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Special Report

iwCLL guidelines for diagnosis, indications for treatment, response assessment, and supportive management of CLL

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The previous edition of the consensus guidelines of the International Workshop on Chronic Lymphocytic Leukemia (iwCLL), published in 2008, has found broad acceptance by physicians and investigators caring for patients with CLL. Recent advances including the discovery of the genomic landscape of the disease, the development of genetic tests with prognostic relevance, and the detection of minimal residual disease (MRD), coupled with the increased availability of novel targeted agents with impressive efficacy, prompted an international panel to provide updated evidence- and expert opinion–based recommendations. These recommendations include a revised version of the iwCLL response criteria, an update on the use of MRD status for clinical evaluation, and recommendations regarding the assessment and prophylaxis of viral diseases during management of CLL. (*Blood*. 2018;131(25):2745-2760)

Introduction

In 2008, the International Workshop on Chronic Lymphocytic Leukemia (iwCLL) published consensus guidelines for the design and conduct of clinical trials for patients with CLL that were revised from those previously published by the National Cancer Institute–sponsored Working Group.[1-3] Those guidelines provided definitions intended to standardize the assessment of patients that were adopted by the US Food and Drug Administration and European Medicines Agency for the evaluation of new drugs. Since the publication of those guidelines, there have been major advances in the biology and treatment of patients with CLL, prompting the iwCLL to evaluate and revise the 2008 criteria.

The following major changes or additions were introduced in these updated guidelines.

- The clinical relevance of the recent discoveries on the genomic alterations found in CLL, including mutations of the *TP53* gene.
- The increasingly important prognostic role of the immunoglobulin variable heavy chain mutational status.
- The current use of clinical staging, novel genetic or biological prognostic markers, and prognostic scores.
- An improved assessment of splenomegaly, hepatomegaly and lymphadenopathy, which was harmonized with the relevant sections of the updated lymphoma response guidelines.
- An updated response assessment for novel targeted drugs (kinase inhibitors, Bcl2 inhibitors) that need to be evaluated during continuous therapy.
- The increasing role of assessing minimal residual disease.
- Updates regarding the baseline assessment and prophylaxis of viral diseases before and under therapy of CLL.

1. Diagnosis of CLL

The World Health Organization classification of hematopoietic neoplasias describes CLL as leukemic, lymphocytic lymphoma, being only distinguishable from small lymphocytic lymphoma (SLL) by its leukemic manifestation.[4] In the World Health Organization classification, CLL, by definition, is always a disease of neoplastic B cells, whereas the entity formerly described as T-CLL is now called T-cell prolymphocytic leukemia.[5]

It is important to verify that the patient has CLL and not some other lymphoproliferative disease that can masquerade as CLL, such as hairy cell leukemia or leukemic manifestations of

mantle cell lymphoma, marginal zone lymphoma, splenic marginal zone lymphoma with circulating villous lymphocytes, or follicular lymphoma. To achieve this, it is necessary to evaluate the blood smear, the immunophenotype, and, in some cases, the genetic features of the circulating lymphoid cells (see sections 1.1, 1.2, and 1.3).

1.1. Blood

The diagnosis of CLL requires the presence of $\geq 5 \times 10^9$/L B lymphocytes in the peripheral blood, sustained for at least 3 months. The clonality of these B lymphocytes needs to be confirmed by demonstrating immunoglobulin light chain restriction using flow cytometry. The leukemia cells found in the blood smear are characteristically small, mature lymphocytes with a narrow border of cytoplasm and a dense nucleus lacking discernable nucleoli and partially aggregated chromatin. Gumprecht nuclear shadows, or smudge cells, found as cellular debris, are additional morphologic features commonly associated with CLL. A small percentage of larger or atypical cells or prolymphocytes can be found admixed with morphologically typical CLL cells. Finding ≥55% prolymphocytes would favor a diagnosis of prolymphocytic leukemia; however, this diagnosis remains difficult and is solely based on morphological criteria, because no reliable immunological or genetic marker has been identified. A significant proportion of circulating prolymphocytes (≥10%) seems to indicate a more aggressive form of CLL (with *NOTCH1* or genetic *TP53* aberrations).[6]

CLL or SLL might be suspected in otherwise healthy adults who have an absolute increase in clonal B lymphocytes, but who have $<5 \times 10^9$/L B lymphocytes in the blood. However, in the absence of lymphadenopathy or organomegaly (as detected by physical examination or imaging studies), or of disease-related cytopenias or symptoms, the presence of $<5 \times 10^9$/L B lymphocytes is defined as monoclonal B lymphocytosis (MBL).[7] The presence of a cytopenia caused by a typical marrow infiltrate establishes the diagnosis of CLL regardless of the number of peripheral blood B lymphocytes or of the lymph node involvement. MBL has been observed to progress to CLL, requiring treatment at a rate of 1% to 2% per year.[8,9] Subjects with MBL appear to share an increased risk of secondary cancers with CLL patients, in particular of the skin, and should be encouraged to participate in the appropriate screening programs (eg, for carcinomas of the skin or colon).[9]

The definition of SLL requires the presence of lymphadenopathy and the absence of cytopenias caused by a clonal marrow infiltrate. Additionally, the number of B lymphocytes in the peripheral blood should be $<5 \times 10^9$/L. In SLL, the diagnosis should be confirmed by histopathological evaluation of a lymph node biopsy or biopsy of other tissues. Some patients may present with enlarged lymph nodes that are not suspicious for solid tumors and with peripheral blood B lymphocytes $<5 \times 10^9$/L that carry a typical CLL immunophenotype (see section 1.2). In these cases, a tissue or lymph node biopsy to establish the diagnosis of SLL may have limited clinical consequences and be omitted.

1.2. Immunophenotype

CLL cells coexpress the surface antigen CD5 together with the B-cell antigens CD19, CD20, and CD23. The levels of surface immunoglobulin, CD20, and CD79b are characteristically low compared with those found on normal B cells.[10-12] Each clone of leukemia cells is restricted to expression of either κ or λ immunoglobulin light chains.[10] The expression of CD5 can also be observed in other lymphoid malignancies, however, such as mantle cell lymphoma.[13] A recent, large harmonization effort has confirmed that a panel of CD19, CD5, CD20, CD23, κ, and λ is usually sufficient to establish the diagnosis.[14] In borderline cases, markers such as CD43, CD79b, CD81, CD200, CD10, or ROR1 may help to refine the diagnosis.[14]

1.3. Other tests

The tests described in this section are not essential to diagnose CLL, but may help predict the prognosis or assess the tumor burden. Of these different tests, only a few are needed to establish a prognostic profile in addition to the clinical staging (see section 2.3). Moreover, the indication for treatment does *not* depend on the results of these tests, but on the patient's clinical stage and symptoms (see section 4).

1.3.1. Molecular genetics Interphase fluorescence in situ hybridization (FISH) can be performed with peripheral blood lymphocytes and identifies cytogenetic lesions in >80% of all CLL cases.[15] The most common deletions are in the long arm of chromosome 13 (del(13q)). Additional, frequent chromosomal aberrations comprise trisomy of chromosome 12 and deletions in the long arm of chromosomes 11 (del(11q)) and in the short arm of chromosome 17 (del(17p)).[15]

Appropriate stimulation of CLL cells in vitro has enabled the performance of conventional karyotyping with enhanced reliability.[16] With this methodology, additional chromosomal aberrations of potential prognostic significance can be identified.[16-18] Moreover, stimulated metaphase karyotyping has demonstrated that leukemia cells with a complex karyotype (ie, ≥3 chromosomal abnormalities) may have adverse prognostic significance.[19-22] However, more data from prospective trials are needed to validate the prognostic and predictive value of stimulated metaphase karyotyping before we can recommend it for routine practice (Table 1).

Furthermore, FISH and conventional karyotyping can help distinguish CLL from other lymphoproliferative diseases, which have distinct disease-associated chromosomal abnormalities (eg, t(11;14), which is usually associated with mantle cell lymphoma). So far, other technologies such as array-based assays or next-generation sequencing have not been able to completely replace FISH or conventional karyotyping.

Prospective clinical trials indicate that certain genetic abnormalities are associated with adverse outcomes in response to standard chemo(immuno)therapy regimens. Patients with leukemia cells that carry del(17p) and/or *TP53* mutations (as determined by DNA sequencing, with a cutoff of 10%)[23] have an inferior prognosis and appear relatively resistant to standard chemotherapy regimens using alkylating drugs and/or purine analogs.[23-26] In a retrospective analysis of several chromosomal aberrations detected by FISH, patients who had CLL cells with chromosomal aberrations del(11q) and del(17p) had an inferior outcome compared with that of patients who had leukemia cells with a normal karyotype or del(13q) as the sole genetic abnormality.[15] On the other hand, patients who have leukemia

Table 1. Baseline evaluation of patients with CLL

Diagnostic test	General practice	Clinical trial
Tests to establish the diagnosis		
CBC and differential count	Always	Always
Immunophenotyping of peripheral blood lymphocytes	Always	Always
Assessment before treatment		
History and physical, performance status	Always	Always
CBC and differential count	Always	Always
Marrow aspirate and biopsy	When clinically indicated (unclear cytopenia)	Desirable
Serum chemistry, serum immunoglobulin, and direct antiglobulin test	Always	Always
Chest radiograph	Always	Always
Infectious disease status	Always	Always
Additional tests before treatment		
Molecular cytogenetics (FISH) for del(13q), del(11q), del(17p), add(12) in peripheral blood lymphocytes	Always	Always
Conventional karyotyping in peripheral blood lymphocytes (with specific stimulation)	NGI*	Desirable
TP53 mutation	Always	Always
IGHV mutational status	Always	Always
Serum β_2-microglobulin	Desirable	Always
CT scan of chest, abdomen, and pelvis	NGI	Desirable
MRI, PET scans	NGI	NGI
Abdominal ultrasound†	Possible	NGI

General practice is defined as the use of accepted treatment options for a CLL patient not enrolled on a clinical trial.

CBC, complete blood count; MRI, magnetic resonance imaging; NGI, not generally indicated; PET, positron emission tomography.

*Conventional karyotyping in peripheral blood lymphocytes (with specific stimulation) may be useful before therapy, if established methodology is available.

†Used in some countries to monitor lymphadenopathy and organomegaly.

cells with del(17p) and/or *TP53* mutation respond poorly to chemo(immuno)therapy but fare significantly better when treated with nonchemotherapeutic agents, such as small molecule inhibitors of BTK, phosphatidylinositol 3-kinase, or BCL2. It has been demonstrated that the progression-free survival and overall survival of CLL patients carrying a del(17p) and patients carrying a *TP53* mutation, as detected by Sanger sequencing in the absence of del(17p), are similar.[27] Therefore, the assessment of both del(17p) and *TP53* mutation has prognostic and predictive value and should guide therapeutic decisions in routine practice. For clinical trials, it is recommended that molecular genetics be performed before treating a patient on protocol. Because additional genetic abnormalities may be acquired during the course of the disease,[28] genetic analyses (in particular for del(17p)/*TP53* mutations) should be repeated before any subsequent second- or third-line treatment.

Next-generation whole exome or whole genome sequencing have identified additional genomic abnormalities, such as mutations in *NOTCH1* or *SF3B1* that have pathogenic as well as prognostic significance. However, more data from prospective trials are needed to validate the prognostic and predictive value of these genomic abnormalities before we can advocate using them in routine practice.

1.3.2. Mutational status of IGHV and variable heavy stereotypes The leukemia cells use immunoglobulin variable heavy chain (IGHV) genes that may or may not have undergone somatic mutations.[29-31] The outcome of patients with leukemia cells that use an unmutated IGHV gene (usually defined as 98% or more sequence homology to the nearest germ line gene) is inferior to that of patients with leukemia cells that use a mutated IGHV gene.[32,33] Moreover, the presence of mutated IGHV genes, in particular when combined with additional prognostic factors such as favorable cytogenetics or attainment of a minimal residual disease (MRD) negative state after therapy, characterizes a CLL patient subgroup with excellent outcome following chemoimmunotherapy with fludarabine, cyclophosphamide, and rituximab.[34-36]

The discovery of almost identical or "stereotyped" B-cell receptor immunoglobulins among unrelated CLL patients suggests that (auto)antigen selection may play a role in disease pathogenesis.[37] Approximately one-third of patients can be grouped into subsets based on shared sequence motifs within the IGHV region complementarity determining region 3.[37] It seems that some of these subgroups share a similar prognosis. For example, *IGHV3-21* gene usage (of stereotype subset 2) may be associated with an unfavorable prognosis independent of the IGHV mutational status.[38,39] As of today, assessment of IGHV stereotypes is not an element of the routine prognostic work up in CLL.

1.3.3. Immunophenotypic markers Leukemia cell expression of ZAP-70 and CD38 correlates with the expression of unmutated IGHV genes and can be associated with poor prognosis.[25,40-47] The association between expression of ZAP-70 or CD38 with the presence of unmutated IGHV genes is not absolute, and discordant cases are more frequently found in patients with high-risk cytogenetics.[48] CD49d, the α chain of the alpha4beta1 integrin heterodimer, has been associated with an unfavorable prognosis in CLL and was shown to be the strongest flow cytometry-based predictor of overall survival and treatment-free survival in a large, multicenter effort.[49,50]

1.3.4. Serum markers Several studies have found that serum markers such as levels of soluble CD23, thymidine kinase, and β_2-microglobulin are associated with overall survival or progression-free survival.[51-58] Of these, β_2-microglobulin has retained independent prognostic value in several multiparameter scores.[55,58,59] Assays for these markers should be standardized and used in prospective clinical trials to validate their relative value in the management of patients with CLL.

1.3.5. Marrow examination In CLL, typically >30% of the nucleated cells in the aspirate are mature lymphoid cells. The extent and pattern of marrow infiltration (diffuse vs nondiffuse) may reflect the tumor burden.[60] A marrow aspirate and biopsy generally are *not* required for the diagnosis of CLL; however, a marrow biopsy and aspirate can help clarify whether cytopenias (neutropenia, anemia, thrombocytopenia) are related or unrelated to leukemic infiltration of the marrow. In these cases, a marrow biopsy may provide important information, in particular before starting therapies with cytotoxic agents. It is recommended to repeat a marrow biopsy in patients with persisting cytopenia after treatment to clarify disease- vs therapy-related causes. A marrow biopsy is mandatory to confirm a complete remission (CR; see section 5.1).

2. Clinical staging

There are 2 widely accepted staging systems for use in both patient care and clinical trials: Rai[61] and Binet.[62] The original Rai classification was modified to reduce the number of prognostic groups from 5 to 3.[63] As such, both systems now describe 3 major subgroups with distinct clinical outcomes. These 2 staging systems are simple, inexpensive, and can be readily and consistently applied by physicians worldwide. Both rely solely on a physical examination and standard laboratory tests and do not require imaging studies.

2.1. Rai staging system

The modified Rai classification defines low-risk disease as occurring in patients who have lymphocytosis with leukemia cells in the blood and/or marrow (formerly considered Rai stage 0). Patients with peripheral blood lymphocytosis, enlarged lymph nodes in any site, and splenomegaly and/or hepatomegaly (lymph nodes being palpable or not) are defined as having intermediate-risk disease (formerly considered Rai stage I or II). High-risk disease includes patients with disease-related anemia (as defined by a hemoglobin [Hb] level < 1 g/dL) (formerly stage III) or thrombocytopenia (as defined by a platelet count of $<100 \times 10^9$/L; formerly stage IV).

2.2. Binet staging system

The Binet staging system is based on the number of involved lymphoid areas, as defined by the presence of enlarged lymph nodes ≥1 cm in diameter or organomegaly, and on whether there is anemia or thrombocytopenia.

Areas of involvement considered for staging include:

1. Head and neck, including the Waldeyer ring (this counts as 1 area, even if ≥1 group of nodes is enlarged).
2. Axillae (involvement of both axillae counts as just 1 area).
3. Groins, including superficial femorals (involvement of both groins counts as just 1 area).
4. Palpable spleen.
5. Palpable liver (clinically enlarged).

Stage A. Hb ≥10 g/dL and platelets $\geq 100 \times 10^9$/L and up to 2 of these areas involved.

Stage B. Hb ≥10 g/dL and platelets $\geq 100 \times 10^9$/L and 3 or more of the lymphoid areas involved.

Stage C. Hb <10 g/dL and/or a platelet count $<100 \times 10^9$/L.

2.3. Use of additional prognostic factors in practice including prognostic scores

In daily practice, Rai or Binet stages help stratify patients according to the disease risk; however, there are a large number of biomarkers that can provide additional prognostic information.[64-66] The most relevant prognostic parameters are *IGHV* mutational status, serum β_2-microglobulin, and the presence of del(17p) and/or *TP53* mutations. Usually, high-risk CLL is defined, at least in part, by a genetic aberration of the *TP53* gene (ie, del(17p) or *TP53* mutation).

Following the identification of new prognostic parameters, several prognostic scores and stratification systems have been proposed based on multivariate analyses to extract the most significant independent prognostic information from the plethora of known prognostic markers.[55,58,59] These models are very useful to identify high-risk patient populations for experimental protocols, but also those patients with a very good prognosis even at advanced stages. One of these prognostic scores, the CLL international prognostic index (CLL-IPI) consists of a weighed score that includes the clinical stage, age, *IGHV* mutational status, serum β_2-microglobulin, and the presence of del(17p) and/or *TP53* mutations.[59] It was originally developed using datasets of ≥4500 patients treated within or outside of clinical trials, divides 4 different prognostic subgroups, and has been validated extensively in various cohorts.[67-73] The value of prognostic markers or scores might change with the application of novel therapies.

3. Eligibility criteria for clinical trials

The selection of CLL patients for clinical trials is similar to that for patients with other malignancies. Phase 1-2 clinical trials commonly, although not invariably, are intended for patients who have received prior therapy. The inclusion of patients with SLL in clinical trials for CLL is encouraged. The combination of new agents with standard therapy as part of phase 2 studies may be investigated in both untreated and previously treated patients. Phase 3 clinical trials are used to compare the clinical outcome using new treatment modalities with that attained using current standard therapy. Other requirements for eligibility with respect to age, clinical stage, performance status, biomarkers, organ function, or status of disease activity should be defined for each study.

3.1. Performance status and fitness

Before inclusion in a trial, the performance status by the Eastern Cooperative Oncology Group (ECOG) should be determined. Future clinical trials involving elderly patients ideally should

assess the comorbidity (fitness) and/or functional activity of patients by appropriate scores.[74-76] The cumulative illness rating scale has been used successfully in the setting of multicenter clinical trials to characterize patients with concomitant morbidity,[77] although other scores to identify unfit patients are available as well.[78]

3.2. Organ function eligibility for clinical trials

Most chemotherapy agents have potential toxicity to the liver, kidneys, heart, lungs, nervous system, or other organ systems; therefore, organ function requirements should be guided by the known or suspected toxicity profile of each agent based on preclinical studies or prior clinical studies. Patients enrolled on protocols evaluating agents with known or suspected toxicity for a given organ should have the specific organ function documented before therapy.

3.3. Infectious disease status

The status of specific infectious diseases, as outlined in section 3.5, should be documented. Patients with active infections requiring systemic antibiotics or antifungal or antiviral drugs should have their infection controlled before initiating therapy in a clinical trial.

3.4. Second malignancies

Patients with active second malignancies generally are not considered candidates for entry into clinical trials. Second cancers in remission and nonmelanoma skin cancers should not necessarily be an exclusion criterion for clinical trials involving patients with CLL.

3.5. Required baseline evaluation

Parameters considered necessary for a complete baseline evaluation might differ depending on whether the patient is treated in a clinical protocol. A clear distinction is made in sections 3.5 and 5 between recommendations for routine practice and the requirements for clinical trials (Tables 1, 2, and 3). Unless indicated, recommendations are the same for clinical trials and routine practice. In general, baseline evaluation studies for defining these parameters should be performed within 2 weeks of clinical trial enrollment, except for molecular cytogenetics (FISH), marrow aspirate and biopsy, as well as computed tomography (CT) scans (see sections 3.5.1 and 3.5.2).

3.5.1. Essential baseline tests

3.5.1.1. Physical examination. The bidimensional diameters of the largest palpable lymph nodes in each of the following sites should be recorded: cervical, axillary, and inguinal (Table 1). The dimensions of the liver and spleen below their respective costal margins, as assessed by palpation, should also be recorded. Note that any of these manifestations of CLL, in particular hepatomegaly, could be caused by a variety of other diseases.

3.5.1.2. Assessment of performance status (ECOG score).

3.5.1.3. A complete blood cell count (white blood cell count, Hb, hematocrit, reticulocyte, and platelet count) and differential leukocyte count, including both percent and absolute number of lymphocytes. Reporting the proportion of prolymphocytes is desirable when these are present.

3.5.1.4. Marrow biopsy. Before initiating treatment within in a clinical trial, a unilateral marrow aspirate and biopsy are recommended. Repeat marrow biopsies may be compared with the baseline marrow specimen to assess the cause of cytopenias (eg, bone marrow toxicity, disease progression).

3.5.1.5. Serum chemistry (eg, creatinine, bilirubin, lactate dehydrogenase, haptoglobin, transaminases, alkaline phosphatase, β_2-microglobulin).

3.5.1.6. Serum immunoglobulin levels.

3.5.1.7. Direct antiglobulin test.

3.5.1.8. Chest radiograph (when a CT scan is not performed).

3.5.1.9. HIV serology. Patients who are infected with HIV should be given special consideration because of the potential risks for immune suppression with most antileukemia therapies and the potential for compounded myelotoxicity of treatment with antiretroviral therapy.

3.5.1.10. CMV. Therapies associated with the potential for reactivation of infection with cytomegalovirus (CMV), such as alemtuzumab, idelalisib, or allogeneic stem cell transplantation, should include plans for monitoring for active CMV disease and/or for providing anti-CMV therapy (eg, ganciclovir, valganciclovir).[79-81] These should cover screening or early diagnosis of CMV reactivation and its subsequent management. However, positive CMV serology does not represent a contraindication to treatment of CLL. As a general recommendation, patients treated with immune-suppressive agents (eg, alemtuzumab, idelalisib), should be monitored for CMV and be considered for antiviral therapy if found to have increased levels of CMV in the blood by the polymerase chain reaction (PCR), even in the absence of clinical symptoms. Moreover, antiviral therapy is recommended for infected patients with clinical symptoms of active CMV infection.

3.5.1.11. Hepatitis B and hepatitis C. Before initiating treatment, patients should be evaluated for infection with hepatitis B virus (HBV) or hepatitis C virus (HCV) because reactivation of HBV and HCV may occur following treatment with immunosuppressive or myelosuppressive drugs, including anti-CD20 antibodies. Patients found to be chronic carriers of HBV, as defined by positive surface antigen, HB core antibody, and/or low HBV titers in serum, should receive prophylactic antiviral agents while undergoing therapy for CLL with immunosuppressive drugs.[82] In patients with high titers of HBV or HCV DNA, initiation of antiviral therapy before antileukemic treatment should be considered.

3.5.2. Additional tests The following recommendations are made for clinical trials or for the assessment in specific clinical situations (Table 1).

3.5.2.1. Examination of leukemia-cell cytogenetics (eg, metaphase karyotyping, FISH [in particular for del(17p)]) and analysis for inactivating mutations in *TP53* should be performed before any line of therapy. The recommended threshold for reporting of mutations detected by next-generation sequencing should reflect the Sanger-like threshold of ~10% variant allele

Table 2. Recommendations regarding indications for treatment in CLL

	General practice	Clinical trial
Treat with Rai stage 0	NGI*	RQ
Treat with Binet stage A	NGI*	RQ
Treat with Binet stage B or Rai stage I or II	Possible*	Possible*
Treat with Binet stage C or Rai stage III or IV†	Yes	Yes
Treatment of active/progressive disease	Yes	Yes
Treat without active/progressive disease	No	RQ

General practice is defined as the use of accepted treatment options for a CLL patient not enrolled on a clinical trial. Early therapy of CLL is generally not recommended outside of clinical trials; however, we recognize the need to conduct clinical trials testing the early use of novel agents.

RQ, research question.

*Treatment is indicated, if the disease is active as defined in section 4.

†Anemia and/or thrombocytopenia from CLL-unrelated causes should be excluded.

frequency.[83] Patients with genetic *TP53* aberrations should not receive chemotherapy, if other options are available.

3.5.2.2. CT scans. CT scans generally are *not* required for initial evaluation or for follow-up. The staging of CLL does not use CT scans but relies on physical examination and blood counts. Enlarged lymph nodes, if detected only by CT scan, do not change the Binet or Rai stage. It has been shown that patients with Rai stage 0 but abdominal disease detectable by CT scans may have a more aggressive course.[84] This requires further investigation before recommending CT scans for routine initial evaluation of patients with CLL. On the other hand, it has been demonstrated that the majority of relapses or progressions in CLL are detected by physical examination and blood counts, not by imaging studies.[85] Moreover, the decision for relapse treatment was determined by imaging studies in only 1% of patients[85]; therefore, the routine follow-up evaluation of CLL patients does not require CT scans.

In clinical trials where the treatment intent is to maximize the overall response rate, neck, chest, abdominal, and pelvic CT scans are recommended to evaluate the response to therapy. One CT scan should be performed before the start of therapy and a second CT scan at the final response assessment (usually the first restaging after the end of therapy) within a study protocol, if abnormal at baseline. For the assessment of continuous therapies, CT scans should be performed at the time point of clinically evaluated maximal response or, alternatively, at a time point defined by the protocol. Additional, repetitive CT scan monitoring is usually not clinically relevant and potentially harmful for the patient.

3.5.2.3. Other imaging methods. Except in patients with proven or suspected Richter transformation, positron emission tomography (PET) scans do *not* provide information that is useful in the management of CLL.[86] Similarly, nuclear magnetic resonance imaging generally does *not* provide useful information beyond that of CT scanning in the management of CLL and therefore is not recommended outside of clinical trials.

3.5.2.4. Ultrasound imaging. In some countries, ultrasound imaging is used to assess the extent of lymphadenopathy and organomegaly in CLL. Although ultrasound imaging may be very useful in the clinical management of individual patients, the results obtained by this methodology are investigator-dependent and difficult to centrally verify. Therefore, ultrasound imaging is currently not recommended for response evaluation in clinical trials.

Table 3. Recommendations regarding the response assessment in CLL patients

Diagnostic test	General practice	Clinical trial
History, physical examination	Always	Always
CBC and differential count	Always	Always
Marrow aspirate and biopsy	At cytopenia of uncertain cause	At CR or cytopenia of uncertain cause
Assessment for minimal residual disease	NGI	Desirable
Ultrasound of the abdomen*	Possible, if previously abnormal	NGI
CT scans of chest, abdomen, and pelvis	NGI	Recommended if previously abnormal and otherwise with a CR and PR

For a detailed description of these parameters, see section 5. General practice is defined as the use of accepted treatment options for a CLL patient not enrolled on a clinical trial.

*Used in some countries to monitor lymphadenopathy and organomegaly.

3.5.2.5. A lymph node biopsy is generally *not* required, unless it is necessary for companion scientific studies or in rare cases in which the diagnosis is difficult. A lymph node or tissue biopsy is performed to establish the diagnosis of a transformation into an aggressive lymphoma (Richter transformation). A PET scan can be useful to identify the best lymphoid area for establishing the diagnosis by biopsy.

4. Indications for treatment

4.1. Primary treatment decisions

Criteria for initiating treatment may vary depending on whether the patient is treated in a clinical trial (Table 2). In general practice, patients with asymptomatic early-stage disease (Rai 0, Binet A), should be monitored without therapy unless they have evidence of disease progression or disease-related symptoms. Several studies have shown that treating patients with early-stage disease does not result in a survival benefit[87-90]; therefore, an early-intervention therapy with antileukemia drugs, including signaling inhibitors or BCL2 antagonists, alone or in combination with monoclonal antibodies, currently is not indicated.

Although patients with intermediate-risk (stages I and II) and high-risk (stages III and IV) disease according to the modified Rai classification or at Binet stage B or C usually benefit from the initiation of treatment, some of these patients (in particular, Rai intermediate risk or Binet stage B) can be monitored without therapy until they have evidence for progressive or symptomatic disease (summarized as "active disease").

Active disease should be clearly documented to initiate therapy. At least 1 of the following criteria should be met.

1. Evidence of progressive marrow failure as manifested by the development of, or worsening of, anemia and/or thrombocytopenia. Cutoff levels of Hb <10 g/dL or platelet counts $<100 \times 10^9$/L are generally regarded as indication for treatment. However, in some patients, platelet counts $<100 \times 10^9$/L may remain stable over a long period; this situation does not automatically require therapeutic intervention.
2. Massive (ie, ≥6 cm below the left costal margin) or progressive or symptomatic splenomegaly.
3. Massive nodes (ie, ≥10 cm in longest diameter) or progressive or symptomatic lymphadenopathy.
4. Progressive lymphocytosis with an increase of ≥50% over a 2-month period, or lymphocyte doubling time (LDT) <6 months. LDT can be obtained by linear regression extrapolation of absolute lymphocyte counts obtained at intervals of 2 weeks over an observation period of 2 to 3 months; patients with initial blood lymphocyte counts $<30 \times 10^9$/L may require a longer observation period to determine the LDT. Factors contributing to lymphocytosis other than CLL (eg, infections, steroid administration) should be excluded.
5. Autoimmune complications including anemia or thrombocytopenia poorly responsive to corticosteroids.
6. Symptomatic or functional extranodal involvement (eg, skin, kidney, lung, spine).
7. Disease-related symptoms as defined by any of the following:
   a. Unintentional weight loss ≥10% within the previous 6 months.
   b. Significant fatigue (ie, ECOG performance scale 2 or worse; cannot work or unable to perform usual activities).
   c. Fevers ≥100.5°F or 38.0°C for 2 or more weeks without evidence of infection.
   d. Night sweats for ≥1 month without evidence of infection.

Hypogammaglobinemia, or monoclonal or oligoclonal paraproteinemia does not by itself constitute a basis for initiating therapy. However, it is recommended to assess the change in these protein abnormalities, if patients are treated. Also, patients with CLL may present with a markedly elevated leukocyte count; however, leukostasis rarely occurs in patients with CLL. Therefore, the absolute lymphocyte count should not be used as the sole indicator for treatment.

4.2. Second- and subsequent-line treatment decisions

Disease relapse alone is not a criterion to restart therapy unless the disease is symptomatic (see active disease criteria). Asymptomatic increases of lymphocyte counts alone without other signs of progression are generally not an indication to restart therapy. Second- and subsequent-line treatment decisions should generally follow the same indications as those used for first-line treatment. Where the original indication for treatment has not resolved with initial therapy, provided treatment-related toxicities have recovered, it is reasonable to initiate second-line treatment without waiting for formal disease progression to be manifest. Also, the rate of disease progression after some newer therapies can be rapid; in such circumstances, it can be acceptable to initiate subsequent therapy before formal progression where there is substantial persisting disease burden.

Patients with any of the following features generally do not respond to second-line chemo(immuno)therapy: primary resistance to first-line chemo(immuno)therapy; time to progression after first-line, fludarabine-based chemo(immuno)therapy of 2 to 3 years[91,92]; or leukemia cells with del(17p)/*TP53* mutations. These patients should be offered a nonchemotherapy regimen and/or entrance into clinical trials. In selected cases, allogeneic hematopoietic stem cell transplantation should be considered.[93-95]

5. Definition of response, relapse, and refractory disease

Assessment of response should include a careful physical examination and evaluation of the blood and bone marrow (Tables 3 and 4). The timing of response assessment for therapies with a defined treatment duration (such as chemoimmunotherapeutic approaches) should be at least 2 months after completion of therapy. To define the response to therapy, 2 groups of parameters need to be assessed and documented: parameters of group A assess the lymphoid tumor load and constitutional symptoms; parameters of group B assess the hematopoietic system (Table 4).

For continued therapies or treatment strategies that contain a maintenance phase, the assessment of response should be performed at least 2 months after patients achieve their maximum response or at a time point that is predefined in the protocol; in this case, it is not necessary to interrupt therapy for response assessment. Maximum response can be defined as a treatment phase in which no additional improvement is seen during at least 2 months of therapy. In clinical trials, any response

Table 4. Response definition after treatment of CLL patients

Group	Parameter	CR	PR	PD	SD
A	Lymph nodes	None ≥1.5 cm	Decrease ≥50% (from baseline)*	Increase ≥50% from baseline or from response	Change of −49% to +49%
	Liver and/or spleen size†	Spleen size <13 cm; liver size normal	Decrease ≥50% (from baseline)	Increase ≥50% from baseline or from response	Change of −49% to +49%
	Constitutional symptoms	None	Any	Any	Any
	Circulating lymphocyte count	Normal	Decrease ≥50% from baseline	Increase ≥50% over baseline	Change of −49% to +49%
B	Platelet count	≥100 × 10^9/L	≥100 × 10^9/L or increase ≥50% over baseline	Decrease of ≥50% from baseline secondary to CLL	Change of −49 to +49%
	Hemoglobin	≥11.0 g/dL (untransfused and without erythropoietin)	≥11 g/dL or increase ≥50% over baseline	Decrease of ≥2 g/dL from baseline secondary to CLL	Increase <11.0 g/dL or <50% over baseline, or decrease <2 g/dL
	Marrow	Normocellular, no CLL cells, no B-lymphoid nodules	Presence of CLL cells, or of B-lymphoid nodules, or not done	Increase of CLL cells by ≥50% on successive biopsies	No change in marrow infiltrate

For a detailed description of the response parameters, see section 5.

*Sum of the products of 6 or fewer lymph nodes (as evaluated by CT scans and physical examination in clinical trials or by physical examination in general practice).

†Spleen size is considered normal if <13 cm. There is not firmly established international consensus of the size of a normal liver; therefore, liver size should be evaluated by imaging and manual palpation in clinical trials and be recorded according to the definition used in a study protocol.

CR, complete remission (all of the criteria have to be met); PD, progressive disease (at least 1 of the criteria of group A or group B has to be met); PR, partial remission (for a PR, at least 2 of the parameters of group A *and* 1 parameter of group B need to improve if previously abnormal; if only 1 parameter of both groups A and B is abnormal before therapy, only 1 needs to improve); SD, stable disease (all of the criteria have to be met; constitutional symptoms alone do not define PD).

(eg, CR, partial remission) should be sustained for at least 2 months before using this response in the assessment. In addition, where appropriate, a further assessment of response (ie, marrow assessment) may be performed at least 2 months after the patient has cleared MRD from the peripheral blood.

5.1. Complete remission

CR requires all of the following criteria (Table 4).

5.1.1. Peripheral blood lymphocytes (evaluated by blood and differential count) <4 × 10^9/L.

5.1.2. Absence of significant lymphadenopathy by physical examination. In clinical trials, a CT scan of the neck, abdomen, pelvis, and thorax is desirable if previously abnormal. Lymph nodes should be <1.5 cm in longest diameter. Once this is determined, further imaging should not be required until disease progression is apparent by clinical examination or on blood testing.

5.1.3. No splenomegaly or hepatomegaly by physical examination. In clinical trials, a CT scan of the abdomen should be performed at response assessment and should show no evidence for lymphadenopathy and splenomegaly. We propose to use a recent consensus response cutoff for splenomegaly of 13 cm in craniocaudal length.[96,97] However, the persistence of splenomegaly may not correlate with outcome.[96] The quantitative determination of hepatomegaly seems more difficult; changes such as focal or disseminated hepatic nodules support liver involvement.

5.1.4. Absence of disease-related constitutional symptoms.

5.1.5. Blood counts need to show the following values:

5.1.5.1. Neutrophils ≥1.5 × 10^9/L.

5.1.5.2. Platelets ≥100 × 10^9/L.

5.1.5.3. Hemoglobin ≥11.0 g/dL (without red blood cell transfusions).

5.1.6. MRD assessment.

In clinical trials aimed at maximizing the depth of remission, the presence of MRD after therapy should be assessed (see section 5.9). The sensitivity of the method used to evaluate for MRD should be reported, as should the tissue studied (blood or marrow). The proportion of patients achieving undetectable MRD should be reported with the total number of patients treated with the specific therapy as the denominator (not as a proportion of responders or those in CR).

5.1.7. For patients in clinical trials (Table 3). A bone marrow aspirate and biopsy should be performed if clinical and laboratory results listed in sections 5.1.1 to 5.1.5 demonstrate that a CR may have been achieved. To define a CR, the cytological or pathological evaluation of the bone marrow smear or biopsy must be at least normocellular for age, without evidence for typical CLL lymphocytes by morphological criteria. This evaluation is not based on a flow cytometry–based MRD assessment (see section 9).

In a clinical trial, the time point of marrow biopsy should be defined by the protocol. For example, in patients receiving chemo(immuno)therapy, the time point of marrow biopsy is typically 2 months posttherapy.

When performing marrow biopsies in clinical trials, lymphoid nodules can be found that may reflect residual disease.[98,99] These nodules may be recorded as "nodular partial remission." Immunohistochemistry may be performed to define whether the nodules comprise primarily T cells, B cells other than CLL cells, or CLL cells. If nodules are not composed of CLL cells, a CR can be documented provided all other criteria are met. If the marrow is hypocellular, a repeat determination should be performed 4 weeks or later, when peripheral blood counts have recovered; however, this interval should not exceed 6 months after the last treatment. In cases in which a marrow biopsy was obtained at baseline, a comparison of pre- vs posttherapy biopsies should be performed. In general practice, the use of a marrow biopsy for evaluating a CR is at the discretion of the physician.

In clinical trials aimed at maximizing the response rate, the quality of the response should be assessed in the marrow for MRD by highly sensitive molecular-based assays or immunophenotyping (see section 5.9).

5.1.8. Some patients fulfill all the criteria for a CR (including the marrow examinations described in section 5.1.7), but have a persistent anemia, thrombocytopenia, or neutropenia apparently unrelated to CLL, but related to drug toxicity. These patients should be considered as a different category of remission, CR with incomplete marrow recovery (CRi). For the definition of this category, the marrow evaluation (see section 5.1.7) should be performed with scrutiny and not show any clonal disease infiltrate. In clinical trials, patients having CR with incomplete marrow recovery should be monitored prospectively to determine whether their outcome differs from that of patients with detectable residual disease or with noncytopenic CR.

5.2. Partial remission

To define a partial remission, at least 2 parameters of group A *and* 1 parameter of group B need to improve, if previously abnormal (Table 4; sections 5.2.1 to 5.2.5). If only 1 parameter of both groups A and B was abnormal before therapy, only 1 needs to improve. Constitutional symptoms persisting for >1 month should be recorded.

5.2.1. A decrease in the number of blood lymphocytes to 50% or less from the value before therapy.

5.2.2. Reduction in lymphadenopathy compared with baseline (by cross-sectional imaging scans in clinical trials or by palpation in general practice) as defined by:

5.2.2.1. A decrease in lymph node size by 50% or more in

- the sum of the products of the same enlarged lymph nodes selected at baseline as assessed by imaging (an established number in clinical trials of lymph nodes has been up to 6).
- and the sum of longest diameters of the same enlarged lymph nodes selected at baseline as assessed by physical examination (an established number in clinical trials of lymph nodes has been a maximum of 6).

5.2.2.2. No increase in any lymph node and no new enlarged lymph node (diameter ≥1.5 cm). For small lymph nodes (longest diameter <1.5 cm), an increase <25% is not considered significant.

5.2.3. A regression ≥50% of the extent of enlargement of the spleen below the costal margin defined by palpation, or normalization in size. When assessed by CT, scan spleen size must have regressed by ≥50% in length beyond normal.[96] A persistence of splenomegaly posttherapy may have limited influence on outcome in CLL.[96]

5.2.4. A regression of ≥50% of the extent of enlargement of the liver below the costal margin defined by palpation, or normalization in size. Given the impact of numerous medical conditions, liver size by physical examination or CT scan is not a reliable measure of hepatic involvement by CLL and should only be counted if hepatomegaly is clearly attributable to lymphoid involvement.

5.2.5. The blood count should show 1 of the following results:

5.2.5.1. Platelet counts >100 × 10^9/L or 50% improvement over baseline.

5.2.5.2. Hb >11.0 g/dL or 50% improvement over baseline without red blood cell transfusions or erythropoietin support.

5.3. Progressive disease

Progressive disease (PD) during or after therapy is characterized by at least 1 of the following, when compared with nadir values (Table 4):

5.3.1. Lymphadenopathy. Progression of lymphadenopathy is often discovered by physical examination and should be recorded at regular intervals. In CLL, the use of imaging (CT scans) usually does not add much information for the detection of progression or relapse.[100] Disease progression occurs if 1 of the following events is observed.

- Appearance of any new lesion such as enlarged lymph nodes (≥1.5 cm), splenomegaly, hepatomegaly, or other organ infiltrates. Transient increases of lymph node size during treatment with novel inhibitors may occur and should not be counted as PD.
- An increase by ≥50% in greatest determined diameter of any previous site (≥1.5 cm).

5.3.2. An increase in the spleen size by ≥50% or the de novo appearance of splenomegaly. In the setting of splenomegaly, the splenic length must increase by ≥50% of the extent of its prior increase beyond baseline (eg, a 15-cm spleen must increase to ≥16 cm). If no prior splenomegaly was observed at baseline or if splenomegaly has resolved with treatment, the spleen must increase by at least 2 cm from baseline.[96]

5.3.3. An increase in the liver size of ≥50% of the extent enlargement of the liver below the costal margin defined by palpation,

Table 5. Grading scale for hematological toxicity in CLL studies

Grade*	Decrease in platelets† or Hb‡ (nadir) from baseline value, %	Absolute neutrophil count (nadir)§ × 10^9/L
0	No change to 10	≥2
1	11-24	≥1 and <2
2	25-49	≥1 and <1
3	50-74	≥0.5 and <1
4	≥75	<0.5

*Grades: 1, mild; 2, moderate; 3, severe; 4, life-threatening; 5, fatal. Death occurring as a result of toxicity at any level of decrease from baseline will be recorded as grade 5.

†Platelet counts must be below normal levels for grades 1-4. If, at any level of decrease the platelet count is <20 × 10^9/L, this will be considered grade 4 toxicity unless a severe or life-threatening decrease in the initial platelet count (eg, 20 × 10^9/L) was present at baseline, in which case the patient is not evaluable for toxicity referable to platelet counts.

‡Hb levels must be below normal levels for grades 1-4. Baseline and subsequent Hb determinations must be performed before any given transfusions. The use of erythropoietin is irrelevant for the grading of toxicity, but should be documented.

§If the absolute neutrophil count (ANC) reaches <1 × 10^9/L, it should be judged to be grade 3 toxicity. Other decreases in the white blood cell count or in circulating granulocytes are not to be considered because a decrease in the white blood cell count is a desired therapeutic end point. A gradual decrease in granulocytes is not a reliable index in CLL for stepwise grading of toxicity. If the ANC was <1 × 10^9/L before therapy, the patient is not evaluable for toxicity referable to the ANC. The use of G-CSF is irrelevant for the grading of toxicity, but should be documented.

or the de novo appearance of hepatomegaly. Given the impact of numerous medical conditions, liver size by physical examination or by CT scan is not a reliable measure of hepatic involvement by CLL and should only be counted if hepatomegaly is clearly attributable to lymphoid involvement.

5.3.4. An increase in the number of blood lymphocytes by 50% or more with at least 5 × 10^9/L B lymphocytes. Certain therapies (eg, kinase inhibitors) may cause lymphocytosis. In the setting of therapy with such agents, an increase in blood lymphocyte count by itself does not uniformly indicate an increased tumor burden, but may reflect redistribution of leukemia cells from lymphoid tissues to the blood. This should be predefined in the protocol of clinical trials for therapies in which redistribution of disease occurs. In such cases, increased lymphocytosis alone is not a sign of treatment failure or PD.[101]

5.3.5. Transformation to a more aggressive histology (Richter syndrome or Richter transformation). The diagnosis of Richter transformation should be established by lymph node or other tissue biopsy.

5.3.6. Occurrence of cytopenia (neutropenia, anemia, or thrombocytopenia) directly attributable to CLL and unrelated to autoimmune cytopenias.

5.3.6.1. During therapy. Cytopenias may occur as a side effect of many therapies and should be assessed according to Table 5. During therapy, cytopenias cannot be used to define disease progression. Each protocol should define the amount of drug(s) to be administered with such cytopenias.

5.3.6.2. Posttreatment. The progression of any cytopenia (unrelated to autoimmune cytopenia), as documented by a decrease of Hb levels ≥2 g/dL or <10 g/dL, or by a decrease of platelet counts ≥50% or <100 × 10^9/L, which occurs at least 3 months after treatment, defines disease progression, if the marrow biopsy is consistent with the cytopenia resulting from increased marrow infiltration of clonal CLL cells and is not considered a treatment related toxicity.

5.4. Stable disease

Patients who have not achieved a CR or a partial remission, and who have not exhibited PD, will be considered to have stable disease (which is equivalent to a nonresponse).

5.5. Treatment failure

Responses that should be considered clinically beneficial include CR and PR; all others (eg, stable disease, nonresponse, PD, death from any cause) should be rated as a treatment failure.

5.6. Time to next treatment, progression-free survival, event-free survival, and overall survival

Progression-free survival is defined as the interval between the first treatment day (in phase 3 trials: day of randomization for intent-to-treat analysis) to the first sign of disease progression or death from any cause. Event-free survival is defined as the interval between the first treatment day (in phase 3 trials: day of randomization for intent-to-treat analysis) to the first sign of disease progression or start of a new treatment or withdrawal from the trial because of toxicity or death (whichever occurs first). Overall survival is defined as the interval between the first treatment day (in phase 3 trials: day of randomization for intent-to-treat analysis) to death. Time to next treatment is defined as interval between the first treatment day until the patient starts an alternative therapy for progressive CLL.

Note that the response duration may be assessed during therapy for continuous treatment, in particular with oral agents, as well as after the end of treatment, in particular with chemo(immuno) therapy. Study protocols should provide detailed specifications of the planned time points for the assessment of the treatment response under continuous therapy. Response durations <6 months are not considered clinically relevant (see refractory disease, section 5.8).

5.7. Relapse

Relapse is defined as evidence of disease progression (see section 5.3) in a patient who has previously achieved the above criteria of a CR or partial remission (sections 5.1-5.2) for ≥6 months.

5.8. Refractory disease

Refractory disease is defined as treatment failure (as defined in section 5.5) or as progression within 6 months from the last dose of therapy.

5.9. Minimal residual disease

The complete eradication of the leukemia is a desired end point. Use of sensitive multicolor flow cytometry, PCR, or next-generation sequencing can detect MRD in many patients who achieved a complete clinical response. Prospective clinical trials have provided substantial evidence that therapies that are able to eradicate MRD usually result in an improved clinical outcome.[97,102-106] The techniques for assessing MRD have undergone a critical evaluation and have become well standardized.[107,108] Six-color flow cytometry (MRD flow), allele-specific oligonucleotide PCR, or high-throughput sequencing using the ClonoSEQ assay are reliably sensitive down to a level of <1 CLL cell in 10 000 leukocytes.[108] Refinement and harmonization of these technologies has established that a typical flow cytometry–based assay comprises a core panel of 6 markers (ie, CD19, CD20, CD5, CD43, CD79b, and CD81).[108] As such, patients will be defined as having undetectable MRD (MRD-neg) remission if they have blood or marrow with <1 CLL cell per 10 000 leukocytes. The blood generally can be used for making this assessment because the marrow will have detectable CLL when it is also found in the peripheral blood. However, there are therapies that preferentially clear the blood but not the marrow (such as monoclonal antibodies); therefore, it may be important to confirm that the marrow aspirate also is MRD-neg when the blood is found to be MRD-neg. Clinical trials aimed at maximizing the depth of remissions should include at least 1 test to assess for MRD, because the lack of leukemia persistence using these sensitive tests has a strong, positive prognostic impact. The report should be clear as to whether blood and/or marrow have been assessed and should report the proportion of MRD-neg patients on an intent-to-treat basis using the total number of patients in that treatment arm as the denominator (not those assessed or those who responded to treatment).

6. Factors requiring stratification at inclusion in a clinical phase 3 trial

6.1. Patients ideally should be stratified with regard to previous treatment vs no previous treatment, *and* as purine analog-sensitive vs purine analog-refractory in studies for which prior therapy is allowed.

6.2. If >1 clinical stage is allowed, patients ideally should be stratified with respect to clinical stage.

6.3. If the specific patient subgroup is not excluded, then they should be stratified or analyzed as a subgroup analysis based upon whether they have leukemia cells with del(17p) or del(11q), and for mutations of the *TP53* gene.

6.4. Patients should be stratified for the mutational status of the *IGHV* gene locus (mutated vs unmutated). If it is not possible to stratify prospectively, then the *IGHV* mutated and *IGHV* unmutated patients should be analyzed as a planned subgroup analysis.

7. Assessment of toxicity

Evaluation of treatment-related toxicity requires careful consideration of both the manifestations of the underlying disease and the anticipated adverse reactions to the agents used in therapy. For this reason, some of the conventional criteria used for assessing toxicity are not applicable to clinical studies involving patients with hematological malignancies in general or CLL in particular. An example is hematological toxicity; patients with advanced CLL generally have cytopenias that may be caused by the underlying CLL and/or prior therapy. A few recommendations are presented to help evaluate for treatment-induced toxicity in CLL.

7.1. Hematological toxicity

Evaluation of hematological toxicity in patients with CLL must take into consideration that many patients have low blood cell counts at the initiation of therapy. Therefore, the standard criteria used for solid tumors cannot be applied because many CLL patients then would be considered to have grade 2 to 4 hematological toxicity at the initiation of treatment. Furthermore, the absolute blood neutrophil counts are rarely used at the initiation of therapy to modify the treatment dose because these values typically are unreliable in CLL patients with lymphocytosis. However, the increasing use of more effective therapeutic agents, particularly those with neutropenia as a dose-limiting toxicity (eg, nucleoside analogs), can result in clinically significant myelosuppression. Therefore, the 1996 guidelines proposed a new dose-modification scheme for quantifying hematological deterioration in patients with CLL, which included alterations in the dose of myelosuppressive agents based on the absolute neutrophil count. This dose modification scheme has proven very helpful in the context of several large prospective trials in CLL and is therefore retained in the current version of the guidelines (Table 5).

7.2. Infectious complications

Patients with CLL are at increased risk for infection because of compromised immune function, which might be related to the disease itself and/or to the consequences of therapy. Nevertheless, the rate of infection following treatment can be used in assessing the relative immune-suppressive effects of a given therapy. The etiology of the infection should be reported and categorized as bacterial, viral, or fungal, and as proven or probable. The severity of infections should be quantified as minor (requiring either oral antimicrobial therapy or symptomatic care alone), major (requiring hospitalization and systemic antimicrobial therapy), or fatal (death as a result of the infection).

Particular attention should be given to monitoring for symptoms or laboratory evidence of opportunistic infections such as *Pneumocystis jirovecii* or *Herpesviridae* (herpes simplex virus, varicella-zoster virus, cytomegalovirus, Epstein-Barr virus) in patients treated with agents such as alemtuzumab and idelalisib (alone or in combination) or with allogeneic stem cell transplantation. Patients receiving anti-CD20 antibodies may experience reactivation of HBV infections.[109] HBV serological status should be evaluated before treatment with such agents; therefore, appropriate antiviral prophylaxis should be initiated in patients with a history of HBV infection.[109] In contrast, the infection rate seems low in patients age <65 years treated with fludarabine-based first-line

therapy, in which no monitoring or routine anti-infective prophylaxis is required.[110] Progressive multifocal leukoencephalopathy has been reported in a few CLL patients treated with anti-CD20 antibodies; therefore, infections with JC virus should be ruled out in situations of unclear neurological symptoms.[111-114]

7.3. TLS

Patients with CLL rarely experience tumor lysis syndrome (TLS) after therapy with purine analog–based regimens.[115] However, TLS might occur following treatment with drugs such as lenalidomide,[116] venetoclax,[117] or type II anti-CD20 monoclonal antibodies.[118] For this reason, patients in early-phase clinical trials should be monitored for possible TLS, which should be treated appropriately. If observed, the occurrence and severity of TLS should be recorded in clinical trials using established criteria.[119]

7.4. Nonhematological toxicities

Other nonhematological toxicities should be graded according to the latest version of the National Cancer Institute Common Terminology Criteria for Adverse Events. Some of the newer agents show a new spectrum of side effects that should be carefully monitored, including cardiac arrhythmias (for ibrutinib), autoimmune colitis (for idelalisib) or diarrhea (ibrutinib), bleeding (ibrutinib), and autoimmune pneumonitis (idelalisib).

8. Reporting of clinical response data

Clear and careful reporting of data are essential parts of any clinical trial. In clinical studies involving previously treated patients, patients who are relapse, or are refractory should be clearly distinguished. Relapse and refractory disease are defined previously (sections 5.7 and 5.8). For those patients who have relapsed, it is also useful to describe the quality and duration of their prior response.

9. Treatment end points

Given the recent increase of treatment options for CLL patients, the choice of treatment and the end points of clinical trials may depend on the fitness of the patients (see section 3.1). For example, the number of MRD-neg complete remissions or the overall survival might be appropriate end points in physically fit patients. In contrast, trials in patients with reduced physical fitness might choose the time to progression or health-related quality of life as trial end points. Moreover, the quality of life in patients with CLL may be reduced compared with that seen in the normal population and only moderately increased by some of the current treatment options.[120-122] Further studies assessing the health-related quality of life in CLL are encouraged.

10. Supportive care and management of complications

10.1. Indications for growth factors in CLL

While patients are under myelosuppressive chemo(immuno) therapy, growth factors such as granulocyte-colony-stimulating factor (G-CSF) should be given according to the guidelines of the American Society of Clinical Oncology.[123] G-CSF might also benefit patients who experience prolonged cytopenias following treatment with alemtuzumab. Similarly, some patients with anemia may benefit from erythropoiesis-stimulating factors, if used according to recently published guidelines.[124] However, CLL-related cytopenias are often efficiently corrected by an appropriate antileukemic therapy.

10.2. Prevention of infections by vaccination and immunoglobulin substitution

Infections are frequent problems during management of CLL patients. Excellent reviews regarding their prevention and therapy have been published recently.[125-127] Unfortunately, there are no randomized studies showing that vaccination may alter infection rates or outcomes from acquired infections in CLL. It is generally recommended that routine vaccinations be performed before initiation of treatment if possible. Vaccinations achieve reasonable rates of seroprotection and seroconversion in immunocompromised cancer patients, with minimal side effects.[127] Conjugate vaccines have proved to be highly immunogenic and are to be preferred, where available, in CLL patients.[128] Vaccines against seasonal influenza and H1N1 can be recommended, given the severity of the H1N1 pandemic and the highly severe flu impact in immunocompromised CLL patients.[129] Live vaccines are contraindicated in CLL patients because severe or even fatal complications have been reported.[125]

Hypogammaglobulinemia (low serum levels of IgG and IgA with variable IgM) is a well-recognized complication associated with CLL. Regarding the substitution of CLL patients with hypogammaglobulinemia and history of infections, 6 randomized studies have shown that the prophylactic use of intravenous immunoglobulins decreases the rate of bacterial infections and prolongs the time to first infection, but does not produce differences in survival or other outcome parameters (summarized in Sánchez-Ramón et al[125]). Therefore, the use of intravenous immunoglobulin cannot be routinely recommended, but should be reserved to individual situations of hypogammaglobulinemia and repeated infections.

10.3. AIHA or ITP

The relationship between CLL and autoimmune cytopenias is well established. The potential mechanisms, particularly the role of leukemic cells in stimulating the production of polyclonal autoantibodies, are increasingly understood.[130] Autoimmune thrombocytopenia (ITP) and autoimmune hemolytic anemia (AIHA) as a single abnormality caused by CLL initially should be treated with glucocorticoids and not initially with chemotherapy or targeted agents. Second-line treatment options for AIHA include rituximab, splenectomy, intravenous immunoglobulins, and/or immunosuppressive therapy with agents such as cyclosporine A, azathioprine, low-dose cyclophosphamide, or alemtuzumab.[131-133] Some ITP patients not responding to glucocorticoids may benefit from rituximab, immunosuppressive agents (eg, mycophenolate), or thrombopoietin analogs.[134] Refractoriness of autoimmune cytopenias to therapy is an indication for treatment directed at the underlying CLL.[135] In this regard, the Binet or Rai staging systems do not distinguish between ITP/AIHA or bone marrow infiltration as the cause for anemia or thrombocytopenia that results in classifying a patient as having stage C or high-risk disease.

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Footnote

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REFERENCES

1. Cheson BD, Bennett JM, Rai KR, et al. Guidelines for clinical protocols for chronic lymphocytic leukemia (CLL). Recommendations of the National Cancer Institute-sponsored working group. *Am J Hematol*. 1988;29(3):152-163.
2. Cheson BD, Bennett JM, Grever M, et al. National Cancer Institute-sponsored Working Group guidelines for chronic lymphocytic leukemia: revised guidelines for diagnosis and treatment. *Blood*. 1996;87(12):4990-4997.
3. Hallek M, Cheson BD, Catovsky D, et al; International Workshop on Chronic Lymphocytic Leukemia. 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.
4. Catovsky D, Müller-Hermelink HK, Montserrat E, Harris NL. B-cell prolymphocytic leukaemia. In: Harris NL, Stein H, Vardiman JW, eds. World Health Organization Classification of Tumours Pathology and Genetics of Tumours of Haematopoietic and Lymphoid Tissues. Lyon, France: IARC Press; 2001:131-132.
5. Catovsky D, Ralfkiaer E, Müller-Hermelink HK. T-cell prolymphocytic leukaemia. In: Harris NL, Stein H, Vardiman JW, eds. World Health Organization Classification of Tumours Pathology and Genetics of Tumours of Haematopoietic and Lymphoid Tissues. Lyon, France: IARC Press; 2001:195-196.
6. Oscier D, Else M, Matutes E, Morilla R, Strefford JC, Catovsky D. The morphology of CLL revisited: the clinical significance of prolymphocytes and correlations with prognostic/molecular markers in the LRF CLL4 trial. *Br J Haematol*. 2016;174(5):767-775.
7. Marti GE, Rawstron AC, Ghia P, et al; International Familial CLL Consortium. Diagnostic criteria for monoclonal B-cell lymphocytosis. *Br J Haematol*. 2005;130(3):325-332.
8. Rawstron AC, Bennett FL, O'Connor SJ, et al. Monoclonal B-cell lymphocytosis and chronic lymphocytic leukemia. *N Engl J Med*. 2008;359(6):575-583.
9. Strati P, Shanafelt TD. Monoclonal B-cell lymphocytosis and early-stage chronic lymphocytic leukemia: diagnosis, natural history, and risk stratification. *Blood*. 2015;126(4):454-462.
10. Moreau EJ, Matutes E, A'Hern RP, et al. Improvement of the chronic lymphocytic leukemia scoring system with the monoclonal antibody SN8 (CD79b). *Am J Clin Pathol*. 1997;108(4):378-382.
11. Ginaldi L, De Martinis M, Matutes E, Farahat N, Morilla R, Catovsky D. Levels of expression of CD19 and CD20 in chronic B cell leukaemias. *J Clin Pathol*. 1998;51(5):364-369.
12. Matutes E, Owusu-Ankomah K, Morilla R, et al. The immunological profile of B-cell disorders and proposal of a scoring system for the diagnosis of CLL. *Leukemia*. 1994;8(10):1640-1645.
13. Morice WG, Kurtin PJ, Hodnefield JM, et al. Predictive value of blood and bone marrow flow cytometry in B-cell lymphoma classification: comparative analysis of flow cytometry and tissue biopsy in 252 patients. *Mayo Clin Proc*. 2008;83(7):776-785.
14. Rawstron AC, Kreuzer KA, Soosapilla A, et al. Reproducible diagnosis of chronic lymphocytic leukemia by flow cytometry: an European Research Initiative on CLL (ERIC) & European Society for Clinical Cell Analysis (ESCCA) harmonisation project. *Cytometry B Clin Cytom*. 2018;94(1):121-128.
15. 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.
16. Buhmann R, Kurzeder C, Rehklau J, et al. CD40L stimulation enhances the ability of conventional metaphase cytogenetics to detect chromosome aberrations in B-cell chronic lymphocytic leukaemia cells. *Br J Haematol*. 2002;118(4):968-975.
17. Mayr C, Speicher MR, Kofler DM, et al. Chromosomal translocations are associated with poor prognosis in chronic lymphocytic leukemia. *Blood*. 2006;107(2):742-751.
18. Haferlach C, Dicker F, Schnittger S, Kern W, Haferlach T. Comprehensive genetic characterization of CLL: a study on 506 cases analysed with chromosome banding analysis, interphase FISH, IgV(H) status and immunophenotyping. *Leukemia*. 2007;21(12):2442-2451.
19. Puente XS, Beà S, Valdés-Mas R, et al. Non-coding recurrent mutations in chronic lymphocytic leukaemia. *Nature*. 2015;526(7574):519-524.
20. Landau DA, Tausch E, Taylor-Weiner AN, et al. Mutations driving CLL and their evolution in progression and relapse. *Nature*. 2015;526(7574):525-530.
21. Thompson PA, O'Brien SM, Wierda WG, et al. Complex karyotype is a stronger predictor than del(17p) for an inferior outcome in relapsed or refractory chronic lymphocytic leukemia patients treated with ibrutinib-based regimens. *Cancer*. 2015;121(20):3612-3621.
22. Herling CD, Klaumünzer M, Rocha CK, et al. Complex karyotypes and KRAS and POT1 mutations impact outcome in CLL after chlorambucil-based chemotherapy or chemoimmunotherapy. *Blood*. 2016;128(3):395-404.
23. Pospisilova S, Gonzalez D, Malcikova J, et al; European Research Initiative on CLL (ERIC). ERIC recommendations on TP53 mutation analysis in chronic lymphocytic leukemia. *Leukemia*. 2012;26(7):1458-1461.
24. Döhner H, Fischer K, Bentz M, et al. p53 gene deletion predicts for poor survival and non-response to therapy with purine analogs in chronic B-cell leukemias. *Blood*. 1995;85(6):1580-1589.
25. 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.
26. Stilgenbauer S, Schnaiter A, Paschka P, et al. Gene mutations and treatment outcome in chronic lymphocytic leukemia: results from the CLL8 trial. *Blood*. 2014;123(21):3247-3254.
27. Zenz T, Eichhorst B, Busch R, et al. TP53 mutation and survival in chronic lymphocytic leukemia. *J Clin Oncol*. 2010;28(29):4473-4479.
28. Shanafelt TD, Witzig TE, Fink SR, et al. Prospective evaluation of clonal evolution during long-term follow-up of patients with untreated early-stage chronic lymphocytic

leukemia. *J Clin Oncol*. 2006;24(28):4634-4641.

29. Schroeder HW Jr, Dighiero G. The pathogenesis of chronic lymphocytic leukemia: analysis of the antibody repertoire. *Immunol Today*. 1994;15(6):288-294.

30. Fais F, Ghiotto F, Hashimoto S, et al. Chronic lymphocytic leukemia B cells express restricted sets of mutated and unmutated antigen receptors. *J Clin Invest*. 1998;102(8):1515-1525.

31. Hashimoto S, Dono M, Wakai M, et al. Somatic diversification and selection of immunoglobulin heavy and light chain variable region genes in IgG+ CD5+ chronic lymphocytic leukemia B cells. *J Exp Med*. 1995;181(4):1507-1517.

32. Damle RN, Wasil T, Fais F, et al. Ig V gene mutation status and CD38 expression as novel prognostic indicators in chronic lymphocytic leukemia. *Blood*. 1999;94(6):1840-1847.

33. Hamblin TJ, Davis Z, Gardiner A, Oscier DG, Stevenson FK. Unmutated Ig V(H) genes are associated with a more aggressive form of chronic lymphocytic leukemia. *Blood*. 1999;94(6):1848-1854.

34. Rossi D, Terzi-di-Bergamo L, De Paoli L, et al. Molecular prediction of durable remission after first-line fludarabine-cyclophosphamide-rituximab in chronic lymphocytic leukemia. *Blood*. 2015;126(16):1921-1924.

35. Fischer K, Bahlo J, Fink AM, et al. Long-term remissions after FCR chemoimmunotherapy in previously untreated patients with CLL: updated results of the CLL8 trial. *Blood*. 2016;127(2):208-215.

36. Thompson PA, Tam CS, O'Brien SM, et al. Fludarabine, cyclophosphamide, and rituximab treatment achieves long-term disease-free survival in IGHV-mutated chronic lymphocytic leukemia. *Blood*. 2016;127(3):303-309.

37. Stamatopoulos K, Agathangelidis A, Rosenquist R, Ghia P. Antigen receptor stereotypy in chronic lymphocytic leukemia. *Leukemia*. 2017;31(2):282-291.

38. Thorsélius M, Kröber A, Murray F, et al. Strikingly homologous immunoglobulin gene rearrangements and poor outcome in VH3-21-using chronic lymphocytic leukemia patients independent of geographic origin and mutational status. *Blood*. 2006;107(7):2889-2894.

39. Baliakas P, Agathangelidis A, Hadzidimitriou A, et al. Not all IGHV3-21 chronic lymphocytic leukemias are equal: prognostic considerations. *Blood*. 2015;125(5):856-859.

40. Orchard JA, Ibbotson RE, Davis Z, et al. ZAP-70 expression and prognosis in chronic lymphocytic leukaemia. *Lancet*. 2004;363(9403):105-111.

41. Crespo M, Bosch F, Villamor N, et al. ZAP-70 expression as a surrogate for immunoglobulin-variable-region mutations in chronic lymphocytic leukemia. *N Engl J Med*. 2003;348(18):1764-1775.

42. Rassenti LZ, Huynh L, Toy TL, et al. ZAP-70 compared with immunoglobulin heavy-chain gene mutation status as a predictor of disease progression in chronic lymphocytic leukemia. *N Engl J Med*. 2004;351(9):893-901.

43. Ibrahim S, Keating M, Do KA, et al. CD38 expression as an important prognostic factor in B-cell chronic lymphocytic leukemia. *Blood*. 2001;98(1):181-186.

44. Lin K, Sherrington PD, Dennis M, Matrai Z, Cawley JC, Pettitt AR. Relationship between p53 dysfunction, CD38 expression, and IgV (H) mutation in chronic lymphocytic leukemia. *Blood*. 2002;100(4):1404-1409.

45. Ghia P, Guida G, Stella S, et al. The pattern of CD38 expression defines a distinct subset of chronic lymphocytic leukemia (CLL) patients at risk of disease progression. *Blood*. 2003;101(4):1262-1269.

46. Boonstra JG, van Lom K, Langerak AW, et al. CD38 as a prognostic factor in B cell chronic lymphocytic leukaemia (B-CLL): comparison of three approaches to analyze its expression. *Cytometry B Clin Cytom*. 2006;70(3):136-141.

47. 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.

48. Kröber A, Bloehdorn J, Hafner S, et al. Additional genetic high-risk features such as 11q deletion, 17p deletion, and V3-21 usage characterize discordance of ZAP-70 and VH mutation status in chronic lymphocytic leukemia. *J Clin Oncol*. 2006;24(6):969-975.

49. Bulian P, Shanafelt TD, Fegan C, et al. CD49d is the strongest flow cytometry-based predictor of overall survival in chronic lymphocytic leukemia. *J Clin Oncol*. 2014;32(9):897-904.

50. Dal Bo M, Tissino E, Benedetti D, et al. Functional and clinical significance of the integrin alpha chain CD49d expression in chronic lymphocytic leukemia. *Curr Cancer Drug Targets*. 2016;16(8):659-668.

51. Hallek M, Langenmayer I, Nerl C, et al. Elevated serum thymidine kinase levels identify a subgroup at high risk of disease progression in early, nonsmoldering chronic lymphocytic leukemia. *Blood*. 1999;93(5):1732-1737.

52. Keating MJ, Lerner S, Kantarjian H, Freireich EJ, O'Brien S. The serum β2-microglobulin (β2m) level is more powerful than stage in predicting response and survival in chronic lymphocytic leukemia (CLL) [abstract]. *Blood*. 1995;86(10 suppl I):606a. Abstract 2412.

53. Reinisch W, Willheim M, Hilgarth M, et al. Soluble CD23 reliably reflects disease activity in B-cell chronic lymphocytic leukemia. *J Clin Oncol*. 1994;12(10):2146-2152.

54. Sarfati M, Chevret S, Chastang C, et al. Prognostic importance of serum soluble CD23 level in chronic lymphocytic leukemia. *Blood*. 1996;88(11):4259-4264.

55. Wierda WG, O'Brien S, Wang X, et al. Prognostic nomogram and index for overall survival in previously untreated patients with chronic lymphocytic leukemia. *Blood*. 2007;109(11):4679-4685.

56. Magnac C, Porcher R, Davi F, et al. Predictive value of serum thymidine kinase level for Ig-V mutational status in B-CLL. *Leukemia*. 2003;17(1):133-137.

57. Matthews C, Catherwood MA, Morris TC, et al. Serum TK levels in CLL identify Binet stage A patients within biologically defined prognostic subgroups most likely to undergo disease progression. *Eur J Haematol*. 2006;77(4):309-317.

58. Pflug N, Bahlo J, Shanafelt TD, et al. Development of a comprehensive prognostic index for patients with chronic lymphocytic leukemia. *Blood*. 2014;124(1):49-62.

59. International CLL; International CLL-IPI working group. An international prognostic index for patients with chronic lymphocytic leukaemia (CLL-IPI): a meta-analysis of individual patient data. *Lancet Oncol*. 2016;17(6):779-790.

60. Rozman C, Montserrat E, Rodríguez-Fernández JM, et al. Bone marrow histologic pattern–the best single prognostic parameter in chronic lymphocytic leukemia: a multivariate survival analysis of 329 cases. *Blood*. 1984;64(3):642-648.

61. Rai KR, Sawitsky A, Cronkite EP, Chanana AD, Levy RN, Pasternack BS. Clinical staging of chronic lymphocytic leukemia. *Blood*. 1975;46(2):219-234.

62. 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.

63. Rai KR. A critical analysis of staging in CLL. In: Gale RP, Rai KR, eds. Chronic Lymphocytic Leukemia: Recent Progress and Future Directions. New York, NY: Alan R. Liss; 1987:253-264.

64. Cramer P, Hallek M. Prognostic factors in chronic lymphocytic leukemia-what do we need to know? *Nat Rev Clin Oncol*. 2011;8(1):38-47.

65. Amaya-Chanaga CI, Rassenti LZ. Biomarkers in chronic lymphocytic leukemia: clinical applications and prognostic markers. *Best Pract Res Clin Haematol*. 2016;29(1):79-89.

66. Parikh SA, Shanafelt TD. Prognostic factors and risk stratification in chronic lymphocytic leukemia. *Semin Oncol*. 2016;43(2):233-240.

67. Gentile M, Shanafelt TD, Rossi D, et al. Validation of the CLL-IPI and comparison with the MDACC prognostic index in newly diagnosed patients. *Blood*. 2016;128(16):2093-2095.

68. da Cunha-Bang C, Christiansen I, Niemann CU. The CLL-IPI applied in a population-based cohort. *Blood*. 2016;128(17):2181-2183.

69. Rigolin GM, Cavallari M, Quaglia FM, et al. In CLL, comorbidities and the complex karyotype are associated with an inferior outcome independently of CLL-IPI. *Blood*. 2017;129(26):3495-3498.

70. Molica S, Giannarelli D, Mirabelli R, Levato L, Kay NE, Shanafelt TD. Chronic lymphocytic leukemia international prognostic index (CLL-IPI): a systematic review and meta-analysis. *Blood*. 2018;131(3):365-368.

71. Molica S, Giannarelli D, Levato L, Mirabelli R, Gentile M, Morabito F. Assessing time to first treatment in early chronic lymphocytic leukemia (CLL): a comparative performance analysis of five prognostic models with inclusion of CLL-international prognostic index (CLL-IPI). *Leuk Lymphoma*. 2017;58(7):1736-1739.

72. Gentile M, Shanafelt TD, Mauro FR, et al. Comparison between the CLL-IPI and the Barcelona-Brno prognostic model: analysis of 1299 newly diagnosed cases. *Am J Hematol*. 2018;93(2):E35-E37.

73. Delgado J, Doubek M, Baumann T, et al. Chronic lymphocytic leukemia: a prognostic model comprising only two biomarkers (IGHV mutational status and FISH cytogenetics) separates patients with different outcome and simplifies the CLL-IPI. *Am J Hematol*. 2017;92(4):375-380.

74. Extermann M, Overcash J, Lyman GH, Parr J, Balducci L. Comorbidity and functional status are independent in older cancer patients. *J Clin Oncol*. 1998;16(4):1582-1587.

75. Balducci L, Extermann M. Management of cancer in the older person: a practical approach. *Oncologist*. 2000;5(3):224-237.

76. Bellera CA, Rainfray M, Mathoulin-Pélissier S, et al. Screening older cancer patients: first evaluation of the G-8 geriatric screening tool. *Ann Oncol*. 2012;23(8):2166-2172.

77. Goede V, Fischer K, Busch R, et al. Obinutuzumab plus chlorambucil in patients with CLL and coexisting conditions. *N Engl J Med*. 2014;370(12):1101-1110.

78. Städler N, Shang A, Bosch F, et al. A systematic review and network meta-analysis to evaluate the comparative efficacy of interventions for unfit patients with chronic lymphocytic leukemia. *Adv Ther*. 2016;33(10):1814-1830.

79. O'Brien SM, Keating MJ, Mocarski ES. Updated guidelines on the management of cytomegalovirus reactivation in patients with chronic lymphocytic leukemia treated with alemtuzumab. *Clin Lymphoma Myeloma*. 2006;7(2):125-130.

80. El Chaer F, Shah DP, Chemaly RF. How I treat resistant cytomegalovirus infection in hematopoietic cell transplantation recipients. *Blood*. 2016;128(23):2624-2636.

81. Baden LR, Swaminathan S, Angarone M, et al. Prevention and treatment of cancer-related infections, version 2.2016, NCCN Clinical Practice Guidelines in Oncology. *J Natl Compr Canc Netw*. 2016;14(7):882-913.

82. Kim HY, Kim W. Chemotherapy-related reactivation of hepatitis B infection: updates in 2013. *World J Gastroenterol*. 2014;20(40):14581-14588.

83. Malcikova J, Tausch E, Rossi D, et al; European Research Initiative on Chronic Lymphocytic Leukemia (ERIC)—TP53 network. ERIC recommendations for TP53 mutation analysis in chronic lymphocytic leukemia—update on methodological approaches and results interpretation [published online ahead of print 2 February 2018]. *Leukemia*. doi:10.1038/s41375-017-0007-7.

84. Muntañola A, Bosch F, Arguis P, et al. Abdominal computed tomography predicts progression in patients with Rai stage 0 chronic lymphocytic leukemia. *J Clin Oncol*. 2007;25(12):1576-1580.

85. Eichhorst BF, Fischer K, Fink AM, et al; German CLL Study Group (GCLLSG). Limited clinical relevance of imaging techniques in the follow-up of patients with advanced chronic lymphocytic leukemia: results of a meta-analysis. *Blood*. 2011;117(6):1817-1821.

86. Conte MJ, Bowen DA, Wiseman GA, et al. Use of positron emission tomography-computed tomography in the management of patients with chronic lymphocytic leukemia/small lymphocytic lymphoma. *Leuk Lymphoma*. 2014;55(9):2079-2084.

87. Dighiero G, Maloum K, Desablens B, et al; French Cooperative Group on Chronic Lymphocytic Leukemia. Chlorambucil in indolent chronic lymphocytic leukemia. *N Engl J Med*. 1998;338(21):1506-1514.

88. Shustik C, Mick R, Silver R, Sawitsky A, Rai K, Shapiro L. Treatment of early chronic lymphocytic leukemia: intermittent chlorambucil versus observation. *Hematol Oncol*. 1988;6(1):7-12.

89. Spanish Cooperative Group Pethema. Treatment of chronic lymphocytic leukemia: a preliminary report of Spanish (Pethema) trials. *Leuk Lymphoma*. 1991;5(suppl 1):89-91.

90. 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.

91. Fink AM, Böttcher S, Ritgen M, et al. Prediction of poor outcome in CLL patients following first-line treatment with fludarabine, cyclophosphamide and rituximab. *Leukemia*. 2013;27(9):1949-1952.

92. Tam CS, O'Brien S, Wierda W, et al. Long-term results of the fludarabine, cyclophosphamide, and rituximab regimen as initial therapy of chronic lymphocytic leukemia. *Blood*. 2008;112(4):975-980.

93. Dreger P, Schetelig J, Andersen N, et al; European Research Initiative on CLL (ERIC) and the European Society for Blood and Marrow Transplantation (EBMT). Managing high-risk CLL during transition to a new treatment era: stem cell transplantation or novel agents? *Blood*. 2014;124(26):3841-3849.

94. Kharfan-Dabaja MA, Kumar A, Hamadani M, et al. Clinical practice recommendations for use of allogeneic hematopoietic cell transplantation in chronic lymphocytic leukemia on behalf of the Guidelines Committee of the American Society for Blood and Marrow Transplantation. *Biol Blood Marrow Transplant*. 2016;22(12):2117-2125.

95. Montserrat E, Dreger P. Treatment of chronic lymphocytic leukemia with del(17p)/TP53 mutation: allogeneic hematopoietic stem cell transplantation or BCR-signaling inhibitors? *Clin Lymphoma Myeloma Leuk*. 2016;16(suppl):S74-S81.

96. Cheson BD, Fisher RI, Barrington SF, et al; United Kingdom National Cancer Research Institute. Recommendations for initial evaluation, staging, and response assessment of Hodgkin and non-Hodgkin lymphoma: the Lugano classification. *J Clin Oncol*. 2014;32(27):3059-3068.

97. Kovacs G, Robrecht S, Fink AM, et al. Minimal residual disease assessment improves prediction of outcome in patients with chronic lymphocytic leukemia (CLL) who achieve partial response: comprehensive analysis of two phase III studies of the German CLL Study Group. *J Clin Oncol*. 2016;34(31):3758-3765.

98. Oudat R, Keating MJ, Lerner S, O'Brien S, Albitar M. Significance of the levels of bone marrow lymphoid infiltrate in chronic lymphocytic leukemia patients with nodular partial remission. *Leukemia*. 2002;16(4):632-635.

99. Noy A, Verma R, Glenn M, et al. Clonotypic polymerase chain reaction confirms minimal residual disease in CLL nodular PR: results from a sequential treatment CLL protocol. *Blood*. 2001;97(7):1929-1936.

100. Blum KA, Young D, Broering S, et al. Computed tomography scans do not improve the predictive power of 1996 national cancer institute sponsored working group chronic lymphocytic leukemia response criteria. *J Clin Oncol*. 2007;25(35):5624-5629.

101. Cheson BD, Byrd JC, Rai KR, et al. Novel targeted agents and the need to refine clinical end points in chronic lymphocytic leukemia. *J Clin Oncol*. 2012;30(23):2820-2822.

102. Böttcher S, Hallek M, Ritgen M, Kneba M. The role of minimal residual disease measurements in the therapy for CLL: is it ready for prime time? *Hematol Oncol Clin North Am*. 2013;27(2):267-288.

103. Böttcher S, Ritgen M, Fischer K, et al. Minimal residual disease quantification is an independent predictor of progression-free and overall survival in chronic lymphocytic leukemia: a multivariate analysis from the randomized GCLLSG CLL8 trial. *J Clin Oncol*. 2012;30(9):980-988.

104. Dreger P, Döhner H, Ritgen M, et al; German CLL Study Group. Allogeneic stem cell transplantation provides durable disease control in poor-risk chronic lymphocytic leukemia: long-term clinical and MRD results of the German CLL Study Group CLL3X trial. *Blood*. 2010;116(14):2438-2447.

105. 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.

106. Wendtner CM, Ritgen M, Schweighofer CD, et al; German CLL Study Group (GCLLSG). Consolidation with alemtuzumab in patients with chronic lymphocytic leukemia (CLL) in first remission–experience on safety and efficacy within a randomized multicenter

phase III trial of the German CLL Study Group (GCLLSG). *Leukemia*. 2004;18(6):1093-1101.

107. Rawstron AC, Villamor N, Ritgen M, et al. International standardized approach for flow cytometric residual disease monitoring in chronic lymphocytic leukaemia. *Leukemia*. 2007;21(5):956-964.

108. Rawstron AC, Fazi C, Agathangelidis A, et al. A complementary role of multiparameter flow cytometry and high-throughput sequencing for minimal residual disease detection in chronic lymphocytic leukemia: an European Research Initiative on CLL study. *Leukemia*. 2016;30(4):929-936.

109. Riedell P, Carson KR. A drug safety evaluation of rituximab and risk of hepatitis B. *Expert Opin Drug Saf*. 2014;13(7):977-987.

110. Eichhorst BF, Busch R, Schweighofer C, Wendtner CM, Emmerich B, Hallek M; German CLL Study Group (GCLLSG). Due to low infection rates no routine anti-infective prophylaxis is required in younger patients with chronic lymphocytic leukaemia during fludarabine-based first line therapy. *Br J Haematol*. 2007;136(1):63-72.

111. Herold T, Seiler T, Egensperger R, et al. Progressive multifocal leukoencephalopathy after treatment with rituximab, fludarabine and cyclophosphamide in a patient with chronic lymphocytic leukemia. *Leuk Lymphoma*. 2012;53(1):169-172.

112. Chakraborty S, Tarantolo SR, Treves J, Sambol D, Hauke RJ, Batra SK. Progressive multifocal leukoencephalopathy in a HIV-negative patient with small lymphocytic leukemia following treatment with rituximab. *Case Rep Oncol*. 2011;4(1):136-142.

113. Garrote H, de la Fuente A, Oña R, et al. Long-term survival in a patient with progressive multifocal leukoencephalopathy after therapy with rituximab, fludarabine and cyclophosphamide for chronic lymphocytic leukemia. *Exp Hematol Oncol*. 2015;4(1):8.

114. D'Souza A, Wilson J, Mukherjee S, Jaiyesimi I. Progressive multifocal leukoencephalopathy in chronic lymphocytic leukemia: a report of three cases and review of the literature. *Clin Lymphoma Myeloma Leuk*. 2010;10(1):E1-E9.

115. Cheson BD, Frame JN, Vena D, Quashu N, Sorensen JM. Tumor lysis syndrome: an uncommon complication of fludarabine therapy of chronic lymphocytic leukemia. *J Clin Oncol*. 1998;16(7):2313-2320.

116. Moutouh-de Parseval LA, Weiss L, DeLap RJ, Knight RD, Zeldis JB. Tumor lysis syndrome/tumor flare reaction in lenalidomide-treated chronic lymphocytic leukemia. *J Clin Oncol*. 2007;25(31):5047.

117. Roberts AW, Davids MS, Pagel JM, et al. Targeting BCL2 with venetoclax in relapsed chronic lymphocytic leukemia. *N Engl J Med*. 2016;374(4):311-322.

118. Howard SC, Trifilio S, Gregory TK, Baxter N, McBride A. Tumor lysis syndrome in the era of novel and targeted agents in patients with hematologic malignancies: a systematic review. *Ann Hematol*. 2016;95(4):563-573.

119. Howard SC, Jones DP, Pui CH. The tumor lysis syndrome. *N Engl J Med*. 2011;364(19):1844-1854.

120. Eichhorst BF, Busch R, Obwandner T, Kuhn-Hallek I, Herschbach P, Hallek M; German CLL Study Group. Health-related quality of life in younger patients with chronic lymphocytic leukemia treated with fludarabine plus cyclophosphamide or fludarabine alone for first-line therapy: a study by the German CLL Study Group. *J Clin Oncol*. 2007;25(13):1722-1731.

121. Molica S. Quality of life in chronic lymphocytic leukemia: a neglected issue. *Leuk Lymphoma*. 2005;46(12):1709-1714.

122. Holzner B, Kemmler G, Kopp M, Nguyen-Van-Tam D, Sperner-Unterweger B, Greil R. Quality of life of patients with chronic lymphocytic leukemia: results of a longitudinal investigation over 1 yr. *Eur J Haematol*. 2004;72(6):381-389.

123. Smith TJ, Bohlke K, Lyman GH, et al; American Society of Clinical Oncology. Recommendations for the use of WBC growth factors: American Society of Clinical Oncology Clinical Practice Guideline update. *J Clin Oncol*. 2015;33(28):3199-3212.

124. Rizzo JD, Brouwers M, Hurley P, et al; American Society of Hematology and the American Society of Clinical Oncology Practice Guideline Update Committee. American Society of Hematology/American Society of Clinical Oncology clinical practice guideline update on the use of epoetin and darbepoetin in adult patients with cancer. *Blood*. 2010;116(20):4045-4059.

125. Sánchez-Ramón S, Dhalla F, Chapel H. Challenges in the role of gammaglobulin replacement therapy and vaccination strategies for hematological malignancy. *Front Immunol*. 2016;7:317.

126. Young JA. Epidemiology and management of infectious complications of contemporary management of chronic leukemias. *Infect Disord Drug Targets*. 2011;11(1):3-10.

127. Tsigrelis C, Ljungman P. Vaccinations in patients with hematological malignancies. *Blood Rev*. 2016;30(2):139-147.

128. Pasiarski M, Rolinski J, Grywalska E, et al. Antibody and plasmablast response to 13-valent pneumococcal conjugate vaccine in chronic lymphocytic leukemia patients–preliminary report. *PLoS One*. 2014;9(12):e114966.

129. van der Velden AM, Mulder AH, Hartkamp A, Diepersloot RJ, van Velzen-Blad H, Biesma DH. Influenza virus vaccination and booster in B-cell chronic lymphocytic leukaemia patients. *Eur J Intern Med*. 2001;12(5):420-424.

130. Hodgson K, Ferrer G, Montserrat E, Moreno C. Chronic lymphocytic leukemia and autoimmunity: a systematic review. *Haematologica*. 2011;96(5):752-761.

131. Rodon P, Breton P, Courouble G. Treatment of pure red cell aplasia and autoimmune haemolytic anaemia in chronic lymphocytic leukaemia with Campath-1H. *Eur J Haematol*. 2003;70(5):319-321.

132. Hamblin TJ. Autoimmune complications of chronic lymphocytic leukemia. *Semin Oncol*. 2006;33(2):230-239.

133. Zaja F, Vianelli N, Sperotto A, et al. Anti-CD20 therapy for chronic lymphocytic leukemia-associated autoimmune diseases. *Leuk Lymphoma*. 2003;44(11):1951-1955.

134. Koehrer S, Keating MJ, Wierda WG. Eltrombopag, a second-generation thrombopoietin receptor agonist, for chronic lymphocytic leukemia-associated ITP. *Leukemia*. 2010;24(5):1096-1098.

135. Gupta N, Kavuru S, Patel D, et al. Rituximab-based chemotherapy for steroid-refractory autoimmune hemolytic anemia of chronic lymphocytic leukemia. *Leukemia*. 2002;16(10):2092-2095.