The initial molecular response predicts the deep molecular response but not treatment-free remission maintenance in a real-world chronic myeloid leukemia cohort

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Abstract

In chronic myeloid leukemia, the identification of early molecular predictors of stable treatment-free remission (TFR) after tyrosine kinase inhibitor (TKI) discontinuation is challenging. The predictive values of residual disease (*BCR::ABL1* quantification) at month 3 and 6 and more recently, *BCR::ABL1* transcript halving time (HT) have been described, but no study compared the predictive value of different early parameters. Using a real-world cohort of 408 patients, we compared the performance of the EUTOS long-term survival (ELTS) score, *BCR::ABL1* HT, and residual disease at month 3 and 6 to predict the molecular response, achievement of the TKI discontinuation criteria, and TFR maintenance. The performances of *BCR::ABL1* HT and residual disease at month 3 were similar. Residual disease at month 6 displayed the best performance for predicting the optimal response (area under the ROC curve between 0.81 and 0.92; cut-off values: 0.11% for MR4 at month 24 and 0.12% for MR4.5 at month 48). Conversely, no early parameter predicted reaching the TKI discontinuation criteria and TFR maintenance. We obtained similar results when patients were divided in subgroups by first-line treatment (imatinib *vs.* second-generation TKI [2G-TKI]). We identified a relationship between ELTS score, earlier milestones and TFR maintenance only in the 2G-TKI group. In conclusion, this first comparative study of early therapeutic response parameters showed that they are excellent indicators of TKI efficacy (*BCR::ABL1* transcript reduction) and best responders. Conversely, they did not predict the achievement of the TKI discontinuation criteria and TFR maintenance, suggesting that other parameters are involved in TFR maintenance.

Introduction

Since the introduction of the first tyrosine kinase inhibitor (TKI), imatinib, for the management of chronic-phase chronic myeloid leukemia (CP-CML),[1] the challenges have changed considerably. The lifespan of patients with CP-CML treated with TKI is considered similar to that of an age- and sex-matched control population,[2] and <5% of

patients are resistant to treatment.[3] TKI efficacy is such that CML becomes undetectable in approximately 30% of patients treated with imatinib[3,4] and the major molecular response (MMR) after TKI withdrawal is maintained in 50% to 70% of patients. The development of second-generation (2G-TKI; nilotinib, dasatinib, bosutinib) and third-generation TKI (ponatinib) offers a large therapeutic arsenal and their optimal use is constantly improved to ensure efficacy and safety.[5,6]

The progress made in CP-CML monitoring has led to increasingly precise guidelines,[7-9] particularly concerning the initial phase of treatment that is related to the risk of disease progression. For example, residual disease assessment by *BCR::ABL1* transcript quantification at month 3 and month 6 of treatment is an important predictive parameter of low risk of progression.[10-12] This justified its inclusion in the European LeukemiaNet (ELN) recommendations[7] and National Comprehensive Cancer Network (NCCN)[13] guidelines. This initial phase is also essential to give patients the best chance of achieving the deepest molecular response and could influence the choice of TKI, depending on the patient's age and individual objective, particularly treatment-free remission (TFR).[14,15] In this context, early parameters have been analyzed to determine whether they can predict the deep molecular response. Moreover, the ELN and NCCN recommendations take into account studies showing that a satisfactory reduction in residual disease at month 3 and month 6 can predict the subsequent molecular response, but with subtle differences.[15] In order to identify more effective early endpoints, several research groups evaluated the value of the initial *BCR::ABL1* transcript decrease kinetics, particularly Branford *et al.*, who defined the initial halving time (HT).[16] This parameter appears useful for TFR prediction.[17] Few other groups used the same or similar approaches.[18-21] However, the relationship between *BCR::ABL1* HT and TFR has not been assessed in other cohorts and the predictive performance of the main early molecular response parameters (*BCR::ABL1* HT, residual disease at month 3 and 6) have not been compared, particularly in a real-life cohort.

Here, we used data from the French CML Observatory[4] to compare the value of these parameters for predicting the achievement of molecular responses, TKI discontinuation criteria, and TFR maintenance. We added in our analysis also the EUTOS long-term survival (ELTS) score.[22,23] This score is frequently used in clinical practice for the initial therapeutical decisions because of its stronger link with the chance of obtaining a deep response compared with the Sokal score.[24,25]

Methods

Chronic myeloid leukemia Observatory database

The CML Observatory database is a secure database to collect the real life laboratory and clinical data of patients with CML after obtaining their informed consent[4] (*Online Supplementary Appendix*). The registration of patients is done, on a voluntary basis, by their physician at each center. All registered patients have a CML ID that is automatically generated at the first registration. The CML Observatory is hosted by a professional health data host (MIPIH) and has been authorized (authorization no. 914456) by CNIL, the French data protection authority, in accordance with its ethical standards and with the 1964 Declaration of Helsinki and its later amendments or comparable ethical standards. It is promoted by the Clermont-Ferrand University Hospital that is also the coordinating center. The data used for the present study are from nine participating centers.

Design and patient selection

At the time of database freezing for data collection (June 30, 2022), 1,123 patients were eligible for inclusion on the basis of the following inclusion criteria: CP-CML with a follow-up for at least 12 months, treated with TKI only. A pretreatment with hydroxyurea was accepted because it does not have any significant effect on the *BCR::ABL1/ABL1* ratio[16] (*Online Supplementary Figure S1*). From the 801 selected patients, we retained only 408 patients with data on all the four early parameters analyzed in this study: ELTS score, *BCR::ABL1* transcript quantification at diagnosis, month 3, and month 6 (±45 days for these two time points) (Figure 1). We excluded patients with a *BCR::ABL1/ABL1* ratio <20% at diagnosis not confirmed by the registering center. The characteristics of the 393 patients excluded were similar to those of the included patients, except for longer TKI duration that explained the high rate of missing data (longer follow-up and impossibility of recovering early data not included in previous recommendations) (*Online Supplementary Table S1*). Then, we divided the selected patients (N=408) in subgroups using an "intention-to-treat" approach according to the first-line TKI used: imatinib (IMA; N=274), 2G-TKI (N=118; N=101 nilotinib and N=17 dasatinib), and other TKI (N=16; N=7 bosutinib and N=9 ponatinib).

We defined the response to TKI therapy according to the usual residual disease assessment criteria using the *BCR::ABL1/ABL1%*^{IS} transcript level (hereafter, *BCR::ABL1* transcript level) to determine the thresholds that correspond to the MMR (≤0.1%), MR4 (≤0.01%), MR4.5 (≤0.0032%), and MR5 (≤0.001%). We defined the deep molecular response (DMR) as the achievement of at least MR4.5. This definition was used in clinical practice during the study period. We considered the therapeutic response as stable when *BCR::ABL1* transcript level was below the considered threshold in three successive assessments. The date of the first value below the considered threshold was the date of therapeutic response achievement. TKI discontinuation was recommended when at least three consecutive values were below the considered threshold within 24 months. In this real-life cohort, 29 patients (7% of the whole cohort and

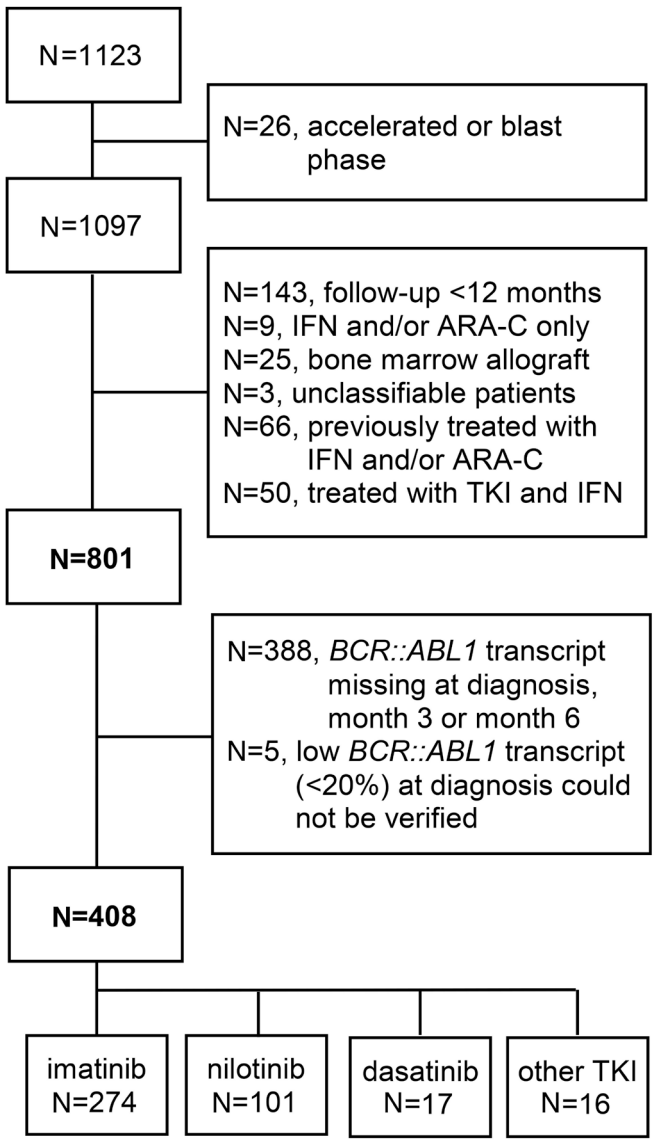


Figure 1. Study flowchart. ARA-C: aracytine; IFN: interferon; TKI: tyrosine kinase inhibitor; other TKI: patients treated with bosutinib (N=7) or ponatinib (N=9).

10.6% of the IMA subgroup) received the diagnosis before 2009 (i.e., before the introduction of the International Scale to measure *BCR::ABL1* transcript level). These patients were not excluded because they were all monitored by the same laboratory, thus using the same technique. All participating laboratories are reference laboratories that are part of the French national group of molecular biology laboratories.

Halving time of the *BCR::ABL1/ABL1* ratio and early response milestones

We used the formula described by Shanmuganathan *et al.*[17]: ln(2)*(number of days between the diagnosis and the day of the 3-month *BCR::ABL1* measurement/ln (transcript at diagnosis/transcript at month 3) (formula [a]) because the decay kinetics are logarithmic whatever the initial and 3-month transcript levels.[16] In formula (a), the initial time point is the diagnosis day because the *BCR::ABL1* transcript levels are similar at diagnosis and at treatment initiation (Branford 2014). Nevertheless, due to the variability of the interval between diagnosis and TKI initiation in our sample (median [25th; 75th percentiles]: 0.8 [0.5; 1. 2] months), we used also formula (b) where the initial time point was the first day of TKI treatment (information available in the CML Observatory). This can be considered the first day of TKI action and therefore, the real start of the tumor mass decrease kinetics and of the residual disease assessment. Formula (b) gave shorter HT (*Online Supplementary Table S2*), but the relationship between HT and the likelihood of therapeutic response and of TFR maintenance after TKI discontinuation were identical for both HT values (*data not shown*). As the *BCR::ABL1/ABL1* transcript level can be considered stable between diagnosis and TKI start[16] and as the variability of the interval between diagnosis and treatment initiation might lead to an evaluation bias, we chose to show only the results obtained with formula (b).

Statistical analysis

Categorical data are expressed as numbers and percentages, and continuous data as mean ± standard deviation or median [25th; 75th percentiles] in function of their statistical distribution.

The patients not included in the study (N=393) were compared to the included patients (N=408) with the χ^2 or Fisher's exact test for categorical data, and with the Student's *t* or Mann-Whitney test for quantitative data.

The cumulative MMR, MR4, MR4.5, and MR5 incidences were measured from the first TKI initiation to the molecular response date. Death before achieving the therapeutic response was considered as a competing event and the cumulative incidence curves were compared using the Gray's test.[4]

Receiver operating characteristic (ROC) curves were plot-

ted to assess whether ELTS score, HT, residual molecular disease at month 3 and at month 6 could predict the therapeutic response. Areas under the ROC curves (AUC) were presented with their 95% confidence intervals (95% CI) and compared using the method described by DeLong ER *et al.*[26] The "optimal" thresholds of HT and residual molecular disease at month 6 to predict the molecular response were defined using the method described by Liu *et al.*[27] that is based on the maximization of the product of sensitivity (Se) and specificity (Sp). These thresholds were presented with their Se and Sp.

ROC curves were also plotted to assess whether ELTS score, HT, residual molecular disease at month 3 and month 6 could predict the achievement of the TKI discontinuation criteria. Patients who did or did not meet such criteria were compared using generalized linear mixed models with logit link function, considering the centers as random effect.

Censored data were estimated using the Kaplan-Meier method and the factors associated with TFR loss were studied with the log-rank test.

Statistical analyses were performed with the Stata software (version 15; StataCorp, College Station, Texas, USA). All tests were two-sided, with an α level set at 5%. No correction for multiple testing was applied in subgroup analyses.[28] The findings obtained from these analyses were interpreted as exploratory.

Results

Baseline characteristics of the patients with chronic-phase chronic myeloid leukemia

The mean age of the 408 patients included in the analysis was 56.2±15.9 years, and 51.7% of them were men (Table 1). The median molecular follow-up was 62 (interquartile range [IQR], 33-98) months, and the median TKI treatment duration was 70 (IQR, 41-101) months. The distribution of patients with low, intermediate, and high prognostic risk, based on the Sokal and ELTS scores, was globally similar to published data.[22] Overall, the median HT was 17 (IQR, 12-26) days and was significantly shorter in the 2G-TKI than IMA subgroup (13 [IQR, 10-18] *vs.* 20 [IQR, 13-31] days; *P*<0.001).

BCR::ABL1 transcript level at month 6 and 12 are the best predictors of therapeutic molecular responses

In the sample, 318 (77.9%), 209 (51.2%), 160 (39.2%), and 130 (31.9%) patients reached MMR, MR4, MR4.5, and MR5 in a median time of 0.8 (IQR, 0.5-1.5), 1.6 (IQR, 0.8-3.0), 2.3 (IQR, 1.3-4.1), and 3.1 (IQR, 2.0-5.1) years, respectively. At year 4 of follow-up, the cumulative MMR, MR4, MR4.5, and MR5 incidence rates were 87.0%, 52.9%, 35.6%, and 26.6%, respectively (*Online Supplementary Figure S2*).

HT was significantly shorter (10-14 days *vs.* 18-23 days; *P*<0.001) in patients who achieved each milestone than in

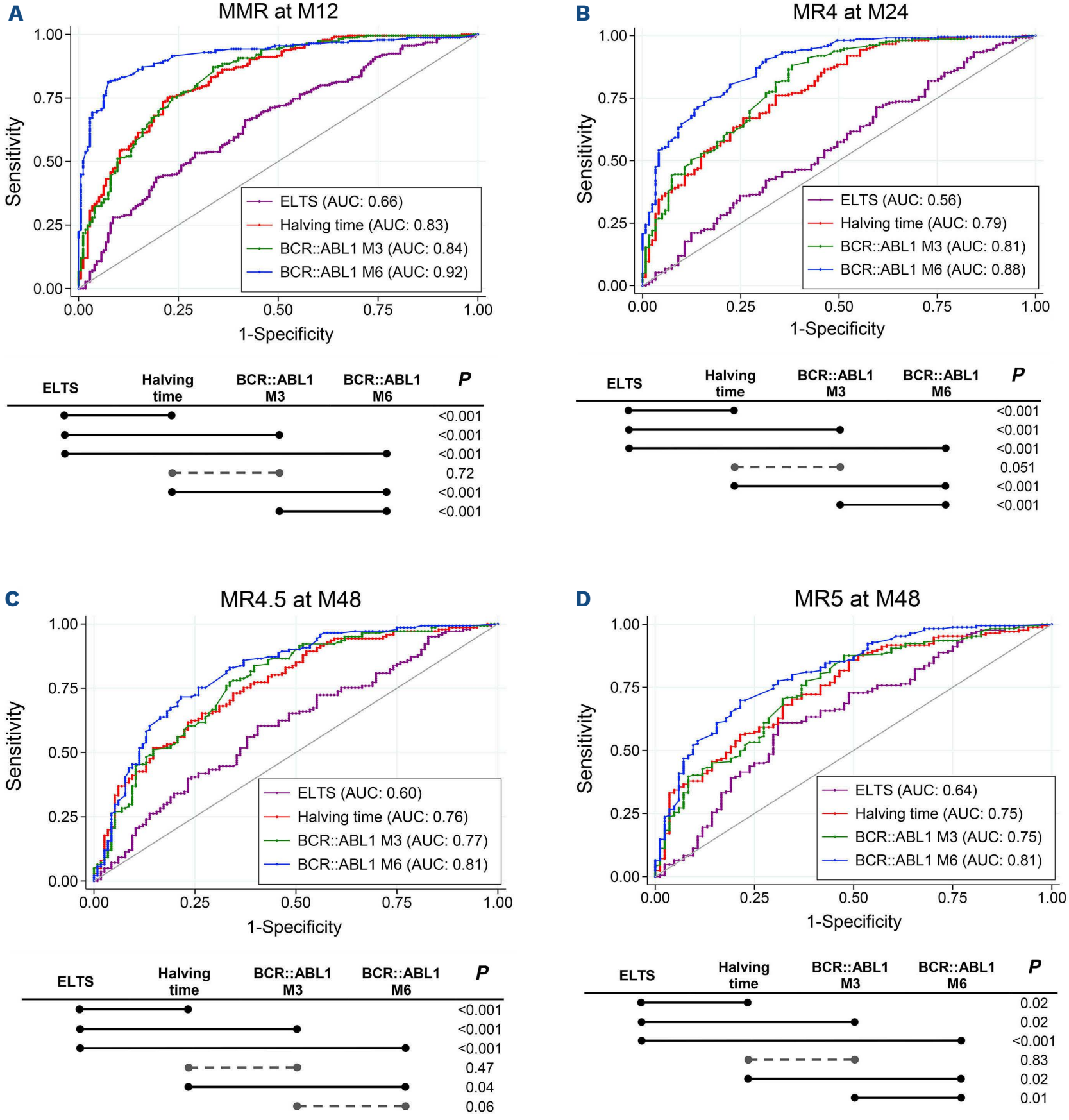
Table 1. Characteristics of the patient subgroups according to the first-line tyrosine kinase inhibitor.

	Total N=408	Imatinib N=274	2G-TKI N=118	Other TKI N=16
Men, N (%)	211 (51.7)	139 (50.7)	63 (53.4)	9 (56.3)
Age in years at diagnosis, mean ± standard deviation	56.2±15.9	59.3±16.0	50.6±13.9	44.7±12.9
Sokal score, N (%) Low <0.8 Intermediate 0.8-1.2 High >1.2 Unknown	 139 (34.1) 178 (43.6) 83 (20.3) 8 (2.0)	 95 (34.7) 125 (45.6) 49 (17.9) 5 (1.8)	 40 (33.9) 45 (38.1) 30 (25.4) 3 (2.5)	 4 (25.0) 8 (50.0) 4 (25.0) 0 (0.0)
ELTS, N (%) Low ≤1.5680 Intermediate 1.5680-2.2185 High >2.2185 Unknown	 223 (54.7) 127 (31.1) 50 (12.3) 8 (2.0)	 154 (56.2) 80 (29.2) 35 (12.8) 5 (1.8)	 62 (52.5) 41 (34.7) 12 (10.2) 3 (2.5)	 7 (43.8) 6 (37.5) 3 (18.8) 0 (0.0)
Molecular follow-up duration in months[a], median (IQR)	62 (33-98)	63 (34-103)	65 (37-96)	18 (14-24)
TKI treatment duration in months[b], median (IQR)	70 (41-101)	70 (42-104)	72 (45-100)	22 (19-25)
Time in months from diagnosis to TKI treatment start, median (IQR)	0.8 (0.5-1.2)	0.7 (0.4-1.1)	0.9 [0.5; 1.2]	0.7 (0.4-1.2)
Halving time in days, median (IQR)	17 (12-26)	20 (13-31)	13 (10-18)	18 (13-30)
BCR::ABL1/ABL1^{IS} at month 3, median (IQR)	1.7 (0.4-7.5)	2.8 (0.6-10.7)	0.6 (0.1-1.7)	1.8 (0.5- 8.5)
BCR::ABL1/ABL1^{IS} at month 6, median (IQR)	0.3 (0.0-1.3)	0.5 (0.1-1.6)	0.1 (0.0-0.4)	0.2 (0.0-0.6)

ELTS: EUTOS long-term survival; Other TKI: patients treated with bosutinib (N=7) or ponatinib (N=9); TKI: tyrosine kinase inhibitor; 2G-TKI: second-generation tyrosine kinase inhibitor (N=101 patients treated with nilotinib and N=17 patients treated with dasatinib). [a]: duration defined as the time between the diagnosis date and the date of the last available transcript quantification; [b]: from the first day of treatment to the date of discontinuation or the date of extraction from the database.

those who did not (*Online Supplementary Figure S3A*). Then, we used ROC curves to compare the predictive performance of the main early follow-up parameters: the initial ELTS score that is considered to be the initial best prognostic score for patients treated with TKI,[25] the *BCR::ABL1/ABL1* ratio HT, and residual molecular disease at month 3 and 6 (Figure 2). The initial ELTS score performed badly, as indicated by the ROC-AUC values between 0.56 (to predict MR4 at month 24) and 0.66 (to predict MMR at month 12). Overall, HT and residual disease at month 3 performed similarly (P>0.05) to predict MMR at month 12 (AUC: 0.83, 95% confidence interval [CI]: 0.79-0.87, and AUC: 0.84, 95% CI: 0.80-0.88, respectively), MR4 at month 24 (AUC: 0.79, 95% CI: 0.74-0.84, and AUC: 0.81, 95% CI: 0.76-0.86), MR4.5 at month 48 (AUC: 0.76, 95% CI: 0.71-0.82, and AUC: 0.77, 95% CI: 0.72-0.83), and MR5 at month 48 (AUC: 0.75, 95% CI:0.68-0.81, and AUC: 0.75, 95% CI: 0.68-0.81) (Figure 2A-D). The HT cut-off values used to classify patients according to the therapeutic response (MMR at month 12, MR4 at month 24, MR4.5, and MR5 at month 48) were 17 (Se: 0.75; Sp: 0.78), 14 (Se: 0.76; Sp: 0.67), 14 (Se: 0.73; Sp: 0.66), and 14 (Se: 0.68; Sp: 0.68) days, respectively.

Conversely, residual disease at month 6 had a better predictive value (P<0.05) than the other parameters, as indi-



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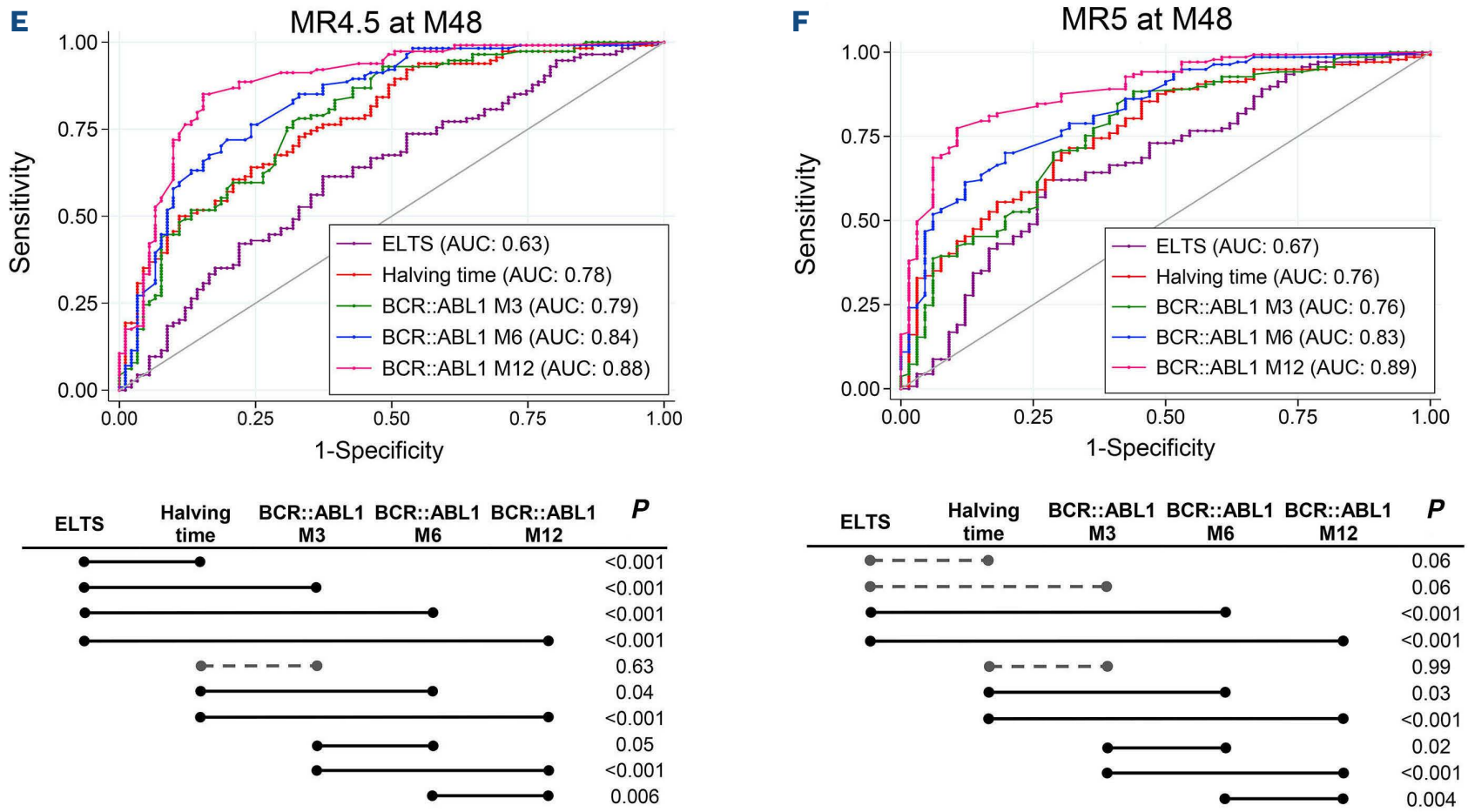


Figure 2. Receiver operating characteristic curves. Receiver operating characteristic (ROC) curves to evaluate early parameters (i.e., EUTOS long-term survival score [ELTS] score, Halving time [HT], *BCR::ABL1/ABL1*^{IS} ratio at months 3 and 6) as predictive markers for the achievement of major molecular response (MMR) at month 12 (A, N=400 patients), molecular responses 4-log reduction (MR4) at month 24 (B, N=330), 4.5-log reduction (MR4.5) (C, N=257) and 5-log reduction (MR5) (D, N=253) at month 48 of tyrosine kinase inhibitor (TKI) treatment. A subgroup analysis was performed in patients with *BCR::ABL1* transcript measurement at month 12 to evaluate early parameters as predictive markers of MR4.5 (E, N=205) and MR5 (F, N=203) at month 48. ROC curve analyses to evaluate early parameters (i.e., ELTS, HT, *BCR::ABL1/ABL1*^{IS} ratio at month 3 and month 6) as predictive markers of the achievement of the criteria for TKI discontinuation (treated for at least 5 years and with MR4.5 or MR5 for at least 24 months), regardless of when they were obtained (G, N=127). AUC: area under the curve; M3: month 3; M6: month 6; M12: month 12; M24: month 24; M48: month 48. The pairwise comparisons are shown below each panel (only if *P* value <0.05, Omnibus test), with black full lines indicating significant differences and gray dashed lines non-significant differences.

cated by the AUC values: 0.92 (95% CI: 0.89-0.95) for MMR at month 12, 0.88 (95% CI: 0.85-0.92) for MR4 at month 24, 0.81 (95% CI: 0.76-0.87) for MR4.5 at month 48, and 0.81 (95% CI: 0.75-0.87) for MR5 at month 48 (Figure 2). The *BCR::ABL1/ABL1* ratio cut-off values at month 6 to classify patients according to their therapeutic response were 0.11% (Se: 0.80; Sp: 0.77) for MR4 at month 24, 0.17% (Se: 0.71; Sp: 0.78) for MR4.5 at month 48, and 0.12% (Se: 0.70; Sp: 0.78) for MR5 at month 48. Therefore, we included the assessment of residual disease at month 12 in the analysis of the predictive performance of early parameters for achieving MR4.5 and MR5 at month 48. In a smaller subgroup due to

the limited data availability (N=205 for MR4.5 and N=203 for MR5), the *BCR::ABL1* transcript level at month 12 was the most effective parameter (AUC: 0.88, 95% CI: 0.84-0.93, and AUC: 0.89, 95% CI: 0.84-0.94, respectively) for predicting MR5 at month 48 (Figure 2E, F).

As we used an 'intention-to-treat' approach to analyze data from patients who received a TKI as first-line treatment, we evaluated the percentages of patients who changed lines and found that they were similar in the IMA (37%) and 2G-TKI (32%) subgroups. Then, we carried out a sensitivity analysis by including only patients who did not change treatment (N=258). The results were similar as those for the whole sample (*Online Supplementary Figure S4*).

All early parameters poorly predict the achievement of the treatment discontinuation criteria

According to the French CML study group[29] and the ELN recommendations,[23,29] patients treated for at least 5 years and with MR4.5 or MR5 for at least 24 months are candidates for TKI discontinuation. In our cohort, 127 patients (31.1%) reached these criteria (Table 2). This subgroup included 59.1% of women and had a significantly shorter HT than the subgroup who did not meet these criteria (14 [IQR, 11-20] *vs.* 19 [IQR, 13-30] days; $P<0.001$). This group also had lower *BCR::ABL1* transcript level at month 3 (0.6 [IQR, 0.1-3.4] *vs.* 2.5 [IQR, 0.6-9.9]; $P<0.001$) and at month 6 (0.1 [IQR, 0.0-0.4] *vs.* 0.5 [IQR, 0.1-1.7]; $P<0.001$). However, HT and *BCR::ABL1* transcript quantification at month 3 and month 6 moderately predicted the achievement of the TKI discontinuation criteria: AUC=0.65 (95% CI: 0.60-0.71), 0.66 (95% CI: 0.60-0.71) and 0.66 (95% CI: 0.61-0.72), respectively (Figure 2G). Among these 127 patients, 60 (47.2%) discontinued treatment due to optimal therapeutic response (according to the clinician's judgment). We divided these 127 patients in four groups using the HT quartiles: <10.60, 10.60 to 13.93, 13.94 to 20.50, and ≥20.50 days. The percentage of patients who discontinued treatment was higher in first quartile HT group (33.3% *vs.* 17.9% of patients who continued their treatment), and lower in the fourth quartile (20.0% *vs.* 28.4%).

Table 2. Characteristics of candidates who met the criteria for tyrosine kinase inhibitor discontinuation and of patients who stopped treatment in real life.

	Met the withdrawal criteria[a] N=127	Stopped treatment due to optimal therapeutic response in real life N=70
First-line TKI, N (%) Imatinib Nilotinib Dasatinib Other	 88 (69.3) 31 (24.4) 7 (5.5) 1 (0.8)	 39 (55.7) 21 (30.0) 10 (14.3) 0 (0.0)
Men, N (%)	52 (40.9)	28 (40.0)
Age at diagnosis in years, mean ± standard deviation	56.4±16.0	55.1±14.6
Sokal score, N (%) Low Intermediate High Unknown	 38 (29.9) 59 (46.5) 28 (22.0) 2 (1.6)	 26 (37.1) 25 (35.7) 18 (25.7) 1 (1.4)
ELTS score, N (%) Low Intermediate High Unknown	 77 (60.6) 31 (24.4) 17 (13.4) 2 (1.6)	 51 (72.9) 11 (15.7) 7 (10.0) 1 (1.4)
Molecular follow-up duration in months, median (IQR)	103 (78-129)	107 (81-135)
TKI treatment duration in months, median (IQR)	91 (76-115)	80 (68-104)
Time from diagnosis to TKI initiation in months, median (IQR)	0.8 (0.5-1.1)	0.7 (0.5-1.2)
Halving time in days, median (IQR)	14 (11-20)	12 (9-16)
BCR::ABL1/ABL1^{IS} at month 3, median (IQR)	0.6 (0.1-3.4)	0.4 (0.1-2.0)
BCR::ABL1/ABL1^{IS} at month 6, median (IQR)	0.1 (0.0-0.4)	0.1 (0.0-0.3)

ELTS: EUTOS long-term survival; TKI: tyrosine kinase inhibitor; Other (N=1): patient treated with bosutinib. [a]At least 5 years of treatment and at least MR4.5 for 2 years.

Early parameters are not correlated with treatment-free remission maintenance in patients who discontinued tyrosine kinsase inhibitors due to optimal therapeutic response

In the study cohort, 70 patients (17.2% of the whole sample and 55% of patients who reached the discontinuation criteria) discontinued treatment due to optimal therapeutic response (according to the clinician's judgement). Their characteristics are presented in Table 2. Compared with the subgroup that did not stop treatment, this subgroup had shorter HT (12 [IQR, 9-16] *vs.* 18 [IQR, 13-29] days; *P*<0.001) and lower *BCR::ABL1* transcript level at month 3 (0.4 [IQR, 0.1-2.0] *vs.* 2.3 [IQR, 0.5-8.2] %; *P*<0.001) and at month 6 (0.1 [IQR, 0.0-0.3] *vs.* 0.4 [IQR, 0.1-1.5]; *P*<0.001). Twenty-eight patients (40.0% of 70) restarted treatment due to molecular relapse. TFR loss was associated only with intermediate (*vs.* low) ELTS score (hazard ratio [HR]=1.86, 95% CI: 1.31-2.66; *P*=0.001), but not with age, sex, Sokal score, follow-up duration, first-line TKI, treatment duration, and HT. Particularly, HT was not significantly different in patients who relapsed and those with TFR maintenance (12 [IQR, 9-15] and 13 days [IQR, 10-19]; *P*=0.52). We divided these 70 patients in four groups using the HT quartiles: <9.40, 9.40 to 12.20, 12.20 to 17.40, and ≥17.40 days. Despite low patient numbers, the percentage of patients who relapsed was higher in the first HT quartile group (35.7% *vs.* 19.0% of patients with TFR maintenance) and lower in the fourth quartile group (21.4% *vs.* 26.2% of patients with TFR maintenance).

Analysis of factors associated with relapse at specific time points after TKI discontinuation (12 months [N=22/63 patients with relapse], 18 months [N=24/61 patients with relapse] and 24 months [N=24/57 patients with relapse]) did not show any relationship between TFR maintenance and HT or *BCR::ABL1* transcript quantification at month 3 and 6 (*data not shown*).

First-line tyrosine kinase inhibitor influence

As previous studies reported faster initial residual disease reduction with 2G-TKI,[3,30-33] we classified patients into two subgroups (IMA, N=274; 2G-TKI, N=118) to assess the influence of the first-line TKI on the predictive performance of

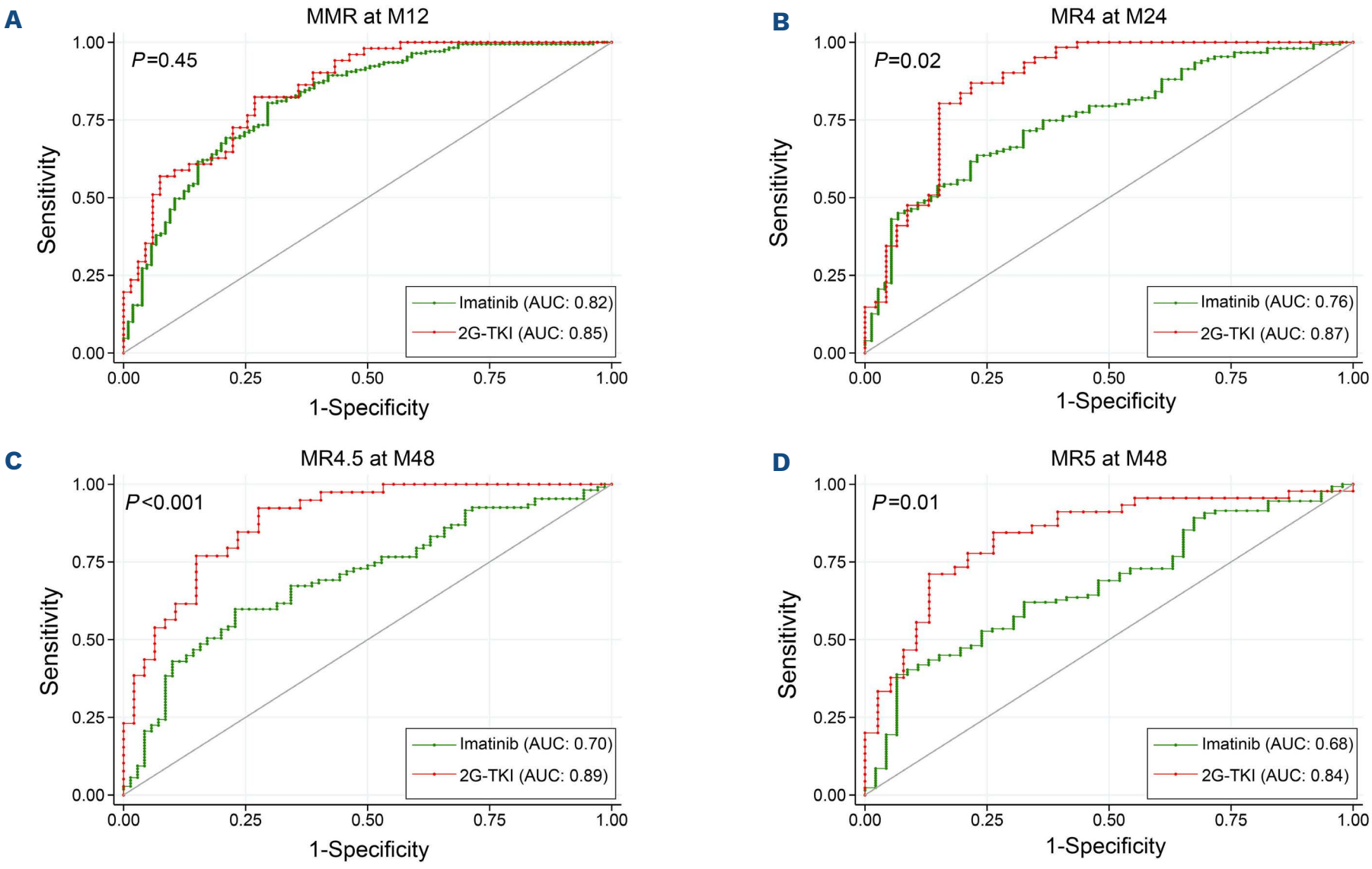
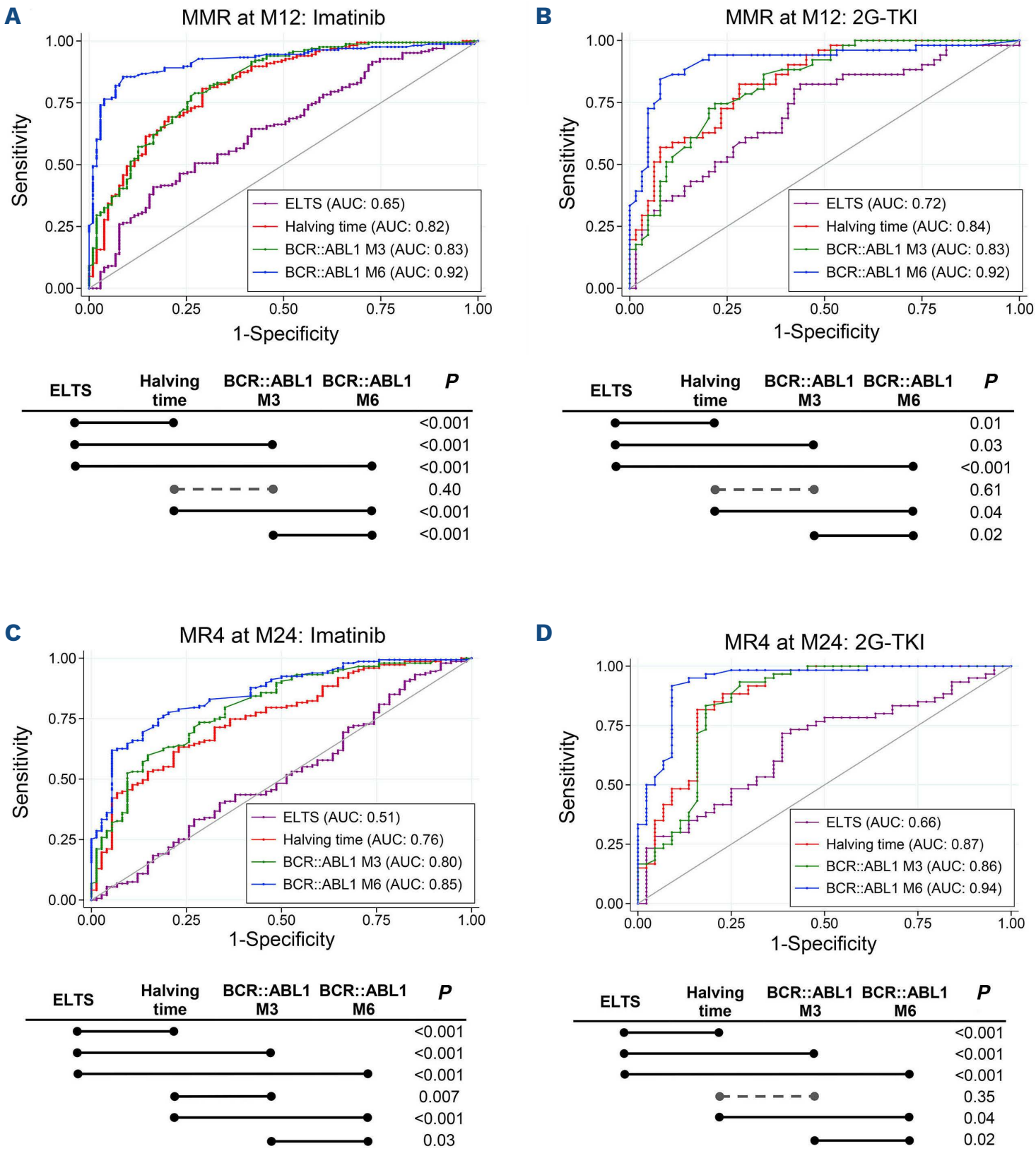


Figure 3. Receiver operating characteristic curves. Receiver operating characteristic (ROC) curves to evaluate the predictive performance for achieving the major molecular response (MMR) at month 12 (A, N=392 patients), molecular response 4-log reduction (MR4) at month 24 (B, N=332), 4.5-log reduction (MR4.5) (C, N=263) and 5-log reduction (MR5) (D, N=258) at month 48 of tyrosine kinase inhibitor (TKI) treatment in function of the first-line treatment (imatinib or second-generation tyrosine kinase inhibitor [2G-TKI] [nilotinib and dasatinib]). AUC: area under the curve; M3: month 3; M6: month 6; M12: month 12; M24: month 24; M48: month 48. The pairwise comparison (*P* value) is shown in each graph.

early therapeutic response parameters. The percentages of patients who subsequently changed TKI were broadly similar in the IMA and 2G-TKI subgroups (37.2% and 32.2%). The cumulative incidence analysis according to the first-line TKI confirmed the higher initial efficacy of 2G-TKI (*Online Supplementary Figure S2C-J*). Moreover, HT was shorter in the 2G-TKI subgroup than in the IMA subgroup, particularly in patients who reached each molecular response milestone at the relevant time point (*Online Supplementary Figure S2B-C*). Consequently, the ROC analyses showed that HT was a better predictor of MR4 at month 24, MR4.5 at month 48, and MR5 at month 48 in the 2G-TKI subgroup than in the IMA subgroup, but not of MMR at month 12 (Figure 3A-D). Overall, the relative performance of the four early parameters was equivalent in the two subgroups, with the exception of a higher performance of the ELTS score in the 2G-TKI group. The 6-month *BCR::ABL1* quantification was the best predictor of early therapeutic responses (MMR and MR4) in both subgroups (Figure 4A-H).

Concerning the achievement of the criteria for TKI discontinuation, all four early parameters performed poorly in the IMA subgroup (*P*=0.23) (Figure 5A). In the 2G-TKI subgroup, the ELTS score performed worst. HT and residual disease at month 3 and month 6 performed moderately and similarly (AUC values between 0.68 and 0.73) (Figure 5B). Despite the low number of patients who discontinued treatment (N=39 and N=31 in the IMA and 2G-TKI subgroups) and who relapsed (N=14/39 and N=14/31 in the IMA and 2G-TKI subgroups), TFR loss was associated only with intermediate (*vs.* low) ELTS score (HR=3.53, 95% CI: 2.42-5.16; *P*<0.001)



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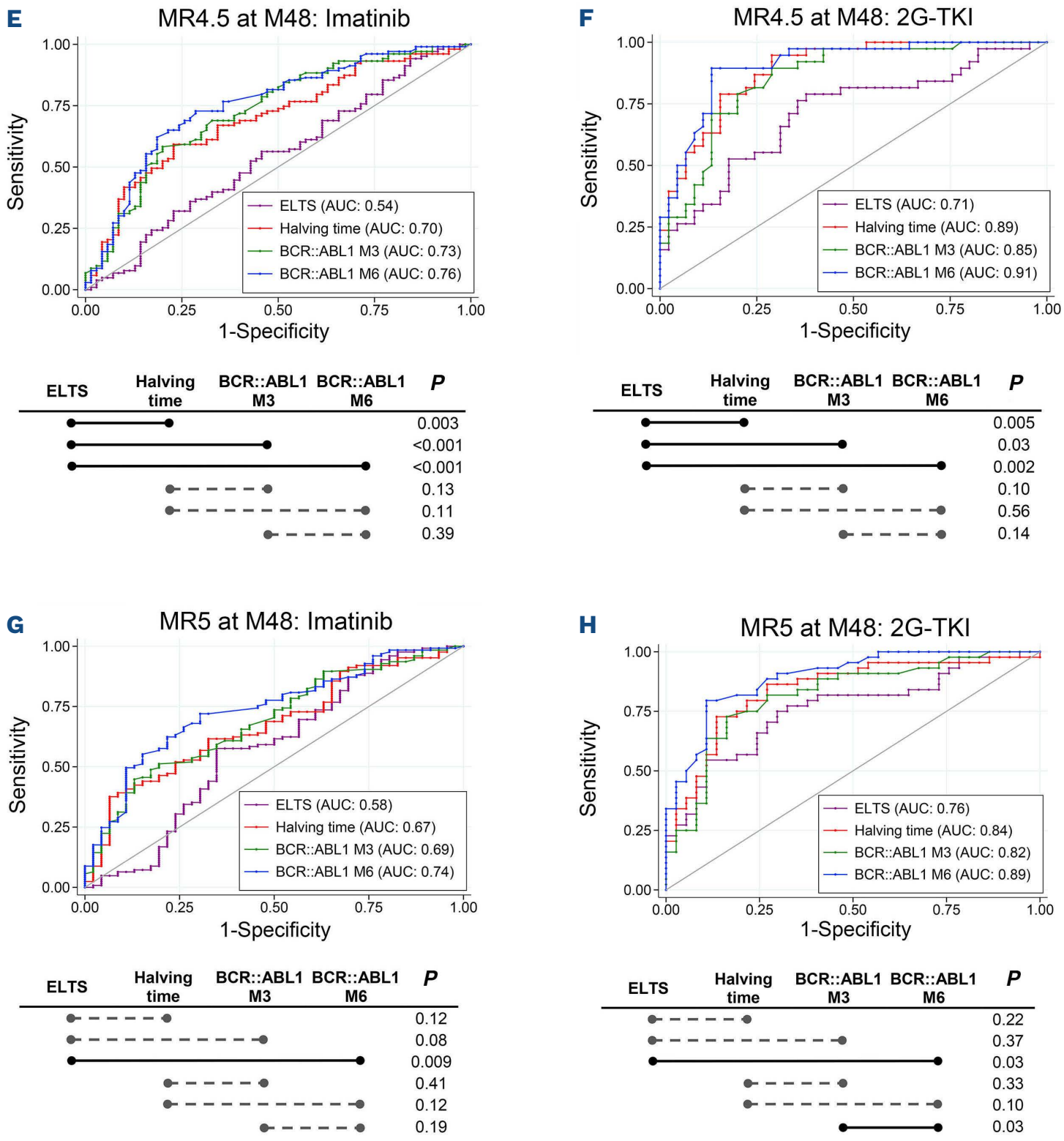

Figure 4. Receiver operating characteristic curves. Receiver operating characteristic (ROC) curves to compare the predictive performance of the 4 early parameters (i.e., EUTOS long-term survival score [ELTS] score, Halving time [HT], *BCR::ABL1/ABL1*^{IS} ratio at month 3 and 6) to achieve molecular response in function of the first-line treatment (imatinib or second-generation tyrosine kinase inhibitor [2G-TKI] [nilotinib and dasatinib]): major molecular response (MMR) at month 12 (A, N=269; B, N=115), 4-log reduction (MR4) at month 24 (C, N=221; D, N=104), 4.5-log reduction (MR4.5) (E, N=173; F, N=83) and 5-log reduction (MR5) (G, N=171; H, N=81) at month 48 of TKI) treatment in the imatinib and 2G-TKI subgroups, respectively. AUC: area under the curve; M3: month 3; M6: month 6; M12: month 12; M24: month 24; M48: month 48. The pairwise comparisons are shown below each panel (only if *P* value <0.05, Omnibus test), with black full lines indicating significant differences and gray dashed lines non-significant differences.

in the IMA subgroup and with female sex (HR=2.73, 95% CI: 1.40-5.34; *P*=0.003), HT (HR=2.69, 95% CI: 1.49-4.86; *P*=0.001) and *BCR::ABL1* level at month 3 (HR=1.63, 95% CI: 1.04-2.54; *P*=0.03) in the 2G-TKI subgroup.

Discussion

With the availability of particularly effective targeted therapies, new challenges have been defined for the personalized management of CP-CML, particularly the identification of early parameters predictive of the therapeutic response, achievement of the treatment discontinuation criteria and of TFR maintenance after TKI discontinuation.

In this study, we analyzed and compared for the first time the predictive performance of one prognostic parameter (ELTS score) and three early parameters related to the initial molecular response (HT, *BCR::ABL1* transcript quantification

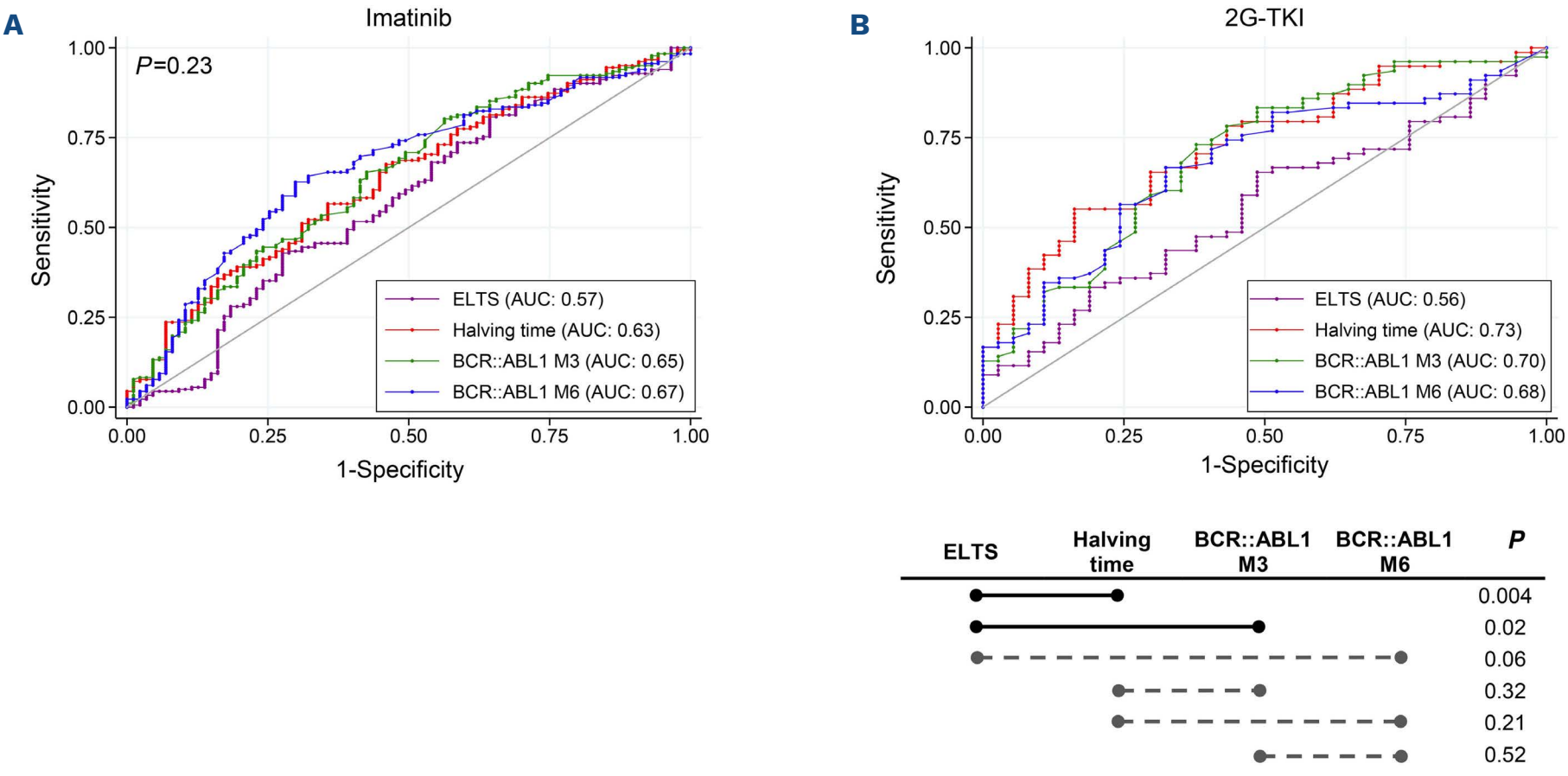

Figure 5. Receiver operating characteristic curves. Receiver operating characteristic (ROC) curves to compare the predictive performance of the 4 early parameters (i.e., EUTOS long-term survival score [ELTS], Halving time [HT], *BCR::ABL1/ABL1*^{IS} ratio at month 3 and 6) to meet the tyrosine kinase inhibitor (TKI) discontinuation criteria (A, N=269; B, N=115) in the imatinib and second-generation TKI (2G-TKI) (nilotinib and dasatinib) subgroups, respectively. AUC: area under the curve; M3: month 3; M6: month 6. The pairwise comparisons are shown below each panel (only if *P* value <0.05, Omnibus test), with black full lines indicating significant differences and gray dashed lines non-significant differences.

at months 3 and 6) for predicting the subsequent molecular response, the achievement of the TKI discontinuation criteria, and TFR maintenance after treatment discontinuation. In this real-life cohort from the French CML Observatory,[4] the ELTS score was the least effective parameter, confirming recent observations.[33] The ELTS score has been proposed as a first-line tool to better assess the risk of death.[22] It has been validated as the most effective prognostic score for patients treated with 2G-TKI and for predicting MMR, MR4 as well as overall, failure-free and progression-free survival in a real-life cohort.[24,25,34] However, in this first comparative study, it clearly performed less well than the initial assessments of the *BCR::ABL1* transcript. This helps to explain why some patients with high-risk score have a deep molecular response and others with an intermediate or low score have a sub-optimal therapeutic response.[24,25] It also suggests that the early assessment of the molecular response is more informative than prognostic scores for predicting the optimal molecular response.

In this cohort, we confirmed that the median HT was shorter in patients who achieved the different levels of molecular response than in those who did not. However, its predictive value is not better than that of residual disease at month 3 and at month 6, which therefore, remain essential milestones for the individual follow-up, confirming the ELN recommendations.[8] Among the four early parameters studied, residual disease at month 6 was the best parameter for predicting MR4.5 and MR5 at year 4 of treatment. The clinical benefit of assessing residual disease in the same patient at month 3 and 6 remains debated.[11,35,36] Our results strengthen the ELN and NCCN recommendations to confirm 1-3 months later any non-optimal response at month 3[8,37] and support the possibility of obtaining a MMR even in the event of an insufficient result at month 3.[21] In view of these results, we added also residual disease at month 12 in the comparative analysis. Although this analysis concerned a smaller subgroup, residual disease at month 12 was the best parameter for predicting optimal molecular responses, strengthening previous results.[33] This also reflects the growing importance of achieving therapeutic objectives during the first year, and indirectly the possibility of correcting a time point with a non-optimal therapeutic response at the next time point. As suggested recently, the requirement to reach the earliest molecular milestones could be made more flexible by integrating other factors (e.g., prognostic score, comorbidities, toxicities, dose adaptation) before changing treatment.[14]

As the initial *BCR::ABL1* transcript kinetics are influenced by the first-line TKI, we confirmed that 2G-TKI resulted in a more rapid molecular response and therefore, shorter HT compared with imatinib. Nevertheless, the performances of the four early parameters were globally similar in the 2G-TKI and IMA subgroups, with the exception of a tendency to improved performance of the ELTS score and a

weaker performance of residual disease at month 6 in the 2G-TKI subgroup.

On the other hand, all four early parameters were poor predictors of the achievement of the TKI discontinuation criteria (i.e., >5 years of TKI treatment and DMR >2 years. Notably, unlike for the analysis of the molecular response levels reached at defined times, we included all patients who reached the TKI discontinuation criteria, regardless of the time required to achieve this. Therefore, this subgroup included patients who reached a DMR later, in line with the observed progressive accumulation of responses over time.[4] Thus, despite their strong relationship with the overall treatment efficacy, the predictive value of the early parameters concerning the TKI discontinuation criteria was low.

Furthermore, analysis of patients who discontinued TKI after optimal response did not identify any relationship between the early quantification of residual disease, including HT, and the likelihood of TFR maintenance. Only the ELTS score was moderately associated with TFR maintenance. The low interest of *BCR::ABL1* transcript quantification at month 3 confirmed previous results,[38] but not the low interest of HT, even when divided in quartiles. Several differences between our study and that by Shanmuganathan *et al.*[17] should be considered: i) our cohort included more patients with high ELTS scores (12.3% *vs.* 6.4%), similarly to previous studies;[4,22,23] ii) the proportion of patients treated with first-line 2G-TKI was higher (28.9% *vs.* 16.8%); iii) the presence of a smaller subgroup of patients with TKI discontinuation (N=70 *vs.* N=115) and with longer post-discontinuation follow-up (at 12, 18 and 24 months *vs.* 12 months only) that allowed a more exhaustive collection of the relapse rate given the late relapses observed in this real-life cohort;[4] iv) the group of patients who discontinued TKI was smaller. This could represent a bias. However, we did not observe any major differences between this group and all patients who met the TKI discontinuation criteria (i.e., optimal responders). Moreover, a high percentage of patient who relapsed belonged to the first HT quartile group, arguing against any HT influence on TFR maintenance; and v) the comparative analysis with other early parameters using ROC curves; vi) the slower overall HT kinetics (17 *vs.* 14 days). Moreover, the HT difference was underestimated because we used the date of the first day of treatment and not the date of the initial *BCR::ABL1* transcript quantification. However, our cohort included a higher proportion of patients treated with 2G-TKI and therefore, with shorter median HT compared with imatinib (13 *vs.* 20 days). These differences may partly explain these contradictory results and it should be interesting to measure the HT in larger cohorts.

In our real-life cohort, the poor association between early molecular response parameters and the chances of reaching the criteria for TKI discontinuation or of remaining in TFR suggests that these early parameters remain of limited interest for deciding to stop TKI in real-life. Thus, while the criteria related to the initial kinetics of tumor mass reduction are essential for predicting the optimal response, once this response is obtained, predicting the maintenance of a deep response and the possibility of discontinuing TKI without relapse is no longer dependent on this early phase. This observation suggests the existence of a Markov-type process during the follow-up of TKI-treated patients with CP-CML that we summarized in a graphical abstract (Figure 6). Therefore, we lack biomarkers that are linked to the deep response phase, before stopping treatment, and that could be used to predict TFR maintenance. Future investigations might focus on the pharmacological and/or biological mechanisms involved in the long-term DMR maintenance and the eradication of the clone or at least of CML stem cells that are subtly different from those underlying the initial reduction of the leukemic clone. Indeed, we do not know what happens during the years when the residual disease is <0.1% before becoming undetectable, or during DMR maintenance before TKI discontinuation. This phase appears essential, as shown by the association between the duration of treatment and DMR and the maintenance of TFR.[39,40] Strong arguments have been accumulated in favor of the favorable influence of the lowest possible number of residual CML cells at the time of TKI cessation, as demonstrated by studies using digital PCR.[41,42] However, the question of the gradual elimination of immature CML progenitors during treatment, which is influenced by their cycling rate, the immune system involvement or the bone marrow microenvironment, remains open.[43] Research mainly focused on the period when treatment is stopped, but the mechanisms underlying treatment duration or DMR duration before TKI discontinuation remain unknown. Few initial parameters have been linked to TFR maintenance (ELTS score, age, type of transcript, telomeres), but they are of little use for personalizing treatment.[37] Identifying the parameters that predict TFR maintenance and that can be measured before treatment discontinuation remains a major challenge in CML.

Our study presents the limitations of a study based on retrospective data collected in real life, with patient recruitment left to the physicians' willingness. Nonetheless, the patient characteristics were broadly equivalent to those of clinical trial cohorts, suggesting the absence of a significant recruitment bias, probably mitigated by the multicenter organization. Moreover, the "intention to treat" approach did not take into account potential subsequent changes in treatment lines. However, the percentage of line changes was similar in all subgroups, and the sensitivity analyses on data from the group that did not change treatment or dose early gave similar results. Lastly, this study did not take into account dose adaptations because their analysis would have been difficult due to the many different therapeutic trajectories. Therefore, we proceeded on the principle that patient care and therapeutic adaptations in real life were decided pragmatically. Therefore, any treat-

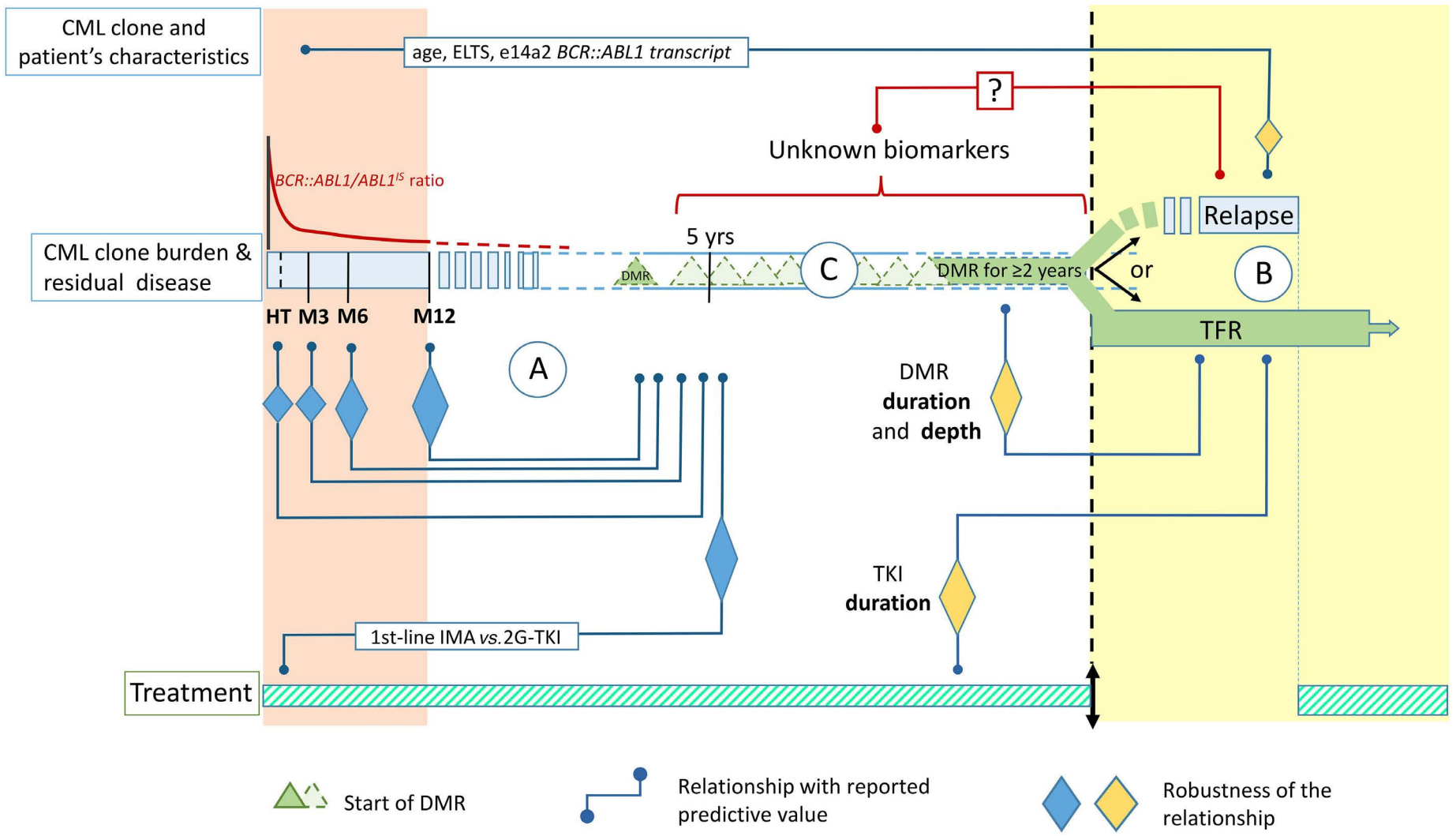

Figure 6. Summary diagram of the parameters with predictive value during the typical trajectory of a patient with chronic myeloid leukemia treated with tyrosine kinase inhibitor. The patient follow-up is schematized by showing the tumor burden changes and the residual disease, the achievement of a deep molecular response (DMR), and then the eventual tyrosine kinase inhibitor (TKI) withdrawal based on the discontinuation criteria. According to the results of this study, early parameters (i.e., Halving time [HT], *BCR::ABL1/ABL1*^{IS} ratio at month 3, 6 and 12) and the choice of first-line TKI can be used to predict DMR (A). The relationship is represented by a connector; its robustness is indicated with a proportional blue diamond. The parameters predicting treatment-free remission (TFR) maintenance after TKI discontinuation (i.e., DMR duration and depth, treatment duration, EUTOS long-term survival score (ELTS), age, e14a2 transcript) are indicated (B). The relationship is represented by a connector; its robustness is indicated with a proportional yellow diamond. Parameters relating to the period of therapeutic maintenance of the optimal molecular response are currently missing (C). CML: chronic myeloid leukemia; DMR: deep molecular response (MR4.5 or MR5); IMA: imatinib; M3: month 3; M6: month 6; M12: month 12; 2G-TKI: second generation TKI (nilotinib or dasatinib); yrs: years.

ment adaptation (change of line or dose adjustment) was considered to be based on the ELN recommendations or on the summary of the drug characteristics. Consequently, the analysis carried out implies that the early parameters of the therapeutic response were considered as the result of real-life care, regardless of the modifications made, and the analysis was focused on their predictive value related to the initial CML clone kinetics. The sensitivity analysis of the subgroup that did not change treatment supports this view.

In conclusion, in this real-life cohort, the results of this first comparative study of early therapeutic response parameters show that they are excellent markers of TKI efficacy and for identifying the best responders. They reinforce the current recommendations for monitoring residual disease during the first year. However, this study also highlights their inadequacy for predicting the achievement of TKI discontinuation criteria and TFR maintenance. The identification of parameters predictive of TFR maintenance remains a major challenge, and would require a more detailed analysis of patient trajectories before TKI cessation in large cohorts and better understanding the biological mechanisms associated with maintenance of the remission status.

Disclosures

No conflicts of interest to disclose.

Contributions

MGB designed the study, participated in the data analysis, and drafted the manuscript. SS is responsible for the LMC Observatory data, extracted the data, and participated in the analysis of the results and the writing of the manuscript. CL carried out all statistical analyses and participated in the writing of the manuscript. SS and CL contributed equally to the study. ED, GRG, FH, PCM, HJA, MEB, ATu, PR, EH and MGB were investigators in the partner centers of the CML Observatory, included patients, recorded data, and approved the submitted version of the manuscript. ATc verified the

molecular biology data. DH developed and maintains the computer database. BP supervised the statistical strategy. All authors validated the submitted manuscript.

Data-sharing statement

Agreement to share the data for non-commercial academic use upon request to the corresponding author.

References

1. Hochhaus A, Larson RA, Guilhot F, et al. Long-term outcomes of imatinib treatment for chronic myeloid leukemia. N Engl J Med. 2017;376(10):917-27.
2. Bower H, Björkholm M, Dickman PW, Höglund M, Lambert PC, Andersson TM-L. Life expectancy of patients with chronic myeloid leukemia approaches the life expectancy of the general population. J Clin Oncol. 2016;34(24):2851-2857.
3. Kantarjian HM, Hughes TP, Larson RA, et al. Long-term outcomes with frontline nilotinib versus imatinib in newly diagnosed chronic myeloid leukemia in chronic phase: ENESTnd 10-year analysis. Leukemia. 2021;35(2):440-453.
4. Saugues S, Lambert C, Daguenet E, et al. Real-world therapeutic response and tyrosine kinase inhibitor discontinuation in chronic phase-chronic myeloid leukemia: data from the French observatory. Ann Hematol. 2022;101(10):2241-2255.
5. García-Gutiérrez V, Hernández-Boluda JC. Tyrosine kinase inhibitors available for chronic myeloid leukemia: efficacy and safety. Front Oncol. 2019;9:603.
6. Vener C, Banzi R, Ambrogi F, et al. First-line imatinib vs second- and third-generation TKIs for chronic-phase CML: a systematic review and meta-analysis. Blood Adv. 2020;4(12):2723-2735.
7. Baccarani M, Deininger MW, Rosti G, et al. European LeukemiaNet recommendations for the management of chronic myeloid leukemia: 2013. Blood. 2013;122(6):872-884.
8. Hochhaus A, Baccarani M, Silver RT, et al. European LeukemiaNet 2020 recommendations for treating chronic myeloid leukemia. Leukemia. 2020;34(4):966-984.
9. Baccarani M, Cortes J, Pane F, et al. Chronic myeloid leukemia: an update of concepts and management recommendations of European LeukemiaNet. J Clin Oncol. 2009;27(35):6041-6051.
10. Marin D, Ibrahim AR, Lucas C, et al. Assessment of BCR-ABL1 transcript levels at 3 months is the only requirement for predicting outcome for patients with chronic myeloid leukemia treated with tyrosine kinase inhibitors. J Clin Oncol. 2012;30(3):232-238.
11. Neelakantan P, Gerrard G, Lucas C, et al. Combining BCR-ABL1 transcript levels at 3 and 6 months in chronic myeloid leukemia: implications for early intervention strategies. Blood. 2013;121(14):2739-2742.
12. Branford S, Yeung DT, Ross DM, et al. Early molecular response and female sex strongly predict stable undetectable BCR-ABL1, the criteria for imatinib discontinuation in patients with CML. Blood. 2013;121(19):3818-3824.
13. Gerds AT, Gotlib J, Ali H, et al. Myeloproliferative Neoplasms, Version 3.2022, NCCN Clinical Practice Guidelines in Oncology. J Natl Compr Canc Netw. 2022;20(9):1033-1062.
14. Senapati J, Sasaki K, Issa GC, et al. Management of chronic myeloid leukemia in 2023 - common ground and common sense. Blood Cancer J. 2023;13(1):58.
15. Narlı Özdemir Z, Kılıçaslan NA, Yılmaz M, Eşkazan AE. Guidelines for the treatment of chronic myeloid leukemia from the NCCN and ELN: differences and similarities. Int J Hematol. 2023;117(1):3-15.
16. Branford S, Yeung DT, Parker WT, et al. Prognosis for patients with CML and >10% BCR-ABL1 after 3 months of imatinib depends on the rate of BCR-ABL1 decline. Blood. 2014;124(4):511-518.
17. Shanmuganathan N, Pagani IS, Ross DM, et al. Early BCR-ABL1 kinetics are predictive of subsequent achievement of treatment-free remission in chronic myeloid leukemia. Blood. 2021;137(9):1196-1207.
18. Iriyama N, Fujisawa S, Yoshida C, et al. Shorter halving time of BCR-ABL1 transcripts is a novel predictor for achievement of molecular responses in newly diagnosed chronic-phase chronic myeloid leukemia treated with dasatinib: results of the D-first study of Kanto CML study group. Am J Hematol. 2015;90(4):282-287.
19. Fava C, Rege-Cambrin G, Dogliotti I, et al. Early BCR-ABL1 reduction is predictive of better event-free survival in patients with newly diagnosed chronic myeloid leukemia treated with any tyrosine kinase inhibitor. Clin Lymphoma Myeloma Leuk. 2016;16 Suppl:S96-S100.
20. Pennisi MS, Stella S, Vitale SR, et al. BCR-ABL1 doubling-times and halving-times may predict CML response to tyrosine kinase inhibitors. Front Oncol. 2019;9:764.
21. Cai Z, Jia X, Zi J, et al. BCR-ABL1 transcript decline ratio combined BCR-ABL1IS as a precise predictor for imatinib response and outcome in the patients with chronic myeloid leukemia. J Cancer. 2020;11(8):2234-2240.
22. Pfirrmann M, Baccarani M, Saussele S, et al. Prognosis of long-term survival considering disease-specific death in patients with chronic myeloid leukemia. Leukemia. 2016;30(1):48-56.
23. Hochhaus A, Baccarani M, Silver RT, et al. European LeukemiaNet 2020 recommendations for treating chronic myeloid leukemia. Leukemia. 2020;34(4):966-984.
24. Sato E, Iriyama N, Tokuhira M, et al. The EUTOS long-term survival score predicts disease-specific mortality and molecular responses among patients with chronic myeloid leukemia in a practice-based cohort. Cancer Med. 2020;9(23):8931-8939.
25. Zhang X-S, Gale RP, Huang X-J, Jiang Q. Is the Sokal or EUTOS long-term survival (ELTS) score a better predictor of responses and outcomes in persons with chronic myeloid leukemia receiving tyrosine-kinase inhibitors? Leukemia. 2022;36(2):482-491.
26. DeLong ER, DeLong DM, Clarke-Pearson DL. Comparing the areas under two or more correlated receiver operating characteristic curves: a nonparametric approach. Biometrics. 1988;44(3):837-845.
27. Liu X. Classification accuracy and cut point selection. Stat Med. 2012;31(23):2676-2686.
28. Feise RJ. Do multiple outcome measures require p-value adjustment? BMC Med Res Methodol. 2002;2:8.
29. Cayuela J-M, Chomel J-C, Coiteux V, et al. Recommandations du France Intergroupe des leucémies myéloïdes chroniques (Fi-LMC) pour l'examen des mutations du domaine kinase de

BCR-ABL1 dans la leucémie myéloïde chronique. Bull Cancer. 2020;107(1):113-128.
30. Kantarjian H, Shah NP, Hochhaus A, et al. Dasatinib versus imatinib in newly diagnosed chronic-phase chronic myeloid leukemia. N Engl J Med. 2010;362(24):2260-2270.
31. Hochhaus A, Saglio G, Hughes TP, et al. Long-term benefits and risks of frontline nilotinib vs imatinib for chronic myeloid leukemia in chronic phase: 5-year update of the randomized ENESTnd trial. Leukemia. 2016;30(5):1044-1054.
32. Cortes JE, Gambacorti-Passerini C, Deininger MW, et al. Bosutinib versus imatinib for newly diagnosed chronic myeloid leukemia: results from the randomized BFORE trial. J Clin Oncol. 2018;36(3):231-237.
33. Okamoto Y, Hirano M, Morino K, et al. Early dynamics of chronic myeloid leukemia on nilotinib predicts deep molecular response. NPJ Syst Biol Appl. 2022;8(1):39.
34. Geelen IGP, Sandin F, Thielen N, et al. Validation of the EUTOS long-term survival score in a recent independent cohort of "real world" CML patients. Leukemia. 2018;32(10):2299-2303.
35. Hwan Kim DD, Hamad N, Lee HG, Kamel–Reid S, Lipton JH. BCR/ABL level at 6 months identifies good risk CML subgroup after failing early molecular response at 3 months following imatinib therapy for CML in chronic phase. Am J Hematol. 2014;89(6):626-632.
36. Stella S, Zammit V, Vitale SR, et al. Clinical implications of discordant early molecular responses in CML patients treated with imatinib. Int J Mol Sci. 2019;20(9):2226.
37. Mikhaeel S, Atallah E. SOHO state of the art updates and next questions/update on treatment-free remission in chronic myeloid leukemia (CML). Clin Lymphoma Myeloma Leuk. 2023;23(5):333-339.
38. Chen K, Du T, Xiong P, Fan G, Yang W. Discontinuation of tyrosine kinase inhibitors in chronic myeloid leukemia with losing major molecular response as a definition for molecular relapse: a systematic review and meta-analysis. Front Oncol. 2019;9:372.
39. Saussele S, Richter J, Guilhot J, et al. Discontinuation of tyrosine kinase inhibitor therapy in chronic myeloid leukaemia (EURO-SKI): a prespecified interim analysis of a prospective, multicentre, non-randomised, trial. Lancet Oncol. 2018;19(6):747-757.
40. Dulucq S, Nicolini FE, Rea D, et al. Kinetics of early and late molecular recurrences after first-line imatinib cessation in chronic myeloid leukemia: updated results from the STIM2 trial. Haematologica. 2022;107(12):2859-2869.
41. Nicolini FE, Dulucq S, Boureau L, et al. Evaluation of residual disease and TKI duration are critical predictive factors for molecular recurrence after stopping imatinib first-line in chronic phase CML patients. Clin Cancer Res. 2019;25(22):6606-6613.
42. Atallah E, Schiffer CA, Radich JP, et al. Assessment of outcomes after stopping tyrosine kinase inhibitors among patients with chronic myeloid leukemia: a nonrandomized clinical trial. JAMA Oncol. 2021;7(1):42-50.
43. Patterson SD, Copland M. The bone marrow immune microenvironment in CML: treatment responses, treatment-free remission, and therapeutic vulnerabilities. Curr Hematol Malig Rep. 2023;18(2):19-32.