# A Phase 2 Clinical Trial of Deforolimus (AP23573, MK-8669), a Novel Mammalian Target of Rapamycin Inhibitor, in Patients with Relapsed or Refractory Hematologic Malignancies

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**Abstract**

**Purpose:** Deforolimus (AP23573), a novel non-prodrug rapamycin analogue, inhibits the mammalian target of rapamycin, a downstream effector of the phosphatidylinositol 3-kinase/Akt and nutrient-sensing pathways. A phase 2 trial was conducted to determine the efficacy and safety of single-agent deforolimus in patients with relapsed or refractory hematologic malignancies.

**Experimental Design:** Eligible patients were assigned to one of five disease-specific, parallel cohorts and given 12.5 mg deforolimus as a 30-minute infusion once daily for 5 days every 2 weeks. A Simon two-stage design was used for each cohort. Safety, pharmacokinetics, pharmacodynamics, and antitumor response were assessed.

**Results:** Fifty-five patients received deforolimus as follows: cohort 1 23 acute myelogenous leukemia, two myelodysplastic syndrome and one chronic myelogenous leukemia in nonlymphoid blast phase; cohort 2, one acute lymphocytic leukemia; cohort 3, nine agnogenic myeloid metaplasia; cohort 4, eight chronic lymphocytic leukemia; cohort 5, nine mantle cell lymphoma and two T-cell leukemia/lymphoma. Most patients were heavily pretreated. Of the 52 evaluable patients, partial responses were noted in five (10%), two of seven agnogenic myeloid metaplasia and three of nine mantle cell lymphoma. Hematologic improvement/stable disease was observed in 21 (40%). Common treatment-related adverse events, which were generally mild and reversible, were mouth sores, fatigue, nausea, and thrombocytopenia. Decreased levels of phosphorylated 4E-BP1 in 9 of 11 acute myelogenous leukemia/myelodysplastic syndrome patients after therapy showed mammalian target of rapamycin inhibition by deforolimus.

**Conclusions:** Deforolimus was well-tolerated in patients with heavily pretreated hematologic malignancies, and antitumor activity was observed. Further investigation of deforolimus alone and in combination with other therapeutic agents is warranted in patients with selected hematologic malignancies.

The prognosis of patients with relapsed or refractory leukemias or lymphomas is poor. Furthermore, for some hematologic diseases, such as agnogenic myeloid metaplasia (AMM), patients generally receive only supportive care because no curative therapy has been defined. Developing new therapies against appropriate molecular targets offers the best hope of improving care for those with advanced hematologic cancer.

One promising therapeutic target is the serine-threonine kinase mammalian target of rapamycin (mTOR). mTOR is an attractive antitumor target because of its essential role in modulating translation of key regulators of the cell cycle. mTOR is a member of the phosphatidylinositol-3-kinase (PI3K) family and has a central role in regulating multiple pathways involved in cell growth, division, metabolism, and angiogenesis in normal and neoplastic cells (1). Activation of mTOR in response to a diverse array of growth and nutrient stimuli depends on upstream signaling through the PI3K/Akt pathway. Once activated, mTOR phosphorylates key translational regulators, such as the 40S ribosomal protein S6 kinase (S6K1) and the eukaryotic initiation factor 4E binding protein 1 (4E-BP1).

Through activation of downstream effectors, mTOR controls processes required for cell growth, cell division/proliferation (progression through the cell cycle), and cell survival (regulation of apoptosis; ref. 2). Dysregulation of the PI3K/Akt pathway upstream and downstream of mTOR is associated with transformation (3, 4). Furthermore, up-regulation of Akt and PI3K activity has been observed in several malignancies, including breast, gastric, non–small cell lung, ovarian, pancreatic, and prostate cancers (5–10).

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Accumulating evidence suggests that hyperactivation of the PI3K/Akt/mTOR pathway plays an important role in transformation of leukemias, lymphomas, and multiple myeloma (4, 11–14). Constitutive activation of PI3K and overexpression of eukaryotic initiation factor 4E give rise to lymphoproliferative disorders in transgenic mice (15). In addition, constitutive expression of cyclin D, a downstream effector of mTOR, is thought to contribute to the high rate of proliferation and poor prognosis in mantle cell lymphoma (MCL; refs. 3, 16, 17). Furthermore, Vega and coworkers found evidence that mTOR activation was critical to the survival of anaplastic large cell lymphoma (18).

The present trial focused on a subset of well-defined disease-specific categories of leukemias and lymphoid malignancies that have limited treatment options or only palliative therapies available. The categories were chosen primarily based on the scientific implication of the PI3K/Akt/mTOR pathway and the potential of mTOR inhibition to affect angiogenesis (2). In addition to MCL and anaplastic large cell lymphoma described above, PI3K/Akt/mTOR pathway-related abnormalities have been identified for the other leukemias and lymphoid malignancies studied here. These pathway aberrations include increased expression and constitutive activation of the catalytic subunit of PI3K and Akt in acute myelogenous leukemia (AML) and myelodysplastic syndrome (MDS; ref. 11), up-regulated pathway based on signaling through BCR/Abl in chronic myelogenous leukemia (CML; ref. 11), the role of the PI3K/Akt/mTOR pathway in normal and neoplastic T-cell and B-cell proliferation in acute lymphocytic leukemia (ALL) and chronic lymphocytic leukemia (CLL; ref. 19), up-regulated pathway based on signaling through platelet-derived growth factor, or the role of angiogenesis, a downstream effect of mTOR signaling, in AMM (20).

The earliest recognized mTOR inhibitor, rapamycin, acts by binding to endogenous FKBP-12 to produce a complex that potently inhibits mTOR activity. Deforolimus (AP23573, MK-8669) is a novel non-prodrug analogue of rapamycin with distinct pharmacokinetic properties (21). *In vitro*, deforolimus potently blocks mTOR activity and inhibits proliferation in multiple tumor cell lines, including an erythroleukemic line. Deforolimus has shown activity against a broad range of human tumor xenografts using intermittent dosing schedules (22). In a phase 1 clinical trial, deforolimus treatment was associated with antitumor activity across a broad range of tumor types when dosed intermittently (i.e., once daily for five consecutive days every other week; ref. 21). Deforolimus was also well tolerated (21). Side effects were generally mild or moderate, manageable, and reversible; mouth sores and rash were the most frequently encountered treatment-related events (21). No immunosuppressive side effects were observed. Based on these promising early clinical data, as well as the growing appreciation of the role of the PI3K/Akt/mTOR pathway in many hematologic malignancies, a phase 2 trial was designed to study the safety, efficacy, and pharmacodynamic activity of deforolimus in patients with selected advanced hematologic malignancies.

## Patients and Methods

***Eligibility criteria.*** Eligible patients were ages ≥18 y with a relapsed or refractory hematologic malignancy who were able to provide informed written consent before receiving deforolimus. Enrollees were assigned to one of five cohorts: cohort 1, AML, MDS (subtypes refractory anemia with excess blasts and refractory anemia with excess blasts in transformation), CML in nonlymphoid blast phase; cohort 2, ALL; cohort 3, AMM; cohort 4, CLL; and cohort 5, T-cell leukemia/lymphoma or MCL. They also had to have an Eastern Cooperative Oncology Group performance status of 0 to 2, adequate renal and hepatic function (serum creatinine and bilirubin, ≤1.5× ULN; aspartate aminotransferase/alanine aminotransferase, ≤3× ULN), no uncontrolled infection, and no other life-threatening illness. Patients must not have received prior therapy with rapamycin, rapamycin analogues, or tacrolimus and must not have had cytotoxic chemotherapy, radiotherapy, any investigational anticancer agent, or major surgery within 14 d before study entry. There was no limit on other previous therapies.

***Study design and treatment plan.*** This was a multicenter, open-label, nonrandomized, flat, fixed-dose trial of five parallel disease-specific cohorts to assess the efficacy and safety of deforolimus. Secondary efficacy end points included time to disease progression, progression-free survival, and duration of response. The study was approved by the institutional review boards of each participating center.

Patients received 12.5 mg deforolimus given i.v. over 30 min once daily for consecutively 5 d every 2 wk on a 4-wk cycle. The dose was based on results from a phase 1 trial evaluating the same dosing schedule (21).

***Assessment of toxicity and response.*** Safety assessments included physical examination, 12-lead ECG, and routine clinical laboratory evaluations during therapy and recovery. Follow-up safety evaluations were done 1, 3, and 6 mo after completion of study dosing. Adverse events were graded according to the National Cancer Institute-Common Terminology Criteria for Adverse Events, version 3.0. Investigators and affiliated staff determined whether physical examination or laboratory findings, such as thrombocytopenia, constituted a drug-related effect or were part of the underlying disease of the patient.

Tumor response evaluations were based on standard criteria (23–28). Peripheral blood (PB) cell counts and samples of bone marrow aspirates were assessed for changes in disease status. In cohort 1, hematologic improvement (HI) and stable disease (SD) were defined as described (25). Specifically, HI required improvement in one of the three hematopoietic lineages compared with baseline measurements, and SD required the absence of objective disease progression for at least 2 mo. Relevant responses had to be confirmed by a repeat measurement at least 1 wk later. In cohort 3, response criteria were derived from Tefferi et al. (27), where complete response (CR) represents absence of disease and partial response (PR) represents improvement in two of the three hematopoietic cell lineages or one lineage and improvement in organomegally. Assessments were done at the end of each cycle of treatment, except for those with lymphoma who had assessments done at the end of every even-numbered cycle.

***Pharmacodynamics.*** PB for analysis of plasma vascular endothelial growth factor (VEGF) levels (R&D Systems) was collected from a subset of consenting patients before dosing on cycle 1, day 1 and on cycle 2, day 1. Samples with values below the limit of quantitation (32 pg/mL) were assigned a value of 16 pg/mL for the purpose of calculating average results.

PB and bone marrow aspirates for cytometric analyses were to be collected before dosing on cycle 1, day 1, day 5 (PB only), day 15 (PB), day 21 (PB and bone marrow aspirate) and on cycle 2, day 1 (PB). On cycle 1, between days 5 and 7, a bone marrow aspirate sample was collected except for those with CLL or MCL with no known marrow involvement.

Quantitative flow cytometry to measure levels of total and phosphorylated 4E-BP1 (p-4E-BP1) and Akt was done as follows: PB or bone marrow aspirate samples were incubated with antihuman CD3 antibody conjugated with fluoroisothiocyanate, antihuman CD19 antibody conjugated with phycoerythrin, and antihuman CD34 antibody conjugated with allophycocyanin (Becton, Dickinson, and Company). Samples were washed, fixed with formaldehyde, treated

with a PBS-buffered saponin-based permeabilizing solution (Beckman Coulter), and incubated with primary antibodies against 4E-BP1 (Cell Signaling Technology), p-4E-BP1 ($Ser^{65}$/$Thr^{70}$; Santa Cruz Biotechnology), phosphorylated AKT ($Ser^{473}$), phosphorylated AKT ($Thr^{308}$), or AKT (Cell Signaling Technology). Samples were then washed and incubated with a phycoerythrin-conjugated secondary antibody (Santa Cruz Biotechnology) that had been previously standardized with QuantiBRITE PE beads (Becton, Dickinson, and Company) to measure antibody-binding capacity in each cell population. Cells were subsequently washed, resuspended, and analyzed using a FACSCalibur (Becton, Dickinson, and Company) flow cytometer to determine the percentage of positive cells, as well as the antibody-binding capacity (mol/100) for each analyte in each cell population. The product of these variables allows the overall levels of each analyte to be calculated.

Measurements of apoptosis and proliferation were done as previously described (29).

***Statistical analysis.*** The study was designed to test the null hypothesis that, within each disease category, the CR and PR rate would be ≤5%. The alternative hypothesis was that, within each disease category, the CR and PR rate would be ≥20%. To determine whether deforolimus has sufficient biological activity to warrant further single-agent testing, the trial used Simon's optimal two-stage design (30) with a minimum of 21 patients in stage 1 per cohort and expansion to a total of 41 evaluable patients if ≥2 CRs or PRs were observed in the initial 21 patients. All statistical tests were two-sided, setting the type I error level at 0.05, unless otherwise specified. There was no correction for multiple hypothesis testing. Summary statistics (number of patients, mean, minimum, median, maximum, standard deviation) were used to summarize all of the continuous variables.

## Results

***Study group.*** As of November 2006, a total of 55 patients had received at least one dose of deforolimus between July 2004 and May 2006. Pretreatment characteristics are summarized in Table 1. In general, patients were heavily pretreated; 32 had received three or more prior chemotherapy regimens for their disease, and three had stem cell transplants before entry onto the study.

Over the course of the study and follow-up period, 32 patients died, with five deaths occurring during treatment with deforolimus. None of the deaths were considered by the caregivers as related to deforolimus therapy. A total of eight discontinued treatment due to adverse events; three due to mouth sores and one each due to increased white cell count, rash, anemia, worsening congestive heart failure, and venous thrombosis.

Enrollment in the AML cohort was not reopened after stage 1 patient accrual, because the expansion criteria based on the Simon design was not met. For the other histologic cohorts (ALL, AMM, CLL, and lymphoma), most of which included rare hematologic malignancies, enrollment was discontinued after the planned enrollment period had elapsed considerably. Additional considerations contributing to the decision to close enrollment included the challenges of the daily i.v. dosing regimen in this infirm population and the entry of an oral deforolimus formulation into phase 1 clinical development. It should be noted that despite early closure, expansion criteria for stage 2 enrollment had already been met in the AMM and lymphoma cohorts.

***Response to treatment and pharmacodynamics.*** Patients completed a median of one cycle, with a median of 10 doses (range, 1-65) given. Dose reduction from 12.5 to 10 mg was necessary in 17 patients (31%), with a further reduction to 7.5 mg required for six patients.

Best responses to treatment for 52 evaluable patients are summarized in Table 2. There were five patients (10%) with PR, and one patient (2%) with an objective response of HI. Twenty patients (38%) maintained SD. Twenty-three patients (44%) had progressive disease (PD), and three patients with AML (6%) had relapse after HI. The duration of SD ranged from one to four cycles for AML, MDS, or AMM patients, one to five cycles for CLL patients, one to three cycles for MCL patients, and up to two cycles for the two patients with T-cell leukemia.

In cohort 1 (AML, MDS, and CML), one patient with AML (which had evolved from MDS) achieved an objective response of HI (normalization of neutrophils). Four patients with AML and one with MDS had SD for one to three cycles; however, all others in this cohort had either relapse after HI ($n$ = 3) or PD ($n$ = 16). Whereas a modest clinical response was noted in these patients, a significant reduction in mTOR activity was observed with deforolimus treatment. mTOR activity was assessed by measuring levels of p-4E-BP1 by quantitative flow cytometry before dosing and 24 hours after the fourth dose (i.e., day 5, predose) in PB cells collected from 11 patients. A significant reduction in p-4E-BP1 levels was observed after dosing with deforolimus ($P$ = 0.016; Fig. 1A and B, *left*). Overall, levels of 4E-BP1 in these same patients did not change significantly ($P$ = 0.93; Fig. 1B, *right*). No meaningful changes in predose and postdose levels of apoptosis and proliferation were detected. Cohort 2 consisted of one ALL patient whose best response was PD.

**Table 1.** Baseline patient characteristics ($n$ = 55)

| | |
|---|---|
| Age, y, median (range) | 61 (31-84) |
| Gender, male/female | 38 (69%):17 (31%) |
| Current diagnosis, $n$ (%) | |
| Cohort 1 | |
| AML | 23 (42%) |
| MDS | 2 (4%) |
| CML (nonlymphoid blast crisis) | 1 (2%) |
| Cohort 2 | |
| ALL | 1 (2%) |
| Cohort 3 | |
| AMM | 9 (16%) |
| Cohort 4 | |
| CLL | 8 (15%) |
| Cohort 5 | |
| T-cell leukemia/lymphoma | 2 (4%) |
| MCL | 9 (16%) |
| ECOG-PS* 0, 1, 2; $n$ (%) | 14 (26%), 33 (61%), 7 (13%) |
| No. prior chemotherapies | |
| 0 | 7 |
| 1 | 3 |
| 2 | 13 |
| 3 | 15 |
| 4 | 5 |
| 5 | 7 |
| 6 | 3 |
| ≥7 | 2 |

Abbreviation: ECOG-PS, Eastern Cooperative Oncology Group Performance Status.
*$n$ = 54.

**Table 2.** Best response to deforolimus treatment ($n$ = 52)

| Disease cohort | No. evaluable patients | PR | HI | SD | PD |
|---|---|---|---|---|---|
| 1. AML | 22 | 0 | 1 | 4 | 17 |
| MDS | 2 | 0 | 0 | 1 | 1 |
| CML | 1 | 0 | 0 | 0 | 1 |
| 2. ALL | 1 | 0 | N/A | 0 | 1 |
| 3. AMM | 7 | 2 | N/A | 3 | 2 |
| 4. CLL | 8 | 0 | N/A | 6 | 2 |
| 5. T-cell | 2 | 0 | N/A | 2 | 0 |
| MCL | 9 | 3 | N/A | 4 | 2 |

NOTE: 57 patients were enrolled. As of November 2006, 55 patients had treatment information. Of these, two AMM patients were misclassified and are not considered evaluable for antitumor response and 1 of the 23 AML patients was never evaluated for antitumor response, leaving 52 treated patients evaluable for antitumor response. The three AML patients with relapse after HI are included in the 17 patients with PD.
Abbreviations: N/A, not applicable; NR, no response (without progression).

In cohort 3 (AMM), two of seven evaluable patients (29%) achieved a PR (reduction in organomegally by 50% in both, with platelet count reduction in one patient and hemoglobin increase in the other) with SD that lasted five and six cycles, respectively. Both elected to discontinue treatment at the end of cycle 6. Each developed PD within the month of discontinuing therapy. In two of three patients analyzed from this cohort, the percentage of PB cells positive for p-4E-BP1 decreased after 4 days of treatment with deforolimus (from 58% to 44% and from 31% to 15% in those with SD and PD, respectively). In the third patient with a PR, p-4E-BP1 levels were low before treatment and remained low (1-3%). No meaningful changes were detected in the level of apoptosis or proliferation among these patients.

Six of eight evaluable patients in cohort 4 (CLL) achieved SD, two of which lasted for more than four cycles. The eight patients had a median number of three prior cytotoxic regimens. The two patients with SD for more than four 4 cycles had two and three cytotoxic regimens and had progressed on prior therapy before starting deforolimus 3 months later. Of the four other patients who achieved SD on deforolimus, three had a best response of PD to their prior therapy and one had a best response of SD. All four patients began deforolimus within 5 months or less of their prior therapy. Three of nine patients (33%) in cohort 5 (MCL) achieved a PR with deforolimus therapy. Tumor shrinkage (≥50%) was evident after the first cycle in these patients. One response lasted through three cycles, whereas the other two responses were maintained for four and five cycles of therapy and at least until the 1-month evaluation after therapy discontinuation. One patient discontinued therapy due to a venous thrombosis and was lost to follow-up. The other patient was switched to another experimental therapy.

Blood samples from one CLL and two MCL patients were available for pharmacodynamic analyses. Two of these, one with CLL and one with MCL, had SD; the other with MCL had PD. A reduction in phosphorylated AKT(Thr$^{308}$)-positive CD19+ cells was noted in the two patients with SD with levels decreasing from 9% to 1% in one and 9% to 3% in the other. In contrast, there was an increase in the percentage of phosphorylated AKT(Thr$^{308}$)-positive CD19+ cells (from 2% to 5%) for the patient with PD. Furthermore, an increase in apoptosis in lymphoid cells, as measured by mitochondrial potential, was observed in all three tested (1-34%, 5-12%, and 5-17%).

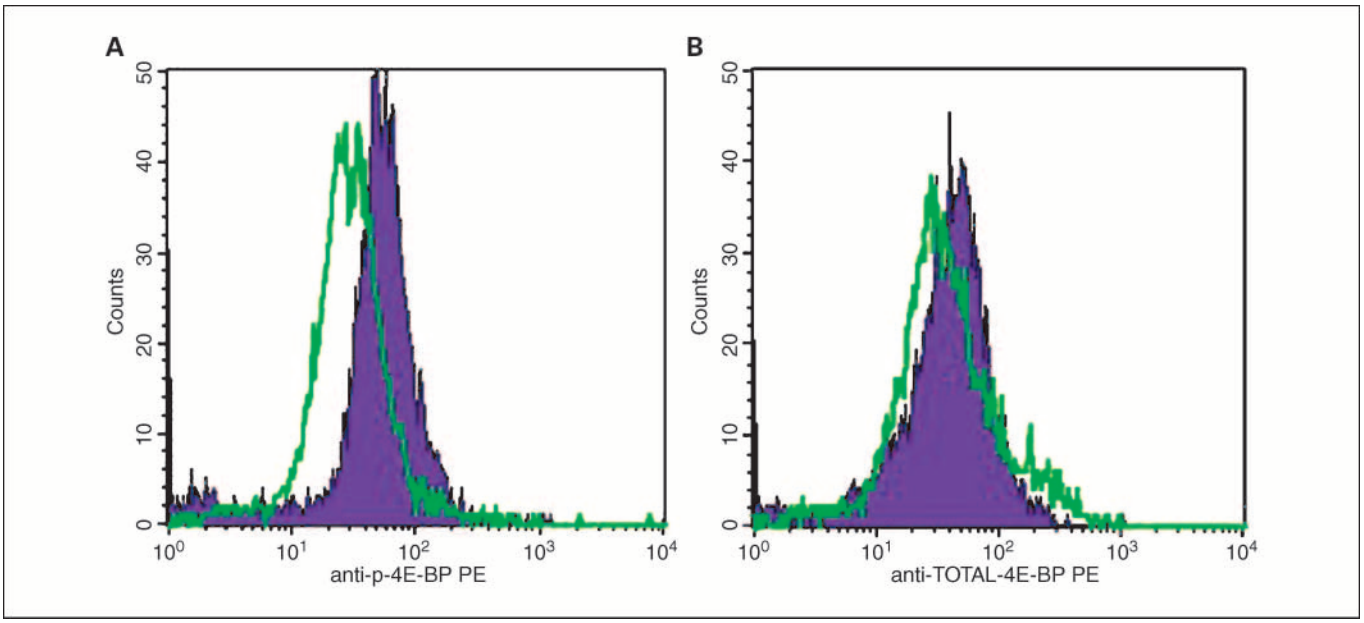


**Fig. 1.** Evidence of mTOR inhibition by deforolimus in AML patients. *A,* cytometric analysis of p-4E-BP1 (*left*) and total 4E-BP1 (*right*) levels in PB cells collected from patient 102 before dosing with deforolimus (day 1; *purple*) and 24 h after the 4th daily dose (day 5; *green*). *B,* percentage of the total cell population positive for p-4E-BP1 (*left*) or total 4E-BP1 (*right*) for samples collected before (day 1) and after (day 5) dosing with deforolimus. The *P* value for each protein, as determined by Wilcoxon matched pairs test, is shown. Patient numbers (all are AML patients) are indicated in the legend. The symbols used reflect the best-response assessment: SD (■; $n$ = 2), PD/HI (●; $n$ = 1), PD (×; $n$ = 6), NE (+; $n$ = 2).

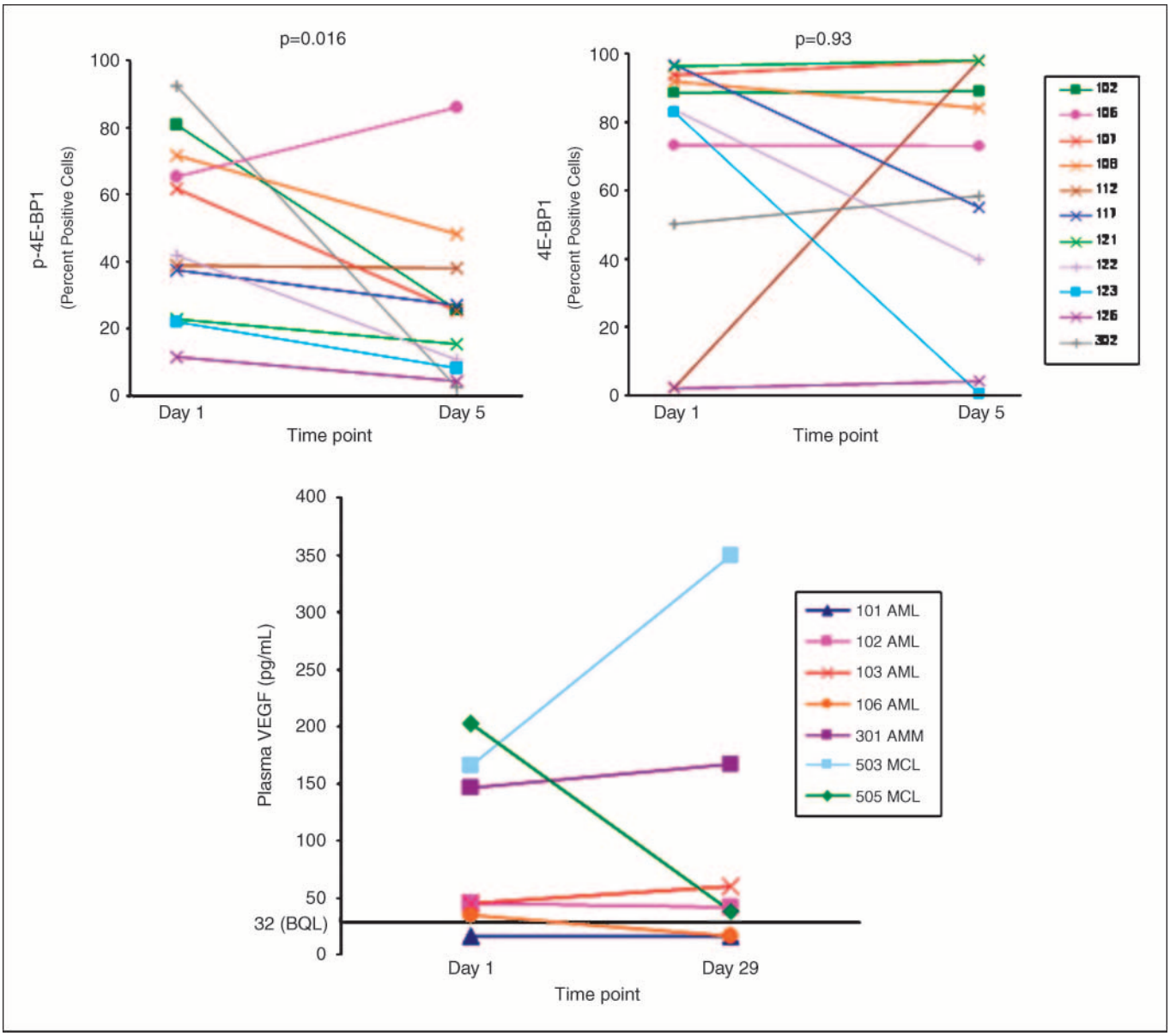


**Fig. 2.** Plasma VEGF levels before and after dosing with deforolimus. VEGF levels in plasma samples collected before dosing (day 1) and at the start of cycle 2 (day 29). Patient numbers and diagnoses are indicated in the legend. The symbols used reflect the best-response assessment: PR (♦; $n = 1$), HI (▲; $n = 1$), SD (■; $n = 3$), PD/HI (●; $n = 1$), PD (×; $n = 1$).

Production of VEGF, which promotes angiogenesis, is regulated by the mTOR pathway (2). Seven patients in the trial had predose and postdose plasma samples available for analysis of VEGF levels and were also evaluable for response. Of these seven, only one, with MCL, exhibited PR. Interestingly, VEGF levels in this patient decreased from 202 to 38 pg/mL after one cycle of treatment. The six remaining patients had a best response of PD (one patient), PD/HI (one patient), SD (three patients), and HI (one patient). Of these, none showed a substantial decrease in VEGF levels, with average levels rising from 78 to 126 pg/mL after one cycle of treatment (Fig. 2).

*Safety profile.* The frequency and grading of investigator-ascribed, treatment-related adverse events occurring in at least 5% of treated patients are summarized in Table 3. The most commonly encountered treatment-related adverse events were mouth sores (45%), fatigue (20%), nausea (20%), and thrombocytopenia (18%).

Grade 4 treatment-related thrombocytopenia occurred in three patients in cohorts 1, 3, and 5. Other treatment-related grade 4 toxicities included neutropenia, mouth sores, respiratory arrest, hypertriglyceridemia, hypocalcemia, and hypokalemia, all occurring in one patient each.

There were no significant reactions to the 30-minute infusion of study medication. Nine patients experienced infections considered at least possibly treatment-related. These included herpes simplex ($n = 3$; grades 1, 2, and 3) and pneumonia/upper respiratory infection ($n = 3$; grades 1 and 2), as well as a central line infection (grade 3), thrush (grade 1), and neutropenic sepsis (grade 3) in one patient each.

## Discussion

Despite the availability of standard chemotherapies, stem cell transplantation, and monoclonal antibody therapy, hematologic malignancies remain challenging to treat and new therapeutic strategies are needed. Dysregulation of the PI3K/Akt/mTOR pathway has been implicated in hematologic malignancies, and therapies targeting this pathway have the potential to inhibit tumor growth and improve survival.

A total of 55 heavily pretreated patients stratified in five independent disease cohorts received the mTOR inhibitor deforolimus as a single agent. Five PRs (10%) were achieved by three patients with MCL, and two patients with AMM; one patient with MDS had HI. In addition, 20 patients (39%) had some SD. None of the 22 evaluable patients with AML had CR or PR.

Of the five disease cohorts, the most favorable response was seen among patients with MCL. Although only nine patients with MCL were treated with deforolimus, the response rate was 33% (three PRs), similar to the 38% response rate (3% CR and 35% PR) observed with single-agent temsirolimus, another mTOR inhibitor (31). The rate of SD upon deforolimus treatment was 44%, and only two patients (22%) had PD on therapy. This promising activity may be due to the effects deforolimus likely has on cyclin D1, which is constitutively expressed in MCL tumor cells due to a characteristic chromosomal translocation. Because mTOR regulates cyclin D1 translation, mTOR inhibition by deforolimus might reduce expression of cyclin D1 and ultimately cell cycle progression.

Activity was also seen in those with AMM. Two of seven patients (29%) had PR and another three patients (43%) had SD. Stabilization was seen in 75% (six of eight) of those with CLL, and both patients with T-cell leukemia/lymphoma had SD.

Pharmacodynamic activity of deforolimus was detected in PB cells obtained from patients with AML, MCL, and CLL. In samples from those with AML, deforolimus was shown to inhibit mTOR as indicated by reduced phosphorylation of 4E-BP1, an established biomarker of mTOR activity. Nevertheless, only modest clinical activity was observed among those with AML, possibly due to activation of alternate signal transduction pathways in this tumor type (32). Consistent with the lack of robust clinical activity, deforolimus treatment had no effect on markers of cellular proliferation or apoptosis in samples from those with AML. In contrast, an increase in apoptosis in lymphoid cells from three of three MCL/CLL patients was observed. Furthermore, an overall decrease in phosphorylated AKT levels in two of three of these, which correlated with SD, was also seen. This result supports the hypothesis that reduced phosphorylation of Akt may be important for a favorable response to mTOR inhibition (33).

Plasma VEGF levels were measured before and after deforolimus infusion in a total of seven patients. A large decrease in VEGF levels was observed in one of these patients who exhibited a PR. No substantial decrease was detected in the other six patients, none of whom had a PR. These preliminary results are consistent with reports that increased angiogenesis and elevated levels of proangiogenic factors, such as VEGF, are found in a variety of hematologic malignancies and may play a role in negative outcome (34). Further study is needed to substantiate these findings.

Deforolimus was well tolerated in this heavily pretreated population. Overall, adverse events were mostly mild in severity and reversible. As in previous deforolimus clinical trials, mouth sores were the most commonly encountered related events. This favorable safety profile supports the potential of combining deforolimus with other targeted or cytotoxic agents.

Single targeted-agent therapy is unlikely to overcome redundancy and cross-talk among activated signal transduction pathways responsible for acute leukemias (32, 35, 36). This is supported by the variable response rates seen in previous trials with other inhibitors and the results of this trial (29, 37, 38).

**Table 3.** Treatment-related adverse events occurring in ≥5% of patients

| Toxicity | Grade 1/2 *n* (%) | Grade 3/4 *n* (%) | Total *n* (%) |
|---|---|---|---|
| Mouth sores* | 17 (31%) | 8 (15%) | 25 (45%) |
| Nausea | 10 (18%) | 1 (2%) | 11 (20%) |
| Fatigue | 11 (20%) | 0 (0%) | 11 (20%) |
| Thrombocytopenia | 2 (4%) | 8 (15%) | 10 (18%) |
| Diarrhea | 6 (11%) | 2 (4%) | 8 (15%) |
| Hypokalemia | 2 (4%) | 3 (6%) | 5 (9%) |
| Hyponatremia | 1 (2%) | 4 (7%) | 5 (9%) |
| Hypertriglyceridemia | 3 (6%) | 2 (4%) | 5 (9%) |
| Neutropenia | 2 (4%) | 2 (4%) | 4 (7%) |
| Pruritus | 4 (7%) | 0 (0%) | 4 (7%) |
| Rash | 3 (6%) | 1 (2%) | 4 (7%) |
| Weight loss | 4 (7%) | 0 (0%) | 4 (7%) |
| Anorexia | 3 (6%) | 0 (0%) | 3 (6%) |
| Herpes simplex | 2 (4%) | 1 (2%) | 3 (6%) |
| Hypocalcemia | 1 (2%) | 2 (4%) | 3 (6%) |
| Fever | 3 (6%) | 0 (0%) | 3 (6%) |
| Headache | 3 (6%) | 0 (0%) | 3 (6%) |
| Vomiting | 1 (2%) | 2 (4%) | 3 (6%) |

*Mouth sores included almost exclusively the verbatim terms "mucositis," "lip ulcer," and "mouth sores."

The activity and tolerability seen in this trial suggest that single-agent therapy with deforolimus could be effective in selected malignancies and might be successfully combined with other drugs for optimal efficacy. Combination studies will be facilitated by development of an oral preparation of deforolimus, which is under way. This study suggests single-agent deforolimus therapy in AML is unlikely to succeed and any further studies in AML should explore combination regiments. It may be particularly worthwhile to explore deforolimus as a treatment for MCL, CLL, AMM, and MDS.

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