A chemotherapy-free future? Novel treatment strategies for chronic lymphocytic leukemia

M. Hallek

Department I of Internal Medicine
Center for Integrated Oncology Köln
Bonn

Center of Excellence on “Cellular Stress Responses in Aging-Associated Diseases”
University of Cologne, Germany

Correspondence:
Michael Hallek
E-mail: michael.hallek@uni-koeln.de

Acknowledgment:
The author wishes to thank Dr. Barbara Eichhorst for giving feedback on the therapeutic algorithm, Drs. Anja Engelke, Gabor Kovacs, and Paula Cramer for carefully reading an earlier version of the manuscript, and the team of the GCLLSG study office for giving important suggestions. Moreover, I thank all patients and physicians participating in the studies of the German CLL Study Group for their continuing support and excellent cooperation. This work is supported by the Deutsche Krebshilfe (German Cancer Aid), the Kompetenznetz Maligne Lymphome (Competence Network Malignant Lymphoma) and the Deutsche Forschungsgemeinschaft (KFO 286, SFB 832 and Center of Excellence on “Cellular Stress Responses in Aging-Associated Diseases”).

Overview

With an age-adjusted incidence of 4.3/100,000 inhabitants in the United States, chronic lymphocytic leukemia (CLL) is the most common type of leukemia in Western countries. More than 15,000 newly diagnosed cases and approximately 4,500 deaths are currently estimated. The median age at diagnosis lies between 67 and 72 years. More male than female patients are affected by this disease.1

Our understanding of the pathogenesis of this B-cell neoplasia continues to grow and has been recently summarized elsewhere.2-3 Fortunately, this tremendous progress has resulted in the development of novel agents for CLL therapy, whose future use is outlined in this article.

Therapy with approved agents

Cytostatic agents

Monotherapy with alkylating agents has served as initial, front-line therapy for CLL, and chlorambucil (CLB) was the gold standard of CLL therapy for several decades.4 This standard has been replaced, at least for physically fit patients, by the use of the more potent purine analogs, fludarabine, pentostatin, and cladribine (2-CdA). Fludarabine is by far the best-studied compound of the three in CLL. In several clinical studies, fludarabine induced more remissions and more CR (7-40%) than purine analogs, fludarabine, pentostatin, and cladribine (2-CdA). Fludarabine is by far the best-studied compound of the three in CLL. In several clinical studies, fludarabine induced more remissions and more CR (7-40%) than

Antibodies

Antibodies were introduced for the treatment of CLL later than for other lymphoid malignancies. In CLL, the anti-CD20 antibody rituximab is less active as a single agent than...
in follicular lymphoma, unless very high doses are used.\textsuperscript{12,13} In contrast, combinations of rituximab with chemotherapy have proven to be very efficacious therapies for CLL (see below). Some newer CD20-antibodies challenge rituximab.\textsuperscript{14-16} Ofatumumab is a fully humanized antibody targeting a unique epitope on the CD20 molecule, resulting in increased binding affinity, prolonged dissociation rate, and increased cell kill due to greater complement-dependent cytotoxicity (CDC), and similar antibody-dependent cellular cytotoxicity (ADCC) activity compared to rituximab, especially in cells expressing low levels of CD20. Ofatumumab has shown some efficacy in patients who are fludarabine and alemtuzumab-refractory or have bulky disease (>5 cm).\textsuperscript{17} The overall response rate was 58% in the FA-refractory group and 47% in the bulky disease group. However, so far, a formal comparison of ofatumumab to rituximab has not been performed in a clinical trial. Therefore, the definitive value of ofatumumab for the treatment of B-cell lymphoma and CLL remains unclear. More recently, a novel, humanized and glyco-engineered monoclonal antibody obinutuzumab (GA101) showed impressive results in vitro with higher rates of apoptosis in malignant B cells in comparison with rituximab.\textsuperscript{18} The humanization of the parental B-Ly1 mouse antibody and subsequent glycol-engineering lead to higher affinity binding to CD20 type II epitope, increased antibody-dependent cellular cytotoxicity (ADCC), low complement-dependent cytotoxicity (CDC) activity, and increased direct cell death induction.\textsuperscript{19} A phase I study with obinutuzumab showed promising results in 13 CLL patients.\textsuperscript{20} Major side-effects included infections, neutropenia, thrombocytopenia and tumor lysis syndrome, which all resolved. There were no dose-limiting toxicities.

Alemtuzumab is a recombinant, fully humanized, monoclonal antibody against the CD52 antigen. Monotherapy with alemtuzumab has produced response rates of 33% to 53%, with a median duration of response ranging from 8.7 to 15.4 months, in patients with advanced CLL who were previously treated with alkylating agents and had failed or relapsed after second-line fludarabine therapy.\textsuperscript{21-23} In addition, alemtuzumab has proven to be effective in patients with high-risk genetic markers such as deletions of chromosome 11 or 17 (\textit{del(11q)} and \textit{del(17p)}) and \textit{TP53} mutations.\textsuperscript{24,25} Therefore, alemtuzumab is a reasonable therapeutic option for patients with these poor prognostic features. In a prospective randomized study, alemtuzumab was tested against CLB.\textsuperscript{26} Alemtuzumab led to a greater OR and CR (\textit{P}<0.0001), superior PFS with a 42% reduction in risk of progression or death (\textit{P}<0.0001), and significantly longer median time to progression (TTP) (\textit{P}=0.0001). Therefore, the drug remains a valuable treatment option for high-risk patients. Unfortunately, the drug is no longer licensed, although it does remain available in some countries on a compassionate use basis.

**Combination therapies**

A major advance in CLL treatment was achieved by the combined use of different treatment modalities. Since purine analogs and alkylating agents have different mechanisms of action and partially non-overlapping toxicity profiles, they were combined both in pre-clinical and clinical studies. The most thoroughly studied combination chemotherapy for CLL is fludarabine plus cyclophosphamide (FC) given as oral drugs or intravenous infusions.\textsuperscript{27} In non-comparative trials, the overall response rates did not appear to be better than with fludarabine alone, but the addition of cyclophosphamide appeared to improve the CR rate up to 50%.\textsuperscript{27} Three randomized trials have shown that FC combination chemotherapy improves the CR and OR rates and PFS as compared to fludarabine monotherapy.\textsuperscript{28-30} The rate of severe infections was not significantly increased by the FC combination despite a higher frequency of neutropenias. An up-dated analysis of the CLL4 trial of the German CLL Study Group (GCLLSG) suggested that the first-line treatment of CLL patients with FC combination might improve the OS of the non-high-risk CLL patients (i.e. all patients who do not exhibit a \textit{del(17p)} or \textit{TP53} mutation). The Polish Adult Leukemia Group (PALG) compared 2-CdA alone to 2-CdA combined with cyclophosphamide (CC) or to cyclophosphamide and mitoxantrone (CMC) in 479 cases with untreated progressive CLL.\textsuperscript{31} Surprisingly, the CC combination therapy did not produce any benefit in terms of progression-free survival or response rates when compared to 2-CdA alone.

Phase II studies suggested that rituximab combinations with fludarabine or fludarabine-based regimens would produce significant improvements in all major outcome parameters such as CR rates, PFS and overall survival (for a review see Hallek and Pfug).\textsuperscript{32} The largest of these trials conducted on 300 patients with previously untreated CLL showed that rituximab plus fludarabine/cyclophosphamide (FCR) achieved an overall response rate of 95%, with CR in 72%.\textsuperscript{33} Six-year OS and failure-free survival were 77% and 51%, respectively, and median TTP was 80 months.

These results led the GCLLSG to conduct a randomized trial, the CLL8 protocol, on 817 patients, median age 61 years, with good physical fitness who received 6 courses of FC (\textit{n}=409) or FCR (\textit{n}=408).\textsuperscript{34} FCR treatment was more frequently associated with CTC grade 3 and 4 neutropenia (FCR 34%; FC 21%), without any formal recommendation to use growth factor support. There was no increase in other grade 3 or 4 side-effects, including infections. FCR induced a higher OR rate than FC (92.8\% vs. 85.4\%) and more cases of CR (44.5\% vs. 22.9\%) (\textit{P}<0.001). Most importantly, this trial was the first to show that the choice of a first-line therapy has an impact on OS of CLL patients. In an up-dated analysis at a median follow up of 5.9 years, 38.0\% of the patients in the FCR group were free of disease progression compared with 27.4\% in the FC group (hazard ratio (HR) 0.6; 95\%CI: 0.5-0.7; \textit{P}<0.0001).\textsuperscript{35} At the same time point, 69.4\% of the patients were alive in the FCR group versus 62.3\% in the FC group (HR 0.7; 95\%CI: 0.5-0.9; \textit{P}=0.001). Median OS was not reached for patients in the FCR group, while median OS was 86.0 months (95\%CI: 78.7-93.2 months) for the FC group (\textit{P}=0.001).\textsuperscript{36} FCR did not improve the survival of patients with a \textit{del(17p)}. Similar results were obtained in a trial comparing FCR to FC in relapsed CLL, without demonstrating a benefit for OS.\textsuperscript{36} Since CLL often occurs in elderly patients with relevant comorbidity, a dose-modified FCR-Lite regimen was designed to maintain the efficacy but decrease the toxicity of the FCR regimen.\textsuperscript{27} This regimen reduced the dose of fludarabine and cyclophosphamide and increased the dose of rituximab, and used a maintenance regimen for rituximab given every three months until progression. The CR
rate was 77% for 50 previously untreated CLL patients with an OR rate of 100%. Grade 3/4 neutropenia was documented in only 13% of cycles, which is lower than observed with the usual FCR regimen.

Following a similar concept, the CLL11 protocol of the GCLLSG was designed to test chemoimmunotherapies with anti-CD20 antibodies combined with a milder chemotherapeutic component, chlorambucil (CLB), in previously untreated CLL patients with comorbidities. The rationale of this study was based on results of phase II trials using CLB in combination with rituximab (RCLB).

Moreover, encouraging results were reported in the run-in phase of the CLL11 trial on CLL patients with increased comorbidity when treated with a combination of CLB and obinutuzumab. In the CLL11 trial, 781 patients with previously untreated CLL and a score higher than 6 on the Cumulative Illness Rating Scale (CIRS) (range 0-56, with higher scores indicating worse health status) or an estimated creatinine clearance of 30-69 mL per minute, were assigned to receive CLB, obinutuzumab plus CLB, or rituximab plus CLB. The patients had a median age of 73 years, creatinine clearance of 62 mL per minute, and CIRS score of 8 at baseline. Treatment with obinutuzumab-CLB or R-CLB, as compared with CLB monotherapy, increased response rates and prolonged progression-free survival: median PFS 26.7 months with obinutuzumab-CLB versus 11.1 months with CLB alone (HR for progression or death 0.18; 95%CI: 0.13-0.24; P<0.001) and 16.3 months with R-CLB versus 11.1 months with CLB alone (HR 0.44; 95%CI: 0.34-0.57; P<0.001). Treatment with obinutuzumab-CLB, as compared with CLB alone, prolonged overall survival (HR for death 0.41; 95%CI: 0.23-0.74; P=0.002). Treatment with obinutuzumab-CLB, as compared with R-CLB, resulted in prolongation of PFS (HR 0.39; 95%CI: 0.31-0.49; P<0.001) and higher rates of complete response (20.7% vs. 7.0%) and molecular response. Infusion-related reactions and neutropenia were more common with obinutuzumab-CLB than with R-CLB, but there was no increase in the risk of infection. Taken together, these results show that combining an anti-CD20 antibody with chemotherapy improved outcomes in patients with CLL and co-existing conditions. In this patient population, obinutuzumab was superior to rituximab when combined with CLB. Other variations have been tested to further improve the efficacy of the FCR regimen. Alemtuzumab (A) was added to FCR (CFAR) in a phase II trial on 60 high-risk untreated patients under 70 years of age with serum beta 2 microglobulin of 4 mg/L or more. CR was achieved in 70%, PR in 18%, and nodular PR in 3%, for an overall response of 92%. Of 14 patients with 17p deletion, 8 (57%) achieved a CR. Grade 3/4 neutropenia and thrombocytopenia occurred with 33% and 13% of courses, respectively. The median PFS was 38 months and median OS was not reached. Therefore, CFAR might be helpful in cases of high-risk CLL where an effective cytoreductive therapy is desired before an allogeneic stem cell transplant. In another study on 72 untreated CLL patients aged 70 years or under, mitoxantrone was combined at 6 mg/m² on Day 1 of each cycle with FCR. The overall response, minimal residual disease (MRD)-negative complete response (CR), MRD-positive CR, and PR rates were 93%, 46%, 36%, and 11%, respectively. Severe neutropenia developed in 13% of patients. These results do not justify the broad use of this regimen outside of clinical trials.

An alternative idea was to replace fludarabine in the FCR regimen with pentostatin (PCR) in order to reduce myelotoxicity. In a phase III randomized trial comparing FCR to PCR in previously untreated or minimally treated CLL patients, there were no statistical differences between treatments in OS or response rates. Moreover, this trial did not demonstrate a lower infection rate with PCR.

Bendamustine has been also combined with rituximab (BR) in 81 patients with relapsed CLL. Patients received 70 mg/m² of bendamustine on Days 1 and 2 and 375 mg/m² of rituximab on Day 0 of the first cycle and 500 mg/m² on Day 1 of subsequent cycles administered every 28 days for up to 6 cycles. On the basis of intent-to-treat analysis, the overall response rate (ORR) was 59.0% (95%CI: 47.3-70.0%). Complete response, partial response, and nodular partial response were achieved in 9.0%, 47.4%, and 2.6% of patients, respectively. Overall response rate was 45.5% in fludarabine-refractory patients and 60.5% in fludarabine-sensitive patients. Among genetic subgroups, 92.3% of patients with del(11q), 100% with trisomy 12, 7.1% with del(17p), and 58.7% with unmutated IGHV status responded to treatment. After a median follow-up time of 24 months, the median event-free survival was 14.7 months. Severe infections occurred in 12.8% of patients. Grade 3 or 4 neutropenia, thrombocytopenia, and anemia were documented in 23.1%, 28.2%, and 16.6% of patients, respectively.

The BR regimen was also investigated as first-line therapy in 117 CLL patients (age 34-78 years). Bendamustine was administered at a dose of 90 mg/m² on Day 1 and 2 combined with 375 mg/m² rituximab on Day 0 of the first course and 500 mg/m² on Day 1 during subsequent courses for up to 6 courses. Overall response rate was 88.0% (95%CI: 80.7-100.0%) with a complete response rate of 23.1% and a partial response rate of 64.9%. Ninety percent of patients with del(11q), 94.7% with trisomy 12, 37.5% with del(17p), and 89.4% with unmutated IGHV status responded to treatment. After a median observation time of 27.0 months, median event-free survival was 33.9 months, and 90.5% of patients were alive. Grade 3 or 4 severe infections occurred in 7.7% of patients. Grade 3 or 4 adverse events for neutropenia, thrombocytopenia, and anemia were documented in 19.7%, 22.2%, and 19.7% of patients, respectively. Overall, these results suggest that when compared to FCR, BR is somewhat less active, yielding lower CR rates, but is also less myelotoxic.

The CLL10 protocol of the GCLLSG compared BR to FCR in first-line therapy of physically fit patients without del(17p). A total of 564 patients with a CIRS score of 6 or less, creatinine clearance over 70 mL/min and without del(17p) were randomly assigned to receive 6 courses of either FCR (n=284) or BR (n=280), with a bendamustine dose of 90 mg/m² on Days 1+2. The ITT population consisted of 561 patients; 22% of patients were Binet A, 38% Binet B and 40% Binet C. Median age was 62 years (range 33-82 years); median CIRS score was 2 (range 0-6). There were significantly more patients with unmutated IGHV in the BR arm (68%) compared to the FCR arm (55%; P=0.003). All other characteristics were well balanced. A median number of 5.27 courses were given in the FCR arm versus 5.41 courses in the BR arm (P=0.022); 70.6% (FCR) and 80.3% (BR) of patients received 6 courses (P=0.008). Dose was reduced by more than 10% in 27.3% (FCR) and 31.6% (BR) of all
courses given ($P=0.012$). The median observation time was 27.9 months in all patients alive. A total of 547 patients (274 FCR; 273 BR) were evaluable for response and all 561 patients (282 FCR; 279 BR) for PFS, event-free survival (EFS) and OS. The overall response rate was identical in both arms with 97.8% ($P=1.0$). The complete response rate was 47.4% for FCR compared to 38.1% for BR ($P=0.031$).

MRD data were available at interim analysis from 192 patients (99 FCR; 93 BR) of the first 300 patients; 71.7% of patients in the FCR and 66.7% in the BR arms achieved MRD levels below $10^{-4}$ in peripheral blood at final staging ($P=0.448$). PFS was 85.0% at two years in the FCR arm and 78.2% in the BR arm ($P=0.041$). EFS was 82.6% at two years in the FCR arm and 75.7% in the BR arm ($P=0.037$). There was no difference in OS. Hazard ratios for PFS, EFS and OS were 1.385, 1.375 and 0.842, respectively. FCR treated patients experienced significantly more frequent CTC grade 3 to 5 adverse events (90.8% vs. 78.5%; $P<0.001$), in particular more hematotoxicity (90.0% vs. 66.9%; $P<0.001$), severe neutropenia (81.7% vs. 56.8%; $P<0.001$) and severe infections (39.0% vs. 25.4%; $P=0.001$). Treatment-related mortality occurred in 3.9% (n=11) in the FCR and 2.1% (n=6) in the BR arm. In conclusion, the FCR regimen seems clearly more potent but also more toxic than BR in CLL patients. Several other combinations have been investigated, like cladribine with rituximab, methylprednisolone plus rituximab followed by alemtuzumab or rituximab plus alemtuzumab. Their detailed description is beyond the scope of this paper, since none of them has been proven to result in higher efficacy when compared to FCR.

**Combinations using alemtuzumab:** the synergistic activity of fludarabine and alemtuzumab was initially suggested by the induction of responses, including one CR, in 5 of 6 patients who were refractory to each agent alone. The combination of fludarabine and alemtuzumab (FA) was investigated in a phase II trial enrolling patients with relapsed CLL using a 4-weekly dosing protocol. This combination has been proven to be feasible, safe, and effective. Among the 36 patients, the ORR was 83% (30 of 36 patients), which included 11 CRs (30%) and 19 PRs (53%); there was one stable disease (SD). Sixteen of 31 evaluated patients (53%) achieved MRD-negativity in the peripheral blood by the 3-month follow up. Resolution of disease was observed in all disease sites, particularly in the blood, bone marrow and spleen. The FA therapy was well tolerated. Infusion reactions (fever, chills and skin reactions) occurred primarily during the first infusions of alemtuzumab, and were mild in the majority of patients.

Two phase III trials tested alemtuzumab in combination with FC (FCA) or fludarabine (FA). One trial comparing FCA to FCR in first-line therapy was closed prematurely due to the higher toxicity and treatment-related mortality observed in the FCA arm. The therapeutic efficacy of FCR was clearly superior to FCA. In this trial, alemtuzumab was given subcutaneously. A second randomized trial compared FA to fludarabine monotherapy in previously treated patients with relapsed or refractory CLL. In this trial, alemtuzumab was given intravenously. FA (n=168) resulted in better PFS than fludarabine monotherapy (n=167; median 23.7 months vs. 16.5 months; HR 0.61; $P=0.0003$) and OS (median not reached vs. 52.9 months; HR 0.65; $P=0.021$) compared with fludarabine alone. Adverse events occurred in 161 (98%) of 164 patients in the FA group and 149 (90%) of 165 in the fludarabine alone group. Patients in the FA group had more cutemegavirus (CMV) events (14% vs. <1%) and grade 1 or 2 infusion-related adverse reactions (62% vs. 13%). Major grade 3 or 4 toxicities in the FA and monotherapy groups were leukopenia (74% vs. 34%), lymphopenia (94% vs. 33%), neutropenia (59% vs. 68%), thrombocytopenia (11% vs. 17%), and anemia (9% vs. 17%). The incidence of serious adverse events was higher in the FA group (33% vs. 25%); deaths due to adverse events were similar between the two groups (6% vs. 12%). Two phase II trials investigated alemtuzumab in combination with high-dose corticosteroids, which seem to be a good therapeutic option, in particular for patients with del(17p). The combination of alemtuzumab and methylprednisolone was tested in the UK CLL206 trial on 17 untreated and 22 previously treated patients with del(17p). The ORR and CR rates were 85% and 36% in the whole cohort, and 88% and 65% in the treatment naïve patient group. In the CLL20 trial of the GCLLSG, the combination of alemtuzumab and high-dose dexamethasone resulted in an ORR above 90% in treatment naïve high-risk patients.

**New drugs targeting pathogenic pathways of CLL cells**

There are an increasing number of interesting new compounds in clinical development (for reviews see Wiestner et al. and Isfort et al.). The common denominator of these compounds is that their mechanism of action targets a relatively specific signaling abnormality (Figure 1A and B) or redirects the immune system against CLL cells. The new inhibitors may be able to correct the pathogenic imbalance between enhanced proliferative and reduced apoptotic signals that are a hallmark of CLL (Figure 1A). A detailed description of these fascinating agents is beyond the scope of this paper and has been given elsewhere. Table 1 illust-
trates selected, recent results of three of the most promising agents, which yield high response rates above 50% even in relapsed and refractory CLL patients. Some of these agents (e.g. obinutuzumab, CART19, ABT-199, idelalisib and ibrutinib) are currently arousing great enthusiasm and hope amongst CLL patients and their treating physicians.59-62

**Agents targeting B-cell receptor signaling**

**Idelalisib** (CAL-101): class I phosphatidylinositol 3-kinases (PI3Ks) regulate cellular functions relevant to oncogenesis.63 Expression of the PI3K p110 δ isoform (PI3Kδ) is restricted to cells of hematopoietic origin where it plays a key role in B-cell proliferation and survival. In CLL, the PI3K pathway is constitutively activated and dependent on PI3Kδ.64 CAL-101 is an oral PI3Kδ-isooform-selective inhibitor which promotes apoptosis in primary CLL cells in a time- and dose-dependent manner without inducing apoptosis in normal T cells or natural killer cells, and without diminishing antibody-dependent cellular cytotoxicity. CAL-101 inhibits CLL cell chemotaxis toward CXCL12 and CXCL13 and migration beneath stromal cells (pseudoe Emperorploiseis). CAL-101 also down-regulates secretion of chemokinase in stromal cocultures and after BCR triggering.64 CAL-101 reduces survival signals derived from the BCR or from nurse-like cells, and inhibits BCR- and chemokine-receptor-induced AKT and MAP kinase (ERK) activation.54

In a phase I clinical trial in 54 heavily pre-treated and high-risk CLL patients, idelalisib showed acceptable toxicity, positive pharmacodynamic effects and favorable clinical activity (high level of lymph node regression and prolonged duration of symptomatic tumor control).60 An ORR of 56% was achieved with 2 CR and 28 PR. Of the 28 patients with PR, 6 showed persistent lymphocytosis. The majority of patients (81%) showed a lymph node response (≥50% reduction in the nodal SPD). Median PFS was 17 months. Side effects were mild, with fatigue, diarrhea, pyrexia, rash and upper respiratory tract infections being the most frequent. More importantly, there were no dose-limiting toxicities.

Idelalisib has been tested in a combination with rituximab versus rituximab plus placebo in a randomized trial including 220 CLL patients with decreased renal function, previous therapy-induced myelosuppression, or major co-existing illnesses.65 Patients received rituximab and either idelalisib (at a dose of 150 mg) or placebo twice daily. The primary end point was PFS. At the first pre-specified interim analysis, the study was stopped early on the recommendation of the DSMB owing to overwhelming efficacy. The median PFS was 5.5 months in the placebo group and was not reached in the idelalisib group (HR for progression or death in the idelalisib group 0.15; 95% CI 0.05-0.37). Patients receiving idelalisib versus those receiving placebo had improved rates of overall response (81% vs. 13%; odds ratio 29.92; 95% CI 9.88-90.00) and overall survival at 12 months (92% vs. 80%; HR for death 0.28; P=0.02). Serious adverse events occurred in 40% of the patients receiving idelalisib and rituximab, and in 35% of those receiving placebo and rituximab. These encouraging results show the potential of combining idelalisib with rituximab in particular in patients with relapsed CLL who are less able to undergo chemotherapy.

**Ibrutinib** (PCI-32765): bruton tyrosine kinase (BTK) leads to downstream activation of cell survival pathways such as NF-kB and MAP kinases via Src family kinases.65 Ibrutinib is an orally active small molecule inhibiting BTK that plays a role in the signal transduction of the B-cell receptor (BCR). Inhibition of BTK might induce apoptosis in B-cell lymphomas and CLL cells.65 Ibrutinib showed significant activity in patients with relapsed or refractory B-cell malignancies including CLL.66 Data of a phase Ib-II multicenter study with single agent ibrutinib on 85 patients with relapsed or refractory CL or small lymphocytic lymphoma, the majority of whom had high-risk disease, were published recently.67 Patients received ibrutinib
once daily (n=51 at a dose of 420 mg, n=34 at 840 mg). Side-effects were mild (predominantly grade 1 or 2) and included transient diarrhea, fatigue, and upper respiratory tract infection. The overall response rate (ORR) was 71%, the majority being partial remissions (68%). Most interestingly, the response was independent of clinical and genomic risk factors, including advanced-stage disease, the number of previous therapies, and del(17p). At 26 months, the estimated PFS rate was 75% and OS rate was 83%. This study illustrates that ibrutinib may soon become an additional treatment option for CLL patients with high-risk genetic lesions.

### Bcl-2 inhibitors

Proteins in the B-cell CLL/lymphoma 2 (Bcl-2) family are key regulators of the apoptotic process. The Bcl-2 family comprises proapoptotic and prosurvival proteins. Shifting the balance toward the latter is an established mechanism whereby cancer cells evade apoptosis. Bcl-2, the founding member of this protein family, is encoded by the BCL2 gene which was initially described in follicular lymphoma as a protein in translocations involving chromosomes 14 and 18.

**Bcl-2 inhibitors** ABT-263 (Navitoclax) and ABT-199: ABT-263 is a small molecule Bcl-2 family protein inhibitor that binds with high affinity (Ki ≤ 1 nM) to multiple anti-apoptotic Bcl-2 family proteins including Bcl-XL, Bcl-2, Bcl-w, as well as Bcl-B and has a high oral bioavailability. Initial studies showed very promising results for this drug as a single agent. However, its therapeutic use seemed somewhat limited by severe thrombocytopenia, being a prominent side effect. Therefore, the compound was re-engineered to create a highly potent, orally bioavailable and Bcl-2-selective inhibitor, ABT-199. This compound inhibits the growth of Bcl-2 dependent tumors in vivo and spares human platelets. A single dose of ABT-199 in 3 patients with refractory chronic lymphocytic leukemia resulted in tumor lysis within 24 h. In a recent update on a phase I study, data of 56 patients have been reported of whom 16 (29%) had a del(17p) and 18 (32%) had F-refractory CLL. Major side effects were tumor lysis syndrome and neutropenia. ABT-199 yielded an ORR of 85%, with 13% CR and 72% PR. Interestingly, 88% and 75% of patients with a del(17p) and F-refractory CLL, respectively, achieved at least a PR. Together, these data indicate that selective pharmacological inhibition of Bcl-2 holds great promise for the treatment of CLL.

### Immunomodulatory drugs

**Lenalidomide:** lenalidomide has shown encouraging results in the treatment of high-risk patients including carriers of a del(17p). In 58% of the patients, lenalidomide causes a tumor flare reaction characterized by a marked and painful increase in lymph node size, malaise and fever. This tumor flare reaction may be life-threatening and is more common in CLL than in other lymphoid malignancies. Lenalidomide may also cause relevant myelosuppression. The overall response rate of lenalidomide monotherapy varied between 32% and 54% in different clinical trials. Most importantly, it seems to have activity as a single agent in fludarabine refractory CLL.

The combination of lenalidomide and rituximab seems to increase the response rate without a higher risk of toxicity, even in patients with del(17p) and/or unmutated IGHV-status. In a phase II trial, 59 patients with relapsed or refractory CLL received a combination of lenalidomide and rituximab. Lenalidomide was started on Day 9 of the first cycle at 10 mg orally and administered daily continuously. Each cycle was 28 days long. Rituximab was administered for 12 cycles; lenalidomide could be continued if patients showed clinical benefit. The overall response rate was 66%, including 12% complete responses and 12% nodular partial remissions. The median time to treatment failure was 17.4 months. The most common grade 3 or 4 toxicity was neutropenia (73% of patients). Fourteen patients (24%) experienced a grade 3 to 4 infection or a febrile episode. In essence, this combination seems a helpful alternative for patients with refractory CLL and warrants further investigation.

In some contrast, the combination of lenalidomide, rituximab and fludarabine may induce severe side-effects (myelosuppression) if all drugs are started simultaneously on Day 1. Using combinations starting with lenalidomide on Day 8 of the first cycle, the treatment is usually better tolerated.

**Chimeric antigen receptors**

Chimeric antigen receptors (CARs) combine the antigen recognition domain of an antibody with intracellular signaling domains into a single chimeric protein. CD19 is an ideal target for CARs since expression is restricted to normal and malignant B cells. Inclusion of the CD137 (4-1BB) signaling domain resulted in potent antitumor activity and in vivo persistence of anti-CD19 CARs in mice. It was recently shown that anti-tumor activity of CAR-modified autologous T cells targeted to CD19 (CART19 cells) yielded sustained responses even in high-risk CLL patients. Recently, the outcome and longer follow up from 10 patients treated with CART19 cells was reported. Autologous T cells collected by leukapheresis were transduced with a lentivirus encoding anti-CD19 scFv linked to 4-1BB and CD3-z signaling domains. Gene-modified T cells were expanded and activated ex vivo by exposure to anti-CD3/CD28 beads. Ten patients have received CART19 cells; 9 adults (median age 65 years, range 51-78 years) were treated for relapsed refractory ALL. Three of 9 CLL patients had a deletion of the p53 gene. All CLL patients received lymphodepleting chemotherapy 4-6 days before infusions (FC, PC or bendamustine, while the ALL patient had an ALC <10 after prior chemotherapy and did not require further lymphodepletion). CART19 homed to the marrow in the CLL patients and marrow and cerebrospinal fluid (CSF) for the ALL patient with detectable CART19 cells in the CSF (21 lymphocytes/µL, 78% CAR+) at Day 23 after infusion. Four of 9 evaluable patients achieved a CR (3 CLL, 1 ALL). Two CLL patients had a PR lasting three and five months, and 3 patients did not respond. In the 4 patients
who achieved CR, maximal expanded cells in the blood were detected at an average of 27-fold higher than the infused dose (range 21-40-fold) with maximal in vivo expansion between Day 10 and Day 31 post infusion. No patient with CR has relapsed. All patients who responded developed a cytokine release syndrome (CRS) manifested by fever, and variable degrees of nausea, anorexia, and transient hypotension and hypoxia. In responding CLL patients, cytokine levels were increased. Five patients with cytokine release required treatment. In summary, CART19 cells can induce potent and sustained responses for patients with advanced, refractory and high-risk CLL. However, the long-term toxicity and efficacy of this approach needs to be further studied.

**Treatment algorithm with currently approved agents**

Given the impressive number of choices, selecting the treatment for a given CLL patient requires experience, a good clinical assessment of the patient and an appropriate use of diagnostic tools. The following parameters should be considered before recommending a treatment for CLL.  

1. The clinical stage of disease.
2. The genetic risk of the leukemia.
3. The fitness of the patient.
4. The treatment situation (first- vs. second-line, response vs. non-response of the last treatment).

It is generally accepted that asymptomatic patients at early stages (Binet A and B, Rai 0-II) should not be treated outside of clinical trials. Whether some of the novel agents create some benefit when given at early stages still needs to be tested in clinical trials.

In patients with advanced stages (Binet C) or with symptomatic disease, treatment should be initiated. For these patients, the following recommendations can be given (Figure 1A and B).

**First-line treatment**

In a patient with advanced stages (Binet C, Rai III-IV) or active, symptomatic disease, treatment should be initiated (Figure 2A). Before treatment, patients need to be evaluated for their physical condition (or comorbidity). For patients in good physical condition (“go go”) as defined by a normal creatinine clearance and a low score at the cumulative illness rating scale (CIRS), patients should be offered FCR chemoimmunotherapy in order to achieve sustained remissions.

Patients with a somewhat impaired physical condition (“slow go”) should be offered a mild chemotherapeutic such as chlorambucil (or bendamustine) in combination with an anti-CD20 antibody.

Patients with symptomatic disease and with del(17p) or TP53 mutations may still receive FCR or BR or an alemtuzumab-containing regimen as first-line treatment. These regimens yield response rates above 50%, but the time to progression tends to be shorter than two years. The recent results obtained with single-agent ibrutinib in refractory or relapsed patients with a del(17p) showed an overall response rate of 68%, with a PFS at 26 months of 57% and an OS of 70%. While these results appear very encouraging, none of these therapies promises long-lasting remissions. Therefore, an allogeneic stem cell transplantation should still be discussed in patients with sufficient physical fitness.

**Second-line treatment**

As a general rule, the first-line treatment may be repeated if the duration of the first remission exceeds 24 months, provided that the first-line therapy was well tolerated (Figure 1B).

The choice becomes more difficult and limited in treatment-refractory CLL, in patients relapsing within 24 months post treatment, or in cases with the chromosomal aberration del(17p). In principle, the initial regimen should be changed. The following options exist as a relapse therapy for patients no longer responding to chemoimmunotherapy with rituximab.
1. Alemtuzumab alone or in combinations, in particular with high-dose steroids.\textsuperscript{22,49,52}
2. Combinations of antibodies with lenalidomide (see below).
3. BTK inhibitors such as ibrutinib,\textsuperscript{65} where available, or experimental protocols using novel agents (Figure 1B).
4. Allogeneic stem cell transplantation with curative intent.\textsuperscript{86}

The choice of one of these options depends on the fitness of the patient, the availability of some drugs and the prognostic risk of the leukemia as defined by molecular cytogenetics. According to recommendations of an EBMT consensus group, physically fit patients with refractory CLL or with a del(17p) should be offered an allogeneic transplantation, since their prognosis so far has remained extremely poor with conventional therapies.\textsuperscript{86} Finally, it is important to emphasize that patients with refractory disease should be treated within clinical trials whenever possible. Some of the new drugs, in particular ibrutinib or ABT-199, show good responses in these risk patients and could become an additional option in the very near future.

The future: treatment of CLL without chemotherapy?

The current challenge is to identify the optimal combination and sequence of the available therapeutic agents to achieve a long-term control of CLL with an optimal quality of life. Currently available data strongly suggest that achieving a very good disease control (using MRD-negativity as a surrogate parameter) prolongs survival of CLL patients.\textsuperscript{34,87} Therefore, I have recently proposed a strategy of combining the best agents in a sequence that tailors the treatment according to the initial tumor load and the response to therapy (CR vs. PR; MRD positive vs. MRD negative).\textsuperscript{87} This, what we call “sequential triple T” strategy (tailored, targeted, total eradication of MRD)\textsuperscript{88} would aim at preventing the outgrowth of adverse leukemic subclones\textsuperscript{88} and at minimizing the use of chemotherapy, thereby reducing the risk for secondary mutations of the CLL clone(s) and for secondary malignancies that are frequent and prognostically unfavorable events in CLL. This “sequential triple T” approach will make use of all the currently available options in a non-aggressive, non-toxic way and will aim at the complete elimination or control of the malignant clone. The approach would be available for most (fit as well as non-fit) CLL patients due to its limited toxicity. It could be given in an outpatient setting. Finally, it would use the current treatment options in a tailored and response-adjusted manner and, therefore, use the drugs in a cost-effective, resource saving way (Figure 3). It might also be used to monitor the evolution of new genetic subclones in CLL that may have clinical and prognostic impact.\textsuperscript{88} The proposed “sequential triple T” strategy

---

**Potential future strategies to achieve long-term control of CLL**

**Debulking**
- Mild chemotherapy with agents like bendamustine or fludarabine

1-2 months (1-2 courses)

**Induction** (combination therapy)
- Kinase inhibitor(s)
- Antibody
- Bcl2 antagonist

6-12 months

**MRD tailored maintenance** (single agents)
- Antibody
- Lenalidomide
- Kinase inhibitor
- Bd2 antagonist

1 year - ∞

---

**Figure 3.** Future treatment concept called “sequential triple T” to illustrate that this approach is comprised of a sequence of tailored measures (according to the risk of the leukemia, the tumor burden, the fitness of the patients), uses targeted agents (i.e. using the novel non-chemotherapeutic agents whose mechanism of action is targeting pathogenic signaling events of CLL cells and their microenvironment), and aims at the total of the leukemic clone (as assessed by MRD-negativity as a clinical end point). The “sequential triple T” strategy is a concept that is currently being tested in several trials of the GCLLSG. Please note that the drugs or classes of drugs in this figure are shown as examples; similar agents of the same class or additional classes of targeted drugs may be used as well.
might consist of three steps: 1) debulking; 2) induction; and 3) maintenance (Figure 3). This strategy is a concept of the GCLLSG that is currently being tested in several trials. It should be stressed that at the present time, with so many exciting therapeutic options for CLL, all hematologists and oncologists should feel the obligation to include their patients in clinical trials to ensure that maximal progress is achieved in the shortest possible time. By doing so, we will likely achieve our historic mission of gaining control over what have so far been incurable diseases such as CLL.

References


