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ORIGINAL REPORT

Long-Term Disease-Free Survival After Gemtuzumab, Intermediate-Dose Cytarabine, and Mitoxantrone in Patients With CD33$^+$ Primary Resistant or Relapsed Acute Myeloid Leukemia

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ABSTRACT

Purpose
To determine the antitumor activity and safety of a combination of gemtuzumab ozogamicin (GO), intermediate-dose cytarabine, and mitoxantrone (MIDAM) in patients with refractory or relapsed CD33$^+$ acute myeloid leukemia (AML).

Patients and Methods
We treated 62 patients with refractory (n = 18) or relapsed (n = 44) CD33$^+$ AML. Median age was 55.5 years. Salvage regimen consisted of GO 9 mg/m^2 on day 4, cytarabine 1 g/m^2 every 12 hours on days 1 through 5, and mitoxantrone 12 mg/m^2/d on days 1 through 3. Median follow-up time was 26.5 months.

Results
Thirty-one patients (50%) achieved complete remission (CR), and eight patients (13%) had CR with delayed platelet recovery (CRp); the overall response (OR; CR + CRp) rate was 63%. A significantly higher OR rate was achieved in patients who had relapsed versus refractory AML (73% *v* 39%, respectively; $P = .007$) and patients with CD33 expression more than 98% of the blast population versus less than 98% (79% *v* 52.3%, respectively; $P = .03$). The overall, event-free, and disease-free survival rates were 41%, 33%, and 53% at 2 years, respectively. Leukocytosis more than 20,000/μL at MIDAM therapy, high-risk cytogenetics, and absence of postremission therapy were adverse prognostic factors. Age, disease status, and/or CD33 expression did not influence survival parameters. Four early toxic deaths occurred; a grade 3 to 4 hyperbilirubinemia rate of 16% was observed, and two patients had veno-occlusive disease (3%).

Conclusion
The MIDAM regimen seems to be an effective salvage regimen for refractory/relapsed CD33$^+$ AML patients. These encouraging results support the need for a randomized phase III trial before considering this combination of GO and chemotherapy as superior or the standard of care treatment for refractory/relapsed CD33$^+$ AML patients.

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INTRODUCTION

Patients with relapsed or refractory acute myeloid leukemia (AML) usually have a poor outcome. In this population, chemotherapeutic salvage regimens can yield complete remission (CR) in less than 50% of patients, with a 3-year overall survival (OS) rate of less than 30%.[1-5] In addition, patients whose first CR (CR1) duration is less than 1 year or who do not achieve CR have a worse outcome[2] and may benefit little from autologous or allogeneic stem-cell transplantation.[6,7]

Gemtuzumab ozogamicin (GO; Mylotarg; Wyeth Oncology, Madison, NJ) is a humanized anti-CD33 antibody conjugated with a cytotoxic antitumor antibiotic, calicheamicin, that can target the CD33 antigen, a sialic acid–dependent adhesion protein found on the surface of leukemic myeloid blasts but not on normal hematopoietic stem cells.[8] Binding of the anti-CD33 antibody portion of GO with the CD33 antigen results in the formation of a complex that is internalized. The calicheamicin derivative is then released in the lysosomes of the myeloid cell, resulting in DNA double-strand breaks

and cell death. With this background, GO at a dose of 9 mg/m^2 (days 1 and 14) has been approved by the US Food and Drug Administration for the treatment of CD33$^+$ AML patients older than 60 years of age in first relapse. GO used as monotherapy for such patients has shown a 26% overall response (OR) rate, with a median disease-free survival (DFS) time of 5.2 months in patients achieving CR or CR with incomplete platelet recovery (CRp).[9,10] More recently, several studies tested the use of GO in combination with chemotherapy, either at diagnosis or at time of relapse.[11,12] However, because of its potential liver toxicity (veno-occlusive disease [VOD]), GO is generally used at lower doses when combined with chemotherapy, especially in patients who are candidates for transplantation within a short interval from GO administration.[9,10,13] We have previously reported our experience with the combination of intermediate-dose cytarabine and mitoxantrone plus a full dose of GO at 9 mg/m^2 (MIDAM) in a cohort of 17 patients with refractory or relapsed CD33$^+$ AML. The OR rate was 76%, and the median OS and DFS times were 11 and 11 months, respectively.[14] The current report provides an update of the results of the MIDAM regimen in a larger cohort of 62 patients.

PATIENTS AND METHODS

Patients

This study included 62 consecutive patients (29 men) treated in four institutions between February 2001 and January 2007. Patients had either refractory (n = 18) or relapsed (n = 44) AML. The relatively small number of patients included during this 6-year period was a result of the limited availability of GO in France and the need for a specific approval delivered by the Agence Française de Securite Sanitaire des Produits de Sante (equivalent to US Food and Drug Administration in France) on a single-patient basis. Median age was 55 years (range, 16 to 71 years), and 22 patients were age ≥ 60 years.

The study inclusion criteria were as follows: CD33$^+$ AML; primary refractory AML (after two courses of induction therapy including high-dose cytarabine in patients age < 60 years, after one course of induction therapy in patients age ≥ 60 years; or relapse < 6 months after CR1); first relapse for patients age ≥ 60 years (> 6 months from CR1) or first relapse within 12 months or after stem-cell transplantation (autograft or allograft) for patients younger than age 60 years. CD33 expression level was determined by dividing the mean fluorescence intensity of CD33 on the leukemic blasts by the mean fluorescence intensity of its isotypic control. The threshold for positivity was defined by a ratio of ≥ 2. Patient characteristics are listed in Table 1.[15-18] According to the French-American-British classification, the series included five M0, 15 M1, 19 M2, one M3, 10 M4, six M5, three M6, and two M7 patients (one patient was unclassified). Six patients had secondary AML (therapy related, n = 3; prior myeloproliferative disorder, n = 3). Prior treatments are listed in Table 1. Among patients experiencing relapse, 22 had received a prior transplantation (autograft, n = 20; allograft, n = 2). The median duration of CR1 in patients who had relapse was 15 months (range, 7 to 120 months). The median percentage of CD33$^+$ blasts was 98% (range, 65% to 100%), with a relative fluorescence intensity of 18.5 (range, 2 to 201). Six, 45, and 11 patients presented with good, intermediate, and poor cytogenetics, respectively (according to Grimwade et al[19]). Data were updated as of October 2007. Patients were fully informed of the treatment modalities and had provided informed consent. The median follow-up time for surviving patients was 26.5 months (range, 7.5 to 78 months).

Salvage Regimen

The MIDAM protocol consisted of GO 9 mg/m^2 intravenously (IV) over 2 hours on day 4, cytarabine 1 g/m^2 every 12 hours IV over 2 hours on days 1 to 5, and mitoxantrone 12 mg/m^2/d IV over 30 minutes on days 1 to 3. Patients received granulocyte colony-stimulating factor from day 7 after chemotherapy until neutrophil recovery. In addition, irrespective of their platelet count, patients received low-dose heparin for VOD prophylaxis. Heparin was admin-

Table 1. Patient Characteristics

Characteristic	No. of Patients (N = 62)	%
Sex		
Male	29	
Female	33	
Age, years		
Median	55.5	
Range	16-71	
< 60	40	
≥ 60	22	
FAB classification		
M0	5	
M1	15	
M2	19	
M3	1	
M4	10	
M5	6	
M6	3	
M7	2	
Unknown	1	
Cytogenetics		
Favorable	6	
Intermediate	45	
High risk	11	
Refractory AML	18	29
Relapsed AML	44	71
Patients < 60 years old		
Relapse between 6 and 12 months	11	25
Relapse between 12 and 24 months	12	27
Relapse > 24 months	4	9
Patients ≥ 60 years old		
Relapse between 6 and 12 months	5	11.5
Relapse between 12 and 24 months	7	16
Relapse > 24 months	5	11.5
CD33 expression, %		
Median	98	
Range	65-100	
Previous treatment	62	
Patients in relapse and < 60 years old		
Goelams-LAM2001 trial*	27	
Patients in relapse and ≥ 60 years old		
Goelams-LAMSA2002 trial†	10	
ALFA-9801 trial‡	6	
ALFA-9803 trial§	1	
Refractory patients < 60 years old		
Goelams-LAM2001 trial (2 inductions)	13	
Refractory patients ≥ 60 years old		
Goelams-LAMSA2002 trial (1 induction)	5	
Previous autograft	22	
Previous allograft	2	
Overall responses (CR + CRp)		
All patients	39	63
< 60 years old, n = 40	24	60
≥ 60 years old, n = 22	15	68
Patients in relapse, n = 44	32	73
First CR between 6 and 12 months, n = 16	13	81
First CR between 12 and 24 months, n = 19	12	63
First CR > 24 months, n = 9	7	77
< 60 years old, n = 27	19	70
≥ 60 years old, n = 17	13	76
Refractory patients, n = 18	7	39
< 60 years old, n = 13	5	38
≥ 60 years old, n = 5	2	40
Secondary AML, n = 6	2	33
High-risk cytogenetics, n = 11	5	45

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Table 1. Patient Characteristics (continued)

Characteristic	No. of Patients (N = 62)	%
Postremission therapy in CR + CRp patients	39	63
None	8	
Chemotherapy alone	16	
Low-dose cytarabine	9	
High-dose cytarabine	7	
Chemotherapy then DLI	1	
Chemotherapy then autograft	1	
Chemotherapy then RIC allograft	7	
Autograft	1	
Allograft	5	
Standard conditioning regimen	1	
RIC	4	
Cause of death	38	
Relapse or progression	28	
Sepsis	4	
Acute GVHD	2	
Hemorrhage	2	
Multiple organ failure	1	
VOD	1	

Abbreviations: FAB, French-American-British; AML, acute myeloid leukemia; CR, complete remission; CRp, complete remission with delayed platelet recovery; DLI, donor lymphocyte infusion; RIC, reduced-intensity regimen; GVHD, graft-versus-host disease; VOD, veno-occlusive disease.

*Goelams-LAM2001 trial[15]: induction included cytarabine 200 mg/m^2 on days 1 to 7 plus daunorubicin 60 mg/m^2 on days 1 to 3 or idarubicin 8 mg/m^2 on days 1 to 5; second induction included cytarabine 1 g/m^2 for four doses over 2 days plus daunorubicin 35 mg/m^2 days 1 to 2 or idarubicin 8 mg/m^2 on days 1 to 2.

†Goelams-LAMSA2002 trial[16]: induction included cytarabine 100 mg/m^2 on days 1 to 7 plus idarubicin 8 mg/m^2 on days 1 to 5 plus lomustine 200 mg/m^2 on day 1.

‡ALFA-9801 trial[17]: induction included cytarabine 200 mg/m^2 on days 1 to 7 plus daunorubicin 80 mg/m^2 on days 1 to 3 or idarubicin 12 mg/m^2 on days 1 to 3 or 4.

§ALFA-9803 trial[18]: induction included cytarabine 200 mg/m^2 on days 1 to 7 plus daunorubicin 45 mg/m^2 or idarubicin 9 mg/m^2 on days 1 to 4.

istered from day 1 of MIDAM until neutrophil recovery (100 U/kg/d).[20] Antibiotics and antifungal prophylaxis were administered according to each center's practice.

Postremission Therapy

Postremission therapy was not predefined. Among patients who achieved CR or CRp (n = 39), eight patients did not receive further therapy after the MIDAM regimen because of poor performance status. Sixteen patients received consolidation with chemotherapy alone including high-dose cytarabine in seven patients (with a second course of GO in two patients) and low-dose cytarabine in nine patients. One patient received chemotherapy followed by an autograft. One patient received chemotherapy followed by donor lymphocyte infusion, and one patient received an autograft as consolidation after MIDAM.

Twelve patients could proceed to allogeneic hematopoietic stem-cell transplantation (HSCT) directly after MIDAM (n = 5) or after consolidation by high-dose cytarabine (n = 7). HSCT consisted of a standard matched unrelated allogeneic HSCT in one patient, a matched related reduced-intensity conditioning (RIC) HSCT in two patients, a matched unrelated RIC HSCT in eight patients, and a cord blood RIC HSCT in one patient. Postremission therapy details are listed in Table 1.

Response Criteria

CR was defined as normalization of blood and bone marrow with $\leq 5\%$ of blasts, neutrophil count more than 1.5×10^9/L, and platelet count more than 100×10^9/L. CRp was defined the same as for CR, except the criteria included platelet transfusion independence and a platelet count remaining less than 100×10^9/L. OR was defined as CR plus CRp.

Statistical Analysis

The primary objective of the study was CR achievement. Secondary objectives included the safety analysis, OS, event-free survival (EFS), and DFS. OS, EFS, and DFS were estimated using the Kaplan-Meier product-limit method. Survival times were calculated from the day of the beginning of the chemotherapy until death or last follow-up for OS; until treatment failure, relapse, death, or last follow-up for EFS; and until death, relapse, or last follow-up in responders (CR + CRp) for DFS. The log-rank test was used to compare the CR rates, OS, EFS, and DFS between the subgroups, according to age, sex, leukocytosis at time of MIDAM administration, CD33 expression (percentage of positive blasts and relative fluorescence intensity), cytogenetics, status at time of MIDAM administration (relapsed or refractory AML), duration of CR1 for relapsed patients, response after MIDAM, and postremission treatments. Data were computed using the STATA software, version 8.2 (STATA Corp, College Station, TX).

RESULTS

Response Rate

In this series, the OR (CR + CRp) rate was 63% (n = 39), and the CR rate was 50% (n = 31). The median time to remission (CR or CRp) was 44 days (range, 22 to 73 days). Treatment failure was observed in 19 patients. Four patients were not assessable because of early toxic death. Response rates according to age, status at transplantation, cytogenetics, and duration of CR1 for relapsed patients are listed in Table 1. Among patients in relapse, there was no difference in terms of OR between patients whose CR1 lasted less than 12 months (81%; 13 of 16 patients) compared with patients who experienced relapse between 12 and 24 months after CR1 (63%; 12 of 19 patients) or patients who experienced relapse more than 24 months after CR1 (77%; seven of nine patients). Also, there was no difference in terms of OR when considering patients younger than age 60 years (60%; 24 of 40 patients) versus $\geq$ age 60 years (68%; 15 of 22 patients); patients in relapse younger than age 60 years (70%; 19 of 27 patients) versus $\geq$ age 60 years (76%; 13 of 17 patients); and refractory patients younger than age 60 years (38%; five of 13 patients) versus $\geq$ age 60 years (40%; two of five patients). Patients in relapse had a significantly higher OR rate compared with refractory patients (73% *v* 39%, respectively; $P = .007$), and patients with CD33$^+$ expression $\geq 98\%$ of blast population had a significantly higher OR rate than patients with $< 98\%$ blasts (79% *v* 53%, respectively; $P = .03$).

OS, EFS, and DFS

The median OS and EFS times were 9.5 months (range, 8 days to 76.5 months) and 4.4 months (range, 8 days to 76.5 months), respectively. Two-year OS and EFS rates were 41% and 33%, respectively (Fig 1A). The median DFS and OS for responders were not yet reached at 2 years (DFS, 53% at 2 years; 95% CI, 27% to 53%; OS, 58% at 2 years; 95% CI, 39% to 72%). The cumulative rates of relapse were 38% and 41% at 1 and 2 years, respectively. Thirty-nine patients (63%) died, including four early toxic deaths. The cause of death was relapse in 28 patients, sepsis in four patients, acute graft-versus-host disease after allogeneic HSCT in two patients, hemorrhage in two patients, multiple organ failure in one patient, and VOD in one patient.

In a Cox multivariate analysis, leukocytosis more than 20×10^9/L at time of MIDAM and high-risk cytogenetics were adverse prognosis factors for OS (hazard ratio [HR] = 7.9; 95% CI, 1.95 to 32.03; $P = .004$; and HR = 4.7; 95% CI, 1.12 to 20.12; $P = .03$, respectively) and DFS (HR = 5.05; 95% CI, 1.35 to 18.87; $P = .01$; and

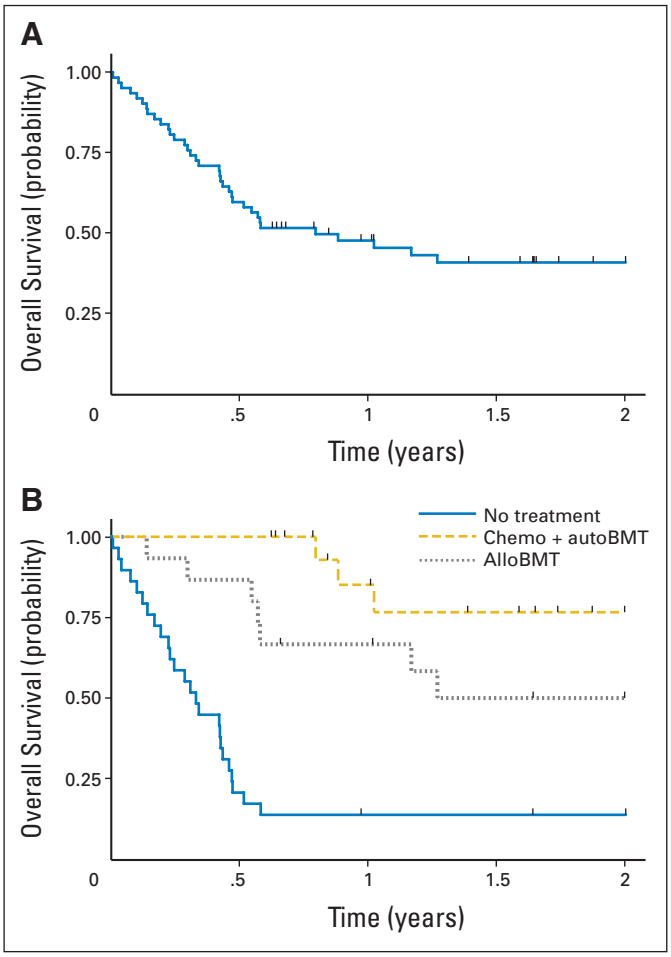

Fig 1. (A) Overall survival (OS) of the whole cohort. (B) OS of responders according to postremission treatment (no treatment: 13% at 2 years; allograft: 50% at 2 years; chemotherapy ± autograft: 76% at 2 years) autoBMT, autologous bone marrow transplantation; AlloBMT, allogeneic bone marrow transplantation.

HR = 4.65; 95% CI, 1.35 to 15.95; $P = .01$, respectively). Conversely, patients in CR (*v* patients in CRp) and patients receiving postremission therapy (chemotherapy ± autograft and/or allograft *v* none) had a significantly improved OS (HR = 0.28; 95% CI, 0.07 to 1.02; $P = .05$; and HR = 0.13; 95% CI, 0.03 to 0.48; $P = .002$, respectively) and/or DFS (HR = 0.39; 95% CI, 0.12 to 1.26; $P = .11$; and HR = 0.17; 95% CI, 0.05 to 0.55; $P = .003$). Age, disease status, and CD33 expression did not influence survival parameters. In addition, allograft was not superior to chemotherapy ± autograft as postremission therapy (OS at 2 years: 50% for allograft *v* 76% for chemotherapy; P = not significant; Fig 1B). Among 12 patients who received allograft, seven are alive in CR, and five have died because of relapse (n = 3) and acute graft-versus-host disease (n = 2).

Toxicity

The median time of hospitalization was 29.5 days (range, 27 to 33 days). All patients developed grade 4 neutropenia and thrombocytopenia. Overall, the median times to neutrophil recovery ($> 0.5 \times 10^9$/L) and to platelet recovery ($> 50 \times 10^9$/L) were 25 days (range, 23 to 27.3 days) and 41 days (range, 34 to 46 days), respectively. The median times to neutrophil and platelet recovery in patients who did not respond to treatment were 30 and 60 days, respectively. Grade 0, 1, 2, 3, and 4 hyperbilirubinemia occurred in 27, 18, seven, six, and four patients, respectively, with an overall grade 3 to 4 hyperbilirubinemia rate of 16%. VOD was observed in only two patients (3%), with complete resolution in one patient and fatal outcome in the other. Four early deaths occurred, on day 8 (intra-alveolar hemorrhage), on day 12 (multiple organ failure), on day 18 (*Pseudomonas aeruginosa* sepsis), and on day 29 (VOD). All patients had fever during hospitalization. MIDAM administration was complicated with documented bacterial sepsis in 34 patients (55%). Moreover, seven patients had invasive aspergillosis, one patient had *Candida tropicalis* fungal infection, and one patient had *Fusobacterium* fungal infection. Cardiac tamponade, cholecystitis, and *Clostridium difficile*–associated pseudomembranous colitis occurred in three patients.

DISCUSSION

Thus far, this study describes the largest series of patients treated with a combination of GO plus chemotherapy for refractory or relapsed CD33+ AML. Responses (CR + CRp) were achieved in 63% of patients, with OS, EFS, and DFS rates of 41%, 33%, and 53% at 2 years, respectively. The median DFS and OS times for responders were not reached at 2 years, with no significant differences between refractory or relapsed AML patients. In this setting of refractory/relapsed AML, these results seem to be superior to those observed with chemotherapy alone, GO as monotherapy, or GO plus chemotherapy.[1-6,9-11] The drug combination we used may help explain these interesting results. First, the combination of mitoxantrone and intermediate-dose cytarabine is known as one of the best salvage regimens for refractory/relapsed leukemia patients, yielding up to a 49% OR rate.[1] Second, we used a full dose of GO (9 mg/m^2) administered at day 4 after the beginning of chemotherapy. Delayed perfusion of GO at day 4 may allow the reduction of CD33 antigen loads in the peripheral blood and, consequently, increase the penetration of GO into bone marrow.[21] These improved results might be also attributed to a relatively low toxicity of the MIDAM regimen because we only observed a 6.4% rate of early toxic death, 16% rate of grade 3 to 4 hyperbilirubinemia, and two patients with VOD. Of note, no VOD was observed after allogeneic transplantation, likely because most of these patients (11 of 12 patients) had received an RIC regimen and the median time between MIDAM and allograft was relatively long (median, 164 days; range, 91 to 316 days). Also, the systematic use of low-dose heparin prophylaxis might have helped to decrease VOD incidence.[20]

Results of this MIDAM regimen are encouraging in relapsed CD33+ AML patients because they achieved long-term DFS, irrespective of age and duration of CR1. However, as expected, a refractory disease status confers a significantly higher probability of treatment failure. Treatment failure in refractory patients can be explained by a higher multidrug resistance activity, which is already known to confer a poor prognosis both to chemotherapy and GO as monotherapy.[1,6,22-27] Other mechanisms may explain resistance to GO. Indeed, reduced binding capacity to leukemic blasts, antiapoptotic mechanisms independent of drug efflux, Bcl-2 antiapoptotic proteins, and resting state of cell cycle have been previously described in this context.[28-31] All of these resistance mechanisms may be targets to improve results of the MIDAM regimen. Interestingly, drugs such

as granulocyte colony-stimulating factor, HSP90 inhibitor, and azacytidine have already been shown to increase efficacy when combined with GO.[32-35]

In terms of response definition, the notion of CRp was developed specifically when GO was introduced because of its platelet-specific toxicity. In the current study, patients achieving CRp had a similar DFS and incidence of relapse compared with patients achieving CR (relapse rate: 37.5% *v* 32%, respectively). A worse DFS has been reported in patients with CRp but only if considering patients without postremission therapy after GO as monotherapy.[10] For the latter result, one explanation is that CRp patients likely express a higher residual tumor burden and should be considered, whenever possible, for consolidation after a salvage regimen because DFS and OS were similar for both the CR and CRp groups after HSCT.[10] In our series, the addition of chemotherapy to GO could explain that CR and CRp patients achieved similar DFS as the percentages of CR or CRp patients receiving consolidation after MIDAM have shown no significant differences (84% *v* 50%; P = NS).

From a biologic standpoint, high CD33 expression was shown to be associated with a better clinical outcome in AML patients receiving GO likely because CD33 expression is inversely correlated with multidrug resistance activity.[25,26] Thus, one can hypothesize that CD33 expression could be a surrogate marker for some unrelated parameters that enhance chemotherapy sensitivity. In the present study, we found a close relationship between high expression of CD33 ($>$ 98% of the blast population) and improved OR, suggesting that higher CD33 expression may help to eradicate all of the leukemic blast population without development of a CD33 negative clonal population.

Despite a high OR rate and acceptable toxicity, postremission therapy after MIDAM is a prerequisite to achieve an improved long-term outcome. Patients who did not receive further treatment after MIDAM had a poor performance status or a persistent infectious complication. Although there was no difference in terms of survival between patients receiving chemotherapy ± autograft or allograft as consolidation, the relatively acceptable toxicity profile of the MIDAM regimen allowed several patients to proceed to an allogeneic stem-cell transplantation, which is a potentially curative option in this context.[6,10] Of note, two patients received a second course of GO plus chemotherapy without additional toxicity as it had already been reported in AML patients at diagnosis, at relapse, or both.[36]

In conclusion, the MIDAM regimen produces a high rate of OR and improves outcome in patients with relapsed/refractory $CD33^+$ AML. Postremission therapy is required to achieve long-term favorable outcome. These encouraging results support the need for a prospective, randomized, phase III trial before considering this combination of GO plus chemotherapy as a superior or standard of care treatment for refractory/relapsed $CD33^+$ AML.

AUTHORS' DISCLOSURES OF POTENTIAL CONFLICTS OF INTEREST

The author(s) indicated no potential conflicts of interest.

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REFERENCES

1. Solary E, Witz B, Caillot D, et al: Combination of quinine as a potential reversing agent with mitoxantrone and cytarabine for the treatment of acute leukemias: A randomized multicenter study. Blood 88:1198-1205, 1996

2. Estey EH: Treatment of relapsed and refractory acute myelogenous leukemia. Leukemia 14: 476-479, 2000

3. Lee S, Tallman MS, Oken MM, et al: Duration of second complete remission compared with first complete remission in patients with acute myeloid leukemia: Eastern Cooperative Oncology Group. Leukemia 14:1345-1348, 2000

4. Tavernier E, Le QH, Elhamri M, et al: Salvage therapy in refractory acute myeloid leukemia: Prediction of outcome based on analysis of prognostic factors. Leuk Res 27:205-214, 2003

5. Leopold H, Willemze R: The treatment of acute myeloid leukaemia in first relapse: A comprehensive review of the literature. Leuk Lymphoma 43:1715-1727, 2002

6. Löwenberg B, Downing J, Burnett A: Acute myeloid leukemia. N Engl J Med 341:1051-1062, 1999

7. Thomas X, Le Q, Botton S, et al: Autologous or allogeneic stem cell transplantation as postremission therapy in refractory or relapsed acute myeloid leukemia after highly intensive chemotherapy. Leuk Lymphoma 46:1007-1016, 2005

8. Dinndorf PA, Andrews RG, Benjamin D, et al: Expression of normal myeloid associated antigens by acute leukemia cells. Blood 67:1048-1053, 1986

9. Sievers EL, Larson RA, Stadtmauer EA, et al: Efficacy and safety of gemtuzumab ozogamicin in patients with CD33 positive acute myeloid leukemia in first relapse. J Clin Oncol 19:3244-3254, 2001

10. Larson RA, Sievers EL, Stadtmauer EA, et al: Final report of the efficacy and safety of gemtuzumab ozogamicin (Mylotarg) in patients with CD33-positive acute myeloid leukemia in first recurrence. Cancer 104:1442-1452, 2005

11. Stasi R, Evangelista ML, Buccisano F, et al: Gemtuzumab ozogamicin in the treatment of acute myeloid leukaemia. Cancer Treat Rev 34:49-60, 2008

12. Burnett AK, Kell WJ, Goldstone AH, et al: The addition of gemtuzumab ozogamicin to induction chemotherapy for AML improves disease free survival without extra toxicity: Preliminary analysis of 1115 patients in the MRC AML15 trial. Blood 108: 8a, 2006 (abstr 13)

13. Wadleigh M, Richardson PG, Zahrieh D, et al: Prior gemtuzumab ozogamicin exposure significantly increases the risk of veno-occlusive disease in patients who undergo myeloablative allogeneic stem cell transplantation. Blood 102:1578-1582, 2003

14. Chevallier P, Roland V, Mahe B, et al: Administration of Mylotarg 4 days after beginning of a chemotherapy including intermediate-dose aracytin and mitoxantrone (MIDAM regimen) produces a high rate of complete hematologic remission in patients with CD33+ primary resistant or relapsed acute myeloid leukaemia. Leuk Res 29:1003-1007, 2005

15. Lioure B, Delaunay J, Blaise D, et al: Allogeneic stem cell transplantation (SCT) with non myeloablative conditioning regimen (NST) following intensive consolidation may be equivalent to conventional alloSCT and superior to auto-SCT for patients over 50 with acute myeloid leukemia (AML) in first CR: First results of the AML 2001 trial. Blood 108:319, 2006 (abstr)

16. Pigneux A, Himberlin C, Hunault-Berger M, et al: A multicenter randomized comparison of maintenance treatment with androgens in elderly acute myeloid leukemia after ICL regimen as induction therapy: Results of the Goelams-2002 study. Blood 108:1983, 2006 (abstr)

17. Pautas C, Thomas X, Merabet F, et al: Randomized comparison of standard induction with daunorubicin (DNR) for 3 days vs idarubicin (IDA) for 3 or 4 days in AML pts aged 50 to 70 and maintenance with interleukin 2: Final analysis of the ALFA 9801 study. Blood 110:162, 2007 (abstr)

18. Turlure P, Gardin C, Malfuson JV, et al: Impact of post-remission therapy on survival of older patients with AML treated with intensive chemotherapy: Results of the multicenter ALFA-9803 trial. Blood 108:421, 2006 (abstr)

19. Grimwade D, Walker H, Oliver F, et al: The importance of diagnostic cytogenetics on outcome in AML: Analysis of 1,612 patients entered into the MRC AML 10 trial—The Medical Research Council Adult and Children's Leukaemia Working Parties. Blood 92:2322-2333, 1998

20. Attal M, Huguet F, Rubie H, et al: Prevention of hepatic veno-occlusive disease after bone marrow transplantation by continuous infusion of low-dose heparin: A prospective, randomized trial. Blood 79:2834-2840, 1992

21. van der Velden VH, Marvelde JG, Hoogeveen PG, et al: Targeting of the CD33-calicheamicin immunoconjugate Mylotarg (CMA-676) in acute myeloid leukaemia: In vivo and in vitro saturation and internalization by leukemic and normal myeloid cells. Blood 97:3197-3204, 2001

22. Tsimberidou AM, Paterakis G, Androutsos G, et al: Evaluation of the clinical relevance of the expression and function of P-glycoprotein, multidrug resistance protein and lung resistance protein in patients with primary acute myelogenous leukaemia. Leuk Res 26:143-154, 2002

23. Biscardi M, Teodori E, Caporale R, et al: Multidrug reverting activity toward leukemia cells in a group of new verapamil analogues with low cardiovascular activity. Leuk Res 30:1-8, 2006

24. Linenberger ML, Hong T, Flowers D, et al: Multidrug-resistance phenotype and clinical responses to gemtuzumab ozogamicin. Blood 98:988-994, 2001

25. Walter RB, Raden BW, Hong TC, et al: Multidrug resistance protein attenuates gemtuzumab ozogamicin-induced cytotoxicity in acute myeloid leukemia cells. Blood 102:1466-1473, 2003

26. Walter RB, Gooley TA, Van der Velden VH, et al: CD33 expression and P-glycoprotein-mediated drug efflux inversely correlate and predict clinical outcome in patients with acute myeloid leukemia treated with gemtuzumab ozogamicin monotherapy. Blood 109:4168-4170, 2007

27. Taksin AL, Legrand O, Raffoux E, et al: High efficacy and safety profile of fractionated doses of Mylotarg as induction therapy in patients with relapsed acute myeloblastic leukaemia: A prospective study of the alfa group. Leukemia 21:66-71, 2007

28. van der Velden VH, Boeckx N, Jedema I, et al: High CD33-antigen loads in peripheral blood limit the efficacy of gemtuzumab ozogamicin (Mylotarg) treatment in acute myeloid leukemia patients. Leukemia 18:983-988, 2004

29. van der Kolk DM, de Vries EG, Muller M, et al: The role of drug efflux pumps in acute myeloid leukaemia. Leuk Lymphoma 43:685-701, 2002

30. Walter RB, Raden BW, Cronk MR, et al: The peripheral benzodiazepine receptor ligand PK11195 overcomes different resistance mechanisms to sensitize AML cells to gemtuzumab ozogamicin. Blood 103:4276-4284, 2004

31. Jedema I, Barge RM, van der Velden VH, et al: Internalization and cell cycle dependent killing of leukemic cells by gemtuzumab ozogamicin: Rationale for efficacy in CD33-negative malignancies with endocytic capacity. Leukemia 18:316-325, 2004

32. Rutella S, Bonanno G, Procoli A, et al: Granulocyte colony-stimulating factor enhances the in vitro cytotoxicity of gemtuzumab ozogamicin against acute myeloid leukaemia cell lines and primary blast cells. Exp Hematol 34:54-65, 2006

33. Leone G, Rutella S, Voso MT, et al: In vivo priming with granulocyte colony-stimulating factor possibly enhances the effect of gemtuzumab-ozogamicin in acute myeloid leukemia: Results of a pilot study. Haematologica 89:634-636, 2004

34. Walter RB, Bernstein ID: The HSP 90 inhibitor, 17-AAG, increases gemtuzumab ozogamicin-induced cytotoxicity in acute myeloid leukemia (AML) cells. 49th Annual Meeting of the American Society of Hematology, Atlanta, GA, December 8-11, 2007 (abstr 1607)

35. Balaian L, Ball ED: 5-Azacytidine augments the cytotoxicity of Mylotarg toward AML blasts in vitro and in vivo. 49th Annual Meeting of the American Society of Hematology, Atlanta, GA, December 8-11, 2007 (abstr 1835)

36. Chevallier P, Touzeau C, Ayari S, et al: Re-administration of a combination of chemotherapy + gemtuzumab at relapse in CD33+ AML patient allows to second remission and is feasible without extra-toxicity: A case report. Leuk Res 32:1321-1322, 2008

