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Discovery of Potent, Selective and Orally Bioavailable SOS1 Inhibitors for KRAS-Driven Cancers

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Background

KRAS is the most frequently dysregulated oncogene with high prevalence in non-small cell lung cancer (NSCLC), colorectal cancer (CRC), and pancreatic cancer (PAC). Currently FDA-approved sotorasib and adagrasib provide breakthrough therapies for cancer patients with $KRAS^{G12C}$ mutation, however there is still high unmet medical need for new agents that target a broader KRAS-driven tumors. An emerging and promising opportunity is to develop a pan KRAS inhibitor by suppressing the upstream guanine nucleotide exchange factor (GEF) protein son of sevenless 1 (SOS1). SOS1 is a key activator of KRAS and facilitates the conversion of GDP-bound KRAS (off) state to GTP-bound KRAS (on) state. Binding to its catalytic domain, small molecule SOS1 inhibitor prevents KRAS activation and suppresses cancer cell proliferation.

Methods

Regor's unique Computer Accelerated Rational Discovery (rCARD™) technology platform was applied to identify potent and selective SOS1 inhibitors with (RGT-X) or without (RGT-018) brain penetration. Biochemical assays and cellular assays were utilized to drive the structure-activity relationship (SAR). *In vitro* and *in vivo* target engagement were confirmed. *In vivo* efficacy study data were generated using lung and pancreatic cancer xenograft mouse models with KRAS mutation.

Results

RGT-018 blocked the interaction of SOS1:KRAS with high potency and selectivity

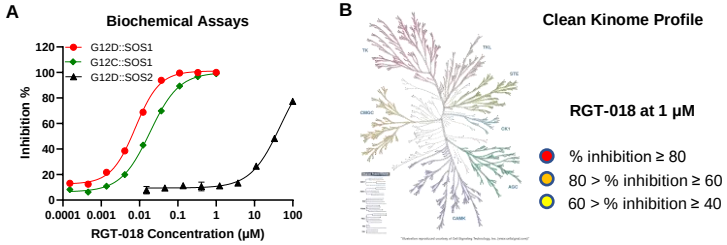

Figure 1. (**A**) Biochemical characterization of RGT-018 was carried out through the analysis of interaction assays using SOS1 and SOS2 recombinant proteins with $KRAS^{G12D}$ or $KRAS^{G12C}$ mutants. (**B**) RGT-018 was tested in a panel of 330 kinases by Kinome Wide Panel (KWP) service to determine the inhibition of kinase activity. RGT-018 at 1 μM concentration didn't show inhibition of the tested kinases greater than 30%.

Results

RGT-018 inhibited KRAS signaling and proliferation of a broad spectrum of KRAS-driven cancer cells as a single agent

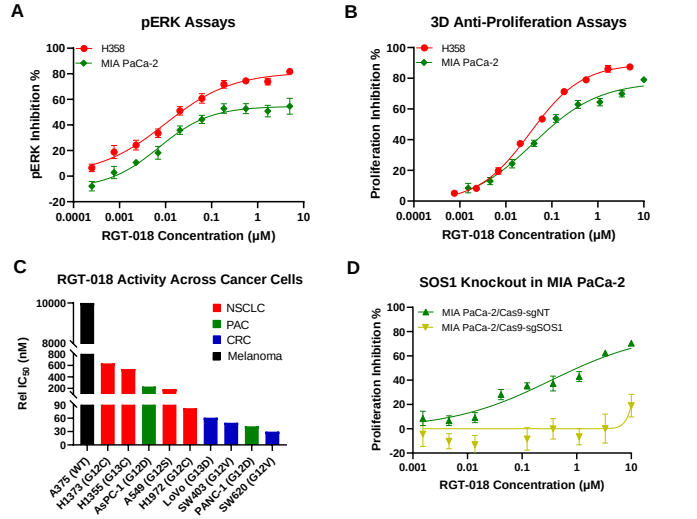

Figure 2. RGT-018 inhibited pERK level (**A**) and 3D growth (**B**) of H358 and MIA PaCa-2 cells harboring $KRAS^{G12C}$ mutation in a dose-dependent manner, suggesting a clear correlation between signaling pathway and growth inhibition by RGT-018 in KRAS-driven cancer cell lines. (**C**) The anti-proliferation activity of RGT-018 was also investigated in a broader panel of cancer cell lines in 3D anti-proliferation assays, including NSCLC, PAC, CRC cells. (**D**) MIA PaCa-2 cells carrying a genetically engineered SOS1 knockout (MIA PaCa-2/Cas9-sgSOS1) were generated to evaluate the anti-proliferation effect of RGT-018 in 3D model, compared with MIA PaCa-2 cells containing Cas9 and non-targeting (NT) sgRNA (MIA PaCa-2/Cas9-sgNT).

RGT-018 exhibited robust anti-proliferation activity when combined with targeted therapies *in vitro*

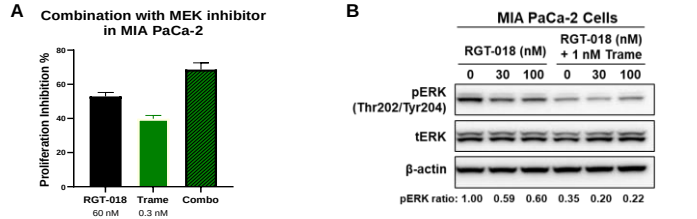

Figure 3. (**A**, **B**) The combination of RGT-018 with MEK inhibitor trametinib (Trame) yielded more robust anti-proliferation effect in 3D model and further suppression of pERK level in MIA PaCa-2 cells.

Results

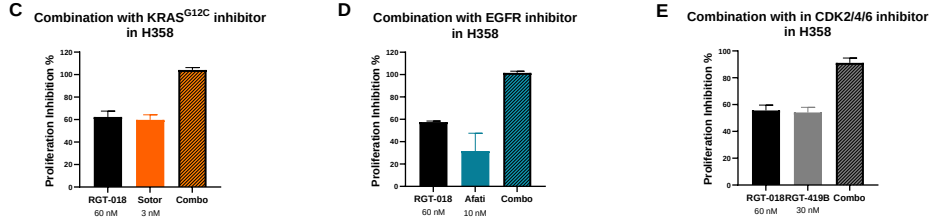

Figure 3. (**C**, **D**, **E**) The combination of RGT-018 with inhibitors of $KRAS^{G12C}$ (sotorasib, Sotor), EGFR (afatinib, Afati) and CDK2/4/6 (RGT-419B) resulted in more robust anti-proliferation effects in H358 cells.

RGT-018 inhibited tumor growth and suppressed KRAS signaling in tumor xenografts as a single agent and in combination with MEK or $KRAS^{G12C}$ inhibitors

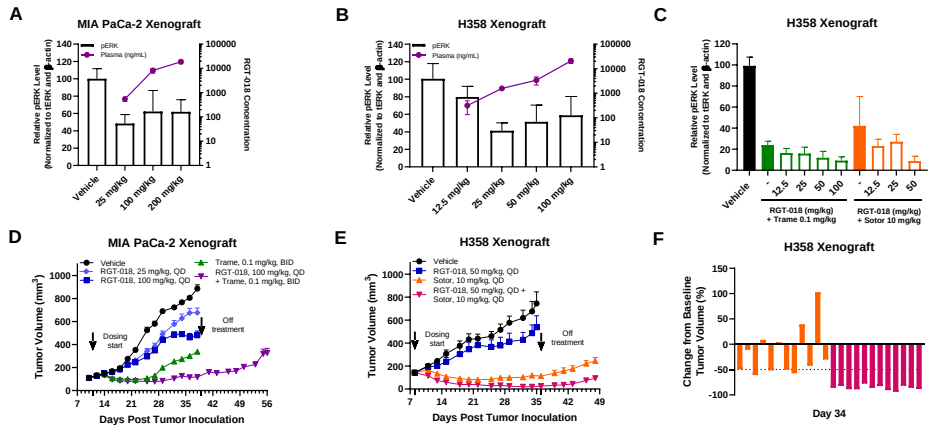

Figure 4. (**A**, **B**, **C**) The MIA PaCa-2 or H358 tumor-bearing mice were administered with RGT-018 (*p.o.* QD) at the doses of 12.5, 25, 50, 100 or 200 mg/kg or a combination of RGT-018 and trametinib (0.1 mg/kg, *p.o.* BID), or RGT-018 and sotorasib (10 mg/kg, *p.o.* QD) for 3 days. Plasma and tumors were collected at 8 h after the last dosage on Day 3. (**D**, **E**, **F**) The efficacy of RGT-018 was evaluated in subcutaneous MIA PaCa-2 or H358 xenografts as a single agent or in combination with trametinib or sotorasib. RGT-018 was well tolerated with less than 10% body weight changes in tumor-bearing mice.

Conclusions

1. RGT-018 displays promising pharmacological properties and represents an attractive drug candidate with oral bioavailability for combination with targeted agents to treat various KRAS-driven tumors (NSCLC, PAC, CRC, etc.). **RGT-018 is IND-ready.**
2. **Regor has generated both non-brain-penetrant (RGT-018) and brain-penetrant (RGT-X) SOS1 inhibitors.**

References

1. *Nat Rev Drug Discov*. 2020 Aug;19(8):533-552.
2. *Biochim Biophys Acta Rev Cancer*. 2020 Dec;1874(2):188445.
3. *Curr Opin Chem Biol*. 2021 Jun;62:109-118.
4. *Nat Rev Clin Oncol*. 2022 Oct;19(10):637-655.

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