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Cyclosporine Modulation of Multidrug Resistance in Combination with Pravastatin, Mitoxantrone, and Etoposide for Adult Patients with Relapsed/Refractory Acute Myeloid Leukemia (AML): A Phase 1/2 Study

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It has been repeatedly observed that outcomes in acute myeloid leukemia (AML) worsen if leukemia cells express active ATP-binding cassette drug transporters, notably P-glycoprotein (Pgp) [1,2]. Indeed, the predictive value of Pgp-mediated multidrug resistance (MDR) for induction failure reaches 90% in chemotherapy-naïve adult patients and remains high for those with relapsed disease [3]. Given the high prevalence of MDR and its significance as independent predictor of induction failure, many studies have tested whether MDR reversal agents could increase the efficacy of conventional chemotherapy [2,3]. Positive results from some randomized trials were obtained with first-generation modulators such as cyclosporine A (CSA) [4], but, overall, results have been inconsistent [2,3].

More recently, HMG-CoA reductase inhibitors (e.g. pravastatin) have been investigated as a novel strategy to overcome drug resistance after *in vitro* investigations indicated that cholesterol synthesis inhibition could restore chemosensitivity of AML cells [5,6]. A phase 1 trial evaluated the addition of pravastatin to chemotherapy with idarubicin and high-dose cytarabine (HiDAC) for newly diagnosed and relapsed AML and showed that doses up to 1,680 mg/day for 4 days prior to and during administration of idarubicin/HiDAC were safe and yielded encouraging responses [7]. Pharmacokinetic data demonstrated higher and more sustained serum pravastatin levels with drug doses of at least 1,280 mg/day, suggesting that this or a higher dose should be selected for further study, with a recommended loading time of 4–5 days [7]. As studies further suggested the HMG-CoA inhibitors could downregulate MDR function [8], we attempted to impair drug transporter activity via two independent mechanisms, i.e. CSA and pravastatin, to improve the efficacy of mitoxantrone/etoposide in

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patients with relapsed/persistent AML in a single-arm, open-label phase 1/2 trial (registered at ClinicalTrials.gov as NCT01342887).

The primary clinical objective of this trial was to determine whether any of 4 dose levels of mitoxantrone and etoposide produced acceptable rates of efficacy and toxicity together with CSA and pravastatin. Thus, this trial differed markedly from the standard phase 1 trial that focuses exclusively on toxicity, although patients enter phase 1 trials to achieve a response and not to determine a maximum tolerated dose of a drug regimen [9]. Adult patients with relapsed/refractory AML (excluding those with acute promyelocytic leukemia) and a treatment-related mortality (TRM) score of <9.2 [10] were eligible if they had an Eastern Cooperative Oncology Group (ECOG) performance status (PS) ≤ 3 and adequate organ function. The latter was defined as bilirubin $\leq 2 \times$ Institutional Upper Limit of Normal (IULN), SGOT and SPGT $\leq 2 \times$ IULN, serum creatinine $\leq 1.5 \times$ IULN, and left ventricular ejection fraction $\geq 40\%$. Prior hematopoietic cell transplantation (HCT) was permissible if relapse occurred >180 days post-HCT. Patients were not eligible if they had 1) another malignancy, unless the patient was diagnosed at least 2 years earlier and had been disease-free for at least 6 months following completion of curative-intent therapy; 2) refractory/relapsing blast crisis of CML; 3) HIV infection if CD4 count was <200 cells/ μ L or if there were active AIDS-related complications. Similarly excluded were pregnant or breastfeeding patients, as well as those with uncontrolled systemic infections. Informed consent was obtained from all study participants in accordance with the Declaration of Helsinki, and the institutional review board at Fred Hutchinson Cancer Research Center approved the clinical protocol.

A phase 2 ECOG trial established doses of etoposide ($80 \text{ mg/m}^2/\text{day} \times 5 \text{ days}$), mitoxantrone ($6 \text{ mg/m}^2/\text{day} \times 5 \text{ days}$), and cytarabine ($1 \text{ g/m}^2/\text{day} \times 5 \text{ days}$) that could be given together with high doses of CSA [11]. Assuming that most patients would have failed cytarabine-containing therapies before enrolling on our trial, and anticipating increased toxicity when pravastatin was added to CSA, we avoided the use of cytarabine and started with mitoxantrone/etoposide doses lower than those previously established in combination with CSA [11], attempting to escalate mitoxantrone/etoposide to doses that are typically used in the absence of MDR reversal agents [11,12]. Specifically, all patients received pravastatin 320 mg orally every 6 hours on days 1–10. The starting dose level (level 1) used etoposide $60 \text{ mg/m}^2/\text{day}$ and mitoxantrone $5 \text{ mg/m}^2/\text{day}$ via continuous infusion over 24 hours on days 5–9. CSA started 6 hours before the first doses of mitoxantrone/etoposide; after loading with 6 mg/kg over 2 hours and 4 mg/kg over 6 hours, CSA was infused at 18 mg/kg/day on days 5–9. CSA levels were monitored on Days 6 and 8 to maintain levels $<2,400 \text{ ng/mL}$ (by liquid chromatography-mass spectrometry). At dose level 2, identical doses of pravastatin and CSA were combined with etoposide ($80 \text{ mg/m}^2/\text{day}$) and mitoxantrone ($6 \text{ mg/m}^2/\text{day}$). Two additional treatment levels were planned to escalate etoposide and mitoxantrone doses step-wise to $100 \text{ mg/m}^2/\text{day}$ and $8 \text{ mg/m}^2/\text{day}$, respectively. After a maximum of 2 cycles of induction therapy, patients could receive up to 2 cycles of consolidation therapy if complete remission (CR) or CR with incomplete blood count recovery (CRi) was achieved after induction.

Dose-limiting toxicities (DLTs) were defined as: 1) any Grade 3 non-hematologic toxicity lasting >48 hours except febrile neutropenia (FN), infection, or hyperbilirubinemia; and 2) any Grade 4 non-hematologic toxicity except FN/infection, constitutional symptoms if recovery to Grade 2 within 14 days, and hyperbilirubinemia. We monitored both toxicity and response during the trial with an “adaptive” Bayesian phase 1–2 method [13–15]. For each dose level, we specified prior probabilities of toxicity and response (e.g. for level 1: toxicity=0.2, efficacy=0.25; for level 2: toxicity=0.3, efficacy=0.35). As toxicity and response data were observed from the first patient cohort, Bayes' law was used to update the

prior probabilities thus yielding posterior probabilities of toxicity and response at each dose. The prior probabilities were relatively “non-informative” allowing the posterior probabilities to be heavily influenced by the data from the trial. These probabilities were then referred to pre-set acceptable rates of toxicity and response. In this study, historical data suggested that an acceptable rate of response would be at least 25%; because the 3+3 rule considers a 1/6 but not a 2/6 toxicity rate acceptable, an acceptable toxicity rate would be <25%. If it was highly likely (i.e. probabilities of >0.90) that no dose was associated with acceptable rates of toxicity and response, the trial concluded. If it was highly likely (i.e. probabilities of >0.90) that more than one dose was associated with acceptable rates of toxicity and response, the next cohort of patients was treated at the dose associated with the highest rate of response. The posterior probabilities would become the new prior probabilities, more patients would be treated, and new posterior probabilities generated. The process would be repeated until either it was concluded that no dose was acceptable or until the maximum sample size (n=45) was reached. At the latter time, if there were more than one dose associated with acceptable rates of response and toxicity, the dose associated with the best rate of response would be chosen as the dose to use in future investigations. Adaptive dose determination for cohorts after the first was done using a web-based program (J. Kyle Wathan, PhD; MD Anderson Cancer Center).

Between April 2011 and February 2012, we treated 6 patients on this protocol. In addition, 4 patients received this experimental therapy at dose level 1 off-protocol after thorough discussion with the patient as standard salvage regimens were considered very unlikely to be successful. Three of these off-study patients were treated during the protocol development phase, while 1 patient was ineligible for protocol enrollment due to early post-HCT relapse but wished to be treated according to this regimen; access to these patient data was obtained via an IRB-approved AML database protocol. Thus, overall, 10 patients (median age 52 [range 40–58] years) were treated with this regimen (Table 1). All patients had received at least 2 prior regimens, and 7 received 3 prior therapies. The majority had received standard induction therapy with “7+3”. One patient had undergone HCT. At baseline, the median TRM score was 4.48 among these 10 patients. The average CSA level on Day 6 was 1,366.3 ng/mL (range 713–2,082 ng/mL) and 1,199.8 ng/mL on Day 8 (range 526–1,593 ng/mL).

Among the patients treated on-protocol, 1 out of 3 patients experienced DLTs at dose level 1 (grade 3 mucositis and grade 5 sepsis/multiorgan failure), and 2 out of 3 patients experienced DLTs at dose level 2 (grade 4 acute kidney injury; grade 5 sepsis/multiorgan failure). Two of these 6 patients died within 28 days of initiation of chemotherapy, while none of the off-study patients experienced early death. Seven additional patients had persistent disease. Only one off-study patient achieved CR after 1 induction therapy course; he received 1 cycle of consolidation with pravastatin/etoposide/mitoxantrone but experienced CNS relapse after CR duration of only 46 days. Based on the results of the on-study patients, the statistical monitoring mandated early study closure for lack of efficacy and excessive toxicity, highlighting one of the advantages of combined efficacy/toxicity monitoring during early phase clinical drug testing [13–15].

Together, these data indicate that CSA/pravastatin/mitoxantrone/etoposide is excessively toxic and failed to achieve acceptable efficacy in this population of heavily pretreated AML patients, limiting enthusiasm for further clinical testing of this regimen.

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References

1. van der Kolk DM, de Vries EGE, Müller M, Vellenga E. The role of drug efflux pumps in acute myeloid leukemia. *Leuk Lymphoma*. 2002; 43:685–701. [PubMed: 12153153]
2. Mahadevan D, List AF. Targeting the multidrug resistance-1 transporter in AML: molecular regulation and therapeutic strategies. *Blood*. 2004; 104:1940–1951. [PubMed: 15217827]
3. Sonneveld P, List AF. Chemotherapy resistance in acute myeloid leukaemia. *Best Pract Res Clin Haematol*. 2001; 14:211–233. [PubMed: 11355932]
4. List AF, Kopecky KJ, Willman CL, et al. Benefit of cyclosporine modulation of drug resistance in patients with poor-risk acute myeloid leukemia: a Southwest Oncology Group study. *Blood*. 2001; 98:3212–3220. [PubMed: 11719356]
5. Li HY, Appelbaum FR, Willman CL, Zager RA, Banker DE. Cholesterol-modulating agents kill acute myeloid leukemia cells and sensitize them to therapeutics by blocking adaptive cholesterol responses. *Blood*. 2003; 101:3628–3634. [PubMed: 12506040]
6. Banker DE, Mayer SJ, Li HY, Willman CL, Appelbaum FR, Zager RA. Cholesterol synthesis and import contribute to protective cholesterol increments in acute myeloid leukemia cells. *Blood*. 2004; 104:1816–1824. [PubMed: 15161671]
7. Kornblau SM, Banker DE, Stirewalt D, et al. Blockade of adaptive defensive changes in cholesterol uptake and synthesis in AML by the addition of pravastatin to idarubicin + high-dose Ara-C: a phase 1 study. *Blood*. 2007; 109:2999–3006. [PubMed: 17158228]
8. Connelly-Smith L, Pattinson J, Grundy M, et al. P-glycoprotein is downregulated in KG1a-primitive leukemia cells by LDL cholesterol deprivation and by HMG-CoA reductase inhibitors. *Exp Hematol*. 2007; 35:1793–1800. [PubMed: 17923246]
9. Thall PF, Cook JD. Dose-finding based on efficacy-toxicity trade-offs. *Biometrics*. 2004; 60:684–693. [PubMed: 15339291]
10. Walter RB, Othus M, Borthakur G, et al. Prediction of early death after induction therapy for newly diagnosed acute myeloid leukemia with pretreatment risk scores: a novel paradigm for treatment assignment. *J Clin Oncol*. 2011; 29:4417–4423. [PubMed: 21969499]
11. Tallman MS, Lee S, Sikic BI, et al. Mitoxantrone, etoposide, and cytarabine plus cyclosporine for patients with relapsed or refractory acute myeloid leukemia: an Eastern Cooperative Oncology Group pilot study. *Cancer*. 1999; 85:358–367. [PubMed: 10023703]
12. Greenberg PL, Lee SJ, Advani R, et al. Mitoxantrone, etoposide, and cytarabine with or without valspodar in patients with relapsed or refractory acute myeloid leukemia and high-risk myelodysplastic syndrome: a phase III trial (E2995). *J Clin Oncol*. 2004; 22:1078–1086. [PubMed: 15020609]
13. Thall PF, Estey EH, Sung HG. A new statistical method for dose-finding based on efficacy and toxicity in early phase clinical trials. *Invest New Drugs*. 1999; 17:155–167. [PubMed: 10638486]
14. Thall PF, Cook JD, Estey EH. Adaptive dose selection using efficacy-toxicity trade-offs: illustrations and practical considerations. *J Biopharm Stat*. 2006; 16:623–638. [PubMed: 17037262]
15. Berry DA. Adaptive clinical trials in oncology. *Nat Rev Clin Oncol*. 2012; 9:199–207. [PubMed: 22064459]

Table 1

Baseline Characteristics of Study Cohort.

Median Age (range), years	52 [40–58]
Gender (male/female), n.	4/6
ECOG Performance status	
0–1	7
2	3
Prior Treatment Regimens, n.	
2	3
3	7
Prior Allogeneic HCT, n.	1
Cytogenetic Risk Group, n.	
Favorable	1
Intermediate	3
Unfavorable	6
Secondary AML, n.	2
Laboratory Findings at Baseline, median (range)	
WBC ($\times 10^3/\mu\text{L}$)	1.47 (0.12–28.59)
Hemoglobin (g/dL)	9.4 (7.3–11.2)
Platelets ($\times 10^3/\mu\text{L}$)	43 (9–165)
Creatinine	0.71 (0.2–1.22)
Total Bilirubin	0.55 (0.4–1.1)
SGOT	29 (12–55)
SGPT	22 (10.79)
Median TRM score (range)	4.48 (1.68–7.79)