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Noninvasive Monitoring of Mantle Cell Lymphoma by Immunoglobulin Gene Next-Generation Sequencing in a Phase 2 Study of Sequential Chemoradioimmunotherapy Followed by Autologous Stem-Cell Rescue

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Abstract

Limited information exists in mantle cell lymphoma (MCL) on the performance of next-generation sequencing–based assay of immunoglobulin gene rearrangements for minimal residual disease (MRD) assessment. Posttreatment peripheral blood samples were collected from 16 MCL patients and analyzed with the Adaptive Biotechnologies MRD assay, which identified early molecular relapse. We observed more sensitivity in the cellular versus acellular compartment.

Background: Minimal residual disease (MRD) monitoring has been used to identify early molecular relapse and predict clinical relapse in mantle cell lymphoma (MCL). Few published data exist in MCL on the performance of next-generation sequencing–based assay of immunoglobulin gene rearrangements for MRD assessment.

Patients and Methods: In a prospective clinical trial (NCT01484093) with intensive induction chemotherapy and autologous stem-cell transplantation, posttreatment peripheral blood samples were collected from 16 MCL patients and analyzed with an earlier version of the Adaptive Biotechnologies MRD assay.

Results: Of the 7 patients whose disease remained in remission, the MRD test remained negative in 5 (71%). Of the 9 patients who experienced relapse, the MRD test was positive at least 3 months

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Disclosure

The authors have stated that they have no conflict of interest.

Supplemental Data

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before relapse in 6 patients (67%) and positive at the time of relapse in 1 patient (11%). All patients with at least 2 positive MRD tests experienced relapse.

Conclusion: The next-generation sequencing–based MRD assay identified early molecular relapse, and we observed more sensitivity in the cellular (circulating leukocytes) versus acellular (plasma cell-free DNA) compartment. This observation may be due to availability of tumor target or a limitation of the assay.

Keywords

High-dose therapy; Surveillance; Minimal residual disease

Introduction

Mantle cell lymphoma (MCL) is a rare subtype of B-cell non-Hodgkin lymphoma considered incurable and associated with a continuous pattern of relapse over time. After completion of first-line therapy in MCL and subsequent complete remission, patients are then followed, typically with clinical evaluations every 3 to 6 months for the first 5 years with intermittent surveillance imaging. Although it is not performed in routine clinical practice, multiple studies have demonstrated that monitoring for minimal residual disease (MRD) after treatment in MCL can be useful in evaluating the quality of remission and in predicting clinical relapse.[1–5] In fact, surveillance MRD monitoring has been utilized to identify patients with early molecular relapse, initiate preemptive rituximab therapy to convert patients to MRD negativity, and potentially delay clinical relapse.[4,6]

The method utilized for MRD detection in most MCL clinical trials has relied on allele-specific oligonucleotide assay for real-time quantitative PCR (ASO-PCR).[2,7] Recombination of the variable (V), diversity (D), and joining (J) gene segments in the immunoglobulin heavy (IGH) and the V and J gene segments of the light chain loci result in unique DNA sequences that are markers of clonality, or clonotypes, in B-cell malignancies, including MCL. The ASO-PCR method uses consensus primers to sequence the immunoglobulin heavy chain region to develop patient-specific IGH primers utilized in subsequent PCR reactions. This method for MRD detection is time-consuming and laborious, as well as restricted to specialized laboratories, thereby limiting its wide applicability. ASO primers are also prone to nonspecific binding that leads to false-positive results. ASO primers are by definition of different sensitivity for each patient, making cross-patient comparisons problematic.[8] Finally, the ASO-PCR MRD method is only standardized in the context of an international consortium.[9]

Adaptive Biotechnologies' next-generation sequencing (NGS) of immunoglobulin rearrangements provides an alternative means for detection of MRD applicable across B- and T-cell malignancies (Adaptive Biotechnologies, Seattle, WA),[10] including MCL.[11–14] This NGS method was systematically compared to the classical ASO method in MCL with superimposable results.[15] Clonal sequences are detected from baseline tumor tissue using universal PCR primers. The clonotypic sequences are identified for each patient, and only these informative MRD markers are followed in subsequent assays. This highly sensitive assay uses PCR and NGS to identify, quantify, and specify every rearrangement

present within a given sample. It has broad clinical applicability and is currently being used in multiple prospective clinical trials in MCL, including the Eastern Cooperative Oncology Group (ECOG) Intergroup's EA4151 trial (NCT03267433), a large randomized study evaluating the necessity of autologous stem-cell rescue (ASCR) consolidation in MCL patients without MRD and who experienced complete response (CR) after induction chemotherapy, as well as the ECOG E1411 (NCT01415752) randomized phase 2 trial in older patients with previously untreated MCL.[16] However, few published data exist on the performance of this NGS-MRD platform in MCL, and little is known about the comparative sensitivity of the platform for the detection of molecular disease from plasma circulating tumor DNA (ctDNA) versus peripheral blood (PB) circulating tumor cells (CTCs) in MCL.

In this study, we aimed to determine whether molecular disease from plasma ctDNA and PB CTCs detected by NGS-MRD assay during the posttreatment surveillance period is predictive of clinical outcome in a cohort of uniformly treated MCL patients who received sequential chemoradioimmunotherapy followed by ASCR in a single-center phase 2 clinical trial.

Patients and Methods

Patients and Treatment

Patients aged 18 to 70 years with previously untreated, histologically confirmed MCL with measurable disease were enrolled (NCT01484093). Other inclusion criteria were Karnofsky performance status ≥ 70, advanced stage disease (clinical stage II with subdiaphragmatic involvement, stage III-IV), and transplant eligibility. Patients received 4 cycles of induction chemoimmunotherapy consisting of dose-intensive R-CHOP (rituximab with cyclophosphamide, doxorubicin, vincristine, prednisone) (cyclophosphamide 1000 mg/m^2) administered every 14 days. R-CHOP induction was followed by 2 cycles of R-HiDAC (rituximab with high-dose cytarabine) (3000 mg/m^2 for patients younger than 65 years and 2000 mg/m^2 for patients 65 years or older) followed by stem-cell collection. Patients with a partial response or CR (as determined by the 1999 Cheson criteria) received consolidation with radioimmunotherapy with tositumomab/iodine-131 followed by high-dose therapy with BEAM (carmustine, etoposide, cytarabine, melphalan) and ASCR. Interim positron emission tomographic (PET) scans were performed after 4 cycles of R-CHOP therapy. If positive after R-CHOP, PET was repeated after 2 cycles of R-HiDAC therapy. PET/computed tomography was performed 3 months after ASCR to determine overall response to therapy. Posttreatment surveillance included clinical evaluations and laboratory tests every 3 months for the first 3 years. A computed tomographic scan was performed every 6 months for 3 years, then annually through year 5. This study was conducted at Memorial Sloan Kettering Cancer Center (MSKCC) and approved by the institutional review board. Written informed consent was obtained for all patients before enrollment.

MRD Assessment

Research blood samples for MRD analysis were collected and processed as part of a biospecimen research protocol approved by the MSKCC institutional review board. Only patients who consented to optional research blood collection and use were included in the

MRD analysis. Pretreatment formalin-fixed, paraffinembedded (FFPE) biopsy specimens were analyzed for tumor-specific clonotypes. NGS-MRD analysis was performed by Sequenta (now Adaptive Biotechnologies, Seattle, WA) using an earlier version of the assay.[11] For patients without FFPE biopsy specimens, tumor clonotypes were determined using pretreatment PB or bone marrow (BM) aspirate samples (8 cases).

Collection of research PB samples and BM aspirate samples for MRD analysis were obtained at the same time as per protocol clinical evaluations. PB and BM aspirate samples were not uniformly collected at all time points for all patients during treatment and surveillance.

PB was collected in 2 different tubes with distinct sample processing algorithms. Blood samples were centrifuged and separated within 2 hours of blood draw. PB mononuclear cells (PBMCs), granulocytes, and plasma were isolated from whole blood using the Becton Dickinson Vacutainer cell preparation tubes (Becton Dickinson [BD], San Diego, CA). Serum was isolated from whole blood using BD serum separation tubes with serum separator gel and clot activator. Separated blood components were transferred into cryovials and immediately frozen at −80°. Biospecimens were shipped to Adaptive Biotechnologies (formerly Sequenta), San Francisco, CA. DNA was extracted using QIAamp Circulating Nucleic Acid Kit (Qiagen, Germantown, MD) from the cell-free compartment of the blood (serum and plasma), and the AllPrep kit (Qiagen) for the cellular compartment (granulocytes and PBMCs).

NGS-MRD analysis was performed by Sequenta (now Adaptive Biotechnologies), as previously described.[11] In brief, genomic DNA from FFPE tumor biopsy was amplified using universal primer sets for the IGH (VDJ and DJ) and immunoglobulin kappa (Igκ) locus. The amplified product was sequenced to determine the sequences and frequencies of the different tumor-specific clonotypes in the sample. Clonotype frequencies were calculated by dividing the number of sequencing reads for each clonotype by the total number of passed sequencing reads within a sample. A frequency threshold of 5% was used to define lymphoma immunoglobulin reporters.

MRD testing was performed at various time points in the PB, analyzing the cell-free and cellular compartments separately when specimens were available. A sample for each time point (ie, granulocytes/PBMCs and plasma/serum) was considered molecular positive for CTCs or ctDNA, respectively, if any lymphoma clonotype molecules were detected in the specimen, quantified as the number of lymphoma clonotype molecules per diploid genome sequenced. MRD testing was also performed on cellular BM aspirate samples when available. For CTCs, the range of DNA input was 0.048 to 22.72 μg. For ctDNA, the range of DNA input was 0.0012 to 1.02 μg and extraction was done on 2 mL of plasma. Raw data are provided in Supplemental Table 1 in the online version. Validation of the assay on ctDNA included reproducibility between replicates, which yielded an r^2 value of 0.94 and 0.93 at the patient and clonotype level respectively (Supplemental Figure 1A and B in the online version). Additionally, correlation between cell-free DNA and cellular DNA (r^2 = 0.66) was observed, supporting detection of ctDNA in patients with known disease burden (Supplemental Figure 1C in the online version).

Statistical Analysis

Descriptive statistics of patient baseline characteristics were reported. Patient outcomes were evaluated on an intent-to-treat basis. Progression-free survival (PFS) was defined as time from beginning of treatment until progression, relapse, death from any cause, or last follow-up visit. Overall survival (OS) was defined as time from diagnosis until death from any cause or last follow-up visit. Survival curves were estimated by the Kaplan-Meier method and survival comparison of the categorical subgroups was performed by log-rank testing. $P < .05$ was considered statistically significant. All statistical analyses were performed by SPSS 22.0 software (IBM, Armonk, NY). The analytical sensitivity of the NGS-MRD assay was estimated at detection of 1 clonal molecule in a background of 1×10^6 nonclonal molecules.

Results

Patients, Response, and Outcomes

Twenty-five patients were enrolled onto the study from February 2012 to October 2013. The data of 2 patients were excluded from final analysis because of revision of histologic diagnosis, one to diffuse large B-cell lymphoma and the other to malignant mesothelioma. Patient characteristics are shown in Table 1, and a clinical flowchart is provided in Figure 1. The median (range) age was 58 (46–69) years, with expected male predominance (70%). Most of the patients presented with Ann Arbor stage IV disease, with 78% of patients with BM involvement and 57% with gastrointestinal tract involvement (baseline endoscopy and colonoscopy were required and performed in all 23 patients). The MCL international prognostic index distribution was low in 48%, intermediate in 30%, and high in 22%. Proliferation index was low (<10%) in 3, intermediate (10%−29%) in 8, high (≥30%) in 10, and missing in 2. Four patients had blastic histology.

The overall response rate was 100% CR (n = 23) to induction chemotherapy, and all but one patient proceeded to high-dose ASCR therapy (n = 22). The survival outcomes are reported after a median follow-up of 61 months. The median PFS (n = 23) was 47 months (Figure 2A). Twelve events have occurred: 12 patients experienced disease relapse, 9 of whom are still alive and 3 of whom subsequently died of disease. Ki-67 percentage of ≥ 30 was associated with a trend toward inferior PFS ($P = .07$, Figure 2C).

MRD Detection and Outcomes

In a subset of patients (n = 17), NGS-MRD assay was used to assess for presence of MRD (Figure 3). A baseline clonotypic sequence was identified in 16 patients (94%) (patient 1 had a calibration failure). Five cases had pretreatment PB evaluated. Five of 5 had detectable disease in baseline cellular compartment samples (patients 2, 8, 9, 10, and 15) Two of 5 (patients 2 and 8) did not have ctDNA samples available. Of the 3 cases (patients 9, 10, and 15) who had baseline plasma/serum samples available, all 3 had detectable disease in baseline cell-free compartment samples. All 16 patients had PET-negative CR at end of treatment.

Patients with Disease Remission.—The disease of 7 patients remained in remission (Figure 3, patients 2–8). Two of these patients (patients 3 and 7) were MRD positive at one time point with multiple MRD-negative results at subsequent time points. Patient 3 had one MRD-positive result 6 months after ASCR from BM aspirate sample in IgH (but not Igκ receptor) at a level of < 1 lymphoma clonotype molecule per 105,147 diploid genomes. Patient 7 had a MRD-positive result at a single time point, 12 months after ASCR, from BM aspirate and PBMC samples in both IgHD and Igκ receptors at a level of < 1 lymphoma clonotype molecule per 195,869 and 1,136,582 diploid genomes, respectively (Supplemental Table 1 in the online version). Of the 7 patients whose disease remained in remission, the MRD test was negative at all time points in 5 (71%).

Patients Who Experienced Disease Relapse.—The disease of 9 patients relapsed within the study follow-up thus far (Figure 3, patients 9–17). Two of these patients (22%; patients 9 and 11) did not have PB available at or around the time of relapse. Patient 9 had negative MRD PB test results 12 months before relapse, and it is possible that at this time, the disease was below the limit of detection with the NGS-MRD assay. Patient 11 experienced relapse shortly after stem-cell rescue, and MRD testing was not performed after transplantation. Of the 9 patients who experienced relapse, the MRD test was positive at least 3 months before relapse in 6 (67%). One patient had an MRD-positive test at the time of relapse (patient 14). The likely explanation for the 2 patients who relapsed with no MRD-positive test (patients 9 and 11) is the timing of the MRD samples in relation to the relapses. For the 6 patients with positive MRD results at least 3 months in advance of relapse, the median (range) anticipation time was 8.5 (5–26) months. Of these 6 patients in whom the MRD test predicted relapse, 3 had detectable CTCs in the cellular compartment at the time of relapse but not the cell-free compartment (patients 10, 16, and 17). The remaining 4 (patients 12, 13, 14, and 15) had detectable disease in both cellular and cell-free compartments at relapse. All cases with at least 2 MRD-positive time points after treatment experienced clinical relapse. Data on BM involvement and leukemic phase disease status for this subset of 17 patients are included in Table 1.

Detection of Molecular Disease by NGS-MRD as CTCs Versus ctDNA

Sixteen patients contributed samples for MRD testing. One patient (patient 1) experienced a calibration failure, so no MRD data were available. MRD testing was performed on 36 BM mononuclear cell samples from 14 patients, 74 PBMC samples from 16 patients, 73 PB plasma samples from 16 patients, and 74 PB serum samples from 16 patients. There were 22 time points with an overlap of both BM mononuclear cell and PBMC samples available, resulting in a total of 88 time points coming from 16 patients in the cellular compartment. There were 72 time points with an overlap of both plasma and serum samples available, resulting in a total of 75 time points coming from 16 patients in the cell-free compartment (Figure 3). A total of 74 time points with both cellular and cell-free compartments from 16 patients were tested. Discordant results between the cellular and cell-free compartments occurred in 14 (19%) of 74 time points when both compartments were available for testing.

All instances of discordance occurred with CTCs detected in the cellular compartment but no ctDNA detected in the cell-free compartment from the same time point. This may be

due to availability of tumor target or a limitation of the assay. Whether this observation is a reflection of disease biology (ie, cell-free DNA was not present in these samples) or because all ctDNA may not be detectable by this assay cannot be addressed by this data set. For the 7 patients who had both cellular and cell-free compartments collected at the time of relapse, all 7 had detectable tumor-specific sequences in the cellular compartment, but only 4 (57%) had detectable disease in the cell-free compartment. Serum was isolated from serum separation tubes and plasma was isolated from cell preparation tubes. Discordant results between serum and plasma samples occurred in 2 (3%) of 72 time points when ctDNA was detected in serum but not plasma. This may suggest that plasma isolation with cell preparation tube processing results in poor preservation and/or increased degradation of ctDNA. Raw data are provided in Supplemental Table 1 in the online version.

MRD Detection Using Multiple Markers for Same Clone

Clonotypes from 2 different loci, IGH and Igκ, were identified in 8 patients. The other 8 patients had only 1 clonotype (IGH) identified. For patients with 2 clonotypes identified, both usually emerged simultaneously when the patient had molecularly detectable disease during MRD tracking. An exception was seen in only one patient at one time point, where the IGH clonotype was detected at a low level (1 lymphoma molecule in sample) whereas the Igκ clonotype was not (patient 3). Of note, the disease of this patient has remained in clinical remission. The other patient (patient 7), whose disease never relapsed, had low-level molecular positivity at 1 time point in both the IGH and Igκ clonotypes. The clonotypes used as the MRD markers for each patient are included in Supplemental Table 1 in the online version.

Discussion

In this study, we described the feasibility of an earlier version of the Adaptive Biotechnologies NGS-MRD assay in monitoring the posttreatment disease status in MCL patients; moreover, our results suggested that this tool might predict future clinical relapse during the surveillance period. We characterized the tumor-specific clonotype in most patients with FFPE tumor-associated biopsy samples (16/17%, 94%). Among patients whose disease relapsed, 6 had MRD-positive results at least 3 months before relapse (6/9%, 67%), 1 had an MRD-positive result at the time of relapse (1/9%, 11%), with a median time of anticipation of 8.5 months. The assay did not detect 2 relapses, but in these cases, PB was not obtained within 12 months before or at the same time as the relapse. MRD results after induction chemotherapy were not uniformly obtained in this pilot study; however, there did not appear to be a strong correlation between MRD results after induction or early in the post-transplantation course and subsequent outcome, which may reflect the limited sensitivity of the assay at early time points.

Timing of PB collection was variable among the subjects because MRD analysis was not a preplanned aspect of the clinical trial. Two MRD-positive time points were not associated with clinical relapse. Similar to the published experience with the NGS-MRD assay for surveillance in diffuse large B-cell lymphoma (DLBCL), low-level positive tests were identified in rare patients whose disease remained in remission and was not confirmed on

repeat testing at the same time point, highlighting the need to monitor MRD over time.[17] A low-level positive result could be due to a small amount of residual disease at or near the limit of detection of the assay, which may not be reproduced as a result of sampling. As a corollary, in our series, we found all patients who had 2 positive MRD tests at distinct time points were destined to have relapsing disease.

There are few published data on the performance of this NGS-MRD assay during the surveillance period in a cohort of uniformly treated MCL patients in a prospective clinical trial.[12–14,16,18,19] Other MCL studies utilizing the NGS-MRD assay for MRD assessment suggest that interim MRD status after 1 to 3 cycles of chemoimmunotherapy may be an important biomarker and correlate with PFS.[13,14,16] However, these studies do not examine the performance of the assay during the posttreatment surveillance period. A phase 2 study of rituximab/bendamustine and rituximab/cytarabine followed by autologous stemcell transplantation (ASCT) consolidation in untreated MCL patients from Dana-Farber Cancer Institute (DFCI) and Washington University in St Louis reported on the outcome of MRD testing during surveillance after ASCT. MRD assessment was performed in 20 of 88 patients using the NGS-MRD assay in PBMCs and plasma at baseline (n = 12), interim (n = 11), and end of induction therapy (n = 12), as well as various time points after ASCT (n = 112). In this study, MRD was detected in 1 (92%) of 12 samples at the end of induction and did not correlate with PFS. Among the 17 patients followed during post-ASCT surveillance, there were 2 relapses, 1 of which was detected 7.2 months before clinical relapse in both PBMCs and plasma using MRD assay. The other relapse was not detected with the NGS-MRD assay; the most proximate blood sample to the relapse had been collected 11 months before. There was a high negative predictive value with all patients with undetectable MRD who were in ongoing remission after ASCT.[12,19] Our anticipation data and the DFCI study demonstrate that this version of the NGS-MRD assay is unlikely to detect molecular relapse 10 to 12 months before clinical relapse, so MRD testing every 3 to 6 months during surveillance is recommended.

Contrary to previous findings in DLBCL,[20,21] we report 100% sensitivity in detecting circulating cells but only a 57% sensitivity detecting plasma ctDNA as assessed by ultimate clinical outcome. This observation may reflect disease biology or may be due to limitations in assessing ctDNA using PCR. Unfortunately, these data do not speak to which. Kurtz et al[21] reported higher sensitivity for detection of molecular disease in the plasma versus circulating cells in DLBCL. This contrast in test performance across compartments may reflect differences in the disease biology and clinical presentation of MCL versus DLBCL because MCL patients more commonly have BM involvement and CTCs in the PB. Recent data from chronic lymphocytic leukemia, a disease with biologic similarities to MCL, also demonstrated that disease is more readily detected in the marrow and PBMCs versus plasma.[22]

While differences in disease detection across compartments may reflect a fundamental difference in the disease biology, it is also possible that preanalytic variables as well as stability of ctDNA affect the results. For example, higher rates of ctDNA degradation might occur when serum is collected in serum separation tubes, as in our study, compared with more modern techniques, such as use of specialized ctDNA collection tubes (ie, Streck

Cell-Free DNA BCT) that contain unique preservatives preventing the release of genomic DNA and allow for isolation of high-quality cell-free DNA.[23] These specialized collection tubes were not available at the time we conducted this study. In an unexpected fashion, the NGS-MRD assay showed more sensitivity in serum than plasma. We would have assumed the sensitivity would have been the same. Hence, we infer that variations in the processing and stability of cell-free DNA across these tubes affected the outcome because the ctDNA content should be the same in serum and plasma at any given time point. DNA quality assessment for ctDNA was not performed. We acknowledge this as a limitation of the study, as they were not available at the time.

Conclusion

The NGS-MRD assay may be a useful tool for monitoring the posttreatment disease status in MCL during the surveillance period. We observed that the assay was more sensitive in detecting CTCs among PBMCs than ctDNA in serum or plasma. However, this data set cannot address whether this observation is due to availability of tumor target or is a limitation of the assay. Although ours is a small study with variable time points for PB collection, it is among the first reports of the NGS-MRD assessment tool in the surveillance context, and the preliminary findings from this study may help to inform its use in future prospective MCL studies. This pilot study provides insights into the sensitivity of the assay across compartments, the need for confirmation of MRD-positive results with more than 1 test, the anticipation time of the assay, and the frequency of MRD testing that is required during surveillance. Although more extensive characterization is required in future studies, including interlaboratory standardization, we hypothesize that the NGS-MRD assessment can identify patients who will ultimately experience relapse and may facilitate early clinical intervention at the time of molecular relapse to improve outcomes in MCL.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Clinical Practice Points

- Minimal residual disease (MRD) monitoring has been used to identify early molecular relapse and predict clinical relapse in mantle cell lymphoma (MCL). Few published data exist in MCL on the performance of next-generation sequencing (NGS)-based assay of immunoglobulin gene rearrangements for MRD assessment.
- In a prospective clinical trial (NCT01484093) with intensive induction chemotherapy and autologous stem-cell transplantation, posttreatment peripheral blood samples were collected from 16 MCL patients and analyzed with an earlier version of the Adaptive Biotechnologies MRD assay.
- Of the 7 patients whose disease remained in remission, the MRD test remained negative in 5 (71%). Of the 9 patients whose disease relapsed, the MRD test was positive at least 3 months before relapse in 6 patients (67%) and positive at the time of relapse in 1 patient (11%). The disease of all patients with at least 2 positive MRD tests relapsed.
- The NGS-MRD assay identified early molecular relapse, and we observed more sensitivity in the cellular (circulating leukocytes) versus acellular (plasma cell-free DNA) compartment. This observation may be due to availability of tumor target or a limitation of the assay.

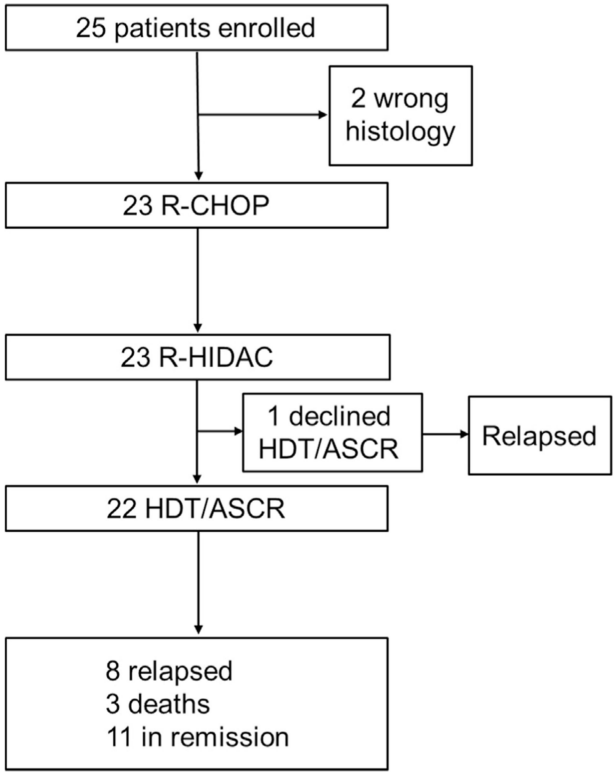

Figure 1.
Clinical Flowchart

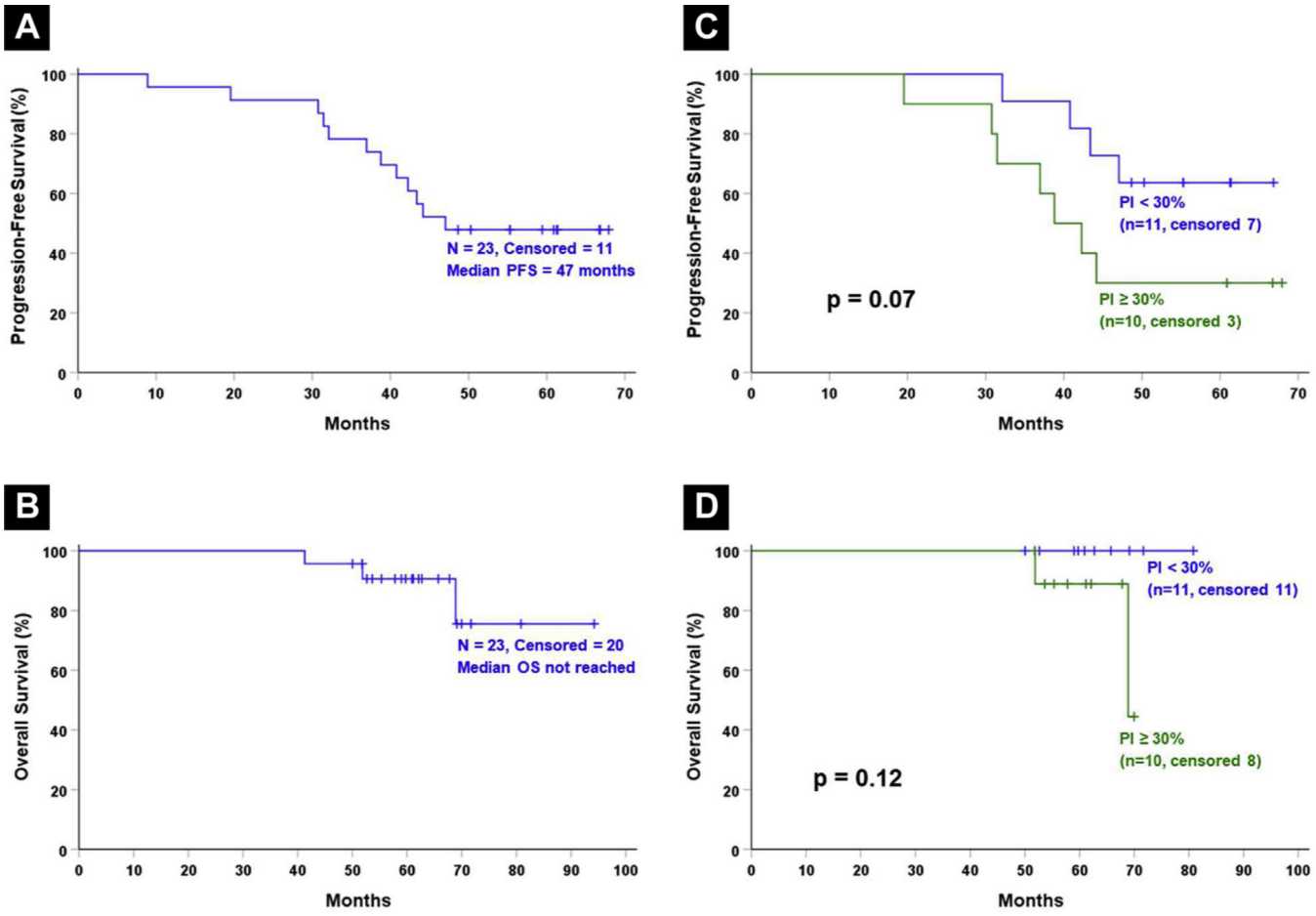

Figure 2. Progression-Free Survival

(A) and Overall Survival (B) and According to Proliferative Index (PI) Higher or Lower Than 30% (C, D) in Intent-to-Treat Population in 23 Patients

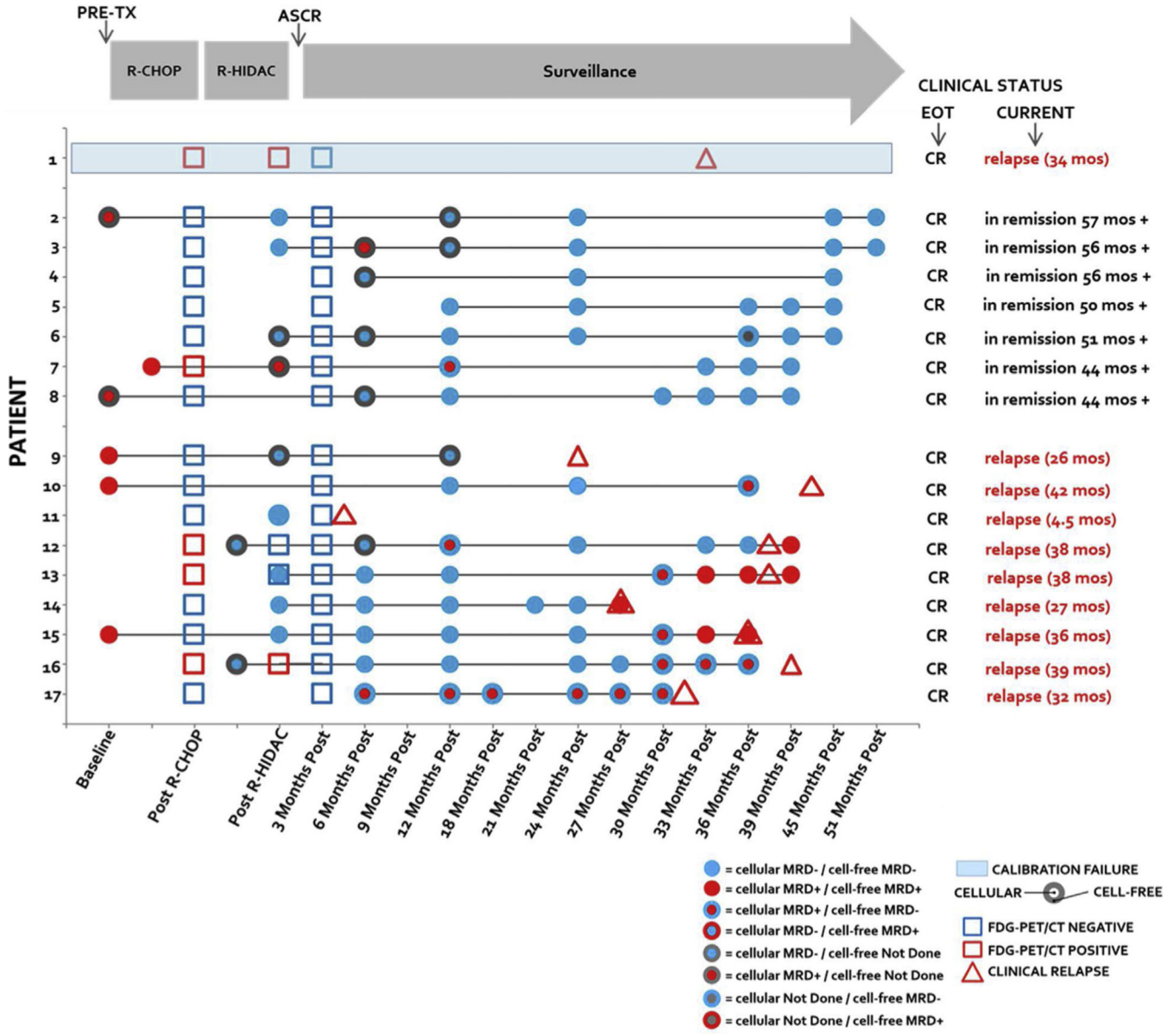

Figure 3. MRD Detection Assessed by Adaptive's NGS-MRD Assay. Cell-free (Plasma and Serum) and Cellular (PBMC) Compartments Were Analyzed Separately

Abbreviations: MRD = minimal residual disease; NGS = next-generation sequencing; PBMC = peripheral blood mononuclear cell.

Table 1

Characteristics of 23 Patients

Characteristic	Value
Age (years), median (range)	58 (46–69)
Gender	
Male	16 (70)
Female	7 (30)
LDH greater than ULN	9 (39)
Ann Arbor stage	
I/II	0
III/IV	23 (100)
Bone marrow involvement	18 (78)
Leukemic phase disease (ALC > 510^9 cells/L)	4 (17)
GI tract involvement (EGD and colonoscopy required)	13 (57)
Other (bone, base of tongue, blood, skin)	5 (22)
Ki-67–positive cells (n = 21)	
<30%	11 (52)
≥30%	10 (48)
MIPI	
Low	11 (48)
Intermediate	7 (30)
High	5 (22)
KPS	
≥80	22 (96)
<80	1 (4)

Data are presented as n (%) unless otherwise indicated.

Abbreviations: ALC = absolute lymphocyte count; EGD = esophagogastroduodenoscopy; GI = gastrointestinal; LDH = lactate dehydrogenase; MIPI = mantle cell lymphoma international prognostic index; KPS = Karnofsky performance status; ULN = upper limit of normal.