Consisting of more than 500 genes, the family of protein kinases are central to nearly every major signal transduction pathway in normal cells, and their dysregulation seems to be a major driver of neoplastic transformation and progression in many human cancers (1). This central role in aberrant cancer signaling, coupled with advances in pharmacologic, biophysical, and medicinal chemistry allowing for the development of selective inhibitors of individual or groups of kinases, has spurred an explosion in the preclinical and clinical testing of kinase inhibitors as cancer therapeutics (2). For such kinase inhibitors to be successful as cancer therapeutics, a given cancer must show a “dependence” or “addiction” to the catalytic activity of the targeted kinase (3). Often, such dependence has been associated with mutations that constitutively activate the kinase signaling pathway (4). Consequently, selective inhibitors of specific kinases that become activated through mutation, amplification, or translocation in specific cancers have shown efficacy for treatment of those cancers. The first major example of kinase inhibitors as targeted therapy for cancer was the use of the ABL tyrosine kinase inhibitor imatinib, and later dasatinib and nilotinib, for treatment of BCR-ABL fusion–positive chronic myeloid leukemia (5). Other successful examples include trastuzumab and lapatinib, targeting the ERBB2 kinase, in ERBB2-amplified breast cancers (6); gefitinib and erlotinib, inhibiting EGFR, in EGFR-mutant lung cancers (7); and the ALK inhibitor crizotinib in ALK fusion–positive lung cancers (8). Although each of these examples involves targeting a kinase that is directly activated by mutation, it is certainly possible that some cancers may have dependence on specific kinases even in the absence of genetic alterations to the kinase gene itself. However, it has been difficult to identify such kinase dependencies in the absence of obvious mutations.

To address this issue, Kothari and colleagues (9) explored the possibility that extreme steady-state mRNA expression of individual kinase genes in specific cancers may indicate a core dependence on those kinases. Using a compendium of RNA-seq genome-wide transcriptome data assembled by their group, they established the reference expression distribution of each of 468 kinase genes in 482 samples from 25 different tissue/cancer types. For each sample, they then identified kinases that showed outlier expression compared with the overall distribution across all samples, hypothesizing that each sample may exhibit a particular dependence on kinases with extreme overexpression. To test this hypothesis in proof-of-principle studies, they examined the sample-specific dependence on such outlier kinases in breast and pancreatic cancer cell lines by inhibiting those kinases using RNA interference (RNAi) and pharmacologic inhibitors.

In the breast cancer samples, as an initial validation that the approach could identify kinase dependencies, they found that all breast cancer cell lines with a known ERBB2 amplification showed outlier expression of the ERBB2 gene. Interestingly, a significant fraction of these also showed outlier expression of FGFR4. Of these, although some did not exhibit any growth inhibition in response to ERBB2 inhibition, targeting FGFR4 in these cell lines did result in growth inhibition, and combined targeting of both FGFR4 and ERBB2 led to additive growth inhibition. In addition, trastuzumab-resistant derivatives of MDA-MB-453 breast cancer cells, which showed outlier FGFR4 and ERBB2 gene expression and were sensitive to inhibition of each kinase alone and in combination, continued to show sensitivity to FGFR4 inhibition despite being rendered resistant to ERBB2 inhibition. These data suggest the possibility of combinatorial targeting of multiple kinases with outlier expression in a sample-specific manner to prevent emergence of resistance to any one kinase inhibitor.

The authors next turned their attention to identifying outlier kinase dependencies as novel targets in pancreatic cancers, which are notoriously refractory to current therapies (10). These studies revealed a strong growth inhibition...
by RNAi-mediated depletion of kinase enzymes in outlier positive, but not negative, cell lines, establishing proof-of-principle for both the potential efficacy and sample specificity of the approach. This efficacy was also confirmed in vivo in tumor xenografts of the pancreatic cell lines BxPC-3 and PANC-1, which displayed outlier expression of the c-Met kinase, using the c-Met inhibitor XL184. Such outlier kinase profiles were also able to identify targets for synthetic lethality. For example, only pancreatic cancer samples with KRAS dependence showed outlier kinase expression of polo-like kinases PLK1 and PLK2. Inhibition of PLK1 has previously been shown to exhibit synthetic lethality with mutant KRAS. Interestingly, Kothari and colleagues (9) show that KRAS-dependent pancreatic cancer cells were sensitive to the pan-PLK inhibitor B16727 only if they had outlier expression of PLK1 or PLK2.

These studies have important implications for cancer precision medicine. First, extreme expression of kinase-encoding genes in a given individual’s cancer, identified as outliers in RNA-seq transcriptome data, can potentially be used to match targeted kinase inhibitors to those patients that might benefit most from them. In addition, such kinase outlier expression profiles seem to hold promise for identification of effective combinations of targeted therapies, as well as for targeting based on synthetic lethality. Implementation of outlier kinase expression profiling in precision medicine paradigms may be accomplished in many different ways, with one type of implementation shown in Fig. 1. Tumor material sampled by biopsy or surgical resection can be used to generate tumor-specific transcriptome data using RNA-seq. To identify outlier kinases, gene expression measures of kinase genes can be extracted and compared with a large compendium of RNA-seq data from reference tissues. Drugs targeting such outlier kinases can then be used alone or in combination with current therapies for individualized treatment of pancreatic cancers. RPKM, reads per thousand base pairs per million reads, refers to normalized absolute gene expression measures for each sample.
reference normal and cancer samples. This process would then allow selection of inhibitors specific to one or more outlier kinases to be used alone or in combination with other therapies for individualized treatment of the patient, resulting, it is hoped, in significant tumor regression and improvement in morbidity and mortality.

Although Kothari and colleagues (9) establish important proof-of-concept for such outlier kinase-driven precision medicine paradigms, significant further testing and study are required for optimal development and deployment of such strategies for cancer clinical care. This study did establish the presence of kinase outlier expression in human cancer tissues as well as cell lines, but all of the functional credentialing of targeting outlier kinase dependencies was carried out in cell lines and in cell line–derived xenografts. Because efficacy in such preclinical cell line models often does not translate to the clinical setting, it will be important to establish the efficacy and safety of inhibiting outlier kinase expression in clinical trials. This point brings us to the question how such clinical trials would be designed, given the sample-specific nature of these outlier kinase expression profiles—a challenge that is common to clinical testing of many precision medicine strategies. Another logistical concern pertains to the assessment of outlier expression in heterogeneous tumor samples, particularly stroma-dense paucicellular neoplasms like pancreatic cancer. Clinical evaluation of inhibitors of outlier kinases that are uncommonly present in various tumors, or those for which inhibitors are currently not available, will present major challenges as well. It will also be important to identify mechanisms by which resistance to inhibition of outlier kinases may emerge. For inhibition of mutated kinases, this often involves development of second-site mutations. Such mechanisms may be fundamentally different for inhibition of kinases that are simply upregulated, but not subject to mutations, and this will be important to understand to avoid the pitfalls that have limited efficacy of many other types of targeted therapies owing to emergence of resistance. For future studies, it will be interesting to see whether outlier expression can signal dependencies on other classes of enzymes and targets for which inhibitors are available, including proteases, phosphatases, nuclear hormone receptors, and so on. If so, the principles identified here can be applied to these other gene families as well.

The explosion of technologies for measurement of gene expression and sequence variation is fueling a growing revolution in precision medicine. Despite the aforementioned challenges, the concept of targeting kinase dependencies showing extreme expression in individual cancers holds great promise for expanding the use of targeted therapies in precision medicine.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

References

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