

Phase II trial of Perifosine in Patients with Relapsed or Refractory Chronic Lymphocytic Leukemia/Small Lymphocytic Lymphoma

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Purpose of the study

The primary objective is to evaluate the response rate of perifosine in patients with relapsed or refractory CLL/SLL.

Secondary objectives are to:

- Monitor toxicity of perifosine
- Evaluate survival, progression free survival and response duration
- Perform laboratory correlates to assess changes in ex vivo CLL cells in phosphorylation, apoptosis, and gene expression with perifosine treatment. To measure in vitro killing of CLL cells with perifosine and to correlate these data with in vivo clinical efficacy.

Background & significance

Chronic lymphocytic leukemia and small B-cell lymphocytic lymphoma represent different manifestations of the same disease. CLL/SLL (hereafter denoted by CLL) is a clonal disorder of small B lymphocytes expressing a characteristic morphology and immunophenotype. The B cells express CD19, dim CD 20, dim CD 5, CD 23, CD 43, CD 79a, and weakly express surface immunoglobulin. CLL can present asymptotically in 25% of patients when diagnosed on a complete blood count. It also can present with diffuse painless lymphadenopathy and, in a smaller number of patients, B symptoms.

CLL is characterized by accumulation of circulating B cells predominantly in the G0 phase of the cell cycle. These cells are resistant to apoptosis. CLL has been found to have aberrant signaling in several pathways including NF- κ B, Akt/PI3K, and JNK/STAT pathways. Akt is important in promoting CLL survival and viability, as seen in *in vitro* experiments where blocking its activity results in apoptosis. Thus an AKT inhibitor may lead to increased apoptosis and may have a role in the treatment of this disease.

Treatment options for CLL range from a watch and wait approach to high dose chemotherapy with stem cell support. Currently, there is no consensus on the best treatment regimen, since no treatment has been shown to improve survival in randomized prospective clinical trials. New approaches to treatment, especially those with lower toxicity rates, are needed.

Perifosine has been shown to inhibit or otherwise modify signaling through a number of different signal transduction pathways, including Akt, MAPK, and JNK. These pathways are involved in the development of cancers and resistance to chemotherapy. Perifosine is of particular interest, especially due to the difficulty in discovery of drugs that inhibit these pathways with minimal toxicity. The effect of perifosine on CLL cells has been tested in the laboratory of Dr. Brice Weinberg and has been shown to be an active agent against primary CLL cells in vitro.

Design & procedures

Perifosine 50 mg will be taken orally, with food, twice a day on an outpatient basis in 28-day cycles, for a total of six cycles. Hematologic responses will be evaluated at cycle by review of the absolute blood count and manual blood film. Subjects will be removed from study treatment for unacceptable treatment-related toxicity or if disease progresses while on therapy. Subjects will be followed for survival for 2 years. Blood and bone marrow will be collected for correlative studies. All other assessments are within standard of care.

Correlative studies

Blood Samples:

- Pre-treatment: 4 green top (heparin) and 1 red top tubes (≤ 50 mL)
- Day 8, cycle 1: 2 green top (heparin)tubes (≤ 20 mL)
- Day 1, cycle 4: 4 green top (heparin) (≤ 40 mL)
- After completion: 4 green top (heparin) and 1 red top tubes (≤ 50 mL)
- 6 months after completion: 1 green top tube (≤ 10 mL)

Bone Marrow Biopsy Samples: Approximately 2-3 mm section of core biopsy placed in tube with RNAlater preservative.

Assays will be performed in the laboratories of Drs. Brice Weinberg, Mark Lanasa, and Daphne Friedman

- Measure in vitro cytotoxicity of perifosine against subjects' pre-treatment CLL cells prior to therapy and on completion of therapy, and correlate the measured ED50 with clinical response to therapy.
- Measure levels of phosphorylated and non-phosphorylated Akt, Erk, Syk, MEK, NF-kB, and PKC-alpha via western blotting and flow cytometry on subjects' leukemia cells collected prior to therapy, on day 8 of cycle 1, on day 1 of cycle 4, and after completion of the study.
- Measure apoptosis using Annexin V staining and/or caspase levels in subjects' leukemia cells collected prior to therapy, at day 8 of cycle 1, on day 1 of cycle 4, and after completion of the study.
- Measure nitric oxide and nitric oxide synthase levels prior to therapy, on day 8 of cycle 1, on day 1 of cycle 4, and after completion of the study.
- Evaluate genomic changes induced by therapy, correlate gene expression with outcome, and compare genomic data in pretreatment blood and bone marrow. Blood collections prior to therapy, at day 1 of cycle 4, and on completion of study. Bone marrow collection prior to study.
- Determine minimal residual disease status on completion of study, and enumerate T-cell subsets prior to therapy, on completion of therapy, and 6 months after completion of therapy.

Selection of Subjects

Inclusion Criteria

- A diagnosis of CLL or SLL based on iwCLL diagnostic criteria.
 - CLL: CD5⁺/CD19⁺ clonal B-cell count of $\geq 5000/\mu\text{L}$ for > 3 months
 - SLL: lymphadenopathy or bone marrow failure due to CD5⁺/CD19⁺ clonal B-cells with circulating CD5⁺/CD19⁺ clonal B-cells $< 5000/\mu\text{L}$
- Prior therapy for CLL (no limit on number of prior regimens).
- Patients requiring therapy, based on at least one of the following iwCLL criteria:
 - Evidence of progressive marrow failure (development or worsening of anemia and/or thrombocytopenia).
 - Massive (>6 cm below left costal margin) or progressive or symptomatic splenomegaly.
 - Massive lymph nodes (> 10 cm in longest diameter) or progressive or symptomatic lymphadenopathy.
 - Progressive lymphocytosis with increase of more than 50% over a 2-month period or lymphocyte doubling time of < 6 months.
 - Disease-related symptoms such as unintentional weight loss $>10\%$ in past 6 months, fatigue with PS ≥ 2 , fevers $> 38^{\circ}\text{C}$ for 2 weeks or more without evidence of infections, night sweats for > 1 month without evidence of infections.
- 18 years of age or older.
- Performance status ECOG 0, 1, or 2.
- An estimated or measured creatinine clearance ≥ 30 ml/min at study enrollment.
- AST, ALT, and total bilirubin ≤ 2.5 times the upper limit of normal, unless due to CLL/SLL.
- Voluntary written informed consent before performance of any study-related procedure not part of normal medical care.
- Female subject is either post-menopausal or surgically sterilized or willing to use an acceptable method of birth control (i.e., a hormonal contraceptive, intra-uterine device, diaphragm with spermicide, condom with spermicide, or abstinence) for the duration of the study therapy and for 2 weeks after study therapy completion.
- Male subject agrees to use an acceptable method for contraception for the duration of the study therapy and for 2 weeks after study therapy completion.

Exclusion Criteria

- Female subject is pregnant or lactating. Confirmation that the subject is not pregnant must be established by a negative serum β -human chorionic gonadotropin (β -hCG) pregnancy test result within 2 weeks of enrollment. Those of child bearing potential with a positive G-hCG test must be shown not to be pregnant with an appropriate ultrasound. Pregnancy testing is not required for post-menopausal or surgically sterilized women.
- Patient has received other investigational drugs for this disease within 14 days of enrollment.
- Patient with known HIV prior to enrollment.
- Serious medical or psychiatric illness likely to interfere with participation in this clinical study.
- Patients with another malignancy within the last three years (from documentation of remission) other than basal or squamous cell skin cancer or CIS of the cervix or early stage prostate cancer not requiring systemic treatment.

- Patients who underwent allogeneic stem cell transplant and have at least 2% donor cells engrafted will be excluded.
- Significant cardiac or vascular events within 6 months: acute MI, unstable angina, severe peripheral vascular disease (ischemic pain at rest class 3 or worse, non-healing ulcers/wounds, congestive heart failure (NHYA class ≥ 2), uncontrolled cardiac arrhythmias.
- Known severe hypersensitivity to perifosine or any component of the formulation.
- Life expectancy less than six months due to co-morbid illness
- Active autoimmune hemolytic anemia or immune thrombocytopenia, requiring current steroid therapy
- *De novo* prolymphocytic leukemia (PLL) or PLL arising from CLL
- Richter's transformation

Subject recruitment and consent

Subjects will be identified from the patient populations of the Duke University Medical Center Hematologic Malignancies program. Physicians from this program will initiate conversation with the patient about the research trial. If interest is expressed, the research nurse or investigator will review the consent form with the subject. Time is allotted to read/review the consent and have questions answered. Documentation of the consenting process is made in the subject's record.

This study will be open to members of all demographic groups who meet the eligibility criteria. It is estimated that 33 DUHS subjects will be consented. There is no compensation for enrollment.

Subject's capacity to give legally effective consent

Only subjects competent to give informed consent will be enrolled on this study.

Risk/benefit assessment

Common events ($\geq 10\%$) related to perifosine are: decreased hemoglobin, lymphopenia, decreased platelet count, fatigue, anorexia, diarrhea, nausea, vomiting, increased alanine aminotransferase, increased bun, increased creatinine, hyperglycemia, hypoalbuminemia hyponatremia, increased LDH.

Dose modifications will be made for persistent nausea, vomiting, diarrhea, or other persistent grade 2 adverse events that, despite symptomatic treatment, remain unacceptable (intolerable) to the subject.

Dose modifications will be made for Grade 3 or 4 events. Perifosine will be until toxicity improves to \leq grade 1 or has returned to baseline level. If the toxicity does not resolve after 2 weeks, study drug will be permanently stopped. If the toxicity resolves within 2 weeks and upon agreement with the clinical investigator, perifosine will be restarted at 50 mg daily (one tablet daily). If the subject has already had a dose reduction, the patient may be removed from study if the adverse event reappears.

Alternatives to this study include taking drugs that are approved for the treatment of CLL [such as Chlorambucil (Leukeran®), Cyclophosphamide (Cytoxan®), and Fludarabine (Fludara®)], bone marrow transplant, taking part in another research study, receiving supportive care only.

A possible benefit to study participation is disease remission.

Costs to the subject and compensation

Study drug will be provided by Keryx Biopharmaceuticals/AOI Pharmaceuticals Inc. Costs for correlative studies will not be charged to subjects. All other assessments are within standard of care and will be billed to the subjects/their insurance. No compensation for study participation will be provided.

Data Analysis & Statistical Considerations

A total of 33 subjects will be accrued at an accrual rate of 10 to 20 subjects per year: we expect about 10% of the patients to be unevaluable for response. While only the approximately 30 patients evaluable for response will be used to estimate response rate, all patients will be used in the analyses of toxicity, time-to-progression, and overall survival. The primary objective of this trial is to estimate the response rate (CR+PR) of perifosine. The historical control response rate in this setting is 0.30, and this study hopes to match this rate. A response rate of 0.10 would be considered poor. Perifosine will be considered worthy of future research if the observed response rate is at least 20% (6/30). If there are 6 responders, then the exact 80% confidence interval will be 0.11-0.42. If the true response rate is 0.30, then the probability of seeing at least 6 responders is 92%. If the true response rate is 0.10, then the probability of seeing at least 6 responders is 7%. While these calculations may sound like they are coming from a hypothesis testing framework, the purpose of this study is simply estimation. Toxicity and other feasibility issues will also have to be taken into account in deciding whether this drug is worthy of further research. There will be no interim analyses, because the median time to response is quite long. However, roughly speaking, if 1 or less responders are observed in the first 12 accrued subjects, we will consider stopping the study. If we were to have considered this a hypothesis-testing study (instead of an estimation study) and designed it as a 2-stage design, then this interim analysis would have resulted in an alpha of 0.06 and a power of 0.87.

Toxicity will be tabulated by Type and Grade. Overall survival will be defined as the length of time between study enrollment and death due to any cause. Progression-free survival will be defined as the length of time between study enrollment and disease progression or death due to any cause, whichever comes first. Response duration will be defined within responders as the length of time between first response and disease progression or death due to any cause. All three of these time-to-event endpoints will be estimated with the Kaplan-Meier method.

Change in ex vivo CLL cells will be described by giving the 1st, 25th, 50th, 75th, and 100th quantiles. This analysis will be done both by using all 33 subjects, as well as within the responders and non-responders.

Data & Safety monitoring

Safety concerns are discussed in the Risk/benefit section above. The research team will monitor toxicities and make dose modifications as outlined above.

Dr. Friedman will meet with members of the study team on a regular basis to review toxicity data, response, accrual and review of study conduct while patients are being treated. All Serious Adverse Events that occur during the study will be assessed by the Principal Investigator for assessment of attribution to study drug and DUHS IRB reporting requirements. Dr. Friedman is responsible for reviewing all SAEs and response to treatment to determine if the study should continue after the first 12 subjects have completed treatment.

The protocol will be monitored and assessed independently via the Duke Comprehensive Cancer Center (DCCC) Protocol Review and Monitoring Process. A determination of the degree of monitoring is made at the time of initial Cancer Protocol Committee (CPC) approval to commensurate with the type and level of intervention, phase, endpoints, degree of risk, size and complexity of the protocol. The minimum level of monitoring for interventional, therapeutic (agent or device) clinical protocols is a routine monitoring visit after the first three subjects have been enrolled. Annual progress reviews will be conducted to assess scientific progress and adequacy of accrual. Subsequent monitoring would be based on monitoring visit(s) and/or the annual progress review.

Data storage & confidentiality

Paper and electronic case report forms (CRFs) will be used for this study. Subject research charts and paper CRFs will be kept in the lock research offices of the Hematologic Malignancies group. Research records will be archived upon completion of the study. Labs are locked and have restricted access.