Chronic Lymphocytic Leukemia:
Something Old, Something New and Something Borrowed...

Kimberly Perez, MD, and Eric S. Winer, MD

Chronic Lymphocytic Leukemia (CLL) is a disease of the elderly: almost 70% of diagnoses are made in patients older than 65 years. This indolent lymphoproliferative disorder, manifested by a clonal expansion of mature but functionally defective lymphocytes, is one of the most common leukemias of adults in Western countries. The incidence rate is roughly 2-6 cases per 100,000 persons per year, and increases with age reaching 12.8 cases per 100,000 at age 65 (the mean age at diagnosis).[1] The disease is more common in men, with an incidence ratio of 1.5-2:1. It also has been found to be more common in the Caucasian population, compared to African American, Hispanic, Native American or Asian populations.

There are no clear occupational or environmental risk factors for CLL. The prevalence in Caucasian populations suggests a genetic versus an environmental influence on the etiology. A recent study supports an association between blood transfusions and CLL development in 66% patients evaluated.[3] However, among the strongest risk factors for the development of CLL is a family history of CLL or other lymphoid malignancy. Retrospective studies have noted increased incidence in first degree relatives. A term coined "genetic anticipation" has also been used to describe a process in which the median age at diagnosis in an affected family decreases in younger generations.[2]

Clinical Presentation

The clinical presentation, course and outcome are extremely heterogeneous. At presentation most patients are asymptomatic; up to 25% are diagnosed when routine blood work reveals an absolute lymphocytosis. For those who present with clinical symptoms, most commonly it is in the form of painless, intermittent swelling of lymph nodes. In 5-10% of patients, clinical presentation consists of B symptoms, which include unintentional weight loss ≥10% within the previous six months, fevers >100.5°F (>38°C) for ≥2 weeks without evidence of infection, drenching night sweats without evidence of infection, or extreme fatigue (ie, ECOG Performance status 2 or worse; cannot work or perform usual activities). Other clinical findings consist of autoimmune hemolytic anemia or thrombocytopenia, recurrent infections, splenomegaly, hepatomegaly, or, rarely, extranodal infiltrates.

Histologic findings on the peripheral blood smear are typically of small lymphocytes with clumped chromatin and scanty cytoplasm. Smudge or basket cells are usually present. (Figure 1) There is usually a small proportion of immature lymphocytes, such as prolymphocytes, at a rate of <2%. Bone marrow findings are variable. Cellularity is normal or increased, and typically the architecture is disrupted, following interstitial, nodular, and/or diffuse patterns. The **International Workshop on CLL (IWCLL)** describes greater than 30% lymphoid cells as present for the diagnosis.[4]

Diagnosis

The diagnosis of CLL can be made utilizing the IWCLL guidelines, based on the identification of cells bearing the unique phenotype of CLL in peripheral blood (CD5-positive/CD19-positive). However, before the diagnosis can be made an important distinction needs to be made between CLL, **small lymphocytic lymphoma (SLL)** and **monoclonal B-cell lymphocytosis (MBL). The World Health Organization (WHO)** classifies both CLL and SLL as different clinical manifestations of the same disease: CLL is the leukemic component with genesis occurring in the marrow, whereas SLL is defined by a lymphomatous origin. To differentiate CLL and MBL, the distinction is made based on the absolute lymphocyte count. MBL is defined in patients who have an absolute increase in the number of clonal B lymphocytes in the peripheral blood that does not exceed 5000/microL and who have no lymphadenopathy, organomegaly, cytopenias, or disease-related symptoms.[5]

The diagnostic criteria for CLL include an absolute B lymphocyte count in the peripheral blood ≥5000/microL for more than 3 months, with a preponderant population of morphologically mature-appearing small lymphocytes and the demonstration of clonality of the circulating B lymphocytes by flow cytometry of peripheral blood. Clonality is determined by extremely low levels of surface immunoglobulin M and IgD with either kappa or lambda (but not both) light chains; expression of B-cell associated antigens (CD19, dim CD20, CD23, CD43 and CD79a); and expression of the T-cell associated antigen CD5.

In CLL, the bone marrow demonstrates infiltration by the clonal population in the range of 30-100%. Prior to the development of cytogenetic markers, bone marrow biopsy was utilized for diagnostic and prognostic indications. However, with the use of cell surface markers for clonality determination, the use of bone marrow aspirate and biopsy has become unnecessary for diagnosis. The same is true in regards to its prognostic use.

After the diagnosis, two clinical staging systems can be utilized: the Rai stage or the Binet system. (Table 1) Both take into account end-organ involvement due to progressive accumulation of neoplastic cells and the development of lymphadenopathy, organomegaly and cytopenias. The Rai system based their classification on the disease process, whereas the Binet system stages based on sites of involvement.[6,7] For both systems, a thorough clinical exam and complete blood count are necessary. The guidelines do not recommend the use of **computed tomography (CT)** scans at diagnosis for assessment of end-organ involvement. Some oncologists may order a CT scan if the patient has poor prognostic features, which can result in upstaging.

Determining Prognosis

The advent of molecular profiling revealed characteristics of CLL as a disease process. The profile consists of cytogenetic

Table 1. Staging systems in chronic lymphocytic leukemia

	Definition	Risk stratum	Median survival
Rai System			
0	Sustained lymphocytosis >10,000mcL	Low	150 months
1	Lymphocytosis with adenopathy	Intermediate	70-100 months
2	Lymphocytosis with splenomegaly		
3	Lymphocytosis with anemia	High	20 months
4	lymphocytosis with thrombocytopenia		
Binet System			
A	Less than 3 areas of lymphadenopathy; no anemia or thrombocytopenia	Low	Comparable to age-matched controls
B	More than 3 involved node areas; no anemia or thrombocytopenia	Intermediate	84 months
C	hemoglobin <10g/dL and/ or platelets <100 cells/mm^3	High	24 months

analysis by **fluorescence in situ hybridization (FISH)** (del 13q24; trisomy 12; deletions or mutations of 11q22-23; deletions or mutations of 6q21 or 17p13) and mutational status of the **immunoglobulin heavy chain variable gene locus (IgVH).** Due to the lack of access to IgVH mutational testing, attempts at securing surrogate markers have been developed. Examples of surrogate markers are ZAP70 and CD38 expression. On final analysis, poor prognostic features consist of IgVH unmutated status, which in turn the expression of ZAP70 or CD38 can act as a surrogate. Some major limitations in these molecular profile markers are the limited access of IgVH, mutational status testing sites, the unreliability of the assays used to determine ZAP70 status, and the question of stability in regards to CD38. Therefore, the use of the biomarkers is limited: they may help predict the behavior of a cohort of patients will behave, but they are not yet precise in predicting the outcome in individual patients.

Treatment – Something Old, Something New, Something Borrowed...and the kitchen sink

Since CLL tends to be an indolent disease and also a disease predominantly of older people, the most important decision to make is when to initiate treatment. Previous meta-analyses have shown that there is no survival benefit for patients who have early intervention versus deferred chemotherapy; in fact there was a slight trend towards worse survival in the early treatment category.[8] The updated IWCLL guidelines offer the following criteria:[4] 1) progressive marrow failure demonstrated by new or progressive anemia or thrombocytopenia, 2) massive or symptomatic splenomegaly, 3) massive of symptomatic lymphadenopathy, 4) a lymphocyte increase of more than 50% over a two-month period or a lymphocyte doubling time of <6 months, 5) autoimmune anemia and or thrombocytopenia poorly responsive to steroids, and 6) presence of B symptoms. If no treatment is indicated, the patient is followed by clinical exam and complete blood counts every 3 months for the first year. These intervals can be modified depending on aggressiveness of disease.

Once the decision to treat has been made, the clinician and patient must decide the goals of treatment. It is uncommon for patients to be cured from their CLL with conventional chemotherapy. While the goal of treatment in a younger patient may be to induce a complete remission of maximum duration, the goal for the older patient may be to manage the disease, such as minimizing anemia and thrombocytopenia. Numerous regimens have been used in CLL, many of which are shown in Table 2.

Something Old: Chlorambucil and purine analogues

Chlorambucil was the first chemotherapeutic to be effectively used in the treatment of CLL. It is given orally as either a low dose daily regimen or pulsed with prednisone on a monthly basis. It is very well tolerated with minimal myelosuppression, nausea, or alopecia. The limitation is that it has a low **complete response (CR)** rate of 0-4%, with an **overall response (OR)** rate of 31-55% when evaluated in multiple modern comparative studies ranging.[9-11] However, despite the lower response rate, very few drugs have conferred a survival advantage when compared to chlorambucil. It remains the treatment of choice for elderly or poor performance status patients due to its efficacy and its excellent side effect profile.

Three purine analogues (fludarabine, pentostatin, and cladribine) are currently used for the treatment of CLL, with the most common being **fludarabine (F).** When compared as a single agent with chlorambucil, fludarabine demonstrated a significantly higher CR (7-20%) and OR (63-72%) rates. However, there was no significant difference in overall survival. The side effect profile shows higher rates of grade 3-4 cytopenias, and therefore more complications with immunosuppression. Similarly, cladribine also produced higher OR rates, but no improvement in overall survival.

Something New: Monoclonal Antibodies

The monoclonal antibodies have revolutionized the treatment of lymphomas. These antibodies directly bind to the surface of the malignant cell and induce cell killing by **antigen dependent cellular cytotoxicity (ADCC), cellular dependent cytotoxicity (CDC)** and/or direct induction of apoptosis. The first mab in

clinical use was rituximab, an anti-CD20 mab used to treat B-cell lymphomas, is now standard in nearly all of the B-cell lymphoma-directed regimens. The CD20 antigen can also be found on 95% of CLL cases. In single agent studies of rituximab in CLL, which used doses of rituximab much higher than conventionally used in other lymphomas, the overall response rates were 36% and 45%. Ofatumumab, a second-generation anti-CD20 mab, has also recently been studied in CLL. In a group of fludarabine refractory CLL patients, the overall response rate was 47-58%.[12]

Another monoclonal antibody widely used in the treatment of CLL is alemtuzumab, an anti-CD52 mab. Alemtuzumab has been compared to chlorambucil and has shown to have higher CR (24%) and OR (83%) rates, but has not demonstrated an overall survival benefit.[20] Initially the most common side effects were severe transfusion reactions with hypotension; however, with the conversion from intravenous to subcutaneous administration, these have been replaced by injection site reactions. Persistent side effects are immunosuppression, particularly viral reactivation (VZV, CMV). Alemtuzumab has proven efficacy in patients considered high-risk with cytogenetic abnormalities such as deletion of chromosome 11 (del 11q) or chromosome 17 (del 17p) and the p53 mutation. Lumiliximab, a novel anti-CD23 mab, was recently evaluated in a phase I trial, but demonstrated limited clinical activity as a single agent.

In treating CLL patients, the first question to ask is "Does this patient need to be treated?"

Something Borrowed: Bendamustine

Bendamustine, an alkylator in the nitrogen mustard family, was first discovered in the former East German Democratic Republic in the early 1960s. Although extensively studied in East Germany, bendamustine had few validation studies and therefore did not gain worldwide acceptance until well after the unification of Germany. One of the initial bendamustine trials in patients with refractory CLL demonstrated an overall response rate of 56%,[13] while the phase III trial against chlorambucil demonstrated a CR rate of 31% and an OR rate of 68%. Bendamustine has a very well tolerated side effect profile notable for mild cytopenias, moderate nausea, and minimal alopecia.

...and the Kitchen Sink

With multiple agents available, the logical progression is the combination of chemotherapeutic agents. The first combination studied extensively was the combination of **fludarabine (F),** a purine analogue, with **cyclophosphamide (C),** an alkylator. Two studies compared F to FC, and one study compared these two regimens to standard chlorambucil.[14-16] FC showed a significant improvement in efficacy, with a CR rate of 23-38% and an overall response rate of 74-94%. Although there was an increase in grade 3-4 neutropenia with this regimen, there was no increase in the rate of severe infections.

The natural progression after this study was the combination of conventional chemotherapy with monoclonal antibodies. **Fludarabine (F)** combined with **rituximab (R)** in the front-line setting yielded a CR rate of 47% and OR of 90%. Impressively,

Table 2. Selected randomized first-line studies in chronic lymphocytic leukemia

Study	Regimen	CR rate (%)	OR rate (%)	PFS (Months)
Rai [9]	Fludarabine	20 (p< 0.001)	63 (p< 0.001)	20 (p< 0.001)
	Chlorambucil	4	37	14
Hillmen [11]	Alemtuzumab	24 (p< 0.001)	83 (p< 0.001)	15 (p<0.001)
	Chlorambucil	2	55	12
Knauf [10]	Bendamustine	31 (p< 0.001)	68 (p< 0.001)	22 (p< 0.001)
	Chlorambucil	2	2	8
Catovsky [15]	Chlorambucil	7	72	5 years-10%
	Fludarabine	15	80	5 Years-10%
	Fludarabine + cyclophosphamide	38 (p<0.0001)	94 (p< 0.001)	5 Years-10% (p<0.00005)
Finn [14]	Fludarabine	5	59	19
	Fludarabine + cyclophosphamide	23 (p< 0.001)	74 (p< 0.001)	32 (p< 0.0001)
Eichhorst [16]	Fludarabine	7	83	20
	Fludarabine + cyclophosphamide	24 (p< 0.001)	94 (p= 0.001)	48 (p=0.001)
Hallek [18]	Fludarabine + cyclophosphamide	23	88	2 Years-76.6%
	Fludarabine + cyclophosphamide + rituximab	44 (p<0.0001)	95 (p= 0.001)	2 Years-62.5% (p<0.0001)

phase II data using FCR in front-line CLL demonstrated a CR rate of 72% and an OR rate of 95%.[17] Phase III data comparing FC to FCR also showed superiority of FCR with similar results, however there was no significant difference in progression free survival or overall survival in the patients with the chromosomal deletion of 17 (del 17p). The addition of rituximab to the fludarabine and cytoxan demonstrated a survival benefit over FC alone.[18] Other purine analogues have been substituted for fludarabine in an attempt to reduce the myelotoxicity, such as the combination of **pentostatin, cyclophosphamide and rituximab (PCR)** but this showed no significant improvement in response rates or decrease in rate of infection. Trials have also studied the addition of alemtuzumab to FC or FCR, but these did not show improved efficacy and had a higher incidence of myelosuppression. Other chemotherapeutic regimens have been attempted with fludarabine but none showed improved efficacy to FCR.

One intriguing combination that demonstrated efficacy was the combination of **bendamustine and rituximab (BR).**[19] This regimen demonstrated a CR rate of 15% and OR rate of 77%. Because of the favorable side effect profile and high response rate, BR is being compared to FCR in a large phase III clinical trial. Similarly, **lumiliximab (L)**, the anti-CD23 antibody, has also been found to be synergistic with FCR;[20] a large phase III trial is ongoing comparing L-FCR with FCR. Other novel agents, such as lenalidomide, flavopiridol and ABT-263, are being studied; it is unclear what role these agents will play in the treatment of CLL.

Relapsed Disease

It is common for patients to relapse after a period of time in a CR. There are few guidelines as to the best second-line treatment. Patients who have high risk disease or relapse in less than one year from treatment and are eligible for high-dose chemotherapy and should be offered allogeneic stem cell transplantation, which has shown to be curative,[21] in a clinical trial setting. Patients with chemosensitive disease who obtain long remissions from their initial treatment are usually treated in the relapse setting with chemotherapy different than that received previously; or, if the remission was longstanding, patients often receive the same regimen.

Recommendations and Summary

In treating CLL patients, the first question to ask is: "Does this patient need to be treated?" Although it is often difficult for patients to gain comfort with "watch and wait," it is important to emphasize that there is no benefit to early intervention. Once criteria have been met for treatment, patients are stratified by performance status and presence of "high risk" molecular features (del 17p or del 11q mutations). Although clinical trials are the preferred treatment, they are not available in all locations. Therefore, older, poor performance patients should be started on chlorambucil, whereas younger patients should be treated with FCR. The treatment for CLL patients with 17p deletion (high-risk) should start with FCR, and include alemtuzumab and allogeneic stem cell transplantation.

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Kimberly Perez, MD, is an Assistant Professor of Medicine, Division of Hematology and Oncology, The Warren Alpert Medical School of Brown University / Rhode Island Hospital.

Eric S. Winer, MD, is an Assistant Professor of Medicine, Division of Hematology and Oncology, The Warren Alpert Medical School of Brown University / Rhode Island Hospital.

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Correspondence

Eric S. Winer, MD
Rhode Island Hospital
593 Eddy St, Providence, RI 02903
Phone: (401) 444-4000
E-mail: ewiner@lifespan.org