ABSTRACT

Chronic lymphatic leukemia (CLL) is a monoclonal hematopoietic disorder characterized by a progressive expansion of lymphocytes of B-cell lineage. The hallmark of CLL cells is that they express CD5 (an antigen commonly found on T cells), CD19, CD20, CD23 are other B-cell markers expressed on CLL cells. The diagnosis of CLL is often incidental, routine blood counts may reveal an elevated absolute lymphocyte count. Diagnostic criteria as proposed by National Cancer Institute (NCI) is followed. Currently we are more and more often using also biological parameters in order to evaluate the degree of aggressiveness of the disease. These parameters are chromosomal abnormalities, presence of IgV gene mutation, expression of certain surface antigens on lymphocytes in particular CD38 and ZAP-70. The well defined staging system for CLL includes Rai's and Binet Staging. CLL is a disease which is often diagnosed in an asymptomatic patient and has a prolonged course, it is not curable with current treatment approaches and survival advantage was not demonstrated with early intervention trials. Considerable progress has been made in the understanding of biology of CLL and developing new and effective treatments. Long term remissions are now possible in a subset of patients. The outlook for CLL patients has improved and ongoing research promises even more progress.

INTRODUCTION

Chronic lymphatic leukemia (CLL) is a monoclonal hematopoietic disorder characterized by a progressive expansion of lymphocytes of B-cell lineage. These small lymphocytes accumulate in the blood, bone marrow, lymph nodes and spleen.

CLL is a common leukemia in the western world, it accounts for 25% to 30% of all adult leukemia's. The disease is uncommon in Asian population; in Japan it accounts for only 2.5% of all leukemia's. The incidence is age dependent with an increase from 5.2 per 1,000,000 persons older than 50 years to 30.4 per 1,000,000 persons older than 80 years. The male to female ratio is 1.5:1.

BIOLOGY:

CLL is a clonal B-cell lymphoid leukemia. The hallmark of CLL cells is that they express CD5 (an antigen commonly found on T cells), CD19, CD20, CD23 are other B-cell markers expressed on CLL cells. Based on the antigen expression profile, CLL appears to arise from an "activated" B cell.

Assessment of somatic hypermutation immunoglobulin variable Gene (IgV) defines two "subsets" of CLL. Approximately 50% of CLL cases have somatic hypermutation of IgV gene and appear to arise from post germinal B cells, while the subset of CLL lacking IgV hypermutation appears to arise from naive B cells. The mutation status of CLL seems fixed and mutational status is not gained or lost during the course of disease. Interestingly, the mutation status provides significant prognostic information. Lack of somatic hypermutation indicates an inferior prognosis. Because of the complexity of testing for IgV gene hypermutation, a search for surrogate marker has ensued. More recently expression of Zeta associated protein 70 (ZAP – 70) was found to correlate inversely with IgV mutation.

Fluorescent in situ hybridization (FISH) using genomic DNA probes have greatly enhanced the ability to detect molecular abnormalities in malignant cells. FISH has demonstrated that molecular abnormalities occur in up to 80% of cases of CLL. 13q deletion is the most common genetic aberration found in CLL (55%) followed by 11q deletion (18%), 12q trisomy (18%) and 17p deletion (7%). The prognosis of CLL varies with the chromosomal abnormality. When divided into five prognostic categories – 17p deletion, 11q deletion, 12q trisomy, normal karyotype and 13q deletion (as sole abnormality) the survival times were 32, 79, 114, 111 and 133 months respectively.

Patients with 17p or 11q deletion had more advanced disease with frequent splenomegaly, mediastinal and abdominal adenopathy and more extensive peripheral lymphadenopathy.

Resistance to apoptosis is a feature of CLL cells and many mechanisms has been proposed for this resistance. Recent experiments have described a subset of peripheral blood mononuclear cells in patients with CLL that differentiate into nurse like cells (NLC) in vitro and protect CLL cells from apoptosis.
When and how to Treat Chronic Lymphatic Leukemia

BCL-2 proto oncogene is elevated in 95% patients with B-cell. BCL-2 is unique among proto oncogenes a known suppressor of programmed cell death (apoptosis). Micro RNAs (Mi RNA s) are a novel class of small noncoding RNAs that modulate the expression of genes at a post transcriptional level and are involved in cellular apoptosis and cell metabolism. Over expression of two micro RNAs (MiR-21 and MiR-155) was seen in most of patients of CLL in one study.

CLINICAL AND LABORATORY FEATURES:

Most of the patients are older than 60 years at diagnosis with more than 90% over 50 years. The diagnosis if CLL is often incidental, routine blood counts may reveal an elevated absolute Lymphocyte count. In symptomatic patients fatigue and infections may be presenting features. A small percentage of patients may present with autoimmune hemolytic anemia (AIHA) or auto immune thrombocytopenia (AIT). Physical examination may reveal cervical, axillary and or inguinal Lymphadenopathy. Splenomegaly and Hepatomegaly are not uncommon. Lymphadenopathy spontaneously wax and wane but do not altogether disappear.

Laboratory findings invariably show Lymphocytes. The absolute Lymphocyte count can range from 5 X 10^9/L to 500 X 10^9/L. CLL cells resemble mature lymphocytes they have dense chromatin as well as scant cytoplasm and lack nucleoli. Smear preparation from peripheral blood may damage these fragile lymphocytes and produce ‘smudge’ cells.

Prolymphocytes < 55% can be seen in blood and peripheral smear of patients with CLL. Bone marrow involvement can be nodular or diffuse. Erythroid, Myeloid and Megakaryocyte precursors may be normal or decreased. Anemia and thrombocytopenia can be due to marrow infiltration or from immune destruction.

Findings of microspherocytes in peripheral blood smear and demonstration of IgG and or complement on red cells support diagnosis of immune hemolytic anemia. Pure red cell aplasia has been described in 1 - 6 % of cases. Patients can often develop hypogammaglobulinemia which can progress in severity with advancing disease. Other laboratory abnormalities include elevated β2 microglobulin (β2 - m), LDh is rarely elevated.

DIAGNOSIS:

Diagnostic criteria as proposed by National Cancer Institute (NCI) - sponsored, CLL working Group (NCIWG) require an Absolute Lymphocyte count of 5 X 10^9/L with lymphocytes coexpressing CDS and B cell marker (CD19, CD20, CD 23) and bone marrow lymphocytes of ≥ 30%. The revised NCIWG criteria allow for atypical cells such as prolymphocytes to account for upto 55% of cells. The presence of more than 55% prolymphocytes establishes a diagnosis of prolymphocytic leukemia (PLL) or indicates progression to PLL.

Clinical, morphological, immunophenotype and cytogenetic methods help to distinguish between B-cell and other lymphoproliferative diseases such as prolymphocytic leukemia, leukemic phase of non-Hodgkin’s lymphoma, Mantle cell Lymphoma, hairy cell leukemia, Marginal zone lymphoma and waldenstrom’s macroglobulinemia.

STAGING AND PROGNOSIS:

The well defined staging system for CLL includes Rai's and Binet Staging.

<table>
<thead>
<tr>
<th>Stage</th>
<th>Description</th>
<th>Binet</th>
<th>Median survival</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>Low risk</td>
<td>A</td>
<td>&gt; 10 Years</td>
</tr>
<tr>
<td>1</td>
<td>Intermediate risk</td>
<td>B</td>
<td>5-7 Years</td>
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<tr>
<td>2</td>
<td>Intermediate risk</td>
<td>C</td>
<td>2-3 Years</td>
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<tr>
<td>3</td>
<td>High risk</td>
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<td></td>
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<tr>
<td>4</td>
<td>High risk</td>
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The original Rai’s classification defined five stages from 0 to 4, this has been modified to three stages by defining Rai stage 0 as low risk, joining stage 1 to 2 to form intermediate risk group and stage 3 to 4 to form high risk group. Both CLL staging system confer significant prognostic information, however they are limited by their inability to identify which patient with early stage disease will develop disease progression.

A lymphocyte doubling time of > 12 months, Rai stage 0 disease, no diffuse bone marrow pattern, hemoglobin ≥ 13gm/dl and Absolute Lymphocyte count < 30 X 10^9/L define a group of smoldering CLL with an excellent prognosis. Women fare better than man independent of age and stage. Several serum factors have been identified as prognostic indicators in early CLL. Elevated serum β2 microglobulin level is an adverse prognostic factor. High serum LDH indicates poor prognosis. IgV gene mutation indicates a good prognosis. Expression of CD38 on surface of CLL cells has been linked to poor prognosis. ZAP-70 was found correlates inversely with IgV gene mutation and expression is associated with shorter time to progression and worse survival. Deletion 13q chromosome and normal karyotype confers better prognosis; while 17p and 11q deletion indicate worse prognosis.

TREATMENT:

An unusual feature of CLL compared to other leukemia’s is that making the diagnosis is not necessarily an indication to initiate the treatment. This is true for several reasons, CLL is a disease of the older population, it may be diagnosed in an asymptomatic patient and has a prolonged course, it is not curable with current treatment approaches and survival advantage was not demonstrated with early intervention trials.

Majority of patients are older than 70 years and have significant
co-morbid conditions associated with aging and may have indolent course, a significant fraction of patients will die of other causes and may never require therapy for CLL. Several European groups conducted clinical trials in 1980’s to evaluate whether immediate treatment in patients with early stage disease could improve survival. These large randomized trials of immediate therapy with chlorambucil versus wait and watch approach, were consistent in showing no survival benefit. Nevertheless, given the significantly better therapies available in 21st century this question has been raised again. The discovery of prognostic factors that can identify early stage disease patients with the likelihood of development of progressive disease may now allow such randomized trials to be conducted.

In the past we had only clinical parameters in order to evaluate the biology of the disease and to find out whether a treatment was necessary or not. E.g.: doubling time of lymphocytes, presence of B symptoms, anemia and or thrombocytopenia, hemolytic episodes, increased tendency to infections etc.

Currently we are more and more often using also biological parameters in order to evaluate the degree of aggressiveness of the disease. These parameters are chromosomal abnormalities, presence of IgV gene mutation, expression of certain surface antigens on lymphocytes in particular CD38 and ZAP-70. In this situation it is very important to understand whether these new parameters should be used routinely and what role they are currently playing and will probably play in future.

**Indications for treatment in CLL (NCIWG Guidelines)**

Active disease should be confirmed prior to initiating treatment.

1. A minimum of any one of the disease related symptoms must be present:
   a. weight loss ≥10% with in previous 6 months
   b. Extreme fatigue (i.e., ECOG PS 2 or worse; cannot work or unable to perform usual activities.
   c. Fevers greater than 100.5° F for ≥ 2 weeks without evidence of infection
   d. Night sweats without evidence of infection
2. Evidence of progressive marrow failure as manifest by the development of worsening of anemia and or thrombocytopenia.
3. Autoimmune anemia and /or thrombocytopenia poorly responsive to corticosteroid therapy
4. Massive (i.e., > 6 cm below the left costal margin) or progressive splenomegaly
5. Massive nodes or clusters (i.e., 10cm in longest diameter) or progressive lymphadenopathy
6. Progressive lymphocytosis with an increase of > 50% over a 2-month period or an anticipated doubling time of less than 6 months

Source: cheson et al.

Prior to initiation of active therapy for patients with symptomatic disease, patients with CLL should undergo pretreatment evaluation to determine the extent of disease, patient performance status and assessment of co-morbidities that are likely to impact upon treatment options. There is no agreed upon standard treatment regimen for symptomatic CLL. As such there are several initial treatment options for these patients.

Treatment options include purine analogs e.g. fludarabine, alkylating agents (e.g.: chloroambucil, bendamustine), monoclonal antibodies (e.g. Rituximab, alemtuzumab or combinations of these agents). When prospective randomized trials have been performed most of these regimens have been compared with chlorambucil, which has been a standard of care for decades prior to the advent of newer agents. While overall survival rates with these different regimens are similar, they differ in their rates of complete remission (CR), time to progression and associated toxicities. A choice between these therapies is made based upon patient characteristics and goals of therapy. Median over all survival is approximately five years.

Fludarabine 25mg/m² on days 1-5 every 28 weeks produces a higher response rates as compared to chlorambucil or a combination of cyclophosphamide, adriamycin and prednisolone, response rate of 70% and progression free survival of 27 months with fludarabine. A combination of fludarabine 25mg/m² on days 1-3 and cyclophosphamide 300mg / m² on day 1-3 offers good result with a response rated of 80% - 90%. Addition of rituximab to this combination increase the response rate to 90% with 47% complete response. Prednisolone has no role in the treatment of CLL except for autoimmune manifestations. The goals and duration of therapy in CLL is poorly defined and there is no evidence that intensification or maintenance therapy is of benefit.

While most of patients with symptomatic or advanced CLL are treated similarly, older patients and patients with Del (17p) or Del (11q) require particular considerations. In patients who relapse on alkylating agents, fludarabine produces a partial response of 40%, Cladurabine, Pentostatin, Bendamustin, Alemtuzumab (anti CD 52 antibody) are indicated for relapse and refractory disease and show variable response. Splectectomy is indicated for hypersplenism or massive splenomegaly. Patients with CLL are usually not fit for allogenic transplant because of their advanced age at presentation. Immunoglobulin can be used for treatment of recurrent infections with hypogammaglobulinemia. The dose is 400mg/m² every 3 weeks for one year.

The National cancer institute working group classified responses as complete response, partial response and stable disease.

**Complete response** - no evidence of clinical disease for more than two months.

**Partial response** - 50% or greater reduction in peripheral blood
lymphocytes and adenopathy and or hepatosplenomegaly.

**Stable disease** – patients who fail to meet the criteria for partial response but do not show evidence of progression.

**HISTOLOGIC TRANSFORMATION**

In a variable percent of patients with CLL and usually as a terminal event, CLL transforms into another lymphoproliferative disorder. The following are the most common reported transformations, prolymphocytic leukemia – 10%, aggressive lymphoma (Richter’s transformation) – 3%

**RICHTER’S SYNDROME**

The term Richter’s syndrome refers to the development of aggressive large cell lymphoma during the course of CLL. Richter’s syndrome is usually associated with worsening of systemic symptoms including B symptoms, elevated LDH, rapid tumor growth. Diagnosis requires tissue biopsy. This disease is usually resistant to therapy and median survival of patients is approximately 6 months.

**CONCLUSIONS**

Considerable progress has been made in the understanding of biology of CLL and developing new and effective treatments. Long term remissions are now possible in a subset of patients. The outlook for CLL patients has improved and ongoing research promises even more progress.

**REFERENCES**

3. Hematology 2003 – Michael J. Keating M.D, Do we have the tools to cure CLL? Page 165-175
5. The M.D Anderson Manual of Medical Oncology – Hagop M. kantarjian, Robert A. Wolff, Charles. A. koller