IN THE SPOTLIGHT

Can the Help Match the Hype? KRAS<sup>G12C</sup>-Specific Inhibitors and Beyond

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Summary: Hallin and colleagues demonstrate the preclinical activity of the KRAS<sup>G12C</sup>-specific inhibitor MRTX849 in a series of in vitro and in vivo studies with supporting pilot clinical efficacy. Variable responsiveness despite effective KRAS<sup>G12C</sup> inhibition highlights both the promise and potential need for combinatorial strategies to optimally target KRAS<sup>G12C</sup>-driven cancers.

See related article by Hallin et al., p. 54 (5).

As with draft picks in professional sports, the majority of hyped small molecules entering phase I clinical trials never achieve more than a few interesting bars on a waterfall plot. The reasons are multifactorial, often owing to incomplete target inhibition, tumor heterogeneity, complex adaptive resistance not adequately modeled in preclinical models, and/or toxicity profiles that limit optimal dosing or combination strategies. Such has been the case for earlier attempts at targeting KRAS-driven cancers with tankyrase inhibitors and MEK-directed agents alone or in combination (1). In fact, prior failures have earned KRAS the dubious moniker of “undruggable.” However, we may now be on the verge of a paradigm shift in KRAS-targeted therapies.

The tumorigenic role of activating mutations in genes encoding the RAS subfamily of small GTP-binding proteins has been known for more than two decades and expertly reviewed elsewhere (1). In 2013, groundbreaking work by Shokat and colleagues demonstrated for the first time that direct inhibition of mutant KRAS could be achieved by small molecules that covalently bind to the mutated cysteine at codon 12 in KRAS<sup>G12C</sup>-mutant cancers (2). These KRAS<sup>G12C</sup>-selective inhibitors bind in an allosteric switch II pocket present in the GDP-bound form of KRAS and lock the protein in its inactive state. This discovery has fueled a number of drug-development efforts to develop clinically active KRAS<sup>G12C</sup> inhibitors, and excitement has continued to build over the past few months with preliminary clinical results of two KRAS<sup>G12C</sup>-specific inhibitors demonstrating safety and clinical activity, primarily in non–small cell lung cancer (NSCLC) where the KRAS<sup>G12C</sup> mutation frequency approaches 15% (Fig. 1A; refs. 3, 4).

In this issue of Cancer Discovery, Hallin and colleagues report on one of these molecules, MRTX849, and provide substantial preclinical characterization and a first look at clinical efficacy (5). Using a panel of KRAS<sup>G12C</sup>-mutant cell lines, the authors demonstrate a wide range of IC<sub>50</sub> values (10 to 973 nmol/L) in in vitro cell viability studies. This early observation highlighted the potential of MRTX849 for targeting KRAS<sup>G12C</sup>, but also exposed a potential therapeutic limitation. Despite effective inhibition of KRAS<sup>G12C</sup> itself, downstream analyses revealed variable suppression of ERK phosphorylation that paralleled the IC<sub>50</sub> levels, perhaps reflecting heterogeneity in RAS addiction. Extending their work to in vivo cell line xenograft and patient-derived xenograft models, the authors demonstrated that MRTX849 is broadly active, achieving tumor regression in 17 of 26 models (65%). Notably, and perhaps sobering, in vivo efficacy did not correlate with in vitro (neither 2-D nor 3-D) sensitivity. Thus, the authors sought to examine correlates of MRTX849 sensitivity in vivo. Neither baseline RNA sequencing, reverse phase protein array analysis, nor co-occurring alterations were sufficient to predict activity or response variability, reminiscent of prior studies on KRAS-mutant models that failed to correlate responses to pharmacologic or genetic approaches targeting KRAS or downstream pathways with co-occurring mutations or other biomarkers. To further evaluate mechanisms of therapy response and resistance and explore potential synergistic vulnerabilities for combination therapy approaches, the authors performed both targeted CRISPR/Cas9 (targeting 400 genes) and pharmacologic screening on several KRAS<sup>G12C</sup> models. The hits of both screens converged on regulators of the RAS pathway [(e.g., SHP2/PTPN11, receptor tyrosine kinases (RTK)], mTOR pathway, and cell cycle, consistent with more limited prior studies using the ARS-1620 KRAS<sup>G12C</sup> inhibitor tool compound (6, 7). Combination activity was explored with a series of in vitro experiments combining the pan-HER inhibitor afatinib, the CDK4/6 agent palbociclib, the ATP-competitive mTOR inhibitor vistusertib, and the SHP2 inhibitor RMC-4550 with MRTX849. The totality of the data confirmed combination benefit in multiple models for each of these vulnerabilities identified in the CRISPR and drug screens, with deeper tumor regressions compared with MRTX849 alone.

An early simultaneous publication by Canon and colleagues has reported the discovery and preclinical characterization of AMG 510, a structurally similar KRAS<sup>G12C</sup> inhibitor that was the first to enter the clinic (8). This study reported similar, albeit less extensive, in vitro and in vivo evidence that the
The efficacy of KRAS<sup>G12C</sup> inhibitors can be improved by combining with inhibitors of RTKs, SHP2, MEK, or AKT/PI3K. Interestingly, in contrast to this study, Canon and colleagues examined the effect of KRAS<sup>G12C</sup> inhibition on the tumor immune microenvironment in a syngeneic mouse model. They demonstrate that KRAS<sup>G12C</sup> inhibitor treatment led to increased CD8<sup>+</sup> T-cell infiltration, upregulation of proinflammatory cytokines in the tumor microenvironment, and synergy with anti–PD-1 agents. Importantly, this provides rationale for yet another KRAS<sup>G12C</sup> inhibitor combination, and clinical investigation of AMG 510 in combination with an anti–PD-1/PD-L1 is planned (NCT03600883, clinicaltrials.gov). It is worth noting that early excitement over increased T-cell infiltration in response to a somewhat analogous combination of MEK and anti–PD-L1 inhibitors in KRAS-mutant microsatellite-stable colorectal cancers did not ultimately translate into improved patient outcomes, although the tumor-selective activity of KRAS<sup>G12C</sup> inhibitors may avoid potential counterproductive actions on T cells (9).

The two presented patient cases provide examples of MRTX849 clinical activity in a patient with KRAS<sup>G12C</sup> colorectal cancer and NSCLC. Responses in additional patients were recently presented, with partial responses seen in 3 of 5 patients with NSCLC, 1 of 2 patients with colorectal cancer, and 0 of 2 patients with appendiceal cancer at the highest dose level (3). Clinical results have also been reported for a larger cohort of patients treated with AMG 510, with an overall response rate of 48% (10) in patients with NSCLC, and 54% (7/13) at the highest dose (3). Similar to MRTX849, the activity of AMG 510 in patients with colorectal cancer and appendiceal cancer appears to be much lower. Although single-agent activity is seen both preclinically and clinically, the optimal pathway forward may indeed be combinatorial approaches (Fig. 1B).

**Figure 1.** Pan-cancer frequency of KRAS<sup>G12C</sup> mutations and reported preliminary clinical activity of KRAS<sup>G12C</sup> inhibitors. A, Cancer types with KRAS<sup>G12C</sup> mutation frequencies greater or equal to 1% are shown. Data are derived from 43,910 patients (46,237 samples) in nonoverlapping studies from The Cancer Genome Atlas (www.cbioportal.org, accessed 10/26/19). CNA, copy-number alteration. B, Comparative clinical data between MRTX849 and AMG 510 reported for all patients with all dose levels reported. ORR, overall response rate; RP2D, recommended phase II dose.
increase. In patients achieving durable benefit, what will be the molecular mechanisms underlying clinical progression? At this time, this question remains understudied and was not specifically addressed by the authors. On the other hand, the insights provided by these studies into the mechanisms of intrinsic and adaptive resistance may provide clues. Will we observe alterations or activation of pathways shown to mediate intrinsic resistance such as RTK signaling, or will other mechanisms emerge? Will additional RAS mutations, which are commonly observed in colorectal cancers treated with EGFR antibodies but rarely seen in NSCLC treated with EGFR or ALK inhibitors, develop? The use of blood-based monitoring strategies including cell-free DNA may be well suited to clinically study the development of resistance to detect alterations that may reestablish ERK signaling (10).

Although these initial preclinical and clinical results are indeed exciting, a number of important questions remain. First, it is not yet clear whether KRAS<sup>G12C</sup> inhibitors will achieve maximal success as monotherapy, or whether drug combination will be necessary for durable responses. In the study presented by Hallin and colleagues, no clear predictors of response to MRTX849 were observed, including the presence of commonly occurring mutations that could be readily determined using clinical genotyping panels. Similarly, there does not appear to be a reliable predictor of RAS dependence. Although adaptive signaling appears to play a significant role in experimental models, the importance of these mechanisms in patients remains incompletely understood. Future reporting with longer-term patient follow-up will begin to clarify this issue. Second, assuming that drug combinations will lead to deeper and more durable responses, the optimal combinations have yet to be defined. The comprehensive studies presented here provide a road map for drug combinations that are likely to be tested in future combination trials. Although the results presented here suggest the potential for genomic predictors of specific combinations (e.g., CDKN2A deletion and sensitivity to CDK4/6 inhibition), most combinations tested lacked obvious biomarkers. Furthermore, many of the models tested responded to multiple MRTX849 combinations, suggesting the potential for many possible therapeutic approaches but offering little clarity on how different combinations should be prioritized. Third, it remains to be determined how the durability of response to KRAS<sup>G12C</sup>-targeted monotherapy (or combinations) will hold up against the current standard of chemotherapy + immune-checkpoint inhibition.

Overall, the authors should be commended for presenting a substantial amount of data regarding MRTX849 in vitro and in vivo and for their comprehensive approach to understanding determinants of drug response and the potential for combination therapy. This impressive effort sets a benchmark for the development and characterization of other forthcoming KRAS inhibitors. We eagerly await further clinical data and hope we are truly entering a new era for KRAS-mutant cancers.

**Disclosure of Potential Conflicts of Interest**

S.J. Klempner has received speakers bureau honoraria from Foundation Medicine, has ownership interest (including patents) in Turning Point Therapeutics, and has consultant/advisory board relationships with Bristol-Myers Squibb, Eli Lilly, Astellas, and Boston Biomedical. A.N. Hata reports receiving commercial research grants from Amgen, Relay Therapeutics, Pfizer, Roche/Gentech, and Novartis. No other potential conflicts of interest were disclosed.

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**REFERENCES**

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