A Decade of Nilotinib and Dasatinib: From *In Vitro* Studies to First-Line Tyrosine Kinase Inhibitors

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See related article by O'Hare et al., Cancer Res 2005;65:4500–5.

In the Wasatch Mountains of Utah, there are many remarkable rock climbing routes. Leading the first ascent of a previously unclimbed route is respected for the accomplishment it signifies and the risk taking that it requires. Similar routes, important variations, and even completely different approaches tend to follow rapidly once a long-eyed project has been "sent." The discovery and successful clinical implementation of imatinib (Gleevec) for the treatment of chronic myeloid leukemia (CML) and Philadelphia chromosome–positive acute lymphoblastic leukemia (Ph^+ ALL) can be regarded as the first ascent in the field of molecularly targeted therapeutics for leukemia. In 2005, we reported on and compared the *in vitro* resistance profiles of AMN107 and BMS-354825, which have gone on to become nilotinib (Tasigna) and dasatinib (Sprycel), respectively (1). These second-generation tyrosine kinase inhibitors (TKI) targeting BCR-ABL1 were developed to contend with clinical resistance to imatinib, and each represents an important variation of the imatinib route. With over a decade of evidence, there can be no argument that nilotinib and dasatinib have positively impacted the treatment of CML and Ph^+ ALL (2, 3).

Nilotinib and Dasatinib in Refractory Disease and in the First Line

Nilotinib bears a strong family resemblance to imatinib. Biochemically, it has all of imatinib's attributes and more than an order of magnitude greater potency. Clinical use of nilotinib can be likened to imatinib dose escalation if one could raise imatinib's dose 20-fold. Both imatinib and nilotinib feature a relatively narrow kinase selectivity profile (c-ABL, BCR-ABL1, ARG, c-KIT, PDGFR) that is traced to preferential binding of an inactive conformation with structural features that are relatively rare in the kinome. As a clinical agent, nilotinib exhibits important dissimilarities compared with imatinib, including twice-daily dosing with fasting and a different cardiotoxicity profile (4). Dasatinib engages BCR-ABL1 in a distinct manner compared with imatinib and nilotinib, binding with extremely high affinity and with a strong preference for the active conformation of the BCR-ABL1 kinase domain. Early in its clinical development, dasatinib dosed twice daily was known for a tendency to induce pleural effusions, but this issue is better managed with the current once-daily dosing prescribing instructions. One lesson derived from the dasatinib clinical experience is that BCR-ABL1 TKIs with a large number of off-targets can be surprisingly well tolerated.

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Nilotinib and dasatinib have worked their way up to consideration for first-line usage by showing clear effectiveness in relapsed CML and, to a lesser extent, Ph^+ ALL. The competition for the first-line, "best TKI for chronic phase CML" crown has obvious market share implications. Results of head-to-head trials of nilotinib versus imatinib (5) and of dasatinib versus imatinib (6) in the first line for newly diagnosed CML patients have given narrow victories to the second-generation TKIs in several categories (faster time to response milestones, higher rates of major molecular response, and lower incidence of disease progression at 5 years), but longer follow-up is needed to determine whether these numerical advantages equate with improved overall survival. Patients and clinicians must consider many factors in evaluating the various first-line TKI treatment options (7).

The BCR-ABL^{T315I} Mutation and BCR-ABL1 Compound Mutations

None of the three TKIs discussed so far has activity against the BCR-ABL1^{T315I} gatekeeper mutant. Once this issue was addressed through the introduction of the third-generation TKI, ponatinib (Iclusig; ref. 8), clinical escape through a BCR-ABL1 mutation–based mechanism has increasingly taken the form of BCR-ABL1 compound mutations, defined as $\geq$2 mutations in the same BCR-ABL1 molecule (9). Nilotinib and dasatinib are effective against certain compound mutants, but only ponatinib works against any of the T315I-inclusive compound mutants and its reach is limited (9). Ponatinib is approved for treatment-refractory CML and Ph^+ ALL. Dose-dependent issues with vascular occlusive events have been a major challenge in the clinical development of ponatinib (4), but improved dosing schedules have mitigated this risk, and ponatinib is the most effective option for patients with the BCR-ABL1^{T315I} mutation.

ABL001 is a first-in-class allosteric inhibitor of BCR-ABL1 that occupies the vestigial myristoyl pocket, inducing an autoinhibited kinase conformation. Thus, this investigational drug achieves BCR-ABL1 inhibition using a completely different approach than the approved BCR-ABL1 TKIs. Given the remote location of the myristoyl allosteric pocket relative to the catalytic site in the kinase domain of BCR-ABL1, a reasonable expectation is that ABL001 should be insulated from point mutations and compound mutations that impart resistance to currently approved TKIs. However, preclinical studies demonstrate that the high level of potency exerted on native BCR-ABL1 is not uniformly maintained against a panel of clinically observed point mutations that impart resistance to nonallosteric BCR-ABL1 TKIs (10). For example, the T315I mutant is considerably less ABL001 sensitive than native BCR-ABL1 in cell line experiments. The mechanistic reasons for these differences are under investigation. Phase I clinical trials with single-agent ABL001 in relapsed patients with chronic or accelerated phase CML previously treated with at least two different TKIs are under way. Patients who exhibit relapsed disease

associated with the presence of the BCR-ABL1^{T315I} mutation after at least one TKI are also eligible if no other effective therapy exists. The unexplored possibility of treating patients with a combination of allosteric inhibitor (e.g., ABL001) and an approved BCR-ABL1 TKI, such as nilotinib, dasatinib, or ponatinib, could prove to be an effective strategy for asserting maximum disease control.

TKIs and Treatment-Free Remission

In summary, the portfolio of BCR-ABL1 TKIs includes an impressive range of agents approved for use in the first line (imatinib, nilotinib, and dasatinib) or second line [ponatinib and bosutinib (Bosulif)] and an investigational agent resulting from clever exploitation of an allosteric mechanism that has reached the point of clinical trials. In the long run, the best drug and the one of most interest to patients is the one that cures the disease. Strictly speaking, no such drug exists for CML. Drug developers and clinical leaders in the BCR-ABL1 TKI field have been clear that TKIs do not effectively eradicate leukemic stem cells and must be considered lifelong therapies. However, some CML patients with sustained optimal responses on TKI therapy have proven willing and often anxious to participate in trials exploring the durability of treatment-free remission (TFR; ref. 11). The results have been spectacular for a subset of patients. Currently, intensive research efforts are aimed at understanding which factors are critical for successful TKI cessation and on determining whether it is possible to guide more patients into the "durable TFR" category. One possibility is that depth of response, even among optimal responders, is a predictor. By current qPCR methods, the measurable dynamic range bottoms out at a molecular response (MR) approximately five logarithmic intervals below an internationally defined baseline, MR$^{5.0}$. Among the several inclusion criteria for TFR trials, maintenance of MR$^{4.5}$ to MR$^{5.0}$ for a period of 2 years is a typical requirement (11). If a new measurement platform, such as droplet digital PCR, can extend accurate quantitation of responses below this current technical limit, it might be possible to learn whether, for example, patients reaching MR$^{6.0}$ can be assigned a higher probability of successful TFR than patients reaching MR$^{4.5}$. The reasons that some optimal responders are successful in sustaining a TFR while other, seemingly similar patients experience molecular relapse remain to be fully established.

Extending the Paradigm: TKIs for Other Malignancies

CML is a simple cancer and drugging it was straightforward. I have been assured of this fact many times, including in the halls of a famous pharmaceutical company. Of note, I have never heard a similar remark from a patient with well-controlled CML. Furthermore, there has been an upsurge in the number of first ascents and important variations of many TKI routes outside of Ph$^+$ leukemia since imatinib and follow-on drugs changed the lives of patients. These disease settings include chronic lymphocytic leukemia (Bruton tyrosine kinase TKIs), acute myeloid leukemia (FLT3 TKIs), myeloproliferative neoplasms (JAK2 TKIs), melanoma (BRAF TKIs), and dozens of others. As in the case of CML, second- and third-generation TKIs are being rapidly developed for many of these targets. Among the most intriguing and encouraging of the newer TKIs are those targeting a subset of non–small cell lung cancer (NSCLC) in patients whose disease is driven by mutated or rearranged kinases, such as EGFR, ALK, or ROS1. These patients are almost exclusively never-smokers, and molecular profiling can definitively identify them. Excellent inhibitors have been discovered for mutated EGFR-driven NSCLC, including erlotinib (Tarceva) and osimertinib (Tagrisso), which is effective against the EGFR^{T790M} gatekeeper resistance mutation, analogous to ponatinib for BCR-ABL1^{T315I}. Crizotinib (Xalkori) is approved for first-line treatment of NSCLC driven by rearranged ALK or ROS1, and a deep group of second- and third-generation TKIs that may be even more extensive than for BCR-ABL1–driven leukemia is in various stages of clinical development. The use of targeted therapies in NSCLC has dramatically improved quality of life and extended survival, but if these targets are entirely analogous to BCR-ABL1 in CML and we have great drugs, why are responses not anywhere near as durable as in CML? The "simple cancer" factor has some validity, it would seem. Patients with CML diagnosed in the chronic phase who adhere to appropriate disease management plans centered on BCR-ABL1 TKIs now have life expectancies approaching those of age-matched controls. For the subsets of NSCLC that can be treated with targeted TKIs, remissions are on the order of a few years. This represents huge progress both in length of overall survival and in the less tangible perception that the additional months or years tend to be much better ones than on chemotherapy regimens. The situation is reminiscent, in fact, of the experience of CML patients with blastic phase disease or with Ph$^+$ ALL. In these instances, blocking BCR-ABL1 kinase activity is clearly warranted for controlling disease, but it is not enough. These are acute leukemias that can access a variety of poorly understood bypass mechanisms to escape the full brunt of BCR-ABL1 TKI treatments. Anecdotally, never-smokers who are afflicted with NSCLC are legendary for not seeking medical treatment until the disease is at an advanced stage, partly because these are generally younger patients and chiefly because they do not have any reason to consider themselves at risk for the disease. In reading an excellent article on a large clinical trial comparing crizotinib in the first line with the standard-of-care chemotherapy regimen (12), I was astonished to note that 98% of newly diagnosed patients presented with metastatic disease. This may reflect a "metastatic-immediately, presents as advanced disease" trait of ALK-positive NSCLC, but it is also possible that identifying these patients earlier in their disease course could be a key for improving the effectiveness of TKIs.

Closing Thoughts

Nilotinib and dasatinib have come a long way in a decade. More generally, TKIs continue to demonstrate their worth as targeted agents for a variety of malignancies. Whether talking about CML, ALK-driven NSCLC, or other kinase-driven diseases, the maxim "more is better" applies to the number and structural diversity of approved TKIs. We are still very much in need of new routes, important variations, and completely different approaches. Climb on!

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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