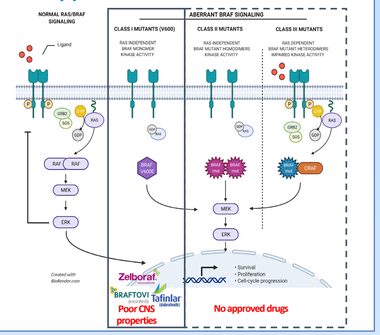
Preclinical efficacy of BDTX-4933, a brain penetrant MasterKey inhibitor targeting oncogenic BRAF Class I/II/III mutations

Pui Yee Ng, Yoon-Chi Han, Luisa Shin Ogawa, Ryan Schulz, Shao Ning Yang, Ivan Jewett, Miao Chen, Noboru Ishiyama, Darlene Romashko, Matthew Lucas, Tai-An Lin, Elizabeth Buck

Black Diamond Therapeutics, Cambridge, MA and New York, NY

34th EORTC-NCI-AACR Symposium | Oct. 26-28, 2022

BDTX-4933 addresses multiple gaps in FDA-approved BRAF V600 inhibitors



Background

FDA-approved BRAF inhibitors target V600 (Class I) mutant monomers and are largely inactive against mutant BRAF dimers. Dimeric mutants found in many solid tumors including primary CNS tumors and brain metastases, can drive RAS-independent (Class II) or RAS-dependent (Class III) oncogenic tumor growth. The approved BRAF inhibitors also induce paradoxical RAF activation that limits their activity.

Although currently approved BRAF V600 mutation-selective inhibitors demonstrated efficacy in brain tumors and metastases when combined with MEK inhibitors in clinical trials, duration of response tends to be short. We believe this is due, at least in part, to limited blood-brain barrier permeability. There remains a high unmet clinical need for a broad Class I/II/III BRAF inhibitor with high CNS penetrant activity for patients with RAF-dependent tumors carrying a broad spectrum of BRAF alterations.

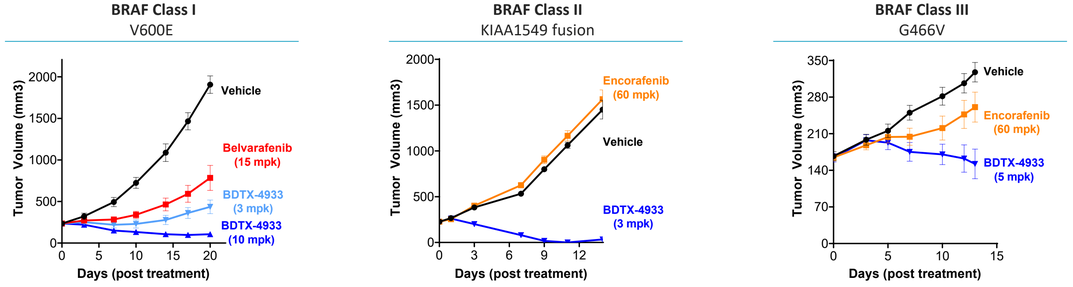
Black Diamond Therapeutics' Mutation-Allostery-Pharmacology (MAP) platform uses NGS data and a proprietary machine-learning algorithm to predict and then scaled biology to validate the oncogenicity of previously uncharacterized groups of oncogenic BRAF mutations across Class I/II/III and to identify small molecule MasterKey inhibitors against this spectrum of mutations.

MasterKey inhibitor BDTX-4933 is a CNS penetrant small molecule inhibitor that targets oncogenic BRAF Class I/II/III mutations without paradoxical RAF activation.

Results

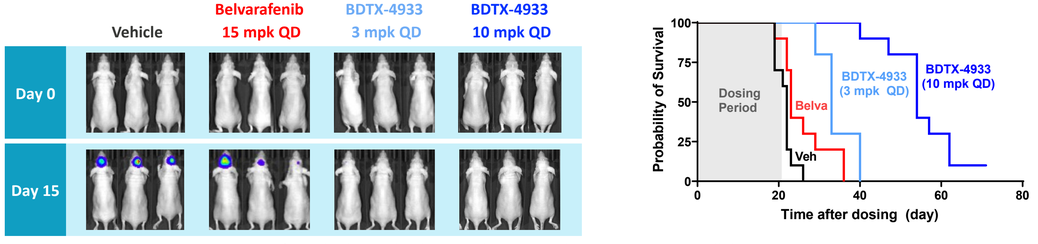
BDTX-4933 is a potent and selective, reversible inhibitor that targets a wide spectrum of oncogenic Class II/III BRAF alterations as well as Class I BRAF V600 mutations along with constitutively active RAF dimers resulting from other upstream oncogenic MAPK pathway alterations, including NRAS alterations. In mouse xenograft and allograft studies, BDTX-4933 achieves regression of tumors carrying BRAF Class I, II, and III mutations. BDTX-4933 possesses good CNS penetrant properties as evidenced by high Kpuu and intracranial anti-tumor activity. BDTX-4933 retains potent activity against PDX cell lines and BRAFi/MEKi combination resistant cell lines.

BDTX-4933 achieves strong anti-tumor activity across all BRAF mutation classes in *in vivo* models



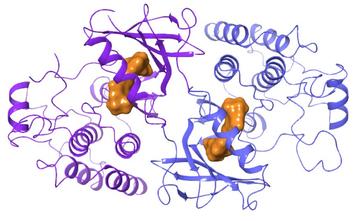
No significant body weight changes observed with BDTX-4933 across all doses tested

BDTX-4933 prolongs survival in intracranial tumor model of BRAF V600E



No significant body weight changes observed with BDTX-4933 across all doses tested

BDTX-4933 is an active-site inhibitor that binds to both protomers of a mutant BRAF dimer



Crystal structure of dimeric BRAF G469A in complex with BDTX-4933

BDTX-4933 achieves on-target inhibition of cell proliferation driven by wide spectrum of oncogenic BRAF and NRAS mutations

Cell Proliferation IC50				
Mutation		BDTX-4933	Encorafenib	Belvarafenib
Class I	V600E			
Class II & III (Non-V600)	G466A			
	G469A			
	L597Q			
	K601N			
	K601E			
	F247L			
	KIAA fusion			
NRAS	Q61K			
WT	WT	>1000 nM	Paradoxical Activation	>1000 nM

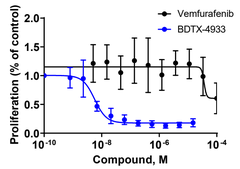
Target Engagement, pERK IC50				
Cancer cell lines with BRAF mutation		BDTX-4933	Encorafenib	Belvarafenib
Class I	V600E			
Class II & III (Non-V600)	K601E			
	Loop deletion			
	Fusion			
NRAS	Q61R			

IC50 color code
- < 50 nM
- 50 – 150 nM
- >150 nM

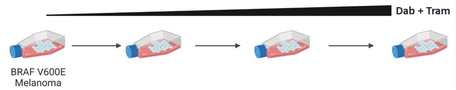
BDTX-4933 potently inhibits melanoma PDX organoids

Cell Proliferation IC50		
Melanoma PDX organoids		BDTX-4933
Class I	V600E	
Class II	G469A	
Class III	L597R	
NRAS	Q61R	

IC_{50} color code: < 100 nM



BDTX-4933 overcomes BRAFi + MEKi combo resistance in BRAF V600E cells



Dabrafenib + Trametinib Resistant cell line

Cell Proliferation	IC50 (nM)	
	Parental	Dab + Tram Resistant
Dabrafenib		
Trametinib		
LXH254		
BDTX-4933		

IC50 color code
- < 100 nM
- 100 – 500 nM
- >500 nM

BDTX-4933 inhibits BRAF & CRAF homodimers and heterodimers in KRAS mutant cells*

pERK IC50			
Compounds	KO: ARAF Expressed: B/CRAF	KO: ARAF + BRAF Expressed: CRAF	KO: ARAF + CRAF Expressed: BRAF
BDTX-4933			
Encorafenib			
Vemurafenib			
LXH254			
Belvarafenib			
Fore-8394			

IC50 color code
- <50 nM
- 50-500 nM
- >500 nM

* ARAF, ARAF+BRAF, or ARAF+CRAF were KO by CRISPR in KRAS mutant cells

Conclusions

BDTX-4933 has all the attributes of a best-in-class CNS penetrant BRAF inhibitor to address patients with and without CNS disease whose tumors express monomeric (Class I) or dimeric (Class II and III) BRAF mutants, or constitutively active RAF dimers resulting from other upstream oncogenic MAPK pathway alterations, including NRAS alterations. BDTX-4933 achieves potent, on-target inhibition of the RAF-MEK-ERK signaling pathway and anti-tumor activity in multiple preclinical tumor models, including intracranial models. IND-enabling studies for BDTX-4933 are underway.

Pui Yee Ng: png@bdtx.com, Yoon-Chi Han: ychan@bdtx.com

BLACK DIAMOND THERAPEUTICS