INTRODUCTION

The introduction of fluorescence in situ hybridization (FISH) of interphase nuclei in the routine diagnostics of chronic lymphocytic leukemia (CLL) has greatly advanced the evaluation of genetic abnormalities. Interphase FISH also allows the detection of genetic abnormalities in nondividing cells.\(^1\) By FISH, genomic aberrations are detected in approximately 80% of CLL cases with a disease-specific probe set. If more than just 1 chromosomal aberration is detected, according to the hierarchical model of Döhner and colleagues,\(^2\) the prognosis is determined by the most unfavorable alteration. Deletions of the short arm of chromosome 17 (17p deletion) are found in 3% to 10% of CLL cases at diagnosis and/or with first-line treatment indication\(^2\)-\(^4\) and in 30% to 50% of relapsed/refractory CLL.\(^5\),\(^6\)

Breakpoints are distributed over the 17p10-p11.2 region.\(^7\) TP53 is located in band 17p13.1, is always affected, and is the centerpiece of pathogenic importance of 17p deletions; 80% to 90% of cases with monoallelic 17p deletion harbor TP53 mutation...
on the remaining allele. *TP53* mutation occurs in 8% to 15% of patients at first-line treatment and up to 35% to 50% of cases in refractory CLL.5,8,9

**RISK STRATIFICATION**

Genomic abnormalities identify subgroups of CLL patients with different time to progression and survival. It is widely acknowledged that patients with 17p deletion and/or *TP53* mutation belong to the ultra–high-risk group. In a multivariate analysis of 100 patients with B-cell CLL, B-prolymphocytic leukemia, or Waldenström macroglobulinemia, *TP53* mutation was the strongest prognostic factor for survival and predicted nonresponse to purine analogs, such as fludarabine and pentostatin.4 Another study of 53 patients with B-cell CLL had reported an association of *TP53* gene mutations with poor clinical outcome and drug resistance.10 In a prospective study of 560 untreated patients, samples were analyzed by conventional cytogenetics. Abnormalities of chromosome 17 were correlated to poor prognosis and observed as the only cytogenetic finding with independent prognostic value in multivariate analysis.11 In a cohort of 325 patients with CLL analyzed by FISH, 17p deletions were found in 7%. In the multivariate analysis, 17p deletion was identified as a significant prognostic factor: patients with 17p deletion had the worst prognosis.2

Nevertheless, there seem to be prognostic differences in the 17p-deleted subgroup itself. There is evidence that acquired 17p deletion by clonal evolution harbors a poor prognosis.12–15 A retrospective study of 99 CLL patients with 17p deletion at diagnosis showed clinical heterogeneity; some patients had an indolent clinical course. No progression within 18 months was a favorable sign for long-term stable disease. Risk factors for poor survival in this study were Rai stage 1 or higher, an unmutated immunoglobulin heavy chain variable (IGHV) status, and a 17p deletion in more than 25% of nuclei.16 Another study described a small subset of patients with loss of *TP53* and stable disease at a median follow-up of 64 months. All patients with an indolent clinical course had mutated IGHV status.17

The cutoff values for determining 17p deletion by FISH vary (usually 3%–12%) and have to be established in every laboratory by hybridization of normal controls.3,4,15,16,18 In one study, initially a level of 20% of 17p-deleted cells was defined as a critical threshold3,19; however, this 20% threshold seems to have resulted from technical artifact. Based on the same patient cohort, the cutoff was later revised and set to 10%.18

The clone with a 17p deletion might be suppressed by effective therapy: 15 patients with 17p deletion at diagnosis and response to therapy (nodular partial remission [PR] or complete remission [CR]) had no evidence for 17p deletion at the time of response. At the time of disease relapse or progression, however, the vast majority of patients again had 17p deletions.16

**THERAPEUTIC APPROACH**

17p deletion and *TP53* mutation are prognostic markers for nonresponse to conventional chemotherapy. Thus, alternative therapeutic approaches are needed, which act independently of the p53 signaling pathway (Fig. 1). As discussed previously, there are also indolent clinical courses in the group of patients with 17p deletion and, therefore, criteria for treatment initiation should be obeyed as set forth in the recommendations of the International Workshop on Chronic Lymphocytic Leukemia.20 Outside clinical trials, treatment approaches for patients with 17p deletion and/or *TP53* mutation are chemoimmunotherapy, such as fludarabine, cyclophosphamide, and rituximab (FCR); alemtuzumab; and allogeneic stem cell transplantation. Novel treatment strategies, like targeted therapies, can be offered in clinical trials.
Fludarabine/Cyclophosphamide/Rituximab

Chemoimmunotherapy with FCR is today the standard of care in treatment-naive physically fit CLL patients. In the CLL8 trial, 17p deletions were found in 10% (29/306) of patients in the fludarabine and cyclophosphamide (FC) arm versus 7% (22/315) of patients in the FCR arm. A significant improvement in overall response rate (ORR) and progression-free survival (PFS) was demonstrated for FCR versus FC treatment (Fig. 2). This was not the case for CR and 3-year overall survival (OS) (see Fig. 2, Table 1).

**Fig. 1.** Novel therapeutic approaches focusing on CLL biology.

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**Fig. 2.** CLL8 trial: OS in the FCR arm for subgroups defined by genomic aberrations. Patients with 17p deletion show the worst prognosis. (From Hallek M, Fischer K, Fingerle-Rowson G, et al. Addition of rituximab to fludarabine and cyclophosphamide in patients with chronic lymphocytic leukemia: a randomised, open-label, phase 3 trial. Lancet 2010;376:1164–74; with permission.)
These experiences with the FCR combination show that physically fit patients with 17p-deleted CLL may derive some benefit from the current standard regimen for first-line therapy.

**Alemtuzumab**

The CD52 antibody, alemtuzumab, has been approved for the treatment of fludarabine-refractory CLL in the United States and European Union since 2001 and, since 2007/2008, for first-line treatment of CLL patients. In this context, it has generally been used primarily for those who are not expected to respond to fludarabine, primarily patients with 17p deletion and/or TP53 mutation. The producing company, however, has recently withdrawn the drug from the market.

Alemtuzumab has proved its efficacy, especially in high-risk CLL patients. One randomized trial investigated efficacy and safety of intravenous alemtuzumab compared with chlorambucil in CLL first-line treatment22; 297 patients were included. Alemtuzumab was significantly superior regarding the ORR (83% vs 55%, \( P < .0001 \)) and rate of complete remissions (24% vs 2%, \( P < .0001 \)). Alemtuzumab was able to eliminate minimal residual disease in 11 of 36 complete responders.

Another important trial on alemtuzumab evaluated efficacy, safety, and clinical benefit of alemtuzumab in 93 fludarabine-refractory CLL patients who had been exposed to alkylating agents.23 The ORR was 33% with 2% CR and 31% PR; the median OS was 16 months. There was no information about cytogenetics. The CLL2H trial of the German CLL Study Group (GCLLSG) showed that subcutaneous administration of alemtuzumab in the refractory situation is equally effective as intravenous administration and harbors less adverse effects.5 A comparatively high number of patients had a 17p deletion (31/103; 30%). Between the different cytogenetic subgroups there were no significant differences in ORR, OS (Fig. 3), PFS, or time to treatment failure. This finding confirms the efficacy of alemtuzumab independent of the p53 signaling pathway.

The combination of alemtuzumab with dexamethasone followed by alemtuzumab maintenance therapy or allogeneic stem cell transplantation in ultra–high-risk CLL (17p deletion or fludarabine refractoriness) was investigated in the CLL2O trial of the GCLLSG.24 The results were updated at the American Society of Hematology annual meeting 2012; 70 of 131 eligible patients had a 17p deletion (31/103; 30%). Between the different cytogenetic subgroups there were no significant differences in ORR, OS (Fig. 3), PFS, or time to treatment failure. This finding confirms the efficacy of alemtuzumab independent of the p53 signaling pathway.

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| Table 1 | CLL8 trial: response and survival data for FC and FCR treatment of 17p deletion groups |
|---|---|---|
| 17p Deletion | FC | FCR | \( P \) Value |
| CR | 0/29 (0%) | 1/22 (5%) | 0.43 |
| ORR | 10/29 (34%) | 15/22 (68%) | 0.025 |
| PFS at 3 y | 0 | 18% | 0.019 |
| OS at 3 y | 37% | 38% | 0.25 |

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study in 39 patients with TP53 deletion tested the combination of alemtuzumab with methylprednisolone\textsuperscript{26}; 39 patients (17 untreated, 22 previously treated) with 17p (TP53) deletion were included. The ORR was 85% and the CR rate was 36%. Concerning the previously untreated subgroup, the CR rate was even higher with 65% versus 14% in previously treated patients ($P = .003$). Also, PFS was longer in untreated compared with previously treated patients (18.3 vs 6.5 months, $P = .010$). Grades 3 to 4 infections occurred in 51% of the patients. In summary, the combination of Alemtuzumab with high-dose steroids led to high response rates, but survival was still inferior to the patients without TP53 alterations in the CLL8 trial of the GCLLSG.

**Allogeneic Stem Cell Transplantation**

According to the European Group for Blood and Marrow Transplantation transplant consensus, indications for allogeneic hematopoietic stem cell transplantation in CLL include patients with p53 abnormalities requiring treatment.\textsuperscript{27} In these patients, transplantation should be considered early during the course of the disease.

Several investigations have shown graft-versus-leukemia activity in CLL.\textsuperscript{28–31} A retrospective review of European Group for Blood and Marrow Transplantation data was conducted to assess the curative potential of allogeneic hematopoietic stem cell transplantation in CLL patients with 17p deletion.\textsuperscript{32} The course of 44 patients with a median age of 54 years was analyzed. The 3-year OS and PFS rates were 44% and 37%. CR after HCT was ongoing in 9 patients with a follow-up time between 4 and 8.5 years. The CLL3X trial of the GCLLSG investigated the long-term outcome of reduced-intensity conditioning allogeneic stem cell transplantation in CLL patients\textsuperscript{33}; 13 of 72 (18%) patients who proceeded to transplantation harbored a 17p deletion. With a median follow-up of 43 months, 7 of these had still an ongoing complete remission. The 4-year event-free survival and OS rates were 45% and 59%. Neither event-free survival nor OS was significantly different between the various cytogenetic subgroups, implicating that allogeneic stem cell transplantation may overcome the cytogenetic risk profile.
In summary, allogeneic stem cell transplantation should be recommended as a consolidation strategy for patients with 17p deletion requiring therapy with younger age, good performance status, and preferentially a remission after induction therapy as well as few prior therapies.

**Flavopiridol**

Flavopiridol (alvocidib) is a cyclin-dependent kinase inhibitor, which induces apoptosis in CLL cells, independently of p53, and decreases the expression of antiapoptotic molecules, such as Mcl-1 and XIAP. A prospective study in 64 relapsed CLL patients with 21 (33%) 17p-deleted cases showed no association of 17p deletion with response ($P > .50$), which was defined as CR, nodular PR, or PR. Furthermore, there was no significant difference in PFS between present and absent 17p deletion (12.1 vs 10.3 months; $P = .94$). In a recently published follow-up of 112 patients, the ORRs did not differ between the cytogenetic subgroups ($P = .17$). CLL with 17p deletion showed an ORR of 48%. Also, PFS was not different between the subgroups with 10.4 months for 17p-deleted patients. Nevertheless, the risk for progression increased over time for patients with 17p deletion or 11q deletion: at 24 months, just 4% of 17p-deleted patients versus 24% in the group without 17p or 11q deletions were still progression-free. The median OS in 17p-deleted patients was 19.8 months and again not different from other cytogenetic subgroups.35 The further commercial development of flavopiridol was stopped, however, based on results of a pivotal trial,36 which showed satisfactory clinical activity in a high-risk subgroup but also considerable toxicity.

**Lenalidomide**

Lenalidomide is an orally bioavailable immunomodulatory drug derived from thalidomide. Its effects in CLL are mediated by stimulation of natural killer cells and cytotoxic T cells and by influencing the tumor microenvironment. Two adverse events are characteristic: tumor flare and tumor lysis. Two phase II trials showed the efficacy of lenalidomide in relapsed or refractory CLL with ORRs of 47% and 32%, respectively.37,38 A separate analysis of cases with high-risk cytogenetics (17p or 11q deletion) was published for the first trial; 6 of 45 patients had a 17p deletion. In this small subgroup, no CR and just one PR were achieved.39 In the second trial, 8 of 44 (18%) patients had a 17p deletion. Overall response in these patients was 13%. A recent phase I trial identified 20 mg as the maximum tolerated dose in relapsed and refractory CLL.40 Neutropenia and thrombocytopenia were the most common adverse events. Another phase II trial of lenalidomide combined with rituximab in 59 patients with relapsed and refractory CLL showed an ORR of 66%, with 12% CR and 12% nodular PR; 25% of the patients had a 17p deletion and the ORR in this subgroup was 53%. Moreover, patients with 17p deletion without refractoriness had a similar time to failure as patients with neither 17p deletion nor refractoriness (Fig. 4).41 Currently, lenalidomide is being further investigated in first-line trials and as maintenance therapy after successful induction treatment (eg, CLLM1 trial of the GCLLSG).

**Ofatumumab**

Ofatumumab is a type I human anti-CD20 antibody (IgG1) with enhanced complement-dependent cytotoxicity. The drug has been approved since 2010 for the treatment of CLL refractory to fludarabine and alemtuzumab. In a phase 2 trial, ofatumumab was investigated in combination with FC in previously untreated patients. Ofatumumab was administered at doses of either 500 mg or 1000 mg and 8 of 61 (13%) patients had a 17p deletion. The OR rate in this subgroup was 63%; the CR
rate was 13%. Overall, the ORR was 75% and CR rate was 41%. Time-to-event analyses were limited by small patient numbers.42

As a single-agent, ofatumumab was investigated in fludarabine-refractory CLL; 138 patients were included in the interim analysis of the single-arm study, which led to the approval of ofatumumab by the FDA. The cohort was divided into 2 subgroups: fludarabine-refractory and alemtuzumab-refractory patients (FA-ref) and fludarabine-refractory patients with bulky disease (BF-ref). In 17p-deleted patients, ORR was 41% in the FA-ref subgroup and 14% in the BF-ref subgroup. Comparing subgroups based on pretreatment characteristics, 17p deletion was the only factor significantly associated with a lower response rate (P = .0073) just in the BF-ref patients.43

Ibrutinib

Ibrutinib (PCI-32765) is an orally bioavailable Bruton tyrosine kinase (BTK) inhibitor that modulates B-cell receptor signaling.44 Loss-of-function mutations cause X-linked (Bruton) agammaglobulinemia with loss of B cells and lack of serum immunoglobulins. BTK inhibition in vitro leads to modest apoptosis induction, decreased proliferation, migration, and abrogation of downstream pathways, such as nuclear factor \( \kappa \)B signaling.44–46 Clinically, transient increase in lymphocytosis and rapid reduction of lymphadenopathy are typical findings in patients treated with ibrutinib, highlighting in vivo mechanisms of action in the tissue compartment.45,46

In a first-dose escalation study, 56 patients with relapsed or refractory B-cell malignancies (B-cell non-Hodgkin lymphoma, CLL, or Waldenström macroglobulinemia) were included. In this heavily pretreated phase I cohort, 11 of 16 CLL/small lymphocytic lymphoma patients achieved responses (2 of them CRs). Ibrutinib was well tolerated with self-limiting adverse events of grades 1 and 2. The median PFS over all cohorts was 13.6 months. Despite rapid absorption and elimination, BTK was occupied by ibrutinib for at least 24 hours, which is in accordance with irreversible inhibition.47

In a phase Ib/II study, 116 treatment-naive or relapsed/refractory CLL patients were treated with 2 fixed doses of ibrutinib (420 mg and 840 mg daily until disease progression). In the high-risk subgroup with either relapse within 2 years after combination chemoinmunotherapy or 17p deletion, the ORR was 50% with all of them...
PRs. Response was independent of poor risk features with an ORR of 67% in relapsed/refractory patients with 17p deletion (3% CR, 64% PR, 20% PR with lymphocytosis). PFS seems somewhat inferior, however, in CLL with 17p deletion (Fig. 5).48

Another study evaluated the combination of ibrutinib plus rituximab in 40 high-risk patients (17p deletion/TP53 mutation or PFS less than 36 months after front-line chemoinmunotherapy or relapsed CLL with 11q deletion); 19 patients had either a 17p deletion or TP53 mutation. Median follow-up time was 4 months. At 3 months, 17 of 20 patients available for response assessment had a PR (ORR 85%).49

**ABT-263 and ABT-199**

ABT-263 (Navitoclax) is an orally bioavailable BH3-mimetic that induces apoptosis by binding to the antiapoptotic BCL-2 family members, BCL-2, BCL-XL, and BCL-W. Compared with ABT-263, the second-generation BH3-mimetic, ABT-199, is a potent selective BCL-2 inhibitor; thus, direct toxic effects on thrombocytes are reduced. An interim analysis of a phase I/IIa study of ABT-263 in 29 patients with relapsed or refractory CLL showed clinical activity in patients with refractory disease and in patients with bulky disease and 17p deletion.50 A PR was achieved in 35% of 26 patients treated with doses of at least 110 mg daily. The median PFS was 25 months, which is astonishing in view of the high-risk nature of this population. Thrombocytopenia was the most common dose-limiting toxicity. Due to its higher specificity to BCL-2, the successor drug, ABT-199, is expected to exhibit similar efficacy paired with improved tolerability.

**GS-1101**

Activation of the B-cell receptor signaling pathway is essential for the survival of CLL cells (Fig. 6).51 Phosphatidylinositol 3-kinase (PI3K) plays a major role in this signaling pathway and is involved in important cellular processes, such as proliferation,
differentiation, migration, and adhesion. In in vitro studies, the orally bioavailable specific PI3Kδ inhibitor, GS-1101 (idelalisib; formerly CAL-101), induced apoptosis in CLL cells by inhibition of B-cell receptor signaling. Results of a phase I trial, combining GS-1101 with rituximab and/or bendamustine in 51 patients with relapsed or refractory CLL, have recently been presented at the American Society of Hematology annual meeting. Similar to ibrutinib treatment, a rapid reduction of lymphadenopathy is accompanied by transient lymphocytosis. The ORR was 78% (rituximab), 82% (bendamustine), and 87% (rituximab/bendamustine) in the 3 treatment groups, with 1-year PFS rates of 74%, 88%, and 87%, respectively. GS-1101 seemed to have activity in CLL with 17p deletion because responses were observed in this subgroup. Several phase III trials of GS-1101 in combination compared with the partner therapy alone are currently recruiting.

SUMMARY

CLL with 17p deletion and/or TP53 mutation remains a major therapeutic challenge due to poor response to conventional approaches, rapid disease progression, and short survival. Although allogeneic hematopoietic stem cell transplantation offers promising long-term results, this approach is not suitable for elderly patients with comorbidities, who represent the largest portion of CLL patients. Alemtuzumab is not only effective but also of considerable toxicity and its approval in the United States and Europe has been withdrawn for economic reasons. Despite satisfactory clinical activity, the development of the cyclin-dependent kinase inhibitor, flavopiridol, has been stopped. Recently, many novel agents with a favorable risk profile, avoiding the p53 signaling pathway, have entered clinical trials: the immunomodulatory drug, lenalidomide; PI3K inhibitors, such as GS-1101; and BH3 mimetics, such as ABT-263 and ABT-199, as well as the BTK inhibitor, ibrutinib. These agents show promising results in phase I/II trials of refractory CLL, including in patients with 17p deletion/TP53, and are, therefore, specifically being investigated in this ultra–high-risk population at desperate unmet therapeutic need.

Fig. 6. The B-cell receptor signaling pathway as a target for novel therapeutic strategies (eg, fostamatinib, GS-1101, and ibrutinib). (From Wiestner A. Targeting B-Cell receptor signaling for anticancer therapy: the bruton’s tyrosine kinase inhibitor ibrutinib induces impressive responses in B-Cell malignancies. J Clin Oncol 2013;31:128–30; with permission.)
REFERENCES


