CLL (chronic lymphocytic leukemia)

Status: July 2020

Continuous research and targeted examinations of blood and bone marrow result in various diagnostic recommendations for patients with chronic lymphocytic leukemia (CLL).

Diagnostic recommendation

Method	Anticoagulant	Recommendation
Cytomorphology	EDTA	mandatory
Immunophenotyping	EDTA or Heparin	mandatory
Chromosome analysis	Heparin	optional
FISH	EDTA or Heparin	mandatory
Molecular genetics	EDTA or Heparin	mandatory

Definition and characteristics

Chronic lymphocytic leukemia (CLL) is the most common leukemic disease in elderly patients. In Germany, the annual incidence in men is 7.4/100.000 and in women 4.8/100.000, and the median age of onset is between 70 and 75 years. The clinical and biological picture of CLL is very heterogeneous. The disease develops from mature B-cells, and is mainly due to the inhibition of apoptosis and dysregulation of proliferation in these cells. In many cases, CLL is preceded by an asymptomatic and unnoticed preliminary stage with proliferation of clonal B cells, which is known as **monoclonal B-cell lymphocytosis (MBL)**.

Classification of CLL

According to the WHO classification 2017, chronic lymphocytic leukemia (CLL), as an indolent lymphocytic lymphoma characterized by a leukemic course, is one of the mature B-cell neoplasms. Differentiation from mantle cell lymphoma, follicular lymphoma or other lymphoma entities must be made by immunophenotyping, cytomorphology, FISH and histology.

Diagnostics of CLL

Cytomorphology

Cytomorphologically, B-CLL frequently shows the phenomenon of "Gumprecht's nuclear shadow" in addition to the proliferation of mature lymphatic cells: These correspond to CLL cells which artificially burst on the slide when being spread. They occur, but do not prove the diagnosis. The typical B-CLL is distinguished from B-prolymphocytic leukemia (B-PLL), in which ≥ 55% of the lymphocytes show an immature appearance with a larger nucleus and a clearly visible nucleolus. In the rare transformation of a B-CLL into a Richter syndrome, blast like cells are found.

Immunophenotyping

CLL has a typical phenotype

Characteristic for CLL is an expression of the B-cell markers CD19, CD20 and cyCD79a with simultaneous aberrant expression of the CD5 antigen. There is a weak surface expression of CD22. Specific for CLL is the weak expression of CD20 and the weak to missing expression of FMC7 and CD79b. Characteristically, weak expression of surface immunoglobulins with clonal light chain restriction is also observed. In general, CD23 is expressed, which is rarely the case in mantle cell lymphoma. In addition, compared to CLL, mantle cell lymphoma shows a stronger expression of surface immunoglobulins as well as a higher expression of CD22 and FMC7 and a higher expression of CD79b. Accordingly, the diagnosis of CLL from a single marker alone is not possible. Rather, the synopsis of all antigens examined and the use of the Matutes score derived from this has proven to be useful (Matutes et al. 1994, Moreau et al. 1997).

Immunophenotypic characteristics of B-CLL
- CD5+
- CD23+
- FMC7-
- sIgM(+)
- sCD22(+) or CD79b(+)

Distinction from MBL, CLL/PL and PLL

Monoclonal B-cell lymphocytosis of the CLL type (MBL) is distinguished from CLL: Here a monoclonal B-cell population < 5 x 10^9/L is found in peripheral blood; the immunophenotype corresponds to that of CLL. The patients are mostly asymptomatic and do not show any laboratory changes typical of neoplasm. Transformations into B-CLL are observed annually in 1-2% of all cases.

In CLL/PL (CLL with 10% to 54% prolymphocytes in peripheral blood) the immunophenotype often shows a stronger expression of CD20 and surface immunoglobulins (s-Ig) than in typical CLL. Furthermore, there is a weaker expression of CD23 and a positivity for CD22 and CD79b.

In prolymphocytic leukemia (B-PLL) (≥ 55% prolymphocytes in peripheral blood), there is no or only weak coexpression of CD5, surface expression of immunoglobulins and of CD20 is stronger; CD22 and FMC7 are also expressed.

Detection of measurable residual disease (MRD)

In addition to establishing the diagnosis, immunophenotyping allows the detection of measurable residual disease (MRD) in CLL. The phenotype of CLL cells - CD19+CD20+CD79-CD5+ - clearly distinguishes them from normal B-lymphocytes. The MRD level in CLL is of prognostic relevance according to the results of several larger studies. A low MRD level during and after therapy is associated with longer progression-free survival (PFS) and prolonged overall survival (OS). Patients with negative MRD levels showed a longer PFS than patients with positive MRD levels, regardless of whether they are in complete or partial remission. Thus, MRD quantification allows an improved PFS prediction in patients who achieve both partial and complete remission (Kovacs et al. 2016, Böttcher et al. 2012).

Chromosome analysis

Chromosome analysis shows more abnormalities than FISH

Chromosomal analysis had previously played only a minor role in CLL, since the necessary cultivation of CLL cells in vitro was hardly successful. However, since the cultivation of CLL cells has been reliably possible, more abnormalities can be detected than with FISH analysis (Dicker et al. 2006).

Chromosome analysis can also be helpful in differentiating B-CLL from other mature B-cell neoplasm. It was shown that patients with inconspicuous findings in the FISH analysis but an aberrant karyotype in the chromosome analysis showed a shorter interval until the need for

therapy and a shorter survival than patients who also showed a normal karyotype in the chromosome analysis (Rigolin et al. 2012).

Important extension to FISH

The results of chromosomal banding analyses showed that the spectrum of cytogenetic alterations in CLL is wider than was apparent using the FISH methodology. Typical for CLL are genomically unbalanced events (Haferlach C et al. 2007). The most frequent alterations are 13q deletion, 11q deletion, trisomy 12, 6q deletion and 17p deletion. Rather rare changes are trisomy 3 or 3q gains, gains of 2p, 8q and 11q as well as translocations involving immunoglobulin loci (2p12 (Ig kappa), 14q32 (IgH), 22q11 (Ig lambda)) (Döhner et al. 1999, Stilgenbauer et al. 2002). The translocations t(11;14)(q13;q32), t(14;18)(q32;q21) and t(14;19)(q32;q13) are also rarely observed.

Importance of the complex aberrant karyotype in CLL

Approximately 20% of CLL patients show a complex aberrant karyotype in chromosome band analysis (≥ 3 aberrations) (Haferlach C et al. 2007; Haferlach C et al. 2010). Herling et al. 2016 showed that a complex aberrant karyotype is an independent prognostic factor that may even exceed the prognostic effect of *TP53* alterations. Recent studies suggest that a complex aberrant karyotype is an independent unfavourable factor only from ≥ 5 aberrations on. However, a complex aberrant karyotype with 3 or 4 aberrations is only associated with an adverse prognosis if there is an additional *TP53* deletion and/or a *TP53* mutation. Complex aberrant karyotypes with a +12 and +19 have a favorable prognosis (Baliakas et al. 2019). Thus, a detailed analysis is necessary for the best possible prognostic classification.

Chromosome band analysis is not yet firmly established in the diagnosis of CLL, but could play an important role in the future due to new prognostic findings in complex aberrant karyotypes. Chromosome band analysis has already been included in the recommendations of the iwCLL (Hallek et al. 2018), the NCCN guidelines (status 2019) and the S3-CLL guideline. Beyond supporting the prognostic classification, chromosome band analysis can help to distinguish CLL from other indolent lymphomas.

FISH

With FISH analysis and a standard panel of FISH probes, genetic changes can be detected in approximately 80% of CLL patients. The most common chromosomal abnormalities are 13q deletion, followed by trisomy 12 and 11q deletion (Döhner et al. 2000, Hallek et al. 2008). The 6q deletion and the 17p deletion are observed less frequently. The FISH analysis is important for the prognostic assessment of both standard and more modern therapies. According to the Onkopedia guideline CLL, a FISH analysis should be performed to detect del(17p13) and further genetic testing for atypical phenotype to differentiate it from other indolent lymphomas. According to S3 guideline CLL, an additional examination for 11q deletion should be performed. Patients with del(11)(q22.3) may benefit from chemoimmunotherapy. The International CLL Workshop (iwCLL) recommends a del(11q) or del(17p) examination, a FISH examination for del(13q) and 12q gains in peripheral blood (Oncopedia Guideline CLL: Stand 04/2019, S3 Guideline 2018, Hallek et al. 2018).

Furthermore, the examination for a t(11;14)/IGH-*CCND1* is useful to distinguish between CLL and mantle cell lymphoma. However, even a few distinct CLL - characterized with all classical criteria - may show a t(11;14).

Molecular genetics

Mutated IGHV status in the majority of CLL patients

In IGHV status, somatic mutations are determined in the variable region (V) of the heavy chain (H) of immunoglobulins (IG). About 60% of all CLL patients have a mutated IGHV status. The load of somatic hypermutations in a specific part of the B cell receptor - the so-called IGHV *(immunoglobulin heavy chain variable*) gene - is determined in comparison to the original DNA sequence. A presence of 2% or less mutations is called "unmutated" and a presence of more than 2% mutations is called "mutated". New classifications for the IGHV mutation status are Unmutated status ≤2% mutations, borderline status: 2.1-3% mutations and mutated status >3% mutations. A prognostic statement about the borderline IGHV mutation status is not yet possible (Davis et al. 2016). Since the result of the IGHV mutation analysis for a specific CLL population does not change over time, repeated analysis is usually not necessary. In about 30% of CLL, similar patterns were found in the B-cell receptor, a so-called stereotype (Messmer et al. 2004, Tobin et al. 2004). Stereotype means similarity of the amino acid sequence in the CDR3 region. Different stereotypic rearrangements have different prognostic meanings (Stamatopoulos et al. 2017). The different subgroups of stereotypic B cell receptors are called "subset". Subsets are more common in CLL with unmutated IGHV status (U-CLL) than in CLL with mutated IGHV status (M-CLL) (40% vs. 10%). Different "subsets" are associated with different prognoses of disease progression.

The IGHV mutation status has purely prognostic significance and can be determined at initial diagnosis according to S3 guidelines CLL, but should be performed at the latest at the time of therapy indication. The International CLL Workshop (iwCLL) also recommends a one-time assessment of the IGHV mutation status prior to therapy (Hallek et al. 2018). According to the Oncopedia Guideline CLL, therapy with ibrutinib should be performed if the IGHV status is not mutated.

Mutations in *TP53*, *SF3B1*, *NOTCH1*, *ATM* and *BIRC3*

***TP53* gene**
For *TP53* alterations a distinction must be made between mutations and deletions (17p-). *TP53* mutations are changes in the base sequence within the gene and can only be detected by sequencing. A *TP53* deletion results in the loss of genetic material of the short arm of chromosome 17 (17p), which spans the entire gene and can be detected by FISH, or by chromosomal analysis if the gene is large enough. At initial diagnosis the incidence of *TP53* mutations is about 8%. *TP53* deletions occur in about 4%, mostly in combination with a *TP53* mutation but also alone (Zenz et al. 2010). In order to clarify the *TP53* status of a patient comprehensively, a FISH analysis for 17p as well as a *TP53* mutation analysis must therefore be performed. According to iwCLL guidelines (Hallek et al. 2018) *TP53* aberrations should be investigated prior to therapy and the test should be repeated at each change of therapy. According to the S3 guidelines on CLL, the last determination of *TP53* aberrations should be made no more than 12 weeks before therapy starts and otherwise it should be repeated, since *TP53* aberrations can also occur only in the course of CLL (S3 guideline CLL 2018). In advanced stages and in refractory CLL the frequency of *TP53* changes increases significantly (see Table 1).

***SF3B1* gene**

SF3B1 mutations occur in about 6% of CLL patients. The incidence is significantly increased in refractory CLL patients (Table 1). *SF3B1* mutations correlate with an unmutated IGHV status, *ATM* changes and a stereotypical IGHV3-21 rearrangement (subset #2). There is increasing evidence that these mutations are associated with an intermediate prognosis (Oscier et al. 2013, Stilgenbauer et al. 2014, Jeromin et al. 2014).

***NOTCH1* gene**
NOTCH1 mutations are common in CLL with an unmutated IGHV status and are associated with trisomy 12. *NOTCH1* mutations are observed in about 10% of initial CLL diagnoses and are associated with an approximately 20-fold increased risk of transformation to DLBCL. There is evidence that patients with *NOTCH1* mutations have an intermediate prognosis and experience no benefit from the addition of rituximab to fludarabin/cyclophosphamide chemotherapy (Stilgenbauer et al. 2014). However, this has yet to be confirmed in further independent studies.

***ATM* gene**
ATM changes can be caused by both mutations and deletions (11q-). Mutations occur in approximately 6% of CLL patients at diagnosis. Deletions of 11q22 always contain the *ATM* gene. In about 30% of the cases with a 11q deletion there is also an *ATM* mutation. These patients show an intermediate prognosis, although there is evidence that if a deletion and a mutation occur simultaneously, a less favourable prognosis is to be expected than with only one *ATM* mutation (Austen et al. 2007, Skowronska et al. 2012). The limited response to chemotherapy in patients with *ATM* alteration seems to be improved by the addition of rituximab (Stilgenbauer et al. 2014). Since the gene is very large and therefore difficult to sequence, an analysis is only recommended for specific questions.

BIRC3 alterations can be divided into *BIRC3* mutations and *BIRC3* deletions. The incidence of *BIRC3* alterations at initial diagnosis is 4%, but is higher in refractory patients. The gene *BIRC3* is located on the long arm of chromosome 11 near the *ATM* gene. In 80% of CLL patients with *ATM* deletion, *BIRC3* is also deleted. An adverse prognosis is assumed for patients with *BIRC3* alterations (Rossi et al. 2013).

Table 1: Frequency of mutations at different points in the course of the disease
(Rossi et al. 2013, Guièze & Wu 2015)

Gene	*TP53*	*SF3B1*	*NOTCH1*	*ATM*	*BIRC3*
Diagnosis	8%	6%	10%	6%	4%
In need of therapy	10%	18%	10%	14%	-
refraktory/relapse	31%	38%	15%	25%	15%

Prognosis of CLL

IGHV status is an important prognostic marker of CLL
IGHV status is a very important prognostic marker in CLL. The non-mutated status is already associated with a less favourable prognosis in early stage CLL (Damle et al. 1999, Hamblin et al. 1999).

The presence of a stereotypic B-cell receptor may affect prognosis. Recently, it has been shown that patients with a stereotypic IGHV3-21 rearrangement (subset #2) have a significantly reduced time to treatment regardless of the mutation status (Baliakas et al. 2015, Jeromin et al. 2016). However, there is evidence that the prognosis is modulated by additionally present molecular and cytogenetic changes (Jeromin et al. 2016).

In addition, it was shown that subset #1 and #8 are frequently associated with a very aggressive clinical course in U-CLL, whereas subset #4, which is mostly present in M-CLL, has an indolent course (Stamatopoulos et al. 2017).

FISH abnormalities have prognostic significance in CLL
The abnormalities detected by FISH have prognostic significance. Döhner et al. (2000) developed a hierarchical model, whereby when multiple aberrations occur, the prognosis is determined by the most unfavourable genetic modification (see Table 2). The presence of a 17p deletion or an 11q deletion indicates a less favourable disease course compared to the "normal karyotype" in FISH, whereas the presence of a 13q deletion alone is associated with a more favourable prognosis. Patients with trisomy 12 show a similar prognosis as those with normal karyotype.

Table 2: Prognostic relevance of genetic changes

	Median survival time in month according to Döhner et al.	Prognosis
17p-Deletion	32	adverse
11q-Deletion	79	
+ 12	114	
"normal" (no abnormalities detectable by FISH panel)	111	
13q-Deletion (alone)	133	favorable

Chromosome band analysis provides further prognostic information
Chromosome band analysis can provide additional prognostic information in addition to the FISH examination. Several studies have shown that a complex aberrant karyotype is an independent prognostic factor that may even exceed the prognostic effect of *TP53* alterations (Herling et al. 2016). Recent results show that only a complex karyotype with ≥ 5 aberrations is an independent unfavourable factor. However, a complex aberrant karyotype with 3 or 4 aberrations is only associated with an adverse prognosis if there is an additional *TP53* deletion and/or a *TP53* mutation. Complex aberrant karyotypes with +12 and +19 show a favourable prognosis (Baliakas et al. 2019).

CLL: *TP53* mutations have an adverse prognosis
An adverse effect on overall survival and time to treatment was shown for *TP53* alterations. The incidence of *TP53* mutations is 8-12% and of *TP53* deletions 4-7%, but it is significantly higher in advanced stages and in refractory CLL. *TP53* mutations are often associated with 17p/*TP53* deletions, but correlate independently with an adverse prognosis (Döhner et al. 2000, Stilgenbauer et al. 2014, Zenz et al. 2010). If one *TP53* allele is mutated and the other is deleted, so that no functional TP53 can be formed, this leads to an additive negative effect (Stengel et al. 2016). Biallelic mutations or monoallelic mutations with CN-LOH (copy-neutral loss of heterozygosity) with a dominant negative effect rarely occur (Goh et al. 2011).

The prognostic panel according to Rossi
The prognostic panel according to Rossi et al. 2013 considers both molecular genetic and cytogenetic markers. Patients are divided into four risk groups:
- **High risk**: *TP53* or *BIRC3* alterations
- **Intermediary risk**: *NOTCH1* or *SF3B1* mutations or 11q deletion
- **Low risk**: trisomy 12 or normal karyotype
- **Very low risk**: only 13q deletion

This model can be used both for initial diagnosis and during the course of the disease.

Subclones with mutations (especially in the gene *TP53*) also show the same unfavourable course as patients with corresponding changes in the main clone (Landau et al. 2013, Rossi et al. 2014). Next-generation sequencing enables the detection of these subclonal mutations up to a clinically relevant mutation load of approx. 3%.

In order to be able to better predict the individual course of events, a systematic prognosis index has been developed (International Prognostic Index CLL-IPI, Table 3 and Table 4).

Table 3: International Prognostic Index for CLL (Variable)

Variable	Risk factors	Points
TP53 status	Deletion and/or mutation	4
IGHV status	unmutated	2
ß2-mikroglobulin	>3.5 mg/L	2
Stage	Rai I-IV or Binet B-C	1
Age	>65 years	1

Table 4: International Prognostic Index for CLL (Risk groups)

Risk group	Score	5-year survival rate (%)
low	0-1	93.2
intermediate	2-3	79.3
high	4-6	63.3
very high	7-10	23.3

Calculation of prognosis
Here you can access the calculation of the CLL-IPI score.

Hoechstetter et al. 2020 showed for stage A CLL according to Binet that in addition to age > 60, ß2-microglobulin >3.5 mg/L, an unmutated IGHV status, a del(17p) in FISH, also lymphocyte doubling time <12 months, del(11q), trisomy 12, male sex and *NOTCH1* mutations are associated with a shorter survival and a shorter time to first treatment (Hoechstetter et al. 2020). It should be noted that the *TP53* mutation status was not evaluated in this study. The final prognostic model includes six independent risk factors weighted relative to the respective hazard ratio. The score can be calculated according to Table 5. Based on the sum of risk points, patients could be stratified into four risk groups, which were of prognostic relevance for the assessment of both overall survival and time to first treatment (TTFT) (Hoechstetter et al. 2020).

Table 5: Prognostic model developed on the basis of patient data from the CLL1 cohort (CLL1-PM)

Independent factor	Status	Points
del(17p)	detectable	3.5
IGHV mutation status	unmutated	2.5
ß2 microglobulin	>3.5 mg/L	2.5
del(11q)	detectable	2.5
Lymphocyte doubling time	<12 months	1.5
Age at baseline	>60 years	1.5
Risk classification by total score Very low risk: 0-1.5 Low risk: 2-4 High risk: 4.5-6.5 Very high risk: 7-14		

A new international prognostic score (IPS-E) was developed for patients with asymptomatic early stage CLL. It provides a prediction of the time to first treatment (TTFT). Three factors with equal weighting (with 1 point each) were determined, which can independently predict TTFT. The factors are: an unmutated IGHV status, lymphocytes > $15x10^9$/L and palpable lymph nodes. This results in 3 risk groups: Low risk (0 points), medium risk (1 point) and high risk (2-3 points). The probability of needing treatment increases from low risk to high risk patients (Table 6 and Table 7). *TP53* aberrations play an important role in therapy decisions, but have no prognostic significance in determining the time to treatment (Condoluci et al. 2020).

Table 6: International Prognostic Score (IPS-E)

Variable	Risk factors	Points
IGHV status	unmutated	1
Lymphocytes	> $15x10^9$/L	1
Lymphe nodes	palpable	1

Table 7: Risk groups

Risk groups	Score	Cumulative incidence of treatment (5-years,%)
low	0	8.4
intermediate	1	28.4
high	2-3	61.2

Associations between genetic changes and prognosis markers
There are associations between molecular cytogenetic changes (in FISH) and other prognostic markers. For example, CD38-positivity associated with an adverse prognosis (Hamblin et al. 2000, Ibrahim et al. 2001, Jelinek et al. 2001) occurs more frequently together with a higher number of FISH abnormalities and with high risk factors (17p-, 11q-). Furthermore, associations between 11q-deletions and 17p-deletions and an unmuted IGHV status are found (Stilgenbauer et al. 2002; Kröber et al. 2002). Associations of *SF3B1* mutations with 11q deletions and *NOTCH1* mutations with trisomy 12 have recently been discussed.

In addition, the detection of CD38 expression and cytoplasmic expression of ZAP70 is associated with an adverse prognosis. However, these parameters are of less clinical and therapeutic relevance today.

Baliakas et al. 2019 examined early stage CLL for different prognostic markers depending on the IGHV status. Unfavorable factors in patients with mutated IGHV status are *TP53* aberrations, trisomy 12 and the stereotype IGHV3-21 (subset #2). In patients with unmutated IGHV status del(11q), *TP53* aberrations and/or *SF3B1* mutations and men showed a shorter time to first treatment.

Diagnostic Recommendations of CLL

If a patient is suspected of having CLL, the various guidelines recommend the following tests:

Onkopedia Guideline CLL (Status: April 2019)

Table 8: Tests to establish the diagnosis CLL

Test	Comment
Complete blood count (CBC)	Leukocytes with differential blood count (microscopic differentiation), thrombocytes, haemoglobin, reticulocytes (for signs of anaemia)
Multiparametric immunophenotyping	Expression of CD19 and CD23 Coexpression of CD5 Weak or missing expression of CD220, CD79b, FMC7 Monoclonality of Igκ and Igλ

Table 9: Additional tests before treatment

Test	Comment
Genetic	• del(17p13)* • *TP53*-mutation analysis • IGHV-mutation status • Further genetic studies in atypical phenotype to differentiate it from other indolent lymphomas

* Data on the adverse prognosis of patients with deletion 17p13 are based on molecular cytogenetic analyses using FISH. The collective of patients with inactivation of p53 by mutations overlaps very much with that of patients with del 17p13, but is not completely congruent

S3 Guideline CLL (March 2018)

Table 10: Overview of tests and indications for initial and follow-up diagnostics

	Initial diagnosis	Course	Prior to Therapy	during/after therapy
Immunophenotyping	shall		can	
FISH del(17p13) and *TP53*-mutation status			shall	
FISH del(11)(q22.3)			should	
FISH del(13)(q14), del(6)(q21~23), +12			can	
IGHV-mutation status			should	
Chromosome band analysis			can	
MRD-evaluation (Cytometry or molecular genetics)			can	can

shall: strong recommendation; should: recommendation; can: recommendation open

iwCLL Guidelines (Hallek et al. 2018)

Table 11: Baseline evaluation of patients with CLL

Tests to establish the diagnosis	Routine	Clinical trial
CBC and differential count	always	always
Immunphenotyping	always	always

Assessment before treatment	Routine	Clinical trial
FISH del(13q), del(11q), del(17p), add(12) in peripheral blood	always	always
TP53 mutation	always	always
IGHV mutational status	always	always
Conventional karyotyping in peripheral blood lymphocytes (with specific stimulation)*	possible	desirable

* Can be useful before therapy if an established methodology is available.

NCCN Guidelines (2019)

Table 12: Overview of NCCN investigation recommendations

Tests	
FISH	+12, del(11q), del(13)(q), del(17p)
Molecular genetics	*TP53* mutation, IGHV mutation status
Further tests	Conventional karyotyping

Therapy of CLL

Cytogenetic and molecular changes influence the decision on therapy in CLL
Cytogenetic changes do not only allow an assessment of the prognosis. Rather, increasing data indicate that it is also possible to assess the response to certain therapies on the basis of the cytogenetic changes. This is of great importance, as many new substances have currently found their way into CLL therapy, whose efficacy also varies depending on the presence of certain genetic abnormalities (Hallek 2013, Byrd et al. 2015, O'Brien et al. 2015, Roberts et al. 2016, Onkopedia Guideline CLL: Status 04/2019).

Del(17p13) and *TP53* mutations influence choice of first-line therapy
Del(17p13) and *TP53* alterations are currently the only prognosis markers that already have a direct influence on the first therapeutic decision. Thus, the presence of *TP53* mutations and/or *TP53* deletions is prognostically unfavourable and associated with a lower response rate to commonly used standard chemotherapy (Oscier et al. 2013, Stilgenbauer et al. 2014). Since *TP53* alterations may be added in the course of the disease, a *TP53* analysis should be performed prior to any therapy decision in case of disease progression. Thus, the current Onkopedia guidelines recommend the use of ibrutinib in first-line therapy in patients with CLL requiring therapy and a *TP53* alteration, regardless of their general condition (Onkopedia guideline CLL: 04/2019).

Recent studies suggest that patients with a complex aberrant karyotype respond less well to ibrutinib-based treatments (O'Brien et al. 2018, Thompson et al. 2015). However, these patients may benefit from treatment with venetoclax-obinutuzumab (Al-Sawaf et al. 2020)

IGHV mutation status
The IGHV status is an important prognosis marker. It does not change during the course of the disease and should therefore be determined once before a decision on therapy is made. Patients with an unmutated IGHV status have a shorter overall survival and need therapy earlier (Hamblin et al. Blood 1999). The Onkopedia guidelines for CLL recommend therapy with ibrutinib.

There are also mutations in other genes whose effects on the course of the disease and relevance to therapy are currently being investigated. These include the following, each with a frequency of less than 10%: *BCOR*, *EGR2*, *FBXW7*, *MYD88*, *NRAS*, *POT1*, *KRAS*, *SAMHD1*, *XPO1*.

Development of resistance under ibrutinib therapy
Treatment with ibrutinib may lead to the development of resistance mutations. Known affected genes are the *BTK* and *PLCG2* genes (Woyach et al. 2014). Since mutations in these genes can only be detected after therapy is administered, an examination is only indicated in the case of non-response and not before the start of therapy.

References

You can find the corresponding references here:
https://www.mll.com/en/diagnostic-offer/chronic-lymphocytic-leukemia-cll/chronic-lymphocytic-leukemia-cll.html#references