Abstract # 34971

The next-generation farnesyltransferase inhibitor KO-2806 constrains compensatory signaling reactivation to deepen responses to KRAS^{G12D} inhibition

Alison Smith, Hetika Vora Patel, Stacia Chan, Francis Burrows, Shivani Malik
Kura Oncology, Inc., San Diego, CA

BACKGROUND

- The landmark approval of KRAS^{G12C} inhibitors has fueled the design and development of small molecules targeting other oncogenic KRAS mutants, including the most prevalent, KRAS^{G12D}.
- MRTX1133 is the first KRAS^{G12D}-selective inhibitor to reach the clinic.
- Preclinical MRTX1133 data and early clinical results from KRAS^{G12C} inhibitor trials suggest that responses to KRAS^{G12D} inhibition will vary, in part due to incomplete and/or transient inhibition of MAPK and mTOR signaling.
- We have previously demonstrated that farnesyltransferase inhibitors (FTIs) can prevent adaptive resistance to PI3K inhibition by blunting compensatory reactivation of MAPK and mTOR.
- KO-2806 is a next-generation FTI with increased potency and improved pharmacokinetic properties, poised to enter a first-in-human clinical trial.
- We hypothesized that KO-2806 would enhance the antitumor activity of MRTX1133 by blocking feedback reactivation of mitogenic and survival signaling; we evaluate the therapeutic potential of this combination here.

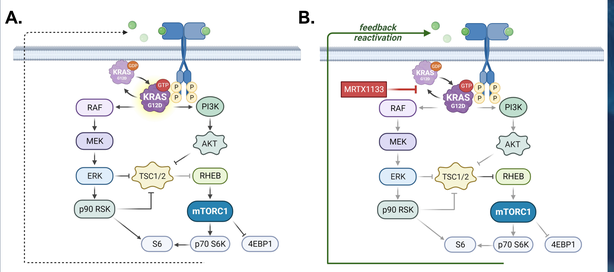


Figure 1. KRAS^{G12D} inhibition relieves inhibitory feedback mechanisms, leading to reactivation of MAPK and mTOR signaling. A. At steady state, KRAS^{G12D} signals through RAF and PI3K, promoting tumor cell growth and survival whilst suppressing upstream pathway activators, limiting the depth and duration of the signal. **B.** KRAS^{G12D} inhibition potently suppresses MAPK and mTOR signaling, relieving feedback inhibitory mechanisms, leading to induction of upstream pathway components and reactivation of signaling through PI3K or WT RAS isoforms.

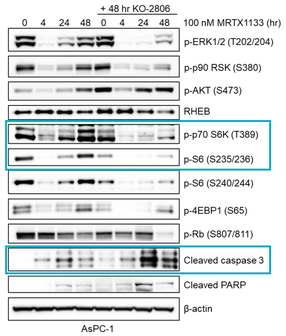
OBJECTIVES

- Evaluate the *in vivo* tolerability and efficacy of KO-2806 and MRTX1133 in KRAS^{G12D}-mutant xenograft models
- Assess impact of combination on MAPK and mTOR signaling *in vitro* and *in vivo* to delineate mechanism of action

RESULTS

KO-2806 blocks reactivation of mTOR following KRAS^{G12D} inhibition and induces apoptosis *in vitro*

Figure 2. Combined KO-2806 and MRTX1133 durably inhibits MAPK/mTOR signaling and induces apoptosis in the PDAC cell line AsPC-1. Immunoblots of the indicated MAPK/PI3K pathway components and phenotypic markers in the KRAS^{G12D}-mutant PDAC cell line AsPC-1 treated with 100 nM MRTX1133 for 0, 4, 24, or 48 hours in the presence or absence of KO-2806. Shift in RHEB mobility is indicative of unfarnesylated state.



Combined KO-2806 and MRTX1133 leads to deeper and more durable antitumor responses

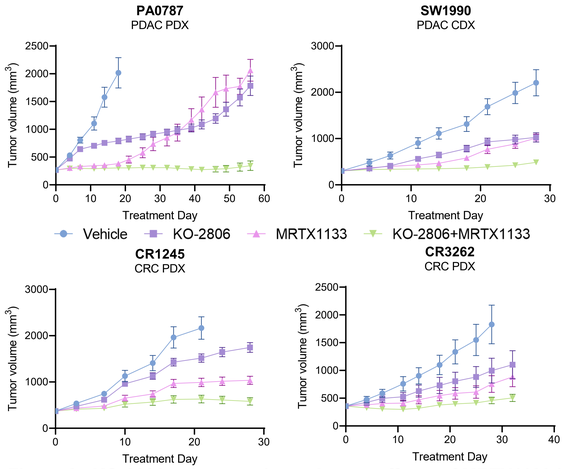


Figure 3. KO-2806 enhances the antitumor effects of MRTX1133 in KRAS^{G12D}-mutant pancreatic and colorectal xenograft models. Growth of PDAC and CRC PDX/CDX models treated with vehicle, KO-2806, MRTX1133 (30 mg/kg IP, BID in PDX models, 10 mg/kg IP, BID in SW1990), or the combination. Combination was well tolerated with minimal BWL observed. Data are means +/- SEM, n=8 animals/group.

KO-2806 and cetuximab have comparable antitumor activity when combined with MRTX1133 in CRC model

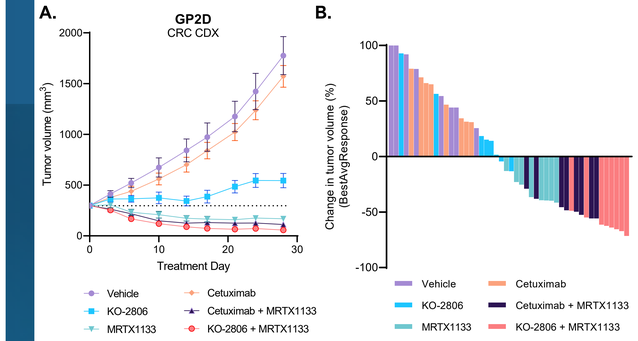


Figure 4. Both KO-2806 and cetuximab deepen response of colorectal CDX model to MRTX1133. A. Growth of GP2D CDX tumors treated with vehicle, KO-2806, MRTX1133 (20 mg/kg IP, BID), cetuximab (0.25 mg/dose IP, Q3D), or combinations for 28 days. **B.** Best average response of GP2D tumors from **A.**, per mRECIST.

Combined KO-2806 and MRTX1133 treatment more potently inhibits mTOR *in vivo* compared to MRTX1133 monotherapy

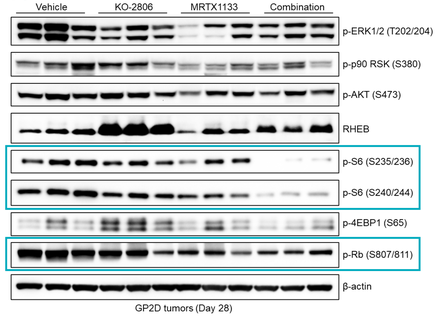


Figure 5. The KO-2806 MRTX1133 combination potently inhibits mTOR and inhibits cell cycling in G12D-mutant CRC tumors. GP2D CDX tumors were treated with vehicle, KO-2806, MRTX1133 (20 mg/kg IP, BID), or the combination for 28 days. Tumors were harvested and snap frozen for immunoblot analysis 2 hours post-treatment on day 28. Combination treatment resulted in stronger inhibition of mTOR (as indicated by lower levels of S6 and 4EBP1 phosphorylation) and cell cycle arrest (decreased Rb phosphorylation) compared with MRTX1133 monotherapy-treated tumors.

CONCLUSIONS

- The combination of KO-2806 and MRTX1133 has superior antitumor activity compared to MRTX1133 monotherapy in KRAS^{G12D}-mutant CRC and PDAC xenograft models while remaining well-tolerated.
- KO-2806 performs as well or better than cetuximab when paired with MRTX1133 in a CRC model, making it a viable combination partner.
- Mechanistically, KO-2806 blocks the compensatory reactivation of mTOR signaling that follows KRAS^{G12D} inhibition, resulting in tumor cell cycle arrest and apoptosis.

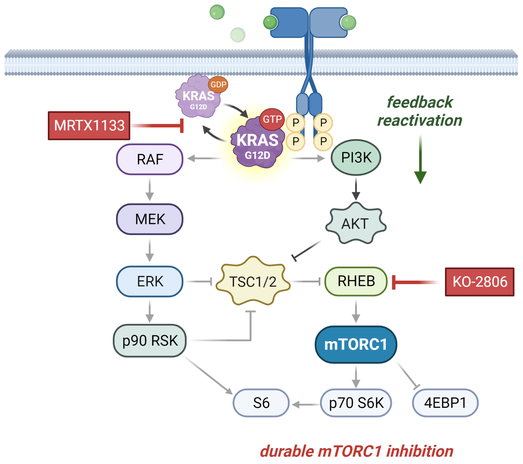


Figure 6. Mechanistic rationale for combining a farnesyltransferase inhibitor with an inhibitor of KRAS^{G12D}. KRAS inhibition with a small molecule inhibitor such as MRTX1133 transiently blocks MAPK-mTOR signaling, relieving negative feedback on upstream pathway components such as receptor tyrosine kinases. Induction of these pathway activators leads to reactivation of mTOR signaling through PI3K or WT RAS isoforms. By inhibiting the obligately-farnesylated mTOR activator RHEB, farnesyltransferase inhibitors such as KO-2806 impede reactivation of mTOR, resulting in durable mTOR inhibition, cell cycle arrest, and cell death.