counts.

Table 1. Clinical characteristics of randomized patients

Characteristic	HiDAC (n = 389)	Multiagent CT (n = 392)	P
Age, y, median (range)	46 (15-64)	47 (15-64)	.697
WBC, ×10^9/L, median (range)	15.6 (0.1-382)	14.9 (0.2-260)	.323
Karyotype, n			.210
Favorable	108	110	
Intermediate	242	256	
Adverse	27	14	
Unknown	12	12	
Induction, n			.914
IDR	196	196	
DNR	193	196	
Induction 1 cycle, %	81.0	81.4	.886

Bone marrow examination was performed to confirm CR in both groups before each consolidation therapy and at the end of all consolidation therapy.

Best supportive care, including administration of antibiotics and platelet transfusions, was given if indicated. When patients had life-threatening documented infections during neutropenia, the use of granulocyte colony-stimulating factor was permitted.

After the completion of consolidation therapy, patients received no further chemotherapy. Allogeneic stem cell transplantation (allo-SCT) was offered during the first CR to patients of age 50 years or less with a histocompatible donor in the intermediate or adverse cytogenetic risk groups. Stem cell source was related donor or unrelated donor. Cord blood was not used. Conditioning before transplantation and prophylaxis for graft-versus-host disease were performed according to each institutional standard.

Responses were evaluated by the recommendations of the International Working Group.[9] CR was defined as the presence of all of the following: less than 5% of blasts in bone marrow, no leukemic blasts in peripheral blood, recovery of peripheral neutrophil counts more than 1.0 × 10^9/L and platelet counts more than 100.0 × 10^9/L, and no evidence of extramedullary leukemia. Relapse was defined as the presence of at least one of the

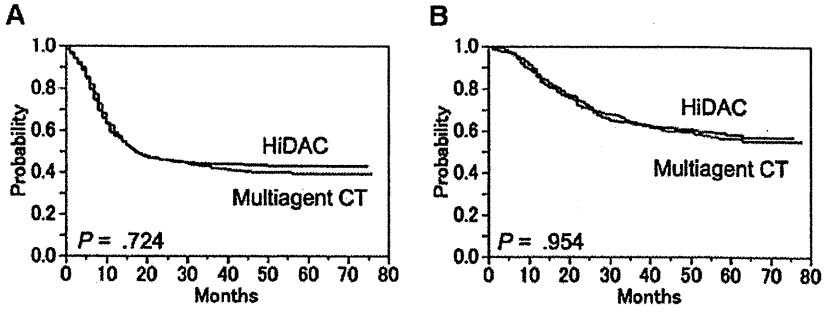

Figure 2. DFS and OS according to treatment arm. (A) DFS of CR patients. Predicted 5-year DFS was 43% for the HiDAC group (n = 389; red line) and 39% for the multiagent CT group (n = 392; blue line; P = .724). (B) OS of CR patients. Predicted 5-year OS was 58% for the HiDAC group (n = 389; red line) and 56% for the multiagent CT group (n = 392; blue line; P = .954).

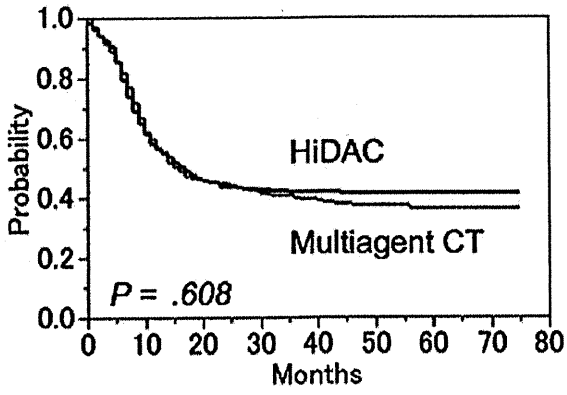

Figure 3. DFS according to treatment arm, after censoring the observation in transplanted patients. Predicted 5-year DFS was 41% for the HiDAC group (n = 389; red line) and 36% for the multiagent CT group (n = 392; blue line; $P = .608$).

following: reappearance of leukemic blasts in peripheral blood, recurrence of more than 5% blasts in bone marrow, and appearance of extramedullary leukemia.

Statistical analysis

This was a multi-institutional randomized phase 3 study with a 2 × 2 factorial design. The primary endpoint of the first randomization was CR rate, and a sample size of 420 patients per group was estimated to have a power of 90% at a 1% level of significance to demonstrate noninferiority (assuming 80% CR rate for both groups). For the second randomization (ie, this study), the primary endpoint was DFS, and the secondary end points were OS and adverse events of grade 3 or more by National Cancer Institute Common Toxicity Criteria. A sample size of 280 patients per group was estimated to have a power of 80% at a 5% level of significance to demonstrate 10% superiority in 5-year DFS for the HiDAC arm (40% vs 30%). OS was defined as the time interval from the date of diagnosis to the date of death. DFS for patients who had achieved CR was defined as the time interval from the date of CR to the date of the first event (either relapse or death). Patients who underwent allo-SCT were not censored. The Kaplan-Meier method was used to estimate probabilities of DFS and OS. For comparison of DFS and OS, the log-rank test was used for univariate analysis and the proportional hazard model of Cox for multivariate analysis. Cumulative incidence of relapse and treatment-related mortality were estimated according to the competing risk method and were evaluated with Gray test. The Wilcoxon rank-sum test was used for continuous data, such as age and WBC count, whereas the χ^2 test was used for ordinal data, such as risk group and frequency of allo-SCT. Statistical analyses were conducted using the JMP program (SAS Institute) and R software Version 2.9.1 (www.r-project.org).

Results

Response to induction therapy

Of 1064 patients registered, 1057 patients were evaluable. Seven patients (1 misdiagnosis, 1 infectious complication, 1 without therapy, and 4 withdrawal of consent) were excluded. Median age was 47 years (range, 15-64 years). Cytogenetic studies were performed in 99.2% of registered patients and the results were available in 97%. Of 1057 evaluable patients, 823 (78%) achieved CR (662 of them after the first induction course). CR rate in the IDR and DNR arms was similar (78.2% vs 77.5%). Percentage of patients who reached CR after the first induction course was also similar (64.1% vs 61.1%, $P = .321$). Day to achieve CR was longer in the IDR arm than the DNR arm (33.8 vs 32.4 days, $P = .038$). The detailed result of induction phase of this study is reported in a separate paper.[10]

Postremission randomization

Of 823 patients who achieved CR, 42 did not undergo the second randomization for a variety of reasons, which included residual toxicity from induction therapy (12), allo-SCT (8), death (1), refusal (1), and unknown (20). The remaining 781 patients were randomly assigned to receive either the HiDAC regimen (389) or the multiagent CT regimen (392; Figure 1). Clinical characteristics of 2 treatment groups were well balanced in age, initial WBC count, cytogenetic risk, induction arm, and induction cycle (Table 1).

DFS and OS

The median follow-up period of living patients was 48 months (range, 5-78 months). Five-year DFS was 43% for the HiDAC group and 39% for the multiagent CT group ($P = .724$; Figure 2A). Five-year OS was 58% for the HiDAC group and 56% for the multiagent CT group ($P = .954$; Figure 2B). After censoring the observation on the date of SCT in transplanted patients, 5-year DFS was 41% for the HiDAC group and 36% for the multiagent CT group ($P = .608$; Figure 3).

The cumulative incidences of relapse and treatment-related mortality during CR, respectively, were 49% and 8% for the HiDAC group and 56% and 5% for the multiagent CT group ($P = .294$, $P = .172$; Figure 4A). After censoring the observation in transplanted patients, those were 55% and 4% for the HiDAC group and 61% and 3% for the multiagent CT group ($P = .402$, $P = .409$), respectively (Figure 4B).

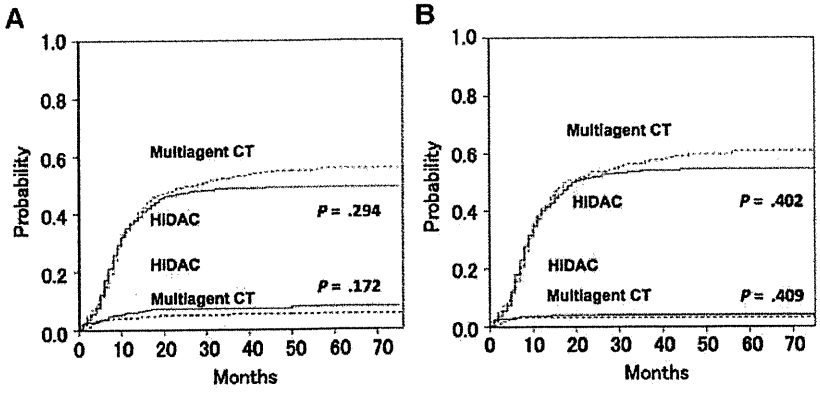

Figure 4. Cumulative incidence of relapse and treatment-related mortality in CR by treatment arm. (A) The incidences of relapse and mortality, respectively, were 49% and 8% for the HiDAC group (solid line) and 56% and 5% for the multiagent CT group (dotted line; $P = .324$, $P = .172$). (B) After censoring the observation in transplanted patients, the incidences of relapse and mortality, respectively, were 55% and 4% for the HiDAC group (solid line) and 61% and 3% for the multiagent CT group (dotted line; $P = .402$, $P = .409$).

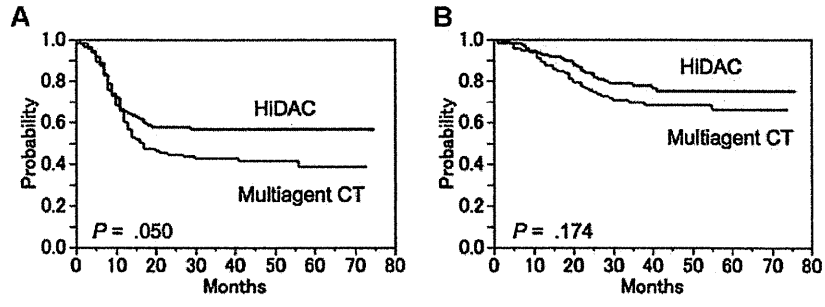

Figure 5. DFS and OS by treatment arm for the favorable cytogenetic risk group. (A) Predicted 5-year DFS was 57% for the HiDAC group (n = 108; red line) and 39% for the multiagent CT group (n = 110; blue line; $P = .050$). (B) Predicted 5-year OS was 75% for the HiDAC group (n = 108; red line) and 66% for the multiagent CT group (n = 110; blue line; $P = .174$).

In patients with the favorable cytogenetics, core-binding factor (CBF) leukemia with t(8;21) or inv(16), 5-year DFS was 57% in the HiDAC group and 39% in the multiagent CT group ($P = .050$; Figure 5A), and 5-year OS was 75% and 66%, respectively ($P = .174$; Figure 5B).

In patients with the intermediate cytogenetics, 5-year DFS was 38% in the HiDAC group and 39% in the multiagent CT group ($P = .403$; Figure 6A), and 5-year OS was 53% and 54%, respectively ($P = .482$; Figure 6B). In patients with the adverse cytogenetics, 5-year DFS was 33% in the HiDAC group and 14% in the multiagent CT group ($P = .364$; Figure 7A), and 5-year OS was 39% and 21%, respectively ($P = .379$; Figure 7B). Among younger patients ($\leq$ 50 years), 5-year DFS was 45% in the HiDAC group and 46% in the multiagent CT group ($P = .590$), and 5-year OS was 62% and 66%, respectively ($P = .228$). Among the older patients (> 50 years), 5-year DFS was 40% in the HiDAC group and 28% in the multiagent CT group ($P = .230$), and 5-year OS was 51% and 40%, respectively ($P = .159$). In patients treated with the IDR regimen at induction, 5-year DFS was 42% in the HiDAC group and 41% in the multiagent CT group ($P = .641$), and 5-year OS was 58% and 57%, respectively ($P = .790$). In patients treated with the DNR regimen at induction, 5-year DFS was 44% in the HiDAC group and 37% in the multiagent CT group ($P = .339$), and 5-year OS was 58% and 56%, respectively ($P = .713$). There was no relationship between the duration of myelosuppression and DFS or OS.

Significant unfavorable prognostic features for DFS by the Cox proportional hazard model were WBC more than 20×10^9/L, the number of induction therapies, and age more than 50 years, and for OS, age more than 50 years, the number of induction therapies, WBC more than 20×10^9/L, and myeloperoxidase-positive blast less than 50%. Induction therapy, consolidation therapy, and cytogenetic risk group were not independent prognostic factors for DFS or OS by this multivariate analysis (Table 2).

Tolerance and toxicity of postremission therapy

All courses of consolidation were administered to 72.5% of patients in the HiDAC group and 70.2% in the multiagent CT group (Table 3). In the HiDAC group, 110 patients (28%) did not receive all 3 courses. The reasons included relapse (18), death in CR (10), allo-SCT (34), adverse events (27), patient's refusal (11), and unknown (10). In the multiagent CT group, 118 patients (30%) did not receive all 4 courses. The reasons included relapse (31), death in CR (8), allo-SCT (42), adverse events (13), patient's refusal (5), and unknown (19). The most common reason was allo-SCT in both groups. Of 125 patients received SCT in first CR, 49 (25 in HiDAC and 24 in multiagent CT) received SCT after completion of full courses of consolidation therapy. The second common reason was adverse events in the HiDAC group and relapse in the multiagent CT group. The patients older than 50 years could tolerate both regimens. Table 4 shows a comparison of both groups regarding the nadir of WBC count and the number of days of WBC less than 1.0×10^9/L. After each course of consolidation, the nadir of WBC count was significantly lower ($P < .0001$) and the day of WBC less than 1.0×10^9/L was significantly longer in the HiDAC group ($P < .001$). During each course of consolidation, the frequency and the number of days of granulocyte colony-stimulating factor administration were significantly higher in the HiDAC group. Table 5 shows toxic adverse events, excluding hematologic side effects. The frequency of documented infections was significantly higher in the HiDAC group ($P < .001$). The subset analysis showed the high incidence of documented infection in HiDAC regimen only in intermediate cytogenetic risk group ($P < .001$).

Discussion

To determine the best postremission therapy, there have been several prospective randomized studies comparing chemotherapy

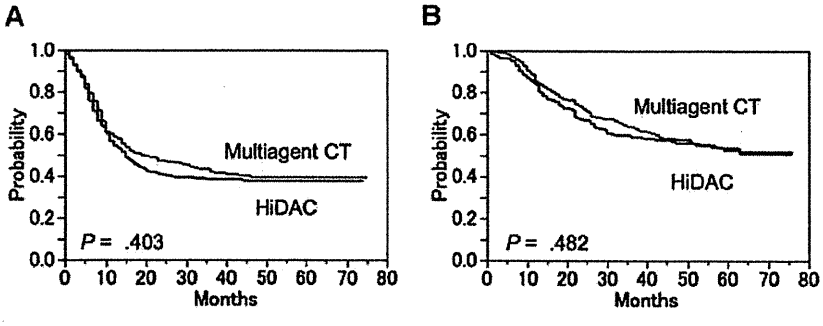

Figure 6. DFS and OS by treatment arm for the intermediate cytogenetic risk group. (A) Predicted 5-year DFS was 38% for the HiDAC group (n = 242; red line) and 39% for the multiagent CT group (n = 256; blue line; $P = .403$). (B) Predicted 5-year OS was 53% for the HiDAC group (n = 242; red line) and 54% for the multiagent CT group (n = 256; blue line; $P = .482$).

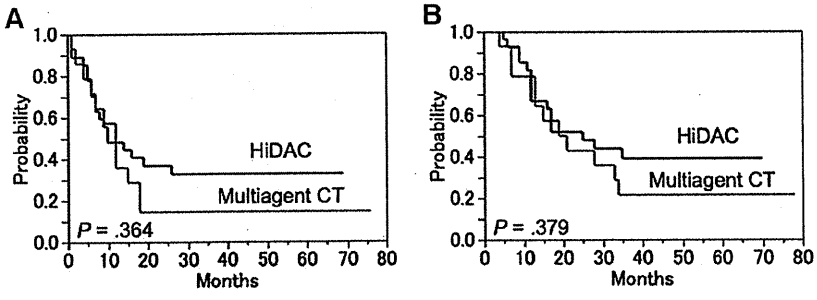

Figure 7. DFS and OS by treatment arm for the adverse cytogenetic risk group. (A) Predicted 5-year DFS was 33% for the HiDAC group (n = 27; red line) and 14% for the multiagent CT group (n = 14; blue line; $P = .364$). (B) Predicted 5-year OS was 39% for the HiDAC group (n = 27; red line) and 21% for the multiagent CT group (n = 14; blue line; $P = .379$).

with SCT. Although there is some limitation in SCT, such as patient age and availability of human leukocyte antigen–identical donors, most randomized studies demonstrate that SCT, the most intensive postremission modality, provides superior or at least noninferior prognosis in high- or intermediate-risk adult AML.[11-13]

As for postremission chemotherapy, HiDAC therapy is generally used in the United States and other countries after the landmark Cancer and Leukemia Group B-8525 (CALGB-8525) study.[14] In Japan, however, because HiDAC therapy was not approved by our national medical insurance system until 2001, combination chemotherapy using non–cross-resistant agents was commonly used in previous studies for adult AML. Therefore, in the current study, we compared conventional multiagent CT with HiDAC therapy.

Our study demonstrated that there is no difference in DFS and OS between the multiagent CT regimen and the HiDAC regimen. The HiDAC regimen, however, was accompanied with more frequent infectious events resulting from more severe and longer-lasting neutropenia. In the CALGB-8525 study,[14] patients randomized to 4 cycles of HiDAC regimen were administered 3 g/m^2 of Ara-C by 3-hour infusion, twice daily on days 1, 3, and 5, and our patients randomized to 3 cycles of HiDAC regimen were given 2 g/m^2 of Ara-C by 3-hour infusion, twice daily for 5 days. Although there were some differences in schedule and dose administered, the total dose of Ara-C was almost the same (72 g/m^2 vs 60 g/m^2). The Acute Leukemia French Association Group compared a timed-sequential consolidation consisting of etoposide, mitoxantrone, and Ara-C with a postremission chemotherapy, including 4 cycles of HiDAC (3 g/m^2), and reported that there were no statistically significant differences between the 2 groups in the rates of event-free survival and OS at 3 years.[15] The British Medical Research Council also compared a conventional Medical Research Council schedule (MACE/MidAC) with 2 courses of HiDAC regimens (3 g/m^2 or 1.5 g/m^2) and reported that there were no significant differences in DFS and OS at 5 years.[16]

On the contrary, the CALGB-8525 study[14] revealed that their HiDAC regimen was superior to the intermediate dose of Ara-C (400 mg/m^2 for 5 days) or to the conventional dose of Ara-C (100 mg/m^2 for 5 days) regimens in DFS and OS; this plausibly comes from the lower dose intensity of the intermediate- or standard-dose Ara-C regimens. Indeed, the CALGB-9222 study[17] showed no difference in DFS and OS between the HiDAC group and the intensified sequential multiagent chemotherapy group.

Cytogenetics is considered one of the most valuable prognostic determinants in adult AML.[8,18] In the present study, although in the intermediate-risk group, the DFS and OS of both consolidation groups were almost identical; in the favorable risk group, the outcome of the HiDAC group (n = 108) tended to be superior to that of the multiagent CT group (n = 110) in DFS (57% vs 39%; $P = .050$) and OS (75% vs 66%; $P = .174$) but not at statistically significant level; and in the adverse risk group, the similar but statistically nonsignificant trend in DFS (33% vs 14%) and OS (39% vs 21%) was noted. Bloomfield et al[19] reported that the HiDAC regimen is the most effective to CBF leukemia. In their study, patients with CBF leukemia (n = 18) had a 78% chance of remaining CR at 5 years when treated with the HiDAC regimen. However, our study showed that DFS of CBF leukemia (n = 108) treated with the HiDAC regimen was only 57% at 5 years.

There are 2 possible explanations of difference between our results and those reported by Bloomfield et al.[19] One is that their superior results may come from a small number of patients (n = 18). Indeed, the CALGB-9222 study,[17] including 28 patients with CBF leukemia, demonstrated that the 5-year DFS and OS of CBF leukemia treated with HiDAC was 60% and 70%, respectively. These data are similar to our results. The other is that CBF leukemia reveals different sensitivity to HiDAC therapy. Some patients with CBF abnormality have KIT mutations, which confer

Table 2. Factors to predict unfavorable prognostic features for DFS and OS by multivariate analysis

Survival type/variable	Category	Hazard ratio	P
DFS			
Initial WBC count	≥ 20 × 10^9/L	1.49	< .0001
No. of induction therapies	2 courses	1.50	.0006
Age, y	> 50	1.33	.0028
Consolidation therapy	Multiagent CT	1.04	.7128
OS			
Age, y	> 50	2.00	< .0001
No. of induction therapies	2 courses	1.58	.0033
Initial WBC count	≥ 20 × 10^9/L	1.41	.0070
MPO-positive blast	< 50 %	1.42	.0149
Consolidation therapy	Multiagent CT	0.96	.7768

MPO indicates myeloperoxidase.

Table 3. Tolerance of consolidation

	% receiving the full courses	
	HiDAC	Multiagent CT
All patients	72.5	70.2
Patients ≤ 50 y	71.9	69.0
Patients > 50 y	73.4	71.9
Reason for not receiving the full courses (no. of patients)		
Relapse	18	31
Death	10	8
SCT in first CR	31	42
Adverse event*	27	13
Patient refusal	11	5
Unknown	10	19

*$P < .05$.

Table 4. Intensity of consolidation

	HiDAC	Multiagent CT	*P*
After first consolidation			
Lowest WBC, $\times 10^9$/L	0.17	0.40	< .0001
Days WBC < 1.0 × 10^9/L	13 (0-40)	12 (0-36)	.0005
After second consolidation			
Lowest WBC, $\times 10^9$/L	0.10	0.40	< .0001
Days WBC < 1.0 × 10^9/L	14 (0-34)	13 (0-241)	.0007
After third consolidation			
Lowest WBC, $\times 10^9$/L	0.10	0.40	< .0001
Days WBC < 1.0 × 10^9/L	14 (0-38)	11.5 (0-28)	< .0001
After fourth consolidation			
Lowest WBC, $\times 10^9$/L		0.40	
Days WBC < 1.0 × 10^9/L		12 (0-34)	

Values are median (range).

higher relapse risk on CBF AML.[20,21] CALGB reported that 29.5% of patients with inv(16) and 22% of patients with t(8;21) had KIT mutations, and the cumulative incidence of relapse was higher for patients with mutated KIT than for those with wild-type KIT.[20] The difference of mutation rates of KIT might result in the difference in DFS. Unfortunately, in our present study, KIT mutations were not prospectively evaluated. However, a high mutation rate of KIT is reported among Asian patients with t(8;21) from Japan (37.8%)[22] and China (48.1%).[23] Consequently, JALSG is prospectively evaluating KIT mutation and its impact on the outcome in patients with CBF leukemia treated with repetitive HiDAC therapy. In the adverse cytogenetic risk group, the outcome of the HiDAC group also tends to be better than that of the multiagent CT group, but the difference is not statistically significant. The small number of this cohort may explain the statistical insignificance. Nevertheless, HiDAC therapy may be recommended to this group if patients have no human leukocyte antigen–matched donor.

Recently, IDR is frequently included into induction regimen for AML because of its better effectiveness compared with DNR.[24-26] A meta-analysis of randomized trials showed that the use of IDR instead of DNR results in a high CR rate.[27] However, a German group reported that the advantage of IDR in response rate may be lost during HiDAC consolidation therapy because of increased toxicity in the IDR group.[28] However, our current study demonstrated that, among the HiDAC group, there is no difference in DFS and OS between patients receiving IDR or DNR in induction phase. In our study, although one or 2 courses of the IDR regimen were given before the HiDAC consolidation, only 19% of patients required 2 courses to obtain CR. In contrast, the German group gave 2 courses of IDR induction regimen before the HiDAC consolidation. Thus, severe adverse events during HiDAC therapy probably depend on the total dose of prior IDR. Nevertheless, the HiDAC regimen could be given safely in our patients who had received IDR as induction therapy.

Table 5. Adverse events (CTC grades 3 and 4) during consolidation therapy

	HiDAC, %	Multiagent CT, %	*P*
Documented infection	20.9	14.5	< .001
Febrile neutropenia	66.5	66.4	.311
Bleeding	0.8	0.7	.601
Early death*	0.9	0.6	.389

*Death within 30 days after consolidation chemotherapy.

In conclusion, postremission consolidation regimen should be selected on the basis of prognostic factors, such as cytogenetics. Although several types of HiDAC regimen have been widely adopted as the optimal postremission therapy, the conventional multiagent CT may be recommendable for the intermediate or adverse cytogenetic risk groups. However, our HiDAC regimen should be recommended to the favorable cytogenetic risk group.

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Authorship

Contribution: S.M. designed and performed research, interpreted data, and wrote the manuscript; S.O. designed and performed research, collected and analyzed data, and participated in writing the manuscript; S.F., H.K., K.S., N.U., T.S., K.M., C.N., Y.M., M. Taniwaki, T. Nagai, T.Y., A.F., M. Takahashi, F.Y., Y.K., N.A., H.S., H.H., S.H., K.O., and T. Naoe performed research; and R.O. interpreted data and participated in writing manuscript.

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Randomized study of induction therapy comparing standard-dose idarubicin with high-dose daunorubicin in adult patients with previously untreated acute myeloid leukemia: the JALSG AML201 Study

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We conducted a multi-institutional randomized study to determine whether high-dose daunorubicin would be as effective as standard-dose idarubicin in remission-induction therapy for newly diagnosed adult patients younger than 65 years of age with acute myeloid leukemia. Of 1064 patients registered, 1057 were evaluable. They were randomly assigned to receive either daunorubicin (50 mg/m^2 daily for 5 days) or idarubicin (12 mg/m^2 daily for 3 days) in combination with 100 mg/m^2 of cytarabine by continuous infusion daily for 7 days as induction therapy. Complete remission was achieved in 407 (77.5%) of 525 patients in the daunorubicin group and 416 (78.2%) of 532 in the idarubicin group ($P = .79$). Patients achieving complete remission received intensive postremission therapy that consisted of either 3 courses of high-dose cytarabine or 4 courses of standard-dose therapy. Overall survival rates at 5 years were 48% for the daunorubicin group and 48% for the idarubicin group ($P = .54$), and relapse-free survival rates at 5 years were 41% and 41% ($P = .97$), respectively. Thus, high-dose daunorubicin and standard-dose idarubicin were equally effective for the treatment of adult acute myeloid leukemia, achieving a high rate of complete remission and good long-term efficacy. This study is registered at http://www.umin.ac.jp/ctrj/ as C000000157. (*Blood*. 2011;117(8): 2358-2365)

Introduction

The combination of anthracycline and cytarabine (Ara-C) with or without other antileukemia drugs is a standard induction therapy for acute myeloid leukemia (AML),[1-3] and a combination of daunorubicin at a dose of 45 to 50 mg/m^2 given daily for 3 days and Ara-C at a dose of 100 to 200 mg/m^2 given daily for 7 days generally has been used. In the late 1980s, however, idarubicin was introduced into clinics, and 3 randomized studies comparing idarubicin with daunorubicin reported significantly higher complete remission (CR) rates in favor of idarubicin.[4-6] A meta-analysis also confirmed a superior effect of idarubicin at a dose of 10 to 12 mg/m^2 for 3 days versus daunorubicin at a dose of 45 to 60 mg/m^2 for 3 days in the achievement of CR.[7] Nevertheless, the long-term follow-up of the above-mentioned 3 randomized studies comparing idarubicin with daunorubicin revealed that the idarubicin group had better overall survival (OS) than the daunorubicin group in only 1 study.[8]

The Japan Adult Leukemia Study Group (JALSG) used idarubicin and Ara-C as induction therapy in the AML95 and AML97 studies,[9-11] after idarubicin was registered and approved for the national health insurance system in 1995. Both studies resulted in satisfactorily high CR rates (80% and 79%, respectively); however, these CR rates were not superior to those of our earlier AML87, AML89, and AML92 studies, which used daunorubicin in combination with other antileukemia drugs.[12-14] In these 3 previous studies,

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daunorubicin and other drugs were administered in a response-oriented individualized manner; that is, additional drugs were given for a few days when the bone marrow at day 8 was not hypoplastic, containing a substantial number of blasts. Therefore, the total doses of daunorubicin administered during the first course of induction therapy were 240 to 280 mg/m^2 given for more than 5 to 7 days, which was more than the conventional dose of 40 to 60 mg/m^2 given for 3 days. Usui et al also reported that the optimal dose of daunorubicin in their induction therapy for newly diagnosed adult AML was approximately 280 mg/m^2 (40 mg/m^2 for 7 days).[15]

Because there had been no prospective randomized study comparing a higher dose of daunorubicin with the standard dose of idarubicin (12 mg/m^2) in adult AML, in the present multi-institutional randomized study, we prospectively compared idarubicin (12 mg/m^2 for 3 days) with daunorubicin (50 mg/m^2 for 5 days), in combination with Ara-C (100 mg/m^2 for 7 days), as induction therapy for previously untreated adult AML. High-dose daunorubicin resulted in the same CR rate and predicted 5-year OS compared with standard-dose idarubicin.

Methods

Patients

From December 2001 to December 2005, 1064 newly diagnosed adult patients 15 to 64 years of age with de novo AML were consecutively registered from 129 participating institutions. AML was first diagnosed by the French-American-British (FAB) classification at each institution. Peripheral blood and bone marrow smears from all registered patients were sent to Nagasaki University and examined by May-Giemsa, peroxidase, and esterase staining. Next, diagnosis was reevaluated by the central review committee. Patients with the FAB M3 subtype were not registered in the present study. Eligibility criteria included adequate function of liver (serum bilirubin level < 2.0 mg/dL), kidney (serum creatinine < 2.0 mg/dL), heart, and lung and an Eastern Cooperative Oncology Group performance status between 0 and 3. Patients were not eligible if they had prediagnosed myelodysplastic syndrome, but they were eligible if they had no definite diagnosis of myelodysplastic syndrome confirmed by bone marrow histologic analysis even when they had a previous history of hematologic abnormality. Cytogenetic abnormalities were grouped by standard criteria and classified according to the Medical Research Council classification.[16] The study was approved by the institutional review boards at each participating institution. Written informed consent was obtained from all patients before registration in accordance with the Declaration of Helsinki. The study was registered at http://www.umin.ac.jp/ctr/ as C000000157.

Treatments

Patients were randomly assigned by use of a centralized computer system to receive either idarubicin or daunorubicin. Randomization was stratified by age (younger or older than 50 years) and type of AML (FAB classification). All patients received 100 mg/m^2/d Ara-C by 24-hour continuous infusion from days 1 to 7. In the idarubicin group, patients received 12 mg/m^2/d idarubicin for 3 days, and in the daunorubicin group, they received 50 mg/m^2/d daunorubicin for 5 days. If patients did not achieve CR by the first course, the same induction therapy was repeated after an approximately 3- to 4-week interval. If patients did not achieve CR with 2 courses, they were judged as failure cases.

All patients who achieved CR were again randomized to receive either 4 courses of conventional consolidation therapy or 3 courses of high-dose Ara-C therapy. In the conventional consolidation-therapy group, the first course consisted of mitoxantrone (7 mg/m^2 by 30-minute infusion on days 1 to 3) and Ara-C (200 mg/m^2 by 24-hour continuous infusion on days 1 to 5). The second course consisted of daunorubicin (50 mg/m^2 by 30-minute infusion on days 1 to 3) and Ara-C (200 mg/m^2 by 24-hour continuous infusion on days 1 to 5). The third course consisted of aclarubicin (20 mg/m^2 by 30-minute infusion on days 1 to 5) and Ara-C (200 mg/m^2 by 24-hour continuous infusion on days 1 to 5). The fourth course consisted of Ara-C (200 mg/m^2 by 24-hour continuous infusion on days 1 to 5), etoposide (100 mg/m^2 by 1-hour infusion on days 1 to 5), vincristine (0.8 mg/m^2 by bolus injection on day 8), and vindesine (2 mg/m^2 by bolus injection on day 10). Each consolidation was administered as soon as possible after the neutrophils, white blood cells (WBCs), and platelets recovered to more than 1.5×10^9/L, 3.0×10^9/L, and 100×10^9/L, respectively. In the high-dose Ara-C group, 3 courses of 2.0 g/m^2 Ara-C were given by 3-hour infusion every 12 hours on days 1 to 5. Each course was administered 1 week after the neutrophils, WBCs, and platelets recovered to the above counts.

The best supportive care, including administration of antibiotics and platelet transfusions, was given as indicated. When patients had life-threatening documented infections during neutropenia, the use of granulocyte colony-stimulating factor was permitted.

After completion of consolidation therapy, no patients received further chemotherapy. Allogeneic stem cell transplantation (SCT) was offered during the first CR to patients 50 years of age or younger and with a histocompatible donor in the intermediate or adverse cytogenetic risk groups.

Definitions and study end points

Responses were evaluated according to the recommendations of the International Working Group.[17] CR was defined as the presence of all of the following: fewer than 5% blasts in bone marrow, no leukemic blasts in peripheral blood, recovery of peripheral neutrophil counts to more than 1.0×10^9/L and platelet counts to more than 100×10^9/L, and no evidence of extramedullary leukemia. Relapse after CR was defined as the presence of at least 1 of the following: reappearance of leukemic blasts in the peripheral blood, recurrence of more than 5% blasts in the bone marrow not attributable to any other cause (eg, bone marrow regeneration after consolidation therapy), and appearance of extramedullary leukemia.

This was a multi-institutional, randomized, phase 3 study with a 2×2 factorial design. The primary end point of the first randomization was CR rate. The result of the second randomization is reported here in part but will be presented fully in a separate paper. OS was calculated from the date of entry into the study until death due to any cause and was censored at the last follow-up. Relapse-free survival (RFS) for patients who achieved CR was measured from the date of CR until the date of AML relapse or death of any cause and was censored at the last follow-up. Patients who underwent allogeneic SCT were not censored at the date of SCT.

Statistical analysis

This study was prospectively powered to demonstrate noninferiority of daunorubicin compared with idarubicin. With a sample size of 420 patients per group (840 in total), the study had a power of 90% at a 1% level of significance to demonstrate noninferiority (assuming an 80% CR rate for both groups). Statistical testing for the noninferiority trial was performed according to the method of Blackwelder.[18] The Kaplan-Meier method was used to estimate probabilities of OS and RFS.[19] To test factors that predict CR, the χ^2 test and Wilcoxon rank sum test were used for univariate analysis, and the multiple logistic regression model was used for multivariate analysis. For comparison of OS and RFS, the log-rank test was used for univariate analysis and the proportional hazard model of Cox for multivariate analysis.[20,21] Cumulative rates of CR, neutrophil recovery, and platelet recovery were estimated according to the Kaplan-Meier method and were evaluated with the log-rank test. The JMP program (SAS Institute Inc) was used for these analyses. All analyses were performed according to the intention-to-treat principle. All statistical tests except the method of Blackwelder were 2-sided, and the significance level was set at .05.