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2021 L Street NW, Suite 900,
Washington, DC 20036
Phone: 202-776-0544 | Fax 202-776-0545
bloodadvances@hematology.org

Biomarker analysis of the ASPEN study comparing zanubrutinib to ibrutinib in patients with Waldenström Macroglobulinemia

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Constantine Tam (Alfred Health and Monash University, Australia) Stephen Opat (Monash Medical Centre, Australia) Shirley D'Sa (University College London Hospitals NHS Foundation Trust, United Kingdom) Wojciech Jurczak (Maria Sklodowska-Curie National Research Institute of Oncology, Poland) Hui-Peng Lee (Flinders Medical Centre, Australia) Gavin Cull (Sir Charles Gairdiner Hospital, Australia) Roger Owen (The Leeds Teaching Hospitals NHS Trust, United Kingdom) Paula Marlton (Princess Alexandra Hospital, Australia) Björn Wahlin (Karolinska Institutet, Sweden) Ramon Garcia-Sanz (University Hospital of Salamanca, Spain) Helen McCarthy (Royal Bournemouth and Christchurch Hospital, United Kingdom) Stephen Mulligan (Royal North Shore Hospital, Australia) Alessandra Tedeschi (ASST Grande Ospedale Metropolitano Niguarda, Niguarda Cancer Center, Italy) Jorge Castillo (Dana-Farber Cancer Institute, United States) Jaroslaw Czyz (Szpital im Jana Biziela Bydgoszcz, Poland) Carlos Fernández de Larrea (Hospital Clínic de Barcelona, Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS), Universitat de Barcelona, Spain) David Belada (University Hospital and Medical School, Hradec Kralove, Czech Republic) Edward Libby (Fred Hutchinson Cancer Center, United States) Jeffrey Matous (Colorado Blood Cancer Institute, United States) Marina Motta (ASST Spedali Civili, Italy) Tanya Siddiqi (City of Hope Medical Center, United States) Monica Tani (Ospedale Civile S.Maria delle Croci, Italy) Marek Trněný (Charles University General Hospital, Czech Republic) Monique Minnema (UMC Utrecht, Netherlands) Christian Buske (Institute of Experimental Cancer Research, Germany) Véronique Leblond (Hopital Pitie-Salpetriere, France) Steven Treon (Dana Farber Cancer Institute, United States) Judith Trotman (Concord Repatriation General Hospital and University of Sydney, United Kingdom) Binghao Wu (BeiGene (Shanghai) Co., Ltd., China) Yiling Yu (BeiGene (Shanghai) Co., Ltd., China) Zhirong Shen (BeiGene, China) Wai Chan (BeiGene USA, Inc., United States) Jingjing Schneider (BeiGene USA, Inc., United States) Heather Allewelt (BeiGene USA, Ltd., United States) Aileen Cohen (BeiGene USA, Inc., United States) Meletios-Athanasios Dimopoulos (National and Kapodistrian University of Athens, Greece)

Abstract:
The phase 3 ASPEN trial (NCT03053440) compared Bruton tyrosine kinase inhibitors (BTKi), zanubrutinib and ibrutinib, in patients with Waldenström macroglobulinemia (WM). Post-hoc biomarker analysis was performed using next-generation sequencing on pretreatment bone marrow samples from 98 zanubrutinib-treated and 92 ibrutinib-treated patients with mutated (MUT) MYD88 and 20 zanubrutinib-treated patients with wild-type (WT) MYD88. Of 329 mutations in 52 genes, mutations in CXCR4 (25.7%), TP53 (24.8%), ARID1A (15.7%), and TERT (9.0%) were most common. TP53MUT, ARID1AMUT, and TERTMUT were associated with higher rates of CXCR4MUT ($P<0.05$). Patients with CXCR4MUT (frameshift [FS] or nonsense [NS] mutations) had lower very good partial response and complete response rate (VGPR+CR) (17.0% vs 37.2%, $P=0.020$) and longer time to response (11.1 vs 8.4 months) than CXCR4WT BTKi-treated patients. CXCR4NS was associated with inferior progression-free survival (PFS; HR=3.39, $P=0.017$) in ibrutinib-treated, but not zanubrutinib-treated patients (HR=0.67, $P=0.598$), but VGPR+CR rates were similar between treatment groups (14.3% vs 15.4%). Compared to ibrutinib, zanubrutinib-treated patients with CXCR4NS had a favorable major response rate (MRR; 85.7% vs 53.8%, $P=0.09$) and PFS (HR=0.30, $P=0.093$). In patients with TP53MUT, significantly lower MRR was observed in ibrutinib- (63.6% vs 85.7%, $P=0.04$) but not zanubrutinib-treated patients (80.8% vs 81.9%, $P=0.978$). In TP53MUT, compared to ibrutinib, zanubrutinib-treated patients had higher VGPR+CR (34.6% vs 13.6%, $P<0.05$), numerically improved MRR (80.8% vs 63.6%, $P=0.11$), and longer PFS (not reached vs 44.2 months, HR=0.66, $P=0.37$). Collectively, WM patients with CXCR4MUT or TP53MUT had worse prognosis compared to patients with WT alleles and zanubrutinib led to better clinical outcomes.

Conflict of interest: COI declared - see note

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Biomarker analysis of the ASPEN study comparing zanubrutinib to ibrutinib in patients with Waldenström macroglobulinemia

Running head: Genomic landscape of Waldenström macroglobulinemia

Constantine S. Tam,[1] Stephen Opat,[2] Shirley D'Sa,[3] Wojciech Jurczak,[4] Hui-Peng Lee,[5] Gavin Cull,[6] Roger G. Owen,[7] Paula Marlton,[8] Bjorn E. Wahlin,[9] Ramón García-Sanz,[10] Helen McCarthy,[11] Stephen Mulligan,[12] Alessandra Tedeschi,[13] Jorge J. Castillo,[14] Jaroslaw Czyż,[15] Carlos Fernández De Larrea Rodriguez,[16] David Belada,[17] Edward Libby,[18] Jeffrey Matous,[19] Marina Motta,[20] Tanya Siddiqi,[21] Monica Tani,[22] Marek Trněný,[23] Monique C. Minnema,[24] Christian Buske,[25] Veronique Leblond,[26] Steven P. Treon,[14] Judith Trotman,[27] Binghao Wu,[28] Yiling Yu,[28] Zhirong Shen,[28] Wai Y. Chan,[28] Jingjing Schneider,[28] Heather Allewelt,[28] Aileen Cohen,[28] and Meletios Dimopoulos[29]

[1]Alfred Health and Monash University, Melbourne, Victoria, Australia; [2]Monash Health and Monash University, Clayton, Victoria, Australia; [3]Centre for Waldenström's Macroglobulinemia and Associated Disorders, University College London Hospital Foundation Trust, London, United Kingdom; [4]Maria Sklodowska-Curie National Institute of Oncology, Krakow, Poland; [5]Flinders Medical Centre, Adelaide, SA, Australia; [6]Sir Charles Gairdner Hospital, University of Western Australia, Perth, WA, Australia; [7]St. James University Hospital, Leeds, United Kingdom; [8]Princess Alexandra Hospital, and University of Queensland, Brisbane, Queensland, Australia; [9]Karolinska Universitetssjukhuset and Karolinska Institutet, Stockholm, Sweden; [10]Hospital Universitario de Salamanca, Salamanca, Spain; [11]Royal Bournemouth and Christchurch Hospital, Bournemouth, United Kingdom; [12]Royal North Shore Hospital, Sydney, New South Wales, Australia; [13]ASST Grande Ospedale Metropolitano Niguarda, Milan, Italy; [14]Dana-Farber Cancer Institute, Boston, MA, USA; [15]Collegium Medicum in Bydgoszcz, Nicolaus Copernicus University in Toruń, Bydgoszcz, Poland; [16]Hospital Clínic de Barcelona, Barcelona, Spain; [17]Department of Internal Medicine – Haematology, University Hospital and

Faculty of Medicine, Hradec Králové, Czech Republic; [18]Fred Hutchinson Cancer Center, Seattle, WA, USA; [19]Colorado Blood Cancer Institute, Denver, Colorado, USA; [20]AO Spedali Civili di Brescia, Lombardia, Italy; [21]City of Hope National Medical Center, Duarte, CA, USA; [22]Ospedale Civile Santa Maria delle Croci, AUSL Ravenna, Italy; [23]Všeobecná fakultní nemocnice v Praze, Prague, Czechia; [24]University Medical Center Utrecht, Utrecht, Netherlands; [25]CCC Ulm - Universitätsklinikum Ulm, Ulm, Baden-Württemberg, Germany; [26]Sorbonne University, Pitié Salpêtrière Hospital, Paris, France; [27]Concord Repatriation General Hospital, Sydney, New South Wales, Australia; [28]BeiGene USA, Inc., San Mateo, CA, USA, and BeiGene (Shanghai) Co., Ltd., Shanghai, China; and [29]National and Kapodistrian University of Athens, Athens, Greece

Corresponding author:

Name: Constantine S. Tam, MBBS, MD

Address: The Alfred Hospital

55 Commercial Road

Melbourne VIC, Australia 3004

Email: constantine.tam@alfred.org.au

Phone: 613 9903 0122

Fax: 613 9903 0228

Data sharing statement

All authors had access to the original data for the analyses described here. On request, and subject to certain criteria, conditions, and exceptions, BeiGene, Ltd will provide access to individual deidentified participant data from BeiGene-sponsored global interventional clinical studies conducted for medicines (1) for indications that have been approved or (2) in programs that have been terminated. BeiGene will also consider requests for the protocol, data dictionary, and statistical analysis plan. Data requests may be submitted to DataDisclosure@beigene.com.

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KEY POINTS

- Patients with Waldenström macroglobulinemia and mutations in *CXCR4* or *TP53* had poorer prognosis after treatment with BTK inhibitors.
- Patients with *CXCR4* or *TP53* mutations had more favorable outcomes when treated with zanubrutinib versus ibrutinib.

ABSTRACT

The phase 3 ASPEN trial (NCT03053440) compared Bruton tyrosine kinase inhibitors (BTKi), zanubrutinib and ibrutinib, in patients with Waldenström macroglobulinemia (WM). Post-hoc biomarker analysis was performed using next-generation sequencing on pretreatment bone marrow samples from 98 zanubrutinib-treated and 92 ibrutinib-treated patients with mutated (MUT) *MYD88* and 20 zanubrutinib-treated patients with wild-type (WT) *MYD88*. Of 329 mutations in 52 genes, mutations in *CXCR4* (25.7%), *TP53* (24.8%), *ARID1A* (15.7%), and *TERT* (9.0%) were most common. $TP53^{MUT}$, $ARID1A^{MUT}$, and $TERT^{MUT}$ were associated with higher rates of $CXCR4^{MUT}$ (P<0.05).

Patients with $CXCR4^{MUT}$ (frameshift [FS] or nonsense [NS] mutations) had lower very good partial response and complete response rate (VGPR+CR) (17.0% vs 37.2%, P=0.020) and longer time to response (11.1 vs 8.4 months) than $CXCR4^{WT}$ BTKi-treated patients. $CXCR4^{NS}$ was associated with inferior progression-free survival (PFS; HR=3.39, P=0.017) in ibrutinib-treated, but not zanubrutinib-treated patients (HR=0.67, P=0.598), but VGPR+CR rates were similar between treatment groups (14.3% vs 15.4%). Compared to ibrutinib, zanubrutinib-treated patients with $CXCR4^{NS}$ had a favorable major response rate (MRR; 85.7% vs 53.8%, P=0.09) and PFS (HR=0.30, P=0.093). In patients with $TP53^{MUT}$, significantly lower MRR was observed in ibrutinib- (63.6% vs 85.7%, P=0.04) but not zanubrutinib-treated patients (80.8% vs 81.9%, P=0.978). In $TP53^{MUT}$, compared to ibrutinib, zanubrutinib-treated patients had higher VGPR+CR (34.6% vs 13.6%, P<0.05), numerically improved MRR (80.8% vs 63.6%, P=0.11), and longer PFS (not reached vs 44.2 months, HR=0.66, P=0.37).

Collectively, WM patients with $CXCR4^{MUT}$ or $TP53^{MUT}$ had worse prognosis compared to patients with WT alleles and zanubrutinib led to better clinical outcomes.

INTRODUCTION

Waldenström macroglobulinemia (WM) is a B-cell malignancy characterized by lymphoplasmacytic bone marrow infiltration of monoclonal immunoglobulin M–secreting cells that constitutively activate the B-cell receptor (BCR) signaling complex.[1] A critical component of the BCR signaling pathway is Bruton tyrosine kinase (BTK), an important regulator of B-cell proliferation and survival.[2] *Myeloid differentiation factor 88* (*MYD88*) and *C-X-C chemokine receptor type 4* (*CXCR4*) are the most frequently mutated genes in patients with WM.[3] A point mutation in *MYD88* that switches leucine to proline at amino acid position 265 ($MYD88^{L265P}$) is present in more than 90% of patients, and this activating mutation triggers tumor cell growth via BTK signaling.[4] Mutations in *CXCR4* ($CXCR4^{WHIM}$) that are similar to germline mutations detected in patients with warts, hypogammaglobulinemia, infection, and myelokathexis syndrome–like symptoms have been found in up to 40% of WM patients and are thought to promote cell survival signaling and confer ibrutinib resistance.[5] The mutational status of *MYD88* and *CXCR4* impacts the efficacy of BTK inhibitors (BTKi) in patients with WM. Patients with wild-type (WT) *MYD88* had lower major response rate (MRR) and shorter overall survival (OS) than patients with $MYD88^{L265P}$ when treated with ibrutinib.[6,7] Ibrutinib-treated patients with $CXCR4^{WHIM}$ mutations had lower response rates and shorter progression-free survival (PFS) compared to WT, and patients with nonsense (NS) mutations in this gene had lower rates of major response and worse PFS than patients with frameshift (FS) mutations.[8] Additionally, mutations or deletions, often in the DNA binding domain, of *tumor protein P53* (*TP53*) have been reported in <14% of patients with WM[3,9-11] and are associated with worse survival outcomes.[9,10]

BTKi have led to substantial improvements in outcomes for patients with WM, reflected by the approval of ibrutinib and zanubrutinib by the FDA. The ASPEN trial is a phase 3 randomized, open-label, multicenter study comparing zanubrutinib to ibrutinib treatment in patients with

WM.[12] While the primary efficacy endpoint was not met, the study's long-term follow-up confirmed patients who received zanubrutinib treatment had a higher very good partial response and complete response (VGPR+CR) rate over time (zanubrutinib, 36% vs ibrutinib, 25%; descriptive P=0.07) and lower rates of atrial fibrillation, diarrhea, hypertension, localized infection, hemorrhage, muscle spasms, pneumonia, and adverse events leading to discontinuation or death compared to ibrutinib. *MYD88*^{WT} patients treated with zanubrutinib also demonstrated major responses with MRR of 63% and 65.4% in two separate studies[13] compared to none reported in patients treated with ibrutinib.[14]

While BTKi are effective therapies for many patients, not all patients may have clinical benefit. Many patients with WM and disease progression during covalent BTKi treatment acquire mutations in *BTK* at the covalent BTKi binding site (*BTK*^{CYS481}) or its downstream mediator, *PLCG2*.[15] These mutations lead to reactivation of downstream signaling pathways and cytokine release resulting in resistant *BTK*^{WT} cells.[16] In addition to resistance mutations in *BTK* and *PLCG2*, other genetic alterations that may be associated with *BTK* include deletions on chromosomes 6q and 8p, which disrupt *BTK*, *MYD88/NF-kB*, and apoptotic signaling, as well as recurring mutations in ubiquitin ligases, innate immune signaling, and *TLR/MYD88* pathway regulators.[17,18]

The objectives of this post-hoc biomarker analysis were 1) to evaluate baseline genetic alterations in WM patients enrolled in the ASPEN study and their association with the efficacy of ibrutinib and zanubrutinib treatment and 2) to explore acquired mutations potentially conferring resistance to BTKi.

MATERIALS AND METHODS

Patients

The phase 3 ASPEN study (NCT03053440) enrolled patients with *MYD88*^{MUT} WM (confirmed by qualitative *MYD88* allele-specific PCR with 0.2%-0.5% limit of detection[19]) to compare responses to zanubrutinib (n=102) versus ibrutinib (n=99). Twenty-eight patients without *MYD88* mutations (*MYD88*^{WT}) were enrolled in a separate cohort 2. Details of the study design have been described previously.[19,12] One hundred ninety patients with *MYD88*^{MUT} (zanubrutinib, 98; ibrutinib, 92) and 20 patients with *MYD88*^{WT} provided residual DNA from pretreatment bone marrow aspirate (BMA) samples without B-cell enrichment for baseline genetic alterations testing. In addition, five patients with disease progression after zanubrutinib treatment (three *MYD88*^{MUT} and two *MYD88*^{WT}) gave informed consent to provide BMA samples at the time of clinical progression to explore resistance mechanisms. The trial was approved by the Institutional Review Board or Independent Ethics Committee at each study site and conducted in accordance with applicable regulatory requirements, the principles of the Declaration of Helsinki, and Good Clinical Practice guidelines of the International Conference on Harmonization. All patients provided written informed consent.

Biomarker assessments

MYD88 mutational status for patients' enrollment was assessed by a qualitative allele-specific PCR with 0.2%-0.5% limit of detection (LOD) as previously described.[19] To detect other baseline genetic alterations, next-generation sequencing (NGS) was performed using the PredicineCARE panel, a Clinical Laboratory Improvement Amendments–certified NGS assay which was validated to have a high sensitivity (LOD=0.1%~0.25%), representing 152 genes. To examine mutations that may be associated with resistance to zanubrutinib, sequencing was performed using the PredicineHeme panel containing 106 hematologic malignancy-related genes, including whole exons of *BTK* and *PLCG2*, validated to have 0.1%~0.25% assay sensitivity. A variant was considered a true mutation only when 1) at least three distinct fragments (at least one of them double-stranded) contained the mutation and 2) the variant allele frequency (VAF) was ≥0.25% and hotspot variants with allele frequency ≥0.1%. Mutations annotated as benign, likely benign, or common germline variants were filtered out based on OncoKB,[20] 1000 genomes,[21] ExAC,[22] gnomAD,[23] and KAVIAR[24] with population allele frequency >0.5%. Finally, the hematopoietic expansion-related variants including those in *DNMT3A*, *ASXL1*, *TET2*, specific alterations within *ATM* (residue 3008), *GNAS* (residue 201, 202), or *JAK* (residue 617) were excluded. The clinical efficacy endpoints used for biomarker analysis included: response rate (VGPR+CR, major response), time to response, and PFS assessed by investigator according to response criteria in the NCCN WM guidelines and modified Owen criteria[25] as of data cutoff date October 31, 2021.

To evaluate the dosage effect of *TP53* mutations, patients were classified into four subgroups based on $TP53^{MUT}$ VAF and deletion status: 1) $TP53^{WT}$ (mutation and deletion not detected), 2) $TP53^{MUT}$ with VAF between 0.25% and <1%, 3) $TP53^{MUT}$ with VAF between 1% and <10%, and 4) $TP53^{MUT}$ with VAF ≥10% or deletion present. For patients with multiple *TP53* mutations, the variant with the maximum VAF was used for classification. The clinical efficacy endpoints were compared among subgroups.

Statistical analysis

Fisher's exact tests were used to evaluate the correlations among different mutations and between mutation status and treatment status (treatment naïve vs relapsed/refractory) or treatment arms. Univariate and multivariate logistic regression models were used to compare the response rates between the mutation status. Covariates in the multivariate logistic regression models include mutation status of *CXCR4* (WT, NS, FS or WT, MUT), *TP53* (WT, MUT), *TERT* (WT, MUT), and the treatment arm to account for the treatment differences. Odds ratios (MUT vs WT) and *P* values were estimated.

The median PFS was analyzed using the Kaplan-Meier method. Hazard ratio and *P* values of PFS between mutation status (MUT vs WT) were estimated using a Cox regression model with treatment arm, *CXCR4*, *TP53*, and *TERT* mutation status as covariates. *P* values ≤0.05 were considered statistically significant without multiplicity adjustment.

RESULTS

Baseline genetic alteration profiling from 210 WM patients treated with BTKi

Across 210 WM patients, including 190 patients with $MYD88^{MUT}$ (zanubrutinib, 98; ibrutinib, 92) and 20 patients with $MYD88^{WT}$ (all zanubrutinib), baseline genetic profiling revealed 329 genetic alterations in 52 genes from 124 patients that are enriched in genes involved in DNA damage response, cell cycle, chromatin remodeling, and kinase pathways (**Table S1**). In 86 patients, genetic alterations were not observed, and the percentage of bone marrow infiltrated CD20+ cells in these patients was not different from patients with detectable genetic alterations. While $MYD88^{L265P}$ mutations were the most common, mutations in *CXCR4*, *TP53*, *ARID1A*, and *TERT* were also observed, with mutation rates of 25.7%, 24.8%, 15.7%, and 9.0%, respectively. Mutation rates of the remaining 48 genes were ≤4.3% (**Figure 1, Table S2**). Post-hoc analysis revealed FS and NS mutations were most common in *CXCR4* and patients in the zanubrutinib arm had a higher rate of $CXCR4^{MUT}$ than patients in the ibrutinib arm (33.7% vs 21.7%, *P*=0.08). Specifically, $CXCR4^{FS}$ mutations were more prevalent in patients in the zanubrutinib arm compared to the ibrutinib arm of the study (19.4% vs 7.6%, *P*=0.02; **Table S4**). FS and NS mutations were also common in patients with $ARID1A^{MUT}$; $ARID1A^{MUT}$ were more often detected in patients with $CXCR4^{MUT}$ compared to $CXCR4^{WT}$ (44.4% vs 5.8%, *P*<0.001; **Table S3**). Mutations in *TP53* consisted of missense, truncation, and splice site mutations with the majority localized in the DNA binding domain. *TP53* mutations were more common in patients with $CXCR4^{MUT}$ compared to patients with $CXCR4^{WT}$ (35.2% vs 21.2%, *P*<0.05) (**Table S3**). *TERT* mutations were localized to promoter regions including -57A>C, -124C>T, and -146C>T, and were detected more frequently in patients with *CXCR4*^{MUT} compared to *CXCR4*^{WT} (24.1% vs 3.9%, *P*<0.001) (**Table S3**).

Response to BTKi therapy was inferior in patients with mutations in CXCR4 and TP53

To evaluate the association between genetic alterations and clinical efficacy of the BTKi zanubrutinib and ibrutinib in *MYD88*^{MUT} WM, a pooled analysis of patients with *MYD88*^{MUT} (N=190) was conducted. Lower rates of VGPR+CR and longer times to response were generally observed in patients with the common mutations compared to patients with respective WT alleles. Univariate analysis revealed that patients with *CXCR4*^{MUT} and *TERT*^{MUT} had significantly lower VGPR+CR and major response, respectively (**Table 1**) compared to patients with WT alleles; *CXCR4*^{MUT} and *TERT*^{MUT} co-occurred in 13 patients (**Table S3**). In patients with *CXCR4*^{MUT}, *TP53*^{MUT}, and *TERT*^{MUT}, a dosage-dependent effect on PFS was observed; patients harboring mutations in these genes with VAF ≥1% trended toward less favorable outcomes than patients with the respective WT alleles (**Figure S1**). Because *TP53*^{MUT} and *TERT*^{MUT} were associated with higher rates of *CXCR4*^{MUT}, we performed multivariate analyses including *CXCR4*, *TP53*, and *TERT* mutational status and treatment as covariates (**Table S3**). Compared to patients with WT alleles, patients with *CXCR4*^{MUT} had significantly lower VGPR+CR rates (17.0% vs 37.2%, *P*=0.020; **Table 1**) and patients with *TP53*^{MUT} had worse PFS (*P*=0.008; **Figure 2**).

Of patients with *MYD88*^{MUT} WM, 11.6% (22/190) had *TP53*^{MUT} at VAF <1% while 13.7% (26/190) of these patients had *TP53*^{MUT} at VAF ≥1% or had a deletion of *TP53*. Patients with *TP53*^{MUT} at VAF ≥1% or *TP53* deletion had higher rates of *CXCR4*^{NS} (**Table S5**). A dosage effect of *TP53* mutations in patients treated by both zanubrutinib and ibrutinib was observed. Compared to patients with *TP53*^{WT}, ibrutinib-treated patients with *TP53*^{MUT} VAF <1% had lower rates of VGPR or better and major response, but patients with *TP53*^{MUT} VAF ≥1% or deletion had much lower rates of response and 42-month PFS (**Table S6, Figure S3**). Similarly, a lower 42-month PFS rate was observed in zanubrutinib-treated patients with *TP53*^{MUT} VAF <1%, and PFS was even lower in patients with *TP53*^{MUT} VAF ≥1% or deletions (**Table S6, Figure S3**). Because patients with *TP53*^{MUT} VAF ≥1% or deletion had higher rates of *CXCR4*^{NS} and trended toward less favorable outcomes, a 1% VAF cutoff was used to assess *TP53*^{MUT} associations with PFS. In both BTKi treatment groups, PFS in this population was inferior compared to patients with *TP53*^{WT} or *TP53*^{MUT} VAF <1% (ibrutinib: HR=3.792, *P*=0.008; zanubrutinib: HR=2.239, *P*=0.140; **Figure S3**).

TERT^{MUT} were detected in 19 patients (zanubrutinib: n=10; ibrutinib: n=9). Patients with *TERT*^{MUT}, especially those with PFS events, had high rates of *CXCR4* or *TP53* co-mutations (**Table S7**) and less favorable PFS compared to patients with *TERT*^{WT} (**Table 1**, **Table 2**).

Among the 20 patients with *MYD88*^{WT}, four patients with *TP53*^{MUT} had a lower MRR (50%) and none achieved VGPR or CR, compared with *TP53*^{WT} patients who had 62.5% MRR and a 25% VGPR+CR rate (**Table 3**). Generally, patients with *TP53*^{MUT} tended to have less favorable PFS than *TP53*^{WT} patients; the 12-month PFS for *TP53*^{MUT} versus *TP53*^{WT} was 25% versus 75%, respectively (**Figure S4**).

Zanubrutinib demonstrated deeper and faster responses, as well as favorable PFS, compared with ibrutinib in patients with CXCR4^{NS}, CXCR4^{FS}, or TP53^{MUT}

To evaluate the response to different BTKi in *MYD88*^{MUT} patients with *CXCR4*^{MUT}, *TP53*^{MUT}, and *TERT*^{MUT}, multivariate analysis was conducted in zanubrutinib- and ibrutinib-treated patients separately. As has been previously reported, patients in the ASPEN study with *CXCR4*^{MUT} had a lower VGPR+CR rate, lower MRR, and longer time to response than *CXCR4*^{WT} patients, independent of treatment. However, patients with *CXCR4*^{MUT} treated with zanubrutinib demonstrated deeper and faster responses, as well as more favorable PFS compared with ibrutinib-treated patients.

To address whether *CXCR4* mutation type affected the patients' response to BTKi, we compared treatment responses in patients with $CXCR4^{NS}$ versus $CXCR4^{FS}$. Patients with either $CXCR4^{NS}$ or $CXCR4^{FS}$ had lower VGPR+CR response rates than $CXCR4^{WT}$ patients regardless of treatment, but zanubrutinib-treated patients trended toward a higher VGPR+CR rate than ibrutinib-treated patients with $CXCR4^{FS}$ and $CXCR4^{WT}$ (**Table 2**). Further, relative to patients with $CXCR4^{WT}$, patients with $CXCR4^{NS}$ had lower MRR with ibrutinib treatment, but not with zanubrutinib (**Table 2**). A logistic regression model with treatment group, *TERT* (WT, MUT), and *TP53* (WT, MUT) mutational status as covariates was used to compare VGPR+CR rate and MRR between zanubrutinib and ibrutinib in patients with $CXCR4^{NS}$ and $CXCR4^{FS}$. Compared to ibrutinib, zanubrutinib-treated patients with $CXCR4^{FS}$ had a more favorable VGPR+CR rate (26.3% vs 0%, P=0.06) although MRR was similar (73.7% vs 85.7%). In patients with $CXCR4^{NS}$, the VGPR+CR rates were similar between treatment groups (14.3% vs 15.4%). Zanubrutinib-treated patients had a more favorable MRR (85.7% vs 53.8%, P=0.09). Patients with $CXCR4^{NS}$ and $CXCR4^{FS}$ had longer times to response to both ibrutinib and zanubrutinib than patients with $CXCR4^{WT}$, but zanubrutinib treatment led to faster response compared to ibrutinib in all *CXCR4* subgroups (**Table 2**). Compared to $CXCR4^{WT}$, $CXCR4^{NS}$ was significantly associated with inferior PFS (HR=3.39, P=0.02) and ibrutinib-treated patients with $CXCR4^{FS}$ trended towards a less favorable PFS (HR=2.08, P=0.185). PFS was not significantly affected by *CXCR4* mutational status in zanubrutinib-treated patients ($CXCR4^{NS}$: HR=0.67, P=0.60; $CXCR4^{FS}$: HR=0.62, P=0.473) (**Table 2**). Zanubrutinib-treated patients with either $CXCR4^{NS}$ or $CXCR4^{FS}$ had similar PFS, but PFS in ibrutinib-treated patients with these mutations was less favorable (**Figure S2**). The median PFS (months) in patients receiving ibrutinib by $CXCR4^{NS}$, $CXCR4^{FS}$, and $CXCR4^{WT}$ was 39.8, 44.2, and not reached (NR), respectively, and NR in all subpopulations receiving zanubrutinib. Zanubrutinib treatment was associated with trends towards more favorable PFS in patients with *CXCR4*^{NS} (*P*=0.09), *CXCR4*^{FS} (*P*=0.07), and *CXCR4*^{WT} (*P*=0.32) versus ibrutinib (**Figure S2**).

In addition to detecting high rates of *TP53*^{MUT} in the ASPEN study population, we further observed that, in patients treated with ibrutinib, these mutations were significantly associated with lower MRR (*P*=0.04) and a trend towards a lower VGPR+CR rate (*P*=0.20) than *TP53*^{WT}. In zanubrutinib-treated patients, MRR (*P*=0.98) and VGPR+CR rates (*P*=0.64) were similar between patients with *TP53*^{MUT} and *TP53*^{WT} (**Table 2**). A logistic regression model with treatment group, *TERT* (WT, MUT), and *CXCR4* (WT, FS, NS) mutational status as covariates was used to compare the VGPR+CR rate and MRR between zanubrutinib and ibrutinib in patients with *TP53*^{MUT} WM. Compared to ibrutinib, zanubrutinib treatment resulted in a better VGPR+CR rate (34.6% vs 13.6%, *P*<0.05) and a more favorable MRR (80.8% vs 63.6%, *P*=0.11) in *TP53*^{MUT}. Compared to patients with *TP53*^{WT}, patients with *TP53*^{MUT} had a longer time to response in both ibrutinib- and zanubrutinib-treated patients, though zanubrutinib-treated patients responded faster (**Table 2**). Similarly, patients with *TP53*^{MUT} had worse PFS compared to patients with *TP53*^{WT} in both zanubrutinib and ibrutinib treatment groups. However, in ibrutinib-treated patients, *TP53*^{MUT} was associated with significantly inferior PFS versus *TP53*^{WT} (HR=2.36, *P*=0.027) compared to zanubrutinib-treated patients (HR=2.20, *P*=0.120) (**Table 2**); the median PFS in patients with *TP53*^{MUT} was not reached in the zanubrutinib treatment group and was 44.2 months in the ibrutinib treatment group (HR=0.66, *P*=0.37) (**Table 2**). In addition, three patients harbored both a *TP53* deletion and mutation. One patient treated with ibrutinib had disease progression after 2.8 months of treatment, while two patients treated with zanubrutinib achieved VGPR and PR but progressed after 35.6 months and 16.9 months of treatment, respectively.

Ibrutinib-treated patients with *TP53*^{MUT} exhibited a dosage effect on response rate by VAF, where patients with *TP53*^{MUT} with VAF≥10% or *TP53* deletion had no VGPR+CR and a lower MRR (**Table S6**). This dosage effect on response rate was not observed in patients with *TP53*^{MUT} treated with zanubrutinib. PFS analysis revealed that patients with *TP53*^{MUT} at VAF ≥1% or a *TP53* deletion had worse PFS when treated with ibrutinib (HR=3.792, *P*=0.008) relative to patients treated with zanubrutinib (HR=0.491, *P*=0.22) (**Figure S3**). In addition, patients with *TP53*^{MUT} at VAF ≥1% or *TP53* deletion achieved 35.3% VGPR rate with zanubrutinib versus 0% VGPR with ibrutinib treatment (**Table S6**). Collectively, these data demonstrate that *TP53* mutations confer a worse prognosis in patients treated with BTKi, but zanubrutinib had a more favorable outcome than ibrutinib in this higher risk population.

In summation, the adverse impact of *CXCR4*^{MUT} and *TP53*^{MUT} on response and PFS was more marked in ibrutinib-treated patients than zanubrutinib-treated patients.

Resistance mutations analysis

To begin to explore how acquired mutations may confer resistance to BTKi, we sequenced BMA samples from five WM patients who progressed after achieving a response on zanubrutinib (three *MYD88*^{MUT}, two *MYD88*^{WT}) to analyze the mutational status of *BTK* and other hematological malignancy-related genes. The median treatment duration was 27.9 months (range, 10.2, 34.5) and paired baseline-progression samples were available for four of five patients. *BTK*^{C481}, a mutation associated with resistance, was identified in one patient with progressive disease after zanubrutinib treatment, but unknown at baseline (**Table S9**). Additionally, 80% (4/5) of progressive patients had *TP53* mutations and 40% (2/5) had mutations within the *TERT* promoter (**Table S9**).

DISCUSSION

In this follow-up to the ASPEN study, we examined NGS data and the mutational status of 210 patients with WM to document the association of common mutations with treatment outcomes in patients treated with either ibrutinib or zanubrutinib. Data from the ASPEN study suggests that, overall, *MYD88*^{MUT} patients with *CXCR4*^{MUT} and *TP53*^{MUT} have a worse response to BTKi treatment compared to patients with WT alleles, and that patients with these mutations have generally better treatment outcomes with zanubrutinib.

In our study population, an NGS panel with 0.1%-0.25% sensitivity in non-enriched BMA samples identified mutation rates of *CXCR4* and *ARID1A* comparable to previous studies,[3,8] indicating that a highly sensitive NGS panel may compensate for non-enrichment of samples. However, compared to previous studies that observed a <14% mutation rate in *TP53,*[3,9,10 11] in our study, the *TP53* mutation rate (24.8%) was considerably higher. This observation may potentially be because the high-sensitivity assay used in this study detected a high rate (11.6%) of low-frequency (VAF <1%) mutations compared to less sensitive assays such as Sanger sequencing, whole-exome sequencing, whole-genome sequencing, or targeted sequencing.

A previous study reported that patients with *CXCR4*^{NS} who were treated with ibrutinib had worse odds of major response and worse PFS than patients with *CXCR4*^{FS} and *CXCR4*^{WT}.[26] *CXCR4* mutations may potentially activate cell survival pathways, therefore bypassing BTK to promote ibrutinib resistance. In cell culture, *CXCR4*^{NS} and *CXCR4*^{FS} mutations have been shown to cause impaired membrane internalization leading to prolonged phosphorylation of AKT1 and MAPK1 and, thus, prolonged survival signals for cancer cells.[27] In our study, we also found that patients with *CXCR4*^{NS} mutations had worse clinical outcomes than patients with *CXCR4*^{FS} or *CXCR4*^{WT} in treatment with ibrutinib, but response to zanubrutinib was not affected by *CXCR4* mutation type. This may suggest that zanubrutinib, a more potent BTKi due to prolonged exposure to, or sustained occupancy on, BTK, can potentially offset the deleterious effects of *CXCR4* mutations.[28]

Our current study, consistent with a previous report,[9] found that *TP53*^{MUT} are commonly associated with *MYD88* and *CXCR4* mutations. We demonstrated that patients with *TP53* VAF <1% have similar PFS to patients with *TP53*^{WT}, whereas patients with *TP53* VAF between 1%-10% or >10% fared worse. This observation suggests that low-frequency (VAF<1%) *TP53* mutations may be small subclones detected with a highly sensitive NGS panel and of less clinical significance. The NGS assay used in this study has a validated limit of detection of 0.1-0.25% VAF. Further, variant calls were made using a strict bioinformatic analysis pipeline defined by at least three distinct fragments containing mutations and VAF ≥0.25% or hotspot VAF ≥0.1%. Thus, the low frequency mutations observed in this study are unlikely to be false positives. Three patients harbored both *TP53* deletions and mutations and all of them had disease progression after 16.9 months (range, 2.8, 35.6) following BTKi treatment (two treated with zanubrutinib). These data indicate deficiencies in *TP53* lead to a poor prognosis in WM patients treated with BTKi, though validation in additional WM populations is warranted. Our finding that *TP53* mutations have a significant negative impact on MRR and PFS in ibrutinib-, but not zanubrutinib-treated patients, suggests that more potent BTKi, like zanubrutinib, could improve the suboptimal response of *TP53*^{MUT} WM patients.

While mutations in *CXCR4*, *ARID1A*, and *TP53* have been reported previously in WM, we also identified *TERT* promoter mutations in 19 patients with *MYD88*^{MUT}. Though patients with *TERT*^{MUT} had high rates of *CXCR4* or *TP53* co-mutations, no associations between *TERT*^{MUT} and clinical response were observed (**Table 1**, **Figure2**). While more PFS events were observed in zanubrutinib-treated patients with *TERT*^{MUT}, this may be due to a higher rate of co-occurring *CXCR4*^{MUT} and *TP53*^{MUT} in these patients compared to those in the ibrutinib treatment arm (60% vs 22.2%). Due to unsorted BMA samples used in this study, we are not able to confirm whether the *TERT* mutations that we observed were derived from tumor cells or clonal hematopoiesis of indeterminate potential. Subsequent studies to validate *TERT* mutations in additional WM populations or using single cell sequencing to elucidate its functional role in WM pathogenesis are needed to determine if these mutations represent a risk factor or have prognostic value in WM.

The main limitation of the current study is the small sample size of patients available for analysis with each mutation type. Further comparisons of zanubrutinib and ibrutinib in patients with these risk mutations from other clinical studies or real-world data are warranted. A second potential limitation is that baseline BMA samples were not enriched for tumor cells prior to NGS sequencing. A third limitation is that because paired baseline and disease progression samples were only available for four zanubrutinib-treated patients, comprehensive analysis of potential zanubrutinib resistance mechanisms in patients with WM is limited. Finally, it is unclear if *TERT* mutations are pathogenic and additional studies to validate and elucidate its function in WM pathogenesis are needed.

In conclusion, patients with WM harboring mutations in *CXCR4* and *TP53* were found to have poorer prognosis after treatment with BTKi compared to those without mutations in these genes, and patients with these mutations who received zanubrutinib had favorable outcomes compared with those who received ibrutinib. Thus, inclusion of the mutational status of *MYD88*, *CXCR4*, and *TP53* in genomic-based treatment algorithms may be valuable when assessing BTKi treatment options for patients with WM. Our data further suggest that zanubrutinib can be effective independent of *MYD88*, *CXCR4*, and *TP53* mutational status.

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AUTHORSHIP

Contributions: B.W., Y.Y., Z.S., W.Y.C., H.A., and A.C. designed the research, analyzed the data, generated figures, and assisted in drafting the manuscript. J.S. provided statistical data analysis. C.S.T, S.O., S.D., W.J., H.L., G.C., R.G.O., P.M., B.E.W., R.G., H.M., S.M., A.T., J.J.C., J.C., C.F.R., D.B., E.L., J.M., M.M., T.S., M.T., M.T., M.C.M, C.B., V.L., S.P.T., and J.T. enrolled patients and collected clinical data. All authors contributed to data interpretation and reviewed the manuscript.

NON-AUTHOR CONTRIBUTIONS AND DISCLOSURES

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Table 1. Response assessment by *CXCR4*, *TP5*3, *TERT*, *ARID1A* mutational status in WM patients with $MYD88^{MUT}$.

Patients with $CXCR4^{MUT}$, $TP53^{MUT}$, and $TERT^{MUT}$ trended toward lower VGPR+CR rate, lower major response rate, and/or longer median time to response than patients with the respective WT alleles. To compare the response rates, univariate logistic regression models were performed, and the OR (95%CI) and the corresponding *P* values[1] are shown. In addition, OR (95% CI) and *P* values[2] were estimated using a multivariate logistic regression model with treatment arm, *CXCR4* (WT, MUT), *TERT* (WT, MUT), and *TP53* (WT, MUT) mutational status as covariates to account for the correlations among the mutations and the treatment differences between treatment arms. WT is the reference group. The same models were performed to further compare the response rates $CXCR4^{WT}$ and $CXCR4^{NS}$ or $CXCR4^{FS}$ (CXCR4 [WT, FS, NS]).[3] *MYD88* status was assessed by PCR-based assay, with a total of 190 patients with $MYD88^{MUT}$ WM. CR, complete response; FS, frameshift; MR, minor response; MUT, mutated; NE, not evaluable due to discontinuation prior to first assessment; NS, nonsense; OR, odds ratio; PD, progressive disease; PR, partial response; SD, stable disease; VGPR, very good partial response; WM, Waldenström macroglobulinemia; WT, wild-type.

	CXCR4^{WT} (n=137)	*CXCR4^{MUT}* (n=53)	*CXCR4^{FS}* (n=26)	*CXCR4^{NS}* (n=27)	*TP53^{WT}* (n=142)	*TP53^{MUT}* (n=48)	*TERT^{WT}* (n=171)	*TERT^{MUT}* (n=19)	*ARID1A^{WT}* (n=158)	*ARID1A^{MUT}* (n=32)
Best overall response, n (%)										
CR	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
VGPR	51 (37.2)	9 (17.0)	5 (19.2)	4 (14.8)	48 (33.8)	12 (25.0)	58 (33.9)	2 (10.5)	52 (32.9)	8 (25.0)
PR	64 (46.7)	30 (56.6)	15 (57.7)	15 (55.6)	71 (50.0)	23 (47.9)	85 (49.7)	9 (47.4)	77 (48.7)	17 (53.1)
MR	16 (11.7)	10 (18.9)	4 (15.4)	6 (22.2)	16 (11.3)	10 (20.8)	20 (11.7)	6 (31.6)	21 (13.3)	5 (15.6)
SD	3 (2.2)	3 (5.7)	2 (7.7)	1 (3.7)	4 (2.8)	2 (4.2)	5 (2.9)	1 (5.3)	6 (3.8)	0 (0.0)
PD	2 (1.5)	1 (1.9)	0 (0.0)	1 (3.7)	2 (1.4)	1 (2.1)	2 (1.2)	1 (5.3)	2 (1.3)	1 (3.1)
NE	1 (0.7)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.7)	0 (0.0)	1 (0.6)	0 (0.0)	0 (0.0)	1 (3.1)
VGPR or better, n (%)	51 (37.2)	9 (17.0)	5 (19.2)	4 (14.8)	48 (33.8)	12 (25.0)	58 (33.9)	2 (10.5)	52 (32.9)	8 (25.0)
Major response, n (%)	115 (83.9)	39 (73.6)	20 (76.9)	19 (70.4)	119 (83.8)	35 (72.9)	143 (83.6)	11 (57.9)	129 (81.7)	25 (78.1)
Time to response, median (min, max), months										
VGPR or CR	8.4 (1.9, 50.0)	11.1 (2.8, 46.0)	11.1 (2.8, 26.0)	13.9 (9.4, 46.0)	9.3 (1.9, 50.0)	11.1 (3.0, 46.9)	9.3 (1.9, 50.0)	34.1 (22.2, 46.0)	9.8 (1.9, 49.9)	10.7 (2.8, 46.0)
Major response	2.8 (0.9, 49.8)	4.6 (1.0, 49.8)	6.6 (1.8, 49.8)	3.7 (1.0, 38.7)	2.9 (0.9, 49.8)	2.9 (1.0, 13.8)	2.8 (0.9, 49.8)	5.6 (1.8, 22.2)	2.8 (0.9, 49.8)	3.0 (1.0, 38.7)

Statistical analysis for rates of VGPR or better						
	Univariate analysis [1]		Multivariate analysis [2]		Multivariate analysis [3]	
	Odds ratio (95% CI)	P value	Odds ratio (95% CI)	P value	Odds ratio (95% CI)	P value

***CXCR4**^{MUT}*	**0.345 (0.156, 0.765)**	**0.009**	**0.370 (0.160, 0.855)**	**0.020**	**-**	**-**
***CXCR4**^{FS}*	**0.401 (0.143, 1.130)**	**0.084**	**-**	**-**	**0.379 (0.130, 1.101)**	**0.075**
***CXCR4**^{NS}*	**0.293 (0.096, 0.896)**	**0.031**	**-**	**-**	**0.360 (0.112, 1.156)**	**0.086**
***TP53**^{MUT}*	**0.653 (0.311, 1.368)**	**0.259**	**0.780 (0.360, 1.690)**	**0.529**	**0.781 (0.360, 1.692)**	**0.530**
***TERT**^{MUT}*	**0.229 (0.051, 1.026)**	**0.054**	**0.353 (0.075, 1.671)**	**0.189**	**0.355 (0.075, 1.687)**	**0.193**
Statistical analysis for major response rate						
	Univariate analysis [1]		**Multivariate analysis [2]**		**Multivariate analysis [3]**	
	Odds ratio (95% CI)	**P value**	**Odds ratio (95% CI)**	**P value**	**Odds ratio (95% CI)**	**P value**
***CXCR4**^{MUT}*	**0.533 (0.249, 1.142)**	**0.106**	**0.718 (0.308, 1.677)**	**0.444**	**-**	**-**
***CXCR4**^{FS}*	**0.638 (0.230, 1.768)**	**0.387**	**-**	**-**	**0.737 (0.251, 2.167)**	**0.579**
***CXCR4**^{NS}*	**0.454 (0.177, 1.167)**	**0.101**	**-**	**-**	**0.701 (0.242, 2.027)**	**0.512**
***TP53**^{MUT}*	**0.520 (0.239, 1.132)**	**0.100**	**0.625 (0.276, 1.411)**	**0.258**	**0.626 (0.277, 1.415)**	**0.260**
***TERT**^{MUT}*	**0.269 (0.099,**	**0.010**	**0.345 (0.118,**	**0.053**	**0.348 (0.117,**	**0.058**

	0.729)		**1.014)**		**1.038)**		

Table 2. Response assessment by *CXCR4, TP53,* and *TERT* mutational statuses and by treatment in patients with *MYD88*^{MUT}.

Response rates, time to response, and PFS were compared according to the mutational status of *CXCR4*, *TP53*, and *TERT* genes in 92 ibrutinib-treated and 98 zanubrutinib-treated patients with *MYD88*^{MUT} WM, respectively. Odds ratio and *P* values were estimated using a logistic regression model with *CXCR4* (WT, NS, FS), *TP53* (WT, MUT), and *TERT* (WT, MUT) mutational status as covariates. Median PFS was estimated by Kaplan-Meier method, HR and *P* values were estimated using a Cox regression model with *CXCR4* (WT, FS, NS), *TP53* (WT, MUT), and *TERT* (WT, MUT) mutational statues as covariates. WT is the reference group. *MYD88* status was assessed by a PCR-based assay, with a total of 190 patients with *MYD88*^{MUT} WM. FS, frameshift; HR, hazard ratio; MUT, mutated; NE, not estimable; NS, nonsense; PFS, progression-free survival; VGPR, very good partial response; WM, Waldenström macroglobulinemia; WT, wild-type.

Patients with *MYD88*^{MUT} treated with ibrutinib (n=92)							
	CXCR4^{WT} **(n=72)**	***CXCR4***^{FS} **(n=7)**	***CXCR4***^{NS} **(n=13)**	***TP53***^{WT} **(n=70)**	***TP53***^{MUT} **(n=22)**	***TERT***^{WT} **(n=83)**	***TERT***^{MUT} **(n=9)**
VGPR or better, n (%)[a]	22 (30.6)	0	2 (15.4)	21 (30.0)	3 (13.6)	23 (27.7)	1 (11.1)
OR (95% CI)	-	0.14 (0.00, 3.23)	0.64 (0.13, 3.08)	-	0.44 (0.12, 1.55)	-	0.52 (0.07, 3.79)
P value	-	0.223	0.579	-	0.202	-	0.525
Major response, n (%)[a]	61 (84.7)	6 (85.7)	7 (53.8)	60 (85.7)	14 (63.6)	70 (84.3)	4 (44.4)
OR (95% CI)	-	1.06 (0.10, 10.36)	0.33 (0.07, 1.41)	-	0.29 (0.09, 0.95)	-	0.23 (0.04, 1.22)
P value	-	0.958	0.135	-	0.040	-	0.085
Time to VGPR or better Median (min, max), months	11.3 (2.0, 49.9)	-	31.3 (16.6, 46.0)	11.4 (2.0, 49.9)	24.9 (5.6, 46.9)	11.4 (2.0, 49.9)	46.0 (46.0, 46.0)
Time to major response Median (min, max), months	2.8 (0.9, 49.8)	7.0 (2.8, 41.5)	2.9 (1.2, 13.6)	2.9 (0.9, 49.8)	3.0 (1.0, 13.8)	2.8 (0.9, 49.8)	10.3 (2.9, 13.8)
PFS[b]							
Event-free rate at 42 months, %	74.6	57.1	43.5	72.1	57.9	68.4	74.0
Median, months	NE	44.2	39.8	NE	44.2	NE	48.2
HR (95% CI)	-	2.08 (0.70, 6.16)	3.39 (1.23, 9.31)	-	2.36 (1.10, 5.09)	-	0.44 (0.10, 1.81)
P value		0.185	0.017		0.027	-	0.257
Patients with *MYD88*^{MUT} treated with zanubrutinib (n=98)							
	CXCR4^{WT} **(n=65)**	***CXCR4***^{FS} **(n=19)**	***CXCR4***^{NS} **(n=14)**	***TP53***^{WT} **(n=72)**	***TP53***^{MUT} **(n=26)**	***TERT***^{WT} **(n=88)**	***TERT***^{MUT} **(n=10)**
VGPR or better, n (%)[b]	29 (44.6)	5 (26.3)	2 (14.3)	27 (37.5)	9 (34.6)	35 (39.8)	1 (10.0)
OR (95% CI)	-	0.51 (0.16, 1.66)	0.24 (0.04, 1.26)	-	1.27 (0.46, 3.52)	-	0.25 (0.02, 2.26)
P value	-	0.269	0.093	-	0.636	-	0.219
Major response, n (%)[b]	54 (83.1)	14 (73.7)	12 (85.7)	59 (81.9)	21 (80.8)	73 (83.0)	7 (70.0)
OR (95% CI)	-	0.66 (0.18, 2.36)	1.52 (0.25, 9.01)	-	1.01 (0.29, 3.47)	-	0.47 (0.09, 2.39)
P value	-	0.524	0.639	-	0.978	-	0.362
Time to VGPR or better Median (min, max), months	6.5 (1.9, 42.0)	11.1 (2.8, 26.0)	10.3 (9.4, 11.1)	6.5 (1.9, 42.0)	11.1 (3.0, 26.0)	6.7 (1.9, 49.8)	22.2 (22.2, 22.2)
Time to major response Median (min, max), months	2.8 (0.9, 28.5)	2.9 (1.8, 49.8)	4.1 (1.0, 38.7)	2.8 (0.9, 49.8)	2.8 (1.0, 5.6)	2.8 (0.9, 49.8)	3.7 (1.8, 22.2)
PFS							
Event-free rate at 42 months, %	81.3	76.4	66.7	84.6	62.0	83.4	37.5
Median, months[c]	NE	NE	NE	NE	NE	NE	25.0
HR (95% CI)[d]	-	0.62 (0.17, 2.25)	0.67 (0.15, 2.88)	-	2.20 (0.81, 5.98)	-	5.78 (1.75, 19.13)
P value[d]		0.473	0.598		0.120	-	0.004

Table 3. Response assessment by *TP53* mutation status in patients with *MYD88*^{WT}.

In twenty patients with *MYD88*^{WT}, four patients with *TP53*^{MUT} had a lower major response rate (50%), and none achieved VGPR or CR, compared with *TP53*^{WT} patients. CR, complete response; MR, minor response; MUT, mutant; PD, progressive disease, PR, partial response; SD, stable disease; VGPR, very good partial response; WT, wild-type.

	Total (N=20)	*TP53*^{WT} (n=16)	*TP53*^{MUT} (n=4)
Best overall response, n (%)			
CR	1 (5.0)	1 (6.3)	0 (0.0)
VGPR	3 (15.0)	3 (18.8)	0 (0.0)
PR	8 (40.0)	6 (37.5)	2 (50.0)
MR	4 (20.0)	3 (18.8)	1 (25.0)
SD	3 (15.0)	2 (12.5)	1 (25.0)
PD	1 (5.0)	1 (6.3)	0 (0.0)
VGPR or better, n (%)	4 (20.0)	4 (25.0)	0 (0.0)
Major response, n (%)	12 (60.0)	10 (62.5)	2 (50.0)
Time to response, median (min, max), months			
VGPR or CR	9.6 (2.8, 22.1)	9.6 (2.8, 22.1)	-
Major response	3.4 (1.8, 44.6)	3.3 (1.8, 44.6)	4.3 (3.0, 5.5)

FIGURE LEGENDS

Figure 1. Baseline genetic alteration landscape in 210 WM patients treated with BTK inhibitors. DNA mutation profile of WM patients and the distribution of mutations among different study cohorts by mutation type and treatment status (TN and RR). Each column represents one patient, and each row represents one gene (represented by the gene symbol). Mutation rates of each gene are shown on the left. Mutation type, treatment status, and best overall response are color-coded as shown in the figure legend. BM, bone marrow; BOR, best overall response; BTK, Bruton tyrosine kinase; CR, complete response; HR, hazard ratio; MR, minor response; MUT, mutated; PD, progressive disease; PR, partial response; RR, relapsed/refractory; SD, stable disease; TN, treatment naïve; VGPR, very good partial response; WM, Waldenström macroglobulinemia; WT, wild-type.

Figure 2. Kaplan-Meier curves of PFS in WM patients with *MYD88*^{MUT} in relation to *CXCR4*, *TP53*, and *TERT* mutational status. Pooled analysis of patients with *MYD88*^{MUT} including 98 treated with zanubrutinib and 92 treated with ibrutinib. Kaplan-Meier curves of PFS were presented according to the mutational statuses of *CXCR4* (A), *TP53* (B), and *TERT* (C). PFS in patients with *CXCR4*^{MUT}, *TP53*^{MUT}, and *TERT*^{MUT} trended toward less favorable outcome than patients with the respective WT alleles. HR and *P* values were estimated using a Cox regression model with *CXCR4* (WT, MUT), *TP53* (WT, MUT), *TERT* (WT, MUT) mutational statuses and treatment arms as covariates. WT is the reference group. HR, hazard ratio; MUT, mutated; PFS, progression-free survival; WM, Waldenström macroglobulinemia; WT, wild-type.

Figure 1

Figure 1. Baseline genetic alteration landscape in 210 WM patients treated with BTK inhibitors

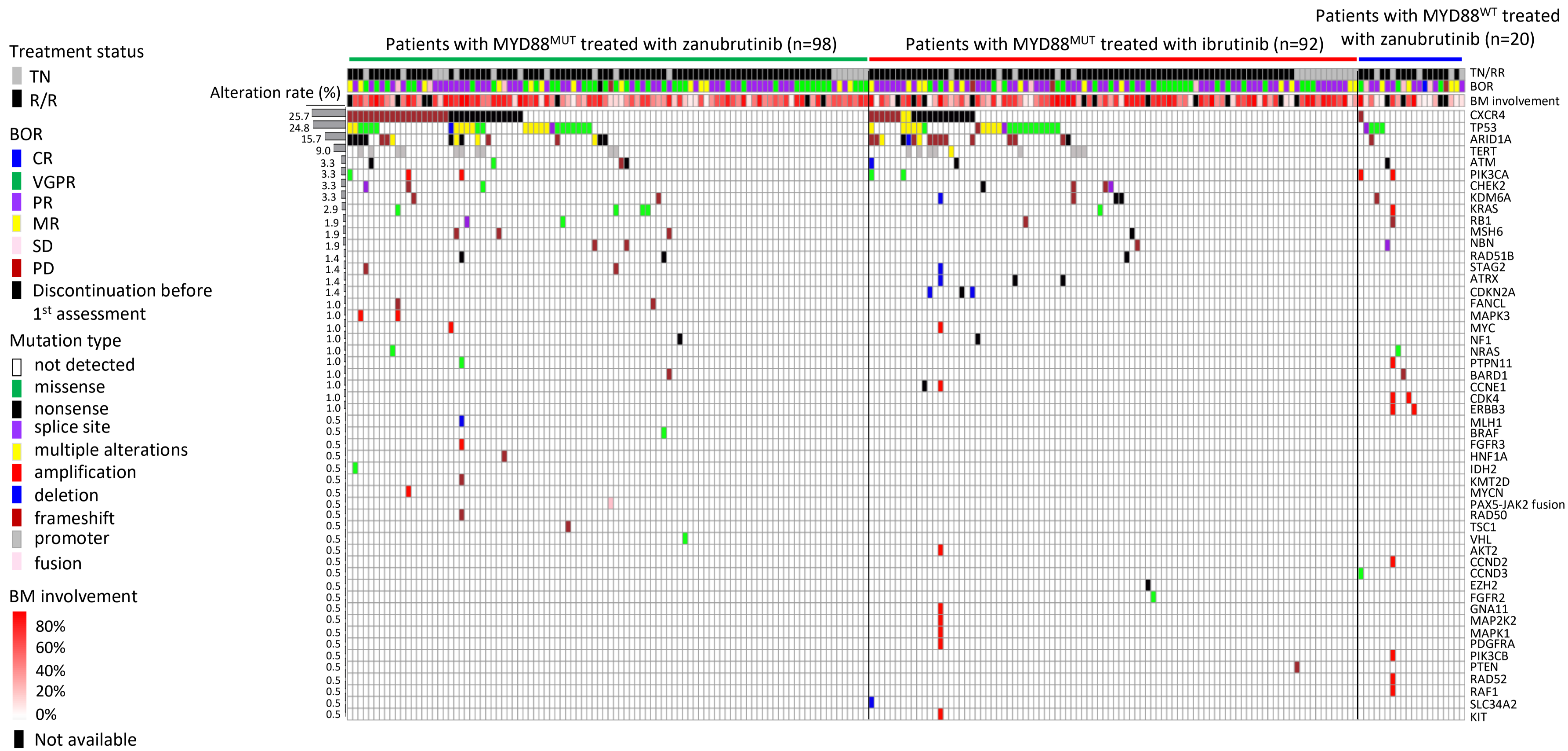
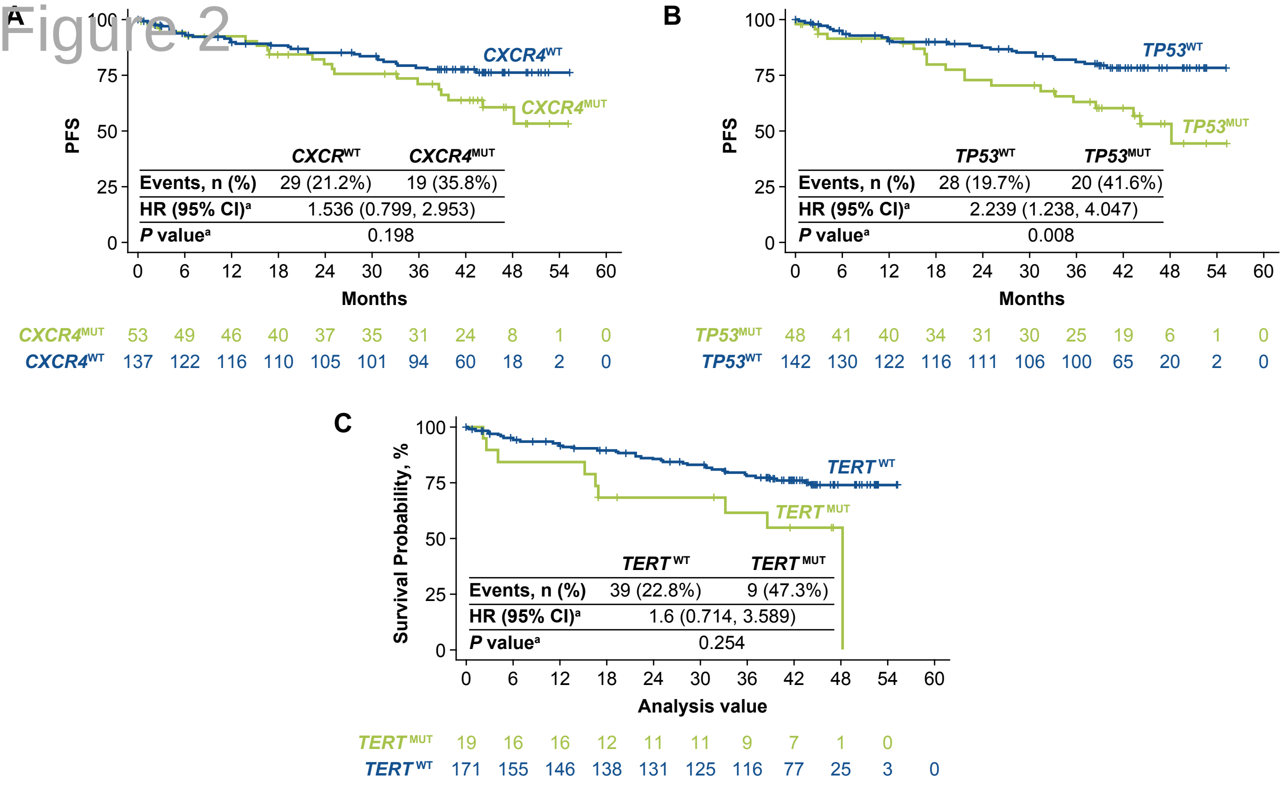


Figure 2


A
100
75
50
25
0
PFS
CXCR4WT
CXCR4MUT
CXCRWT
CXCR4MUT
Events, n (%)
29 (21.2%)
19 (35.8%)
HR (95% CI)a
1.536 (0.799, 2.953)
P valuea
0.198
0 6 12 18 24 30 36 42 48 54 60
Months
CXCR4MUT 53 49 46 40 37 35 31 24 8 1 0
CXCR4WT 137 122 116 110 105 101 94 60 18 2 0
B
100
75
50
25
0
PFS
TP53WT
TP53MUT
Events, n (%)
28 (19.7%)
20 (41.6%)
HR (95% CI)a
2.239 (1.238, 4.047)
P valuea
0.008
0 6 12 18 24 30 36 42 48 54 60
Months
TP53MUT 48 41 40 34 31 30 25 19 6 1 0
TP53WT 142 130 122 116 111 106 100 65 20 2 0
C
100
75
50
25
0
Survival Probability, %
TERT WT
TERT MUT
Events, n (%)
39 (22.8%)
9 (47.3%)
HR (95% CI)a
1.6 (0.714, 3.589)
P valuea
0.254
0 6 12 18 24 30 36 42 48 54 60
Analysis value
TERT MUT 19 16 16 12 11 11 9 7 1 0
TERT WT 171 155 146 138 131 125 116 77 25 3 0