RESEARCH ARTICLE

Long-term bosutinib for chronic phase chronic myeloid leukemia after failure of imatinib plus dasatinib and/or nilotinib

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Bosutinib is an Src/Abl tyrosine kinase inhibitor (TKI) indicated for adults with Ph+ chronic myeloid leukemia (CML) resistant/intolerant to prior TKIs. This long-term update of an ongoing phase 1/2 study evaluated the efficacy and safety of third-/fourth-line bosutinib in adults with chronic phase (CP) CML. Median durations of treatment and follow-up were 8.6 (range, 0.2–87.7) months and 32.7 (0.3–93.3) months, respectively. Cumulative confirmed complete hematologic response (cCHR) and major cytogenetic response (MCyR) rates were 74% (95% CI, 65–81%) and 40% (31–50%), respectively; Kaplan–Meier (K–M) probability of maintaining cCHR or MCyR at 4 years was 63% (95% CI, 50–73%) and 69% (52–81%). Cumulative incidence of on-treatment disease progression (PD)/death at 4 years was 24% (95% CI, 17–33%); K–M 4-year overall survival was 78% (68–85%). Baseline Ph+ cells ≤35 vs. ≥95% was prognostic of MCyR and CCyR by 3 and 6 months, increased baseline basophils was prognostic of PD/death, and no prior response to second-line TKI was prognostic of death. Common adverse events included diarrhea (83%), nausea (48%), vomiting (38%), and thrombocytopenia (39%). Bosutinib demonstrates durable efficacy and a toxicity profile similar to previous bosutinib studies in CP CML patients resistant/intolerant to multiple TKIs, representing an important treatment option for patients in this setting. This trial is registered at www.clinicaltrials.gov (NCT00261846).
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Introduction

Tyrosine kinase inhibitors (TKIs) are the standard of care for patients with chronic myeloid leukemia (CML) [1]. Although many patients are successfully treated with imatinib [2–4] or the second-generation TKIs dasatinib [5,6] or nilotinib [7,8], some patients develop resistance or intolerance and require alternate therapy [9–11].

Bosutinib (SKI-606), an oral, dual Src/Abl TKI, has demonstrated efficacy in prospective clinical trials as second-, third-, and fourth-line therapy in Philadelphia chromosome–positive (Ph+) CML patients with treatment failure on prior TKIs [12–14] and is effective against most BCR–ABL1 mutations conferring resistance to TKIs, except T315I and V299L [12,15]. The toxicity profile of bosutinib differs from those of other TKIs, which may relate to its minimal activity against c-KIT and platelet-derived growth factor receptor, targets of imatinib, dasatinib, and nilotinib that could be associated with certain toxicities (e.g., fluid retention and bleeding disorders) [16–20]. The most common adverse event (AE) reported with bosutinib is diarrhea, which is typically transient (1–3 days/event) and easily managed [21,22].

This report describes the long-term (≥48 months) efficacy and safety of third- and fourth-line bosutinib therapy in an ongoing phase 1/2 trial in patients with CP CML resistant/intolerant to imatinib plus dasatinib and/or nilotinib. Exploratory analyses assessing baseline predictors of long-term outcomes are also reported.

Methods

Patients and study design. This analysis includes adults (≥18 years) enrolled prospectively in an ongoing 2-part, phase 1/2 study [12,14] with a confirmed diagnosis of Ph+ CP CML who had received imatinib followed by dasatinib and/or nilotinib. Additional eligibility criteria are described in the Supporting Information.

The present analysis was based on patients with CP CML that was imatinib-resistant (≥600 mg/day) or imatinib-intolerant (any dose) plus ≥1 of the following: resistant to dasatinib ≥100 mg/day (IM + D − R), intolerant to any dose of dasatinib (IM + D − I), resistant to nilotinib 800 mg/day (IM + N − R), or intolerant to any dose of nilotinib or resistant/intolerant to dasatinib and nilotinib (IM + N ± D). Dose escalation to bosutinib 600 mg/day was allowed for lack of efficacy (no complete hematologic response

Additional Supporting Information may be found in the online version of this article.

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[CHR] by week 8 or no complete cytogenetic response [CCyR] by week 12) unless treatment-related grade ≥3 AEs occurred. Bosutinib treatment continued until disease progression/death, unacceptable toxicity, or withdrawal of consent.

The study protocol was approved by each sites' ethics board and conducted in accordance with the principles of Good Clinical Practice and the Declaration of Helsinki.

Assessments. Responses were assessed as described previously [12,14]. Hematologic response was defined as achievement of a confirmed CHR (cCHR) or a baseline cCHR that was maintained for ≥5 weeks. Cytogenetic response was defined as one newly achieved during treatment or, if present at baseline, maintained for ≥4 weeks. Evaluable patients received ≥1 dose of bosutinib and had a valid baseline assessment for the respective endpoint. Duration of response (DOR) was evaluated among responders from the first response date until confirmed loss of response, treatment discontinuation due to progressive disease (PD)/death, or death within 30 days after last dose; patients without events were censored at their last assessment visit.

Disease progression was defined as described previously [12,14]. Time from first dose to (1) PD/death and (2) transformation to accelerated phase (AP)/blast phase (BP) CML were evaluated through 30 days after last dose; patients without events were censored at their last assessment visit. Overall survival (OS) was evaluated for up to 2 years after treatment discontinuation (per protocol) and included data from patients enrolled in an ongoing extension study (Clinicaltrials.gov ID: NCT01903733) [23]; patients not deceased were censored at the last known alive date.

The safety analysis included all patients who received ≥1 bosutinib dose. AEs were reported up to 30 days after the last dose and graded according to National Cancer Institute Common Terminology Criteria for Adverse Events Version 3.0. Treatment-emergent AEs (TEAEs) were assessed overall and by year of first occurrence.

Statistical analyses. Time-to-event distributions and probabilities were estimated using the Kaplan−Meier (K–M) method (DOR, OS) or cumulative incidence adjusting for the competing risk of treatment discontinuation without the event (response, PD/death, transformation). Two-sided 95% confidence intervals (CIs) for response rates and K–M quartiles were based on the exact binomial and Brookmeyer–Crowley linear transformation method, respectively; 2-sided 95% CIs for K–M and cumulative incidence yearly probability estimates were based on Greenwood's formula and Gray's method, respectively.

Retrospective backward elimination (criteria: $P = 0.20$) multivariable analyses evaluated baseline characteristics as predictors of (1) major cytogenetic response (MCyR) or CCyR by 3 or 6 months or cumulatively using logistic regression, (2) progression-free survival (PFS) distribution (truncated at 4 years) using a Cox proportional, cause-specific hazard model, (3) OS distribution using a Cox proportional hazard model, and (4) time to first diarrhea AE (grade 3/4) or liver-related AEs (any grade and grade 3/4) using a Cox proportional cause-specific hazard model. Results are presented as odds and hazard ratios (95% CI); *P* values were not adjusted for multiple testing.

Results

Patients

This analysis included 119 patients (IM + D − R, $n = 38$; IM + D − I, $n = 50$; IM + N − R, $n = 26$; IM + N ± D, $n = 5$; Supporting Information Table I). The time from the last patient's first dose to data cutoff was 50.2 months. As of the data snapshot (May 23, 2014) based on an unlocked trial database for this interim publication, 29 (24%) patients in the phase 1/2 study were still receiving bosutinib at the 4-year follow-up (IM + D − R, $n = 4$; IM + D − I, $n = 16$; IM + N − R, $n = 8$; IM + N ± D, $n = 1$; 1 year = 48 weeks). The median (range) follow-up duration for all patients was 32.7 (0.3–93.3) months. The median (range) treatment duration was 8.6 (0.2–87.7) months overall and 60.1 (45.8 − 87.7) months for patients still on treatment at year 4 (Supporting Information Table II). Ninety (76%) patients discontinued treatment by year 4, with the most common primary reasons being AEs (28 [24%]), PD (24 [20%]), and lack of efficacy (22 [18%]). Sixty (50%) patients received dose reductions due to AEs. Twenty-two (18%) patients escalated to 600 mg/day for lack of efficacy, 2 of whom were still on therapy and receiving 600 mg/day at the data snapshot (33.7 and 41.1 months on-treatment with 600 mg/day).

Compared with patients aged <40 or 40 − 64 years, those aged ≥65 years were more frequently intolerant to prior imatinib, more often had worse Eastern Cooperative Oncology Group performance scores, and more frequently had medical history events of hypertension, pleural effusion, peripheral edema, osteoarthritis, and tonsillectomy (Supporting Information Table III). The median (range) CML duration since diagnosis for patients aged ≥65, 40 − 64, and <40 years, respectively, was 7.3 (1.7–18.3), 6.8 (0.6–17.6), and 3.9 (1.2–14.4) years. The median (range) follow-up duration was generally similar across age groups (≥65 years, 29.1 [0.3–80.9] months; 40–64 years, 36.6 [2.5–93.3] months; <40 years, 30.5 [5.8–68.8] months); median (range) bosutinib treatment duration was as follows: ≥65 years, 6.6 (0.2–71.5) months; 40-64 years, 10.2 (0.3–87.7) months; <40 years, 7.4 (1.1–68.7) months. Compared with patients aged ≥65 or 40–64 years, those aged <40 years had lower rates of permanent treatment discontinuation due to AEs (≥65 years, 44%; 40–64 years, 20%; <40 years, 6%), dose reduction (≥65 years, 67%; 40–64 years, 49%; <40 years, 33%), and dose interruption due to AEs (≥65 years, 89%; 40–64 years, 62%; <40 years, 50%) within 4 years.

Efficacy

The 4-year cumulative cCHR rate (attained or maintained) was 74% (95% CI, 65–81%) in 117 evaluable patients (Supporting Information Table IV). Among 52 patients with a CHR at baseline, 45 (87%) maintained a cCHR on bosutinib; among 65 patients without a CHR at baseline, 41 (63%) newly attained a cCHR. Among 112 evaluable patients, the cumulative MCyR rate was 40% (95% CI, 31–50%) in patients who newly attained or maintained an MCyR from baseline for ≥4 weeks (newly attained, 33%; maintained, 7%), including 32% (95% CI, 24–42%) who attained/maintained a CCyR (newly attained, 26%; maintained, 6%; Supporting Information Table IV). Newly attained/maintained cytogenetic response rates were lower in older (≥65 years) vs. younger (<40 or 40–64 years) patients (Supporting Information Table V). K–M-estimated probabilities of maintaining a CHR, MCyR, or CCyR at 4 years remained high in the IM + D − I (70, 87, and 66%, respectively) and IM + N − R (62, 78, and 63%) cohorts but appeared lower in the IM + D − R cohort (57, 43, and 17%; Fig. 1 and Supporting Information Fig. 1). The median durations of CHR, MCyR, and CCyR were not reached prior to the minimum follow-up of 48 months.

Of the 53 patients with a dose reduction to 400 mg/day due to an AE, 21 attained/maintained MCyR: 15 (28%) first achieved MCyR after dose reduction, 4 (8%) achieved MCyR before and maintained it after dose reduction, and 2 (4%) lost MCyR after dose reduction. Of 22 patients with a dose reduction to 300 mg/day, 9 attained/maintained MCyR, including 4 (18%) first achieving an MCyR, 4 (18%) maintaining MCyR, and 1 (5%) losing MCyR after dose reduction, respectively (Supporting Information Table VI). Among patients with a dose reduction, the median cumulative number of days on 400 mg/day was smaller than with 300 mg/day (Supporting Information Table II). Median duration of attained/maintained MCyR (non-K–M) was slightly longer for patients with a dose reduction to 400 mg/day vs. 300 mg/day (21 days longer in patients with a first MCyR after dose reduction and 24 days longer in patients with an MCyR before and after dose reduction).

Of the 95/119 (80%) patients with known baseline mutational status, 39 (41%) had ≥1 BCR–ABL1 kinase domain mutation (IM + D − R, $n = 13/31$ [42%]; IM + D − I, $n = 9/36$ [25%]; IM + N − R, $n = 15/23$ [65%]; IM + N ± D, $n = 2/5$ [40%]); 11 patients had ≥2 mutations. A total of 20 unique BCR–ABL1 mutations were evident, with varying degrees of TKI sensitivity; 7 patients had the TKI-resistant T315I mutation. CHR and MCyR were observed across baseline BCR–ABL1 mutations, including several associated with TKI resistance [15] (Supporting Information Table VII). Among evaluable patients with ≥1 BCR–ABL1 mutation (excluding T315I), CHR and MCyR rates were 24/32 (75%) and 12/30 (40%), respectively. CHR and MCyR rates were similar in patients without mutations (77 and 37%, respectively) or with 1 mutation (including T315I; 75 and 38%) and lower in patients with ≥2 mutations (45 and 27%).

Of 57 patients evaluated for mutations before and during therapy (at disease progression and/or at treatment completion), 13 had ≥1

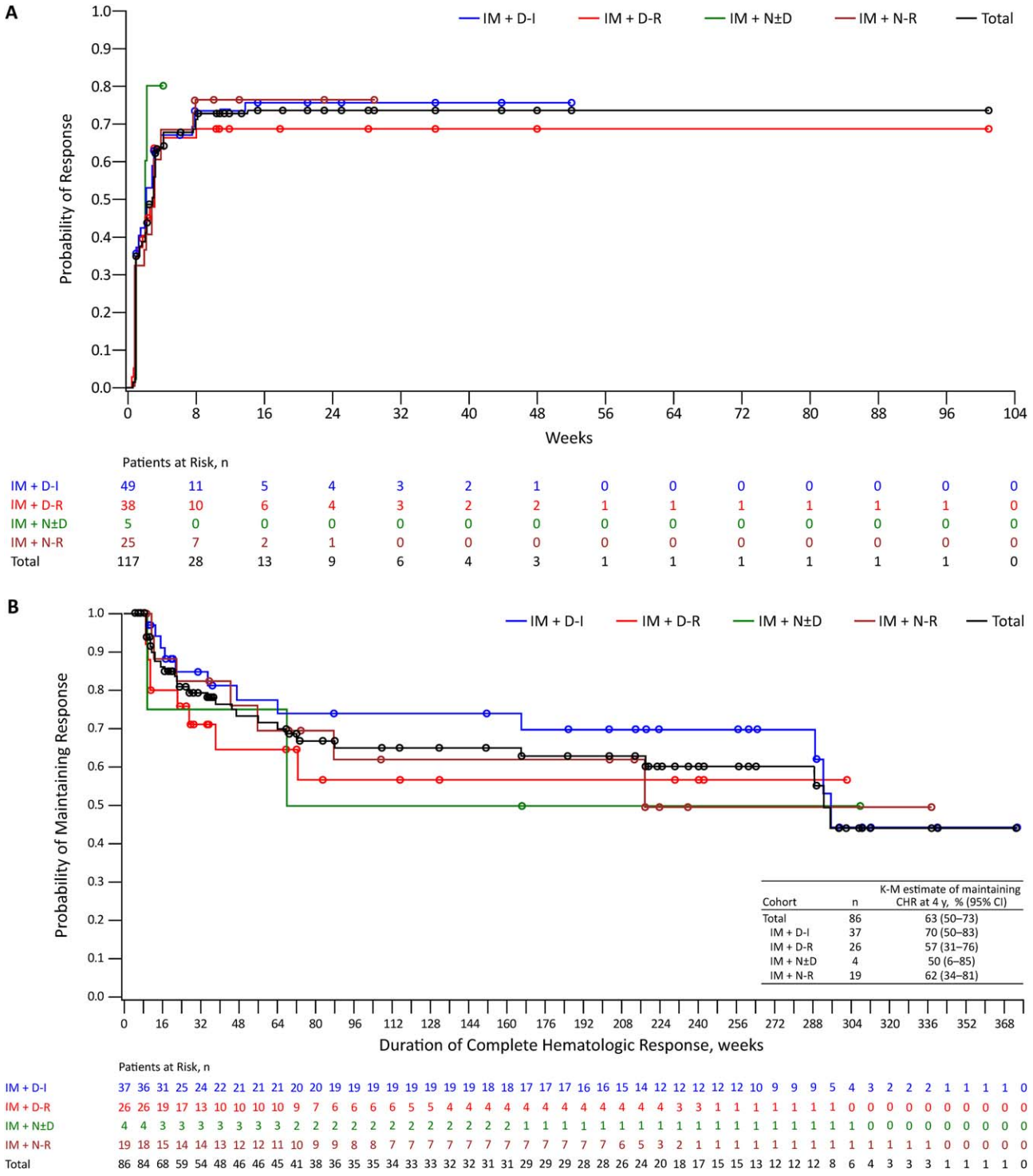

Figure 1. Cumulative incidence of response and duration of response. (A) Cumulative incidence of CHR adjusting for the competing risk of treatment discontinuation without the event; (B) duration of CHR among responders. (C) Cumulative incidence of MCyR adjusting for the competing risk of treatment discontinuation without the event; (D) duration of MCyR among responders. CHR = complete hematologic response; CI = confidence interval; D = dasatinib; I = intolerant; IM = imatinib; K–M = Kaplan–Meier; MCyR = major cytogenetic response; N = nilotinib; NE = not evaluable; R = resistant.

new BCR–ABL1 mutation emerge during bosutinib treatment, including V299L ($n = 6$), T315I ($n = 3$), G250E ($n = 2$), F359C, L248V, and L273M ($n = 1$ each); one IM + D − I patient with F317L at baseline had 2 emergent mutations (V299L and G250E). Twelve of these 13 patients discontinued treatment; 1 IM + N − R patient with a baseline Y253H mutation and an emergent L273M mutation continued therapy [23]. Reasons for discontinuation were lack of efficacy ($n = 7$), PD ($n = 4$), and AE (grade 3 neutropenia; $n = 1$). Seven of these 13 patients achieved only a CHR as a best response; the other 6 patients either achieved a CCyR ($n = 2$), a partial cytogenetic response ($n = 1$), a complete molecular response ($n = 1$; patient who entered the extension study), or no response ($n = 2$).

Long-term outcomes

The cumulative incidence of on-treatment AP transformation at 4 years was 4% (95% CI, 2–10%), including IM + D − R, 8% ($n = 3$;

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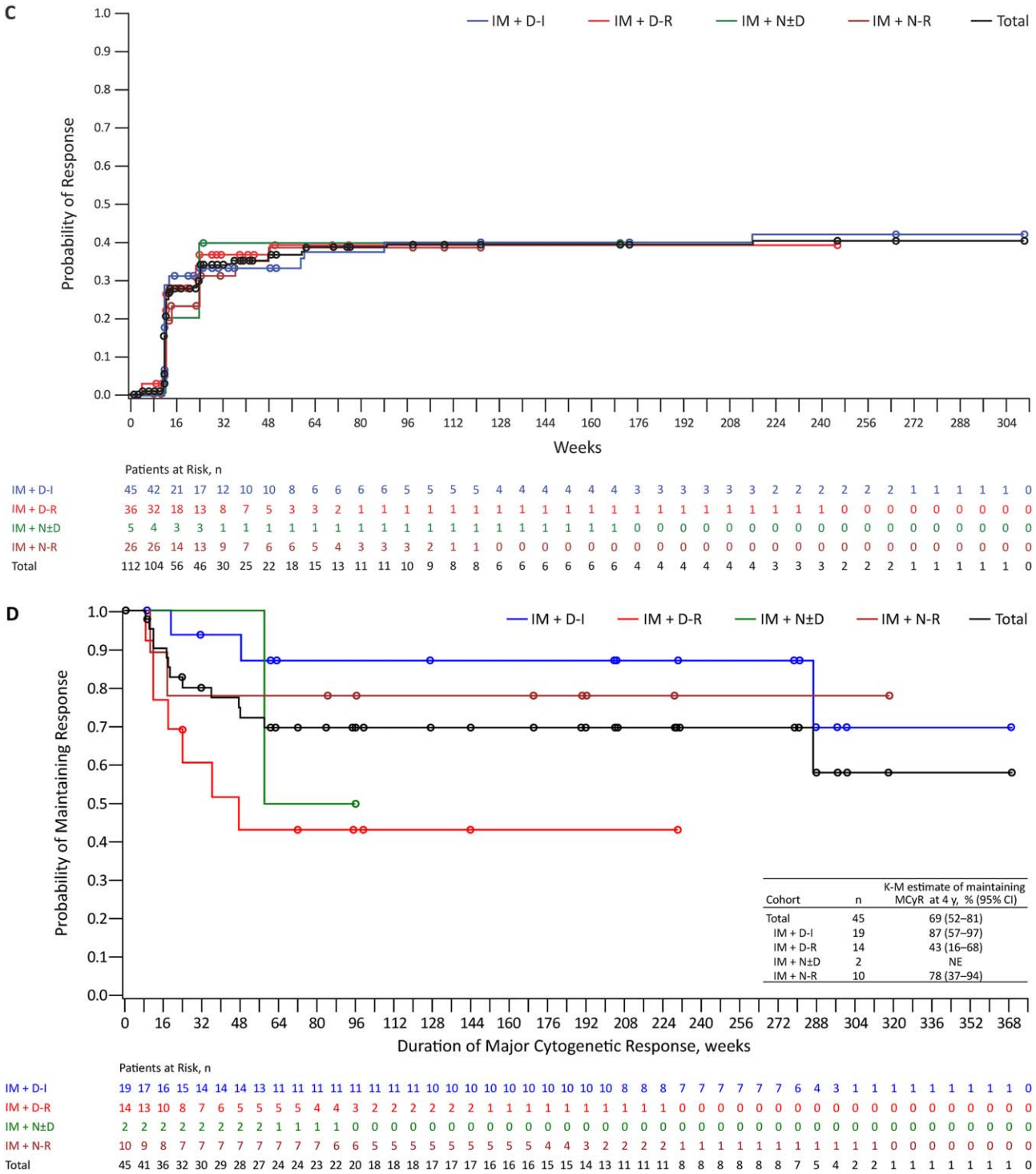

Figure 1. (Continued).

95% CI, 3–23%); IM + D − I, 0%; IM + N − R, 4% ($n = 1$; 95% CI, 1–26%); and IM + N ± D, 20% ($n = 1$; 95% CI, 4–100%). No BP transformations or new on-treatment transformations occurred after 2 years. Overall, 85 (71%) patients discontinued bosutinib before year 4 without transformation. Among the 5 patients progressing to AP, 1 IM + N − R patient had 6 temporary interruptions due to AEs (cumulative duration, 127 days) and dose reductions to 400 mg/day and to 300 mg/day due to AEs (cumulative durations of 28 and 211 days, respectively). One additional IM + N ± D patient had a 1-day treatment interruption due to AEs with no dose reductions. The IM + N − R patient was diagnosed with nonsmall cell lung cancer and discontinued treatment 7 days before documented AP transformation (428 days after first bosutinib dose).

At 4 years, the cumulative incidence of on-treatment PD (including AP/BP transformation) or death was 24% (95% CI, 17–33%) overall (IM + D − R, 24% [13–42%]; IM + D − I, 16% [9–30%]; IM + N − R, 35% [20–59%]; IM + N ± D, 40% [14–100%]); 54% of patients discontinued through year 4 without on-treatment PD or death (Supporting Information Fig. 2). The K–M-estimated 4-year OS was 78% (95% CI, 68–85%) overall (IM + D − R, 67% [95% CI, 45–81%]; IM + D − I, 80% [64–89%]; IM + N − R, 87% [66–96%]; IM + N ± D, 80% [20–97%]); approximately 48% of patients were

TABLE I. Baseline Predictors of Cytogenetic Response and Survival

	Cytogenetic response, OR (95% CI)[a]	
	MCyR	CCyR
Age ≥65 years ($n = 24$) vs. <65 years ($n = 87$)		
3 months		0.20 (0.02–2.30); $P = 0.1966$
6 months		**0.10 (0.01–0.92); $P = 0.0419$**
Cumulative		
Women ($n = 62$) vs. men ($n = 49$)		
3 months		
6 months	**0.25 (0.07–0.89); $P = 0.0319$**	
Cumulative	0.40 (0.14–1.12); $P = 0.0807$	0.39 (0.14–1.13); $P = 0.0825$
Prior IM response: yes ($n = 56$) vs. no ($n = 40$)[a]		
3 months	1.69 (0.34–8.34); $P = 0.5204$	
6 months	3.01 (0.80–11.42); $P = 0.1046$	
Cumulative	2.84 (0.92–8.76); $P = 0.0696$	
Prior D/N response: yes (64) vs. no ($n = 30$)		
3 months	0.87 (0.13–5.59); $P = 0.8806$	
6 months	0.64 (0.17–2.45); $P = 0.5115$	
Cumulative	1.20 (0.37–3.83); $P = 0.7641$	3.75 (0.94–14.94); $P = 0.0606$
% Ph+ cells: <95% to >35% ($n = 17$) vs. ≤35% ($n = 20$)[b]		
3 months	**0.16 (0.03–0.98); $P = 0.0475$**	**0.023 (<0.01–0.27); $P = 0.0028$**
6 months	**0.11 (0.02–0.76); $P = 0.0258$**	**0.03 (<0.01–0.23); $P = 0.0005$**
Cumulative	0.19 (0.03–1.15); $P = 0.0711$	**0.14 (0.03–0.73); $P = 0.0201$**
% Ph+ cells: ≥95% ($n = 66$) vs. ≤35% ($n = 20$)[b]		
3 months	**0.02 (<0.01–0.14); $P < 0.0001$**	**<0.01 (<0.01–0.05); $P < 0.0001$**
6 months	**0.01 (<0.01–0.09); $P < 0.0001$**	**<0.01 (<0.01–0.04); $P < 0.0001$**
Cumulative	**0.04 (<0.01–0.21); $P = 0.0002$**	**0.04 (0.01–0.19); $P < 0.0001$**
% Ph+ cells: unknown ($n = 8$) vs. ≤35% ($n = 20$)[b]		
3 months	**0.02 (<0.01–0.51); $P = 0.0176$**	**0.04 (<0.01–0.78); $P = 0.0337$**
6 months	**0.01 (<0.01–0.27); $P = 0.0054$**	**0.03 (<0.01–0.38); $P = 0.0068$**
Cumulative	**0.08 (<0.01–0.87); $P = 0.0381$**	0.14 (0.02–1.19); $P = 0.0718$
Prior resistance to last TKI: yes ($n = 63$) vs. no ($n = 48$)		
3 months		
6 months		
Cumulative		
IFNα treatment before IM or diagnosis to IM initiation ≥6 months [$n = 39$] vs. <6 months [$n = 72$]		
3 months	0.13 (0.02–1.03); $P = 0.0529$	**0.08 (<0.01–0.72); $P = 0.0244$**
6 months	**0.09 (0.01–0.63); $P = 0.0156$**	
Cumulative	0.24 (0.05–1.07); $P = 0.0608$	
Prior IFNα: yes ($n = 63$) vs. no ($n = 48$)		
3 months	7.17 (0.91–56.40); $P = 0.0611$	
6 months	**18.4 (2.36–142.91); $P = 0.0054$**	
Cumulative	**5.59 (1.24–25.22); $P = 0.0253$**	
BCR–ABL1 mutation status: sensitive mutation ($n = 6$) vs. no mutation ($n = 54$)[c]		
3 months	11.92 (0.99–142.90); $P = 0.0506$	
6 months		
Cumulative		
BCR–ABL1 mutation status: insensitive mutation ($n = 23$) vs. no mutation ($n = 54$)[c]		
3 months	0.44 (0.05–4.22); $P = 0.4768$	
6 months		
Cumulative		
BCR–ABL1 mutation status: unknown sensitivity mutation ($n = 8$) vs. no mutation ($n = 54$)[c]		
3 months	0.16 (<0.01–6.71); $P = 0.3393$	
6 months		
Cumulative		
BCR–ABL1 mutation status: unknown/missing mutation ($n = 20$) vs. no mutation ($n = 54$)[c]		
3 months	2.48 (0.48–12.93); $P = 0.2807$	
6 months		
Cumulative		
Disease duration (years)		
3 months		
6 months		
Cumulative		
Basophils, %		
3 months	1.08 (0.96–1.22); $P = 0.1991$	
6 months		
Cumulative		

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	PFS and OS, HR (95% CI)[a]	
	PD/Death	Death
Age ≥65 years ($n = 26$) vs. <65 years ($n = 91$)		2.18 (0.95–5.01); $P = 0.0673$
Women ($n = 64$) vs. men ($n = 53$)		
Prior IM response: yes ($n = 58$) vs. no ($n = 42$)[a]	0.95 (0.41–2.23); $P = 0.9086$	0.94 (0.36–2.48); $P = 0.9057$
Prior D/N response: yes (68) vs. no ($n = 31$)		**0.34 (0.14–0.85); $P = 0.0203$**
% Ph+ cells: <95% to >35% ($n = 18$) vs. ≤35% ($n = 22$)[b]		
% Ph+ cells: ≥95% ($n = 66$) vs. ≤35% ($n = 22$)[b]		
% Ph+ cells: unknown ($n = 11$) vs. ≤35% ($n = 22$)[b]		
Resistance to last TKI: yes ($n = 65$) vs. no ($n = 52$)	2.11 (0.93–4.78); $P = 0.0738$	
IFNα treatment before IM or diagnosis to IM initiation ≥6 months ($n = 74$) vs. <6 months ($n = 43$)		
Prior IFNα: yes ($n = 65$) vs. no ($n = 52$)		
BCR–ABL1 mutation status: sensitive mutation ($n = 6$) vs. no mutation ($n = 56$)[c]		
BCR–ABL1 mutation status: insensitive mutation ($n = 25$) vs. no mutation ($n = 56$)[c]		
BCR–ABL1 mutation status: unknown sensitivity mutation ($n = 8$) vs. no mutation ($n = 56$)[c]		
BCR–ABL1 mutation status: unknown/missing mutation ($n = 22$) vs. no mutation ($n = 56$)[c]		
Disease duration (years)		
Basophils (%)	**1.10 (1.04–1.16); $P = 0.0012$**	

Shading corresponds to parameters failing to meet elimination criteria ($P < 0.20$; not shown); significant predictors are bolded. Odds ratios and hazard ratios >1 indicate worse outcome. *P* values were not adjusted for multiple comparisons.
BCR–ABL1 = Breakpoint Cluster Region protein/Abelson tyrosine-protein kinase 1; CCyR = complete cytogenetic response; CML = chronic myeloid leukemia; CP = chronic phase; D = dasatinib; FISH = fluorescence *in situ* hybridization; HR = hazard ratio; IFNα = interferon alpha; IM = imatinib; MCyR = major cytogenetic response; N = nilotinib; OR = odds ratio; OS = overall survival; PD = progressive disease; PFS = progression-free survival; Ph+ = Philadelphia chromosome–positive; TKI = tyrosine kinase inhibitor; Unk = unknown.
[a] Prior response was defined as achievement of at least a minimal cytogenetic response (standard cytogenetic criteria: 66% to 95% Ph+ cells from bone marrow or BCR–ABL1 fusion product from FISH).
[b] Required ≥20 metaphases for standard cytogenetics or ≥200 cells for FISH.
[c] Bosutinib-sensitive mutations are those resulting in half maximal inhibitory concentration (IC_{50}) ≤2-fold higher than wild type (M244V, Q252H, Y253H/F, D276G, E279K, E292L, M343T, M351T, F359V, L384M, H396P/R, and G398R) and bosutinib-insensitive mutations are those resulting in IC_{50} values >2-fold higher than wild type (L248R/V, G250E, E255K/V, V299L, T315A/I/V, F317L/R/V, F359I, and F486S); the sensitivity of all other mutations is unknown. If patients had >1 mutation with different sensitivities, they were categorized based on the following hierarchy: bosutinib-insensitive, unknown sensitivity, and bosutinib-sensitive [15,28]. The effect of (1) insensitive mutations was not estimable because there were no CCyRs by month 3 in the insensitive mutation groups, and (2) prior response to D/N was not estimable because there were no CCyRs in the no prior response to D/N group. Two patients with missing values for baseline covariates were not included in any of the predictors analyses.

censored prior to year 4. A total of 26 (22%) deaths occurred on study (IM + D − R, $n = 10$; IM + D − I, $n = 12$; IM + N − R, $n = 3$; and IM + N ± D, $n = 1$). Deaths were most commonly because of PD ($n = 12$ [10%]) or AEs ($n = 11$ [9%]); 3 deaths were due to unknown causes. Ten of the AEs resulting in death were considered by the investigator to be unrelated to treatment (intracranial hemorrhage, internal bleed secondary to kidney infarction, myocardial infarction, respiratory insufficiency, internal hemorrhage, advanced gastric cancer, mesenteric ischemia, pneumonia, severe heart failure, and acute myocardial infarction [$n = 1$ each]); 1 was considered treatment-related by the investigator (IM + D − I cohort; lower gastrointestinal bleeding occurring at 78 days on therapy in the setting of grade 4 thrombocytopenia within 30 days of the last bosutinib dose).

Cumulative incidence of on-treatment AP/BP transformation at 4 years was similar across age groups, whereas K–M-estimated OS at 4 years was lower in older patients. Cumulative incidence of on-treatment PD/death at 4 years appeared higher in younger vs. older patients (Supporting Information Table V); however, this is likely a function of more older patients discontinuing prior to PD/death.

Factors affecting long-term response and survival outcomes

Significant baseline predictors of attaining/maintaining an MCyR or CCyR were as follows (Table I): younger age (<65 years; CCyR by 6 months [$P = 0.0419$]); male sex (MCyR by 6 months [$P = 0.0319$]); Ph+ ratio ≤35 vs. ≥95% (MCyR and CCyR by 3, 6 months or anytime on treatment [$P \leq 0.0002$]); Ph+ ratio ≤35 vs. >35 to ≤95% (MCyR and CCyR by 3, 6 months, and CCyR anytime on treatment [$P \leq 0.0475$]); prior interferon (IFN) treatment (MCyR by 6 months or anytime on treatment [$P \leq 0.0253$]); and time from diagnosis to IM initiation of <6 months (with no prior IFNα) vs. ≥6 months or IFNα treatment anytime before IM (MCyR by 6 months or CCyR by 3 months [$P \leq 0.0244$]). No prior response to dasatinib or nilotinib was the only significant predictor of decreased OS. Increased basophils was the only significant predictor of decreased PFS ($P = 0.0012$; Table I).

Safety and tolerability

The most common nonhematologic TEAEs (all grades; grade 3/4) were gastrointestinal (diarrhea [83%; 9%], nausea [48%; 1%], and vomiting [38%; 1%]; Supporting Information Table VIII). Common hematologic TEAEs were thrombocytopenia (39%; 26%), neutropenia (21%; 16%), and anemia (20%; 7%). TEAEs by age (<40 vs. 40–64 vs. ≥65 years) are shown in Supporting Information Table V; grade 3/4 laboratory abnormalities occurring in ≥10% of patients are presented in Supporting Information Table IX.

Overall incidence of newly occurring TEAEs (MedDRA preferred terms not reported for the same patient previously for those on treatment during a given year) was most frequent in year 1 (99%) and somewhat lower in years 2 (74%), 3 (64%), and 4 (72%; Fig. 2). Increased blood creatinine and pleural effusion, which occurred in 13% and 17% of patients overall, had a higher incidence of first occurrence in year 4 (13 and 16%) vs. years 1, 2, and 3, respectively

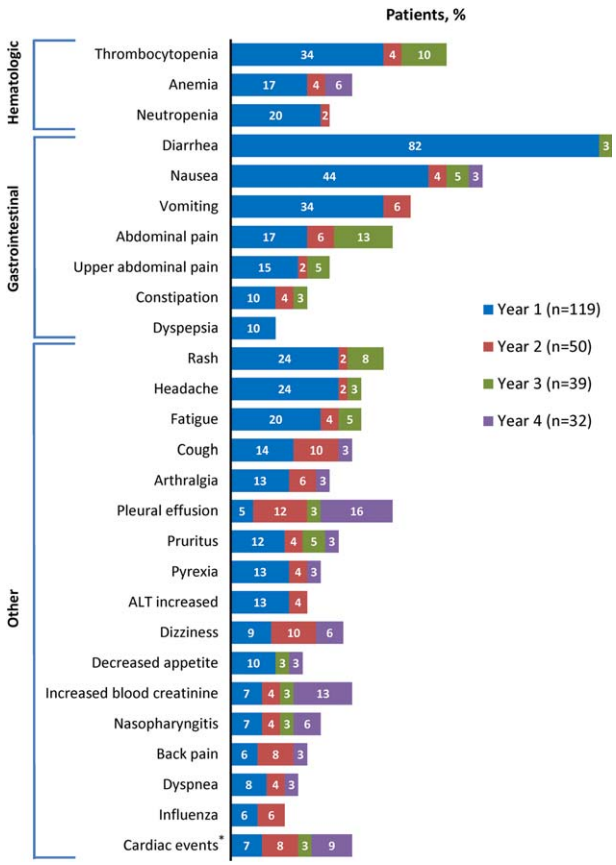

Figure 2. Percentage of patients with TEAEs occurring in year 1 and newly occurring in years 2, 3, and 4 in ≥10% of patients overall (any grade). AE = adverse event; ALT = alanine aminotransferase; TEAE = treatment-emergent adverse event. Denominators are the number of patients on treatment during the specific years (note: the incidence of certain TEAEs appears higher in later years compared with previous years due to a lower number of patients on treatment: pleural effusion [year 1, $n = 6/119$; year 2, $n = 6/50$; year 3, $n = 1/39$; year 4, $n = 5/32$]; increased blood creatinine [year 1, $n = 8/119$; year 2, $n = 2/50$; year 3, $n = 1/39$; year 4, $n = 4/32$]; cardiac AEs [year 1, $n = 8/119$; year 2, $n = 4/50$; year 3, $n = 1/39$; year 4, $n = 3/32$]). 1 year = 365.25 days. Newly occurring TEAEs were defined as those MedDRA preferred terms (PTs) not experienced by the same patient previously for patients on treatment during a given year. *Includes all PTs under the high-level group terms cardiac arrhythmias, pericardial disorders, and heart failures under the cardiac disorders system organ class (SOC) and the following PTs: cardiac death, sudden cardiac death, sudden death, decreased ejection fraction, abnormal electrocardiogram QT interval, prolonged electrocardiogram QT, congenital long QT syndrome.

(increased blood creatinine: 7, 4, 3%; pleural effusion: 5, 12, and 3%). However, only 29 patients remained on treatment through year 4. Among the 20 patients who experienced pleural effusions, 14 (70%) had a history of these events and 19 had received prior dasatinib.

Thirty-three (28%) patients discontinued treatment due to AEs as the primary reason (IM + D − R, $n = 7/38$ [18%]; IM + D − I, $n = 22/50$ [44%]; IM + N − R, $n = 3/26$ [12%]; IM + N ± D, $n = 1/5$ [20%]; Table II), most commonly thrombocytopenia ($n = 7/119$ [6%]; all 7 were IM + D − I patients, with 6 discontinuing in year 1). Across cohorts, the median (range) time to discontinuation due to an AE was 170 (15–2171) days. Among 34 patients who discontinued bosutinib due to an AE (including 1 patient who discontinued due to PD as the primary reason), 14 (41%) discontinued without attempting dose reduction to <500 mg/day for that AE. There were 5 deaths within 30 days of last dose, 2 due to PD and 3 due to AEs (myocardial infarction, acute myocardial infarction, and lower gastrointestinal bleeding).

Diarrhea was the most common TEAE (99 [83%]); all but one initial event occurred within year 1 and only 2 patients (IM + D − R, $n = 1$; IM + D − I, $n = 1$) discontinued bosutinib because of this AE (see Supporting Information text for details). Diarrhea was typically of low severity (maximum grade 1/2, 74%; grade 3, 9% [1% serious]; grade 4, 0%), transient (median duration of 2 [range: 1–413] days/any grade event; median [range] cumulative duration of 25 [range: 1–852] days), and initially occurred early (median time to onset of 2 [range: 1–210] days). Patients with diarrhea were managed with concomitant medications (65% [$n = 64$]), transient treatment interruptions (19% [$n = 19$]), and dose reductions (9% [$n = 9$]). Significant predictors of time to first grade 3/4 diarrhea were Ph+ ratio ≤35% vs. ≥95% at baseline ($P = 0.0412$) and concomitant medication (Loperamide and Lomotil) use at baseline ($P = 0.0129$; see Supporting Information text for a list of all factors assessed).

Alanine aminotransferase (ALT)/aspartate aminotransferase (AST) elevations occurred in 19 (16%; grade 3, 6%; no grade 4 or serious) patients (IM + D − R, $n = 7$; IM + D − I, $n = 6$; IM + N − R, $n = 5$; IM + N ± D, $n = 1$). These TEAEs were considered treatment-related by the investigator in 16 (13%) patients (IM + D − R, $n = 6$; IM + D − I, $n = 4$; IM + N − R, $n = 5$; IM + N ± D, $n = 1$) and led to discontinuation in 3 (3%) patients (IM + D − I, $n = 1$; IM + N − R, $n = 2$). ALT/AST elevations initially occurred early (median time to first event, 81 [range: 8–492] days) and had a median duration of 15 (range: 4–236) days/any grade event (cumulative duration: 29 [range: 7–250] days). Patients with ALT/AST events were managed with dose interruptions (6 [32%]), dose reductions (5 [26%]), and concomitant medications (1 [5%]). Liver-related AEs (increased ALT or AST, increased conjugated or blood bilirubin, hepatic enzyme increased, liver function test abnormal, increased transaminases, and hyperbilirubinemia) occurred in 22 (18%; grade 3, 7%; no grade 4 or serious AEs) patients. Significant baseline predictors of time to first grade 3/4 liver-related AE were duration of <6 months (with no prior IFNα) vs. ≥6 months from diagnosis to IM initiation or IFNα treatment anytime before IM ($P = 0.0366$) and increased basophils ($P = 0.0285$) (see Supporting Information text for a list of all factors assessed). No significant baseline predictors of all-grade liver-related AEs were identified.

Cardiac TEAEs were reported in 18 (15%; grade ≥3, 8%; serious, 6%) patients (IM + D − R, $n = 5$; IM + D − I, $n = 11$; IM + N − R, $n = 1$; IM + N ± D, $n = 1$; Supporting Information Table VIII), among whom 5 (28%) had a history of cardiac events. Cardiac AEs were considered by the investigator to be treatment-related in 13 (11%) patients (IM + D − R, $n = 2$; IM + D − I, $n = 10$; IM + N − R, $n = 0$; IM + N ± D, $n = 1$) and led to treatment discontinuation in 5 patients (IM + D − I cohort: cardiac failure, atrial fibrillation, and cardiac failure/pericarditis [$n = 1$ each]; pericardial effusion [$n = 2$]). Cardiac TEAEs did not lead to death in any patients. Individual cardiac AEs (any grade) occurring in ≥5% of patients aged ≥65 ($n = 27$), 40 − 64 ($n = 74$), or <40 years ($n = 18$) or with ≥5% difference between age groups were atrial fibrillation (11, 4, and 0%, respectively), cardiac failure (7, 1, and 0%), congestive cardiac failure (7, 1, and 0%), and pericardial effusion (11, 5, and 0%).

Seven (6%; grade ≥3, 4%; serious, 4%) patients reported vascular TEAEs (IM + D − R, $n = 1$; IM + D − I, $n = 5$; IM + N − R, $n = 1$; IM + N ± D, $n = 0$), none of which were considered treatment-related by the investigator. Among these 7 patients, 4 had a history of vascular events. One patient had a vascular TEAE that led to treatment discontinuation (IM + D − R cohort: myocardial infarction) and 2 patients had events that led to death (IM + D − I cohort: myocardial infarction and acute myocardial infarction). Hypertension-related events occurred in 9 (8%; grade ≥3, 2%; no grade 4 or serious AEs) patients (IM + D − R, $n = 2$; IM + D − I, $n = 6$; IM + N − R, $n = 1$; IM + N ± D, $n = 0$), none of which were considered treatment-

TABLE II. AEs Resulting in Treatment Discontinuation in Years 1–4 (≥1% of Patients Overall)[a]

	Year 1 ($n = 119$)	Year 2 ($n = 50$)	Year 3 ($n = 39$)	Year 4 ($n = 32$)	Total[b] ($n = 119$)
Any AE, *n* (%)	23 (19)	2 (4)	2 (5)	3 (9)	34 (29)
Thrombocytopenia	6 (5)	0	0	0	7 (6)
Neutropenia	5 (4)	0	0	0	5 (4)
Pleural effusion	0	0	0	1 (3)	4 (3)
ALT increased	3 (3)	0	0	0	3 (3)
Dyspnea	1 (1)	0	0	1 (3)	3 (3)
Vomiting	3 (3)	0	0	0	3 (3)
Anemia	1 (1)	0	0	1 (3)	2 (2)
Cardiac failure	1 (1)	1 (2)	0	0	2 (2)
Diarrhea	2 (2)	0	0	0	2 (2)
Pericardial effusion	0	0	0	1 (3)	2 (2)
Renal failure	0	0	1 (3)	0	2 (2)

ALT = alanine aminotransferase.
[a] Denominators for year 1, year 2, year 3, and year 4 are the number of patients remaining on treatment during those time periods (1 year = 365.25 days).
[b] Includes discontinuations due to AEs occurring after 4 years and 1 discontinuation due to disease progression as the primary reason.

related by the investigator; 2 of these 9 patients (both IM + D − I) had a history of hypertension. One grade 2 event of peripheral arterial occlusive disease was reported in an IM + N − R patient with prior nilotinib exposure, which was considered serious, was unrelated to bosutinib, and resolved within 10 days.

Cross-intolerance

Of the 119 patients who received third-/fourth-line bosutinib, 35 were intolerant (defined as permanent discontinuation due to an AE) to prior imatinib (1 additional patient did not have AEs reported), 50 were intolerant to prior dasatinib (2 additional patients did not have AEs reported), and 3 were intolerant to prior nilotinib (Supporting Information Table X). Among the 35 imatinib-intolerant patients, 7 (20%) were cross-intolerant to bosutinib (i.e., discontinued due to the same AE that led to prior imatinib discontinuation: thrombocytopenia [IM + D − I, $n = 3$] and bone marrow failure [IM + D − I, $n = 3$; IM + D − R, $n = 1$). Among the 50 dasatinib-intolerant patients, 12 (24%) were cross-intolerant to bosutinib (all IM + D − I patients), most commonly due to thrombocytopenia ($n = 4$), pleural effusion ($n = 2$), and bone marrow failure ($n = 2$). One of the 3 nilotinib-intolerant patients discontinued bosutinib due to the same AE that led to prior nilotinib discontinuation (pleural effusion; IM + N ± D cohort). No deaths occurred on bosutinib due to the same AE that led to intolerance to any of the 3 prior TKIs.

Discussion

The cumulative rates of newly attained/maintained cCHR or newly attained cCHR in this 4-year update of the ongoing phase 1/2 study (data snapshot: May 23, 2014) were similar to those reported at year 1 (data snapshot: March 28, 2011; 74% [$n = 86/117$] vs. 73% [$n = 85/116$] and 63% [$n = 41/65$] vs. 65% [$n = 44/68$], respectively) [14]. Cytogenetic responses were also similar; here, cumulative rates of newly attained/maintained MCyR and CCyR were 40% ($n = 45/112$) and 32% ($n = 36/112$), respectively, compared with 39% ($n = 42/108$) and 31% ($n = 33/108$) in the 1-year update [14]. Although the cohorts have changed since year 1, these findings suggest that a substantial number of patients receiving third-/fourth-line bosutinib may attain clinical benefit and that response is most likely to occur within the first year of treatment.

Responses were also durable, with 1-year vs. 4-year K–M estimates of maintaining CHR, MCyR, and CCyR of 73 vs. 63%, 72 vs. 69%, and 58 vs. 54%, respectively. These rates were lower in the IM + D − R cohort (57, 43, and 17%) vs. the IM + D − I (70, 87, and 66%) and IM + N − R (62, 78, and 63%) cohorts, respectively (NE for IM + N ± D). However, estimates of long-term outcomes may be biased due to early discontinuation of patients for reasons such as unacceptable toxicity or inadequate response, and because survival was only followed for up to 2 years after treatment discontinuation.

Importantly, cytogenetic responses were attained across a spectrum of baseline BCR–ABL1 mutations, except T315I, G250E, and V299L. The overall cumulative response rates observed here (cCHR, 74%; MCyR, 40%; CCyR, 32%) are consistent with those reported with third-line dasatinib and nilotinib in CP CML patients resistant/intolerant to ≥2 prior TKIs (CHR, 67–81% [24]; MCyR, 22 − 48% [24–26]; CCyR, 11 − 32% [24–26]). However, differences in follow-up duration and design (the present study is a prospective analysis of third-/fourth-line therapy whereas the reports of dasatinib and nilotinib were retrospective) complicate direct comparisons of efficacy across studies. MCyR and CCyR rates in CP CML patients receiving ponatinib in a previous phase 2 study (median follow-up, 15 months) were 67 and 56%, respectively, for third-line therapy and 45 and 39% for fourth-line therapy [27].

Analyzing predictive factors for outcome is important for identifying patients most likely to benefit from treatment. Consistent with present observations, a previous study of third-line dasatinib or nilotinib in patients with CP CML found that age was not a significant predictor of cytogenetic response [26]. Furthermore, in the previous study, best response of at least a minimal CyR (MiCyR; ≤95% Ph+ cells) with prior second-line dasatinib or nilotinib was associated with subsequent cytogenetic response and improved survival on third-line nilotinib or dasatinib [26]. In this analysis, having had at least an MiCyR with prior dasatinib and/or nilotinib was a significant predictor of survival and a lower Ph+ ratio at baseline (≤35%) was a significant predictor of cytogenetic response. However, marginally significant predictors should be interpreted with caution, as *P* values were not adjusted for multiple testing.

Gastrointestinal toxicities remained the most commonly reported AEs, with similar percentages of patients reported to have gastrointestinal AEs in the 4-year vs. 1-year updates (e.g., diarrhea [83 vs. 81%, respectively], nausea [48 vs. 43%], and vomiting [38 vs. 32%]), indicating the early incidence of these events. Although diarrhea was common, grade 3 events were experienced by only 9% of patients and no grade 4 events were reported. Cumulative rates of grade 3/4 hematologic laboratory abnormalities were also similar in the 4-year vs. 1-year [14] updates (e.g., thrombocytopenia [26 vs. 25%], anemia [8 vs. 8%], and neutropenia [18 vs. 19%]). Except for elevated blood creatinine and pleural effusions, newly occurring AEs generally decreased in frequency after the first year of treatment. However, most discontinuations due to AEs (64%) occurred during year 1 and only 29 patients remained on treatment after year 4; thus, these results should be interpreted with caution. The observed low rates of cross-intolerance between bosutinib and prior imatinib, dasatinib, or nilotinib treatment suggests that most patients intolerant to prior TKI therapy may be successfully treated with bosutinib.

In conclusion, high response rates were observed in CP CML patients receiving long-term bosutinib as third- or fourth-line treatment. Most responses (attained or maintained) were observed within the first year of treatment and, among responders, the likelihood of maintaining a response was high at 4 years. Overall, 4 years after last enrollment, bosutinib continues to demonstrate durable efficacy and manageable toxicity for the majority of patients with CP CML resistant or intolerant to multiple prior TKIs who are able to remain on treatment after the first year, representing an important treatment option for patients in this setting.

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