BBO-11818: an orally bioavailable, highly potent and selective non-covalent pan-KRAS(ON) and (OFF) inhibitor with robust anti-tumor activity in KRAS-mutant preclinical models

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# Equal contribution

Overview

- Oncogenic variants of KRAS drive tumor growth and metastasis through aberrant signaling, making them important therapeutic targets.[1] Inhibitors against KRAS^{G12C} have recently been approved, but a major clinical need for agents against other KRAS variants remains.[1, 2]
- We have developed BBO-11818: a potent, selective, orally bioavailable non-covalent KRAS inhibitor with activity against multiple KRAS mutants, including KRAS^{G12D} and KRAS^{G12V}.[3]
- BBO-11818 targets KRAS in both its inactive GDP-bound and active GTP-bound states, potently suppressing MAPK signaling and inhibiting cell proliferation in KRAS-mutant cell lines.
- BBO-11818 monotherapy induces strong anti-tumor responses, including strong dose- and time-dependent inhibition of pERK and regressions at well-tolerated doses in CDX models of KRAS-mutant pancreatic, non-small cell lung, and colorectal cancer.
- In combination with BBO-10203, a selective RAS:PI3Kα breaker that blocks RAS-mediated activation of AKT, or cetuximab, an anti-EGFR monoclonal antibody, BBO-11818 shows significantly enhanced efficacy in CDX/PDX models harboring KRAS^{G12D} or KRAS^{G12V} mutations. Similarly, the combination of BBO-11818 and anti-PD-1 antibody improves survival in a KRAS^{G12D} syngeneic model.

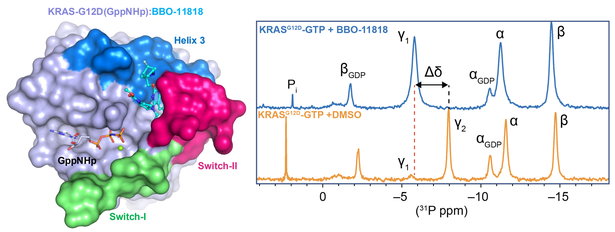
Methods

SPR: Surface plasmon resonance direct binding assays to determine affinity of BBO-11818 to GppNHp- or GDP-loaded avi-tagged KRAS proteins were performed as described in (3).
Crystallization and Structure Determination: Protein samples for crystallization were prepared as described in (3). Diffraction data were collected from Brookhaven National Laboratory National Synchrotron Light Source II beamline 17-ID-2. Data processing and model building were performed as described in (3). Structure figure was generated with PyMOL.
^{31}P NMR: NMR data were collected as described in (3). Briefly, ^{31}P NMR data were collected on a sample of KRAS^{G12D}-GTP in the absence and in presence of BBO-11818 (1:1.5 P:L stoichiometric ratio). NMR data were processed and analyzed in Bruker TopSpin 4.1.4.
Protein:protein interaction: A PPI Homogeneous Time-Resolved Fluorescence (HTRF) assay was used to determine compound effectiveness in disrupting KRAS protein and effector (RAF1) binding.
ERK phosphorylation. Cells were seeded and the next day treated with BBO-11818. Two hours post-treatment, pERK phosphorylation was assessed by HTRF.
3D viability. Cells were seeded and treated with BBO-11818 three days post-seeding after spheroid formation. Four days post-treatment, viability was assessed with the CellTiter-Glo viability assay.
Long-term 2D clonogenic assay. Cells were seeded, treated 24 hours later with BBO-11818, or cetuximab and incubated for 15 or 20 days. Media and compounds were changed biweekly. Confluence was measured twice daily using an Incucyte Live-Cell Analysis System.
Pharmacokinetics (PK) and pharmacodynamics (PD). Dose and time response PK/PD analyses were performed following a single oral dose of BBO-11818. Plasma and tumors were collected for PK and pERK analysis using the MesoScale Discovery platform.
***In vivo* efficacy and survival studies.** BBO-11818 efficacy was assessed following twice daily (BID) oral dosing at the indicated dose levels in cell line-derived or patient-derived xenograft (CDX, PDX) or syngeneic models bearing KRAS^{G12D} or KRAS^{G12V} mutations. BBO-10203 was dosed orally once daily (QD). Anti-PD-1 or cetuximab were administered twice weekly (BIW) by intraperitoneal administration. Tumor growth inhibition (TGI), mean tumor regression (REG), and number of complete regressions (CR) were calculated.
Statistical analyses: Two-way repeated measures ANOVA followed by post hoc Tukey's multiple comparison test through day 15 or 16 were performed for clonogenic assays. Two-way repeated measures ANOVA for in vivo efficacy studies were performed with Dunnett's test vs the vehicle group or between the indicated groups.

BBO-11818 is a potent and selective pan-KRAS binder and KRAS:RAF1 PPI inhibitor

		BBO-11818	
	RAS Allele	**GppNHp**	**GDP**
RAS SPR, K_D (nM)	KRAS^{G12D}	7.40	<0.003
	KRAS^{G12V}	13.2	0.037
	KRAS^{G13D}	17.5	0.30
	KRAS^{WT}	20.0	0.25
	NRAS^{WT}	725,000	2,455
	HRAS^{WT}	>200,000	831
PPI: KRAS(GTP)/RAF1 effector, IC_{50} (nM)	KRAS^{G12D}	28	
	KRAS^{G12V}	61	
	KRAS^{G12C}	47	
	KRAS^{G12R}	51	
	KRAS^{WT}	120	

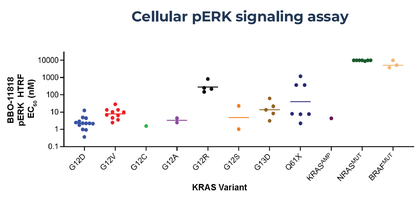
BBO-11818 binds Switch-II and Helix 3 of KRAS^{G12D}-GTP and shifts it to the inactive state 1 conformation



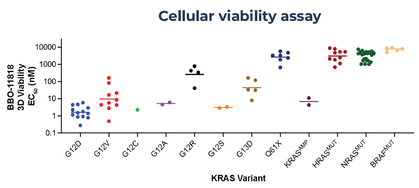
BBO-11818 has a favorable ADME and PK profile and is orally bioavailable

Parameter	BBO-11818
Mouse PK CL (mL/min/kg) / V (L/kg) / % F	45 / 4.9 / 18
Rat PK CL (mL/min/kg) / V (L/kg) / % F	30 / 7.8 / 16
Dog PK CL (mL/min/kg) / V (L/kg) / % F	11 / 5.8 / 28
Minipig PK CL (mL/min/kg) / V (L/kg) / % F	47 / 7.8 / 27
Selectivity: hERG & safety panel	No red flags

BBO-11818 inhibits ERK phosphorylation and cell proliferation in KRAS-mutant cell lines

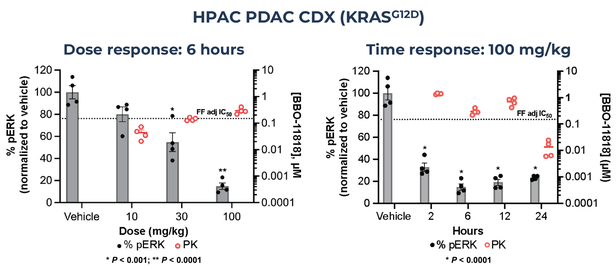


KRAS variant	Mean EC_{50} (nM)
KRAS^{G12D}	3.08
KRAS^{G12V}	9.91
KRAS^{G12C}	1.53
KRAS^{G12A}	3.46
KRAS^{G12R}	357
KRAS^{G12S}	11.9
KRAS^{G13D}	21.7
KRAS^{Q61X}	278
KRAS^{AMP}	4.35
NRAS^{MUT}	> 10 µM
BRAF^{MUT}	> 10 µM

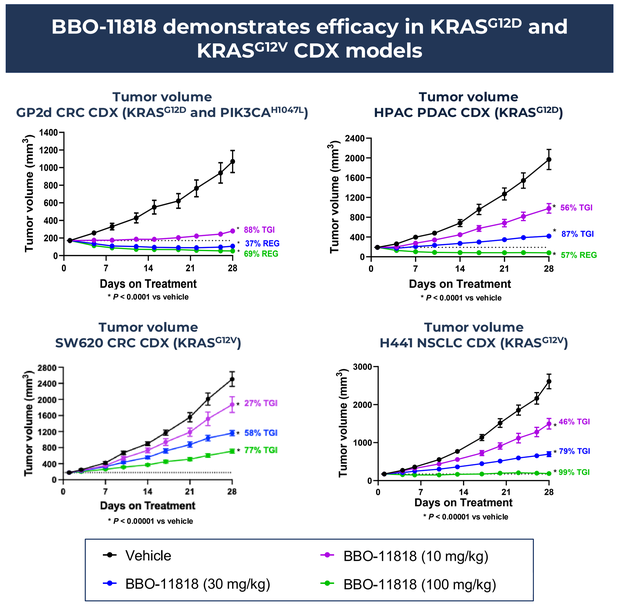


KRAS variant	Mean EC_{50} (nM)
KRAS^{G12D}	2.21
KRAS^{G12V}	31.2
KRAS^{G12C}	2.26
KRAS^{G12A}	5.32
KRAS^{G12R}	400
KRAS^{G12S}	3.09
KRAS^{G13D}	71.7
KRAS^{Q61X}	3,170
KRAS^{AMP}	7.62
HRAS^{MUT}	4,030
NRAS^{MUT}	3,720
BRAF^{MUT}	7,430

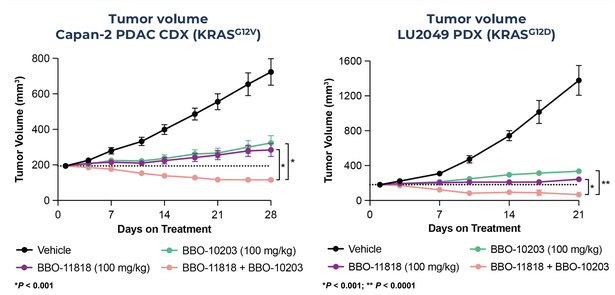
BBO-11818 demonstrates dose- and time-dependent inhibition of pERK in a KRAS^{G12D} model



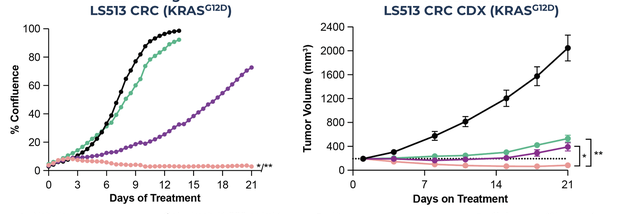
BBO-11818 demonstrates efficacy in KRAS^{G12D} and KRAS^{G12V} CDX models



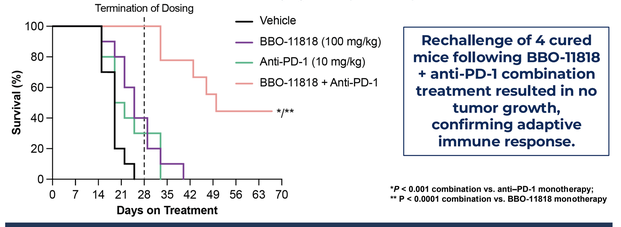
BBO-11818 and BBO-10203 (RAS:PI3Kα breaker) show a combination effect in CDX and PDX models



BBO-11818 and EGFR inhibitors demonstrate combination benefit *in vitro* and *in vivo*



The combination of BBO-11818 and anti-PD-1 antibody improves survival *in vivo*



Rechallenge of 4 cured mice following BBO-11818 + anti-PD-1 combination treatment resulted in no tumor growth, confirming adaptive immune response.

Conclusions

- BBO-11818 is a potent pan-KRAS inhibitor targeting both GTP-bound and GDP-bound forms of KRAS, with good selectivity over HRAS and NRAS.
- BBO-11818 binds in the Switch-II/Helix 3 pocket, inducing conformational changes incompatible with effector binding.
- BBO-11818 potently inhibits ERK phosphorylation and proliferation in KRAS-dependent cell lines *in vitro*.
- BBO-11818 has favorable PK and oral bioavailability and shows dose- and time-dependent inhibition of pERK in *in vivo* PD studies.
- BBO-11818 demonstrates robust efficacy in KRAS^{G12D} and KRAS^{G12V} CDX models.
- BBO-11818 exhibits *in vivo* combination effect with the RAS:PI3Kα breaker BBO-10203 and cetuximab in KRAS-mutant CDX and PDX models.
- BBO-11818 also shows a combination benefit with anti-PD-1 treatment, resulting in complete tumor regressions in the CT26 syngeneic model.
- The Phase 1a/1b KONQUER-101 study (NCT06917079) has been initiated and is enrolling patients globally with KRAS G12A, G12C, G12D, G12S, or G12V mutation, or KRAS-amplification.

References and acknowledgements

1- Prior IA., Hood FE, and Hartley JL. The frequency of Ras mutations in cancer. *Cancer Research 2020*; 80(14): 2969-2974.
2- Liu J, Kang R, Tang D. The KRAS-G12C inhibitor: activity and resistance. *Cancer Gene Therapy* 2022; 29(7):875-8.
3- Stahlhut C, Maciag AE, Sullivan KA, Singh K, Gitego N, Zhang Z et al. Discovery of BBO-11818, a Potent and Selective Noncovalent Inhibitor of (ON) and (OFF) KRAS with Activity against Multiple Oncogenic Mutants. *Cancer Discovery* 2026; 16 (4): 740–759.

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