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Clinical Significance of Cytogenetic Abnormalities Detected by FISH in CLL: Insights from a Real-World Single-Centre Cohort

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Abstract

Objective: Chronic lymphocytic leukemia (CLL) is a hematologic malignancy that predominantly affects the elderly. This study aimed to investigate cytogenetic alterations that could be used for risk assessment and treatment decisions in patients with CLL in relation to clinical parameters and outcomes.

Materials and Methods: Peripheral blood samples from 101 CLL patients either newly diagnosed or previously diagnosed but untreated were analyzed using interphase fluorescence *in situ* hybridization (I-FISH) method with a CLL panel probe set. The associations among cytogenetic abnormalities, clinical features, treatment indications, and outcomes were statistically evaluated.

Results: FISH analysis revealed statistically significant associations between specific cytogenetic abnormalities and clinical features. del(13q) and ATM deletion were significantly associated with large/symptomatic/progressive lymphadenopathy (LAP); ATM deletion was also linked to progressive splenomegaly. Regarding the causes of death, P53 deletion was significantly associated with secondary solid neoplasms and acute decompensated heart failure; trisomy 12 with disease progression and secondary solid neoplasms; and immunoglobulin heavy chain gene (IGH) rearrangement with tumor lysis syndrome. No significant difference in the overall survival was observed between the treated and untreated CLL patients.

Conclusion: If validated in larger, multicenter cohorts, the novel associations identified between cytogenetic abnormalities and both treatment indications and causes of death could help guide the management of CLL.

Keywords Chronic lymphocytic leukemia · Fluorescence in situ hybridization · Prognosis · Cytogenetic abnormalities · Clinical marker

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INTRODUCTION

Chronic lymphocytic leukemia (CLL), which originates from B cells, is the most common neoplasm in adults. It has significant clinical and genetic heterogeneity that influences disease progression. The median age at diagnosis is 70 years, with a male predominance. Approximately 10% of CLL cases occur in individuals under 45 years of age (1). 70-80% of CLL patients are asymptomatic at the time of diagnosis. The watch and wait approach is the standard of care for this subgroup, with nearly one-third never requiring intervention during their lifetime (2).

Clinically, CLL is staged using the Rai and Binet classification systems, which assess lymphadenopathy, splenomegaly/hepatomegaly, and hematologic parameters such as anemia or thrombocytopenia (3, 4).

Approximately 2-10% of patients develop Richter's syndrome (RS) - a progression to a high-grade aggressive lymphoma. In 95-99% of these cases, the transformation is to diffuse large B-cell lymphoma (DLBCL), in 0.5-5% to Hodgkin's Lymphoma (HL), and less frequently to plasmablastic lymphoma (5).

Genetic abnormalities are critical prognostic markers and play an important role in determining the right treatment decisions in CLL. Interphase fluorescence *in situ* hybridization (I-FISH) is the most effective method for detecting cytogenetic abnormalities, with approximately 80% sensitivity (6). Standard I-FISH panels screen for trisomy 12, del(13)(q14.3), del(11)(q22.3) (*ATM* gene deletion), and del(17)(p13.1) (TP53/P53 gene deletion). Some panels also include del(6)(q23) (*MYB* gene deletion) and immunoglobulin heavy chain (IGH) genes rearrangements. The prognostic impact of each abnormality is different (6, 7). These abnormalities are also associated with RS, which worsens the prognosis of the patient (8).

In this single-centre cohort study, we investigated the prognostic significance of cytogenetic abnormalities detected by I-FISH in patients with CLL. By correlating these genetic alterations with clinical parameters, treatment indications, and survival outcomes, we aimed to identify specific FISH-detectable aberrations that enhance risk stratification and inform therapeutic decisions in real-world settings.

MATERIALS AND METHODS

Study Population

Peripheral blood samples were collected from 101 patients with CLL, either newly diagnosed or previously diagnosed but treatment-naïve. None of the patients were receiving cytotoxic therapy at the time of sampling, and all had no history of other malignancies. Informed consent was obtained from each patient before participation. The study protocol was approved by the Clinical Research Ethics Committee (Decision date/number: November 17, 2023/23), in accordance with the Declaration of Helsinki.

I-FISH Analysis

I-FISH was performed using the direct culture method in accordance with our laboratory's standard procedures. Peripheral blood samples from the patients were drawn into sodium heparinized tubes. For each patient, 0.1 mL of blood was added to a polystyrene tube containing 5 mL of RPMI 1640 medium. Then, the tubes were centrifuged at 3,000 rpm for six minutes, and the supernatant was removed. The pellet was resuspended in 8 mL of a 0.075 M potassium chloride (KCl) solution and incubated at 37 °C for 15 min. Following the second centrifugation and removal of the supernatant, the pellet was resuspended in 8 mL of the fixative (Carnoy's solution: 3:1 methanol/acetic acid) and centrifuged again. The supernatant was removed. This procedure was repeated three times. Finally, the resulting pellet was vortexed, spread onto a microscope slide for each target, and air-dried at room temperature for 10 min.

The CLL panel probe set with the I-FISH method was applied to the patients according to the manufacturer's instructions. For each patient, 200 interphase nuclei were evaluated. Analyses were evaluated with the Isis Fluorescent Imaging System (Metasystems, Germany). The CLL FISH Panel was used for interphase FISH analysis; Alpha satellite 12 plus (Cytocell, UK) for trisomy 12, D13S319 plus deletion (Cytocell, UK) for 13q deletion, and P53 (TP53)/ATM combination (Cytocell, UK) for 17p/11q deletions, IGH plus breakapart (Cytocell, UK) for IGH rearrangements (deletion/translocation) and *MYB* gene deletion (Cytocell, UK) for 6q deletion.

Statistical Analyses

SPSS 20.0 (SPSS Inc., Chicago, IL, USA) was used for the statistical analysis. Shapiro-Wilk and Levene's tests were used to investigate the continuous variables. ANOVA and t-test were used for the parametric variables; Mann–Whitney U and Kruskal-Wallis tests were used for the non-parametrical variables. Categorical data were examined with chi-square tests. Correlations between expression data and clinical parameters were evaluated using Pearson's correlation coefficient.

RESULTS

Of the 101 CLL patients included, 40 (39.6%) were female and 61 (60.4%) were male, yielding a male:female ratio of 1.52. The mean age was 59.7 years (range: 35-86). Clinical follow-up

revealed that 38 patients (37.6%) did not require treatment, 62 (61.4%) received treatment, and 1 patient (1%) was lost to follow-up. According to the Rai staging system, patients were classified as stage 0 (n=39, 38.6%), stage I (n=27, 26.7%), stage II (n=19, 18.8%), stage III (n=9, 8.9%), and stage IV (n=7, 6.9%). Based on the Binet staging system, 69 patients (68.3%) were in stage A, 17 (16.8%) in stage B, and 15 (14.9%) in stage C. The clinical findings of the patients are presented in Table 1.

Comparisons between RAI stages of the patients and treatment indications demonstrated significant associations with progressive lymphocytosis, progressive bone marrow failure, and refractory autoimmune hemolytic anemia (p=0.01, p=0.0003, p=0.00009, respectively). According to the Binet staging system, a significant relationship was found between progressive bone marrow failure and refractory autoimmune hemolytic anemia (p=0.0003 and p=0.01, respectively) (Table 2).

Cytogenetic abnormalities were detected in 64 patients (63.4%), whereas 37 patients (36.6%) had no detectable abnormalities for the regions examined by I-FISH. The frequency of specific abnormalities was as follows: (13q) deletion in 42 patients (41.6%), ATM deletion in 9 patients (8.9%), (P53) deletion in 9 patients (8.9%), trisomy 12 in 13 patients (12.9%), IGH rearrangement in 12 patients (11.9%), and MYB deletion in 1 patient (1%). Images from the CLL FISH panel are shown in Figure 1. A total of 19 patients (18.8%) harbored two or more abnormalities, with the 13q deletion most frequently co-occurring with other abnormalities (Table 3).

When comparing patients' treatment indications with their FISH abnormality results, a significant association was found between 13q deletion and bulky/symptomatic/progressive lymphadenopathy (LAP) (p=0.01), as well as between ATM deletion and both bulky/symptomatic/progressive LAP (p=0.05) and progressive splenomegaly (p=0.03) (Table 4).

During follow-up, 28 patients (27.7%) died (25 of the 63 patients from the treated group and 3 of the 37 patients from the untreated group). The survival statistics (mean and median) are detailed in Table 5. Kaplan–Meier analysis revealed no statistically significant difference in survival between the treated and untreated patients (p=0.154) (Figure 2).

Infection was the leading cause of death with a rate of 8%. Notably, P53 deletion was significantly associated with secondary solid neoplasms (p=0.003) and acute decompensated heart failure (p=0.03); trisomy 12 correlated with disease progression (p=0.0001) and secondary solid neoplasms (p=0.02); and IGH rearrangements were linked to tumor lysis syndrome (p=0.006) (Table 6).

Table 1. Clinical findings of the patients

Characteristic	n = 101
Age at diagnosis, mean ± SD (range)	59.7 ± 10.7 (35-86)
Gender, n(%)	
Female	40 (39.6)
Male	61 (60.4)
Binet stage, n(%)	
A	69 (68.3)
B	17 (16.8)
C	15 (14.9)
Rai stage, n(%)	
0	39 (38.6)
1	27 (26.7)
2	19 (18.8)
3	9 (8.9)
4	7 (6.9)
Number of untreated patients, n(%)	38 (37.6)
Binet stage	
A	30 (78.9)
B	6 (15.4)
C	2 (5.1)
Rai stage	
0	22 (57.9)
1	7 (17.9)
2	6 (15.4)
3	2 (5.1)
4	1 (2.7)
Number of treated patients, n(%)	62 (61.4)
Binet stage	
A	38 (61.3)
B	11 (17.7)
C	13 (21)
Rai stage	
0	16 (25.8)
1	20 (32.3)
2	13 (20.9)
3	7 (11.3)
4	6 (9.7)
Number of patients with an unknown treatment status, n(%)	1 (0.99)
Alive, n (%)	
Yes	73 (72.3)
No	28 (27.7)
Mean time to first treatment (months), mean (range)	26.95 (1-119)
Overall survival (months), mean (range)	86 (29-201)
Patients ≥65 years	55.9
Patients between the ages of 50-65	95
Patients ≤50 years	102.7

Table 2. Comparison of the patient's clinical phase results and reasons for therapy

	Clinical phase			
	Rai stage		Binet stage	
Reason for the therapy	n	p value	n	p value
progressive lymphocytosis	101	**0.01***	101	0.76
progressive bone marrow failure	101	**0.0003***	101	**0.0003***
b symptoms	101	0.796	101	0.44
bulky/symptomatic/progressive LAP	101	0.07	101	0.39
unresponsive to therapy autoimmune hemolytic anemia	101	**0.00009***	101	**0.01***
unresponsive to therapy ITP	101	0.78	101	0.49
progressive splenomegaly	101	0.63	101	0.62
symptomatic/progressive splenomegaly	101	0.44	101	0.16

* Significant difference (p≤0.05); LAP: lymphadenopathy; ITP: immune thrombocytopenic purpura.

Six patients developed RS. Among these, one patient had no detected abnormalities, two had ATM deletion, one had both 13q deletion and IGH rearrangement, another had 13q deletion and trisomy 12, and one had IGH rearrangement. A correlation was found between the time elapsed from diagnosis to death and the RS (p=0.07). The reason for treatment in 3 of 6 patients with RS was bulky/symptomatic/progressive LAP.

DISCUSSION

CLL is the most common type of leukemia. Although most patients are over 65 years of age at the time of diagnosis, approximately 10% are younger than 45 years and a male predominance (male:female ratio 1.9:1) (1).

In our cohort of 101 CLL patients, the mean age was slightly younger 59.7 years (range: 35-86). Nevertheless, the proportion of patients under the age of 45 (8.9%) and the male:female ratio (1.52) remained consistent with the literature.

CLL is a heterogeneous disease diagnosed via blood tests and flow cytometry, with patients monitored until the need for treatment (1). The presence of genetic abnormalities provides valuable prognostic information in CLL requiring therapy (9).

Trisomy of chromosome 12 is one of the most frequent cytogenetic abnormalities (approximately 20%) and remains the only abnormality in 40-60% of cases. Its prognostic significance is debated. It is typically related to shorter survival in the intermediate risk group (10, 11). However, according to the results of a nine-year follow-up of 250 CLL patients with untreated trisomy 12, these patients were reported to have increased CD38 positivity and an atypical immunophenotype, as well as a high incidence of thrombocytopenia, RS, and secondary malignancies (12). It is hypothesized that concurrent NOTCH1 mutations intensify

Table 3. Genetic abnormalities detected in patients by I-FISH testing

FISH abnormalities	Patient n	Percentage (%)
(13q) deletion*	42	41.6
sole abnormality	27	
with one or more abnormalities	15	
ATM deletion	9	8.9
sole abnormality	4	
with one or more abnormalities	5	
(P53) deletion	9	8.9
sole abnormality	7	
with one or more abnormalities	2	
(12) trisomy	13	12.9
sole abnormality	5	
with one or more abnormalities	8	
IGH rearrangement	12	11.9
sole abnormality	3	
with one or more abnormalities	9	
MYB deletion (sole abnormality)	1	1
Complex FISH abnormalities	19	18.8
(13q) monoallelic deletion + ATM deletion	4	3.96
(13q) monoallelic deletion + P53 deletion	2	1.98
(12) trisomy + IGH rearrangement	2	1.98
(13q) deletion + IGH rearrangement	3	2.97
(12) trisomy + (13q) biallelic deletion	2	1.98
(12) trisomy + (13q) monoallelic deletion	2	1.98
(13q) trisomy + IGH rearrangement	1	0.99
(12) trisomy + P53 deletion + IGH rearrangement	1	0.99
(13q) monoallelic deletion + ATM deletion + IGH rearrangement	1	0.99
(12) trisomy + (13q) monoallelic deletion + IGH rearrangement	1	0.99

* 13q deletions are biallelic in only 2 patients and monoallelic in the other 40 patients.

prognosis and that trisomy 12 alone may predispose to disease progression and subsequent alterations such as TP53 and NOTCH1 (11, 13).

In our cohort, trisomy 12 was present in 13 patients (12.9%), and in five cases (38.5%) was the only abnormality. These rates were consistent with the literature findings. The other abnormalities accompanying trisomy 12 were 13q deletion, IGH rearrangement, and P53 deletion. We also identified a significant correlation between trisomy 12 abnormalities and disease progression as well as the development of secondary solid neoplasms.

13q deletion is observed in approximately 50% of CLL patients, either mono- or biallelic. Although it is

Table 4. Comparison of patients' FISH abnormalities results and reasons for therapy

	FISH abnormalities											
	(13q) deletion		ATM deletion		(P53) deletion		(12) trisomy		IGH rearrangement		MYB deletion	
Reason for the therapy	n	p value	n	p value	n	p value	n	p value	n	p value	n	p value
progressive lymphocytosis	15	0.08	4	0.20	3	0.63	3	0.75	2	0.40	1	0.09
progressive bone marrow failure	3	0.30	1	0.98	1	0.98	1	0.69	2	0.49	0	0.72
B symptoms	5	0.78	1	0.98	2	0.25	1	0.69	0	0.19	0	0.72
bulky/symptomatic/progressive LAP	17	**0.01***	5	**0.05***	4	0.24	2	0.28	2	0.36	0	0.53
Unresponsive to therapy autoimmune hemolytic anemia	3	0.39	0	0.47	1	0.37	1	0.62	0	0.40	0	0.81
unresponsive to therapy ITP	2	0.37	1	0.13	0	0.58	0	0.49	0	0.51	0	0.86
progressive splenomegaly	1	0.80	1	**0.03***	0	0.65	0	0.58	0	0.60	0	0.88
symptomatic/progressive splenomegaly	3	0.66	0	0.43	0	0.43	1	0.77	2	0.09	0	0.80

* Significant difference (p≤0.05); LAP: lymphadenopathy; ITP: immune thrombocytopenic purpura.

Table 5. Mean and median survival time of patients

Treatment	Mean[a]				Median			
	Estimate	Std. Error	95% confidence interval		Estimate	Std. Error	95% confidence interval	
			Lower Bound	Upper Bound			Lower Bound	Upper Bound
No	222.333	21.647	179.906	264.761	.	.	.	.
Yes	180.053	14.491	151.651	208.455	147.000	4.429	138.318	155.682
Overall	190.634	14.333	162.541	218.727	150.000	4.667	140.853	159.147

[a] Estimation is limited to the largest survival time if it is censored.

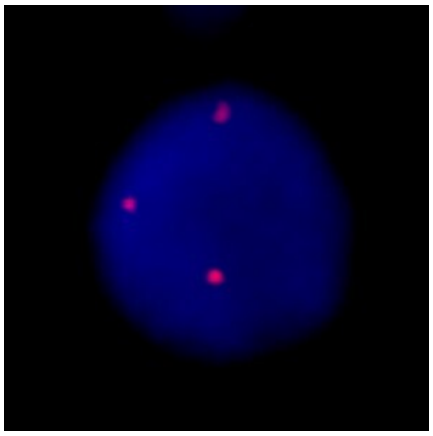
Table 6. Comparison of patients' FISH abnormalities results and the cause of death

	FISH abnormalities											
	(13q) deletion		ATM deletion		(P53) deletion		(12) trisomy		IGH rearrangement		MYB deletion	
Cause of death	n	p value	n	p value	n	p value	n	p value	n	p value	n	p value
Disease progression	1	0.49	1	0.24	0	0.52	3	**0.0001***	1	0.40	0	0.83
Secondary Solid Neoplasm	0	0.08	0	0.52	2	**0.003***	2	**0.02***	1	0.40	0	0.83
Infection	5	0.21	2	0.09	1	0.71	0	0.25	1	0.95	0	0.76
Acute Decompensated Heart Failure	1	0.80	0	0.65	1	**0.03***	0	0.58	0	0.60	0	0.88
Other: Hepatitis	0	0.22	0	0.65	0	0.65	0	0.58	1	0.09	0	0.88
Tumor lysis syndrome	0	0.39	0	0.75	0	0.75	0	0.69	1	**0.006***	0	0.92

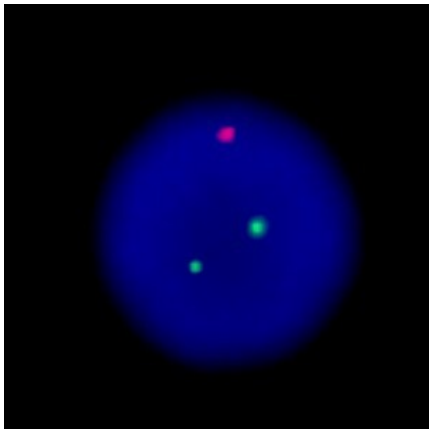
* Significant difference (p≤0.05)

clinically heterogeneous, its presence in the absence of additional cytogenetic abnormalities is considered a favorable prognostic marker (11). It is also a protective factor that reduces the risk of RS development (8). The deletion region at 13q14 spans an area from 300 kbp to 70 Mbp, including the *DLEU1, DLEU2, RB1,* and *TRIM13* genes, as well as microRNAs (miRs 15a and 16-1) (7). A higher number of deleted cells (>70) and the larger deletions shorten the treatment-free intervals. It has been reported that mono/biallelic deletion has no effect on prognosis. Although the 13q deletion is isolated in 36% of patients, it frequently accompanies other abnormalities (14).

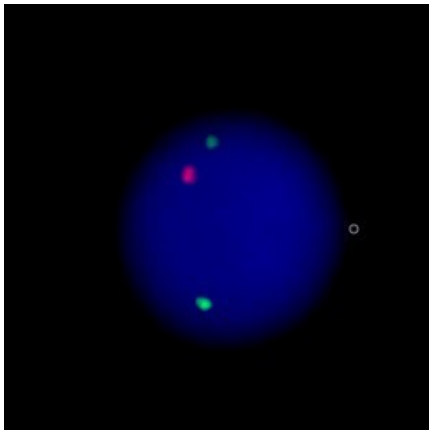
Consistent with previous studies, we detected 13q deletion in 42 patients (41.6%) in our cohort. Among those, 27 patients (64%) had it as the sole abnormality. This rate was higher than that reported in the literature. In patients with two or more abnormalities, the 13q deletion frequently co-occurred. We newly identified a significant association between 13q deletion and bulky/symptomatic/progressive lymphadenopathy in the treated patients. To our knowledge, this relationship has not been reported in the literature.



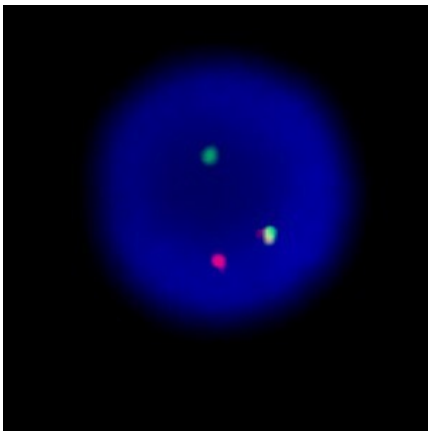
a. Trisomy 12 (red signal D12Z3, 12p11.1-q11.1; three red signals indicate trisomy 12).



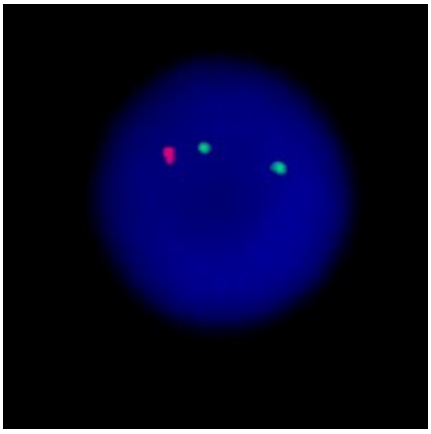
b. Del
(13q) (red signal 13q14.2-q14.3, green signal 13q34; 1R,2G signal indicates a deletion in 13q).



c. del (17p) (red signal P53(17p13), green signal ATM(11q22.3); 1R,2G signal indicates P53 deletion and normal ATM presence).



d. IGH rearrangement (red signal IGHC, 14q32.3, green signal IGHV, 14q32.3 ; 1F,1R,1G signal indicates rearrangement).



e. del (6q) (red signal 6q23.3, green signal D6Z1, 6p11.1-q11.1 ; 1R,2G signal indicates MYB deletion).

Figure 1. FISH images of different patients.

The 11q deletion, observed in 5-20% of CLL cases, is a highly heterogeneous locus ranging from 2 to 20 megabase pairs (Mb) in the 22.3-q23.1 segment interval. It includes the *ATM, ACAT, BIRC3, CUL5, EXPH2, FRDX1, H2AX, KDELC2, MRE11, NPAT, RAB39,* and *RDX* genes. While the 11q deletion results in the loss of the *ATM* tumor suppressor gene in CLL, other genes within this region are also thought to contribute to the disease pathogenesis. It is linked to an early onset, poor prognosis, unmutated IGH variable (*IGHV*) genes, and lymphadenopathy (11, 15). It is also a risk factor for the development of RS (8). Mutations in the *ATM* gene are detected in approximately 30% of CLL patients with del 11q. Furthermore, mutations and deletions in the *BIRC3* gene, which is located near the *ATM* gene at 11q22, have been identified in 4% at diagnosis and 24% in fludarabine-resistant patients. This provides evidence that the *BIRC3* gene is associated with the chemotherapy-resistant CLL phenotype (7, 11).

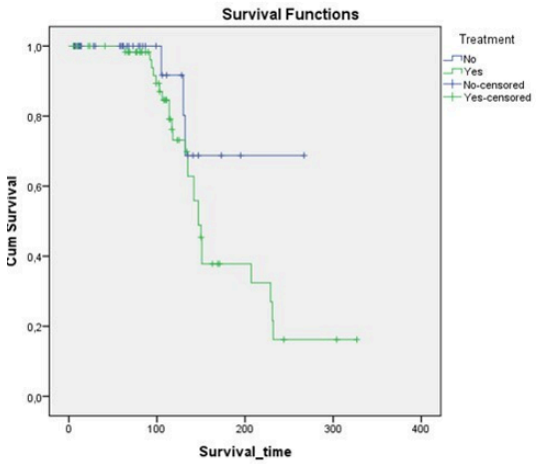


Figure 2. Kaplan–Meier plots of survival estimate for CLL patients who received and did not receive treatment.

When we compared our findings with published data, we found that the 11q deletion rate (8.9%) in our study was consistent with previously reported values. This abnormality was identified as the sole cytogenetic finding in four patients (44.4%). In our cohort, ATM deletion was frequently accompanied by 13q deletion. There was a significant association between ATM deletion and bulky/symptomatic/progressive LAP, consistent with the literature, and with progressive splenomegaly, which has not been previously reported.

The del (17p) abnormality, seen in approximately 5-10% of newly diagnosed CLL patients and in 40-50% of those with relapsed or refractory disease, results in the loss of the tumor suppressor gene *P53* (*TP53*). This deletion, which is associated with a poor prognosis, also confers resistance to treatment. In addition to deletions, somatic gene mutations were detected in the p53 gene. These mutations are observed in approximately 10% of CLL patients and are often associated with p53 deletion (16, 17). Biallelic loss of the *p53* gene, defined as the deletion of one allele and mutation of the other, disrupts the protective barrier against genomic instability, leading to increased DNA damage (18). Notably, CLL patients with monoallelic *TP53* abnormalities exhibit better survival compared with those with biallelic alterations (19). Approximately 40% of patients with p53 abnormalities have biallelic loss. While the mutation frequency is low in untreated CLL patients, it is higher among those who experience disease progression and develop treatment resistance. Although the prognostic impact of *TP53* deletion and mutation is independent, the presence of both abnormalities is associated with a shorter time to first treatment, progression-free survival, and overall survival (20).

In our study, p53 deletion was identified in nine patients (8.9%), which is lower than the frequency reported in the literature. This abnormality was the only cytogenetic finding in seven patients (77.7%). When the causes of death were compared with the FISH abnormalities, a significant association was observed between p53 deletion and both secondary solid neoplasms and acute decompensated heart failure.

The 6q deletion is observed in approximately 3-7% of CLL cases and is typically considered a secondary abnormality within a complex karyotype. It is rarely detected as an isolated aberration (21–23). The 6q deletion region is highly heterogeneous and contains genes with different breakpoints. In patients with CLL, this deletion most frequently occurs between q21 and q23 (24). The minimal deleted region detected by array comparative genomic hybridization (array CGH) included the *SCML4, SEC63, STM1, NR2E1, SNX3, LACE1,* and *FOXO3* genes at 6q21, spanning 107.7–108.7 Mb. It has been reported that the *FOXO3* gene triggers apoptosis by regulating the expression of genes required for cell death. It shows low expression with 6q21 deletion and may represent a potential target for therapy in CLL (23). The 6q23 deletion, which involves the *MYB* gene (the most frequently investigated *MYB* gene), is more prevalent among patients with advanced-stage CLL (25), but remains rare in treatment-naïve patients, who tend to have shorter survival (22).

In our study, MYB deletion was detected in only one patient, representing a lower frequency than that reported in the literature. This patient was clinically diagnosed with RAI stage II and Binet stage A disease and required treatment 22 months after diagnosis.

IGH rearrangements are an important cytogenetic abnormality in CLL. Deletions, translocations, and mutations involving the *IGH* gene can be detected. Translocations of the IGH locus on 14q32 are seen with a frequency of 4-9% in CLL. *BCL2* (18q21) and *BCL3* (19q13) are the most common recurrent partner genes (26). CLL patients with 14q32 rearrangements typically have a shorter treatment-free interval (27). Approximately 60% of CLL patients have a mutation in the *IGHV* gene. In these patients, the disease tends to progress more slowly, usually does not require early treatment, and is associated with a favorable prognosis (28, 29). In contrast, patients without IGHV mutations exhibited high expression of CD38, ZAP-70, and CD49d as well as had a more aggressive disease with shorter overall survival. Unlike other genetic markers in CLL, the IGHV mutation status remains stable over the course of the disease. This observation suggests the existence of two distinct biological subtypes of CLL according to the IGHV mutation status, which plays a role in the pathogenesis of the

disease (28). Del (14q), which is associated with trisomy 12, has also been linked to the unmutated IGHV status, NOTCH1 mutations, and shorter treatment-free survival (29).

In our study, the frequency of IGH rearrangements was higher than that previously reported in the literature, at 11.9%. In nine of the 12 patients (75%) with IGH rearrangements, the abnormality was associated with one or more additional cytogenetic alterations. The other abnormalities accompanying the IGH rearrangements were 13q deletion, trisomy 12, ATM, and P53 deletions. Furthermore, a significant association was found between IGH rearrangement and tumor lysis syndrome when the causes of death were analyzed in relation to the FISH results.

Most patients with CLL are asymptomatic at diagnosis and are monitored without immediate treatment (1).

In our study, 38 of 101 patients (37.6%) did not receive treatment, 62 (61.4%) received treatment, and one patient (1%) was lost to follow-up, with the treatment status unknown. Among those who received treatment, 23 patients (37.1%) initiated therapy within 12 months of diagnosis, 16 (25.8%) between 12 and 24 months, and 23 (37.1%) after more than 24 months. The mean treatment-free survival was 26.95 months (range: 1-119). The overall survival was 55.9 months for patients aged ≥65 years, 95 months for those aged 50-65 years, and 102.7 months for patients aged ≤50 years.

In a study of 1143 CLL patients, the causes of death in 225 patients who died were reported as disease progression (46%), other causes unrelated to CLL (27%), secondary solid neoplasms (19%) and infection (8%) (30).

In our study, 28 of 101 patients (27.7%) died. The cause of death was unknown in 8 cases. In the remaining 20 patients, the most common cause of death was infection (8%). The infection-related mortality rate in our cohort was consistent with that reported in the literature.

In the literature, del (11q) and del (17p) have been reported as risk factors for the development of RS, whereas del (13q) has been described as a protective factor associated with lower risk (8).

In our study, among the 6 patients who developed RS, 1 had no cytogenetic abnormality, 2 had ATM deletions, 1 had both 13q deletion and IGH rearrangement, 1 had 13q deletion with trisomy 12, and 1 had IGH rearrangement. No statistically significant associations were identified.

Given the clinical heterogeneity of CLL, a multifaceted approach is required for disease monitoring and prognosis prediction. Although CLL is generally characterized by advanced age and prolonged survival without treatment, our cohort exhibited a younger average age at diagnosis. The findings of our single-centre study were largely consistent with previously published data. However, the novel associations we observed between cytogenetic abnormalities and both treatment indications and causes of death could help guide the CLL management if validated in larger, multicenter cohorts.

Ethics Committee Approval	Ethics Committee Approval: Ethical approval for this study was obtained from the Clinical Research Ethics Committee of Istanbul Faculty of Medicine, dated 17/11/2023 and numbered 23.
Peer-review	Externally peer-reviewed.
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