PS6 Invited

Kinase inhibitors in cancer: lessons from chronic myeloid leukemia

C.L. Sawyers

Howard Hughes Medical Institute, UCLA, USA

The success of kinase inhibitor therapy in chronic myeloid leukemia (CML) has validated the long-held thesis in the cancer research community that a precise molecular understanding of cancer can directly impact cancer therapy. Now that several years have passed since the approval of imatinib/gleevec for CML treatment, we have a greater appreciation for the challenges involved in effectively deploying these agents in the clinic.

A central theme that has emerged from this work is the notion of "kinase-dependent" cancer, i.e. those cancers whose growth is driven by a specific kinase or set of kinases. By definition, such cancers should respond (shrink) when exposed to an inhibitor that effectively blocks the enzymatic activity of the responsible kinase. While CML serves as a paradigm, it is unique in that essentially all patients with clinical CML share a common molecular abnormality, activation of the ABL tyrosine kinase by chromosome translocation. It is perhaps more instructive to consider other diseases where only subsets of patients respond to these drugs. Examples include gastrointestinal stromal tumors (GIST), chronic myelomonocytic leukemia (CMML), hypereosinophilic syndrome (HES) and broncoalveolar lung cancer. In each of these cases clinical sensitivity to a kinase inhibitor is precisely correlated with a mutation in the gene encoding the target kinase that alters its biological potency, creating an oncogenic allele that drives the growth of that cancer.

If this paradigm continues to hold up using inhibitors that target additional mutant kinases implicated in other cancers (B-Raf in melanoma, Her2 in lung cancer, Flt3 in AML), we should have a very clear clinical development path for future kinase inhibitors. Specifically, human cancer kinome sequencing efforts currently underway should provide a catalogue of the kinase mutations found in human cancers. Small molecule inhibitors to these kinases can be identified, and clinical evaluation will be conducted in patients most likely to respond, i.e. those whose tumors contain mutations in the target kinase. Obviously, this will require the use of molecular diagnostics, presumably through PCR-based mutation detection, for informed patient selection.

Importance of downstream signaling pathway status in response to kinase inhibitors: While I fully expect this scenario to be played out in some fashion over the coming years, it is important to recognize certain complexities that may influence tumor response to kinase inhibitors, even in tumors with a mutation in the target kinase. One result from analysis of glioblastoma patients treated with EGFR inhibitors by the UCLA Neurooncology Group (Ingo Mellinghoff, Tim Cloughesy, Paul Mischel) serves and an example. The response rate to the drugs (Iressa or Tarceva) in relapsed glioma is ~15% in phase II studies. Of 49 such patients treated at UCLA, 7 had dramatic, radiographically documented objective responses. Molecular profiling of the EGFR in all these patients revealed that 6 of these 7 patients had mutations in the extracellular domain (called EGFR variant iii). (Of note, no patients had mutations in the EGFR kinase domain, thereby distinguishing this mechanism from that found in lung cancer.) However, 6 of 19 patients who rapidly progressed through EGFR inhibitor therapy also had EGFR viii mutations. The variable that distinguishes these two groups is presence or absence of the PTEN tumor suppressor gene, which was expressed in wild-type form in all of the responding patients but in only 2 of the progressive disease patients with EGFR viii mutations. These data were confirmed on an independent sample set from UCSF (Mike Prados and colleagues), and suggest that molecular assessment of both EGFR viii and PTEN is required to define sensitivity to EGFR inhibitors in glioblastoma patients. The broader implication is that future application of patient-tailored kinase inhibitor therapy is likely to require evaluation of a suite of molecular variables for optimal predictive power.

Acquired resistance to kinase inhibitors: mechanisms and potential solutions: A second issue complicating the success of kinase inhibitor therapy is acquired resistance, defined as disease relapse on continuous therapy after an initial response. First recognized as a significant problem initially in advanced stage CML patients, acquired resistance also occurs in chronic phase CML, GIST, HES and bronchoalveolar

lung cancer. The best understanding of resistance mechanisms comes from CML, where >85 percent of relapsed patients have mutations in the ABL kinase domain that alter drug sensitivity. Of note, similar mechanisms have been reported for GIST and HES but explored in less detail.

In the case of ABL, 38 different mutations have been reported to date, but 3–4 mutations account for 60–70 percent of all cases. Insights into how these mutations cause resistance became apparent through the solution of the co-crystal structure of imatinib bound to ABL by John Kuriyan's group at UC-Berkeley. Curiously, only a small number of the mutations occur at contact residues where substitution of the new amino acid leads to loss of a hydrogen bond donor or steric hindrance due to bulkier side chains. Rather, the majority of mutations are at residues that appear, based on structural modeling studies, to later the conformational flexibility of ABL such that it can no longer achieve the closed, inactive conformation required for optimal imatinib binding.

These structural studies raise the possibility that a second ABL kinase inhibitor which binds in a less conformation-dependent fashion may have activity against certain imatinib resistant mutants. We tested this hypothesis using dual SRC/ABL kinase inhibitors which can bind ABL in the active or inactive conformation. One such compound BMS-354825 blocked the growth of murine hematopoietic cells transformed by all but one imatinib resistant BCR-ABL mutant in culture and showed anti-leukemic activity in mouse models (Shah et al., Science, 2004). This compound also induced hematologic and cytogenetic remissions in a high fraction of imatinib-resistant CML patients in a phase I trial with minimal side effects (Sawyers et al., ASH 2004; Talpaz et al., ASH 2004). Phase II studies are currently underway.

We have also examined potential mechanisms of resistance to BMS-345825 by saturation mutagenesis. Unlike imatinib we find that resistance occurs almost exclusively through mutations at drug contact residues, presumably due to less conformation-stringent binding requirements. In addition some mutations were isolated that confer resistance to BMS-354825 but not to imatinib. These data provide evidence in favor of either sequential or combination therapy with these two compounds for CML.

References

[1] Burgess MR, Skaggs BJ, Shah NP, Lee FY, Sawyers CL. Comparative analysis of two clinically active BCR-ABL kinase inhibitors reveals the role of conformation-specific binding in resistance. *Proc Natl Acad Sci USA* 2005, **102**, 3395–3400.

[2] Mellinghoff IK, Wang MY, Vivanco I, *et al.* Molecular determinants of EGFR kinase inhibitor response in glioblastoma. *N Engl J Med*, in press.

[3] Sawyers CL. Opportunities and challenges in the development of kinase inhibitor therapy for cancer. *Genes Dev* 2003, **17**, 2998–3010.

[4] Shah NP, Nicoll JM, Nagar B, et al. Multiple Bcr-Abl kinase domain mutations confer resistance to the tyrosine kinase inhibitor imatinib in chronic phase and blast crisis chronic myeloid leukemia. *Cancer Cell* 2002, **2**, 117–125.

[5] Shah NP, Tran C, Lee FY, Chen P, Norris D, Sawyers CL. Overriding imatinib resistance with a novel ABL kinase inhibitor. *Science* 2004, **305**, 399–401.