Safety and Efficacy of Gemtuzumab Ozogamicin (Mylotarg®) in
Pediatric Patients With Advanced CD33-Positive
Acute Myeloid Leukemia

Running head: Antibody-Targeted Chemotherapy of Pediatric AML

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*The following co-authors were employed by the study sponsor, Wyeth Pharmaceuticals, Philadelphia, Pennsylvania, at the time of the study. They both worked directly on the Wyeth product, Gemtuzumab Ozogamicin, which is discussed in the above-referenced article. Mark S. Berger, MD and Matthew L. Sherman, MD
Both authors are now employed by Glaxo SmithKline Pharmaceuticals, Philadelphia, Pennsylvania and no longer have a financial interest in the above mentioned product.

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Scientific Heading: Clinical Observations, Interventions, and Therapeutic Trials
ABSTRACT

This open-label dose-escalation study evaluated the safety and efficacy of single-agent gemtuzumab ozogamicin (Mylotarg®), a humanized anti-CD33 antibody–targeted chemotherapeutic agent, for pediatric patients with multiple relapsed or primary refractory acute myeloid leukemia (AML). Twenty-nine children 1 to 16 years (relapse=19/refractory=10) received gemtuzumab ozogamicin ranging from 6 to 9 mg/m² per dose for 2 doses (separated by 2 weeks) infused over 2 hours. All patients had anticipated myelosuppression. Other toxicities included grade 3/4 hyperbilirubinemia (7%) and elevated hepatic transaminases (21%); the incidence of grade 3/4 mucositis (3%) or sepsis (24%) was relatively low. One patient treated at 9 mg/m² developed veno-occlusive disease (VOD) of the liver and defined the dose-limiting toxicity. Thirteen patients received hematopoietic stem cell transplantation <3.5 months after the last dose of gemtuzumab ozogamicin; 6 (40%) developed VOD. Eight of 29 patients achieved overall remission (28%). Remissions were comparable in refractory (30%) and relapsed (26%) patients. Mean multidrug resistance protein–mediated drug efflux was significantly lower in the leukemic blasts of patients achieving remission (P<0.005). Gemtuzumab ozogamicin was relatively well tolerated at 6 mg/m² for 2 doses and was equally effective in refractory and relapsed patients. Further studies in combination with standard induction therapy for childhood AML are warranted.
INTRODUCTION

Acute myeloid leukemia (AML) is generally considered to be a disease of older adults. The annual incidence of AML in people older than 65 years of age exceeds 10 per 100,000, making it the second most common form of leukemia in these adults. However, a substantial number of children and adolescents also are diagnosed with AML. The estimated incidence of AML in children up to 4 years of age is 0.9 per 100,000 and for 15 to 19 years of age, 0.8 per 100,000.1,2

With currently used induction regimens, more than 80% of children with AML achieve remission.3-6 Dose intensification by time compression or increased exposure during induction chemotherapy has improved survival.3,4 However, only about 50% of all children with newly diagnosed AML will be cured with currently available treatment.7 Although several randomized studies have demonstrated that allogeneic hematopoietic stem cell transplantation (HSCT) from a human leukocyte antigen–matched family donor can improve survival in children with AML in first remission compared with chemotherapy alone or autologous HSCT, some centers prefer to perform allogeneic HSCT in second remission for some favorable subgroups.8 Thus, the goal of chemotherapy for patients with relapsed AML is to achieve a remission in order for patients to undergo subsequent HSCT.

Gemtuzumab ozogamicin (Mylotarg®, CMA-676; Wyeth Pharmaceuticals, Philadelphia, PA) is a humanized monoclonal antibody directed against the CD33 surface antigen that is conjugated to a derivative of the cytotoxic antibiotic calicheamicin.9 Approximately 90% of patients with AML have myeloid blast cells that express significant levels of CD33.10 Upon binding, the antibody-antigen complex is internalized into target cells, and the calicheamicin molecule is released through hydrolysis. Calicheamicin binds to the minor groove of cellular DNA and causes double-strand breaks that lead to apoptosis.11
Gemtuzumab ozogamicin is approved by the US Food and Drug Administration (FDA) for the treatment of patients 60 years of age or older with CD33-positive AML in first relapse who are not eligible for other cytotoxic chemotherapy.\textsuperscript{12,13} Response rates of approximately 25\% have been observed in adult patients with refractory AML treated with gemtuzumab ozogamicin.\textsuperscript{14-16} The most clinically important toxicities associated with this drug have been abnormalities in hepatic function.\textsuperscript{15,16} Only limited data exist regarding the use of gemtuzumab ozogamicin for the treatment of refractory or relapsed AML in children.\textsuperscript{17-20} This phase I clinical trial was undertaken to evaluate the efficacy and tolerability of gemtuzumab ozogamicin as monotherapy in children younger than 18 years of age with relapsed or refractory CD33-positive AML.

METHODS

Patients

Twenty-nine children with CD33-positive AML who either had failed to respond to primary induction therapy (n=10) or had suffered a relapse (n=19) participated in this prospective, open-label, dose-escalation study conducted at 7 clinical research centers in the United States from 1999 to 2002 to evaluate the efficacy and safety of gemtuzumab ozogamicin as monotherapy. Children were considered to have CD33-positive AML if they had more than 80\% of leukemic blast cells with CD33-immunofluorescence staining 4 times above background staining. Other enrollment criteria included a Lansky play index or Karnofsky performance status of $\geq 60\%$, normal renal and liver function, and a peripheral white blood cell (WBC) count of $<30,000/\mu L$. Patients who developed AML secondary to chemotherapy or exposure to toxins were ineligible for enrollment. Patients with known central nervous system leukemia or testicular involvement with leukemia or patients who had undergone prior HSCT were also ineligible. Concomitant chemotherapy was prohibited.
The legal guardians of all patients provided signed informed consent, which was often also obtained from the patient. The study was approved and monitored by the Institutional Review Board at each center and was performed in a manner consistent with the Declaration of Helsinki and Good Clinical Practice guidelines.

**Gemtuzumab Ozogamicin Dosage**

Determining the maximum tolerated dose of gemtuzumab ozogamicin in pediatric patients was a primary objective of the study. Based on results obtained in the phase II trials using 9 mg/m² as a 2-hour intravenous infusion conducted in 142 adults with AML in first untreated relapse, the first pediatric patient cohort received gemtuzumab ozogamicin monotherapy starting with a dose of 6 mg/m² (0.2 mg/kg for patients <3 y of age) for 2 doses with a 14-day interval between doses. Dose-limiting toxicity (DLT) was defined as National Cancer Institute (NCI) grade 3 gemtuzumab ozogamicin–related nonhematologic toxicity. Exceptions were grade 3 infections, bleeding, nausea, vomiting, febrile neutropenia, or infusion-related adverse events that resolved within 24 hours or elevations in hepatic enzyme and bilirubin levels that were transient and reversible within 28 days of last dose. If DLT did not develop in 3 patients, the next 3-patient cohort was escalated to 9 mg/m² (0.3 mg/kg for <3 y of age) for 2 doses, 14 days apart. The final escalation planned was 12 mg/m² (0.4 mg/kg for <3 y of age). Toxicity was assessed after each infusion at all dose levels. In the event that ≥3 patients experienced a grade 3 or 1 patient experienced a grade 4 nonhematologic event, or any patient experienced veno-occlusive disease (VOD), de-escalation to the maximum tolerated dose (MTD) was allowed. The goal was to have 10 to 15 patients treated at the MTD, defined as the dose level just below the level at which DLT occurs.
Before receiving gemtuzumab ozogamicin, patients were required to have peripheral WBC counts of <30,000/µL; hydroxyurea treatment was allowed to reduce peripheral WBC counts to that level. Patients were routinely premedicated with acetaminophen (15 mg/kg intravenously or orally) and diphenhydramine (1 mg/kg) approximately 1 hour before gemtuzumab ozogamicin infusion. Two additional doses of acetaminophen were permitted: 1 at approximately 4 hours and the other at approximately 8 hours after the initial pretreatment dose. Steroids were not used as prophylactic medication before gemtuzumab ozogamicin administration. Patients were eligible to receive the second dose of gemtuzumab ozogamicin if they had recovered from reversible nonhematologic toxicities due to the previous dose and had no evidence of uncontrolled infection, disease progression, or detectable formation of antibodies reactive with the drug.

The outcome of gemtuzumab ozogamicin therapy was evaluated for the treatment period, defined as the time beginning from administration of the drug until 28 days after completion of therapy. Following recovery from treatment with gemtuzumab ozogamicin, patients received additional therapy thought most appropriate by their treating physician.

Safety Assessments

Adverse Events

The primary safety assessment was monitoring of adverse events, defined as treatment-emergent or infusion-related adverse events or any clinically significant abnormal laboratory finding. All patients who received at least 1 dose of study drug were included in the safety analysis. Safety was monitored continually during study drug administration and immediate follow-up, and then monthly for approximately 6 months. Treatment-emergent adverse events were events not present at baseline or those present at baseline that worsened during treatment. The severity of adverse events was evaluated using the NCI Common Toxicity Criteria version 1. Events with a
severity of grade 1 or 2 were considered to be mild or moderate and manageable. Grade 3 or 4
events were considered to be severe or life threatening. The presence and severity of VOD was
determined according to the criteria established by McDonald et al.21

Analysis for Antibodies Directed Against Components of Gemtuzumab Ozogamicin

Gemtuzumab ozogamicin contains 3 components: humanized monoclonal antibody hP67.6
against the CD33 antigen, a derivative of calicheamicin, and a linker connecting the antibody and
the calicheamicin derivative. Patients were screened for the production of antibodies directed
against any of these components. Details regarding the methodology of enzyme-linked
immunosorbent assays in gemtuzumab ozogamicin antibody analyses have been described
elsewhere.15

Efficacy Assessments

The primary efficacy end point in this study was the rate of complete remission (CR). Patients
were assessed monthly for remission status. A patient had to meet the following criteria to be
classified as having CR: (1) leukemic blasts absent from peripheral blood; (2) percentage of
blasts in the bone marrow ≤5% as measured by morphologic studies, either bone marrow aspirate
or biopsy; (3) peripheral blood counts with hemoglobin ≥9 g/dL, absolute neutrophil count
≥1500/µL, and platelets ≥100,000/µL; and (4) red blood cell transfusion independence for 2
weeks and platelet transfusion independence for at least 1 week.

In adult phase II trials with gemtuzumab ozogamicin, a portion of patients met all the criteria for
CR with the exception of full recovery of platelet counts (CRp) before additional therapy was
received. CRp is defined in the same way as CR except that the platelet count is not specified,
although platelet transfusion independence for at least 1 week was required. The rate of CRp was
included as a secondary efficacy end point. The overall remission rate was the sum of the CR and CRp rates.

Patients were considered to have no remission (NR) if they did not meet all the criteria for CR or CRp. The NR category included patients who had leukemic blasts in the peripheral blood or whose percentage of blasts in the bone marrow was >5%. Patients with NR also included those who met the bone marrow criteria for remission but did not meet the criteria for peripheral count recovery or were not transfusion independent.

**Drug Efflux Studies**

The analysis of drug efflux based on adenosine triphosphate (ATP)–dependent multidrug resistance (MDR) transport proteins was determined using the fluorescent dye DiOC2 as a surrogate substrate, as previously described. Multiparameter flow cytometry was used to assess the residual DiOC2 in cells treated with or without cyclosporin A ([CSA]; Novartis Pharmaceuticals, East Hanover, NJ). CSA was used at concentrations that inhibit MDR protein–mediated dye efflux. Efflux ratios in the presence or absence of CSA were then calculated as the ratio of loading fluorescence intensity and fluorescent intensity following incubation. Statistical significance was calculated using the Wilcoxon rank sum test for the difference between the mean efflux ratios of responders and nonresponders. Previous analysis of MDR and response to gemtuzumab ozogamicin therapy in adult patients indicated that the median dye efflux ratio in patients failing to achieve CR or CRp was significantly greater than the median value (of 1.2) observed in CR or CRp patients. In the present study, the significance of a 1.2 cutoff value for efflux ratio was assessed with the Fisher exact test.
RESULTS

Patient Characteristics

A total of 29 children with AML that was either refractory to initial induction therapy or in untreated first relapse were enrolled in this phase I study. The median age of the patients was 12 years (range, 1–16 years). Fifteen patients (52%) were boys, and 23 patients (79%) were white. Of the patients who were entered in the protocol and treated, 10 (35%) were refractory to prior therapy and 19 (66%) suffered a relapse following a prior remission. Median first remission before relapse was 144 days (4.7 mo; range, 1.1–27.8 mo). At study entry, most patients (26/29) had ≥20% blasts as determined by histopathology or flow cytometry. Three patients with blasts between 13% and 19% were also enrolled.

Dosing

All 29 patients received a first dose of gemtuzumab ozogamicin (14 at 6 mg/m², 2 at 7.5 mg/m², 13 at 9 mg/m²), and 23 patients were administered a second dose (12 at 6 mg/m², 1 at 7.5 mg/m², and 10 at 9 mg/m²). Six patients did not receive a second dose of gemtuzumab ozogamicin because of refractory leukemia (n=2); adverse event (n=1); death from disease progression 7 days after first gemtuzumab ozogamicin dose (n=1); fever, neutropenia, and disease progression (n=1); or infusion-related nausea and vomiting (n=1). The second dose was administered a median of 14 days following the first dose (range, 14–26 d; mean, 16 d).

Initially, a single patient received 1 and 6 patients received 2 doses at the 6-mg/m² dose with no DLT observed; therefore, the dose was escalated to 9 mg/m² for 2 doses (n=13). Three patients had grade 3/4 elevations of aspartate aminotransferase (AST)/alanine aminotransferase (ALT). One patient was treated at this dose and developed VOD; therefore, the dose in the next 7
patients was de-escalated to 6 mg/m², again without DLT. Subsequently, the dose in 2 patients was escalated to 7.5 mg/m², without DLT, before the study closed.

**Safety Results**

Infusions were associated with low-grade chills (55%), vomiting (41%), fever (35%), nausea (28%), tachycardia (14%), headache (10%), body pain (10%), sweating (7%), hypotension (7%), neck pain (7%), and hypertension (7%). Eight patients developed 12 episodes of infusion-related grade 3 or 4 adverse events (Table 1). These were generally of short duration and tolerable.

There were no instances of anaphylaxis, anaphylactoid reactions, or delayed hypotension.

Almost all patients had transient and reversible elevations in liver function test results (AST, ALT, and total bilirubin). Grade 3 or 4 elevations were observed in 8 patients (28%) (Table 1). The most common grade 3 or 4 treatment-emergent adverse events were leukopenia (48%), thrombocytopenia (35%), sepsis (24%), fever (24%), hypokalemia (21%), hypochromic anemia (17%), pleural effusion (17%), and pneumonia (17%).

Veno-occlusive disease was assessed separately according to the criteria of McDonald et al. and occurred in 7 patients. One patient received the first dose of gemtuzumab ozogamicin and 3 days later developed signs and symptoms of VOD that resolved in 13 days. This patient subsequently received allogeneic HSCT without VOD. Fifteen patients received HSCT following gemtuzumab ozogamicin, with 6 patients developing moderate to severe VOD (Table 2). The mean time from the first dose was 102 days, and 12 of the 15 patients had a second dose a mean of 98 days before HSCT (range, 35–410 days). The median time between the first and second dose was 14 days (range, 14–32 days). Patients who developed VOD following HSCT had their transplant <107 days (3.5 months) from the last dose of gemtuzumab ozogamicin (Table 2). Only 1 patient (3%, 1/29) was diagnosed with VOD without undergoing HSCT. VOD
occurred 3 days after the first dose of gemtuzumab ozogamicin. The patient did not receive a
second dose of gemtuzumab ozogamicin and died approximately 9 months later of refractory
leukemia.

Deaths After Gemtuzumab Ozogamicin Treatment

The causes of death are listed in Table 3. Most patients died from progressive disease. Three
patients died within 28 days of the last dose of gemtuzumab ozogamicin. All were reported to be
refractory to gemtuzumab ozogamicin and died from progressive disease. All other patients died
>28 days after the last dose of gemtuzumab ozogamicin (n=24) or are still living (n=2; 1 CR and
1 CRp, are alive and in remission at 645+ and 112+ days, respectively).

Survival Characteristics of Patients Treated With Gemtuzumab Ozogamicin

Survival of these patients with initially refractory and relapsed AML was typically short. Those
patients who received HSCT had the longest survival. However, HSCT was also associated with
toxicity and early death in some patients (Table 3).

Immunogenicity of Gemtuzumab Ozogamicin in Treated Patients

None of the patients developed detectable antibody responses against the hP67.6 monoclonal
antibody, calicheamicin, or the linker on day 8 or day 22 after each dose.

Efficacy Results

Four patients experienced CR, and 4 experienced CRp, for 8 (28%) overall remissions following
gemtuzumab ozogamicin therapy (Table 3). Of the 8 remissions, 5 (26%) were from the initial
relapse group, and 3 (30%) were from the initial refractory group.

Drug Efflux Results
Previous studies have demonstrated that the MDR phenotype, defined as the ability of leukemic cells to show increased efflux of multiple drug types in association with elevated expression of ATP-dependent drug transporters (such as P-glycoprotein and MDR-related proteins), correlates with resistance to gemtuzumab ozogamicin.\textsuperscript{22} In these studies, a median dye efflux ratio of >1.2 was observed in patients failing to achieve CR or CRp. To assess the applicability of this observation from patient samples in our phase I trial, we determined the relative level of drug efflux with and without the inhibitor of MDR-mediated efflux, CSA. Data available from the samples of 22 patients indicated a significant difference in the mean efflux ratio of responders versus nonresponders ($P=0.005$, Wilcoxon rank sum test). The groups also appeared to be separable by using an efflux ratio cutoff value of 1.2: 5 out of 8 patients with an efflux ratio less than 1.2 had a CR or CRp, whereas none of the 14 patients with a ratio $\geq$1.2 achieved remission ($P=0.002$, Fisher exact test) (Figure 1).

**DISCUSSION**

Limited data have been published on the use of gemtuzumab ozogamicin for the treatment of AML in younger patients. Because children were not enrolled in the pivotal phase II clinical trials of gemtuzumab ozogamicin that led to its US marketing approval,\textsuperscript{14} this dose-escalating study was conducted to evaluate the efficacy and safety of gemtuzumab ozogamicin in patients younger than 18 years of age with refractory and relapsed AML.\textsuperscript{17}

Dose-limiting toxicity was not seen at a dose of 6 mg/m$^2$. Although some reversible hepatic transaminase and bilirubin elevations occurred, these were not dose limiting. At 9 mg/m$^2$, a patient developed VOD considered to be related to gemtuzumab ozogamicin and thus constituted a DLT. The MTD was determined to be 6 mg/m$^2$; however, 2 patients tolerated the 7.5-mg/m$^2$ dose, 1 achieving CRp. Before more patients could be escalated to this dose, the FDA
approved gemtuzumab ozogamicin, additional enrollment was halted, and the study closed. Of interest, responses were observed in all dose categories. The MTD of gemtuzumab ozogamicin for adults is 9 mg/m², and because the pharmacokinetic data for adults is comparable to children for this drug,²⁰ it is possible that the MTD could be higher than 6 mg/m² for children.

The safety and tolerability profile of gemtuzumab ozogamicin presented here appears to be similar to that described in adults.¹⁵,¹⁶ Most of the deaths that occurred during this pediatric study were attributable to disease progression or complications associated with HSCT. Gemtuzumab ozogamicin was generally well tolerated by younger patients. Grade 3 or 4 infusion-related adverse events and grade 3 or 4 elevations in liver function tests were each reported in 8 patients. In previous studies of gemtuzumab ozogamicin, hyperbilirubinemia and elevated hepatic transaminase levels also were common nonhematologic adverse events.¹⁵,¹⁶

The overall incidence of VOD in the current study was 24% (n=7). In the analysis of 277 adult patients with AML in first relapse from 3 pivotal phase II trials of gemtuzumab ozogamicin,²³ the rate of VOD when the drug was administered without prior or subsequent HSCT was <1%. The incidence of VOD increased to 17% when gemtuzumab ozogamicin was given either before or after HSCT.²⁴ It is important to note that 6 of the 7 patients who developed VOD in the present study did so after undergoing HSCT.

Depending on prior therapy and the conditioning regimen received, approximately 15% of transplant patients develop VOD. Two studies have suggested an elevated risk of VOD among patients undergoing transplant within a short time interval after gemtuzumab ozogamicin exposure.²⁴,²⁵ All 6 children who developed VOD after transplant in the present study were exposed to gemtuzumab ozogamicin within 3.5 to 4 months of their transplant. Neither of the 2 patients who underwent transplant more than 4 months from gemtuzumab ozogamicin exposure...
developed VOD. These findings are consistent with those of Erba and colleagues\textsuperscript{24} and Wadleigh and colleagues.\textsuperscript{25} These studies, when taken together with the data from the present trial, suggest that the time from treatment with gemtuzumab ozogamicin to transplant for relapsed or refractory AML appears to be important with respect to the development of posttransplant VOD, regardless of age. Although the current study was not designed to assess the relationship between gemtuzumab ozogamicin, time to HSCT, and VOD, these results suggest such a relationship. At the doses used in this phase I trial, gemtuzumab ozogamicin as a single agent may increase the risk of VOD when bone marrow transplantation occurs in less than 3.5 months from the time of the last dose of the antibody-conjugate. However, the data presented in this report are not necessarily relevant to patients with relapsed or newly diagnosed AML receiving gemtuzumab ozogamicin at lower doses in combination with chemotherapy, such as is being tested in several ongoing studies.

More than one third (35\%) of the patients in this study entered the trial with AML that was refractory to primary therapy, and the median duration of first complete remission for the remaining patients was only 4.7 months. Yet, the response of these patients was 28\% overall. Importantly, patients with refractory disease responded (30\%) as well as patients in first relapse (26\%), suggesting that gemtuzumab ozogamicin has comparable efficacy in these 2 high-risk but distinct patient populations.

The analysis of drug efflux phenotype and response to gemtuzumab ozogamicin is a possible surrogate marker of response, with no remissions observed in patients whose leukemic blasts showed relatively elevated levels of drug efflux. These data are consistent with a previous report correlating clinical response to gemtuzumab ozogamicin with low drug efflux blast phenotype.\textsuperscript{22} The data we present in our study extends these observations to pediatric patients within this
clinical trial. Additional studies are warranted to determine whether the phenotype of low versus high levels of drug efflux helps to define a priori which patients are more or less likely to respond to gemtuzumab ozogamicin. Similar studies with combinations of chemotherapy plus gemtuzumab ozogamicin should be particularly interesting to examine using drug efflux as a predictive surrogate of response. It will also be important to assess whether gemtuzumab ozogamicin is particularly useful in pediatric patients with newly diagnosed AML, given the low level of MDR1 expression in this patient population.²⁶,²⁷

The remission rate observed in our study compares favorably with that observed in adult patients with relapsed AML treated with gemtuzumab ozogamicin reported by Sievers and colleagues (30%)¹⁵ and Larson and colleagues (28%).¹⁶ Additionally, these results are similar to those of 2 published reports in which gemtuzumab ozogamicin was used to treat children with refractory/relapsed AML on a compassionate-use basis.¹⁸,¹⁹ As reported by Zwaan and colleagues,¹⁸ 15 children (4 de novo, 11 relapsed/refractory) were administered gemtuzumab ozogamicin in the dose range of 4 to 9 mg/m² per course. Clinical response included 3 patients achieving CR, 5 patients achieving CRp, and 3 patients with no change in bone marrow blast count. Although survival rates were low, many of these patients had relapsed at least once, if not multiple times, and were resistant to multiple chemotherapeutic agents and combinations. Results reported by Reinhardt and colleagues¹⁹ in 12 relapsed/refractory patients receiving gemtuzumab ozogamicin doses of 1.8 to 9.0 mg/m² per course did not reveal any CRs. In both compassionate-use reports, 1 patient in each report experienced VOD. The results cannot be used to predict efficacy or survival in other patients, such as nonrelapsed patients or those patients treated with combination therapy.
The targeting of myeloid cells by gemtuzumab ozogamicin and our demonstration of comparable remission rates in refractory and relapsed patients suggests that this agent should be further tested in combination with standard chemotherapy in an attempt to increase the initial remission rate of de novo pediatric patients with AML and potentially increase relapse-free survival. The MTD outlined in this trial may not be the appropriate dose in combination therapy. In adults with AML, De Angelo and colleagues\textsuperscript{28} reduced the dose of gemtuzumab ozogamicin by 30% when the drug was used with combination chemotherapy. Thus, a dose-escalation combination chemotherapy clinical trial would be suggested as a next step.

This clinical dose-escalation trial demonstrated that single-agent gemtuzumab ozogamicin can be used with acceptable safety and comparable efficacy at 6 mg/m\textsuperscript{2} in pediatric patients with relapsed and refractory AML. Once these patients enter remission, subsequent therapy is the next issue for physicians to determine. If an appropriate donor has been identified, HSCT is an important consideration in this pediatric population, and our data demonstrate that gemtuzumab ozogamicin can allow patients to achieve sufficient disease control to undergo HSCT. Safely balancing an increased risk of VOD associated with swift application of transplant against the risk of recurrent leukemia that often occurs during the interval between remission induction and transplant remains a complex challenge for future studies. Recent data suggest that the occurrence of VOD may be reduced if the experimental medication defibrotide is used prophylactically.\textsuperscript{29} The timing of transplant following exposure to gemtuzumab ozogamicin and the potential role of prophylaxis for VOD need to be studied further, especially when gemtuzumab ozogamicin is used at reduced doses in combination with chemotherapy.
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P-glycoprotein (P-170) as detected by monoclonal antibody MRK-16 in pediatric acute myeloid


Table 1. Grades 3 and 4 Infusion-Related Adverse Events and Liver Function Test Abnormalities by Treatment Dose

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<td>0</td>
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<td>3</td>
</tr>
<tr>
<td>AST</td>
<td>1</td>
<td>0</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Total bilirubin</td>
<td>1</td>
<td>0</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Total</td>
<td>4</td>
<td>0</td>
<td>4</td>
<td>8</td>
</tr>
</tbody>
</table>

ALT=alanine aminotransferase; AST=aspartate aminotransferase.

Table 2. Timing of VOD After HSCT (A) and HSCT After Treatment Dose (B)
### A

<table>
<thead>
<tr>
<th>Patient</th>
<th>Dose, mg/m²</th>
<th>Time From Last Dose to HSCT, days</th>
<th>Time From HSCT to VOD, days</th>
<th>Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>9</td>
<td>20</td>
<td>1</td>
<td>Died 1 mo after HSCT; disease progression</td>
</tr>
<tr>
<td>2</td>
<td>6</td>
<td>48</td>
<td>10</td>
<td>Died 3 mo after HSCT; sepsis</td>
</tr>
<tr>
<td>3</td>
<td>6</td>
<td>72</td>
<td>9</td>
<td>Died 1 y after HSCT; disease progression</td>
</tr>
<tr>
<td>4</td>
<td>6</td>
<td>45</td>
<td>8</td>
<td>Died 7.5 mo after HSCT; disease progression</td>
</tr>
<tr>
<td>5</td>
<td>6</td>
<td>106</td>
<td>6</td>
<td>Died 1 mo after HSCT; disease progression</td>
</tr>
<tr>
<td>6</td>
<td>6</td>
<td>84</td>
<td>22</td>
<td>Died 1.5 mo after HSCT; sepsis</td>
</tr>
</tbody>
</table>

### B

<table>
<thead>
<tr>
<th>Patients With HSCT</th>
<th>Time From GO Exposure to HSCT, days First Dose</th>
<th>Second Dose</th>
<th>VOD</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>110</td>
<td>84</td>
<td>√</td>
</tr>
<tr>
<td>2</td>
<td>140</td>
<td>121</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>50</td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>50</td>
<td>35</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>71</td>
<td>55</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>95</td>
<td>*</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>64</td>
<td>48</td>
<td>√</td>
</tr>
<tr>
<td>8</td>
<td>424</td>
<td>410</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>86</td>
<td>72</td>
<td>√</td>
</tr>
<tr>
<td>10</td>
<td>120</td>
<td>106</td>
<td>√</td>
</tr>
<tr>
<td>11</td>
<td>67</td>
<td>53</td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>59</td>
<td>45</td>
<td>√</td>
</tr>
<tr>
<td>13</td>
<td>118</td>
<td>104</td>
<td></td>
</tr>
<tr>
<td>14</td>
<td>56</td>
<td>42</td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>20</td>
<td>*</td>
<td>√</td>
</tr>
</tbody>
</table>

GO=gemtuzumab ozogamicin; HSCT=hematopoietic stem cell transplantation; VOD=veno-occlusive disease.

*Did not receive a second GO dose.

√ Presence of VOD.
**Table 3. Causes of Death by Treatment Dose**

<table>
<thead>
<tr>
<th>Outcome</th>
<th>Time From HSCT, days</th>
<th>6 mg/m² (n=14)</th>
<th>7.5 mg/m² (n=2)</th>
<th>9 mg/m² (n=13)</th>
<th>Total (n=29)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Died</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Progressive disease</td>
<td>10 (1 CR; 9 NR)</td>
<td>1 (NR)</td>
<td>11 (2 CR; 1 CRp; 8 NR)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>1</td>
<td>4</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>0</td>
<td>7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pneumonia</td>
<td>52</td>
<td>1 (NR)</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Multiorgan failure</td>
<td>77</td>
<td>1 (NR)</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Sepsis, acute renal failure</td>
<td>53</td>
<td>1 (NR)</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Respiratory failure</td>
<td>42</td>
<td>0</td>
<td>0</td>
<td>1 (CRp)</td>
<td></td>
</tr>
<tr>
<td>Graft loss plus fungal sepsis</td>
<td>60</td>
<td>0</td>
<td>0</td>
<td>1 (CRp)</td>
<td></td>
</tr>
<tr>
<td>Alive</td>
<td>1 (CR, 645+ d)*</td>
<td>1 (CRp, 112+ d)†</td>
<td>0</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>1</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>0</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>14</td>
<td>2</td>
<td>13</td>
<td>29</td>
<td></td>
</tr>
</tbody>
</table>

CR=complete remission; CRp=complete remission with incomplete platelet recovery;

HSCT=hematopoietic stem cell transplantation; NR=no remission.

*Last follow-up date March 15, 2002.

†Last follow-up date February 1, 2000.
Figure Legends

Figure 1. The level of drug efflux and response to single-agent gemtuzumab ozogamicin in pediatric patients with AML. This figure demonstrates the response (defined as complete remission [CR] or complete remission with incomplete platelet recovery [CRp]) for patients whose leukemic blasts showed an efflux ratio <1.2 versus a ratio ≥1.2 ($P=0.005$). The ratio of drug efflux is defined in the Methods. AML=acute myeloid leukemia; NR=no remission.
Figure 1
Safety and efficacy of gemtuzumab ozogamicin (Mylotarg®) in pediatric patients with advanced CD33-positive acute myeloid leukemia


Bernstein and Eric L Sievers