Phase II trial of vorinostat and gemtuzumab ozogamicin as induction and post-remission therapy in older adults with previously untreated acute myeloid leukemia

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ABSTRACT

Histone deacetylase inhibitors such as vorinostat enhance gemtuzumab ozogamicin efficacy *in vitro*. We, therefore, investigated vorinostat+gemtuzumab ozogamicin for adults aged 60 years and over with untreated acute myeloid leukemia. We stratified patients into 2 groups (group 1: patients aged ≥70 years and performance status 2-3; group 2: aged 60-69 years with performance status 0-3 or aged ≥70 years and performance status 0-1). Responses were monitored separately in group 2 patients with normal or favorable cytogenetics (group 2A) and other cytogenetics (group 2B). Among 31 patients, 6 (19.4%) achieved complete remission, and one (3.2%) achieved complete remission with incomplete platelet recovery; these patients had a higher median overall survival than non-responders (553 *vs.* 131 days, *P*=0.0026). Response rates were: group 1, one of 10 (10.0%); group 2A, 6 of 13 (46.2%); and group 2B, none of 8 (0%). These data indicate that vorinostat+gemtuzumab ozogamicin has activity that is mostly confined to patients with normal karyotype disease. *ClinicalTrial.gov: NCT00673153.*

Key words: acute myeloid leukemia, vorinostat, gemtuzumab ozogamicin, induction, post-remission, older adults.

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Introduction

Age remains an important risk factor for poor outcome in acute myeloid leukemia (AML).[1-3] Therefore, novel strategies are needed for older adults, especially for those with poor performance status (PS) who are at high risk of life-threatening or fatal toxicities from intensive therapy. This need for new therapies is reflected by the general recommendation that all older patients with AML should receive investigational treatments whenever possible; this call is supported by current guidelines, e.g. by the National Comprehensive Cancer Network (NCCN).[4] One approach uses monoclonal antibodies, most notably with gemtuzumab ozogamicin (GO), an immunoconjugate between an anti-CD33 antibody (hP67.6) and a toxic calicheamicin-γ_1 derivative.[5] However, when used alone for adults with untreated or relapsed AML, rates of complete remission (CR) and CR with incomplete platelet recovery (CRp) have typically not exceeded 25-35%.[6-8] Research has, therefore, focused on developing means to improve GO monotherapy.

hP67.6 facilitates cellular uptake of the calicheamicin-γ_1 moiety which, after cleavage from the antibody, causes DNA damage, apoptosis, and cell death.[5] This putative mechanism suggests that drugs that augment the DNA damage response and lower the apoptotic threshold could improve GO efficacy. Histone deacetylase inhibitors (HDACis) are of interest in this regard. There is increasing evidence to suggest that HDACis, such as vorinostat, function synergistically or additively with conventional anti-AML chemotherapeutics, partly due to lowering the apoptotic threshold of tumor cells.[9] Furthermore, recent *in vitro* studies indicate that HDACis strongly increase DNA intercalation of the calicheamicin-γ_1 derivative and enhance DNA degradation and GO efficacy.[10] These findings prompted a phase II study of vorinostat as chemosensitizer with GO in patients aged 60 years and over with untreated AML.

Design and Methods

Study population

Patients were eligible if they had untreated primary or secondary AML (other than acute promyelocytic leukemia) according to the 2008 World Health Organization classification.[11] Other inclusion criteria included: an Eastern Cooperative Oncology Group (ECOG) PS of 0-3; a white blood cell (WBC) count <10×10^9/L; total bilirubin ≤2.5 x Upper

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Limit of Normal (ULN) unless elevation was due to hepatic infiltration by AML, Gilbert's syndrome, or hemolysis; SGOT/SPGT ≤1.5 x ULN unless elevation was due to hepatic infiltration by AML; serum creatinine ≤1.5 x ULN; and left ventricular ejection fraction of 40% or over. Exclusion criteria were: diagnosis of another malignancy (unless disease-free for >6 months); prior AML-like systemic therapy or use of GO or HDACi; central nervous system AML; positive HIV test; or uncontrolled systemic infection. Cytogenetic risk-group assignment was carried out according to the Southwest Oncology Group (SWOG)/ECOG criteria.[12] Treatment responses were defined according to standard criteria.[3,13] The study was approved by the institutional review board of all participating institutions (*Fred Hutchinson Cancer Research Center/University of Washington/Seattle Cancer Care Alliance, Stanford University, Wake Forest University,* and *Barbara Ann Karmanos Cancer Center;* trial registered at *ClinicalTrials.gov as NCT00673153*), and patients gave their informed consent in accordance with the Declaration of Helsinki.

Treatment plan

Patients received vorinostat 400 mg orally once daily on Days 1-9 and GO 3 mg/m^2 on Day 8; hydroxyurea was given to reduce the WBC to less than 10×10^9/L before beginning vorinostat. Therapy was repeated for residual disease on Day 14; the protocol was amended after 8 enrolled patients to allow a third induction course before response assessment. Patients who achieved either CR or CRp were eligible for one cycle of consolidation treatment with vorinostat and GO at the same doses. Patients could then proceed with maintenance therapy with vorinostat only (400 mg once daily on Days 1-14 every four weeks for up to 4 cycles) if CR/CRp was maintained. Patients entering a CR/CRp could also receive more intensive consolidation therapy including hematopoietic cell transplantation (HCT).

Statistical analyses

Our main objective was to test whether preliminary efficacy data suggested that further clinical testing of vorinostat and GO in a randomized fashion with 'standard' chemotherapy (e.g. '3+7') was warranted. Given the influence of age and PS on treatment outcome,[2] patients were stratified into two groups: group 1, patients aged ≥70 years with ECOG PS 2-3; and group 2, aged 60-69 years with PS 0-3 or aged ≥70 years and PS 0-1. For group 1, the primary outcome of interest was 30-day survival, using a historical rate of 50% for similar patients undergoing standard induction therapy.[2] For group 2, we focused on the CR/CRp rate. Given the influence of cytogenetics on remission rates,[3] the statistical design monitored CR/CRp rate separately in group 2 patients with normal or favorable cytogenetics (group 2A) and other cytogenetics (group 2B) while allowing results from group 2A to affect stopping in group 2B and vice versa. CR/CRp rates of interest were based on those found in the literature. In group 2A, stopping would occur if the probability was over 97% that the true CR/CRp rate was below 45%, while in group 2B, the criterion probability was 98% and the reference CR/CRp rate was less than 30%. These cut offs were chosen to allow a maximum 10% probability of early stopping if the true CR/CRp rates were at least 45 or 30% in groups 2A or 2B, respectively. A planned interim analysis was performed after enrollment of 15 patients into group 2. Follow-up data are current as of August 16, 2011.

Results and Discussion

Between September 2008 and March 2010, 31 adults with a median age of 74 years were enrolled (Table 1). Based on their age, PS, and cytogenetic risk, 10 patients were assigned to group 1 (including one patient with favorable cytogenetics), 13 to group 2A, and 8 to group 2B.

All patients received study therapy; 30 patients completed at least one course of induction therapy, 29 received at least 2 courses, and 9 received 3 courses. Six of the 31 patients (19.4%, exact 95% confidence interval [CI], 7.5-37.5%) achieved CR, and one patient (3.2%) achieved CRp, for a CR/CRp rate of 22.6% (9.6-41.1%; Table 2). The 3 patients with known NPM1-positive/FLT3-ITD-negative normal karyotype AML achieved CR/CRp; this observation is consistent with a previous report suggesting that patients with NPM1-positive/FLT3-ITD-negative normal karyotype AML may benefit from GO-containing therapy.[14] All patients who achieved CR/CRp received additional consolidation chemotherapy, including vorinostat alone (n=1), non-myeloablative allogeneic HCT (n=3), and/or cytarabine-based chemotherapy (n=4). Three patients relapsed after 82, 291 and 294 days; 4 patients are in ongoing remission after 455, 496, 956 and 988 days. Median overall survival for patients achieving CR/CRp was significantly longer than for those who failed therapy but lived at least 30 days after treatment initiation (553 [range 291-1035]) *vs.* 131 [34-998] days, log rank *P*=0.0026; Figure 1). Three patients (9.7%) died within 30 days after initiating study therapy: 2 patients in group 1 died from progressive disease and one patient in group 2B died from sepsis. Two of 3 patients died before initial treatment assessment ('death from indeterminate cause'). Treatment was otherwise well tolerated with grade 3-4 bleeding (n=4), dyspnea (n=3) and acute renal failure (n=2) as the most common toxicities besides cytopenias and neu-

Table 1. Characteristics of study cohort.

Parameter	n=31
Median age (range), years	73.8 (61.1-80.7)
Sex (male/female), n.	20/11
Performance status, n. (%)	
0	4 (12.9%)
1	13 (41.9%)
2	11 (35.5%)
3	3 (9.7%)
Cytogenetic risk group, n. (%)	
Favorable	1 (3.2%)
Intermediate	18 (58.1%)
Unfavorable	12 (38.7%)
Secondary AML, n. (%)	19 (61.3%)
Laboratory findings at baseline, median (range)	
WBC (×10^3/μL)*	3.3 (0.7-10.4)
Hemoglobin (g/dL)	9.5 (7.4-14.9)
Platelets (×10^3/μL)	48 (15-700)
LDH (U/L)	231 (141-1405)
Creatinine (mg/dL)	0.9 (0.6-1.7)
Total bilirubin (mg/dL)	0.7 (0.4-3.3)
SGOT (U/L)	21 (10-69)
SGPT (U/L)	26 (10-116)
Cohort assignment, n. (%)	
Group 1	10 (32.3%)
Group 2A	13 (41.9%)
Group 2B	8 (25.8%)

**11 patients received hydroxyurea prior to study therapy initiation.*

Table 2. Overall treatment response.

Parameter	Group 1 (n=10)	Group 2A (n=13)	Group 2B (n=8)	All patients (n=31)
Best response after induction therapy, n. (%)				
CR	1 (10.0%)	5 (38.5%)	0 (0.0%)	6 (19.4%)
CRp	0 (0.0%)	1 (7.7%)	0 (0.0%)	1 (3.2%)
CR + CRp	1 (10.0%)	6 (46.2%)	0 (0.0%)	7 (22.6%)
No response	8 (80.0%)	7 (53.9%)	7 (87.5%)	22 (71.0%)
Death from indeterminate cause	1 (10%)	0 (0%)	1 (12.5%)	2 (6.5%)
Early death*	2 (20.0%)	0 (0.0%)	1 (12.5%)	3 (9.7%)
N. of induction cycles to CR/CRp, n. (%)				
2	1	4	0	5 (71.4%)
3	0	2	0	2 (28.6%)

Death within 30 days of initiation of study therapy.

tropenic fever and/or infections (*Online Supplementary Table S1*).

Importantly, our data indicate that the likelihood of achieving CR/CRp depended on age/PS and cytogenetics. Specifically, CR/CRp rates were one of 10 (10.0%) in group 1, 6 of 13 (46.2%) in group 2A (2 patients negative for minimal residual disease by flow cytometry), and none of 8 (0%) in group 2B. This low CR/CRp rate led to discontinuation of accrual in group 2B after a planned first interim analysis. The stopping rule was not met in group 2A, but accrual was stopped after the availability of GO in the US was curtailed.[15] Two of 10 (exact 95% CI, 2.5-55.6%) group 1 patients died within the first 30 days of progressive disease, a finding that would not have led to termination of accrual were it not for the general unavailability of GO.

As this was a single-arm clinical study, the relative contribution(s) of vorinostat and GO to the clinical efficacy is unknown. The currently available clinical data with vorinostat suggest that its activity as single agent is relatively limited.[16] On the other hand, GO monotherapy has shown efficacy in older adults with newly diagnosed AML in several phase II studies, with overall response rates ranging from 8-27%,[17-19] although post hoc stratified analyses in one study indicated better response rates for patients aged 61-75 years than those aged over 75.[18] Relative to our study, however, higher doses of GO were used in the previous studies and data were not stratified prospectively based on cytogenetic risk, limiting the ability to directly compare trial results. Only a better-controlled trial would ultimately be able to determine whether vorinostat indeed augments GO efficacy, as preclinical data suggest.

A particular characteristic of our trial is the statistical design in group 2 which monitored drug efficacy in cohorts with different cytogenetic risk separately, thereby addressing the patient heterogeneity that poses a major challenge in phase II drug development.[20] At the time of trial development, there was little information on the relationship between cytogenetic/molecular abnormalities and likelihood of response to either vorinostat or GO; therefore, our trial design was loosely based on risk stratification schemes that evolved from experience with 'standard' AML therapeutics. However, recent results from the Medical Research Council (MRC) AML15 trial on 1,113 predominantly younger adults with newly diagnosed AML, randomized to receive a single dose of GO (3 mg/m^2) during induction chemotherapy, indicate that the benefit of addition of GO is most prominent for patients with favorable-risk disease, with some benefit extending to subgroups of patients with intermediate-risk disease but no benefit for those with poor-risk disease.[21] These data suggest that the response to GO may be similar to that of conventional AML chemotherapy, providing some basis for the chosen cytogenetic risk stratification in our study. Although direct comparability is limited, our study agrees with data from the MRC AML15, indicating only very limited efficacy of GO in worse-prognosis patients.

While it is well recognized that therapeutic response, likelihood of disease relapse, and chance of cure differs widely between individual patients,[3] most phase II trials combine all patients in one group, thus ignoring heterogeneity. Consequently, a new drug might be called 'promising' ('not promising') purely because of a disproportionate number of favorable ('unfavorable') patients given the drug. The small number of patients treated in many phase II trials increases the probability that this situation will arise. Accounting for patient heterogeneity should increase the likelihood of drawing correct conclusions in phase II trials. The simplest method to account for heterogeneity is to conduct distinct trials in various prog-

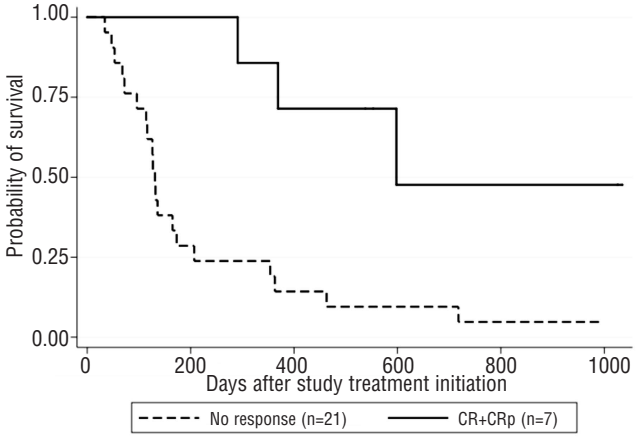

Figure 1. Kaplan-Meier estimates of the probability of survival for responders (i.e. patients who achieved CR or CRp, n=7) and non-responders (n=21); patients who died within the first 30 days of treatment initiation (n=3) were excluded from this analysis. Patients who achieved CR/CRp after induction therapy with vorinostat and GO had a statistically significantly better overall survival than those who failed therapy but lived at least 30 days after treatment initiation (log rank *P*=0.0026).

nostic groups. Although this is preferable to averaging and considering such patients as one group, separate trials increase sample size and study duration. Another method computes observed/expected success ratios with the expected rates accounting for heterogeneity. However, these methods do not formally allow results from one subgroup to adaptively influence trial conduct (e.g. stopping or continuing) in another group. This is particularly problematic when treatment-subgroup interactions exist, i.e. when a treatment has different effects in different prognostic groups. Wathan *et al.* have proposed a hierarchical Bayesian design to address this problem.[22] As with separate trials, stopping rules are subgroup specific. However, in contrast to separate trials, the design examines accumulating data to see whether a given treatment might have similar effects in different prognostic groups and allows data from two groups to be combined to the extent that such 'borrowing of strength' is justified by these data. Although the design is computationally complex, advances in computing algorithms and in computing power will facilitate its use. Our trial shows that this approach can identify subgroups of patients that benefit from drug treatment while preventing unnecessary toxicity in others that are unlikely to respond to therapy.

Together, our study indicates that vorinostat and GO has encouraging anti-AML activity with limited toxicity in a well defined subset of older adults with untreated AML, i.e. those with normal karyotype AML (perhaps in particular those with NPM1-positive/FLT3-ITD-negative AML) while it is largely ineffective in others. These data would support further clinical testing of vorinostat and GO, ideally in a randomized design with established therapies (e.g. '3+7') in the subset of older adults with better-prognosis disease, i.e. favorable-risk and normal karyotype AML.

Authorship and Disclosures

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References

1. Howlader N, Noone AM, Krapcho M, Neyman N, Aminou R, Waldron W, et al. SEER Cancer Statistics Review, 1975-2008, National Cancer Institute. Bethesda, MD, http://seer.cancer.gov/csr/1975_2008/.
2. Appelbaum FR, Gundacker H, Head DR, Slovak ML, Willman CL, Godwin JE, et al. Age and acute myeloid leukemia. Blood. 2006;107(9):3481-5.
3. Döhner H, Estey EH, Amadori S, Appelbaum FR, Büchner T, Burnett AK, et al. Diagnosis and management of acute myeloid leukemia in adults: recommendations from an international expert panel, on behalf of the European LeukemiaNet. Blood. 2010;115(3):453-74.
4. NCCN Clinical Practice Guidelines in Oncology (NCCN Guidelines™): Acute myeloid leukemia. Version 2.2011. Available from: www.nccn.org
5. Linenberger ML. CD33-directed therapy with gemtuzumab ozogamicin in acute myeloid leukemia: progress in understanding cytotoxicity and potential mechanisms of drug resistance. Leukemia. 2005;19(2): 176-82.
6. Abutalib SA, Tallman MS. Monoclonal antibodies for the treatment of acute myeloid leukemia. Curr Pharm Biotechnol. 2006;7(5):343-69.
7. Pagano L, Fianchi L, Caira M, Rutella S, Leone G. The role of Gemtuzumab Ozogamicin in the treatment of acute myeloid leukemia patients. Oncogene. 2007;26(25):3679-90.
8. Hütter ML, Schlenk RF. Gemtuzumab ozogamicin in non-acute promyelocytic acute myeloid leukemia. Expert Opin Biol Ther. 2011;11(10):1369-80.
9. Bolden JE, Peart MJ, Johnstone RW. Anticancer activities of histone deacetylase inhibitors. Nat Rev Drug Discov. 2006;5 (9):769-84.
10. ten Cate B, Samplonius DF, Bijma T, de Leij LF, Helfrich W, Bremer E. The histone deacetylase inhibitor valproic acid potently augments gemtuzumab ozogamicin-induced apoptosis in acute myeloid leukemic cells. Leukemia. 2007;21(2):248-52.
11. Vardiman JW, Thiele J, Arber DA, Brunning RD, Borowitz MJ, Porwit A, et al. The 2008 revision of the World Health Organization (WHO) classification of myeloid neoplasms and acute leukemia: rationale and important changes. Blood. 2009;114(5):937-51.
12. Mrózek K, Heerema NA, Bloomfield CD. Cytogenetics in acute leukemia. Blood Rev. 2004;18(2):115-36.
13. Cheson BD, Bennett JM, Kopecky KJ, Büchner T, Willman CL, Estey EH, et al. Revised recommendations of the International Working Group for diagnosis, standardization of response criteria, treatment outcomes, and reporting standards for therapeutic trials in acute myeloid leukemia. J Clin Oncol. 2003;21(24):4642-9.
14. Chevallier P, Prebet T, Pigneux A, Hunault M, Delaunay J, Perry F, et al. Influence of NPM1 and FLT3-ITD status on outcome in relapsed/refractory AML patients receiving salvage therapy including gemtuzumab ozogamicin. Leukemia. 2010;24(2):467-9.
15. Pfizer Inc. 2010. Available from: http://media.pfizer.com/files/products/mylotarg_hcp_letter.pdf
16. Prebet T, Vey N. Vorinostat in acute myeloid leukemia and myelodysplastic syndromes. Expert Opin Investig Drugs. 2011;20(2):287-95.
17. Estey EH, Thall PF, Giles FJ, Wang XM, Cortes JE, Beran M, et al. Gemtuzumab ozogamicin with or without interleukin 11 in patients 65 years of age or older with untreated acute myeloid leukemia and high-risk myelodysplastic syndrome: comparison with idarubicin plus continuous-infusion, high-dose cytosine arabinoside. Blood. 2002;99(12):4343-9.
18. Amadori S, Suciu S, Stasi R, Willemze R, Mandelli F, Selleslag D, et al. Gemtuzumab ozogamicin (Mylotarg) as single-agent treatment for frail patients 61 years of age and older with acute myeloid leukemia: final results of AML-15B, a phase 2 study of the European Organisation for Research and Treatment of Cancer and Gruppo Italiano Malattie Ematologiche dell'Adulto Leukemia Groups. Leukemia. 2005;19(10): 1768-73.
19. Nabhan C, Rundhaugen LM, Riley MB, Rademaker A, Boehlke L, Jatoi M, et al. Phase II pilot trial of gemtuzumab ozogamicin (GO) as first line therapy in acute myeloid leukemia patients age 65 or older. Leuk Res. 2005;29(1):53-7.
20. Walter RB, Appelbaum FR, Tallman MS, Weiss NS, Larson RA, Estey EH. Shortcomings in the clinical evaluation of new drugs: acute myeloid leukemia as paradigm. Blood. 2010;116(14):2420-8.
21. Burnett AK, Hills RK, Milligan D, Kjeldsen L, Kell J, Russell NH, et al. Identification of patients with acute myeloblastic leukemia who benefit from the addition of gemtuzumab ozogamicin: results of the MRC AML15 trial. J Clin Oncol. 2011;29(4):369-77.
22. Wathen JK, Thall PF, Cook JD, Estey EH. Accounting for patient heterogeneity in phase II clinical trials. Stat Med. 2008;27 (15):2802-15.