Acquired Mutations in Patients With Relapsed/Refractory Chronic Lymphocytic Leukemia Who Progressed in the ALPINE Study

Jennifer R. Brown,[1] Jessica Li,[2] Barbara F. Eichhorst,[3] Nicole Lamanna,[4] Susan M. O'Brien,[5] Constantine S. Tam,[6] Luqui Qiu,[7] Ruiqi Huang,[8] Yang Shi,[9] Adam Idoine,[2] Tommi Salmi,[10] Aileen Cohen,[2] Mazyar Shadman[11]

[1]Dana-Farber Cancer Institute, Boston, MA, USA; [2]BeiGene USA, Inc, San Mateo, CA, USA; [3]Department of Internal Medicine, University of Cologne, Center for Integrated Oncology Aachen Bonn Köln Düsseldorf, Cologne, Germany; [4]Columbia University, New York, NY, USA; [5]University of California, Irvine, CA; [6]Alfred Hospital and Monash University, Melbourne, VIC, Australia; [7]Institute of Hematology and Blood Diseases Hospital, Chinese Academy of Medical Sciences, Tianjin, China; [8]BeiGene (Shanghai) Co, Ltd, Shanghai, China; [9]BeiGene (Beijing) Co, Ltd, Beijing, China; [10]BeiGene International GmbH, Basel, Switzerland; [11]Fred Hutchinson Cancer Center, University of Washington School of Medicine, Seattle, WA, USA

Disclosures for Jennifer R. Brown

Employed with Dana-Farber Cancer Institute (Boston, MA). Consultant for AbbVie, Acerta/AstraZeneca, Alloplex Biotherapeutics, BeiGene, Galapagos NV, Genentech/Roche, Grifols Worldwide Operations, InnoCare Pharma Inc, iOnctura, Kite, Loxo/Lilly, Merck, Numab Therapeutics, Pfizer, and Pharmacyclics. Received research funding from BeiGene, Gilead, iOnctura, Loxo/Lilly, MEI Pharma, and TG Therapeutics.

Introduction

- Patients administered cBTK inhibitors for CLL can develop acquired drug resistance, leading to disease progression
- Often, cBTK inhibitor resistance results from the emergence of subclones with *BTK* mutations at the cBTK inhibitor binding site (C481) and/or *PLCG2* mutations
- Less frequently, non-C481 *BTK* mutations, including gatekeeper residue T474 and kinase-impaired L528 mutations, have been reported in patients with progression on cBTK inhibitors
- Most previous reports of cBTK inhibitor resistance mutations have been retrospective or in small patient populations
- Here, to gain further insight into the genetic mechanisms of cBTK inhibitor resistance in a randomized population of patients with CLL, NGS was performed on samples from patients who progressed on zanubrutinib or ibrutinib in the phase 3 ALPINE study (NCT03734016)[1]

1. Brown JR, et al. *N Engl J Med.* 2023;388(4):319-332

BTK, Bruton tyrosine kinase; cBTK, covalent Bruton tyrosine kinase; CLL, chronic lymphocytic leukemia; NGS, next-generation sequencing.

Baseline Characteristics of Patients With PD

- A total of 57 patients with PD[a] assessed by either investigator (n=132) and/or the independent review committee (n=139) had PD samples collected for this post hoc biomarker analysis
- Peripheral blood samples were collected at baseline and at or after PD and prior to subsequent therapy

	Zanubrutinib (n=26)	Ibrutinib (n=31)
Number of prior treatments, median (range)	1 (1-3)	1 (1-7)
Study follow-up time, median (range), mo	25.4 (10.6-40.5)	28.1 (5.8-42.3)
Duration of treatment, median (range), mo	19.9 (4.3-39.3)	16.6 (3.4-35.7)
del(17p) and/or TP53 mutation, n (%)	5 (19.2)	6 (19.4)
IGHV unmutated, n (%)	22 (84.6)	26 (83.9)

PFS final analysis data cutoff: August 8, 2022

[a] Assessed using Hallek et al. criteria.[1]

1. Hallek M, et al. *Blood*. 2008;111(12):5446-5456.

Del(17p), deletion 17p; IGHV, immunoglobulin variable heavy chain; PD, progressive disease; PFS, progression-free survival; TP53, tumor protein p53.

Blood Samples Available for Biomarker Analysis

- A total of 52 patients with paired baseline and PD samples and without RT as assessed at PD were included in this analysis
- NGS was performed using a 106-gene PredicineHEME panel[a]
  - 27 CLL driver genes identified by Knisbacher et al.[1] were represented in this panel
- The limit of detection was 0.1% for hotspot mutations and 0.25% for non-hotspot mutations; data reported include all *BTK* and *PLCG2* mutations with a VAF of ≥0.25%. For all other genes, pathogenic mutations with a VAF ≥1% were reported

Patients, n		Zanubrutinib (n=26)	Ibrutinib (n=31)	Total (N=57)
No RT at PD	Paired baseline and PD sample	24	28	52
	Without baseline but had PD sample	0	1[b]	1[b]
RT at PD	Paired baseline and PD sample	2[b]	0	2[b]
	Without baseline but had PD sample	0	2[b]	2[b]

RT reported as of data cutoff: August 8, 2022

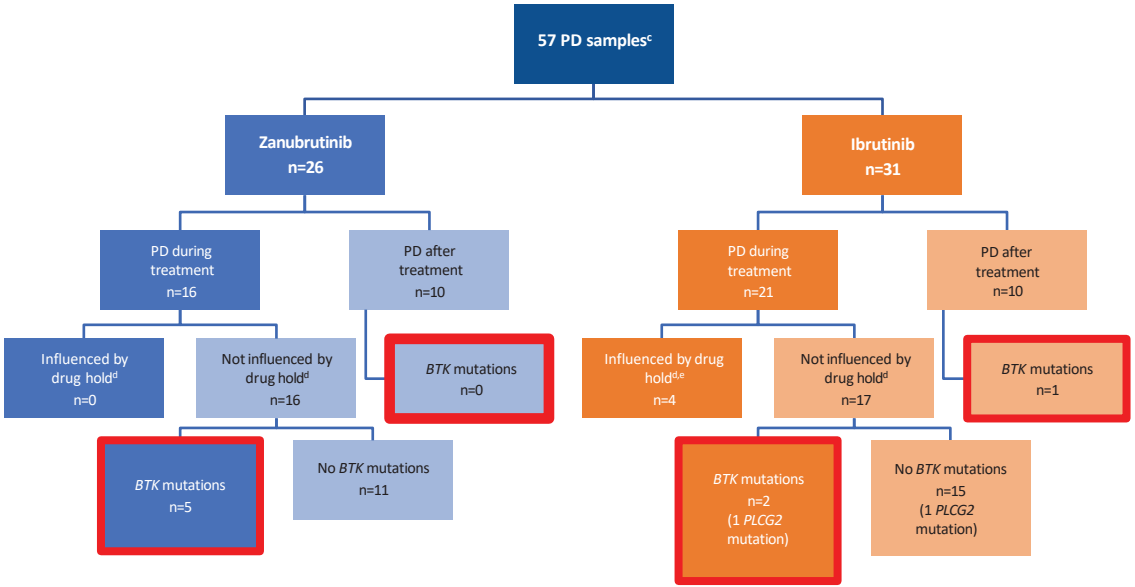
[a] Other assessments included fluorescence in situ hybridization for chromosome abnormalities; cytogenetic analysis for CK ≥3; and NGS for IGHV gene mutation per the European Research Initiative on CLL. [b] No acquired *BTK/PLCG2* mutations were detected.

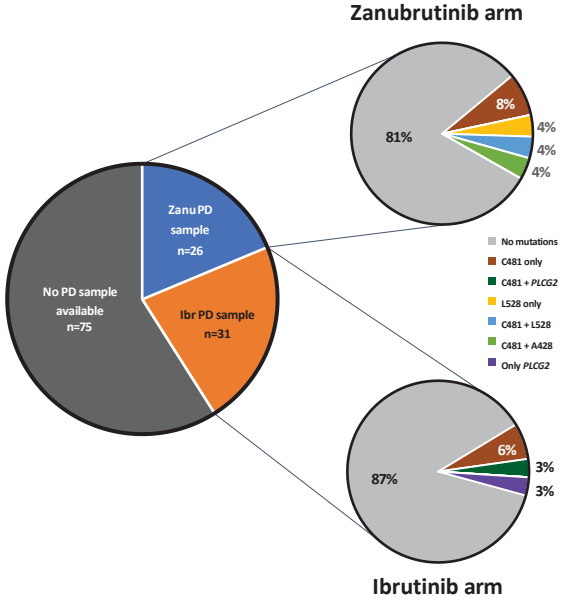
1. Knisbacher BA, et al. Nat Genet. 2022;54(11):1664-1674.

CK, complex karyotype; CLL, chronic lymphocytic leukemia; IGVH, immunoglobulin variable heavy chain; NGS, next-generation sequencing; PD, progressive disease; RT, Richter transformation; VAF, variant allele frequency.

PD Samples With *BTK* and/or *PLCG2* Mutation Distribution

- No *BTK* mutations were identified at baseline
- At PD, 8 patients had acquired mutations in *BTK*, with half of these patients having ≥2 *BTK* mutations[a]
- Among patients without RT who progressed on zanubrutinib (n=24), 5 acquired *BTK* mutations[b]





[a] 77.8% (14/18) of the *BTK* mutations were at C481. One patient had a sole *PLCG2* mutation at PD. [b] L528W only, n=1; C481 only, n=2; L528W and C481, n=1; A428D and C481, n=1. [c] Peripheral blood samples were collected at baseline and at or after PD and prior to subsequent therapy. [d] Hold ≥7 days within 6 weeks before progressive disease. [e] No *BTK* or *PLCG2* mutations.
BTK, Bruton tyrosine kinase; ibru, ibrutinib; PD, progressive disease; RT, Richter transformation; zanu, zanubrutinib.

Acquired *BTK* and *PLCG2* Mutations by Patient

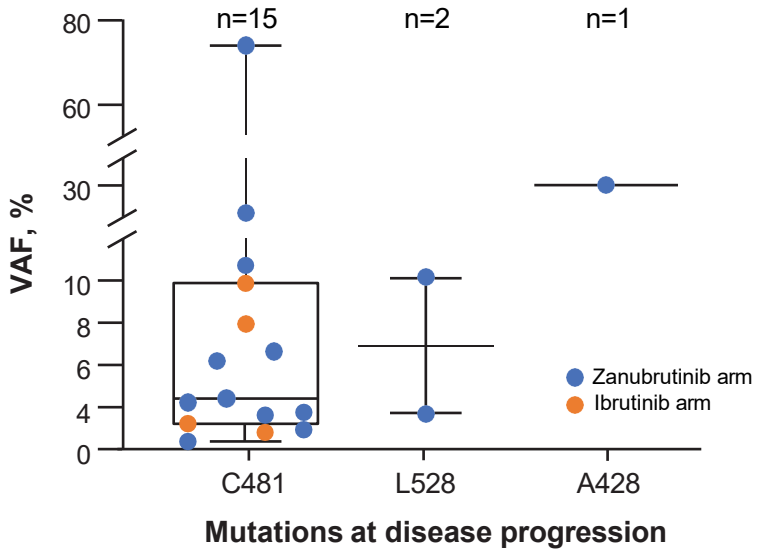
- Overall median treatment duration was 17.0 months (range, 5.0-34.5 months)
- Among the patients with *BTK* mutations at PD, median treatment duration was 29.7 months in those treated with zanubrutinib (n=5) vs 30.8 months in those treated with ibrutinib (n=3)

Patient ID	Treatment arm	Acquired *BTK* mutation at PD: coding DNA description (VAF, %)	Acquired *BTK* mutation at PD: protein description	Acquired *PLCG2* mutation at PD: coding DNA description (VAF, %)	Acquired *PLCG2* mutation at PD: protein description	Duration of treatment, months
1	Ibrutinib	1442G>C (1.29)	C481S	Not detected	Not detected	30.8
2	Ibrutinib	1442G>C (7.95)	C481S	Not detected	Not detected	34.5
3	Ibrutinib	1442G>C (0.88)	C481S	2535A>C (0.60)	L845F	11.8
		127G>C (0.51)	D43H			
4	Ibrutinib	Not detected	Not detected	3422T>A (5.69)	M1141K	18.8
5	Zanubrutinib	1442G>C (8.80)	C481S	Not detected	Not detected	34.2
6	Zanubrutinib	1283C>A (31.10)	A428D	Not detected	Not detected	28.0
		1442G>C (4.72)	C481S			
		1441T>A (2.48)	C481S			
7	Zanubrutinib	1442G>C (16.22)	C481S	Not detected	Not detected	29.7
		1583T>G (8.22)	L528W			
		1441T>A (4.28)	C481S			
		1442G>A (1.83)	C481Y			
		1442G>T (1.70)	C481F			
		1441T>C (1.01)	C481R			
8	Zanubrutinib	1583T>G (1.76)	L528W	Not detected	Not detected	33.8
9	Zanubrutinib	1442G>C (74.39)	C481S	Not detected	Not detected	18.4
		1441T>C (2.30)	C481R			
		1441T>A (0.45)	C481S			

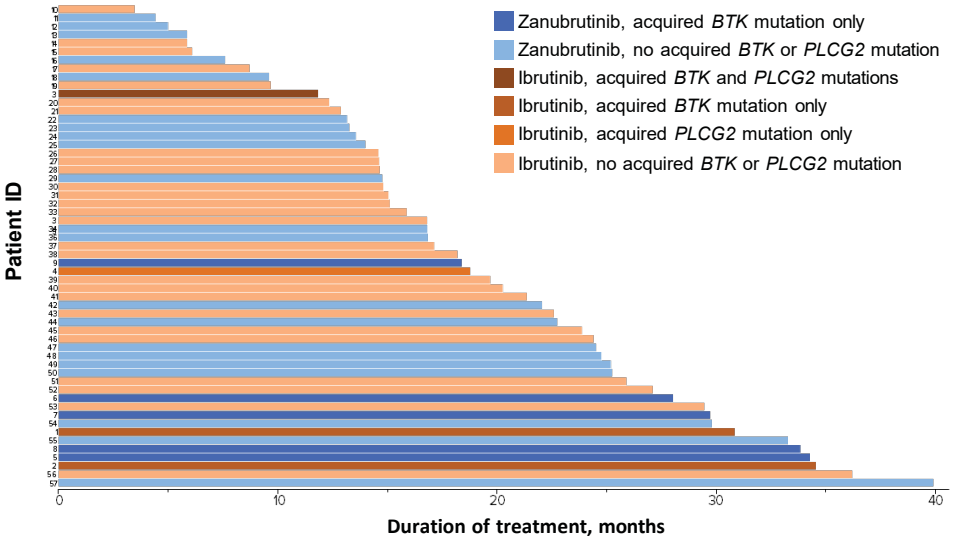
BTK, Bruton tyrosine kinase; PD, progressive disease; VAF, variant allele frequency.

Acquired *BTK* Mutation VAF and Treatment Duration by Mutation Status

- The VAF of the 2 *BTK* L528 mutations was similar to that of the *BTK* C481 mutations

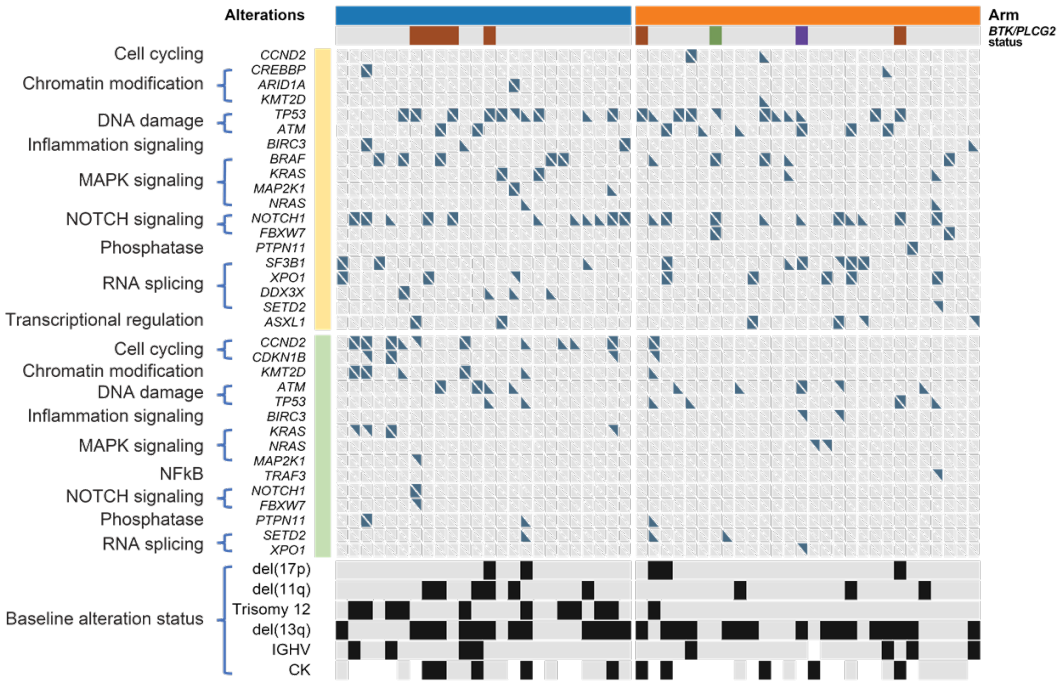


- Median treatment duration at disease progression was shorter in patients with wild-type *BTK* in both the zanubrutinib (n=19, 16.8 months [range, 5.0-33.3 months], *P*<.01) and ibrutinib (n=25, 15.9 months [range, 5.9-29.4 months], *P*=.21) treatment arms



BTK, Bruton tyrosine kinase; VAF, variant allele frequency.

Driver Gene Alterations and Molecular Pathways



- Among the 48 patients who had baseline CLL driver gene mutations, 18 mutated driver genes were identified; the median number of driver genes mutated per patient was 3 (range, 1-5)[a]
- No associations between driver gene mutations and *BTK* mutational status were detected
- Driver gene mutations at either baseline or PD were not associated with del(17p), IGHV mutation, or CK status

[a] Mutations were most frequently observed in *NOTCH1* (n=21), *TP53* (n=19), *BRAF* (n=10), *SF3B1* (n=8), and *ATM* (n=8) at baseline. Acquired driver gene mutations were observed in 1 patient in the zanubrutinib arm (with *TP53* and *XPO1* mutations) and 5 patients in the ibrutinib arm (1 with *TP53*, 1 with *SETD2*, 1 with *SF3B1*, and 2 with *ASXL1* mutations).

BTK, Bruton tyrosine kinase; CK, complex karyotype; CLL, chronic lymphocytic leukemia; CNV, copy number variant; del, deletion; IGHV, immunoglobulin variable heavy chain; PD, progressive disease; SC, screening; SNV, single-nucleotide variant.

Next Line of Treatment After Discontinuation

- The majority of patients received additional treatment following study treatment discontinuation (zanubrutinib, 18/26 [69.2%]; ibrutinib, 21/31 [67.7%]), including all patients with acquired *BTK* and/or *PLCG2* mutations

Next line of treatment after discontinuing study treatment	Zanubrutinib (n=26)		Ibrutinib (n=31)	
	Patients, n	Outcome	Patients, n	Outcome
Chemotherapy	1	Ongoing/completed	0	N/A
Chemoimmunotherapy[a]	4	Ongoing/completed, n=2 (*BTK* C481 mutation, n=1; RT when completing study treatment, n=1); discontinued due to AE, n=2	3	Ongoing/completed
cBTK inhibitor therapy	2	Ongoing/completed, n=1; PD, n=1	5	Ongoing/completed, n=3; PD, n=1 (*BTK* C481 mutation); discontinued due to AE, n=1
Noncovalent BTK inhibitor therapy[b]	2	Ongoing/completed, n=1; PD, n=1 (*BTK* C481 and L528 mutations)	2	Ongoing/completed, n=1; death, n=1
BCL2i monotherapy	3	Ongoing/completed, n=2; discontinued due to AE, n=1	5	Ongoing/completed, n=2 (*BTK* C481 mutation, n=1); PD, n=1; discontinued due to AE, n=1; death, n=1
BCL2i plus mCD20Ab therapy	3	PD, n=1 (*BTK* L528 mutation); discontinued due to AE, n=1 (*BTK* C481 mutation); death, n=1	2	Ongoing/completed, n=1; PD, n=1 (*PLCG2* mutation)
BCL2i plus BTK inhibitor therapy[c]	0	N/A	3	Ongoing/completed, n=1; PD, n=1 (RT when completing study treatment); discontinued due to AE, n=1 (*BTK* C481 and *PLCG2* mutations)
mCD20Ab plus BCL2i plus noncovalent BTK inhibitor	1	Ongoing/completed	0	N/A
Other[d]	2	Ongoing/completed, n=1; unknown, n=1 (*BTK* C481 and A428 mutations)	1	PD
No known treatment after study treatment discontinuation	8	(RT when completing study treatment, n=1)	10	(RT when completing study treatment, n=1)

[a] One patient in the zanubrutinib arm was co-administered venetoclax. [b] One patient in the ibrutinib arm was co-administered mCD20Ab. [c] Two patients were co-administered a cBTK inhibitor and 1 patient a noncovalent BTK inhibitor. [d] Two patients (1 in each arm) were treated with a spleen tyrosine kinase inhibitor and 1 patient with rituximab plus a PI3K-δ inhibitor.

AE, adverse event; BCL2i, B-cell lymphoma 2 inhibitor; BTK, Bruton tyrosine kinase; cBTK, covalent Bruton tyrosine kinase; mCD20Ab, monoclonal CD20 antibody; N/A, not applicable; PD, progressive disease; RT, Richter transformation.

Conclusions

- Of the patients who progressed in ALPINE and were included in this analysis, most (83%) did not acquire *BTK* or *PLCG2* mutations
- Among the 24 patients in this analysis who progressed on zanubrutinib, 5 (21%) acquired *BTK* mutations
- These data suggest that *BTK* and/or *PLCG2* mutations are not the main factors driving PD in this population
- Given the low incidence to date of non-C481 mutations in patients with PD in ALPINE, patients with CLL who have been treated with cBTK inhibitors are likely to remain sensitive to other BTK-targeting therapies

BTK, Bruton tyrosine kinase; cBTK, covalent Bruton tyrosine kinase; CLL, chronic lymphocytic leukemia; PD, progressive disease.

Acknowledgments

- We would like to thank the investigators, site support staff, and especially the patients for participating in this study
- This study was sponsored by BeiGene, Ltd. Editorial assistance was provided by Jenna M. Gaska, PhD, CMPP, of Nucleus Global, an Inizio Company, and was funded by BeiGene
- Previously presented at the 65th ASH Annual Meeting and Exposition; December 9-12, 2023, San Diego, CA. Abstract 1890

Corresponding Author:

Jennifer R. Brown; email: Jennifer_Brown@dfci.harvard.edu