SUPPLEMENTARY DATA

Supplementary Table S1. Mutations Detectable by BEAMing

Primary Mutations			Secondary Mutations		
Exon 9	Exon 11	Exon 13	ATP Pocket Exon 13/14	Activation Loop Exon 17/18	
Y503_F504insAY	W557_K558del	K642E	V654A	C809G	D820V
	V559D		T670E	D816A	D820Y
	V559A		T670I	D816E	N822D
	V560D			D816G	N822H
	L576P			D816H	N822K
				D816V	N822Y
				D820A	Y823D
				D820E	A829P
				D820G	

Only the five most common *KIT* exon 11 primary mutations, of the >100 observed in patients,[1] are included in the BEAMing assay. Mutations highlighted in red were detected in at least one patient sample.
Abbreviation: ATP, adenosine triphosphate; BEAMing, beads, emulsions, amplification, and magnetics.

Reference

1. KIT Gene, Catalogue Of Somatic Mutations In Cancer (COSMIC), 2019. Available at: https://cancer.sanger.ac.uk/cosmic/gene/analysis?ln=KIT

Supplementary Table S2. Patient Disposition

Disposition	*KIT* exon 11-positive n = 30, n (%)	*KIT* exon 11-negative n = 15, n (%)	Total N = 45, n (%)
Discontinued from treatment			
Died on/before clinical cutoff date	22 (73)	10 (67)	32 (71)
Alive within 12–24 weeks before database cutoff date	0	1 (7)	1 (2)
Alive >24 weeks before database cutoff date	8 (27)	3 (20)	11 (24)
Withdrawn consent	0	1 (7)	1 (2)
Primary reason for treatment discontinuation			
Progressive disease (RECIST-defined and clinical progression)	15 (50)	10 (67)	25 (56)
Adverse event	6 (20)	2 (13)	8 (18)
Withdrawal by patient	4 (13)	1 (7)	5 (11)
Physician decision	3 (10)	0	3 (7)
Study terminated by sponsor	0	2 (13)	2 (4)
Other[a]	2 (7)[a]	0	2 (4)[a]

[a]Two patients were discontinued by the FDA during the clinical hold because they had not already responded to ponatinib and were required to be treated with all other approved tyrosine kinase inhibitors for GIST.

GIST, gastrointestinal stromal tumors; RECIST, Response Evaluation Criteria in Solid Tumors.

Supplementary Table S3. Plasma Sample Collection

	Patients With Samples Analyzed, n
Total patients with plasma samples	37 (105 samples analyzed)
Patients with BL sample only	9
Patients with BL and PBL samples	28
Documented disease progression	14 (59 samples analyzed)
No documented disease progression	14
Discontinued (BL, first-PBL, and/or EOT samples analyzed)	10 (25 samples analyzed)
Ongoing (BL, first-PBL, and latest visit samples analyzed)	4 (12 samples analyzed)

BL, baseline; EOT, end of treatment; PBL, postbaseline.

Supplementary Table S4. Mutational Status at Baseline and Clinical Benefit Rate at 16 Weeks

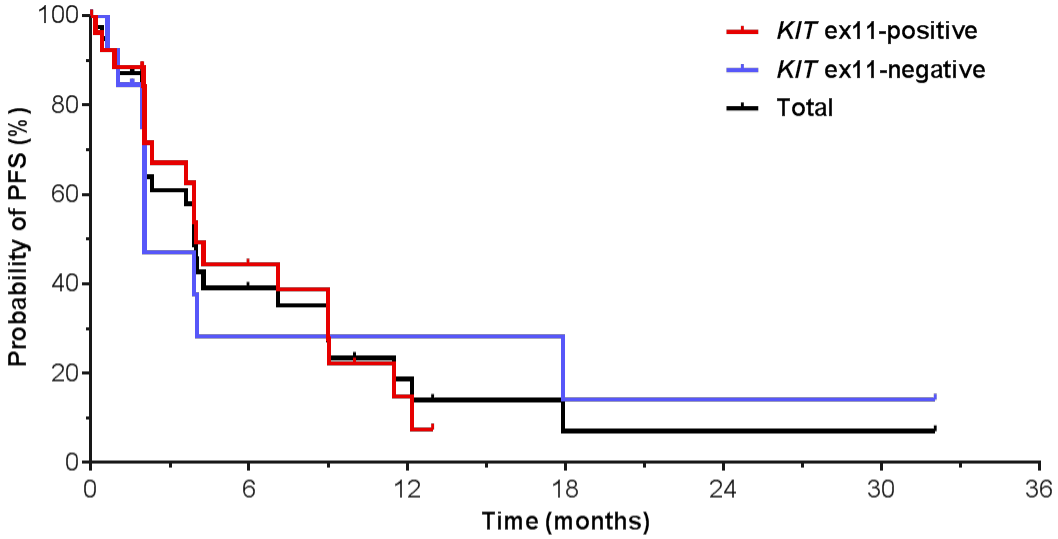
	***KIT* exon 11-positive (n = 24[a])**	***KIT* exon 11-negative (n = 13[a])**	
		***KIT* mutation[b] (n = 8)**	***KIT* WT (n = 5)**
Baseline plasma mutation status in patients, n (%)			
No mutation	6 (25)	2 (25)	5 (100)
Primary mutation only	4 (17)	5 (62)	0
Secondary mutations	14 (58)[c]	1 (13)	0
Exon 17/18 mutations	14 (58)	1 (13)	0
Exon 13/14 mutations	2 (8)[d]	0	0
CBR at 16 weeks, % (n/n):			
Overall CBR[e]	42% (10/24)	14% (1/8)	40% (2/5)
[95% CI]	[22–63]	[3–53]	[5–85]
CBR of patients with:			
Exon 17/18 mutations	43% (6/14)	50% (1/2)[f]	ND
[95% CI]	[18–71]	[13–99]	
No exon 17/18 mutants	40% (4/10)	0% (0/6)	40% (2/5)
[95% CI]	[12–74]	[0–46]	[5–85]

Data are as of March 2, 2015.

Abbreviations: CBR, clinical benefit rate; CI, confidence interval; ND, mutation not detected; WT, wild type.

[a]Analysis is based on 37 patients with baseline plasma samples analyzed; cohort assignment is based on historical mutation data. [b]*KIT* exon 9, n = 6; *KIT* exon 13 (K642E), n = 1; *KIT* exon 17 (D820Y), n = 1. [c]Includes four patients who also had a primary mutation detected at baseline. [d]Two patients had an exon 17/18 mutation in addition to an exon 13/14 mutation. [e]Overall CBR of trial: 36% (10/28) cohort A and 20% (3/15) cohort B; 4/4 cohort A and 2/2 cohort B patients who did not have baseline plasma samples available did not achieve CBR. [f]The responding patient had an exon 17 mutation (D820Y), which was documented as the primary mutation in the patient's tumor.

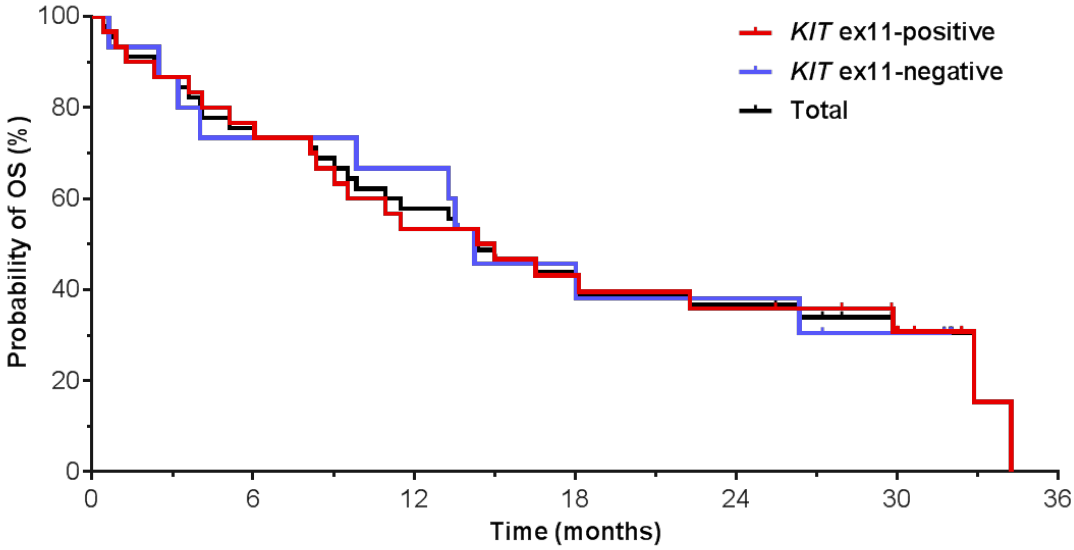
Supplementary Fig. S1a. Progression-free survival (PFS) in the *KIT* exon 11-positive and *KIT* exon 11-negative cohorts and in the total patient population.



No. at risk												
KIT ex11-positive	30	14	8	7	1	0	0	0	0	0	0	0
KIT ex11-negative	15	5	2	2	2	2	1	1	1	1	1	0
Total	45	9	10	9	3	2	1	1	1	1	1	0

	No. (%) of Events	Median PFS, months (range)	PFS Probability, % (95% CI)	
			6 Months	12 months
KIT ex11-positive (n = 30)	19 (63)	4.0 (0.04–13.0)	44 (24–63)	15 (3–35)
KIT ex11-negative (n = 15)	9 (60)	2.0 (0.04–32.0)	28 (7–55)	28 (7–55)
Total (n = 45)	28 (62)	3.9 (0.04–32.0)	39 (23–55)	19 (7–35)

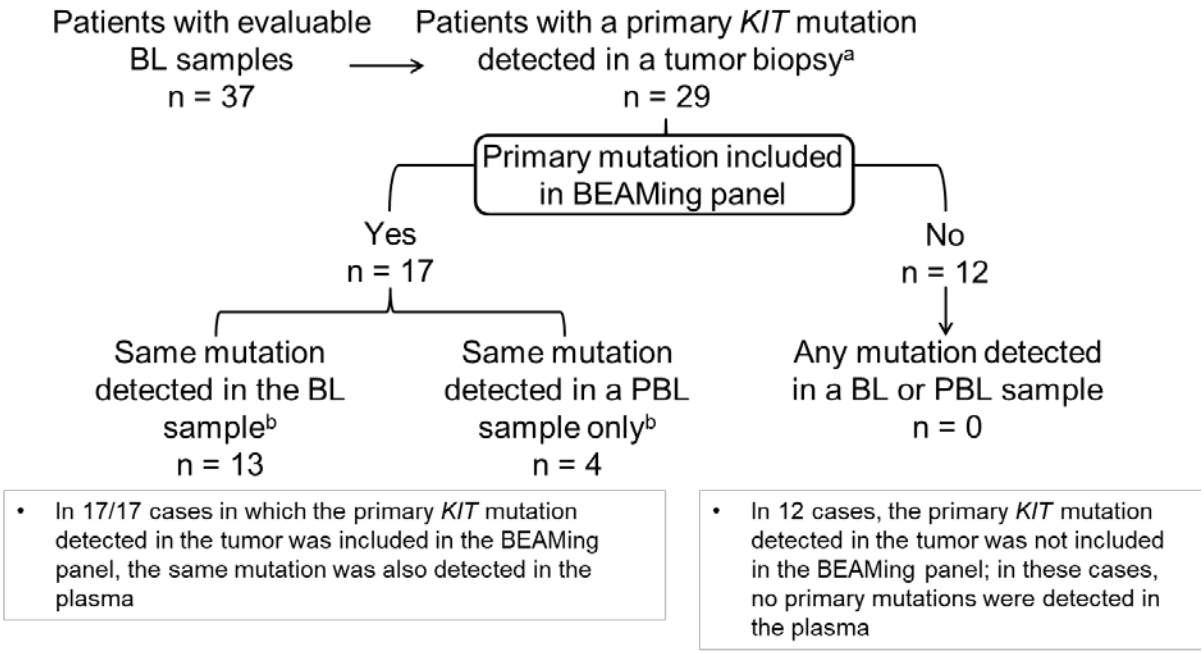
Supplementary Fig. S1b. Overall survival in the *KIT* exon 11-positive, *KIT* exon 11-negative cohorts and in the total patient population.



No. at risk												
KIT ex11-positive	30	25	22	20	16	15	11	11	10	9	5	2
KIT ex11-negative	15	12	11	11	10	6	5	5	5	3	3	0
Total	45	37	33	31	26	21	16	16	15	12	8	2

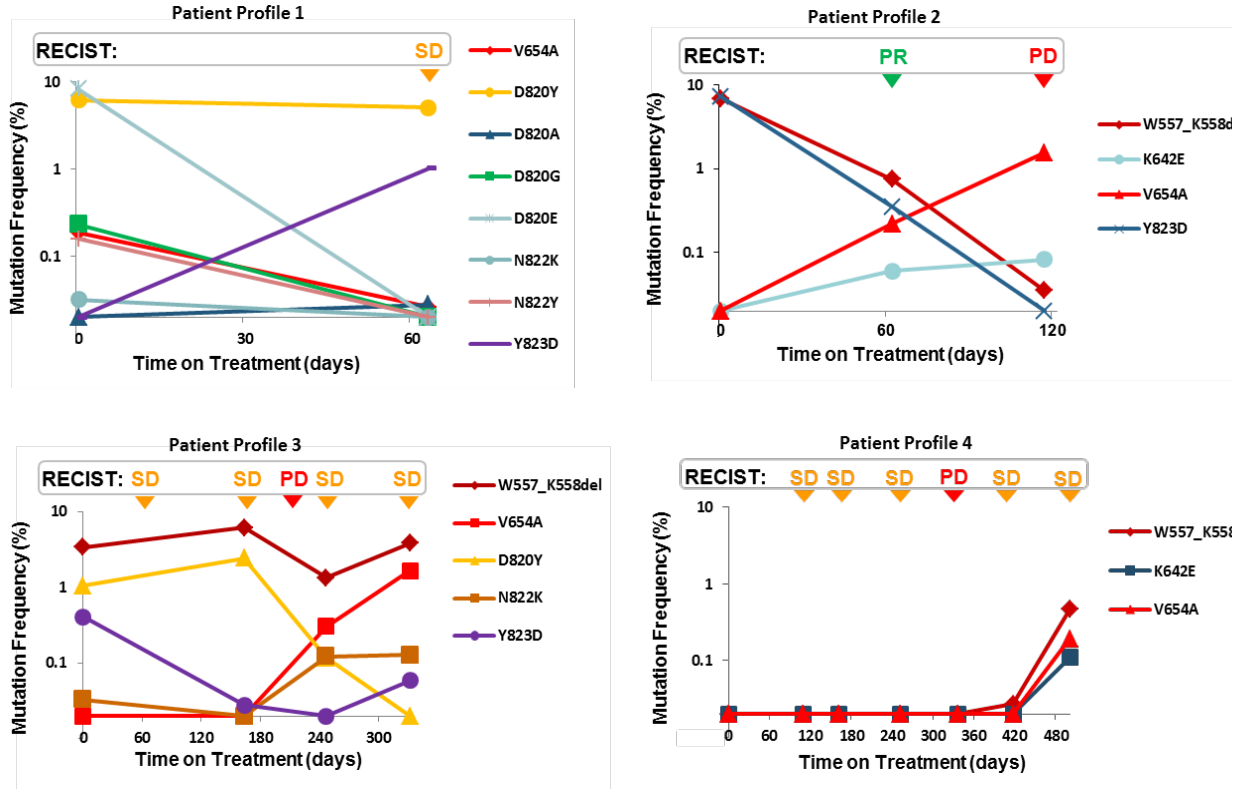
	No. (%) of Events	Median Survival, months (range)	OS Probability, % (95% CI)	
			6 Months	12 months
KIT ex11-positive (n = 30)	22 (73)	14.7 (0.4–34.3)	77 (57–88)	53 (34–69)
KIT ex11-negative (n = 15)	10 (67)	14.3 (0.6 – 32.0)	73 (44–89)	67 (38–85)
Total (n = 45)	32 (71)	14.4 (0.4–34.3)	76 (60–86)	58 (42–71)

Supplementary Fig. S2. Concordance of primary mutations detected in tumor and plasma BEAMing.



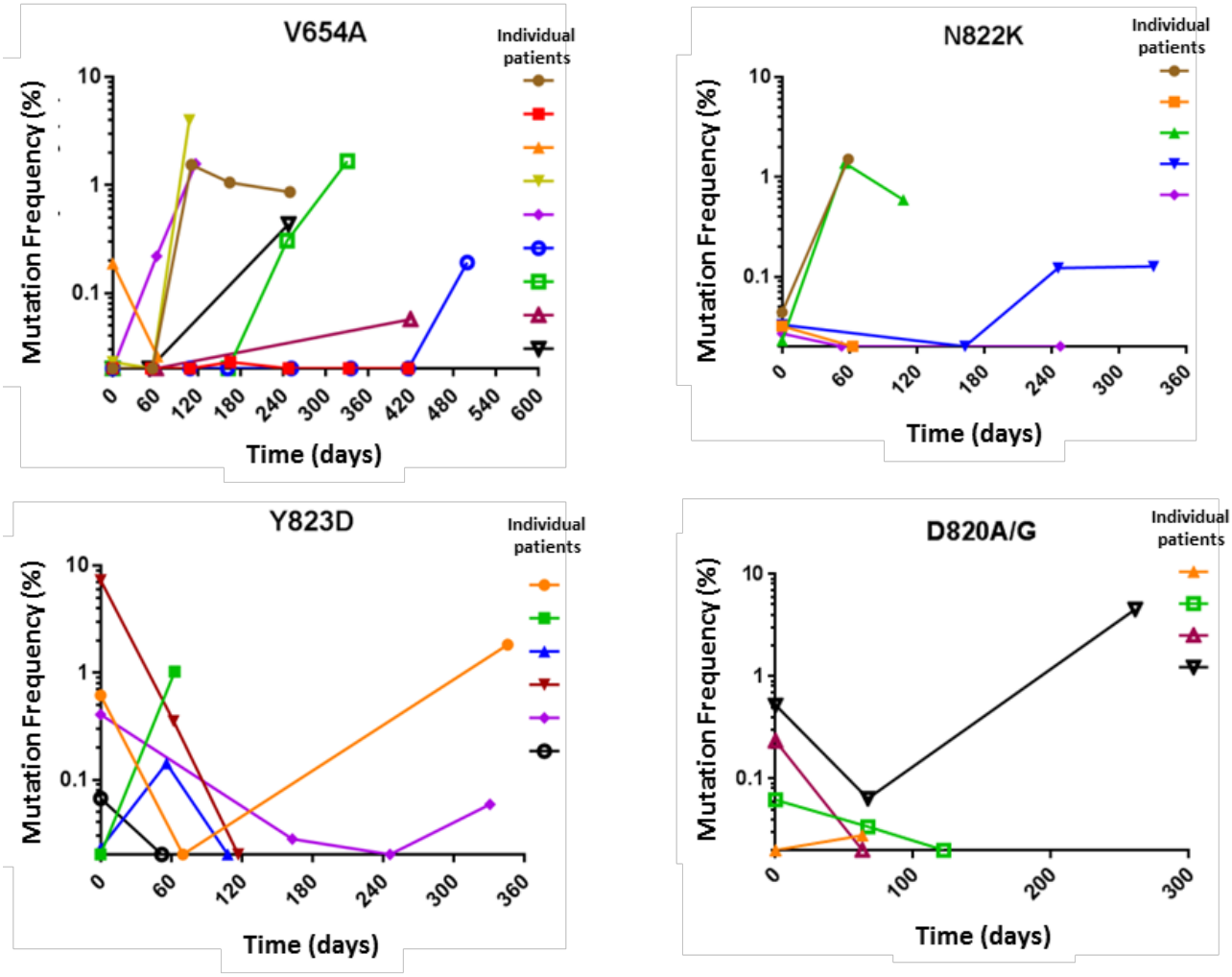
[a]As reported by investigators based on historical analyses; three patients had no mutation history and five had wild-type *KIT*.
[b]Four patients had an additional primary mutant detected in a baseline or post-baseline plasma sample (L576P, V559A, or K642E [twice]) that was not documented in the patient's history.
Abbreviation: BEAMing, beads, emulsions, amplification, and magnetics.

Supplementary Fig. S3. Mutation dynamics in select individual patients.



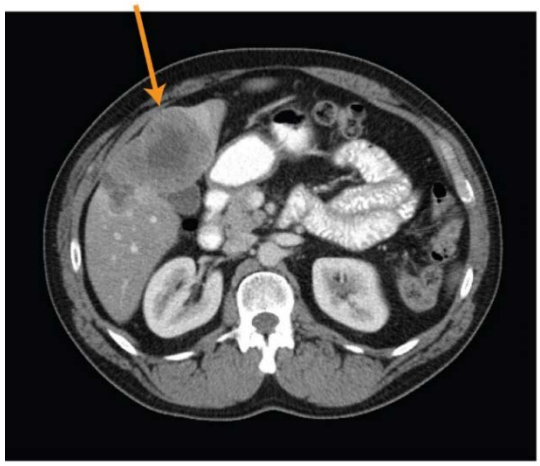
Abbreviation: RECIST, Response Evaluation Criteria In Solid Tumors.

Supplementary Fig. S4. Dynamics of mutations detected in at least four patients.



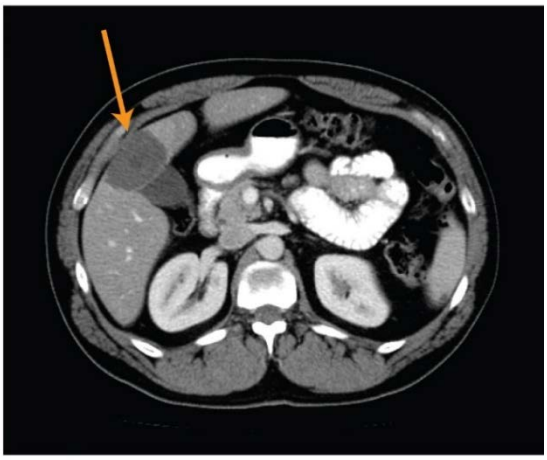
Supplementary Fig. S5. Computer tomography scan and liver biopsy images of a patient treated with ponatinib who had previously progressed on two tyrosine kinase inhibitors. (A) Computed tomography scan at baseline. (B) Computed tomography scan on Cycle 3 Day 1 of treatment. Arrows in Figures A and D indicate the presence of tumor. Liver biopsy at baseline: (C) At low magnification and (E) At high magnification. Arrows in Figure E indicate the presence of mitotic figures. Liver biopsy on Cycle 1 Day 19 of treatment: (D) At low magnification and (F) At high magnification. PET, positron emission tomography. Images courtesy of Leona A. Doyle, MD.

A.



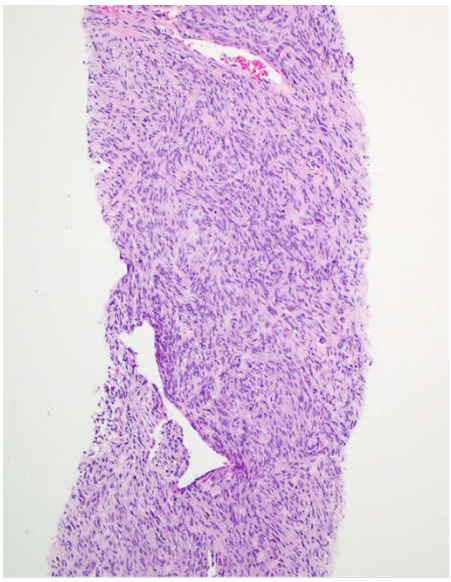
Baseline

B.



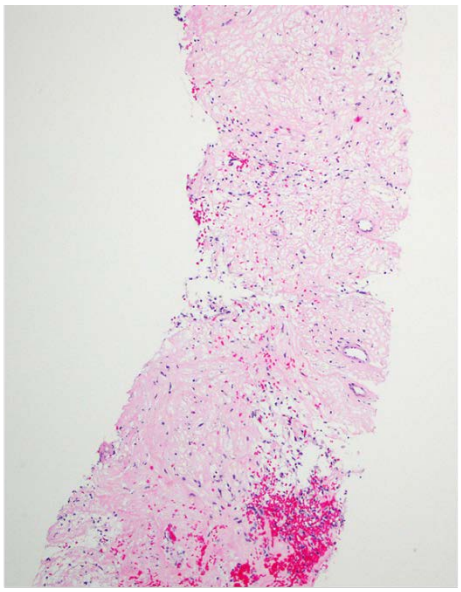
Cycle 3, Day 1 of Ponatinib

C.



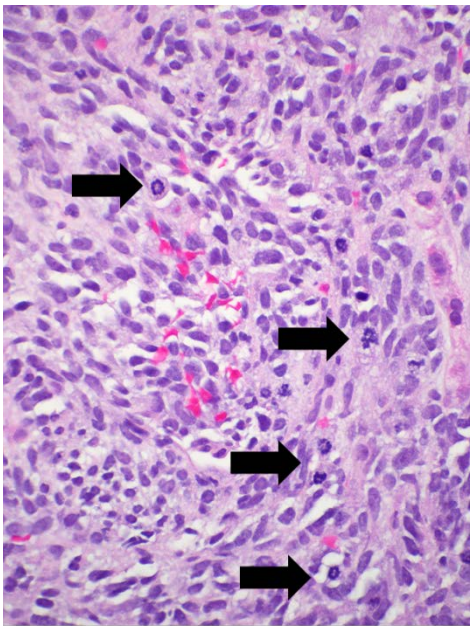
Liver biopsy, baseline

D.



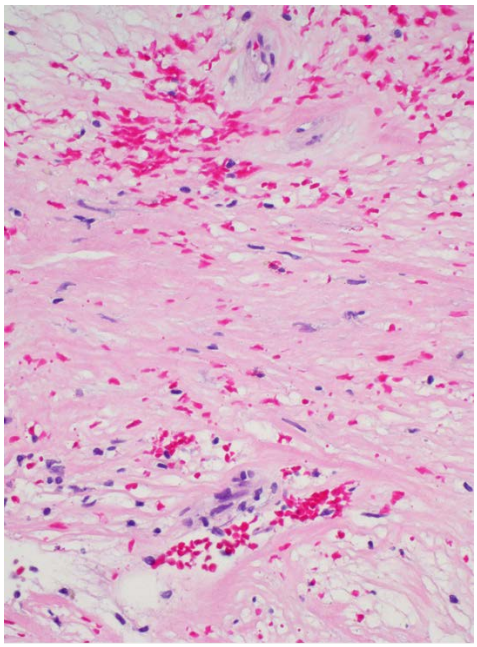
Liver biopsy, cycle 1, day 19
(2 days post-PET)

E.



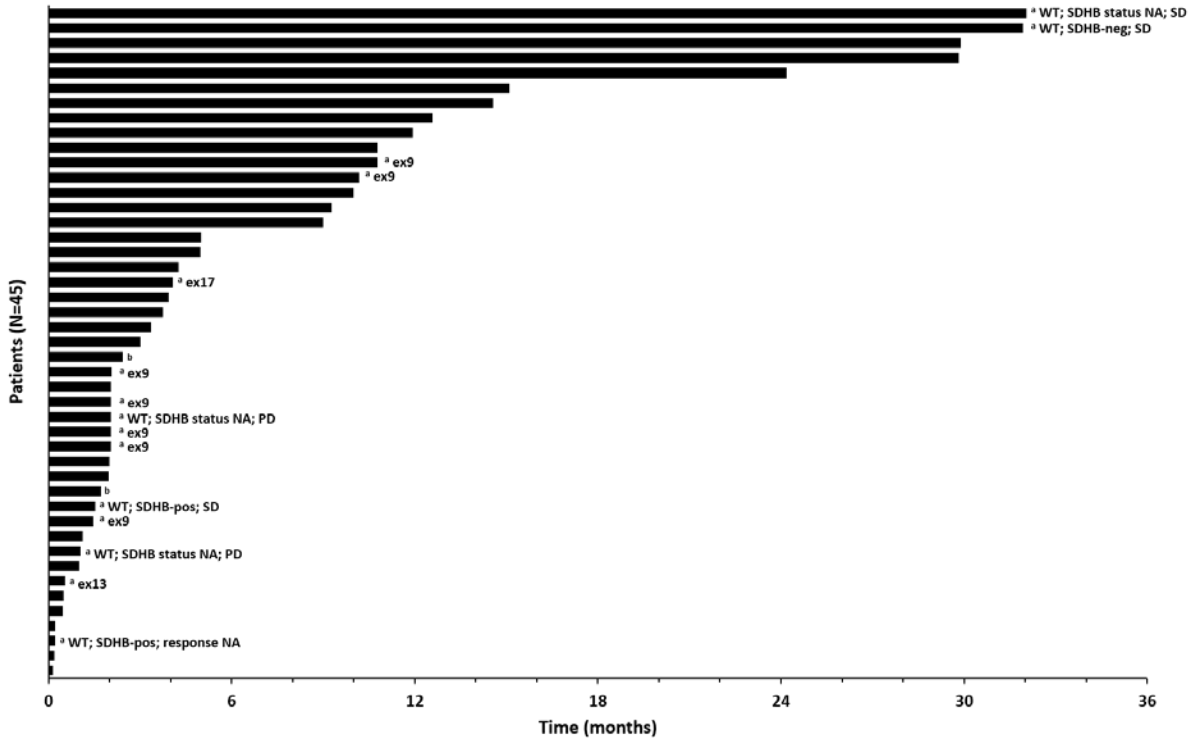
Liver biopsy, baseline

F.



Liver biopsy, cycle 1, day 19
(2 days post-PET)

Supplementary Fig. S6. Time on treatment for individual patients.



ex9, *KIT* exon 9-positive patients; ex13, *KIT* exon 13-positive patients; ex17, *KIT* exon 17-positive patients; NA, not available; neg, negative; PD, progressive disease; pos, positive; SD, stable disease; SDHB, succinate dehydrogenase complex iron sulfur subunit B; WT, *KIT* wild-type patients.

[a]*KIT* ex11-negative patients.

[b]Patients who were discontinued from study per US Food and Drug Administration request.