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ORIGINAL ARTICLE

Lymphadenopathy as a predictor of progression during venetoclax treatment in chronic lymphocytic leukemia. A campus chronic lymphocytic leukemia study

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Abstract

Clinical or biological parameters useful to predict progression during treatment in real-life setting with ibrutinib, idelalisib and venetoclax in relapsed/refractory chronic lymphocytic leukemia (CLL) are still debated. We conducted a multi-center retrospective study on CLL patients treated with ibrutinib and/or idelalisib who were switched to venetoclax for progression or due to adverse events to identify any clinical and/or biological parameters useful to predict progression during treatment with venetoclax. Of all the 128 evaluable patients, 81 had received ibrutinib prior to switching to venetoclax, 35 had received idelalisib and 12 both. When comparing the three subgroups, we did not notice any statistical difference in terms of clinical or biological features. No variable at baseline and at different time points during the follow-up (at 6, 12, 18 and 24 months) was found to predict progression nor to have significance for Progression Free Survival (PFS) in the ibrutinib group and in the idelalisib group and in subgroups according to the line of treatment. Analyzing the data of the venetoclax treatment, after a median follow up of 14.3 months, median PFS was not reached and estimated 3-year PFS was 54%. Of the 128 patients treated with venetoclax, 28 (22%) experienced progressive disease. At multivariate analysis for predictive factors for progression, lymph node diameter >56.5 mm before starting treatment emerged as an independent risk factor for progression. The lymph node

predictive role for progression during venetoclax treatment could be a new parameter that deserves to be investigate in future studies.

KEYWORDS
CLL, lymph node, venetoclax

1 | INTRODUCTION

In recent years, the strategies for the treatment of patients with chronic lymphocytic leukemia (CLL) have constantly evolved. Nowadays, several therapeutic options are available and the use of chemotherapy is increasingly limited, while new drugs are widely prescribed. These include the B-cell receptor inhibitors (BCRis) ibrutinib and idelalisib, and more recently acalabrutinib, and the BCL-2 inhibitor (BCL2i) venetoclax. The choice of treatment is based on clinical and biological factors, such as age, TP53/del17p disruptions, IGHV status, and performance and fitness status evaluated by Eastern Cooperative Oncology Group and Cumulative Illness Rating Scale.[1–3]

The Bruton tyrosine kinases inhibitor (BTKi) ibrutinib has been widely used as continuous treatment after chemotherapy and then in untreated patients, and has been associated with high rates of durable responses. However, over time an increasing number of patients discontinues treatment because of progressive disease or development of adverse events.[4] In such cases, patients should be switched to another BCRi or to venetoclax.[5–7]

Clinical or biological parameters that are useful to predict progression during treatment in the real-life setting with ibrutinib, idelalisib and venetoclax in relapsed/refractory CLL are still debated. While response to chemotherapy agents is well defined, timing and modality of response assessment during treatment with new agents have changed. If it was established that end-of-combination-treatment or end-of-treatment response assessments are highly prognostic with a role of the minimal residual disease (MRD),[8,9] the timing of the switch from a drug to another is debated. Predicting a suboptimal response to a drug and the timing of the response could help in the proper therapeutic management of these patients.

2 | PATIENTS AND METHODS

We conducted a multi-center retrospective study on all consecutive CLL patients treated, outside clinical trials, with ibrutinib and/or idelalisib who were switched to venetoclax for progression or due to adverse events. The primary objective of the study was to define clinical or biological parameters useful to predict progression during treatment with venetoclax.

Patients enrolled in the study started the first inhibitor between March 2014 and March 2020. The drugs were administered according to the label. Main clinical characteristics such as lymphoadenopaties and splenomegaly, and laboratory parameters such as lymphocyte count, hemoglobin levels, platelet count, Lactate DeHydrogenase (LDH) levels were recorded at the start of the treatment and at pre-specified time points during the therapy (at 6, 12, 18 and 24 months). Also biological characteristics studied before treatment were registered and analyzed.

The study was approved by the Ethics Committee of Fondazione Policlinico Gemelli and conducted in accordance with the Declaration of Helsinki.

2.1 | Statistical analysis

Demographic and baseline data including disease characteristics were summarized descriptively. Categorical data were presented as frequencies and percentages. For continuous data, median and range were presented.

Non-parametric tests were performed for comparisons among groups (Chi-Squared and Fisher Exact test in case of categorical variables or response rate, Mann-Whitney and Kruskal-Wallis test in case of continuous variables). Survival distributions (progression-free survival Progression Free Survival (PFS) and overall survival OS) were estimated using the Kaplan-Meier Product Limit estimator. Subgroup comparisons on survival curves were evaluated by means of the Log-Rank test.

Hazard Ratios (HR) and 95% Confidence Interval were reported as parameter results of the Cox regression models. All covariates were evaluated in univariate models and all factors with univariate association within p-value <0.1 were considered in the multivariate models. Backward and stepwise methods were applied to identify the multivariate models with a step-by-step iterative construction that involves the selection of independent variables to be considered in the final model.

All tests were 2-sided, accepting $p < 0.05$ as indicating a statistically significant difference and confidence intervals were calculated at 95% level. All analysis were performed using the *R* software (R Core Team (2022). R: A language and environment for statistical computing. *R* Foundation for Statistical Computing, Vienna, Austria. URL: https://www.R-project.org/).

3 | RESULTS

The evaluable patients for the analysis were 128, which were divided in three subgroups according to the BCRi administered before switching to venetoclax: ibrutinib (81 patients), idelalisib (35 patients) and both (12 patients). Baseline characteristics such as age, gender,

white blood cells, lymphocytes counts, hemoglobin, platelets, LDH levels, spleen and lymph node diameters, p53 disruption, Fluorescence In Situ Hybridization abnormalities, IGHV mutational status, Rai and Binet stage, are presented in Table 1. Clinical and biological features were homogeneously distributed among the three subgroups.

The majority of patients were treated with ibrutinib and idelalisib in third or following line of treatment (53% and 66%, respectively); only 12% and 3% were treated frontline and 35% and 31% in second line with ibrutinib and idelalisib, respectively.

In the ibrutinib group, 57 patients (70.4%) showed a progression during treatment. After a median follow up of 67.1 months, median PFS was 27.6 months. When analyzing the above variables in univariate no one was identified as predictive of progression at baseline and at the time points during the follow-up. There was no statistically significant difference between patients whose treatment discontinuation was due to progression or adverse event ($p = 0.7$).

Thirty-five patients had received idelalisib prior to switching to venetoclax; 13/35 (37.1%) showed a progression during treatment with a median time of treatment of 18.2 months. Median PFS was 13.4 months. No variable predicting progression was found also in the idelalisib group.

When the 12 patients who received both inhibitors (in 7 patients ibrutinib and then idelalisib; in 5 patients idelalisib and then ibrutinib) prior to venetoclax were considered, 9 patients (75%) progressed with a median PFS of 13.3 months, calculated from the last BTK inhibitor between ibrutinib and idelalisib. No variable was identified as predictive in this subgroup neither.

In addition, we performed Cox regression among the subgroups according to the line of treatment; also when considering the different drugs according to first, second or subsequent lines of treatment, no parameter predictive of progression was found.

The curves of PFS and OS on kinase inhibitors were comparable ($p = 0.18$ and $p = 0.93$, respectively), as shown in Figure 1.

We then conducted the analysis on all 128 patients who were treated with venetoclax. After a median follow up of 14.3 months, the median PFS for venetoclax was not reached; the estimated 3-year PFS was 54%. In this group, we registered 28 patients (22%) who progressed. When considering the cause of discontinuation of the BCRi taken before venetoclax (progression vs. toxicity) it could be possible to distinguish two different groups: patients who progressed during ibrutinib (Figure 2A) and idelalisib (Figure 2B) treatment showed a shorter PFS than patients who stopped the treatment due to toxicity: in fact 2-year PFS after ibrutinib discontinuation was 57% for progression versus 95% for toxicity, whereas 2-year PFS after idelalisib discontinuation was 68% for progression versus 89% for toxicity.

The Kaplan-Meier in Figure 2A reached statistical significance ($p < 0.001$) using the restricted mean survival time at 24 months. The restricted mean is a measure of average survival from time 0 to a specified time point and may be estimated as the area under the survival curve up to that point.

We analyzed all clinical and biological features as above to find a predictive factor of progression. Data on univariate analysis are available in Supplemental Table 1. Multivariate analysis showed an impact of the diameter of the biggest lymph node at the start of venetoclax treatment. By Receiver Operating Characteristic analysis, we found a diameter of 56.5 mm of the largest lymph node at baseline to be a predictor of progression (HR 1.01, range 1.01–1.02, $p = 0.005$) (Supplement Table 1). Moreover, the clinical assessment conducted at each time point confirmed the presence of larger lymph nodes in patients who progressed (Figure 3A). When considering only the 72 patients presenting with lymphoadenopathies at the start of treatment with venetoclax, the diameter of 56.5 mm of the largest lymph node retained statistical significance as predictive factor (Figure 3B). In Figure 3C Kaplan-Meier survival curve showed PFS of patients segregated based on lymph node diameter.

Concerning OS, our analysis showed inferior OS for patients undergone to therapy with BCRi beyond second line of treatment (at least 2 lines of therapy before starting target therapy), with a HR 1.24 (1.02–1.51, $p = 0.038$); 2-year OS 95% versus 91% (<2 line of therapy vs. ≥2 line of therapy).

4 | DISCUSSION

Our study reported real-life experience of treatment with venetoclax in relapsed/refractory CLL patients and identified as predictive factor for progression the presence of at least one lymph node larger than 56.5 mm at baseline.

Predictive value of lymphoadenopathies >5 cm should be taken in consideration when treating patients with venetoclax. Recently this consideration has also been made in the last update published of CLL14 trial, where patients treated with venetoclax in combination with obinutuzumab who were at high risk for Tumor Lysis Syndrome at baseline (defined as lymphoadenopathies >5cm and lymphocyte count >25.000/mmc or lymphoadenopathies >10cm) showed a shorter PFS.[8] These data were reported in frontline therapy, while our cohort was composed of relapsed/refractory CLL patients treated with venetocalx +/− rituximab.

The importance of lymph nodes in patients treated with venetoclax should also be considered in patients with clinical PR and MRD negativity, as recently shown in the interim analysis conducted on the cohort of the Phase 2 HOVON 158/Next STEP trial,[10] where patients in PR MRD negative showed the persistence of CLL cells in PET-negative lymph nodes.

Concerning high risk CLL (TP53/del17p distrupted and/or unmutated immunoglobulin variable heavy chain), our cohort was mostly treated with continuative venetoclax therapy and median observation time was 14.3 months, so probably the impact of these prognostic factors could not have emerged yet, as showed previously.[9]

Nowadays, choosing the correct treatment option and timing of the switch between new targeted agents is crucial, especially considering that these treatments are increasingly anticipated and now rarely used in treatment lines after the third one. This partly justifies our slightly worse real-life data compared to recent clinical trial results[9,11]; in fact, patients previously undergone to multiple lines of therapy before BCRi were included in the analysis, and 85.2%

TABLE 1 Baseline characteristics.

Characteristic	Overall, *N* = 128	Ibrutinib, *N* = 81	Ibrutinib-idelalisib, *N* = 12	Idelalisib, *N* = 35	*p*-value[a]
Gender, *n* (%)					0.80
Female	43 (34%)	26 (32%)	5 (42%)	12 (34%)	
Male	85 (66%)	55 (68%)	7 (58%)	23 (66%)	
Age at diagnosis, median (range)	61 (37–89)	59 (37–89)	63 (37–71)	62 (44–86)	0.25
Number of previous lines of treatment, median (range)	2 (0–7)	2 (0–7)	1.5 (0–6)	2 (0–5)	0.66
White blood cells, median (range)	38,250 (1700–287,500)	36,570 (1700–239,470)	43,660 (8540–136,500)	44,395 (2030–287,500)	0.75
Lymphocytes, median (range)	26,177 (180–275,000)	25,290 (230–194,000)	36,333 (3680–132,000)	27,000 (180–275,000)	0.80
Hb, median (range)	119 (76–170)	120 (77–170)	110 (80–132)	114 (76–167)	0.38
Platelets count, median (range)	121,500 (16,000–368,000)	128,500 (19,000–368,000)	110,500 (18,000–358,000)	103,500 (16,000–261,000)	0.44
LDH, *n* (%)	44 (44%)	26 (40%)	7 (78%)	11 (42%)	0.10
Spleen (mm), median (range)	150 (10–1700)	150 (100–240)	140 (100–240)	150 (10–1700)	>0.99
Lymph node max (mm), median (range)	40 (10–270)	40 (10–161)	26 (10–55)	33 (10–270)	0.30
Lymph node threshold 2.5 cm, *n* (%)	75 (71%)	53 (76%)	5 (56%)	17 (63%)	0.27
Lymph node threshold 5 cm, *n* (%)	26 (25%)	20 (29%)	2 (22%)	4 (15%)	0.36
FISH, *n* (%)					0.68
del11q	20 (20%)	12 (18%)	1 (11%)	7 (25%)	
del13q	23 (23%)	12 (18%)	2 (22%)	9 (32%)	
del17	35 (34%)	24 (37%)	4 (44%)	7 (25%)	
neg	14 (14%)	9 (14%)	2 (22%)	3 (11%)	
tri12	10 (9.8%)	8 (12%)	0 (0%)	2 (7.1%)	
Unknown	26	16	3	7	
p53, *n* (%)					0.26
mut	25 (37%)	15 (33%)	4 (67%)	6 (40%)	
wt	42 (63%)	31 (67%)	2 (33%)	9 (60%)	
Unknown	61	35	6	20	
IGHV, *n* (%)					0.85
mut	17 (20%)	11 (20%)	1 (14%)	5 (24%)	
unmutated	66 (80%)	44 (80%)	6 (86%)	16 (76%)	
Unknown	45	26	5	14	
Rai, *n* (%)					0.38
I	9 (8.0%)	8 (11%)	0 (0%)	1 (3.4%)	
II	43 (38%)	30 (42%)	2 (18%)	11 (38%)	
III	17 (15%)	9 (12%)	3 (27%)	5 (17%)	
IV	43 (38%)	25 (35%)	6 (55%)	12 (41%)	
Unknown	16	9	1	6	
Binet, *n* (%)					0.24
A	5 (4.5%)	5 (6.8%)	0 (0%)	0 (0%)	

TABLE 1 (Continued)

Characteristic	Overall, *N* = 128	Ibrutinib, *N* = 81	Ibrutinib-idelalisib, *N* = 12	Idelalisib, *N* = 35	*p*-value[a]
B	52 (46%)	36 (49%)	3 (27%)	13 (46%)	
C	55 (49%)	32 (44%)	8 (73%)	15 (54%)	
Unknown	16	8	1	7	

Abbreviations: FISH, Fluorescence In Situ Hybridization; LDH, Lactate DeHydrogenase.

[a]Pearson's Chi-squared test; Kruskal-Wallis rank sum test.

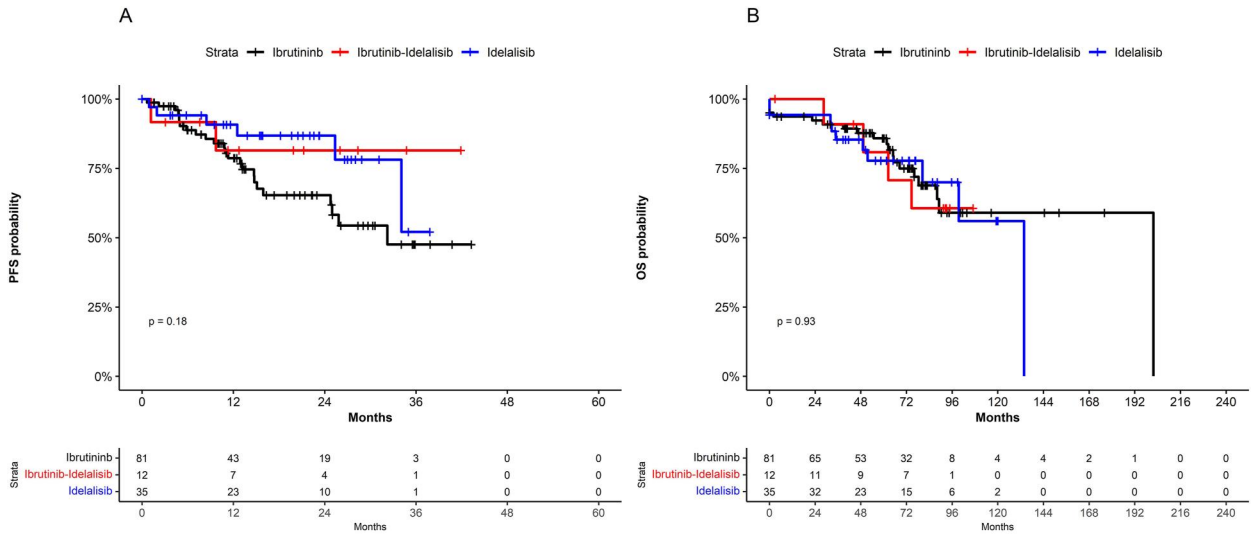

FIGURE 1 A: Progression Free Survival of the three subgroups; B: OS of the three subgroups.

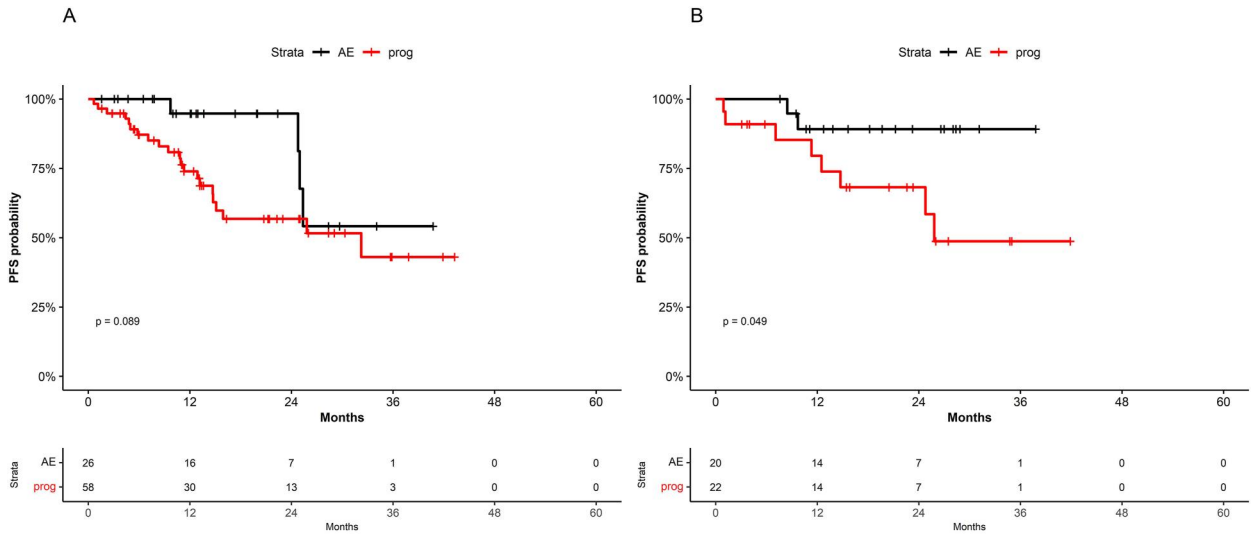

FIGURE 2 Progression Free Survival in venetoclax-treated patients after switching from ibrutinib (A) and from idelalisib (B) for adverse events or for progression.

of the cohort was treated with chemotherapy before the start of targeted therapy. No independent risk factor for progression during BCRi treatment emerged from our analysis. The impact of TP53/del17p disruption on BCRi treatment is not aligned with previous literature,[12] since do not have a predictive value in Cox regression, probably due to the small sample size.

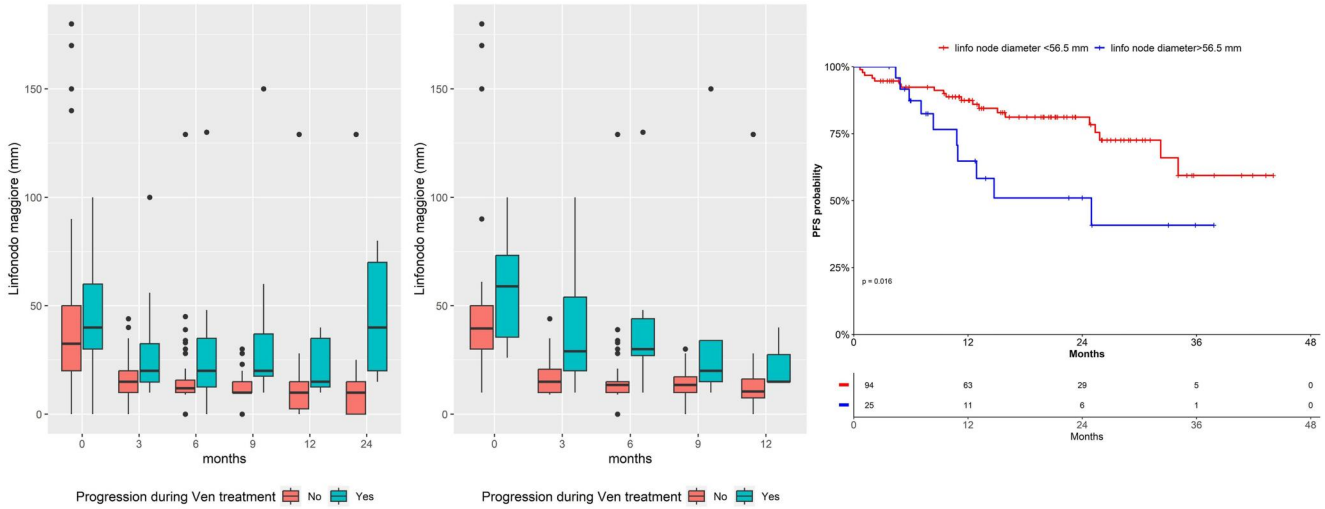

FIGURE 3 Size of the larger lymph node at each time point comparing patients who did not progress (0, red) and patients who progressed (1, blue) in all patients (A) and in patients treated with venetoclax with lymphadenopaties (B). Kaplan-Meier survival curve showing Progression Free Survival of patients segregated based on lymph node diameter (C).

Our data confirm that the cause of discontinuation of BCRi treatment (progression during therapy vs. adverse event) has an impact on the PFS during treatment with venetoclax +/− rituximab.[5]

In conclusion, the presence of a lymphadenopathy greater than 56.5 mm emerged as a predictor of early progression during venetoclax therapy. Therefore, these patients should be monitored closely to identify early signs of progression, even if the treatment change is still guided by iwCLL criteria.[1] In the future, in patients less previously treated or in the frontline setting, this observation needs to be further investigated, especially in novel treatment combinations of BTKi and venetoclax, in order to assess whether the synergic anti-homing effect of the BTKi can overcome the persistence of residual disease in lymph nodes.[10]

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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