Gracia Dayton. *Birch Garden.* Watercolor on paper, 22″ × 30″.

Newer prognostic markers assist management choices for both indolent and aggressive forms of chronic lymphocytic leukemia.

Front-Line Therapy for Chronic Lymphocytic Leukemia

Sheetal Desai, PharmD, and Javier Pinilla-Ibarz, MD, PhD

Background: *Historically, alkylator-based therapy has been used to treat patients with chronic lymphocytic leukemia (CLL). More effective therapies, such as the use of monoclonal antibodies in combination with chemotherapy, have been shown to prolong both progression-free survival and overall survival. Improvements in the identification of prognostic markers for CLL, as well as novel combinations for chemoimmunotherapy regimens, have improved the outcome for patients with CLL.*

Methods: *We examine the diagnosis of CLL, the role of prognostic factors in determining treatment goals, and current data on front-line management of CLL.*

Results*: The benefits of single-agent and combination therapies are associated with prolonged progression-free and overall survival. While more aggressive management may therefore be warranted, each patient's comorbidities and performance status must be weighed against the benefits, availability, cost, treatment goals, and incidence of adverse effects associated with each therapy.*

Conclusions: *New single agents and novel treatment combinations have shown promising results in phase I/II studies. The ultimate therapeutic goals of prolonged survival and improved quality of life will be validated only by ongoing clinical and laboratory research and by continuous enrollment of patients in clinical trials.*

Introduction

Chronic lymphocytic leukemia (CLL) is a clinically heterogeneous disease originating from B lymphocytes that may differ in activation, maturation state, or cellular subgroup. CLL is the most common form of adult leukemia in Western countries; almost 15,000 men and women were diagnosed with this B-cell malignancy in the United States in 2010, resulting in more than 4,000 deaths.[1] An apparent decrease in incidence, from 3.5 per 100,000 population in 1973 to 2.3 per 100,000 in 1990, may reflect progress in our ability to distinguish CLL from other chronic leukemias. CLL is primarily a disease of the elderly, with the median age of 72 years at diagnosis. CLL occurs twice as often in males as in females, and it is most common in the Caucasian population.[2]

No etiologic factor has been identified for CLL, although a few carefully conducted studies have been performed. CLL is one of the few leukemias that does not appear to be associated with prior exposure to ionizing radiation, chemicals, or drugs, and it is the only leukemia where no hard evidence has been found to correlate an association with atomic bomb explosions.[3,4] Approximately 20% of patients with this disease have relatives with CLL or another lymphoid malignancy, although no genetic linkage has been identified.[5]

From the Departments of Pharmacy (SD) and Malignant Hematology (JP-I) at the H. Lee Moffitt Cancer Center & Research Institute, Tampa, Florida.

Submitted January 13, 2011; accepted March 24, 2011.

Address correspondence to Sheetal Desai, PharmD, Department of Pharmacy, Moffitt Cancer Center, 12902 Magnolia Drive, Tampa, FL 33612. E-mail: Sheetal.Desai@moffitt.org

Dr Desai reports no significant relationship with the companies/organizations whose products or services may be referenced in this article. Dr Pinilla receives grants/research support from GlaxoSmithKline Corp and Genentech Inc, and also receives honoraria from GlaxoSmithKline Corp, Genentech Inc, and Cephalon Inc.

Table 1. — Rai and Binet Staging Systems for Chronic Lymphocytic Leukemia

System	Stage	Risk	Definition
Rai[8]	0	Low	Lymphocytosis
	I	Intermediate	Lymphocytosis and lymphadenopathy
	II	Intermediate	Lymphocytosis, lymphadenopathy, and splenomegaly and/or hepatomegaly
	III	High	Lymphocytosis and anemia (hemoglobin < 10 g/dL)
	IV	High	Lymphocytosis and thrombocytopenia (platelets < 100,000/mm^3)
Binet[9]	A	N/A	Lymphocytosis with enlargement of ≤ 2 lymphoid areas, no anemia or thrombocytopenia
	B	N/A	Lymphocytosis with enlargement of ≥ 3 lymphoid areas
	C	N/A	Lymphocytosis with anemia (hemoglobin > 10 g/dL) and/or thrombocytopenia (platelets < 100,000/mm^3)

The modified Rai staging system now defines low-risk disease as patients with lymphocytosis with leukemia cells in the blood and/or marrow (formerly stage 0). Intermediate-risk disease includes patients with lymphocytosis, enlarged nodes in any site, and splenomegaly and/or hepatomegaly (formerly stage I or II). High-risk disease includes patients with disease-related anemia (formerly stage III) or thrombocytopenia (formerly stage IV).[7]

Historically, CLL was considered to be an indolent and incurable disease. Nonetheless, the clinical course is highly variable. Some patients survive for decades, whereas others develop aggressive disease and die within 2 to 3 years of diagnosis.[2,5]

For more than half a century, alkylator-based therapy such as chlorambucil or cyclophosphamide has been used to treat patients with CLL. More effective therapies, including the use of monoclonal antibodies in combination with chemotherapy, have been available since the late 1990s, and these have been shown to prolong both progression-free survival (PFS) and overall survival (OS). More aggressive management may therefore be warranted for patients with early-stage disease. However, since more than two-thirds of CLL patients are older than 65 years, treatment needs to be tailored to each patient's fitness level, and the presence of comorbidities needs to be taken into account when treatment decisions are being contemplated.[6]

This review examines the diagnosis of CLL, the role of prognostic factors in determining treatment goals, and current data on front-line management of CLL.

Clinical Presentation, Diagnosis, Staging, and Risk Stratification

Patients with CLL are generally asymptomatic at presentation, and the diagnosis is often made incidentally when lymphocytosis is noted at the time of a routine evaluation. Workup includes a complete blood count, peripheral blood smear examination, and physical examination with attention to node-bearing areas, including sizes of liver and spleen. Bone marrow biopsy is not required for the diagnosis.[7] CLL cells arise from polyclonal expansion of CD5+ B lymphocytes, which are transformed into a monoclonal population by mutational agents. Neoplastic CD5+ cells accumulate in the lymph nodes and spleen due to the loss of apoptosis, by either the overexpression of an oncogene or the loss of a tumor suppressor gene. The presence of B-cell lymphocytosis of at least 5×10^9/L for 6 months or longer is diagnostic for CLL. An examination of the peripheral blood demonstrates monomorphic, small, round B lymphocytes with a narrow border of cytoplasm and a dense nucleus. Immunophenotyping of CLL cells shows expression of CD5, CD19, and CD23, as well as dim expression of CD20 and CD79b.[7]

The clinical staging of CLL is based on physical examination and complete blood counts alone. The two widely used staging systems are the Rai[8] (used primarily in the United States) and the Binet[9] (used in Europe). The value of each system lies mainly in its prognostic implications for survival. Staging does not identify who may have indolent or progressive disease, and it does not predict response to treatment. With both staging systems, patients with the most advanced stage have a predicted survival time of 1 to 2 years, while patients with the lowest stage of disease have a median survival time of more than 10 years (Table 1).[8,9]

Döhner et al[10] determined that chromosomal abnormalities may be identified in more than 80% of CLL cases. These cytogenetic abnormalities can be identified by fluorescence in situ hybridization (FISH) analysis and may be the most meaningful predictor of disease progression at this time (Table 2). It is important to note that the aberrations are not stable, and it is imperative to analyze the whole panel of FISH markers on repeat testing before later treatment decisions are made.[10]

In addition to clinical staging, traditional prognostic factors for stratifying the risk of disease progression have included high serum levels of β_2-microglobulin and soluble CD23, short lymphocyte doubling time (< 6 months), and diffuse bone marrow infiltration. The mutational status of immunoglobulin heavy chain variable region (IgVH) genes and the expression of ZAP70 and CD38 are other predictors of disease progression that have been identified recently, but these should not be used to individualize treatment selection outside of a clinical trial at this time.[11] Currently, no evidence has

shown that asymptomatic patients with high-risk disease features benefit from early treatment. The only clear exception is symptomatic patients with 17p deletions or p53 mutations since this may warrant therapy with agents that act independently of p53.[10]

Goals of Therapy

Historically, the goal of treatment for CLL patients has been palliation of symptoms, and treatment was usually continued until disease-related symptoms were resolved. In 2008, the International Workshop on Chronic Lymphocytic Leukemia[7] affirmed the guidelines established by the 1996 National Cancer Institute Working Group for initiation of treatment for CLL. These indications include autoimmune cytopenias, progressive bone marrow failure, rapid lymphocyte doubling time, progressive splenomegaly and/or lymphadenopathy, and constitutional symptoms related to CLL, including fatigue, fever ≥ 100.5ºF for ≥ 2 weeks, ≥ 10% weight loss in the preceding 6 months, and night sweats (Table 3).[7] More than 80% of patients who are symptomatic will also have lymphadenopathy and, less commonly, splenomegaly and/or hepatomegaly.

The Cancer and Leukemia Group B (CALGB) 10501 trial is currently studying whether early intervention affects long-term outcome for patients with high-risk features, but currently no data are available to support early therapy for high-risk CLL patients who do not meet established criteria for treatment. In addition, CLL7, a randomized study of the German and French CLL study groups, has recently completed recruitment of patients to determine whether early treatment with chemoimmunotherapy would be beneficial for patients with high-risk features.

Table 2. — Incidence of Chromosomal Abnormalities in 325 Patients With Chronic Lymphocytic Leukemia

Aberration	No. of Patients[a]
13q deletion	178 (55%)
11q deletion	58 (18%)
12q trisomy	53 (16%)
17p deletion	23 (7%)
6q deletion	21 (6%)
8q trisomy	16 (5%)
t(14q32)	12 (4%)
3q trisomy	9 (3%)
Clonal abnormalities	268 (82%)
Normal karyotype	57 (18%)

[a] One hundred seventy-five patients had 1 aberration, 67 had 2 aberrations, and 26 had more than 2 aberrations.

Döhner H, Stilgenbauer S, Benner A, et al. Genomic aberrations and survival in chronic lymphocytic leukemia. *N Engl J Med.* 2000;343(26): 1910-1916.

Table 3. — Indications for Initiation of Treatment in Patients With CLL

Progressive marrow failure
Autoimmune cytopenias
Systemic symptoms: fatigue, night sweats, weight loss, fever
Massive splenomegaly (ie, > 6 cm below the left costal margin)
Massive lymphadenopathy (> 10 cm)
Lymphocyte doubling time < 6 months (or > 50% rise in lymphocyte count within 2 months)

Based on the National Cancer Institute Working Group 1996 guidelines, updated in 2008 by the International Workshop on Chronic Lymphocytic Leukemia.[7]

Single-Agent Chemotherapy

Chlorambucil

Chlorambucil (Leukeran), an alkylating agent, has been the gold standard of treatment for CLL patients for more than 40 years. It is rapidly absorbed from the gastrointestinal tract; peak plasma concentrations occur within 1 hour after ingestion. The dosage and schedule of administration vary greatly, but the two most commonly used approaches are low-dose continuous therapy (0.08 mg/kg per day) every 4 to 8 weeks and pulsed intermittent therapy (0.4 to 1.0 mg/kg) every 3 to 4 weeks until there is a response. Responses are attained in approximately 30% to 70% of previously untreated patients, although few of these are complete responses (CRs).[12] With the advent of newer agents, chlorambucil has fallen out of favor in the United States. However, results of the German CLL Study Group (GCLLSG) CLL5 trial suggest the drug still has a role — particularly in elderly patients with decreased performance status — due to its oral availability, minimal cost, and low incidence of adverse effects.[13]

Fludarabine

Fludarabine (Fludara) is the most extensively studied purine analog for the treatment of patients with CLL. Several prospective randomized phase III trials comparing single-agent analogs (eg, fludarabine, cladribine, and pentostatin) with alkylating agents (eg, chlorambucil) have demonstrated significantly superior overall response (OR) and CR rates for previously untreated patients treated with fludarabine.[13,14] However, fludarabine did not have a statistically significant effect on OS time in any of these trials, including the recent long-term follow-up CLL5 trial of the GLLSG[13] that compared fludarabine with chlorambucil as single-agent treatment for elderly patients (Table 4).[13,14]

Single-agent fludarabine treatment has also proven to be more advantageous than combination regimens

Table 4. — Randomized Trials Comparing Single-Agent Fludarabine With Single-Agent Chlorambucil

Reference	Regimen	No. of Patients	Complete Remission	Overall Response	Median Progression-Free Survival
Rai et al[14]	Fludarabine Chlorambucil	179 193	20% 4%	63% 37%	20 mos 14 mos
Eichhorst et al[13]	Fludarabine Chlorambucil	93 100	7% 0	72% 51%	19 mos 18 mos

such as cyclophosphamide, doxorubicin, vincristine, and prednisone (CHOP) and cyclophosphamide, doxorubicin, and prednisone (CAP) for inducing CR rates in the range of 7% to 40%, but it has not been demonstrated to improve survival.[15]

Bendamustine

Bendamustine (Treanda) is a novel bifunctional alkylating agent that contains a benzimidazole ring and has structural similarities to both alkylating agents and purine analogs. It is a nonspecific cytostatic drug that causes single- and double-stranded DNA breaks. In addition, it activates many proapoptotic genes, thereby restoring p53-dependent tumor suppressor function and causing a strong activation of intrinsic apoptosis. Bendamustine differs from other alkylating agents by its capacity to activate a DNA-damage stress response and apoptosis, inhibit mitotic checkpoints, induce mitotic catastrophe, and activate a base-excision DNA-repair pathway rather than an alkyltransferase DNA-repair mechanism. Also, bendamustine can downregulate genes important in mitotic checkpoint regulation. The unique structure of bendamustine and these observations of function may explain its potential clinical activity in CLL patients with resistance to alkylating agent-based therapy.[16]

Bendamustine has been used for more than 30 years in Germany, where it was originally synthesized in the 1960s, but it was only recently (2008) approved by the US Food and Drug Administration (FDA) for treating CLL patients in the United States. Its approval was based on results of a multicenter European phase III trial in which 319 previously untreated patients with CLL were randomized to one of two treatment groups: oral chlorambucil (0.8 mg/kg on days 1 and 15, every 28 days) or bendamustine (100 mg/m^2 on days 1 and 2, every 28 days). Treatment was continued for 6 cycles or until disease progression. Results of the study demonstrated that bendamustine had a superior outcome, with OR and CR rates of 67% and 31%, respectively, vs OR and CR rates of 30% and 2%, respectively, in the chlorambucil group. It is important to note that the dosage and number of cycles of chlorambucil administered were less than optimal, which may account for the lower OR rate than that previously reported in other studies. Median PFS was significantly extended with bendamustine (22 months with bendamustine vs 8 months with chlorambucil). However, OS was not changed by bendamustine. Nausea and vomiting were only modest with this agent, and there was no alopecia. The incidence of grade 3/4 infection was similar in both groups (8% with bendamustine and 3% with chlorambucil).[17]

Combination Chemotherapy

Fludarabine Combinations

In an attempt to achieve synergism and increase response rates, purine analogs and alkylating agents have been studied in combination due to their different mechanisms of action and toxicity profiles. Fludarabine plus cyclophosphamide (FC) is the most thoroughly studied chemotherapy combination for CLL. Three large prospective, randomized, multicenter studies comparing FC to fludarabine monotherapy have shown that the combination improves OR, CR, and PFS (Table 5).[18-20] However, OS was not significantly increased by the combination therapy in any of these studies, with one exception: the GCLLSG CLL4 trial demonstrated improved OS among the combination-treated non–high-risk patients (ie, those not exhibiting a del(17p) or a p53 mutation). Of note, FC caused more grade 3/4 neutropenias but did not increase the rate of severe infections.

Cladribine Combinations

Cladribine (Leustatin) is another purine analog that has been investigated for possible effectiveness as a first-line therapy for CLL patients. In one multicenter trial, cladribine plus prednisone was compared with chlorambucil plus prednisone.[21] The cladribine group had a significantly higher CR rate than the chlorambucil group had (47% vs 12%, respectively) and a significantly longer PFS. However, as was the case with fludarabine, survival was not extended by treatment with the purine analog. A large randomized Polish trial was undertaken to assess whether cladribine combinations would offer any advantage over fludarabine combination therapy. This study compared single-agent cladribine with cladribine plus cyclophosphamide and with cyclophosphamide plus mitoxantrone in 479 previously untreated patients with progressive CLL.[22] The cyclophosphamide combination did not produce any benefit in terms of PFS or response rates when compared with cladribine alone. In

fact, none of the treatment groups showed a difference in OR, PFS, or OS. Grade 3/4 neutropenia and infections occurred more frequently in the combination arms than in the single-agent cladribine arm.[22] Hence, when used as first-line treatment for CLL, cladribine combination therapies do not seem to offer any advantage.

Monoclonal Antibodies

CLL B cells express surface antigens CD20 and CD52 and thus are potential targets for therapy using the monoclonal antibodies rituximab and alemtuzumab.

Rituximab

Rituximab (Rituxan) is a chimeric murine/human monoclonal antibody directed specifically at CD20, but it has only modest activity in previously treated patients with CLL unless very high doses are administered. This is thought to be a result of the low density of CD20 on CLL cells. In addition, soluble CD20 has been observed in the plasma of patients with CLL, which might interfere with the binding of rituximab to the cancer cells and result in rapid clearance and negative pharmacokinetic effects.[23-25] When tested in a small trial as a front-line single-agent therapy for previously untreated patients with CLL, rituximab resulted in a modest OR rate of 51% after a front-line course, with the majority being only PRs.[26] In contrast, combinations of rituximab with chemotherapy have proven to be efficacious therapies for CLL, as discussed below.

Alemtuzumab

Alemtuzumab (Campath), the anti-CD52 fully humanized monoclonal antibody, was recently approved for use in previously untreated CLL patients. Unlike rituximab, alemtuzumab has been shown to induce cell death in vitro in CLL cells through a mechanism that is independent of p53 status and caspase activation.[27] Several clinical trials have also demonstrated that it is effective, particularly in patients with high-risk cytogenetics such as del(17p) and del(11q).[28-31]

Alemtuzumab administered subcutaneously demonstrates decreased side effects, particularly infusion-related reactions, without altering efficacy rates. Lundin et al[32] treated 41 CLL patients with subcutaneous injections of alemtuzumab as first-line therapy for up to 18 weeks. The OR and CR rates were 87% and 19%, respectively, and the median time to treatment failure was greater than 18 months (the actual median time had not yet been reached at the time of this report). The treatment was generally well tolerated, with adverse events mainly consisting of local injection site reactions. Cytomegalovirus (CMV) reactivation occurred in 10% of patients. These promising results spurred investigators to undertake a phase III randomized trial comparing alemtuzumab with chlorambucil as first-line therapy in 297 patients with treatment-naive progressive CLL.[29] Patients were randomized to receive either intravenous alemtuzumab (30 mg 3 times weekly for up to 12 weeks) or oral chlorambucil (40 mg/m^2 every 4 weeks for up to 12 cycles). This study demonstrated superior response rates for alemtuzumab compared with chlorambucil (OR rates of 83% vs 56% and CR rates of 24% vs 2%). Median PFS time was also increased in the alemtuzumab arm, with a 42% reduction in risk of progression or death. No difference was noted between the two arms in terms of grade 3/4 hematologic toxicities. However, 52% of patients in the alemtuzumab

Table 5. — Recent Randomized Trials Comparing Fludarabine Plus Cyclophosphamide With Single-Agent Fludarabine

Trial Group	Regimen	No. of Patients	Complete Response	Overall Response	Median Progression-Free Survival
GCLLSG[a]	Fludarabine	182	7%	83%	20 mos
	Fludarabine + cyclophosphamide	180	24%	94%	48 mos
ECOG[b]	Fludarabine	137	5%	60%	19 mos
	Fludarabine + cyclophosphamide	141	23%	74%	32 mos
LRF CLL4[c]	Chlorambucil	387	7%	72%	20 mos
	Fludarabine	194	15%	80%	23 mos
	Fludarabine + cyclophosphamide	196	38%	94%	43 mos

[a] The German CLL Study Group (GCLLSG) randomized 375 patients to fludarabine 25 mg/m^2 IV daily for 5 days, vs fludarabine 30 mg/m^2 IV daily for 3 days plus cyclophosphamide 250 mg/m^2 IV daily for 3 days, every 28 days for 6 cycles. Thirteen patients were excluded because of violations of inclusion criteria (4 patients due to wrong diagnoses, 3 due to missing consent forms, and 6 due to concomitant disease). Eleven patients were lost to follow-up. Survival data were available in 351 patients, response data in 328 patients, and toxicity data in 346 patients.[18]

[b] ECOG randomized 278 patients to fludarabine 25 mg/m^2 IV daily for 5 days vs fludarabine 20 mg/m^2 IV daily for 5 days plus cyclophosphamide 600 mg/m^2 IV on day 1, every 28 days for 6 cycles.[19]

[c] LRF randomized 777 patients to chlorambucil 10 mg/m^2 orally daily for 7 days, every 28 days up to 12 cycles or until maximal response, vs fludarabine 25 mg/m^2 IV daily for 5 days or 40 mg/m^2 orally daily for 5 days, vs fludarabine 25 mg/m^2 IV daily plus cyclophosphamide 250 mg/m^2 IV daily for 5 days, every 28 days for 6 cycles.[20]

arm developed CMV reactivation compared with only 2% in the chlorambucil arm.[29] It is important to note that alemtuzumab efficacy in the first-line setting has been established only relative to chlorambucil. For this reason, and because of the immune suppression associated with its use, alemtuzumab is not generally utilized as single front-line therapy for CLL patients. Furthermore, chemoimmunotherapy has provided improved results.

Chemoimmunotherapy

Fludarabine/Rituximab

Since in vitro data showed evidence for a synergy between rituximab and fludarabine, several phase II trials have investigated combinations of rituximab and fludarabine. In the first such trial, Schulz et al[33] examined the safety and efficacy of fludarabine plus rituximab for 31 previously treated or untreated patients with CLL. The OR rate was 87%, and the CR rate was 32%. The median duration of response was 75 weeks.

The CALGB 9712 randomized phase II trial involved 104 previously untreated patients with CLL.[34] The trial compared combinations of fludarabine and rituximab administered either concurrently or sequentially. The patients all received 6 cycles of fludarabine (25 mg/m^2 intravenously per day on days 1–5, every 28 days). In the concurrent treatment arm, rituximab was also administered (375 mg/m^2 per day on days 1 and 4 of cycle 1 and on day 1 only of cycles 2, 3, 4, and 5). Two months after the last fludarabine cycle, patients then received 4 additional weekly doses of rituximab. In the sequential treatment arm, rituximab was administered only as the 4 weekly doses, beginning 2 months after the fludarabine cycle 6. After a median follow-up time of 23 months, the OR and CR rates were higher in the concurrent arm (90% and 47%, respectively) than in the sequential arm (77% and 28%, respectively). The median PFS and OS had not yet been reached. All 104 patients in this CALGB 9712 trial were compared retrospectively with 178 patients in the CALGB 9011 trial who received only fludarabine. The inclusion criteria of the two trials were identical. The patients in the fludarabine/rituximab arm had higher OR and CR rates (84% and 38%, respectively) than the patients receiving fludarabine alone (63% and 20%, respectively).[35]

Fludarabine/Cyclophosphamide/Rituximab

Keating et al[36] performed a single-institution phase II trial adding rituximab to fludarabine and cyclophosphamide (FCR) in 300 patients with previously untreated CLL. FCR resulted in an OR rate of 95%, with a CR in 72%; the 6-year OS and failure-free survival rates were 77% and 51%, respectively. Interestingly, the CR rate was significantly higher for patients with β_2-microglobulin of less than twice the upper limit of normal. Median time to progression was 80 months. Toxicity in this study included predominately cytopenias and associated infection. Eight patients developed treatment-related myelodysplasia. Due to the promising results demonstrated in this trial, the GCLLSG CLL8 undertook a prospective randomized trial involving 817 previously untreated CLL patients that compared FC with FCR. Patients received 6 cycles of fludarabine 25 mg/m^2 on days 1 to 3 and cyclophosphamide 250 mg/m^2 on days 1 to 3, with or without rituximab 500 mg/m^2 on day 1 (375 mg/m^2 on day 1 of cycle 1) repeated every 28 days. After a median observation time of 37.7 months, the OR rate was higher in the FCR group than in the FC group (95.1% vs 88.4%) and more CRs were reported (44.1% vs 21.8%). The median PFS at 2 years was 51.8 months in the FCR group and 32.8 months in the FC group. Superiority of OS was observed only in patients with Binet stage A and B disease. In addition, FCR did not improve the PFS or OS of patients with del(17p). However, those patients with del(11q) appeared to benefit from FC combination therapy, which induced a response rate in 100% of patients treated with this combination. FCR was more frequently associated with Common Toxicity Criteria (CTC) grade 3 and 4 neutropenia (34% vs 21%), but this did not translate into increased infections.[37] Major residual disease (MRD) negativity refers to the eradication of leukemic cells assessed by four-color flow cytometry or allele-specific oligonucleotide polymerase chain reaction. The importance of eradicating MRD was confirmed by Boettcher et al[38] in a trial (GCLLSG CLL8) demonstrating that median PFS depended on the ability to eradicate MRD in the peripheral blood. Clear-cut differences in median PFS were observed between patients demonstrating levels $< 10^{-4}$ (not reached), $\geq 10^{-4}$ and $< 10^{-2}$ (34 months) and $\geq 10^{-2}$ (15 months). Furthermore, 67% of patients receiving FCR achieved MRD $< 10^{-4}$ compared with only 34% of FC patients, thus accounting for the improved PFS with FCR. With the demonstration of significant improvement in PFS, in 2010 the FDA approved the use of FCR for treating previously treated or untreated patients with CD20+ CLL.[39]

With the goal of maintaining the high response rate previously achieved with FCR therapy while reducing the toxicity of that regimen (of particular concern due to the high proportion of elderly patients in the CLL population), Foon et al[40] undertook a study involving FCR therapy with lowered doses of fludarabine and chlorambucil (designated "FCR-Lite"). This study involved 50 previously untreated patients with CLL. The dose of fludarabine was reduced to 20 mg/m^2 per day on days 2–4, and cyclophosphamide was reduced to 150 mg/m^2 per day on days 2–4 during cycle 1 and on days 1–3 in cycles 2–5. Rituximab was dosed as 375 mg/m^2 on day 1 of cycle 1, then at 500 mg/m^2 on day 1 of cycles 2–5 and on day 14 of each cycle. Maintenance rituximab was administered every 12 weeks until progression. Measured according to the 2008 guidelines from the International Working Group on CLL, the CR rate was 77% and the OR rate was 100%. At a median follow-up 2.4 years, all

complete responders remained in CR except 1 patient who died of a myocardial infarction. Grade 3/4 neutropenia was documented in only 13% of the cycles,[40] which is substantially lower than that observed with the traditional FCR regimen.[36] Based on these results, this regimen might warrant further testing in larger trials.

Pentostatin/Cyclophosphamide/Rituximab

In an attempt to reduce myelotoxicity, investigators have substituted pentostatin for the fludarabine in the FCR regimen. Phase II trials studied the safety and effectiveness of pentostatin, cyclophosphamide, and rituximab (PCR) combinations administered to previously treated[41] or untreated[42] patients with CLL. It was concluded that the PCR combination had significant clinical activity with only modest toxicity.[41,42] As was observed with the fludarabine combination, patients with del(11q) had PFS results similar to those of patients without this aberration, again suggesting that cyclophosphamide may be an important addition for this subset of patients.[42]

Reynolds et al[43] conducted a multicenter, community-based phase III trial that compared PCR with FCR in 184 previously untreated or minimally treated CLL patients. The OR rates were similar in the two groups (45% in the PCR group vs 57.5% in the FCR group). However, the CR rate was significantly lower in the PCR group (7%) vs the FCR group (15%). Infection (temperature ≥ 101°F, with or without symptoms, requiring antibiotics) was the primary endpoint of the study, and there were no significant differences in infection rates between the two treatment arms (31% in FCR and 34% in PCR). Even though the study was not powered to show a statistically significant difference in OR rate between FCR and PCR, these findings indicated that results from academic centers may not necessarily be reproducible in the community, due primarily to reduction in the ability to complete planned treatment courses.

Cyclophosphamide/Fludarabine/Alemtuzumab/Rituximab

The cyclophosphamide/fludarabine/alemtuzumab/rituximab (CFAR) regimen has demonstrated activity in high-risk patients, particularly those with β_2-microglobulin levels of ≥ 4 mg/dL. Wierda et al[44] investigated the addition of alemtuzumab to FCR (CFAR). The CFAR regimen consisted of fludarabine 20 mg/m^2 on days 3–5; cyclophosphamide 200 mg/m^2 on days 3–5; rituximab 375 mg/m^2 on day 2 of cycle 1 or 500 mg/m^2 on day 2 of cycles 2–6; and alemtuzumab 30 mg intravenously on days 1, 3, and 5 every 28 days for up to 6 cycles. Patients received pegfilgrastim as well as prophylaxis for *Pneumocystis jirovecii* pneumonia (PJP) and CMV.

Results from a phase II study conducted by Parikh et al[45] in previously untreated patients demonstrated an OR rate of 92% and a CR rate of 70% for the whole population. Grade 3/4 neutropenia was reported to be 31%, an incidence that is comparable to that seen with high-risk patients treated with FCR. At the time of this report, the median time to progression was 38 months, and the median OS had not been reached (49+ months). In the subset of patients with the high-risk features of del(17p) and unmutated IgVH status, remarkable CR rates were observed (57% and 73%, respectively); however, these remissions were not durable. While the results do not appear to be superior to those achieved with FCR, further investigation in a multicenter trial is warranted.

Bendamustine/Rituximab

The combination of bendamustine and rituximab (BR) has recently been studied in a multicenter phase II trial by Fischer et al[46] of the GCLLSG. The trial included 117 previously untreated CLL patients. Bendamustine was administered (90 mg/m^2 on days 1 and 2 every 28 days for up to 6 cycles) plus rituximab (375 mg/m^2 on day 1 in cycle 1 and 500 mg/m^2 in cycles 2–6). The OR rate was 90.9% and the CR rate was 32.7%. After 18 months, the median PFS had not been reached and 75.8% of patients were still in remission. Although the CR rate was lower than would be expected with FCR, the toxicity profile was favorable, with grade 3/4 neutropenia and infection complications in only 6.5% and 5.1%, respectively, of all cycles. As a result of the substantial response rates, the ability of this combination to eradicate detectable MRD, and the modest toxicity observed in this trial, the GCLLSG is currently conducting a phase III study (CLL10) comparing BR with FCR in previously untreated CLL patients. Since the major advantage of the BR combination is reduced toxicity, this regimen may be particularly suitable for treatment of elderly patients or those with multiple comorbidities.

High-Dose Methylprednisolone Plus Rituximab

In a recent phase II trial conducted by Castro et al[47] that involved 28 previously untreated CLL patients, a high dose of methylprednisolone was administered together with rituximab. Methylprednisolone (1,000 mg/m^2 per day) was given on days 1–3 every 4 weeks for 3 cycles, and rituximab (total dose 4,500 to 6,750 mg/m^2) was administered weekly. At a follow-up of more than 3 years, the OR and CR rates were 96% and 32%, respectively. The patients more likely to respond were those who had lower β_2-microglobulin levels and those without splenomegaly. Hematologic toxicity was minimal, with the majority (80%) of all adverse events limited to grade 1/2 in severity. Fever and neutropenia were noted, but there was no episode of sepsis. These results may be attributable to (1) the limiting of corticosteroid dose to 3 days compared with a 5-day dose used in previous studies of this combination, (2) the use of prophylactic antibiotics throughout the treatment and for 2 months afterward, and (3) the fact that this was first-line therapy for these patients.[47]

Cladribine and Rituximab

Bertazzoni et al[48] recently conducted a trial with previously treated (n = 16) and untreated (n = 27) CLL patients to assess the efficacy of combined treatment with rituximab and cladribine. Patients received rituximab (375 mg/m^2 intravenously on day 1) and cladribine (0.1 mg/kg subcutaneously on days 2–6) every 28 days for 4 cycles, with a median follow-up of 2 years. In the previously untreated subset of patients, the CR rate was 54% and median time to treatment failure was 40.7 months. The treatment was fairly well tolerated, with 2 patients developing grade 4 neutropenia. The subcutaneous route of cladribine administration did not require hospitalization, and the absence of any local reaction made this a safe and easily performed regimen in the outpatient setting. However, due to the small numbers of this study, further investigation is warranted before routinely applying this combination to clinical practice.

Newer Agents of Interest

Ofatumumab

Ofatumumab (Arzerra) is a humanized monoclonal antibody directed at CD20. It appears to have greater potency in complement-dependent cellular cytotoxicity (CDC) than rituximab, as well as a slower off-rate and more stable CD20 binding. Furthermore, it appears to bind to a unique epitope of CD20 that is different from the one bound by rituximab.[28] There are no current studies evaluating its use as a single agent in previously untreated CLL patients. However, combination studies are being conducted to enhance the therapeutic efficacy of ofatumumab. Wierda et al[49] recently conducted a phase II trial evaluating the combination of ofatumumab with fludarabine and cyclophosphamide (O-FC). Sixty-one previously untreated patients received either 500 mg or 1,000 mg of ofatumumab on day 1, followed by fludarabine (25 mg/m^2) and cyclophosphamide (250 mg/m^2) on days 1–3 every 4 weeks for 6 cycles. The CR rates were 32% in the 500-mg arm vs 50% in the 1,000-mg arm. The OR rates were 77% vs 73% in the 500-mg arm vs 1,000-mg arm. The most common grade 3/4 toxicities were infections, neutropenia, anemia, and thrombocytopenia.

Clinical trials to determine the efficacy of ofatumumab for previously untreated CLL patients, in chemoimmunotherapy combinations comparable to those explored with rituximab, are ongoing. In addition, phase III trials will be required to prove the superiority of ofatumumab to rituximab in clinical practice.

Lenalidomide

Lenalidomide (Revlimid), an analog of thalidomide, is an immunomodulatory agent that is approved by the FDA for use in multiple myeloma and del(5q) myelodysplastic syndrome. The exact mechanism of action is unknown, but it is thought to be related to its inhibitory effects on angiogenesis and signal transduction as well as its effects of altering the immune system.[50] It has demonstrated activity in the relapsed CLL setting, but it is also recently being shown to have significant efficacy in the first-line setting as well.

Chen et al[51] initiated a phase I study in which 25 previously untreated patients with CLL received lenalidomide (starting at 2.5 mg daily with monthly escalation to a target dose of 10 mg daily) for 21 days out of a 28-day cycle, for a median of 13 cycles. The OR rate was 56%, with no patient having a CR. Patients experienced major toxicity, including fatigue (78%), tumor flare (88%), rash (48%), and grade 3/4 neutropenia (72%). Of note, these patients were reasonably high-risk: 75% had unmutated IgVH, and 33% had 17p or 11q deletions.

Ferrajoli et al[52] administered lenalidomide to 43 previously untreated CLL patients 65 years of age or older. The drug was given orally at 5 mg daily for 2 continuous monthly cycles. The dose was then increased by 5 mg per cycle up to a maximum of 25 mg. The median dose delivered was 5 mg to 10 mg. Grade 3/4 myelosuppression was observed in 26% of the patients and tumor flare in 44%. The OR rate was 54%.

While these results demonstrate that lenalidomide may have activity in CLL, it is important to note that prolonged continuous therapy may be required for the highest-quality responses. In addition, the response rates observed may be lower than optimal for front-line therapy.

GA-101

GA-101 is a type II glycoengineered, humanized anti-CD20 monoclonal antibody that binds CD20 in a completely different orientation than rituximab and over a larger surface area. It has shown promising activity when given as a single agent to heavily pretreated patients with CLL, and it has a safety profile for CLL patients similar to that observed in patients with non-Hodgkin lymphoma.[28] GA-101 is currently being evaluated in combination with chlorambucil in a phase II study with previously untreated elderly patients.

Special Considerations and Discussion

The National Cancer Institute Working Group 1996 guidelines, updated in 2008 by the International Workshop on Chronic Lymphocytic Leukemia, should be followed when deciding the appropriate time to initiate therapy in patients.[7] Indications for initiating therapy are summarized in Table 3.

The addition of cyclophosphamide to fludarabine in first-line therapy resulted in only a partial improvement in CR, suggesting that the addition of rituximab to this combination is responsible for the increased CR rate seen with FCR.[28] The benefit of cyclophosphamide is uncertain as there has not been a direct comparison of FCR vs FR. However, the CALGB 10404 trial is currently ongoing to determine whether FCR has an advantage over FR and, if so, whether the increased risk of second-

ary acute myeloid leukemia associated with cyclophosphamide justifies that advantage.

Hallek[6] recently proposed an algorithm for the selection of the best treatment option for patients requiring initiation of therapy. This algorithm is based on three important factors: the physical condition of the patient (regardless of age), the stage of disease, and the prognostic risk factors of the leukemia. For patients who fall into the "go go" category, as defined by having normal renal function (creatinine clearance > 70 mL/min) and a low score on the Cumulative Illness Rating Scale (CIRS), patients should be offered regimens such as FCR, which produce higher response rates and longer PFS and OS. "Slow go" patients are those who have significant comorbidities; the goal of treatment is to alleviate disease-related symptoms. These patients should be treated with agents such as chlorambucil, bendamustine with or without rituximab, or dose-reduced fludarabine combinations.[6]

Regimens that result in more MRD negativity, such as FCR, may be ideal for the "go go" group of patients, since MRD negativity has been shown to correlate with a longer PFS in some trials. The use of consolidation therapy with lenalidomide or alemtuzumab to achieve MRD negativity cannot be recommended because it has resulted in significant, and sometimes fatal, toxicity despite improved survival rates.[53-55] Maintenance therapy or consolidation therapy with chemotherapy or immunotherapy has no established role in the treatment of CLL as no randomized trials have been performed to determine if any benefit can be derived from this strategy.

To date, three studies have been published that examined the impact of prognostic markers on response to therapy in previously untreated patients with CLL. Results of these studies suggest that patients with high-risk cytogenetic and IgVH features respond unfavorably or have a shorter response duration when treated with chemotherapy or chemoimmunotherapy. Specifically, those who present with del(17p) or p53 mutations have poor responses to therapy and reduced survival time. Ideally, this patient population should be enrolled in clinical trials (Table 6).[20,56-58]

Hematopoietic stem cell transplantation (HSCT) should be considered only as first-line therapy for younger, healthy patients with del(17p) or p53 mutations who are in their first CR. These patients have a poor prognosis and tend to be resistant to fludarabine-based therapies. Recent guidelines outlined by the European Group for Blood and Bone Marrow Transplantation recommend allogeneic HSCT as a reasonable treatment option for the aforementioned patient population. It is generally not a suitable option for the majority of CLL patients, most of whom are elderly and with disease that has an extremely indolent course.[59]

Infections are a major cause of morbidity and mortality in CLL patients, mediated through humoral and cellular immunosuppression inherent to the disease itself and through the further immunosuppression caused by treatment. The frequency of infections is particularly increased after the use of alemtuzumab and high-dose corticosteroids. Bacterial infections are most common among CLL patients. However, because some of the newer therapeutic agents (alemtuzumab in particular) cause T-cell dysfunction, patients are at risk for CMV, *Pneumocystis jirovecii* pneumonia (PJP), herpes simplex virus (HSV), and *Listeria monocytogenes*.[60] Recent guidelines for the use of alemtuzumab include conducting routine monitoring for CMV by polymerase chain reaction analysis and instituting preemptive therapy when positive. The guidelines also include prophylaxis with sulfamethoxazole/trimethoprim and acyclovir for PJP and HSV, respectively.[61]

Hypogammaglobulinemia is the most important immune defect in terms of the risk of severe bacterial infections. Supplementation with intravenous immunoglobulin should be considered in patients with CLL who

Table 6. — Selected Studies Demonstrating the Impact of Prognostic Markers on Outcome in Patients With CLL

Trial	Complete Remission	Progression-Free Survival	Overall Survival
CALGB 9712[56]	Not significant	IgVH del(11q) del(17p)	IgVH del(11q) del(17p)
ECOG 2997[57]	Not significant	IgVH (FC only) del(11q) del(17p)	Not stated
LRF CLL4[20]	del(11q) del(17p)	IgVH del(11q) del(17p)	Not stated

From Gribben JG. How I treat CLL up front. *Blood.* 2010;115(2):187-197. *Blood:* Journal of the American Society of Hematology by American Society of Hematology. Copyright 2010. Reproduced with permission of AMERICAN SOCIETY OF HEMATOLOGY (ASH) in the format Journal via Copyright Clearance Center.[58]

experience frequent severe bacterial infections, particularly patients with encapsulated organisms.[60]

Conclusions

Over the past decade, major improvements in the identification of new prognostic markers for CLL, as well as novel combinations for chemoimmunotherapy regimens, have extended both PFS and OS. Since CLL is primarily a disease of the elderly, treatment should be initiated only when warranted and must consider each patient's performance status, including comorbidities. For patients who do require therapy, several chemoimmunotherapy options (eg, FCR, FCR-Lite, BR) are available in the front-line setting, the choice of which should take into account the condition of the patient and the goals of treatment.

New single agents and novel treatment combinations have achieved promising results in phase I/II studies, results that now await confirmation in randomized phase III trials. This approach will provide answers several questions: What is the optimal front-line therapy? When should treatment be initiated? Should MRD be used to monitor treatment efficacy? Should treatment be tailored by specific prognostic markers? Our ultimate therapeutic goals of prolongation of survival and improvement in quality of life will continue to be validated only by ongoing clinical and laboratory research and by continuous enrollment of patients in clinical trials.

References

1. National Cancer Institute. Surveillance Epidemiology and End Results Stat Fact Sheets: Chronic Lymphocytic Leukemia. http://seer.cancer.gov/statfacts/html/cll.html. Accessed May 8, 2011.
2. Wierda WG, Keating MJ, O'Brien S. Chronic lymphoid leukemias. In: DeVita VT Jr, Lawrence TS, Rosenberg SA, eds. *Cancer: Principles and Practice of Oncology*. 8th ed. Philadelphia PA: Lippincott Williams & Wilkins; 2008:2279-2293.
3. Malone KE, Koepsell TD, Daling JR, et al. Chronic lymphocytic leukemia in relation to chemical exposures. *Am J Epidemiol*. 1989;130(6):1152-1158.
4. Bizzozero OJ Jr, Johnson KG, Ciocco A, et al. Radiation-related leukemia in Hiroshima and Nagasaki 1946-1964. II. *Ann Intern Med*. 1967;66(3):522-530.
5. Yuille MR, Matutes E, Marossy A, et al. Familial chronic lymphocytic leukemia: a survey and review of published studies. *Br J Haematol*. 2000;109(4):794-799.
6. Hallek M. State-of-the-art treatment of chronic lymphocytic leukemia. *Hematology Am Soc Hematol Educ Program*. 2009:440-449.
7. Hallek M, Cheson BD, Catovsky D, et al. Guidelines for the diagnosis and treatment of chronic lymphocytic leukemia: a report from the International Workshop on Chronic Lymphocytic Leukemia updating the National Cancer Institute — Working Group 1996 guidelines. *Blood*. 2008;111(12):5446-5456.
8. Rai KR, Sawitsky A, Cronkite EP, et al. Clinical staging of chronic lymphocytic leukemia. *Blood*. 1975;46(2):219-234.
9. Binet JL, Auquier A, Dighiero G, et al. A new prognostic classification of chronic lymphocytic leukemia derived from a multivariate survival analysis. *Cancer*. 1981;48(1):198-206.
10. Döhner H, Stilgenbauer S, Benner A, et al. Genomic aberrations and survival in chronic lymphocytic leukemia. *N Engl J Med*. 2000;343(26):1910-1916.
11. Rassenti LZ, Jain S, Keating MJ, et al. Relative value of ZAP-70, CD38 and immunoglobulin mutation status in predicting aggressive disease in chronic lymphocytic leukemia. *Blood*. 2008;112(5):1923-1930.
12. CLL Trialists' Collaborative Group. Chemotherapeutic options in chronic lymphocytic leukemia: a meta-analysis of the randomized trials. *J Natl Cancer Inst*. 1999;91(10):861-868.
13. Eichhorst BF, Busch R, Stilgenbauer S, et al. First-line therapy with fludarabine compared with chlorambucil does not result in a major benefit for elderly patients with advanced chronic lymphocytic leukemia. *Blood*. 2009;114(16):3382-3391.
14. Rai KR, Peterson BL, Appelbaum FR, et al. Fludarabine compared with chlorambucil as primary therapy for chronic lymphocytic leukemia. *N Engl J Med*. 2000;343(24):1750-1757.
15. Leporrier M, Chevret S, Cazin B, et al. Randomized comparison of fludarabine, CAP, and ChOP in 938 previously untreated stage B and C chronic lymphocytic leukemia patients. *Blood*. 2001;98(8):2319-2325.
16. Cheson BD, Rummell MJ. Bendamustine: rebirth of an old drug. *J Clin Oncol*. 2009;27(9):1492-1501.
17. Knauf WU, Lissichkov T, Aldaoud A, et al. Phase III randomized study of bendamustine compared with chlorambucil in previously untreated patients with chronic lymphocytic leukemia. *J Clin Oncol*. 2009;27(26):4378-4384.
18. Eichhorst BF, Busch R, Hopfinger G, et al. Fludarabine plus cyclophosphamide versus fludarabine alone in first-line therapy of younger patients with chronic lymphocytic leukemia. *Blood*. 2006;107(3):885-891.
19. Flinn IW, Neuberg DS, Grever MR, et al. Phase III trial of fludarabine plus cyclophosphamide compared with fludarabine for patients with previously untreated chrnic lymphocytic leukemia: US Intergroup Trial E2997. *J Clin Oncol*. 2007;25(7):793-798.
20. Catovsky D, Richards S, Matutes E, et al. Assessment of fludarabine plus cyclophosphamide for patients with chronic lymphocytic leukaemia (the LRF CLL4 trial): a randomised controlled trial. *Lancet*. 2007;370(9583):230-239.
21. Robak T, Blonski JZ, Kasznicki M, et al. Cladribine with prednisone versus chlorambucil with prednisone as first-line therapy in chronic lymphocytic leukemia: reportof a prospective, randomized, multicenter trial. *Blood*. 2000;96(8):2723-2729.
22. Robak T, Blonski JZ, Gora-Tybor J, et al. Cladribine alone and in combination with cyclophosphamide or cyclophosphamide plus mitoxantrone in the treatment of progressive chronic lymphocytic leukemia: report of a prospective, multicenter, randomized trial of the Polish Adult Leukemia Group (PALG CLL2). *Blood*. 2006;108(2):473-479.
23. Huhn D, von Schilling C, Wilhelm M, et al. Rituximab therapy of patients with B-cell chronic lymphocytic leukemia. *Blood*. 2001 98(5):1326-1331.
24. Byrd JC, Murphy T, Howard RS, et al. Rituximab using thrice weekly dosing schedule in B-cell chronic lymphocytic leukemia and small lymphocytic lymphoma demonstrates clinical activity and acceptable toxicity. *J Clin Oncol*. 2001:19(8):2153-2164.
25. O'Brien SM, Kantarjian H, Thomas DA , et al. Rituximab dose-escalation trial in chronic lymphocytic leukemia. *J Clin Oncol*. 2001:19(8):2165-2170.
26. Hainsworth JD, Litchy S, Barton JH, et al. Single-agent rituximab as first-line and maintenance treatment for patients with chronic lymphocytic leukemia or small lymphocytic lymphoma: a phase II trial of the Minnie Pearl Cancer Research Network. *J Clin Oncol*. 2003;21(9):1746-1751.
27. Mone AP, Cheney C, Banks AL, et al. Alemtuzumab induces caspase-independent cell death in human chronic lymphocytic leukemia cells through a lipid raft-dependent mechanism. *Leukemia*. 2006;20(2):272-279.
28. Jaglowski SM, Alinari L, Lapalombella R, et al. The clinical application of monoclonal antibodies in chronic lymphocytic leukemia. *Blood*. 2010;116(19):3705-3714.
29. Hillmen P, Skotnicki AB, Robak T, et al. Alemtuzumab compared with chlorambucil as first-line therapy for chronic lymphocytic leukemia. *J Clin Oncol*. 2007;25(35):5616-5623.
30. Stilgenbauer S, Döhner H. Campath-1H-induced complete remission of chronic lymphocytic leukemia despite p53 gene mutation and resistance to chemotherapy [letter]. *N Engl J Med*. 2002;347(6):452-453.
31. Lozanski G, Heerema NA, Flinn IW, et al. Alemtuzumab is an effective therapy for chronic lymphocytic leukemia with p53 mutations and deletions. *Blood*. 2004;103(9):3278-3281.
32. Lundin J, Kimby E, Björkholm M, et al. Phase II trial of subcutaneous anti-CD52 monoclonal antibody alemtuzumab (Campath-1H) as first-line treatment for patients with B-cell chronic lymphocytic leukemia (B-CLL). *Blood*. 2002;100(3):768-773.
33. Schulz H, Klein SK, Rehwald U, et al. Phase 2 study of a combined immunochemotherapy using rituximab and fludarabine in patients with chronic lymphocytic leukemia. *Blood*. 2002;100(9):3115-3120.
34. Woyach JA, Ruppert AS, Heerema NA, et al. Chemoimmunotherapy with fludarabine and rituximab produces extended overall survival and progression-free survival in chronic lymphocytic leukemia: long-term follow-up of CALGB study 9712. *J Clin Oncol*. 2011;29(10):1349-1355.
35. Byrd JC, Rai K, Peterson BL, et al. Addition of rituximab to fludarabine may prolong progression-free survival and overall survival in patients with previously untreated chronic lymphocytic leukemia: an updated retrospective comparative analysis of CALGB 9712 and CALGB 9011. *Blood*. 2005;105(1):49-53.
36. Keating MJ, O'Brien S, Albitar M, et al. Early results of a chemoimmunotherapy regimen of fludarabine, cyclophophamide and rituximab as initial therapy for chronic lymphocytic leukemia. *J Clin Oncol*. 2005;23(18):4079-4088.
37. Hallek M, Fischer K, Fingerle-Rowson G, et al. Addition of rituximab to fludarabine and cyclophosphamide in patients with chronic lymphocytic leukaemia: a randomised, open-label, phase 3 trial. *Lancet*. 2010;376(9747):1164-1174.

38. Boettcher S, Fischer K, Stilgenbauer S, et al. Quantitative MRD assessments predict progression free survival in CLL patients treated with fludarabine and cyclophosphamide with or without rituximab — a prospective analysis in 471 patients from the randomized GCLLSG CLL8 trial. *Blood*. 2008;112:125-126.

39. FDA Approval for Rituximab. http://www.cancer.gov/cancertopics/druginfo/fda-rituximab. Accessed June 17, 2011.

40. Foon KA, Boyiadzis M, Land SR, et al. Chemoimmunotherapy with low dose fludarabine and cyclophosphamide with high dose rituximab in previously untreated patients with chronic lymphocytic leukemia. *J Clin Oncol*. 2009;27(4):498-503.

41. Lamanna N, Kalaycio M, Maslak P, et al. Pentostatin, cyclophosphamide and rituximab is an active,well-tolerated regimen for patients with previously treated chronic lymphocytic leukemia. *J Clin Oncol*. 2006;24(10):1575-1581.

42. Kay NE, Geyer SM, Call TG, et al. Combination chemoimmunitheraoy with pentostatin, cyclophosphamide, and rituximab shows significant clinical activity with low accompanying toxicity in previously untreated B chronic lymphocytic leukemia. *Blood*. 2007;109(2):405-411.

43. Reynolds C, Di Bella N, Lyons RM, et al. Phase III trial of fludarabine, cyclophosphamide, and rituximab vs. pentostatin, cyclophopshamide, and rituximab in B-cell chronic lymphocytic leukemia. *Blood (ASH Anuual Meeting Abstracts)*. 2008;112(11):327.

44. Wierda WG, O'Brien SM, Faderl SH, et al. CFAR, an active frontline regimen for high risk patients with CLL, including those with del 17p. *Blood (ASH Annual Meeting Abstracts)*. 2008;112:2095.

45. Parikh SA, Keating M, O'Brien S, et al. Frontline combined chemoimmunotherapy with fludarabine, cyclophosphamide, alemtuzumab and rituximab (CFAR) in high-risk chronic lymphocytic leukemia. *Blood (ASH Annual Meeting Abstracts)*. 2009;114(22):208. Abstract.

46. Fischer K, Cramer P, Stilgenbauer S, et al. Bendamustine combined with rituximab (BR) in first-line therapy of advanced CLL: a multicenter phase II trial of the German CLL Study Group (GCLLSG). *Blood (ASH Annual Meeting Abstracts)*. 2009;114(22):205.

47. Castro JE, James DF, Sandoval-Sus JD, et al. Rituximab in combination with high-dose methylprednisoline for the treatment of chronic lymphocytic leukemia. *Leukemia*. 2009;23(10):1779-1789.

48. Bertazzoni P, Rabascio C, Gigli F, et al. Rituximab and subcutaneous cladribine in chronic lymphocytic leukemia for newly diagnosed and relapsed patients. *Leuk Lymphoma*. 2010;51(8):1485-1493.

49. Wierda WG, Kipps TJ, Dürig J, et al. Ofatumumab combined with fludarabine and cyclophosphamide (O-FC) shows high activity in patients with previously untreated chronic lymphocytic leukemia (CLL): results from a randomized, multicenter, international, two-dose, parallel group, phase II trial. *Blood (ASH Annual Meeting Abstracts)*. 2009;114(22):207.

50. Revlimid (Lenalidomide) package insert, 2005-2010, Celgene Corp, Summit, NJ. www.Revlimid.com/pdf/REVLIMID_PI.pdf. Accessed June 5, 2011

51. Chen CI, Bergsagel PL, Paul H. Single-agent lenalidomide in the treatment of previously untreated chronic lymphocytic leukemia. *J Clin Oncol*. 2011;29(9):1175-1181.

52. Ferrajoli A, O'Brien S, Wierda W, et al. Lenalidomide as initial treatment of elderly patients with chronic lymphocytic leukemia (CLL). *Blood (ASH Annual Meeting Abstracts)*. 2008;112(11):45.

53. Moreton P, Kennedy B, Lucas G, et al. Eradication of minimal residual disease in B-cell chronic lymphocytic leukemia after alemtuzumab therapy is associated with prolonged survival. *J Clin Oncol*. 2005;23(13):2971-2979.

54. O'Brien SM, Kantarjian HM, Thomas DA, et al. Alemtuzumab as treatment for residual disease after chemotherapy in patients with chronic lymphocytic leukemia. *Cancer*. 2003;98(12):2657-2663.

55. Schweighofer CD, Ritgen M, Eichhorst BF, et al. Consolidation with alemtuzumab improves progression-free survival in patients with chronic lymphocytic leukemia (CLI) in first remission: long-term follow-up of a randomized phase III trial of the German CLL Study Group (GCLLSG). *Br J Haematol*. 2009;144(1):95-98

56. Byrd JC, Gribben JG, Peterson BL, et al. Select high-risk genetic features predict earlier progression following chemoimmunotherapy with fludarabine and rituximab in chronic lymphocytic leukemia: justification for risk-adapted therapy. *J Clin Oncol*. 2006;24(3):437-443.

57. Grever MR, Lucas DM, Dewald GW, et al. Comprehensive assessment of genetic and molecular features predicting outcome in patients with chronic lymphocytic leukemia: results from the US Intergroup Phase III trial E2997. *J Clin Oncol*. 2007;25(7):799-804.

58. Gribben JG. How I treat CLL up front. *Blood*. 2010;115(2):187-197.

59. Gribben JG. Stem cell transplantation in chronic lymphocytic leukemia. *Biol Blood Marrow Transplant*. 2009;15(1 suppl):53-58.

60. Morrison VA. Update on prophylaxis and therapy of infection in patients with chronic lymphocytic leukemia. *Expert Rev Anticancer Ther*. 2001;1(1):84-90.

61. Osterbog A, Foà R, Bezares RF, et al. Management guidelines for the use of alemtuzumab in chronic lymphocytic leukemia. *Leukemia*. 2009;23(11):1980-1988.