Abstract
LB-098

The Anti-Tumor Activity of the KRAS^{G12C} Inhibitor MRTX849 is Augmented by Cetuximab in CRC Tumor Models

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Background

- MRTX849 is a potent, selective, and covalent KRAS^{G12C} inhibitor presently under evaluation in clinical trials.
- MRTX849 only inhibits inactive GDP-bound KRAS^{G12C}, which is known to be dependent on nucleotide cycling to regulate the activation state of KRAS.
- Comprehensive work in tumor xenograft models shows activation of pathways upstream or downstream of KRAS through additional mutations or relief of feedback inhibition can limit the single agent activity of MRTX849 in less responsive tumors.
- Activation of RTKs through relief of feedback activity is one key mechanism implicated in limited responses to KRAS^{G12C} inhibitors and the EGFR inhibitor, cetuximab, and SHP2 inhibitor, RMC-4550, both impact RTK signaling. Moreover, EGFR functions as a dominant RTK in colorectal cancer.
- The combination of MRTX849 and cetuximab or RMC-4550 in CRC models demonstrated increased KRAS^{G12C} modification and RAS/MAPK pathway inhibition.
- These data suggest that co-targeting KRAS^{G12C} and upstream targets like EGFR or SHP2 may be a critical strategy to overcome mechanisms of intrinsic or adaptive resistance to KRAS^{G12C} targeted inhibition in CRC.

Fig. 1 MRTX849 key small molecule interactions with KRAS G12C protein

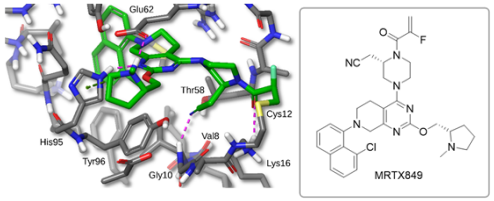
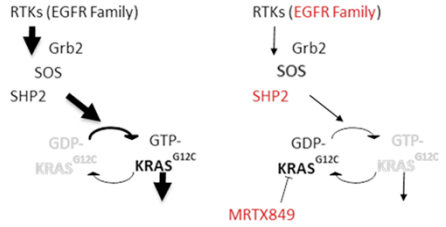


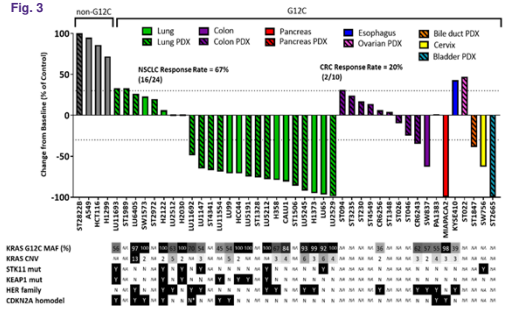
Fig. 2 Co-targeting KRAS^{G12C} and RTK signaling can more fully inhibit downstream signaling



Results

MRTX849 Demonstrates Broad Anti-tumor Activity Across KRAS^{G12C} Mutant Tumor Models with Variable Activity Observed in CRC

Fig. 3



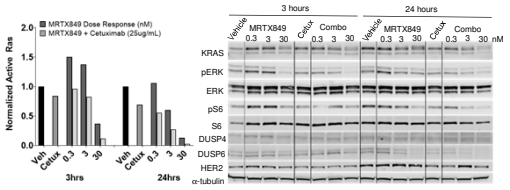
Mice bearing CDX or PDX models were treated with MRTX849 @ 100mg/kg PO, QD in all models shown.
% change from baseline control was calculated on ~ day 22 post initiation of dosing.

Cetuximab or RMC-4550 Synergizes with MRTX849 *In Vitro* Leading to Increased KRAS Modification and Pathway Inhibition

Fig. 4

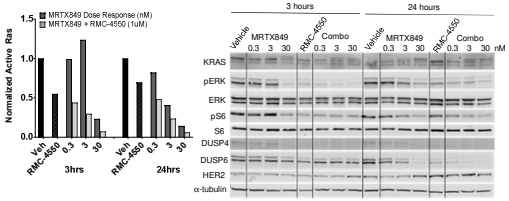
Model	KRAS MAF (%)	Co-Mutations						MRTX849 IC$_{50}$ (nM)			2D MCA Scores				
		TP53	PIK3CA	APC	SMAD 4	BRAF	HER Family	pERK	2D 3 Day Viability	3D 7 Day Viability	Cetuximab SA	Cetux combo	Afatinib SA	Afatinib combo	Afatinib SA	SHP2i combo
SW837	57	R248W		R1450* R213*				17.1	71.8	77.5	no activity	66	60% inhib @ 10uM	27	50% inhib @ 10uM	22
SW1463	100	R248Q	PIK3R4 P578L	V450fs	R361Q			38	15.3	11.2	no activity	88	266	88	4748	88

Unified combination score (MCA) based on multiple algorithms from viability data using a CTG assay; All combinations were calculated as synergistic.



Western blot analysis of SW837 cell line treated with time course of MRTX849 dose response, cetuximab @ 25ug/mL, and a combination of both compounds. Quantitation of band density performed for Active Ras.

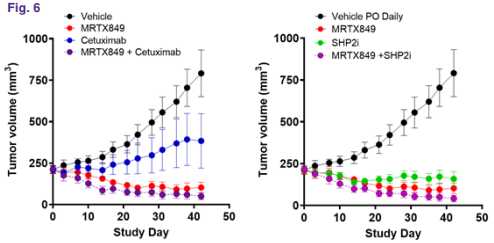
Fig. 5



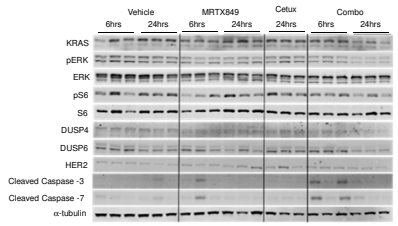
Western blot analysis of SW837 cell line treated with time course of MRTX849 dose response, RMC-4550 @ 1uM, and a combination of both compounds. Quantitation of band density performed for Active Ras.

MRTX849 and EGFR or SHP2 Inhibitor Combinations Lead to More Robust Anti-tumor Efficacy *In Vivo*

Fig. 6

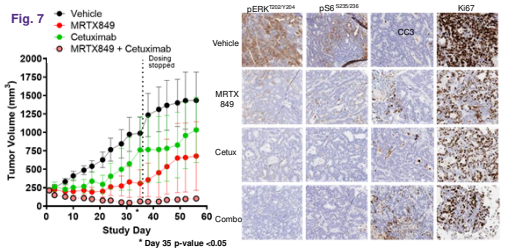


Mice bearing SW837 cell line derived xenografts were treated with Vehicle PO QD, MRTX849 at 30mg/kg PO QD, Cetuximab at 0.25mg IP Q3D, SHP2i (RMC-4550) at 30mg/kg PO QD, or a combination of MRTX849 and the combination partner at the same doses for the duration of the study. Data shown as average tumor volume +/- SEM, n=5/group.



SW837 tumors were harvested at 6 and 24 hours after a single dosing of MRTX849 @ 30mg/kg, Cetuximab @ 0.25mg, or the combination. Tumors were lysed and analyzed by western blot for proteins of interest. n=3/time point.

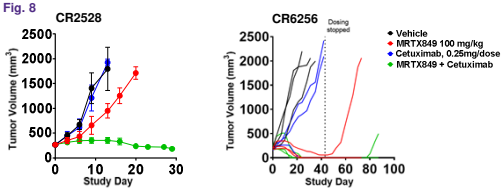
Fig. 7



Mice bearing SW1463 cell line derived xenografts were treated with Vehicle PO QD, MRTX849 at 30mg/kg PO QD, Cetuximab at 0.25mg IP Q3D, or a combination of the two at the same doses for the duration of the study. Data shown as average tumor volume +/- SEM, n=5/group. Tumor-bearing mice were treated as described, and tumors were harvested 24hrs after a single dose. Tumor were formalin-fixed, paraffin embedded, sectioned and stained with pERK, pS6, Ki67 and Cleaved Caspase-3 via immunohistochemistry methods. 10X representative images shown.

MRTX849 with Cetuximab Result in Deep, Durable Regressions in PDX Models

Fig. 8



Mice bearing CR2528 and CR6256 patient derived xenografts were treated with Vehicle PO QD, MRTX849 at 100mg/kg PO QD, Cetuximab at 0.25mg IP Q3D, or the combination for the duration of the study. Data shown as average tumor volume +/- SEM, or individual tumor volumes, n=3/group.

Conclusions

- MRTX849 is a potent, selective, and covalent KRAS^{G12C} inhibitor presently under evaluation in clinical trials in NSCLC, CRC, and other tumor types with confirmed KRAS G12C mutations.
- Activation of RTKs upstream of KRAS may be a key mechanism that limits the single agent activity of KRAS^{G12C} inhibition, especially in CRC tumor types.
- MRTX849 treatment in combination with cetuximab or RMC-4550 can more fully inhibit mutant KRAS signaling, which leads to more robust pERK modulation *in vitro*, and deeper, more durable regressions in CDX and PDX models *in vivo*.
- Co-targeting KRAS^{G12C} and EGFR or SHP2 are likely critical strategies to overcome resistance to single agent KRAS^{G12C} inhibition in CRC.

Acknowledgements

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- Crown Biosciences for their support with in vivo PDX models
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