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Regular Article

CLINICAL TRIALS AND OBSERVATIONS

Ibrutinib lead-in followed by venetoclax plus ibrutinib for relapsed/refractory chronic lymphocytic leukemia: the SAKK 34/17 trial

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KEY POINTS

- **Longer ibrutinib lead-in phase decreases TLS risk, and a minimum of 24-month IV combination enhances uMRD CR/CRi rates.**
- **Plasma circulating tumor DNA did not provide additional insights into immunoglobulin high-throughput sequencing MRD assessment.**

The combination of ibrutinib plus venetoclax (IV) in chronic lymphocytic leukemia (CLL) treatment leverages their complementary mechanisms of action. Studies investigating IV typically begin with a short initial course of ibrutinib, followed by venetoclax introduction for a limited duration, typically 12 months. The Swiss Group for Clinical Cancer Research (SAKK) 34/17 study is a single-arm, multicenter, phase 2 trial evaluating the effectiveness of a modified IV schedule in patients with relapsed/refractory (R/R) CLL. No prior exposure to BTK or BCL2 inhibitors was allowed. The lead-in phase with ibrutinib was extended to 6 months to reduce the tumor burden and related tumor lysis syndrome (TLS) risk. Additionally, the treatment phase with IV is prolonged to a minimum of 24 months to enhance the undetectable minimal residual disease (uMRD; 10^{-4}) rate. The primary end point was the rate of complete response or complete response with incomplete bone marrow recovery (CR/CRi) with uMRD in both bone marrow (BM) and peripheral blood (PB). Secondary end points included assessing the proportion of patients transitioning to a low-risk category for TLS after receiving ibrutinib lead-in. Of the 30 enrolled patients with R/R CLL, 40.0% achieved uMRD CR/CRi by intention-to-treat analysis, and 53.3% showed uMRD in the BM and PB. After the lead-in period with ibrutinib, 57.1% of patients achieved a low risk of TLS. At cycle 31, the progression-free survival rate was 89.9%. These results contribute to the increasing body of evidence supporting the idea that a longer IV duration is beneficial for enhancing therapeutic effectiveness. This trial was registered at www.clinicaltrials.gov as #NCT03708003.

Introduction

The pathophysiology of chronic lymphocytic leukemia (CLL) involves the proliferation of malignant B cells and their migration to tissues, driven by chronic activation of the B-cell receptor signaling and enhanced cell survival owing to overexpression of the antiapoptotic BCL2 protein.[1,2] This dual mechanism results in the accumulation of CLL cells, leading to increasing tissue infiltration and symptomatic progression.

Considering the dual pathophysiology of CLL, the combination of ibrutinib plus venetoclax (IV) was expected to enhance the percentage of patients attaining undetectable minimal residual disease (uMRD).[3,4] Typically, in IV studies, there is a brief initial phase involving ibrutinib monotherapy aimed at diminishing tumor bulk and consequently reducing the risk of tumor lysis syndrome (TLS). However, despite a significant reduction in the size of lymph nodes and spleen within 3 months of ibrutinib monotherapy, the lymphocyte count may remain elevated.

A heightened lymphocyte count is one of the recognized risk factors for TLS.[5-8]

Achieving uMRD after the induction treatment serves as an early surrogate end point for progression-free survival (PFS), granting patients with uMRD status the opportunity for prolonged drug-free intervals and reduced medical interventions.[9] In IV studies, the treatment is often halted after 12 months from the initiation of the combination. However, at this time point, the combination has not fully exploited its effectiveness on minimal residual disease (MRD) as shown by the FLAIR trial and the MD Anderson Cancer Center study.[7,10] Consequently, after 12 months, the uMRD rate was lower than expected considering that IV combines the 2 most effective targeted agents for CLL.[5,11]

The Swiss Group for Clinical Cancer Research (SAKK) 34/17 study (NCT03708003) is a single-arm, open-label, multicenter, phase 2 trial evaluating the effectiveness of a modified schedule of the combination in patients with relapsed/refractory (R/R) CLL. In our modified schedule, the lead-in phase with ibrutinib is extended to 6 months to allow ibrutinib to better exploit its effect in reducing the risk of TLS. In addition, the induction phase with IV is extended to a minimum of 24 months in an MRD/response-driven approach to increase the rate of uMRD.

Methods

Patients

Patients with R/R CLL were recruited from 10 hospitals in Switzerland. The study was approved by the responsible local ethics committee. The study protocol (supplemental Data, available on the *Blood* website) was conducted according to the Declaration of Helsinki and Good Clinical Practice. All patients provided a written informed consent. Key inclusion criteria were age ≥18 years, World Health Organization performance status ≤2, diagnosis of CLL, progression after at least 1 line of therapy, no previous exposure to BTK inhibitors and venetoclax, need for systemic treatment according to the 2018 International Workshop on Chronic Lymphocytic Leukemia (iwCLL) guidelines,[12] creatinine clearance >30 mL/min, and no significant cardiac comorbidities or risk of bleeding.

Treatment

Patients received ibrutinib 420 mg daily for 6 cycles of 28 days each, followed by the addition of venetoclax (5-weekly dose-escalation up to 400 mg daily). Patients who had a high TLS risk after cycle 6 received the first 2 doses of venetoclax in an inpatient setting. Combined therapy was given for 24 cycles of 28 days each. Patients who were not in uMRD complete remission/complete remission with incomplete bone marrow recovery (CR/CRi) after cycle 30 received consolidation treatment with the combination until the achievement of uMRD CR/CRi, progression, unacceptable toxicity or up to 5 years. Dose modification guidelines are included in the supplemental Data.

End points

Safety and efficacy analyses included patients who received at least 1 dose of treatment as per intention to treat. The primary end point was the rate of CR/CRi with uMRD by flow cytometry in blood and marrow at the end of cycle 30. Secondary efficacy end points were the rate of low risk of TLS at the time of venetoclax start, the overall response rate (ORR) at the end of cycle 30, the uMRD rate at the end of cycle 30, the best CR/CRi rate, the best ORR, the best uMRD rate, PFS, and overall survival (OS). The ORR was defined as the proportion of patients having achieved a CR/CRi or partial response (PR) according to the 2018 iwCLL criteria at the end of cycle 30 (±14 days). The CR/CRi rate was defined as the proportion of patients having achieved a CR/CRi at the end of cycle 30 (±14 days). Patients without any response assessment at the end of cycle 30 (±14 days) were counted as nonresponders. PFS was defined as the time from registration to progression or death from any cause, whichever occurred first. Patients not having an event at the time of analysis and patients starting a new antileukemic therapy in the absence of an event were censored at the date of their last tumor assessment showing nonprogression before starting a new antileukemic treatment, if any. OS was defined as the time from registration to death from any cause. Patients not having an event at the time of analysis were censored at the date they were last known to be alive. Efficacy end points were measured according to the 2018 iwCLL guidelines.[12] Safety end points were the rate of adverse events (AEs) defined according to the National Cancer Institute Common Terminology Criteria for Adverse Events (version 5) and the rate of AEs of special interest (cardiovascular events, TLS, and major hemorrhage). Exploratory analyses included the evaluation of MRD in peripheral blood (PB) cells by immunoglobulin high-throughput sequencing (HTS; ClonoSEQ Assay, Adaptive Biotechnologies Corporation) and the evaluation of MRD and clonal evolution in plasma cell-free DNA (cfDNA) by cancer personalized profiling by deep sequencing (CAPP-seq). Further details regarding the methods used are outlined in the supplemental Data.

Assessments

Blood and bone marrow (BM) MRD assessments, clinical assessments, and computed tomography scans were conducted at screening, at the end of cycle 6 (before starting venetoclax, except for BM biopsy), at the end of cycle 30 (after 24 cycles of IV), and yearly thereafter. Immunophenotyping for diagnosis confirmation, conventional karyotyping, fluorescent in situ hybridization, and *TP53* and immunoglobulin heavy-chain variable (IGHV) region mutational status were evaluated centrally. MRD was centrally assessed by 8-color flow cytometry (sensitivity 10^{-4}, uMRD $<10^{-4}$ [MRD4]) on a FACSCanto II System cytometer (Becton Dickinson AG, Eysins, Switzerland), and results were analyzed with Infinicyt CE-IVD 2.0.6 version.

Statistical analysis

The primary assumption was a rate of uMRD CR/CRi exceeding 35% after cycle 30. The sample size was determined according to the A'Hern design, using a 1-sided statistical significance (α) of 5% and statistical power of 80% to test that the rate of uMRD CR/CRi is no greater than 15% (H_0) against the alternative that it exceeds 35% (H_1). Thus, if a minimum of 11 of 30 patients achieved uMRD CR/CRi at the end of cycle 30, then the combined treatment would be deemed worthy of further investigation. Data were frozen on 21 September 2023. A sample size of 28 patients also achieved 83% power to detect an increase of 25% in the rate of patients fulfilling the criteria for low TLS risk after 6 cycles of ibrutinib monotherapy, assuming a proportion of 25% under the null hypothesis and a proportion of 50%

under the alternative hypothesis and using a 1-sided binomial test (1-sided 0.05 type I error). The populations used for the statistical analysis included (1) full analysis set, defined as all registered patients who received at least 1 dose of trial treatment excluding patients with major eligibility violations, and (2) safety analysis set, defined as all patients who received any dose of trial treatment. Fisher exact test was used to assess associations between variables and responses. Secondary end points of PFS and OS were estimated by Kaplan-Meier analysis. Descriptive statistics were calculated for biomarker assays, and log-rank tests were performed to assess associations between variables and survival outcomes. All *P* values were 2 sided with statistical significance evaluated at the .05 alpha level; 95% confidence intervals (CIs) were calculated to assess estimate precision. All analyses were performed on IBM SPSS v.28, SAS 9.4 (SAS Institute Inc, Cary, NC, USA) and R v.4.2.0 software.

Results

Patient characteristics

Between March 2019 and August 2020, 30 patients with previously treated CLL were enrolled (Figure 1). At study entry, median age was 69 years (range, 53-80) and 73.3% of patients (22/30) were male. Ten patients (33.3%) had Rai stage III/IV, 24 (80.0%) had unmutated IGHV, and 9 (30.0%) had del(17p) and/or *TP53* mutation. Complex karyotype, defined as ≥3 unrelated abnormalities on conventional karyotyping, was detected in 8 patients (26.7%). The median number of previous lines of therapy was 1 (range, 1-3). Prior exposure to BTK inhibitors or venetoclax was not allowed per the study's protocol. The baseline patient and disease characteristics are presented in Table 1. The median time on study treatment was 28.6 months (range, 2.4-41.7).

Baseline genetic landscape

The mutation profile of purified PB CLL cells and plasma circulating tumor DNA (ctDNA) was centrally analyzed by CAPP-seq (Figure 2A). Most mutations were detected in the DNA of CLL cells, whereas only a minority of mutations were restricted to the ctDNA compartment. Notably, ctDNA did not disclose any clinical mutation (eg, *TP53* mutations) beyond those detected in the DNA of CLL cells (Figure 2B). Similarly, high concordance between copy number abnormalities detected on cellular DNA and ctDNA was observed (Figure 2C). One patient with the t(14;18) translocation, which can occur in 5% of typical IGHV mutated CLL,[13,14] also harbored 2 distinct mutations of the *BCL2* gene mapping to the 5′-UTR and at codon 21, respectively, which are consistent with AID misfire, but not with drug resistance.[15]

Efficacy

Nineteen patients were assessable for the primary end point, whereas 11 patients were not evaluated because they stopped study treatment before cycle 30. In the intention-to-treatment analysis, these 11 patients were considered and scored as not in uMRD CR/CRi. The SAKK 34/17 trial achieved its primary end point, with an intention-to-treat uMRD CR/CRi rate of 40% (90% CI, 25-57) at the end of cycle 30, exceeding the study hypothesis (H0) of 15%. PR with uMRD was observed in 4 of 30 patients (13.3%), whereas PR with measurable MRD occurred in 3 of 30 patients (10%) (Figure 3A). Four patients (13.3%) underwent maintenance (clinical and genetic characteristics of

Figure 1. CONSORT diagram.

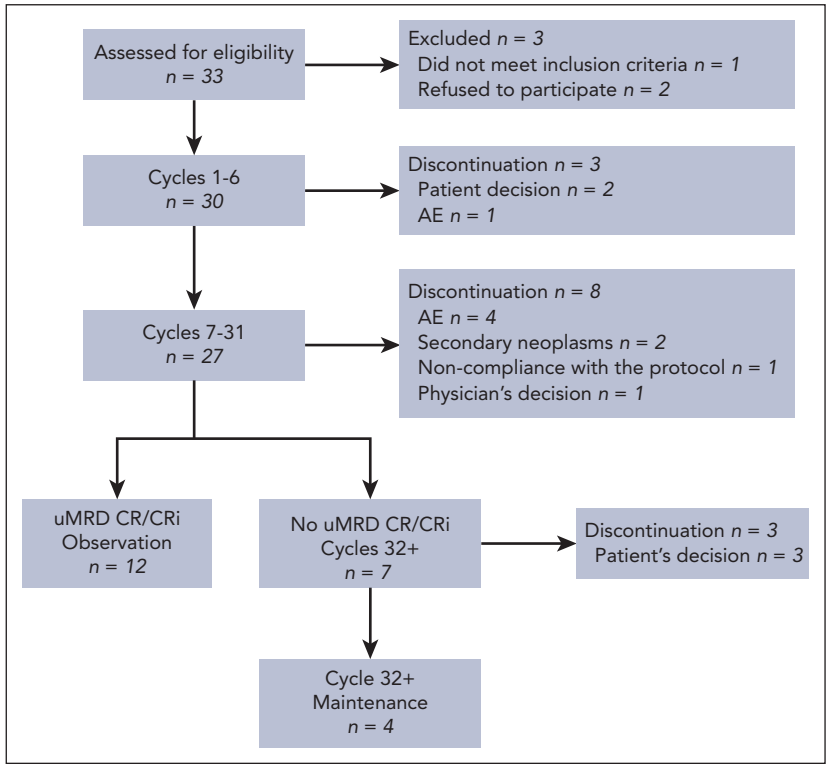

Table 1. Characteristics of the study cohort

Characteristics	N	Summary
Age, median (range), y	30	69 (53-80)
Sex, n (%)	30	
Female		8 (26.7)
Male		22 (73.3)
Rai stage, n (%)	30	
0		3 (10.0)
I		9 (30.0)
II		8 (26.7)
III		4 (13.3)
IV		6 (20.0)
Hemoglobin, median (range), g/L	30	125 (75-166)
Neutrophils, median (range), $\times 10^9$/L	30	4.0 (0.0-8.0)
Platelets, median (range), $\times 10^9$/L	30	136 (44-391)
Lymphocytes, median (range), $\times 10^9$/L	29	15.0 (1.0-347.0)
Creatinine clearance, median (range), mL/min	30	67 (38-124)
Beta-2-microglobulin, median (range), mg/L	30	4.1 (1.7-8.7)
IgG levels, median (range), g/L	30	5.1 (2.4-10.4)
WHO performance status, n (%)	30	
0		16 (53.3)
1		13 (43.3)
2		1 (3.3)
CIRS total score, median (range)	30	2 (0-9)
TLS risk, n (%)	30	
Low risk		10 (33.3)
Medium risk		4 (13.3)
High risk		16 (53.3)
IGHV unmutated, n (%)	30	24 (80.0)
TP53 mutated, n (%)	30	8 (26.7)
17p13 deletion, n (%)	30	6 (20.0)
TP53 abnormality, n (%)	30	9 (30.0)
Complex karyotype, n (%)	30	8 (26.7)
11q23 deletion, n (%)	30	6 (20.0)
CLL IPI, n (%)	30	
Low risk		2 (6.7)
Intermediate risk		6 (20.0)
High risk		13 (43.3)
Very high risk		9 (30.0)
Previous lines of therapy, median (range)	29	1 (1-3)

CIRS, cumulative illness rating scale; CLL IPI, international prognostic index for chronic lymphocytic leukemia; IGHV, immunoglobulin heavy-chain variable; WHO, World Health Organization.

these patients are presented in supplemental Table 1). The primary end point categorized by the initial clinical and genetic characteristics is displayed in Figure 3B.

Among the 19 patients assessable at the end of cycle 30, BM uMRD rate was 84.2% (16/19) and PB uMRD rate was 100% (19/19). Among patients with at least 1 time point assessable, the uMRD rate in PB was 85.7% (24/28).

The high tumor burden category decreased from 53.3% (16/30) at baseline to 14.3% (4/28) after 6 cycles of ibrutinib lead-in. Only 4 patients required hospitalization for TLS monitoring owing to high TLS risk. The remaining hospitalizations during the venetoclax ramp-up phase were attributed to logistical reasons (eg, distance from the study site). Notably, the ibrutinib lead-in phase eliminated the need for hospitalization for TLS monitoring in 12 of 16 patients who were initially classified as at high risk of TLS before starting ibrutinib (Figure 3C). None of the patients experienced laboratory or clinical TLS according to Howard criteria[16] during the venetoclax ramp-up phase.

After a median follow-up of 42 months (range, 35-52), 1 patient had progressed. This patient discontinued study treatment after 2.4 months of ibrutinib lead-in owing to an AE and experienced disease progression 19.7 months after interrupting ibrutinib while on follow-up. Five patients died. One death was deemed probably related to ibrutinib. In particular, this patient passed away from a thrombotic stroke during the observation phase, 1.5 months after completing treatment. The remaining 4 deaths were assessed by investigators as unrelated or unlikely to be related to the study drugs and occurred after study treatment discontinuation. The causes of these unrelated deaths included worsening of pre-existing medical conditions 1 month after discontinuing the ibrutinib lead-in phase, gastroesophageal carcinoma 18 months after the end of study treatment while under observation, urosepsis 17 months after treatment while under observation, and disease progression 33 months after treatment while receiving venetoclax monotherapy as the next line of therapy. No patients developed a transformed disease.

Median PFS and OS were not reached; estimated PFS and OS rates at cycle 31 were 89.9% (95% CI, 71.5-96.6) and 93.3% (95% CI, 75.9-98.3), respectively (Figure 4A-B). One patient started a subsequent treatment for CLL after withdrawing from the study.

Safety

The most frequent AEs of any grade were hypertension (60.0%, 18/30), diarrhea (43.3%, 13/30), contusion (40.0%, 12/30), neutropenia (36.7%, 11/30), and fatigue (33.3%, 9/30). The most frequent grade ≥3 AEs related to ibrutinib were hypertension (30.0%, 9/30) and lung infection (3.3%, 1/30), whereas for venetoclax it was neutropenia (30.0%, 9/30; Table 2). The most frequent site of infections was the upper respiratory tract, followed by urinary tract and pneumonia, with 13.3% of patients (4/30) experiencing severe acute respiratory syndrome coronavirus 2 infection. Cardiovascular events included atrial flutter (3.3%, 1/30), atrial fibrillation (6.7%, 2/30), or other types of arrhythmias (6.7%, 2/30). Cardiovascular events were experienced in 2 patients (6.7%) during the ibrutinib lead-in phase and in 3 patients (10.0%) during the IV combination. Among the

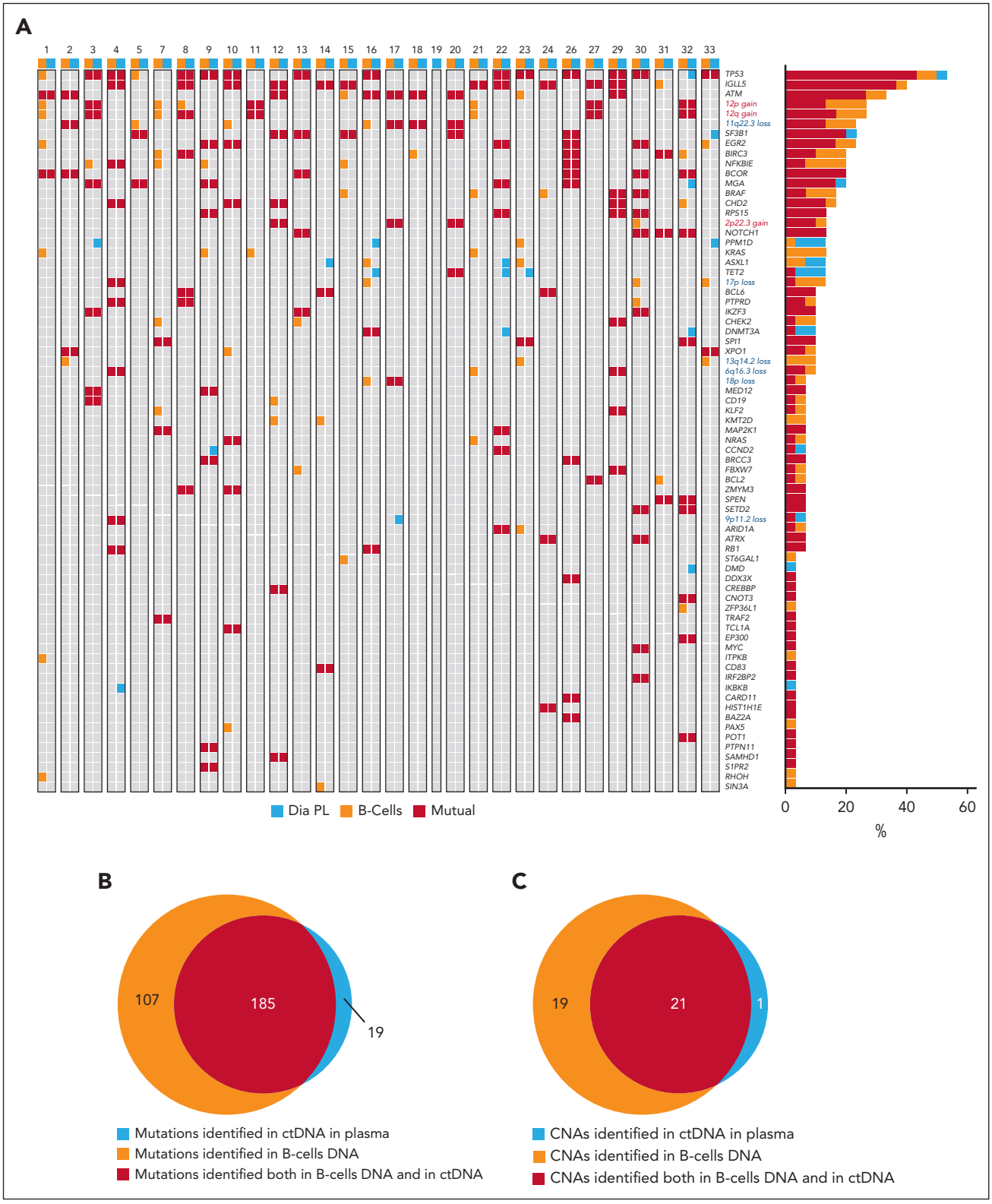

Figure 2. Baseline genomic landscape in paired purified B-cell DNA and ctDNA detected in plasma. (A) The heat map illustrates the paired analysis of purified CLL cells DNA (orange), ctDNA (light blue), and the shared mutations between B cells and ctDNA compartments (red). Each column represents a sample, and genes/copy number abnormalities (CNAs) are represented in rows. Venn diagrams summarizing the overall number of mutations (B) and CNAs (C) discovered only in B-cellular DNA (orange), only in plasma ctDNA (light blue), or in both plasma ctDNA and B-cellular DNA (red). Dia PL, plasma ctDNA at diagnosis.

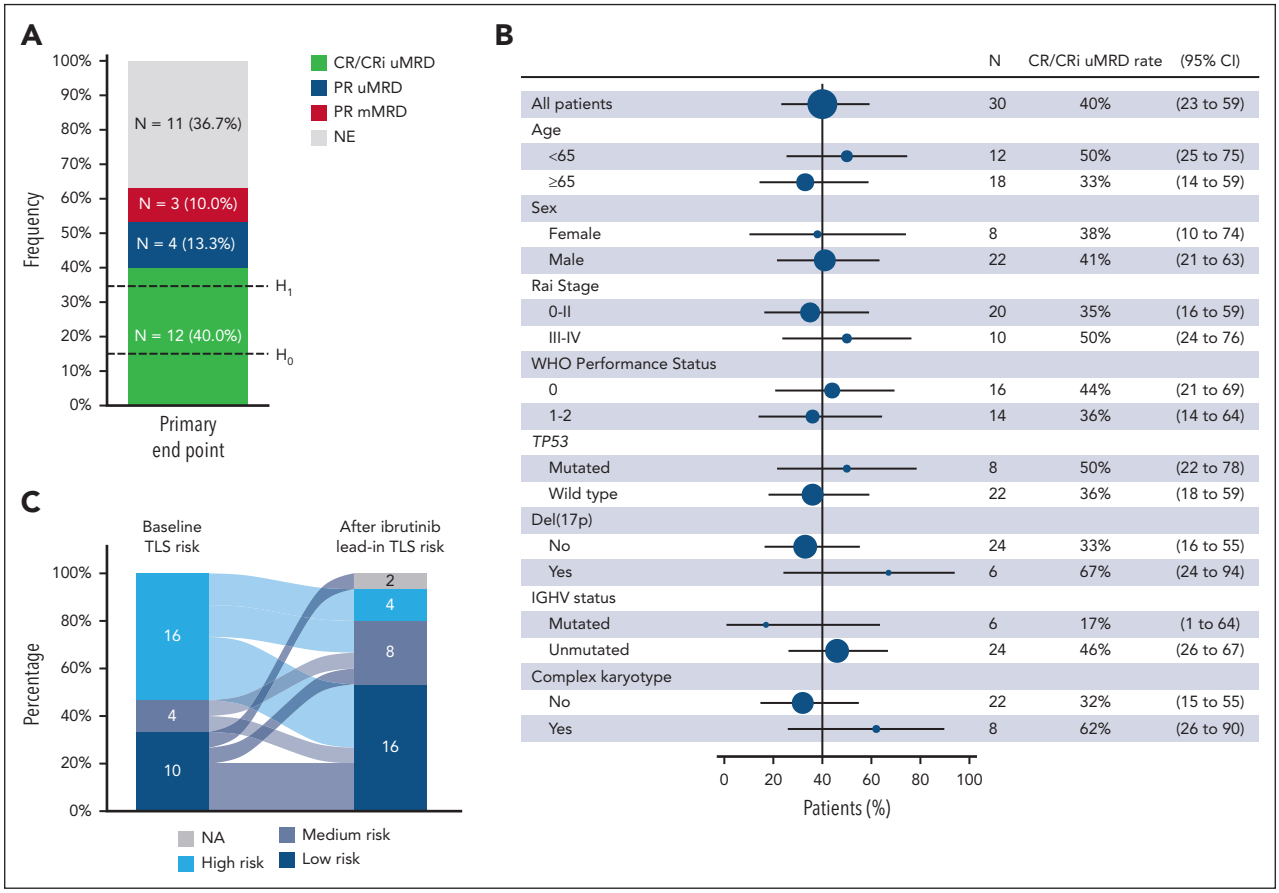

Figure 3. Responses and outcomes. (A) Response according to the primary end point in the intention-to-treat population; patients who discontinued study treatment before evaluation for response at the end of cycle 30 (n = 11) were classified as not evaluable (NE). (B) Forest plot of primary end point based on baseline characteristics. Primary end point was the rate of patients achieving uMRD by flow cytometry in BM and PB with CR/CRi at cycle 31. (C) TLS risk alluvial plot. H1, alternative hypothesis; mMRD, measurable minimal residual disease; NA, not available.

18 documented cases of hypertension, 43.3% were considered ibrutinib related by investigators' assessment. In particular, 5 (16.7%) occurred during the ibrutinib lead-in, 7 (23.3%) during the IV combination, and 1 (3.3%) during the observation phase.

Serious AEs (SAEs) were experienced by 63.3% of patients (19/30). Treatment-related SAEs that were attributed to ibrutinib by the investigator were experienced by 11 patients (36.7%), whereas 9 patients (30.0%) had treatment-related SAEs attributed to the combination (supplemental Table 2).

At the end of cycle 30 landmark, 11 patients (36.7%) have discontinued treatment, including 7 owing to AEs (supplemental Table 3). Three patients came off the study during the

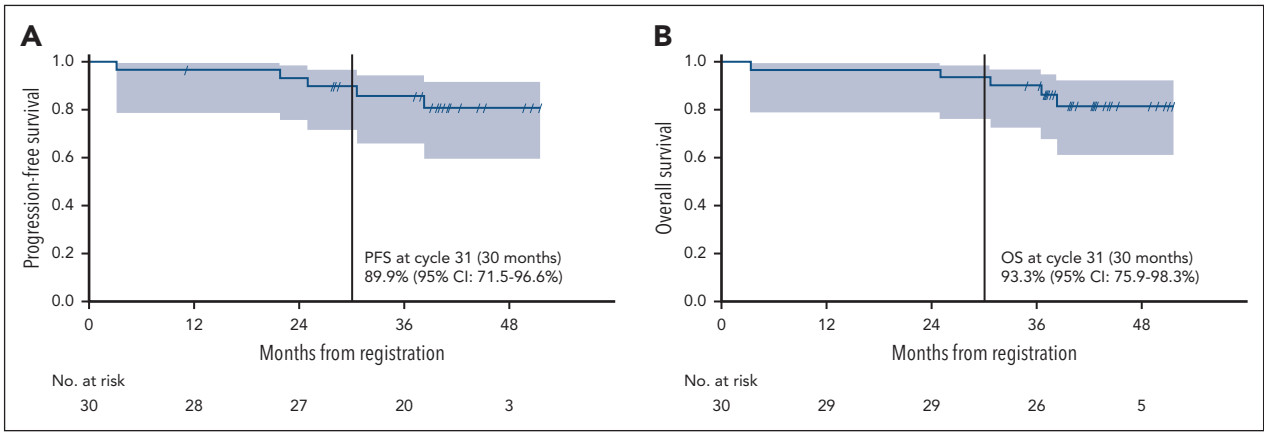

Figure 4. Kaplan-Meier analyses. (A) PFS and (B) OS.

Table 2. AEs occurring in at least 10% of patients (all grades) or at least 2% of patients (grade 3 or above in severity)

	Patients, N = 30				
	Grade 1	Grade 2	Grade 3	Grade 4	Grade 5
AEs, highest grade only, n (%)					
Fatigue	6 (20.0)	4 (13.3)	0	0	0
Dyspnea	2 (6.7)	1 (3.3)	0	0	0
Cough	7 (23.3)	0	0	0	0
Anemia	1 (3.3)	3 (10.0)	0	1 (3.3)	0
Platelet count decreased	2 (6.7)	1 (3.3)	0	1 (3.3)	0
Abdominal pain	2 (6.7)	2 (6.7)	0	0	0
Nausea	6 (20.0)	2 (6.7)	0	0	0
Vomiting	3 (10.0)	1 (3.3)	0	0	0
Rash maculopapular	5 (16.7)	1 (3.3)	0	0	0
AEs of particular interest, highest grade only, n (%)					
Atrial fibrillation/flutter	0	0	3 (10.0)	0	0
Hypertension	0	5 (16.7)	13 (43.3)	0	0
Neutrophil count decreased	0	2 (6.7)	6 (20.0)	3 (10.0)	0
Infections (other than COVID-19)	5 (16.7)	5 (16.7)	2 (6.7)	0	0
COVID-19 infection/pneumonia	0	1 (3.3)	3 (10.0)	0	0
Diarrhea	8 (26.7)	2 (6.7)	3 (10.0)	0	0
Epistaxis	6 (20.0)	0	0	0	0
Arthralgia	4 (13.3)	4 (13.3)	0	0	0
Hematoma	12 (40.0)	0	0	0	0
Fever	3 (10.0)	0	0	0	0
Stroke	0	0	0	0	1 (3.3)

ibrutinib lead-in phase (2 withdrew consent and 1 had a treatment delay of >4 weeks owing to infection). During the 24 cycles of the IV combination, 8 patients discontinued treatment: 6 owing to AEs (diarrhea, peripheral motor neuropathy, ventricular arrhythmia, multifocal atrial tachycardia, and 2 secondary neoplasms), 1 owing to severe noncompliance with the protocol, and 1 owing to physician's decision.

Dose modifications consisting of reductions, delays, and omissions were reported in 21 patients (70.0%; supplemental Table 4).

MRD

Overall, 5 longitudinal time points have been evaluated during treatment. After the ibrutinib lead-in phase, none of the patients achieved uMRD by flow cytometry and immunoglobulin HTS, and 4 patients were negative by CAPP-seq on plasma cfDNA (Figure 5A-C). At the end of cycle 30, by intention to treat, 53.3% of patients (16/30) achieved MRD4 in BM and 63.3% (19/30) in PB by flow cytometry; 20.0% (6/30) had MRD $<10^{-6}$ (MRD6) by immunoglobulin HTS in PB and 50.0% (15/30) had uMRD $<10^{-3}$ by CAPP-seq in plasma cfDNA (Figure 5D-G). Comparing the different assays head to head, immunoglobulin HTS showed the highest sensitivity in detecting MRD-positive cases (Figure 6A). Plasma cfDNA CAPP-seq failed to identify any instances of measurable MRD, which were instead classified as uMRD by immunoglobulin HTS in blood cells. This suggests that immunoglobulin HTS surpasses not only flow cytometry but also plasma-based MRD assays in performance (Figure 6A). CAPP-seq of plasma cfDNA did not detect any acquired resistance mutations of *BTK*, *PLCG2*, or *BCL2* while on treatment (Figure 6B). One patient (depicted by black dots in Figure 6B) had 2 *BCL2* mutations not associated with drug resistance (*BCL2:* NM_000633:exon2: c.C67T:p.L23L; *BCL2*: NM_000633: c.-512T>A), at baseline and cycle 7 (postibrutinib lead-in phase). However, by the end of treatment, these mutations were no longer detectable.

Discussion

The SAKK 34/17 trial confirms and further expands our understanding of how to schedule IV treatment for CLL. Here for the first time, to our knowledge, we are showing that a prolonged ibrutinib lead-in followed by 2 years of IV combination delivers a high rate of MRD clearance in the BM exceeding those previously attained in shorter IV combination trials. This was achieved despite the high prevalence of high-risk molecular and cytogenetic features of the study population, which included 30.0% *TP53* abnormalities, 80.0% unmutated IGHV, and 26.7% complex karyotype.

Despite a comparable median number of prior treatment lines and the absence of BTK or BCL2 inhibitors exposure, previous trials have reported a lower BM uMRD rate in R/R CLL after shorter IV treatment. The CLARITY trial, which tested 2 cycles of ibrutinib lead-in followed by 12 months of IV induction in

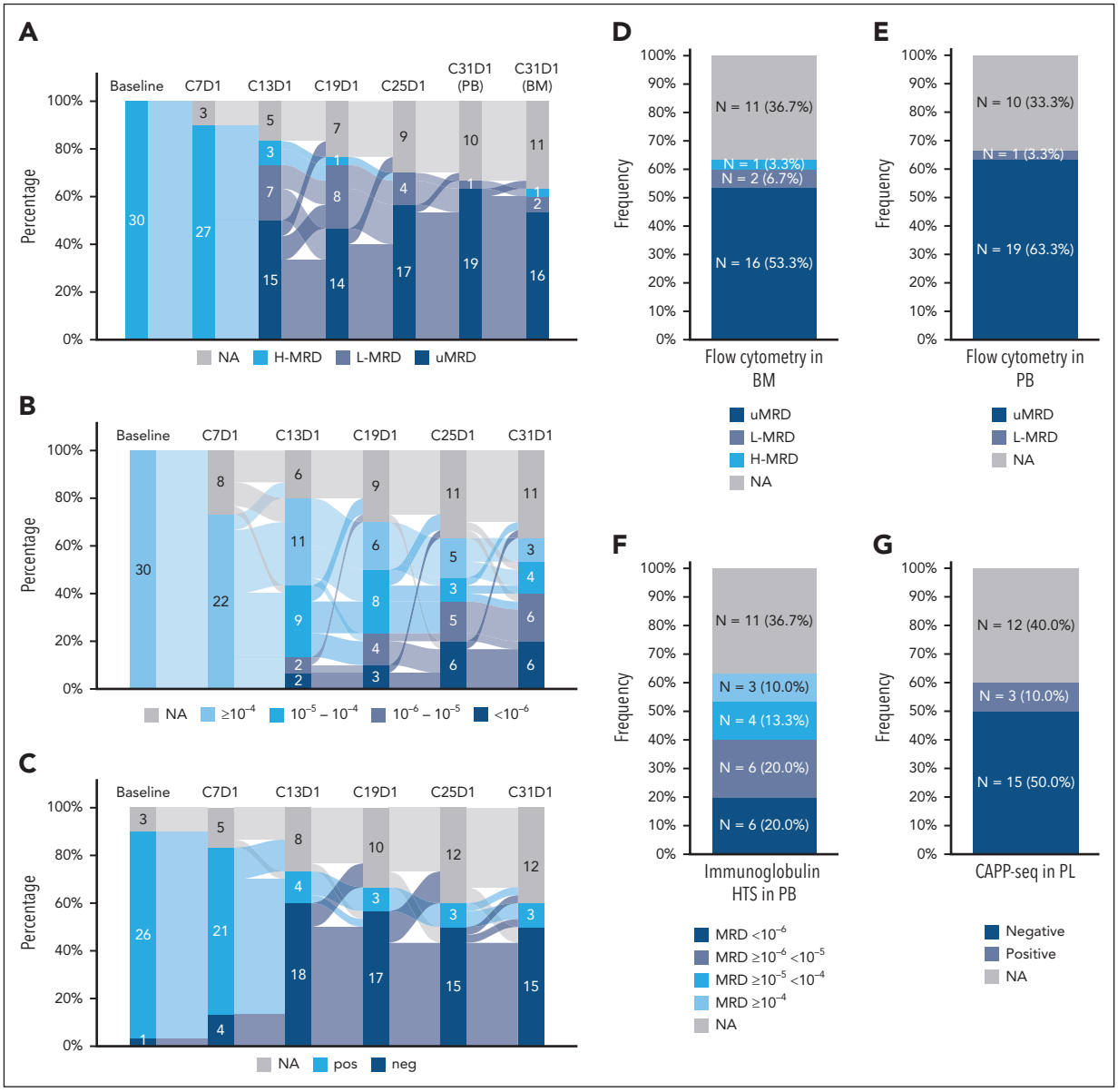

Figure 5. MRD dynamics and rates across compartments at the end of cycle 30. The alluvial plots show the MRD kinetics of the intention-to-treat population (A) by flow cytometry in PB from baseline to cycle 31; to the right, MRD response in BM is shown for comparison with MRD response in PB at cycle 31; (B) MRD kinetic by immunoglobulin HTS in cellular DNA and in which negativity cutoff was $<10^{-6}$ (MRD6); (C) by CAPP-seq in plasma cfDNA in which negativity cutoff was $<10^{-3}$ (uMRD $<10^{-3}$). MRD rates by flow cytometry in BM (D), by flow cytometry in PB (E), by immunoglobulin HTS in purified B-cell DNA (F), and CAPP-seq in plasma cfDNA (G). H-MRD, high MRD ($>10^{-2}$); L-MRD, low-MRD (10^{-4}-10^{-2}); PL, plasma; uMRD, unmeasurable MRD ($<10^{-4}$, MRD4).

53 patients with a median of 1 prior therapy, found a BM uMRD rate at the end of induction of 36%. Deletion of 17p occurred in 22% and unmutated IGHV status in 74%. The HOVON141/VISION trial, which involved 225 patients with a median of 1 previous therapy, evaluated 2 cycles of ibrutinib monotherapy followed by 13 cycles of IV combination therapy, for a total of 15 months of treatment. The uMRD rate in BM at the end of induction was 37%. *TP53* abnormalities occurred in 24% and unmutated IGHV status in 64%.[5,11]

Although cross-trial comparisons should be interpreted cautiously owing to differences in patient populations, our findings reinforce the evidence that IV should be administered for >1 year to improve uMRD rate. The rate of uMRD in BM among evaluable patients in the SAKK 34/17 trial is 84.2%, which aligns with the uMRD rates achieved in BM at 2 (52.4%), 3 (64.0%), 4 (65.9%), and 5 years (65.9%) among evaluable patients treated with IV in the FLAIR trial.[7] Furthermore, findings from the MD Anderson Cancer Center, presented in abstract format in 2019,[17] emphasized the incremental benefits of extending IV combination therapy beyond the initial 12 cycles, with uMRD rates in BM increasing from 48% at cycle 12% to 67% at cycle 24 among patients with previously treated CLL. Among treatment-naïve patients, the MD Anderson Cancer Center group consistently showed an increase of the uMRD rate in BM from 56% after cycle 12 of IV to 66% at cycle 24.[18] Finally, an update of the CLARITY trial presented in abstract format in 2020 showed that the uMRD in BM improved from 36% after

A

UPN	FC PB	FC BM	CAPPseq PL	Ig HTS PB
3417_007	pos	pos	neg	pos
3417_013	neg	pos	neg	pos
3417_030	neg	pos	neg	pos
3417_001	neg	neg	pos	pos
3417_003	neg	neg	pos	pos
3417_020	neg	neg	pos	pos
3417_027	neg	neg	neg	pos
3417_002	neg	neg	neg	pos
3417_004	neg	neg	neg	pos
3417_015	neg	neg	neg	pos
3417_023	neg	neg	neg	pos
3417_033	neg	neg	NA	pos
3417_029	neg	neg	NA	pos
3417_005	neg	neg	neg	neg
3417_011	neg	neg	neg	neg
3417_017	neg	neg	neg	neg
3417_018	neg	neg	neg	neg
3417_019	neg	neg	neg	neg
3417_031	neg	neg	neg	neg
3417_026	neg	NA	neg	NA

B

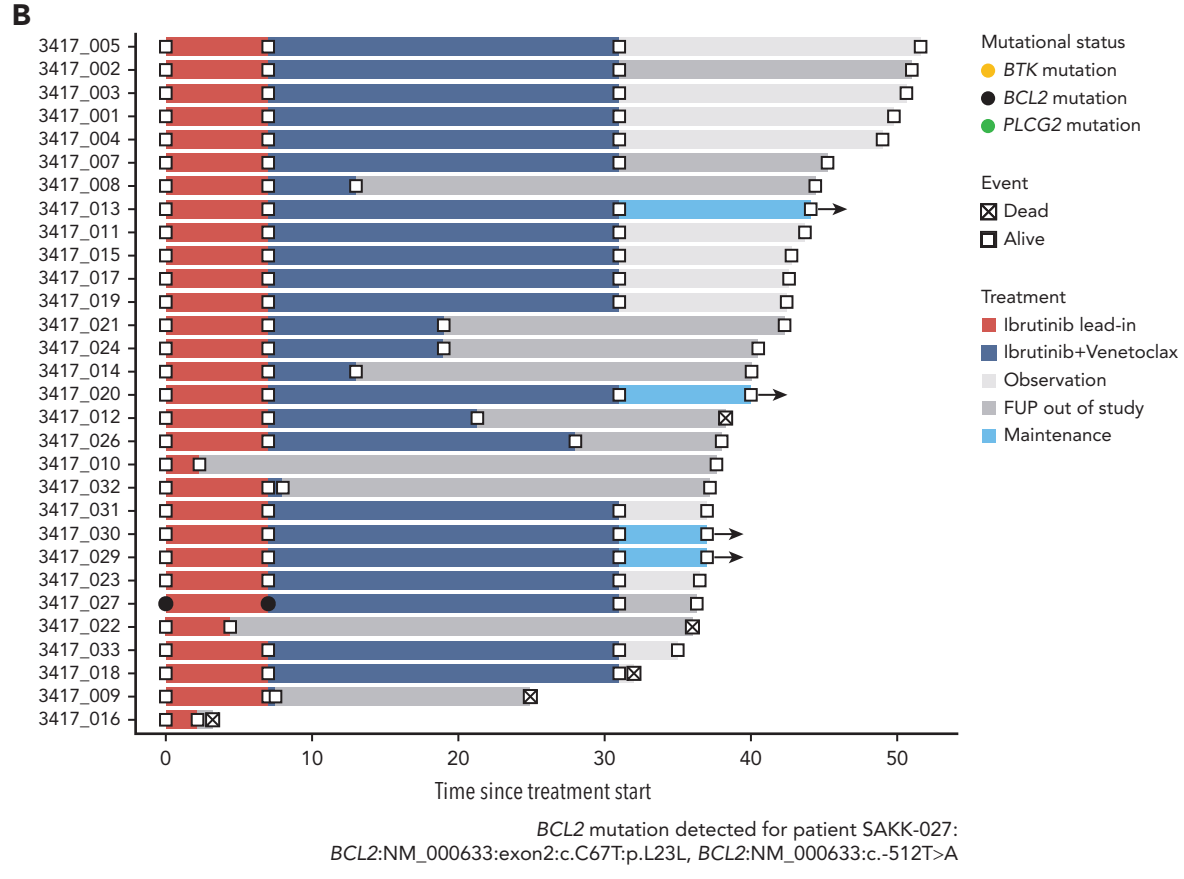

Figure 6. MRD status across compartments at the end of cycle 30 and monitoring of resistance mutations. (A) Head-to-head comparison of uMRD across compartments at the end of cycle 30. (B) Swimmer plot monitoring resistance mutations in ctDNA. Bars depict the duration of treatment for each patient up to cycle 31, representing the time until the final response assessment. Patients undergoing maintenance therapy are indicated by black arrows. FC, flow cytometry; FUP, follow-up; UPN, unique patient number.

12 cycles of IV to 44% after 24 cycles.[19] The FLAIR trial also showed significant improvements in PFS and OS with an MRD-driven approach and a median of 2 years of IV combination in previously untreated patients with standard-risk CLL.[7]

Prolonged exposure to IV combination therapy may impair tolerability, emphasizing the need to balance efficacy and safety in combination regimens. In our trial (median age 69 years, Cumulative Illness Rating Scale-Geriatric [CIRS-G] 2), 3% of patients experienced treatment-emergent deaths and 36.7% discontinued treatment for reasons unrelated to disease progression. By comparison, (1) the GLOW trial (median age 71 years, CIRS-G 9) found 6% of treatment-emergent deaths and 20% of discontinuations not caused by disease progression[20]; (2) the CAPTIVATE trial (median age 60 years, CIRS-G not reported) found <1% of treatment-emergent deaths and 10% of discontinuations not caused by disease progression[21]; (3) the HOVON141/VISION trial (median age 68 years, CIRS-G 2) found 2% of treatment-emergent deaths and 14% of discontinuations not caused by disease progression[11]; and (4) the CLARITY trial (median age 64 years, CIRS-G not reported) found no fatal AEs and 7% of discontinuations not caused by disease progression.[5] These findings underscore the trade-offs in tolerability associated with combination regimens, particularly in the context of prolonged treatment exposure.

It is not clear whether 6 months of ibrutinib lead-in is better than 2 or 3 months. Data on TLS risk of the SAKK 34/17 study have been analyzed in the context of findings from other trials, recognizing that comparisons across studies may be influenced by potential biases. These biases could stem from differences in baseline characteristics, such as patient tumor volume, lymphocyte count, and sensitivity or adherence to ibrutinib during the lead-in phase. Restricting the analysis to studies on R/R CLL, the CLARITY trial publication (8 weeks of ibrutinib lead-in) did not present any information on changes in TLS risk after ibrutinib lead-in or the need for hospitalization for TLS monitoring and prevention, with only 1 TLS event mentioned in its safety report.[5] Similarly, the HOVON141/VISION trial publication (8 weeks of ibrutinib lead-in) did not report these parameters, although 4 TLS events were recorded.[11] In our trial, the high tumor burden category decreased from 53% at baseline to 14% after 6 cycles of ibrutinib lead-in. Only 4 patients required hospitalization for TLS monitoring owing to high TLS risk, and no TLS events were observed. First-line IV trials are less directly comparable because of the expected lower rate of high tumor volume patients at presentation. In the GLOW trial, after 3 cycles of ibrutinib lead-in, the proportion of patients classified as having high tumor burden for TLS risk decreased from 24% at baseline to 1.9%, with no TLS cases reported.[20] In the CAPTIVATE trial, TLS risk among patients with high tumor burden dropped from 23% at baseline to 2% after 3 cycles of ibrutinib lead-in, with 18% of patients indicated for hospitalization.[8] In the MD Anderson Cancer Center trial, the high tumor burden category decreased from 13% at baseline to 3% after 3 cycles of ibrutinib lead-in, with 3 TLS events documented.[18] The SAKK 34/17 protocol allowed maintenance IV in patients who were not in uMRD CR/CRi at the end of cycle 30. The indication for continuing treatment in patients with uMRD but PR according to iwCLL criteria after venetoclax-based therapy remains uncertain and has not yet been fully addressed by clinical trials or meta-analyses. At the time the study was designed in 2016, there was no evidence on the prognostic significance of residual lymph nodes in patients who were otherwise uMRD after venetoclax-based treatment. However, the M13-982 trial reported by Stilgenbauer et al[22] highlighted that the persistence of residual lymph nodes had prognostic implications for patients treated with venetoclax. In addition, at that time, Kovacs et al[9] suggested that residual lymphadenopathy in uMRD patients might indicate a higher risk of relapse in the context of chemoimmunotherapy. Simon et al[23] demonstrated that, in a cohort including patients treated with venetoclax-based combinations, those with uMRD but in PR according to iwCLL criteria had shorter survival than patients with uMRD and CR.

Reported AEs in the SAKK 34/17 trial were consistent with those expected from either drug classes alone. Similar to the CLARITY trial, a comparable rate of grade 3-4 infectious AEs was observed (17%),[5] whereas a higher rate of infections was reported in the HOVON141/VISION study (27.4%).[11] Cardiac AEs (grade 3-4) and SAEs, including atrial fibrillation, atrial flutter, or other types of arrhythmias, were more frequent in our cohort than trials with shorter ibrutinib exposure, but these findings are consistent with those from studies evaluating ibrutinib monotherapy, where their rate increases along with the duration of therapy.[24] In particular, grade 3-4 cardiac AEs occurred at a rate of 10.0% in SAKK 34/17 compared with 3.0% (HOVON141/VISION) and 7.5% (CLARITY).[5,11] In addition, ibrutinib-related cardiac SAEs were reported in 13.3% of patients in our study compared with 3.0% in the HOVON141/VISION trial.[11]

The discontinuation rate was 36.7%, higher than in studies investigating shorter IV combination phases (ranging from 10% to 16%).[5,11] Among patients discontinuing during the combination phase, 50.0% of them discontinued during the first year and 50.0% during the second year of IV therapy. Combination with new generation BTK inhibitors[25,26] might result in a better toxicity profile.

Exploratory translational MRD studies investigated 2 working hypotheses, based on the assumption that ctDNA may capture the anatomical heterogeneity of solid cancer clones[27,28]: (1) whether ctDNA can detect residual disease of tissue-restricted CLL clones and (2) whether ctDNA can early detect mutations of resistance during the treatment period. The addition of ctDNA-based analyses to MRD4 assessment by flow cytometry seems to improve early detection of relapses, although it has not been compared directly with MRD6 by immunoglobulin HTS.[29] In the SAKK 34/17 population, uMRD rates at the end of induction varied significantly across the different assays and compartments, with MRD6 by immunoglobulin HTS of PB outperforming not only MRD4 by flow cytometry (in both BM and PB) but also uMRD $<10^{-3}$ by CAPP-seq in plasma cfDNA. In addition, whether prolonged exposure to IV may increase the risk of selecting resistant clones harboring *BCL2* and/or *BTK* mutations, subsequently hampering treatment sequencing, was explored.[30] CAPP-seq in plasma cfDNA failed to identify resistance mutations in patients with persisting measurable ctDNA, further reassuring the possibility of rechallenge with BTK or BCL2 inhibitors–based therapies even after 24 months of treatment.[11]

Retreatment with IV has been shown to be both feasible and effective[11] and could be an option for patients who had late relapse (eg, the European Society for Medical Oncology guidelines indicate a duration of remission >36 months[31]) and lack mutations of resistance in the re-emerging CLL clone. The time-limited IV regimen, being entirely oral, is more convenient for patients than venetoclax-rituximab, while providing a comparable PFS,[32] which may be beneficial in the relapse setting.

In conclusion, our data indicate that a prolonged ibrutinib lead-in followed by a 24-month IV combination offers an acceptable benefit-risk profile in previously treated patients with CLL. This contributes to the growing evidence that a longer duration of IV therapy increases therapeutic effectiveness while minimizing the risk of acquiring resistance mutations by allowing time off treatment.

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Authorship

Contribution: D.R. was responsible for the conception and design of the study; D.D. and G.M. were responsible for the trial management; A.C., G.S., N.C., R.C., U.M., A.W., T.Z., M.G., D.H., M.A., R.B., and D.R. were responsible for the recruitment and treatment of patients; A.C., I.R., G.M., D.D., and D.R. had access to the raw data; A.C., I.R., and D.R. wrote the first draft of the manuscript and performed a central review of all clinical data; K.P. and G.S. performed the laboratory analyses; A.C., I.R., D.D., and D.R. performed the statistical analysis; all authors interpreted the data and reviewed and approved the manuscript.

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Footnotes

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The data generated and analyzed during this study are not publicly available owing to patient confidentiality and restrictions outlined by the Swiss Group for Clinical Cancer Research (SAKK) protocols. However, anonymized data may be made available on reasonable request to the corresponding author, Davide Rossi (davide.rossi@ior.usi.ch), subject to approval by SAKK and in compliance with institutional and regulatory guidelines.

The online version of this article contains a data supplement.

There is a *Blood* Commentary on this article in this issue.

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