

Long-term outcomes of fixed-duration venetoclax–rituximab in relapsed/refractory chronic lymphocytic leukemia: impact of prior BTK inhibitor exposure and measurable residual disease in a multicenter real-world study

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Abstract

Fixed-duration venetoclax–rituximab (VenR) is established for relapsed/refractory (R/R) chronic lymphocytic leukemia (CLL), but real-world evidence after BTKi exposure remains limited.

We retrospectively evaluated 90 R/R CLL patients receiving 24-month fixed-duration VenR across 12 Italian centers from 2020 to January 2025. The primary endpoint was PFS; secondary endpoints were OS, EOT response, MRD, safety, and feasibility.

Fifty-five percent had prior BTKi exposure. At 40-month median follow-up, 3-year PFS and OS were 90% and 94%, respectively. Prior BTKi exposure was associated with inferior 3-year PFS (84% vs 97%; $P = 0.03$). TP53 disruption was associated with inferior PFS (64% vs 97%; $P = 0.005$), whereas IGHV status had no impact ($P = 0.9$). Among 68 EOT-evaluable patients, 57 (83.8%) achieved CR and 10 (14.7%) PR (ORR, 98.5%), without significant PFS differences. Among 65 MRD-evaluable patients, 55 (84.6%) achieved uMRD, associated with superior 40-month PFS (100% vs 90%; $P = 0.0021$). Treatment modifications did not adversely affect PFS. Safety was manageable, with grade ≥ 3 neutropenia in 6.7% of patients. Fixed-duration VenR provides durable disease control in real-world R/R CLL. Prior BTKi exposure, TP53 disruption, and EOT-MRD identify patients with less durable disease control, highlighting the importance of treatment sequencing, disease biology, and depth of response.

Introduction

The treatment landscape of chronic lymphocytic leukaemia (CLL) has undergone a profound transformation with the introduction of targeted therapies, which have progressively replaced chemoimmunotherapy (CIT) in both frontline and relapsed/refractory (R/R) settings [1]. Bruton tyrosine kinase inhibitors (BTKi) and the B-cell lymphoma 2 (BCL2) inhibitor venetoclax have substantially improved survival outcomes while offering distinct treatment strategies based on continuous or fixed-duration administration [2]. In the phase III MURANO trial, fixed-duration venetoclax plus rituximab (VenR), administered for 24 months, significantly improved progression-free survival (PFS) and overall survival (OS) compared with bendamustine–rituximab, establishing VenR as an effective time-limited treatment for R/R CLL [3]. Long-term follow-up confirmed sustained clinical benefit, with a median PFS of approximately 55 months and durable treatment-free remissions in a substantial proportion of patients [4].

However, contemporary use of VenR differs substantially from the MURANO setting, as an increasing proportion of patients have previously received BTKi [3, 4]. Evidence for fixed-duration VenR after BTKi exposure remains limited. Lew et al. reported an overall response rate of 81% and median PFS of 25.9 months in BTKi-exposed patients [5], while more recent real-world data confirmed meaningful activity after covalent BTKi exposure [6]. Larger cohorts with longer follow-up are therefore needed to better define the impact of treatment sequencing. Measurable residual disease (MRD) represents another clinically relevant determinant of outcome with fixed-duration venetoclax-based therapy. In MURANO, achievement of undetectable MRD (uMRD) at the end of treatment was strongly associated with

prolonged disease control, whereas detectable MRD predicted earlier progression [7]. Despite its established prognostic value in clinical trials, evidence regarding end-of-treatment (EOT) MRD assessment in routine clinical practice remains relatively limited, particularly in multicenter populations enriched for BTKi-exposed patients. Real-world administration of VenR may also require individualized treatment adaptations, including temporary interruptions, dose reductions, and supportive care interventions. Available observational studies suggest that venetoclax-based regimens remain effective and manageable outside clinical trials, including in older, comorbid, and previously treated populations [6, 8]. However, the impact of such treatment modifications on long-term efficacy has not been fully characterized. We therefore conducted a multicenter retrospective study of consecutive patients with R/R CLL treated with fixed-duration VenR across the Campania Hematology Network. The primary objective was to evaluate long-term PFS, with particular emphasis on prior BTKi exposure. Secondary objectives included OS, EOT clinical response and MRD status, treatment feasibility, and safety in routine clinical practice.

Material and methods

Patients' selection and study design

This retrospective, observational, multicenter study included consecutive patients treated across 12 Italian Hematology centers. Eligible patients were aged ≥ 18 years, had R/R CLL requiring treatment according to iwCLL criteria [9], and initiated fixed-duration VenR between June 2020 and January 2025 within the Campania regional network. Patients discontinuing VenR before treatment completion remained included in the study cohort.

For treatment-sequencing analyses, patients were classified as BTKi-naïve if previously treated with CIT alone and BTKi-exposed if previously treated with a BTKi, either alone or after CIT.

The study was conducted in accordance with the Declaration of Helsinki and approved by the local Ethics Committee (protocol No. 163/2026).

Data collection

Clinical and treatment data were retrospectively collected from electronic medical records and institutional databases using a standardized data collection form. Baseline variables included demographic, clinical, treatment, and biological characteristics. Molecular characterization included TP53 mutation analysis by next-generation sequencing (NGS), IGHV mutational status, and FISH for recurrent cytogenetic abnormalities. TP53 disruption was defined as TP53 mutation and/or del(17p). Treatment modifications, adverse events, infectious complications, and supportive care were also recorded. Further details are provided in the Supplementary Methods.

Treatment and supportive care

VenR was administered according to the fixed-duration MURANO schedule [3], including a 5-week venetoclax ramp-up (20–400 mg/day), 24 months of venetoclax, and six cycles of rituximab. Treatment modifications were performed according to physician judgment and systematically recorded.

Supportive care included acyclovir and trimethoprim-sulfamethoxazole prophylaxis, secondary G-CSF prophylaxis for neutropenia ($ANC < 1.0 \times 10^9/L$), and risk-adapted HBV monitoring and prophylaxis [10–12]. TLS risk assessment, prophylaxis, and monitoring followed established recommendations [13, 14]. Additional supportive-care details are provided in the Supplementary Methods.

Response assessment

Clinical response was assessed according to the 2018 iwCLL criteria [9] and categorized as complete remission (CR), partial remission (PR), stable disease (SD), or progressive disease (PD). EOT response was assessed at median time of 24 months (range 24–27) from VenR initiation. Patients who had not yet reached this assessment window at data cut-off or discontinued treatment without an evaluable response assessment were considered not evaluable (NE), whereas patients discontinuing treatment because of disease progression were classified as PD. Bone marrow assessment was performed according to clinical practice. MRD status was assessed separately on peripheral blood by multiparametric flow cytometry according to institutional practice and ERIC recommendations [15].

Study endpoints and statistical analysis

The primary endpoint was PFS, defined as time from VenR initiation to disease progression or death from any cause. Secondary endpoints included OS, EOT clinical response and MRD status, safety, and treatment feasibility. PFS was explored according to prior BTKi exposure, age, CIRS, TP53 disruption, IGHV status, bulky disease, EOT response, and MRD status.

PFS and OS were estimated using the Kaplan–Meier method and compared using the log-rank test. Continuous and categorical variables were compared using appropriate parametric/non-parametric and chi-square/Fisher's exact tests, respectively. AEs were graded according to CTCAE v5.0 [16]. All analyses were performed using R software, with two-sided $P < 0.05$ considered statistically significant. Additional statistical details are provided in the Supplementary Methods.

Results

Patient characteristics

From June 2020 to January 2025, 90 consecutive patients with R/R chronic lymphocytic leukemia (CLL) receiving fixed-duration venetoclax plus rituximab (VenR) met the inclusion criteria and were enrolled in

the final analysis. All patients were treated across the Hematology Units of the Campania regional network according to routine clinical practice. Baseline characteristics are summarized in Table 1. All 90 patients were included in the PFS and OS analyses irrespective of treatment completion or availability of EOT response or MRD assessment.

Table 1
Patients' characteristics at VenR start

Table 1. Patients' characteristics at VenR initiation	n	%
Total population	90	100
Age, median (range)	70 (47–88)	
Male sex	64	71
Treatments before VenR, n (range) ^a	1 (1–3)	
BTKi naïve	41	44
BTKi treated	49	56
Prior bendamustine	39	43
Genetic data at enrolment		
IGHV unmutated	46	51
IGHV mutated	40	44
Unknown	4	5
TP53 WT	62	69
TP53 mutated	20	22
Unknown	8	9
BTKi resistance mutation(s) detected ^b	13	14
FISH data at enrolment ^c		
Del17	20	22
Del11	22	24
Del13	37	41
Tris12	17	19
No alteration	5	6
Unknown	4	4
Clinical characteristics at enrolment		
Splenomegaly	40	44
Bulky lymphadenopathy > 10 cm	32	36
Anemia	38	42

Table 1. Patients’ characteristics at VenR initiation	n	%
Thrombocytopenia	25	28
Lymphocyte doubling time< 6months	20	22
BTKi: Bruton tyrosinase kinase inhibitor; Treatment: R-BENDA: Rituximab-bendamustine; RFC: Rituximab, fludarabine, cyclofosfamide; R-chlorambucil: Rituximab-chlorambucil; IGHV, immunoglobulin heavy-chain variable region gene. FISH: Fluorescent in situ hybridization; del: deletion; tris: trisomy. ^b BTKi Mutation: p.(Cys481Ser). ^c Percentages may exceed 100% due to overlapping cytogenetic abnormalities.		

Table 2. VenR safety and feasibility characteristics	n	%
Total population	90	100
Hematological toxicity		
Neutropenia grade ≥ 3	6	7
Thrombocytopenia grade ≥ 3	2	2
Febrile neutropenia	3	3
Extra-hematological toxicity		
TLS	1	1
Rituximab infusion reaction	7	8
Diarrhea	3	4
Infections		
Sars Cov2 symptomatic infection	3	4
Pneumonitis	4	5
HBV-related hepatitis	1	1
Ven permanently discontinuation*	5	6
Months of Ven exposure, median (range)	18 (12–22)	
Ven temporary discontinuation	21	23
Days of discontinuation, median (range)	17 (5–180)	
Rituximab permanently discontinuation	2	2
Dosage reduction		
Ven < 400 mg/die	7	8
Rituximab 375 mg/mq	26	29
N = is number of patient if not otherwise specified. VenR scheduled: ramp-up from 20 to 400 mg daily of Ven plus R 375 mg/mq i.v for cycle 1 and 500 mg/mq i.v for cycles 2–6. TLS: tumor lysis syndrome (1 clinical and laboratoristic TLS was recorded) <i>*Venetoclax exposure data is related to patients with early discontinuation.</i>		

The median age at VenR initiation was 70 years (range, 47–88), and most patients were male (71%).

Median time from diagnosis to initiation of VenR was 4 [1–21] years.

A comprehensive molecular and cytogenetic characterization was available for the vast majority of patients at VenR initiation. TP53 mutation analysis was available in 82/90 patients (91.1%), IGHV mutational status in 86/90 (95.6%), and FISH data in 86/90 (95.6%). Among patients with available IGHV results, 46/86 (53.5%) had unmutated and 40/86 (46.5%) mutated IGHV. TP53 mutations were detected in 20/82 evaluable patients (24.4%). BTK resistance mutations (C481S) were detected in 13 patients (14%). Cytogenetic abnormalities included del(17p) in 20 patients (22%), del(11q) in 22 (24%), del(13q) in 37 (41%), and trisomy 12 in 17 (19%) (Table 1).

Patients had received a median of one prior line of therapy (range, 1–3): 62 (69%) had received one prior line, 27 (30%) two lines, and 1 (1%) three lines. Overall, 41 patients (46%) were BTKi-naïve, whereas 49 (54%) had previously received a BTKi, including 27 (30%) previously exposed to both CIT and BTKi and 22 (24%) treated with BTKi without prior CIT. Prior BTKi exposure consisted predominantly of ibrutinib (47/49, 95.9%), whereas only 2 patients (4.1%) had previously received acalabrutinib; both were still receiving VenR at data cut-off. Among ibrutinib-exposed patients, 8 discontinued treatment because of intolerance after a median exposure of 10 months (range, 6–12), whereas the remaining patients discontinued because of disease progression after a median exposure of 47 months (range, 24–73). Prior bendamustine exposure was documented in 39 patients (43%). Other prior CIT regimens included R- Fludarabine Cyclophosphamide (FCR) in 30 patients (33%) and rituximab-chlorambucil in 7 (8%). VenR was prescribed in accordance with iwCLL treatment initiation criteria [9]. The primary indications for starting treatment included disease progression with massive/symptomatic splenomegaly or lymphadenopathy (31 patients, 34%), and alteration in peripheral blood count, such as anemia, thrombocytopenia and progressive lymphocytosis (59 patients, 66%).

Primary endpoint

PFS was assessed in the entire study population (N = 90), irrespective of treatment completion or EOT evaluability. After a median follow-up of 40 months (range, 7–72), the estimated 3-year PFS for the overall population was 90% (95% CI, 81%–95%; Fig. 1).

To investigate the impact of previous treatment sequencing, patients were stratified according to prior BTKi exposure. Patients previously treated with BTKi showed a significantly shorter PFS compared with BTKi-naïve patients treated only with CIT before VenR. The estimated 3-year PFS was 84% in BTKi-exposed patients compared with 97% in BTKi-naïve patients ($P = 0.03$) (Fig. 2A).

Biological risk factors were subsequently analyzed, TP53 disruption significantly affected long-term disease control, with an estimated 3-year PFS of 64% in TP53-disrupted patients compared with 97% among TP53 wild-type patients ($P = 0.005$) (Fig. 2B).

Conversely, no significant differences were observed according to IGHV mutational status ($P = 0.9$) (Fig. 2C). Likewise, age, comorbidity burden (CIRS > 6), and bulky disease were not associated with differences in progression-free survival.

Overall, seven disease progressions were observed, occurring at a median of 22 months (range, 10–46). Most events occurred during VenR treatment, whereas only two patients experienced late progression beyond 24 months; both subsequently received pirtobrutinib and remained alive at data cut-off. Two patients progressing at 20 and 22 months, respectively, received zanubrutinib as subsequent therapy. PD was confirmed by BM evaluation in all cases. One patient developed Richter transformation, histologically confirmed by lymph node biopsy, and died after the first cycle of CIT. One additional patient with documented PD remained under active surveillance at data cut-off, as iwCLL criteria for treatment initiation had not yet been met.

Secondary endpoints

Overall survival

OS was assessed in the entire cohort (N = 90), irrespective of treatment completion or availability of EOT response and MRD assessments. After a median follow-up of 40 months (range, 7–72), the estimated 3-year OS was 94%, (95% CI, 86% – 97%; Fig. 3).

EOT clinical response

In the overall cohort, 68 patients were evaluable for EOT clinical response and 65 for EOT-MRD.

Among the 90 patients enrolled, 68 patients underwent response assessment at EOT since 11 were still receiving treatment at data cut-off, and 11 discontinued VenR before completing the planned 24-month treatment. Reasons for early discontinuation were PD (n = 5), infectious complications (n = 3), allogeneic hematopoietic stem cell transplant (allo-HSCT) after achieving a clinical response (n = 1), development of a solid tumor (n = 1), and accidental death (n = 1). The five PD events occurred before the scheduled EOT assessment and were considered treatment failures.

Among the 68 patients who reached the EOT response assessment, 57 (83.8%) achieved CR, 10 (14.7%) PR, and 1 (1.5%) had PD, resulting in an ORR of 98.5% among EOT-evaluable patients. PRs were all characterized by isolated, low-volume residual lymphadenopathy (> 1.5 cm short-axis diameter, all < 3 cm) according to the scheduled EOT-CT. EOT clinical response did not significantly discriminate PFS, with a 40-month PFS of 100% in patients achieving CR compared with 90.9% in those with PR/PD (log-rank P = 0.21; Fig. 4A).

EOT measurable residual disease

EOT-MRD was assessed by MFC in 65 patients. Among the remaining patients, 11 were still receiving treatment at data cut-off, 11 had discontinued VenR before the scheduled EOT assessment, and three did not undergo MFC for logistical reasons. Among the 65 evaluable patients, 55 (84.6%) achieved undetectable MRD (uMRD), whereas 10 (15.4%) remained MRD-positive. Of the 10 patients achieving PR, 6 were MRD-positive and 4 achieved uMRD. Conversely, 4 patients classified as clinical CR remained MRD-positive, highlighting the incomplete overlap between clinical response and MRD status.

MRD status strongly influenced treatment durability: patients achieving uMRD maintained a 100% PFS at 40 months, whereas MRD-positive patients experienced significantly inferior outcomes ($P = 0.0021$). Beyond 40 months, the PFS curve of MRD-positive patients progressively declined, reaching approximately 45% at 48 months (Fig. 4C).

These findings suggest that EOT-MRD provides additional prognostic information beyond conventional clinical response assessment.

Safety and treatment feasibility

VenR was generally well tolerated, and the safety profile was consistent with that reported in pivotal clinical trials (Table 2). Grade ≥ 3 neutropenia occurred in 6 patients (6.7%), while febrile neutropenia was observed in 3 (3.3%). G-CSF was introduced as secondary prophylaxis in patients developing an ANC $< 1.0 \times 10^9/L$. Grade ≥ 3 thrombocytopenia in 2 patients (2%), and febrile neutropenia in 3 patients (3%). Only one case (1%) of TLS was observed, while infusion-related reactions to rituximab occurred in 7 patients (8%). Infectious complications were uncommon and included symptomatic SARS-CoV-2 infection (4%), pneumonitis (5%), and one case of HBV-related hepatitis. Diarrhea was reported in 3 patients (4%).

Treatment modifications were frequently required during routine clinical practice. Temporary venetoclax interruption occurred in 21 patients (23%), with a median interruption of 17 days (range, 5–50), whereas permanent venetoclax discontinuation occurred in 5 patients (6%). Venetoclax dose reduction below 400 mg/day was required in 7 patients, while rituximab dose reduction to 375 mg/m² was performed in 26 patients (29%). The median duration of venetoclax exposure was 18 months (range, 12–22).

Importantly, neither dose reductions nor temporary treatment interruptions adversely affected PFS. Prior bendamustine exposure was associated with a higher incidence of hematologic and infectious complications leading to treatment discontinuation (64.1% vs 29.4%; $P = 0.001$) without a significant difference in OS between bendamustine-exposed and unexposed patients.

Discussion

The therapeutic landscape of R/R CLL has profoundly changed with the introduction of targeted therapies. Although MURANO established fixed-duration VenR for R/R CLL, contemporary practice increasingly includes BTKi-exposed patients, making treatment sequencing a major unresolved issue. More than half of our cohort had prior BTKi exposure. Our study confirms that fixed-duration VenR maintains excellent efficacy outside the setting of prospective clinical trials. After a median follow-up of 40 months, 3-year PFS was 90% and OS 94%, supporting durable disease control despite older age, comorbidities, and adverse biological characteristics. Although cross-study comparisons should be interpreted cautiously, our results are consistent with the long-term efficacy reported in the MURANO trial and subsequent follow-up analyses, supporting the reproducibility of fixed-duration venetoclax-based therapy in routine clinical practice.

A major finding concerns the impact of previous BTKi exposure. Patients previously treated with BTKi experienced significantly shorter PFS than those receiving VenR after chemoimmunotherapy alone. This is particularly relevant because our cohort reflects the current therapeutic scenario in which BTKi are frequently used in earlier treatment lines. Previous retrospective experiences have suggested inferior outcomes with venetoclax after BTKi discontinuation, although available cohorts were generally limited by short follow-up and heterogeneous treatment strategies [5, 6, 17]. Our data strengthen these observations by demonstrating that previous BTKi exposure remains associated with inferior long-term disease control despite the use of fixed-duration VenR. Notably, most BTKi-exposed patients had switched to VenR because of disease progression after prolonged BTKi exposure (median, 47 months), whereas only 8 discontinued BTKi because of intolerance. Thus, the inferior PFS observed after prior BTKi exposure may primarily reflect a population with biologically selected, treatment-resistant disease rather than prior BTKi exposure per se. This interpretation is consistent with a more biologically complex disease characterized by cumulative treatment pressure, clonal evolution, and the selection of resistant subclones. These results highlight the growing importance of therapeutic sequencing in the management of R/R CLL and support the need for prospective studies specifically designed to define the optimal sequence of targeted agents. Of note, prior BTKi exposure in our cohort consisted almost exclusively of ibrutinib; therefore, whether these findings can be extrapolated to patients previously treated with second-generation covalent BTKi, such as acalabrutinib or zanubrutinib, remains to be established.

Disease biology remained a key determinant of treatment outcome. Consistent with previous studies, TP53 abnormalities retained a strong adverse prognostic impact despite treatment with venetoclax, confirming that TP53 disruption continues to identify patients at increased risk of disease progression in the era of targeted therapies [6, 18, 19]. Conversely, IGHV mutational status was not associated with PFS. Although fixed-duration venetoclax-based therapies substantially improve outcomes in patients with unmutated IGHV compared with chemoimmunotherapy, long-term analyses indicate that unmutated IGHV may retain prognostic relevance [6, 19]. The absence of an IGHV effect should be interpreted cautiously given the limited number of progression events. An additional strength of our study is the evaluation of MRD in routine clinical practice. Among evaluable patients, 85% achieved undetectable MRD at the end of treatment, and MRD negativity translated into remarkably durable disease control. In contrast, patients with detectable MRD showed a progressive decline in PFS, reaching approximately 45% at 48 months. These findings closely reproduce the biological model proposed by the MURANO investigators, in which MRD status at the end of treatment represents one of the strongest predictors of subsequent disease progression. Beyond confirming the prognostic value of MRD outside clinical trials, our results support its incorporation into routine clinical practice as an objective tool for post-treatment risk stratification and potentially for future MRD-guided therapeutic strategies.

Treatment feasibility was favorable. Approximately one-quarter of patients required temporary venetoclax interruption, but treatment modifications did not adversely affect PFS, consistent with previous analyses [20]. The low incidence of grade ≥ 3 neutropenia and febrile neutropenia may partly reflect secondary G-CSF prophylaxis after development of neutropenia, although causal inference is not

possible. Prior bendamustine exposure was associated with more frequent neutropenia and/or infectious complications without a detectable impact on OS. Systematic supportive care, including antimicrobial prophylaxis, secondary G-CSF, HBV monitoring/prophylaxis, and vaccination, may have mitigated their clinical impact [10–12].

This study is limited by its retrospective design, incomplete EOT-MRD assessment, relatively modest sample size, and low number of progression events, which precluded robust multivariable modeling and limits subgroup analyses. Conversely, strengths include its multicenter design, relatively long follow-up, high proportion of BTKi-exposed patients, and extensive biological characterization. TP53 mutation analysis by NGS was available in 91.1% of patients, IGHV status in 95.6%, and FISH in 95.6%, allowing outcomes to be evaluated in the context of both disease biology and prior treatment exposure.

In conclusion, fixed-duration VenR remains an effective and feasible treatment for R/R CLL in routine practice. Prior BTKi exposure, TP53 disruption, and detectable EOT-MRD identified patients with less durable disease control, while treatment adaptations did not appear to compromise efficacy. These findings highlight the importance of disease biology, depth of response, and prior treatment history in optimizing therapeutic sequencing.

Figure 1. Progression-free survival in the overall cohort.

Kaplan–Meier estimates of progression-free survival (PFS) in the overall study population (N = 90). At a median follow-up of 40 months, the estimated 3-year PFS was 90% (95% CI, 81%–95%). Tick marks indicate censored observations.

Figure 2. Progression-free survival according to prior treatment exposure and biological risk factors.

Kaplan–Meier estimates of PFS according to (A) prior Bruton tyrosine kinase inhibitor (BTKi) exposure, (B) TP53 disruption, defined as TP53 mutation and/or del(17p), and (C) IGHV mutational status. Three-year PFS was 84% in BTKi-exposed patients versus 97% in BTKi-naïve patients (log-rank P = 0.03) and 64% in patients with TP53 disruption versus 97% in those without TP53 disruption (log-rank P = 0.005). No significant difference in PFS was observed according to IGHV mutational status (log-rank P = 0.9). Tick marks indicate censored observations.

Figure 3. Overall survival in the overall cohort. Kaplan–Meier estimate of overall survival (OS) in the overall study population (N = 90). The estimated 3-year OS was 94% (95% CI, 86%–97%) after a median follow-up of 40 months (range, 7–72). Tick marks indicate censored observations

Figure 4. Progression-free survival according to end-of-treatment measurable residual disease status and to end-of-treatment clinical response

Kaplan–Meier estimates of progression-free survival (PFS) according to (A) end-of-treatment (EOT) measurable residual disease (MRD) status and (B) EOT clinical response. (A) At 40 months, PFS was 100% in patients achieving undetectable MRD (uMRD) compared with 90% in MRD-positive patients (log-

rank $P = 0.0021$). (B) At 40 months, PFS was 100% in patients achieving clinical complete response (cCR) compared with 90.9% in patients with partial response or progressive disease (PR/PD), with no statistically significant difference between groups (log-rank $P = 0.21$). uMRD was defined as < 1 CLL cell per 10^4 leukocytes ($< 10^{-4}$) by multiparametric flow cytometry. Tick marks indicate censored observations.

Declarations

Conflict of Interest

The authors declare no conflicts of interest relevant to this work.

Ethics statement

The study was conducted in accordance with the principles of the Declaration of Helsinki and was approved by the local Ethics Committee (protocol No. 163/2026). Given the retrospective and observational nature of the study, the requirement for written informed consent was waived in accordance with applicable regulations. Patient data were collected and analyzed in anonymized form.

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Author Contribution

Conceptualization: [C.G, G.F; A.F]; Methodology: [M.P, C.G]; Data curation: all authors; Formal analysis: C.G; Investigation: all authors; Writing—original draft preparation: [C.G, A.F]; Writing—review and editing: all authors; Supervision: [M.P, F.P]. All authors have read and agreed to the published version of the manuscript. All authors meet the International Committee of Medical Journal Editors (ICMJE) criteria for authorship and agree to be accountable for all aspects of the work.

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Data Availability Statement

The data supporting the findings of this study are not publicly available because they contain information that could compromise the privacy of research participants. De-identified data may be made available from the corresponding author upon reasonable request and subject to approval by the relevant ethics committee and participating institutions.

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Figures

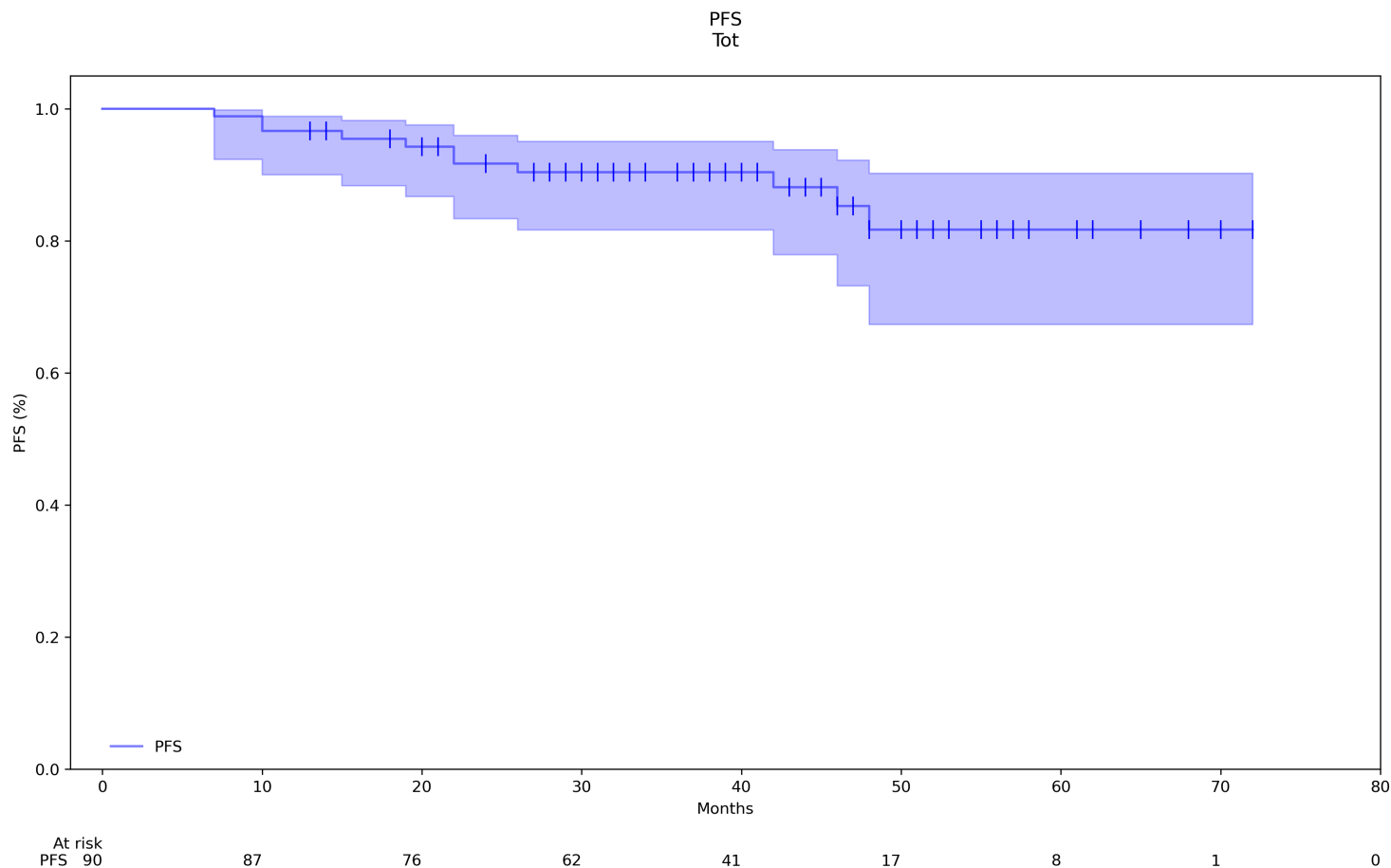


Figure 1

Progression-free survival in the overall cohort.

Kaplan–Meier estimates of progression-free survival (PFS) in the overall study population (N=90). At a median follow-up of 40 months, the estimated 3-year PFS was 90% (95% CI, 81%–95%). Tick marks indicate censored observations.

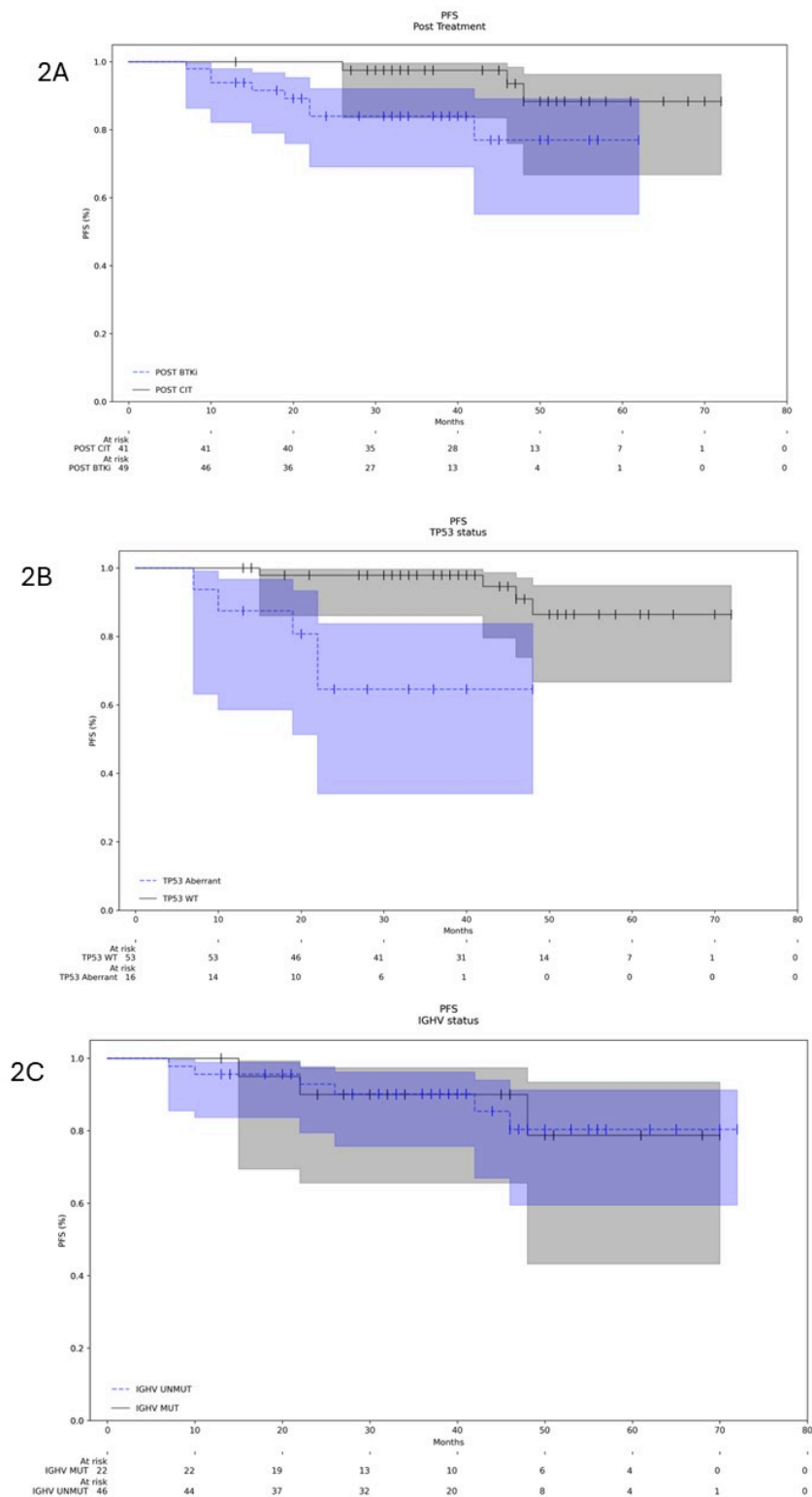


Figure 2

Progression-free survival according to prior treatment exposure and biological risk factors.

Kaplan–Meier estimates of PFS according to (A) prior Bruton tyrosine kinase inhibitor (BTKi) exposure, (B) TP53 disruption, defined as TP53 mutation and/or del(17p), and (C) IGHV mutational status. Three-year PFS was 84% in BTKi-exposed patients versus 97% in BTKi-naïve patients (log-rank $P=0.03$) and 64% in patients with TP53 disruption versus 97% in those without TP53 disruption (log-rank $P=0.005$). No

significant difference in PFS was observed according to IGHV mutational status (log-rank P=0.9). Tick marks indicate censored observations.

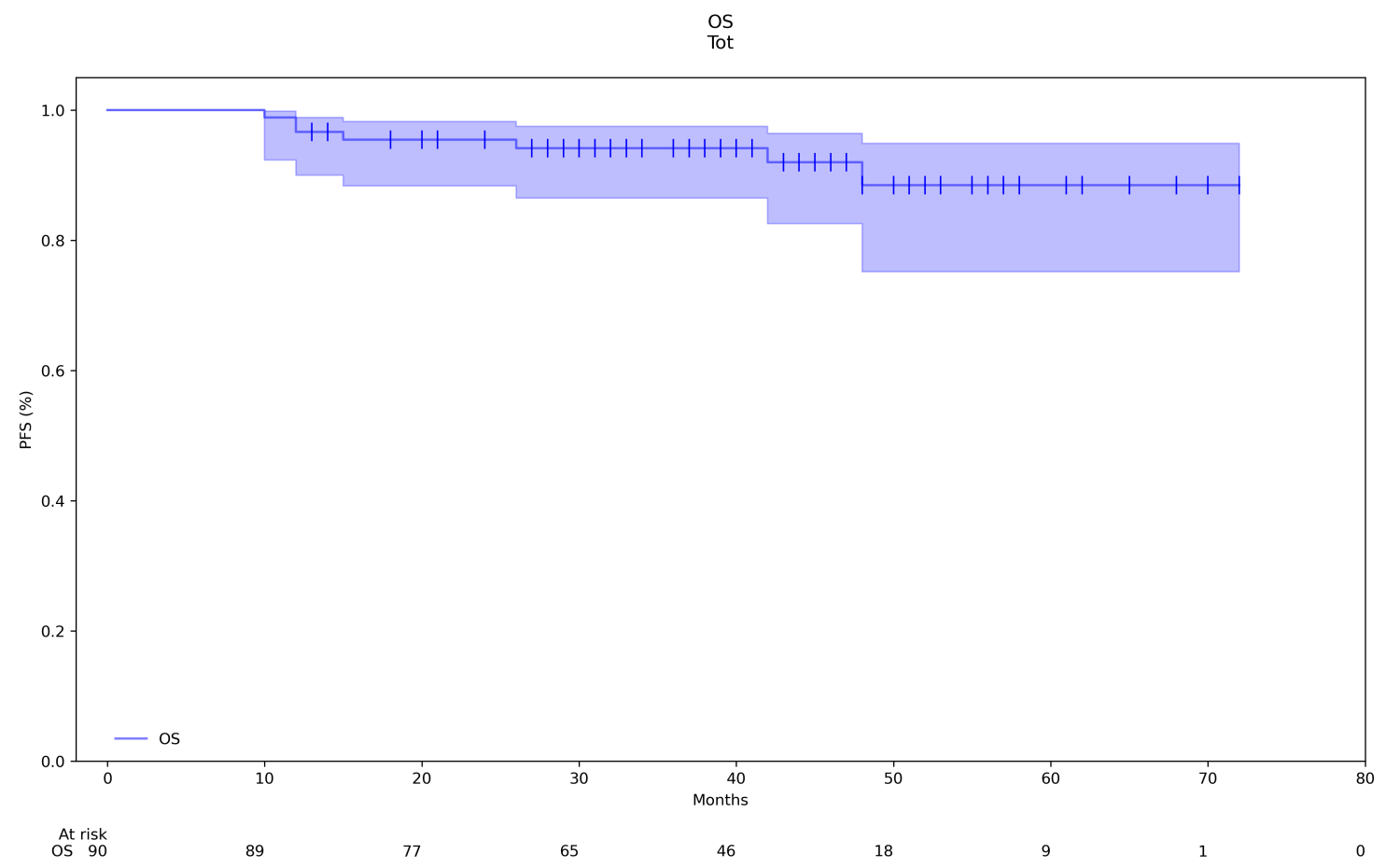


Figure 3

Overall survival in the overall cohort. Kaplan–Meier estimate of overall survival (OS) in the overall study population (N=90). The estimated 3-year OS was 94% (95% CI, 86%–97%) after a median follow-up of 40 months (range, 7–72). Tick marks indicate censored observations

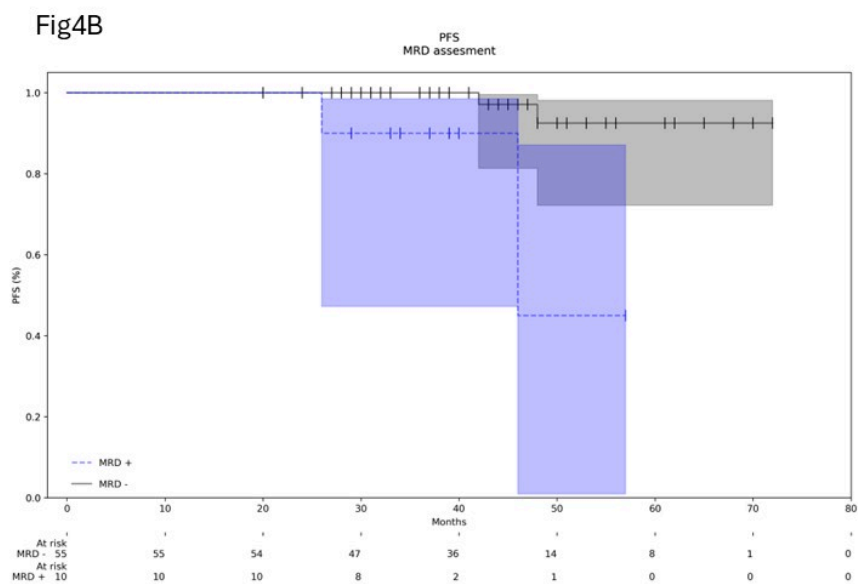
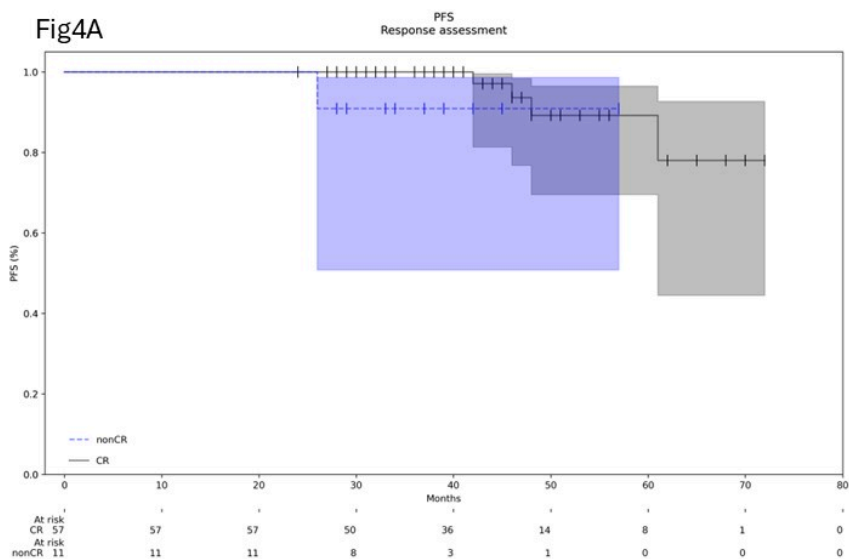


Figure 4

Progression-free survival according to end-of-treatment measurable residual disease status and to end-of-treatment clinical response

Kaplan–Meier estimates of progression-free survival (PFS) according to (A) end-of-treatment (EOT) measurable residual disease (MRD) status and (B) EOT clinical response. (A) At 40 months, PFS was 100% in patients achieving undetectable MRD (uMRD) compared with 90% in MRD-positive patients (log-

rank $P=0.0021$). (B) At 40 months, PFS was 100% in patients achieving clinical complete response (cCR) compared with 90.9% in patients with partial response or progressive disease (PR/PD), with no statistically significant difference between groups (log-rank $P=0.21$). uMRD was defined as <1 CLL cell per 10^4 leukocytes ($<10^{-4}$) by multiparametric flow cytometry. Tick marks indicate censored observations.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- [Table1VenR.docx](#)
- [Table2VenR.docx](#)
- [Supplemntaldata.docx](#)