Prognostic significance of treatment response in CML in view of current recommendations for treatment and monitoring

Nikolas von Bubnoff

Abstract: The use of small-molecule kinase inhibitors has redefined the management of cancer. Chronic myelogenous leukemia (CML) has become the paradigm for targeted cancer treatment. Imatinib has become the gold standard in the treatment of CML with excellent and durable responses and minimal side effects. Molecular diagnostics constitute an integral part of the routine monitoring. Results of cytogenetic analysis and quantitative reverse transcriptase polymerase chain reaction (qRT-PCR) indicate suboptimal response or treatment failure and guide treatment. New Abl kinase inhibitors such as nilotinib or dasatinib are options after the failure of or intolerance to imatinib, and both are available for first-line treatment of newly diagnosed CML. This review focuses on the prognostic significance of achieving a response at specific time points in patients with CML treated with imatinib, nilotinib or dasatinib in view of available data and current treatment recommendations.

Keywords: CML, matinib, myeloid neoplasms

Introduction
The Philadelphia chromosome, first described as a shortened chromosome 22 [Nowell and Hungerford, 1960], results from a reciprocal translocation between the long arms of chromosomes 9 and 22 [Rowley, 1973], and is present in approximately 95% of chronic myelogenous leukemia (CML) patients and up to 20% of adult acute lymphoblastic leukemia (ALL) [Faderl et al., 1999; Sawyers, 1999]. The Philadelphia translocation gives rise to the oncogenic BCR-ABL fusion protein that is characterized by a constitutively active tyrosine kinase. BCR-ABL is sufficient to cause CML in mice [Daley et al., 1990], and its transforming capacity strictly depends on tyrosine kinase activity [Lugo et al., 1990]. This made BCR-ABL an attractive target for therapeutic intervention in CML and Ph+ ALL. The 2-phenylamino pyrimidine class of small-molecule kinase inhibitors was identified using a high-throughput screen of compound libraries at Ciba-Geigy (now Novartis) [Zimmermann et al., 1996]. The phenylamino pyrimidine CGP57148B (Imatinib-mesylate, hereafter imatinib), a derivative of the initial lead compound, was found to inhibit autophosphorylation of Abl and Bcr-Abl [Buchdunger et al., 2000; Beran et al., 1998; Zimmermann et al., 1997; Druker et al., 1996]. Preclinical studies demonstrated activity in Bcr-Abl positive cell lines and in animal models [le Coutre et al., 1999; Druker et al., 1996; Zimmermann et al., 1996]. Based on these observations, clinical trials in BCR-ABL positive CML were initiated in 1998. Phase 2 clinical trials demonstrated the activity of imatinib in chronic-phase as well as in accelerated-phase and blast crisis CML [Kantarjian et al., 2002a; Sawyers et al., 2002; Talpaz et al., 2002] and lead to the approval of imatinib for the treatment of CML in 2002. Activity was reported in patients with chronic phase CML and interferon (IFN) resistance or intolerance [Hochhaus et al., 2008b]. A phase 3 clinical trial (IRIS trial) documented the superiority of imatinib over IFN in combination with low-dose cytarabine in patients with newly diagnosed, untreated chronic phase CML with respect to hematologic, cytogenetic and molecular responses [Hughes et al., 2003; O’Brien et al., 2003], and also with respect to overall survival [Roy et al., 2006]. After 8 years, 55% of patients randomized to receive imatinib were still on study.
medication, and the overall survival rate for patients randomized to imatinib (intention to treat) was 85% or 93% when only CML-related deaths were considered [Deininger et al. 2009]. Imatinib side effects were mainly considered as mild or moderate [Druker et al. 2006; O’Brien et al. 2003]. Similar results were reported outside of the setting of a clinical trial [de Lavallade et al. 2008] and established imatinib 400 mg daily as standard treatment for patients with CML in the chronic phase [NCCN, 2010; Baccarani et al. 2009a]. While imatinib leads to sustained responses in the majority of chronic-phase CML cases, responses in advanced-phase CML are usually short lived [Sawyers et al. 2002; Talpaz et al. 2002]. Therefore, patients in blast crisis should proceed to allogeneic haematopoietic stem cell transplantation as soon as a haematologic response has been achieved [NCCN, 2010; Baccarani et al. 2009a]. For patients in accelerated phase, there is a correlation between complete cytogenetic response (CCyR) achieved with imatinib 600 mg per day (approximately 20% of patients [Palandri et al. 2009]) and survival for more than 5 years [Kantarjian et al. 2005]. These patients should be closely monitored, and in the case of a loss of haematologic or cytogenetic response, should be submitted to stem cell transplantation. For patients failing imatinib treatment, approved second-line Abl kinase inhibitors are available. Both nilotinib and dasatinib have been demonstrated to induce haematologic and cytogenetic responses in imatinib-intolerant and imatinib-resistant CML [Cortes et al. 2008; Hochhaus et al. 2008a; Guilhot et al. 2007; Kantarjian et al. 2007b; le Coutre et al. 2007] and have been approved for the treatment of imatinib-resistant or imatinib-intolerant CML. Moreover, both nilotinib and dasatinib in the setting of first-line treatment in chronic-phase CML demonstrated higher rates of cytogenetic and molecular responses at 12 months and lower rates of progression to accelerated or blast crisis [Kantarjian et al. 2010a; Saglio et al. 2010]. The availability of alternative ABL kinase inhibitors and the need for specialized diagnostic tools makes the clinical management of CML more complex. Prognostic factors have been identified that facilitate treatment decisions, and these include baseline as well as response-related factors.

**Assessing response in CML**

Determining the response to treatment is the prerequisite for the recognition of a treatment failure. According to the techniques used for monitoring, three levels of response can be discriminated (Figure 1) [Baccarani et al. 2006; Hughes et al. 2006; Talpaz et al. 1986]. With decreasing leukaemic burden, the primary finding will be the normalization of blood cell counts (haematologic response). Later on, the decrease of Philadelphia-positive metaphases in the bone marrow indicates cytogenetic response. It is important to note that the well-established association between cytogenetic response and positive outcomes (see the following) is based on conventional cytogenetic analysis of at least 25 bone marrow metaphases. Fluorescence in situ

![Figure 1.](http://tah.sagepub.com)
hybridization (FISH) can be used to assess cytogenetic response if marrow metaphases cannot be obtained, but as 200 peripheral blood interphase cells should be scored, low-level FISH positivity does not exclude CCyR established by conventional bone marrow cytogenetic analysis and therefore should not be used to define treatment failure. Molecular response is reflected by a decrease of $BCR-ABL$ transcripts in peripheral blood or bone marrow using quantitative real-time polymerase chain reaction (qRT-PCR) and is at least 3 logs more sensitive than conventional cytogenetics [Branford et al. 1999]. The magnitude of molecular response is expressed as the ratio of $BCR-ABL$ to a control gene or as log reduction compared with either the pretreatment value or a standard [Hughes et al. 2006]. Efforts of harmonizing the methodology for detection of $BCR-ABL$ transcripts and expressing results have lead to the calculation of lab-specific correction factors allowing conversion of PCR results into the internationally uniform and comparable international scale [Branford et al. 2006; Hughes et al. 2006]. There is a good correlation between bone marrow cytogenetics and peripheral blood transcript levels, with a $BCR-ABL$ ratio of 10% or less being equivalent to major cytogenetic response (MCyR) and 1% or less being equivalent to CCyR (Figure 1 and Table 1) [Ross et al. 2006]. The high sensitivity of PCR allows disease activity and treatment response below the level of CCyR to be monitored. In the case of a negative test result for quantitative PCR, nested PCR allows a qualitative detection of $BCR-ABL$ transcripts with a one log higher sensitivity compared with qRT-PCR [Hughes et al. 2006]. However, the presence of a complete molecular response (CMR; undetectable $BCR-ABL$ transcripts by qRT-PCR and/or nested PCR in two consecutive samples with sensitivity $>10^4$ [Baccarani et al. 2009a]) is also dependent on test sensitivity, which might vary between different assays and laboratories [Hughes et al. 2006].

### Molecular and cytogenetic diagnostics at the time of imatinib failure

Molecular mechanisms that frequently cause clinical resistance to imatinib include $BCR-ABL$ gene amplification and protein overexpression [Hochhaus et al. 2002; Gorre et al. 2001], clonal cytogenetic evolution [Cortes et al. 2003; Marktel et al. 2003; Hochhaus et al. 2002] and most importantly, mutations of the BCR-ABL kinase domain that lead to structural changes that affect drug binding [Branford et al. 2002; Hochhaus et al. 2002; Roche-Lestienne et al. 2002; Shah et al. 2002; von Bubnoff et al. 2002; Gorre et al. 2001]. Importantly, not only treatment failure itself but also molecular mechanisms leading to resistance can be identified by molecular diagnostic procedures that are routinely performed during treatment monitoring: conventional cytogenetic analysis (clonal cytogenetic evolution), FISH ($BCR-ABL$ gene amplification),

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<th>Table 1. Response definitions according to the ELN recommendations [Baccarani et al. 2009a].</th>
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<tr>
<td><strong>CHR (complete hematologic response)</strong></td>
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<tr>
<td><strong>Platelet count $&lt;450^9$/l</strong></td>
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<tr>
<td><strong>Differential without immature myeloid cells and $&lt;5%$ basophils</strong></td>
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<td><strong>Nonpalpable spleen</strong></td>
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<td><strong>Cytogenetic response</strong></td>
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<tr>
<td><strong>Partial (PCyR) 1–35% Ph$^+$</strong></td>
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<tr>
<td><strong>MajorCyR (MCyR) 0–35% Ph$^+$</strong></td>
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<tr>
<td><strong>Minor CyR 36–65% Ph$^+$</strong></td>
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<tr>
<td><strong>Minimal CyR 66–95% Ph$^+$</strong></td>
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<tr>
<td><strong>No CyR $&gt;95%$ Ph$^+$</strong></td>
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<tr>
<td><strong>Molecular response</strong></td>
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<td><strong>(BCR-ABL transcripts PB)</strong></td>
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CHR, complete haematologic response; CMR, complete molecular response; CCyR, complete cytogenetic response; CyR, cytogenetic response; MMR, major molecular response; PB, peripheral blood; PCR, polymerase chain reaction; pCyR, Partial cytogenetic response; WBC, white blood cell; qRT, quantitative real-time; Ph$^+$, Philadelphia positive.
denaturing high-performance liquid chromatography (DHPLC; screening for BCR-ABL gene mutations; sensitivity 0.1–10%) and conventional sequencing of the BCR-ABL kinase domain (sensitivity approximately 20%). Direct sequencing is sufficiently sensitive to detect clinically significant mutant leukaemia subpopulations, and at the present time still is considered as the technology of choice. DHPLC-based methods offer the advantage of sensitive, high-throughput capacity screening for multiple mutations at one time in an automated, cost-effective way [Deininger et al. 2004; Irving et al. 2004; Soverini et al. 2004]. This technology increasingly complements conventional sequencing. However, a mutation detected by HPLC still needs to be confirmed by conventional sequencing. Detection of low-level mutant disease clones using highly sensitive assays such as allele-specific PCR (sensitivity as low as 0.01%) is of low clinical significance.

Sequencing analysis for the presence of BCR-ABL mutations that mediate inhibitor resistance should be initiated in the case of treatment failure and in the case of repeated qRT-PCR results demonstrating an increase of BCR-ABL transcripts. The factor increase that should trigger mutation analysis is not yet established [Kantarjian et al. 2009; Press et al. 2007; Hughes et al. 2006]. If a rise in transcripts is due to expansion of a mutant clone, the proportion of transcripts carrying the mutation might very well be above the threshold of detection of conventional sequencing although the absolute transcript level might be low.

**Prognostic significance of biologic, molecular and cytogenetic parameters at baseline**

In CML, initial workup at the time of diagnosis gives information that is of prognostic relevance and predictive with respect to response to treatment and overall survival (see Table 2).

First, disease phase at diagnosis predicts both response to imatinib and overall survival. In chronic phase, overall survival for patients randomized to first-line imatinib in the IRIS trial (intention to treat) at 8 years was 85% or 93% when only CML-related deaths were considered [Deininger et al. 2009]. In contrast, overall survival was 53% at 4 years and 43% at 7 years, respectively, in the accelerated phase [Palandri et al. 2009; Kantarjian et al. 2005], and 11% at 3 years in blast crisis [Palandri et al. 2008].

Second, in chronic-phase CML the baseline prognostic scores proposed by Sokal and colleagues [Sokal et al. 1984] and Hasford and colleagues [Hasford et al. 1998] are predictive for response and overall survival in patients treated with imatinib [Baccarani et al. 2009b; Marin et al. 2008; Druker et al. 2006; Hughes et al. 2003]. Both scores were developed in the era of hydroxyurea/busulfan (Sokal) and IFN (Hasford) and in terms of biology at least in part reflect features of accelerated-phase disease. In the IRIS trial, CCyR rates at 12 months for Sokal low, intermediate and high patients were 78%, 68% and 51%, respectively [Hughes et al. 2003], and overall survival at 6 years was 94%, 87% and 76%. However, pretreatment Sokal score did not significantly affect outcome, once a CCyR was achieved [Druker et al. 2006]. Interestingly, first-line treatment of chronic phase CML with novel Abl kinase inhibitors such as nilotinib or dasatinib might at least in part overcome the adverse effect of intermediate or high-risk Sokal score. Looking at the ENEST phase 3 trial, which compares imatinib 400 mg OD to nilotinib at 300 mg and 400 mg BID in newly diagnosed chronic-phase CML, CCyR rates at 12 months among patients with Sokal high-risk score were 74% with nilotinib at 300 mg BID, 63% among those receiving 400 mg of nilotinib BID, and 49% with imatinib, and MMR rates were 41% for 300 mg nilotinib BID, 32% for 400 mg nilotinib BID, and 17% with imatinib 400 mg [Saglio et al. 2010]. Similar results were observed in the DASISION phase 3 trial, which compares imatinib 400 mg daily with dasatinib 100 mg daily in newly diagnosed chronic phase CML. Here, the rates of CCyR by 12 months among patients with Hasford high-risk score were 78% with dasatinib

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<th>Factor</th>
<th>Prognostic significance</th>
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<tr>
<td>Disease phase</td>
<td>HR, CR, PFS, OS</td>
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<tr>
<td>Sokal, Hasford score*</td>
<td>CR, PFS, OS</td>
</tr>
<tr>
<td>CCA Ph+</td>
<td>PFS, OS</td>
</tr>
<tr>
<td>hOCT-1 activity*</td>
<td>CR, MR, PFS, OS</td>
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*Measured in peripheral blood mononuclear cells, not routinely available.

CCA Ph+, clonal chromosomal abnormalities in Philadelphia-positive cells; CR, cytogenetic response; HR, haematologic response; MR, molecular response; PFS, progression-free survival; OS, overall survival.
and 64% with imatinib, and the corresponding MMR rates at 12 months were 31% with dasatinib and 16% with imatinib [Kantarjian et al. 2010a].

Third, attempts have been made to correlate disease-specific molecular and cytogenetic features at baseline with outcome. In particular, 5–10% of patients with chronic-phase CML present with clonal chromosomal abnormalities (CCAs) in Ph+ cells [Sokal et al. 1988; Kantarjian et al. 1985; Swolin et al. 1985]. The finding of CCAs at baseline or in the course of treatment was found to be associated with disease progression and inferior overall survival in the pre-imatinib era [Sokal et al. 1988; Kantarjian et al. 1985; Swolin et al. 1985], as well as in CML patients in chronic and accelerated phase treated with imatinib [Marin et al. 2008; Cortes et al. 2003; Marktel et al. 2003], although response to imatinib therapy in one study was a stronger independent predictor for survival than clonal evolution at diagnosis [Cortes et al. 2003]. Current recommendations define CCAs as a ‘warning’ at baseline and as treatment failure during imatinib treatment [Baccarani et al. 2009a]. The significance of CCA in Ph- cells is less clear. CCAs in Ph- cells may occur in a transient manner, and current recommendations identify CCAs in Ph-cells as a ‘warning’ feature, based on rare instances of progression to myelodyplasia or acute myeloid leukaemia [Zaccaria et al. 2010, 2007; Baccarani et al. 2009a; Abruzzese et al. 2007; Deininger et al. 2007; Jabbour et al. 2007; Kovitz et al. 2006]. Recently, the human organic cation transporter-1 (hOCT-1) was found to be involved in the imatinib import into the cell [Thomas et al. 2004]. Three studies demonstrated a correlation of hOCT-1 gene expression or activity in CML mononuclear cells at baseline and response to imatinib [Marin et al. 2010; White et al. 2007b; Crossman et al. 2005], and baseline hOCT-1 activity was also found to be associated with overall and progression-free survival [White et al. 2010]. In addition, it has been demonstrated that low hOCT-1 activity can be overcome by increasing the imatinib dose [White et al. 2010, 2007b]. These data suggest that patients with low baseline hOCT-1 activity in mononuclear cells might benefit from imatinib dose increase or treatment with nilotinib or dasatinib, which both do not utilize hOCT-1 [Giannoudis et al. 2008; Hiwase et al. 2008; White et al. 2006]. Thus, determination of hOCT-1 activity should be prospectively evaluated in future clinical trials and might be incorporated in future treatment recommendations. Other pharmacologic markers that have been proposed to be of prognostic significance and/or predictive for response include measurements of plasma and intracellular imatinib levels. Plasma levels of imatinib and its major metabolite N-desmethyliimatinib can be measured using HPLC and mass spectrometry [Larson et al. 2008; Picard et al. 2007; Titier et al. 2005]. Several reports investigated the significance of imatinib plasma levels for response. In two reports, higher imatinib plasma levels correlated with achieving CCyR and major molecular response (MMR), and in one report not surprisingly also with adverse events [Larson et al. 2008; Picard et al. 2007], while one report did not find such an association [Forrest et al. 2009]. Interestingly, imatinib plasma levels did also correlate with clinical benefit (complete response, partial response, stable disease, time to progression) in patients with unresectable or metastatic gastrointestinal stromal tumours (GISTs) [Demetri et al. 2009]. Both in CML as well in GIST patients, a mean trough imatinib plasma level of approximately 1000 ng/ml or above was associated with response [Demetri et al. 2009; Larson et al. 2008; Picard et al. 2007]. Thus, monitoring of imatinib plasma concentrations in patients with CML might be useful in poorly responding patients, in patients with suspected insufficient compliance and in patients with unusually severe toxicity. However, in chronic-phase CML patients with suboptimal response to or failure of imatinib and documented ‘suboptimal’ plasma levels, it is currently not clear whether increasing the imatinib dose would be equivalent to switching the treatment to nilotinib or dasatinib. The measurement of intracellular imatinib levels or inhibition of BCR-ABL would integrate drug absorption, tissue distribution, drug influx and efflux in leukaemic cells. One study reported a correlation of dephosphorylation of CRKL (which is phosphorylated by BCR-ABL and thus indirectly reflects BCR-ABL kinase activity) measured in peripheral blood mononuclear cells, and the subsequent MMR rate in chronic phase patients treated with imatinib at 600 mg daily [White et al. 2007a]. However, both intracellular measurement of nonradiolabeled imatinib and measurement of p-CRKL is technically challenging and thus implementation in daily routine seems unrealistic. In addition to pharmacokinetic markers and predictors of response, specific gene...
expression signatures of leukaemic cells at baseline might be associated with treatment response or progression [Diaz-Blanco et al. 2007; Frank et al. 2006; Radich et al. 2006; Villuendas et al. 2006; Zheng et al. 2006]. However, the prognostic or predictive value of gene expression studies remains to be determined. As stated above, highly sensitive PCR assays allow the detection of low-level (as low as 0.01%) BCR-ABL kinase domain mutations associated with imatinib resistance [Ernst et al. 2009]. However, even in the setting of accelerated phase or blast crisis, low-level detection of mutations neither at baseline nor in patients responding to imatinib treatment without concomitant rise in BCR-ABL transcript levels correlated with response, relapse, progression-free survival or overall survival [Sherbenou et al. 2007; Willis et al. 2005; Roche-Lestienne et al. 2002]. Thus, mutational analysis at baseline or in patients responding to treatment is not useful.

**Long-term survival in CML requires cytogenetic response**

Hydroxyurea and busulfan can induce haematologic but not cytogenetic responses in chronic-phase CML [Hehlmann et al. 2003; The Italian Cooperative Study Group on Chronic Myeloid Leukemia, 1998]. A meta-analysis demonstrated a survival benefit at 4 years for hydroxyurea compared with busulfan (53.6% versus 45.1%) [Chronic Myeloid Leukemia Trialists’ Collaborative Group, 2000]. Later on, treatment with alpha IFN alone or in combination with low-dose cytarabine-induced haematologic responses in 70–80% of cases and durable CCyRs (for the definition see Table 1) in 5–15% of cases, with higher rates of cytogenetic responses achieved by IFN in combination with low-dose cytarabine [Baccarani et al. 2002; Bonifazi et al. 2001; Guilhot et al. 1997; Hehlmann et al. 1994]. A survival advantage of IFN-based therapy to hydroxyurea or busulfan was demonstrated in a meta-analysis (5-year survival 57% versus 42%) [Chronic Myeloid Leukemia Trialists’ Collaborative Group, 1997], and survival at 10 years was 72% with IFN-based therapy when a CCyR was attained [Bonifazi et al. 2001]. Thus, already in the pre-imatinib era, long-term survival in nontransplant patients was linked to cytogenetic response.

It was therefore not unexpected to see that in patients with chronic-phase CML treated with imatinib, outcome was determined by haematologic and cytogenetic response to first-line imatinib at specific time points. Based on these observations, recommendations for the management and monitoring of CML patients receiving imatinib were developed [NCCN, 2010; Baccarani et al. 2009, 2006; Hochhaus et al. 2006], including criteria defining suboptimal response and treatment failure which are based on haematologic and cytogenetic response to first-line imatinib in patients with chronic-phase CML (Table 3).

**Table 3. ELN response criteria** [Baccarani et al. 2009a] and prognostic significance of suboptimal response or treatment failure at 3, 6, 12 and 18 months imatinib in early chronic-phase chronic myelogenous leukaemia.

<table>
<thead>
<tr>
<th>Time</th>
<th>Failure</th>
<th>Suboptimal response</th>
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<tr>
<td>3 months</td>
<td>&lt;CHR*</td>
<td>No CR</td>
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<tr>
<td>6 months</td>
<td>No CR</td>
<td>&lt;PCyR*</td>
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<td>12 months</td>
<td>&lt;PCyR*</td>
<td>&lt;CCyR*</td>
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<tr>
<td>18 months</td>
<td>&lt;CCyR*</td>
<td>&lt;MMR</td>
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<tr>
<td>Anytime</td>
<td>Loss of CHR*</td>
<td>Loss of MMR</td>
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<tr>
<td></td>
<td>Loss of CCyR*</td>
<td>BCR-ABL mutation§</td>
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<td></td>
<td>BCR-ABL Mutation§</td>
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<td></td>
<td>CCA Ph+*</td>
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*Low probability of subsequent CCyR.
<Predictive for progression-free survival.
>Predictive for overall survival [see the text for details].
>Exchange poorly imatinib sensitive.
>Exchange imatinib sensitive.
CHR, complete haematologic response; CCA Ph+, clonal chromosomal abnormalities in Philadelphia-positive cells; CCyR, complete cytogenetic response; CR, cytogenetic response; MMR, major molecular response; PCyR, partial cytogenetic response.

**Prognostic significance of response to imatinib in CML**

Importantly, it became evident very soon that most criteria for suboptimal response and treatment failure at 3, 6, 12 and 18 months in chronic-phase CML patients receiving imatinib as defined by the original and revised European Leukemia Net (ELN) recommendations [Baccarani et al. 2009a, 2006] indeed proved to identify patients with inferior outcome with respect to overall survival, progression-free survival, gain of a CCyR, or loss of a previously achieved CCyR. Haematologic imatinib failures in early chronic phase are rare (<5% of cases) [Marin et al. 2008; O’Brien et al. 2003]. In contrast, primary cytogenetic failures are more prevalent and were reported to occur in 3–18% at 6 months [Marin et al. 2008], 15–27% at 12 months [Marin et al. 2008; Druker et al. 2006] and
23–49% at 18 months [Marin et al. 2008; Roy et al. 2006]. Lack of a complete haematologic response (CHR) at 3 months and lack of any cytogenetic response (Ph+ >95%) at 6 months, lack of a partial cytogenetic response (PCyR; Ph+ >35%) at 12 months, and lack of a CCyR (Ph+ ≥1%) at 18 months indicate primary haematologic or cytogenetic failure (see also Table 3) and were associated with inferior progression-free survival [Kantarjian et al. 2008; Marin et al. 2008; Druker et al. 2006; Roy et al. 2006]. Patients without any cytogenetic response (>95% Ph+) at 3 and 6 months had a low probability of achieving a CCyR later on [Alvarado et al. 2009; de Lavallade et al. 2008]. Looking at the IRIS study, for patients not achieving a MCyR at 12 months (failure according to ELN), a significant disadvantage was also seen for overall survival and moreover, a similar significant disadvantage was seen for patients failing CCyR after 12 months (suboptimal response according to ELN) [Roy et al. 2006]. A single institution analysis performed by the Hammersmith group confirmed these findings demonstrating that treatment failure at 3, 6 and 12 months affected overall survival [Marin et al. 2008]. In addition, it was shown that suboptimal responders either at 6 months (less than PCyR) or 12 months (less than CCyR, see Table 3) had a significantly inferior progression-free survival, a lower probability of CCyR, and in the case of 12 months suboptimal responders also worse overall survival after 5 years (98% versus 85%). Similar findings were reported from a single-institution analysis performed by the Houston group [Kantarjian et al. 2008], although this analysis mainly included patients receiving an increased imatinib dose of 800 mg instead of the standard dose of 400 mg. Looking at the 18 months time point, failure to achieve a CCyR constitutes a treatment failure according to ELN, and in the IRIS study was associated with inferior overall survival after 8 years (74.9% versus 94.9%) [Deininger et al. 2009]. These findings substantiate the importance for early monitoring and demonstrate the prognostic significance of the ELN response criteria. Criteria for suboptimal response are very close to failure with respect to prognostic significance, and therefore the consequence in either case will rather be a switch to nilotinib or dasatinib than a dose increase of imatinib.

Acquired treatment failure (secondary resistance) denotes loss of a previously achieved haematologic or cytogenetic remission or progression to accelerated phase or blast crisis despite continued imatinib treatment [Baccarani et al. 2009a]. In early chronic-phase CML patients receiving imatinib treatment, the annual rates of secondary resistance (loss of CHR, loss of MCyR, progression to accelerated phase/blast crisis) or death in the IRIS study continuously decreased from the second (7.5%) to the sixth year (0.4%) and was 1.3% in the eighth year, including 2.8%, 0% and 0.4% with progression to accelerated phase or blast crisis at the second, sixth and eighth year [Deininger et al. 2009]. Looking at all of the 457 IRIS patients who achieved a CCyR, 82 patients (18%) had a documented loss of CCyR during treatment and 15 (3%) progressed to accelerated phase/blast crisis [Deininger et al. 2009]. Any loss of haematologic or cytogenetic response predicted shorter progression-free and overall survival [Marin et al. 2008].

Several studies have established a correlation between achieving a MMR (BCR-ABL ratio ≤0.1%) and improved durations of CCyR [Marin et al. 2008; Press et al. 2007; Iacobucci et al. 2006; Cortes et al. 2005]. In the IRIS study, the probability of loss of CCyR at 7 years was 3% for patients in MMR at 18 months compared to 26% for patients with CCyR without MMR [Hughes et al. 2010]. Two studies demonstrated a small, but significant advantage in progression-free survival for patients achieving a MMR at 12 [Hughes et al. 2010], or 18 months [Kantarjian et al., 2008, Hughes et al., 2010]. However, achieving MMR at 12 or 18 months was not demonstrated to be predictive for overall survival [Hughes et al. 2010; Kantarjian et al. 2008; Marin et al. 2008; Druker et al. 2006; Iacobucci et al. 2006; Cortes et al. 2005]. These findings might well indicate that MMR can be regarded as a ‘safe haven’ [Hughes et al. 2008], and suggest that cytogenetic analysis is dispensable once a CCyR has been attained and confirmed, provided that regular molecular monitoring can be assured [Baccarani et al. 2009a; Ross et al. 2006].

In advanced-phase CML, primary haematologic failure occurs more frequently and in phase 2 clinical trials was reported in 18–30% of patients with accelerated phase and in 60% of patients with blast crisis. After 4 years, resistance to imatinib had emerged in 45–70% (accelerated phase) and 90% (blast crisis) [Palandri et al. 2009; Kantarjian et al. 2005, 2002b, 2002c;
Importantly, in accelerated phase approximately 20% of patients treated with imatinib at 600 mg daily achieve a CCyR [Palandri et al. 2009], and these patients in phase 2 trials survived more than 5 years [Palandri et al. 2009; Kantarjian et al. 2005]. Thus, accelerated-phase patients achieving CCyR should be continuously monitored and should be submitted to allogeneic transplant if cytogenetic or haematologic response is lost.

Together, haematologic and cytogenetic response determine progression-free and overall survival, and regular monitoring of imatinib treatment in CML is indispensable to confirm adequate response and to identify patients with suboptimal response or treatment failure early enough to make appropriate treatment changes [Baccarani et al. 2009a]. Patients who continue with imatinib despite the lack of cytogenetic response face the risk of progression to accelerated phase and blast crisis. In contrast, achieving a CCyR is associated with excellent progression-free survival, provided that imatinib is continued without dose reduction or interruptions [Kantarjian et al. 2008; Marin et al. 2008; Druker et al. 2006; Roy et al. 2006; O’Brien et al. 2003]. These findings underscore the importance of a regular monitoring particularly in the beginning of treatment. However, treating physicians and patients must also be aware that nonadherence to imatinib is prevalent [Darkow et al. 2007]. Poor adherence might be a frequent cause for suboptimal outcome in CML treated with oral tyrosine kinase inhibitors, and in the setting of stable CCyR in chronic-phase CML patients receiving imatinib was a strong predictor of not achieving a subsequent MMR [Marin et al. 2010]. Moreover, improving adherence may not only optimize outcomes of treatment but also reduce the economic burden, since adherence was found to be inversely associated with healthcare costs excluding imatinib [Darkow et al. 2007].

New BCR-ABL inhibitors after imatinib failure: baseline and response-related prognostic factors

The finding of clinical resistance to imatinib triggered the development of novel ABL kinase inhibitors. Preclinical models revealed a higher inhibitory activity of these drugs against wild-type BCR-ABL in cell lines and animal models, and also demonstrated activity of these novel compounds against many of the known imatinib-resistant BCR-ABL exchanges. Examples include nilotinib (AMN107) [Weisberg et al. 2005] and dasatinib (BMS354825) [Shah et al. 2004]. Both compounds have been demonstrated to induce haematologic responses in imatinib-intolerant and imatinib-resistant CML [Kantarjian et al. 2011; Apperley et al. 2009; Cortes et al. 2008; Hochhaus et al. 2008a; le Coutre et al. 2008; Giles et al. 2007;] and demonstrated superior rates of cytogenetic and molecular responses at 12 months compared with imatinib given as first-line treatment in chronic-phase CML [Kantarjian et al. 2010a; Saglio et al. 2010]. Both compounds have been approved for the treatment of imatinib-resistant or imatinib-intolerant CML, and recently also for the first-line treatment of CML. Baseline prognostic factors with respect to response to nilotinib or dasatinib have not been identified for patients with imatinib-intolerant disease but, however there is evidence for baseline prognostic factors in imatinib-resistant patients. Specifically, one study established and validated a scoring system based on cytogenetic response to imatinib, Sokal score and recurrent neutropenia during imatinib treatment [Milojkovic et al. 2010]. The three resulting risk categories predicted a cumulative incidence of CCyR at 2.5 years of 100%, 52.2% and 13.8% [Milojkovic et al. 2010]. In a second study, three risk categories based on previous cytogenetic response to imatinib, and performance status predicted the 12-month probability of achieving a MCyR to nilotinib or dasatinib (64%, 36% and 20%), and in addition discriminated 2-year rates for event-free survival (78%, 49% and 20%) and overall survival (95%, 85% and 40%) [Jabbour et al. 2011]. In addition, it was demonstrated that haematologic resistance to imatinib, clonal cytogenetic evolution, and the presence of specific, but not any BCR-ABL kinase domain mutations mediating imatinib resistance affects response to nilotinib and dasatinib [Milojkovic et al. 2010; Hochhaus et al. 2008a; Shah et al. 2008; Tam et al. 2008; Kantarjian et al. 2007a]. Both inhibitors display a largely nonoverlapping profile of resistance mutations that well correlates to mutations that were predicted by in vitro studies [Ray et al. 2007; Braden et al. 2006; von Bubnoff et al. 2006; Burgess et al. 2005; O’Hare et al. 2005]. Of note, two studies demonstrated that patients with history of a BCR-ABL resistance mutation might have a higher likelihood of developing further mutations receiving second- and third-line
tyrosine kinase inhibitors, and noted that some patients fail several lines of Abl tyrosine kinase inhibitors without ever developing a BCR-ABL kinase domain mutation [Soverini et al. 2009; Cortes et al. 2007], probably reflecting a difference in biology of the disease. Regarding response to nilotinib or dasatinib after imatinib failure, two studies demonstrated that the probability of achieving a CCyR is low if a patient does not achieve any cytogenetic response at 3 months and less than a minor cytogenetic response at 6 months [Milojkovic et al. 2010; Tam et al. 2008]. Both studies included patients in chronic phase, with 28% and 23% of the patients displaying additional cytogenetic abnormalities. In addition to the predictive value of cytogenetic response at 3 and 6 months for achieving a subsequent CCyR, a significant progression-free survival and overall survival advantage was demonstrated for patients achieving at least a minor cytogenetic response at 3 months [Milojkovic et al. 2010] and a MCyR at 6 [Milojkovic et al. 2010] or 12 months [Tam et al. 2008], respectively. Thus, both studies demonstrate that early cytogenetic response to nilotinib or dasatinib after imatinib failure predicts outcome. For patients with CML in advanced phase or Ph+ ALL treated with nilotinib or dasatinib after imatinib failure, a recent analysis identified patients failing a complete haematologic response at the time of achievement of a MCyR due to incomplete neutrophil or platelet recovery to have a particular poor outcome, pointing to the prognostic significance of recovery of Ph- haematopoiesis [Fava et al. 2009]. Overall survival at 2 years in these patients was similar to that of patients failing a MCyR.

**Imatinib versus new BCR-ABL inhibitors as first-line treatment**

As outlined above, the outcome of chronic-phase CML patients treated with imatinib is determined by response to treatment. A patient who rapidly achieves CCyR or MMR will have a lower risk of treatment failure. Second, although long-term outcome with imatinib in chronic-phase CML is impressive, one has to be aware that approximately 25% of patients in chronic-phase CML fail imatinib, and only 63% of the patients in the IRIS study were in CCyR and on study treatment after 6 years [Hochhaus et al. 2009]. As already pointed out, not achieving a CCyR determines inferior progression-free and overall survival, and of all patients in the IRIS study who achieved CCyR, 18% lost CCyR, and 3% progressed to accelerated phase or blast crisis [Deininger et al. 2009]. With these presumptions in mind, it might well be that superior rates of CCyR and MMR observed at 12 months with nilotinib [Saglio et al. 2010] or dasatinib [Kantarjian et al. 2010a] given as first-line treatment in chronic-phase CML will translate into long-term outcomes superior to imatinib. In the short term, both drugs reduced the rate of early progression to accelerated phase or blast crisis (in the case of dasatinib not yet statistically significant). On the other hand, it will be interesting to analyse whether baseline prognostic parameters (as described previously) allow the identification of subgroups that either do or do not gain benefit from frontline treatment with nilotinib or dasatinib. In addition, it might well be that treatment can be initiated with imatinib, and early response monitoring allows the identification of patients at risk for failing treatment which then would be switched to nilotinib or dasatinib. Unfortunately, there will probably never be a trial comparing the strategy of switching suboptimal imatinib responders/failures versus upfront nilotinib or dasatinib. Thus, treating physicians and patients might discuss and decide on an individual basis, taking into account specific circumstances and probably also costs. Some patients with ongoing complete molecular responses may be able to discontinue imatinib without subsequent relapse [Mahon et al. 2010]. Thus, long-term costs do not necessarily need to be higher with nilotinib or dasatinib, since rapid and deep remissions might lead to a higher proportion of patients qualifying for investigational treatment discontinuation. However, discontinuation of any Abl kinase inhibitor is not recommended outside the setting of a clinical trial. Alternative frontline treatment strategies might include induction-maintenance concepts with initial debulking using nilotinib or dasatinib and response-guided maintenance treatment using continued nilotinib or dasatinib, imatinib or alpha IFN, depending on the pace and quality of response. The Italian GIMEMA is currently examining the feasibility and response rates of a rotating regime of nilotinib and imatinib for frontline treatment of CML and Ph+ ALL [Saglio et al. 2009]. In addition to alternating strategies, the upfront combination of alpha IFN with imatinib is currently being investigated in France, Germany and Italy. The French SPIRIT study reported superior molecular response rates in the combination arm [Preudhomme et al. 2010]. This observation...
was confirmed in a retrospective analysis by the Italian GIMEMA [Palandri et al. 2010]. In contrast, the German CML IV study so far did not detect differences in response rates between imatinib and imatinib + alpha IFN [Hehlmann et al. 2009]. The difference might well be attributed to disparities in adherence to the combination and the formulation of IFN. It will be interesting to see whether improved molecular responses seen in the SPIRIT trial will translate into superior long-term outcome. Finally, a small study from Germany suggests that alpha IFN maintenance might sustain CCyR and MMR achieved during induction with imatinib and IFN.

Conclusions
In chronic-phase CML, treatment decisions today are mainly based on imatinib response-related prognostic factors that predict outcome and can be determined by routine monitoring. The challenge of the coming years will be to integrate nilotinib and dasatinib in the first-line treatment in a reasonable and also cost-effective way. Unfortunately current and future trials are intended to replace imatinib rather than to complement it. On the other hand, the availability of nilotinib and dasatinib simplifies matters in patients that do not achieve optimal response to imatinib. The ELN categories of suboptimal response and failure might well amalgamate since the outcome of both categories is similar, and a switch to nilotinib or dasatinib for the majority of patients failing imatinib would be more appropriate than to increase the imatinib dose. The second challenge will be the identification of patients that may safely temporarily or permanently discontinue ABL kinase inhibitor treatment. In the future, the majority of CML patients may not die from their leukaemia, and some of them may not do so even after treatment discontinuation.

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