

AACR

American Association
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**ANNUAL
MEETING**

2023

APRIL 14-19 • #AACR23



The Discovery and Characterization of CFT1946: A Potent, Selective, and Orally Bioavailable Degradator of Mutant BRAF for the Treatment of BRAF-driven Cancers

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on behalf of the C4T BRAF Project Team

C4 Therapeutics, Inc.

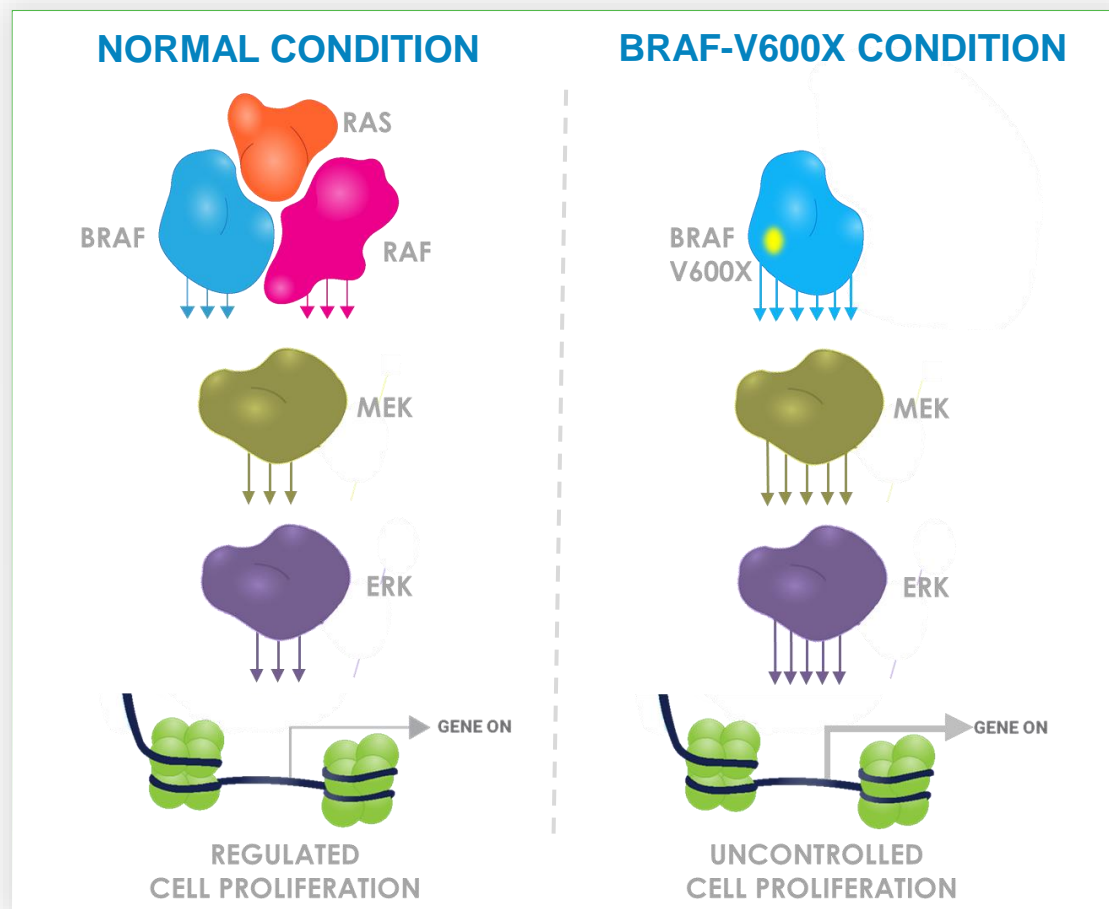
Watertown, MA

Disclosure Information

Yanke Liang, Ph.D.

- I have the following financial relationships to disclose:
 - Stockholder in: C4 Therapeutics, Inc.
 - Employee of: C4 Therapeutics, Inc.

Mechanism of Action for BRAF-V600X Driven Human Cancers



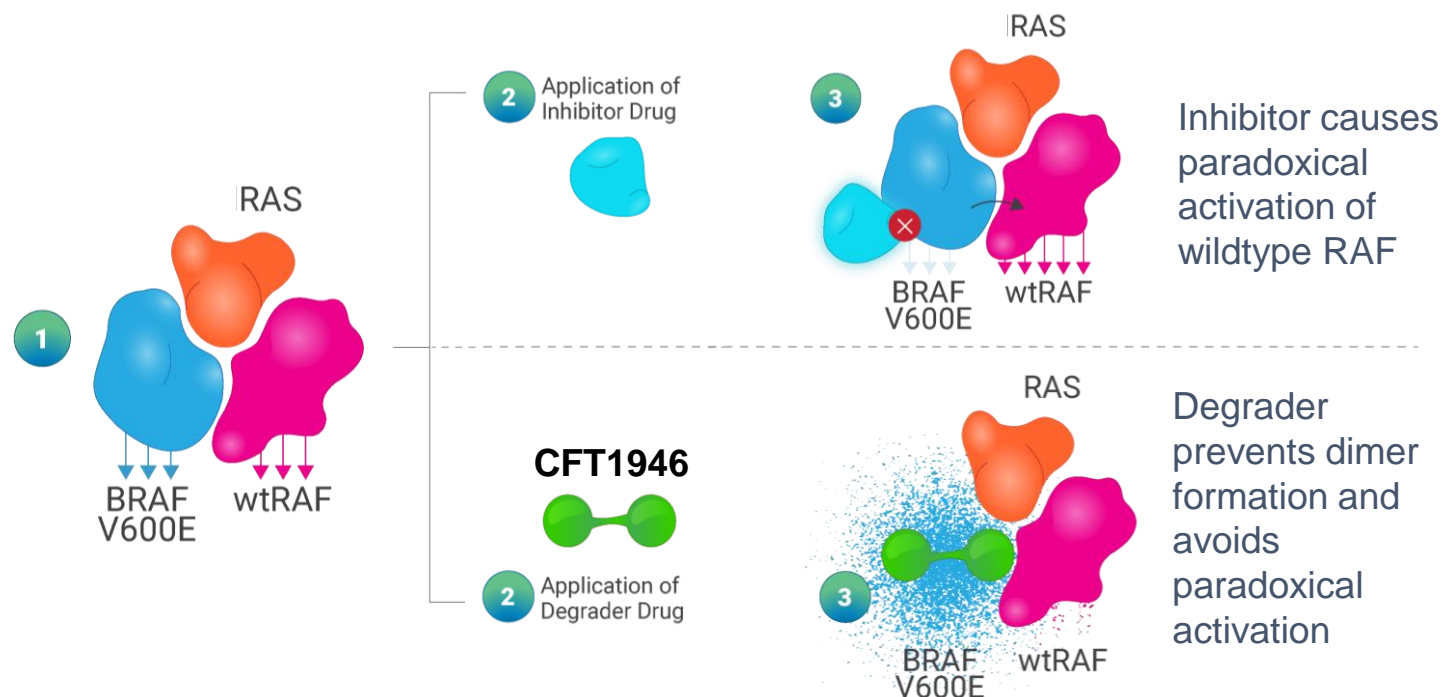
MAPK, MAP kinase.

Mechanism of BRAF-V600X Driven Cancer

- BRAF is a serine/threonine protein kinase in the MAPK pathway that promotes cell proliferation and survival when activated through extracellular signals
- Constitutively active BRAF-V600X causes uncontrolled MAPK signaling, leading to tumorigenesis and tumor growth
- Decreasing BRAF-V600X activity in these cancers leads to growth arrest, cell death, and tumor regression
- BRAF-V600X is a clinically validated oncology target, however limitations in currently approved inhibitors highlight the need for additional BRAF-V600X targeted therapies

Utilizing a Degradator Approach to Overcome Limitations of BRAF Inhibition

Degradator Rationale



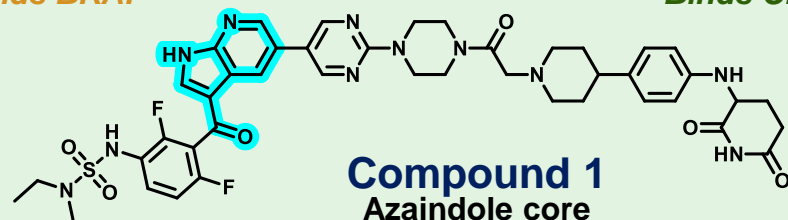
Advantages of BRAF V600X Degradation

- Specifically target mutant BRAF-V600X over wildtype BRAF
- Prevent mutant BRAF-V600X incorporation into RAF dimers
- Avoid paradoxical activation of RAF dimers
- Address failures in inhibitor-based therapy due to resistance mechanisms
- Effect deep elimination of mutant BRAF signaling and create durable responses

Scaffold Hopping to Address Pharmacokinetics (PK) and Solubility Challenge

Binds BRAF

Binds CRBN



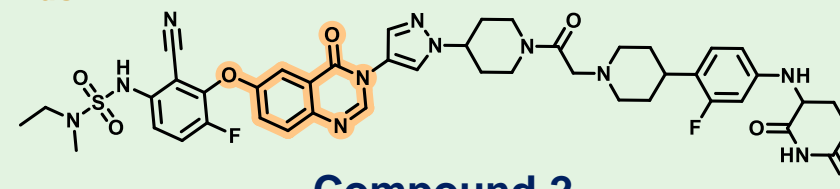
Compound 1
Azaindole core
(medchem previously described)

Mouse IV Cl = **10.8** mL/min/kg
PO DN_AUC = **152** (ng*h/mL)/(mg/kg)
F = **10%**

Scaffold hopping

Binds BRAF

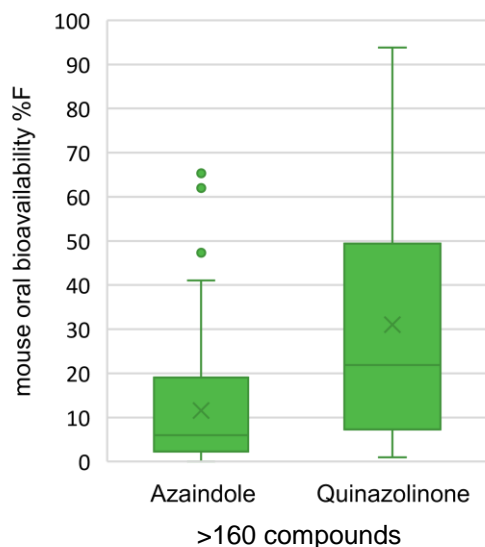
Binds CRBN



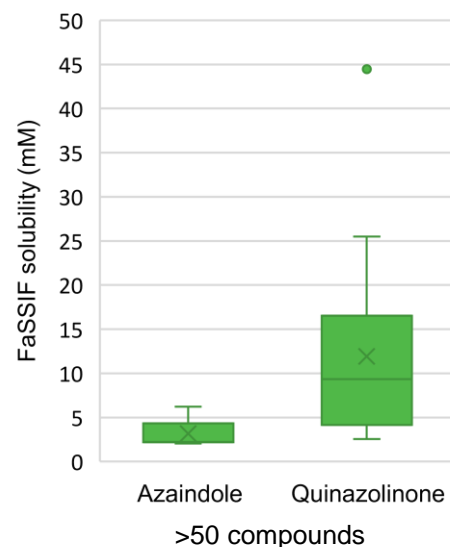
Compound 2
Quinazolinone core

Mouse IV Cl = **1.8** mL/min/kg
PO DN_AUC = **1345** (ng*h/mL)/(mg/kg)
F = **14%**

Oral bioavailability



Solubility

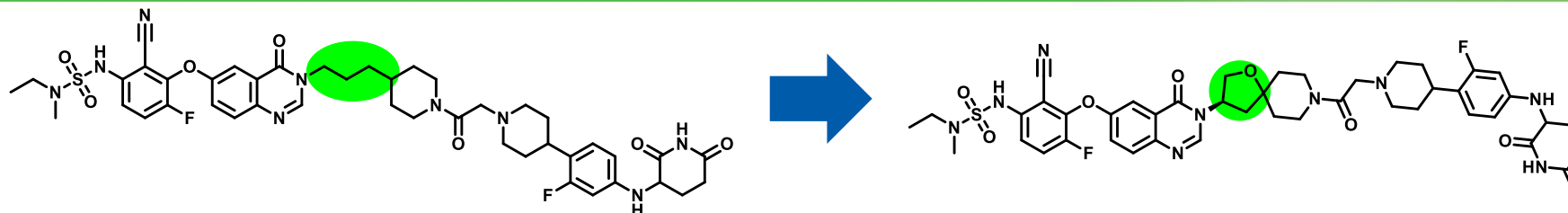


Advantages of BRAF quinazolinone BiDAC™ degraders:

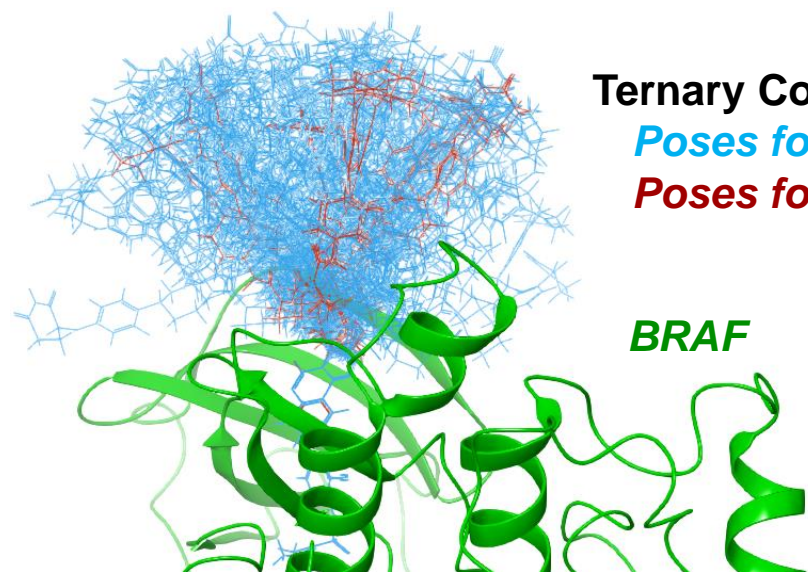
- Lower mouse IV PK clearance
- Higher mouse oral exposure and bioavailability
- Generally more soluble

Strategy: Identify a quinazolinone-based BRAF degrader suitable for oral dosing

Rigidifying Spacer Region Improved Degradation Efficiency and PK



| | Compound 3 | Compound 4 |
|--|-------------|-------------|
| Rotatable Bonds | 15 | ▼ 12 |
| BRAF-V600E DC ₅₀ / E _{max} [6 h] | 71 nM / 28% | 53 nM / 19% |
| Mouse IV Cl [mL/min/kg] | 12.8 | 3.5 |
| Mouse F [%] | 14 | 40 |



Ternary Complex Modeling

Poses for Compound 3

Poses for Compound 4

BRAF

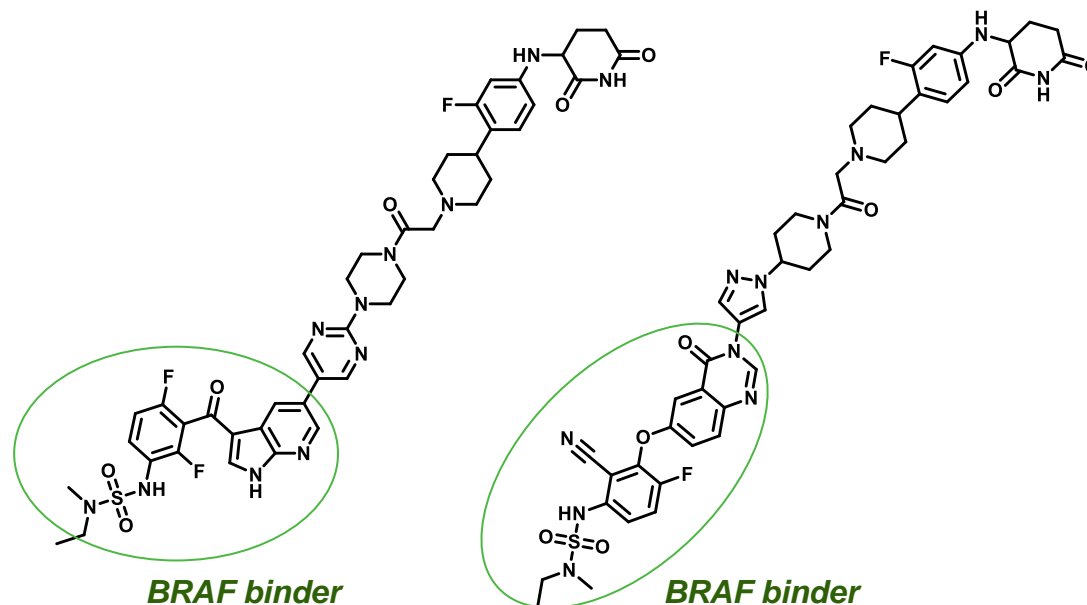
* CRBN proteins removed for clarity

BRAF BiDAC degrader with spirocyclic spacer:

- Sample narrower, focused ternary complex regions that favor more catalytically efficient poses
- Lowered mouse IV PK clearance
- Improved mouse oral bioavailability

DC₅₀, [degrader] needed for 50% target depletion
E_{max}, % remaining target at the incubation timepoint

Binary Binding Potency on Both Ends of a BiDAC Degradator Impacted Degradation Efficiency

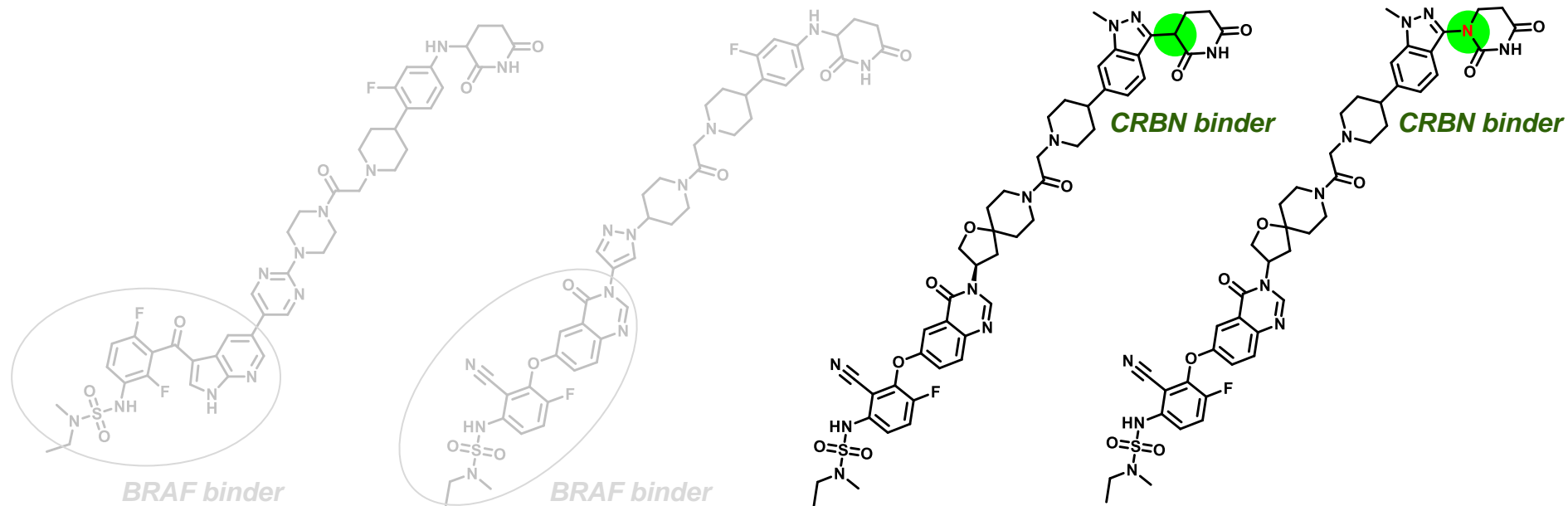


Compound 5

Compound 2

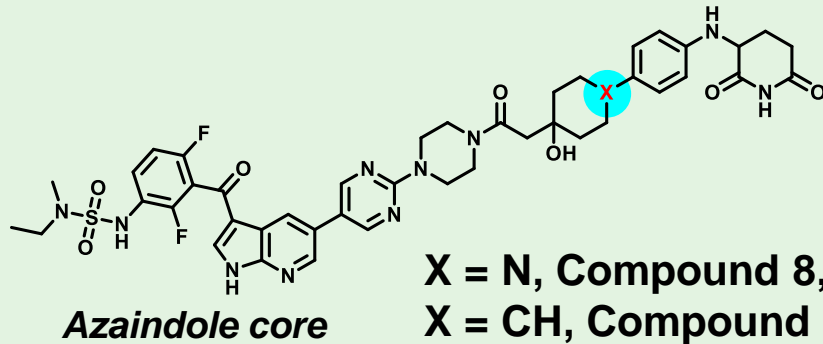
| Scaffold | Compound 5 | Compound 2 |
|---|-------------------|--------------------|
| BRAF-V600E Ki [nM] | 17 | ▼ 0.1 |
| CRBN FP Kd [nM] | 92 | 96 |
| BRAF-V600E DC₅₀ / E_{max} [6 h] | 8 nM / 18% | 92 nM / 41% |

Binary Binding Potency on Both Ends of a BiDAC Degradator Impacted Degradation Efficiency

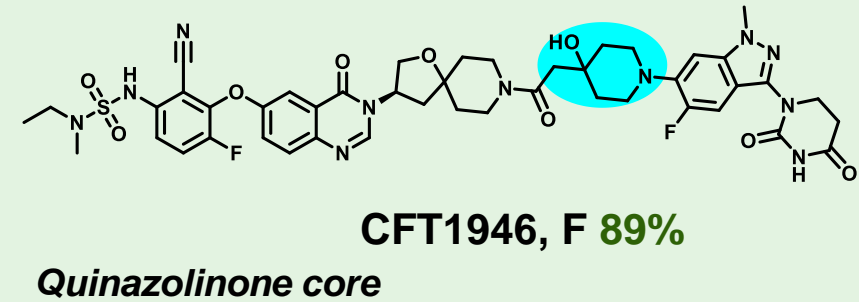


| | Compound 5 | Compound 2 | Compound 6 | Compound 7 |
|---|------------|---------------|--------------------|--------------------|
| Scaffold | Azaindole | Quinazolinone | Quinazolinone | Quinazolinone |
| BRAF-V600E K_i [nM] | 17 | ▼ 0.1 | 0.2 | 0.6 |
| CRBN FP K_d [nM] | 92 | 96 | 4 | ▲ 111 |
| BRAF-V600E DC₅₀ / E_{max} [6 h] | 8 nM / 18% | 92 nM / 41% | 59 nM / 31% | 39 nM / 17% |

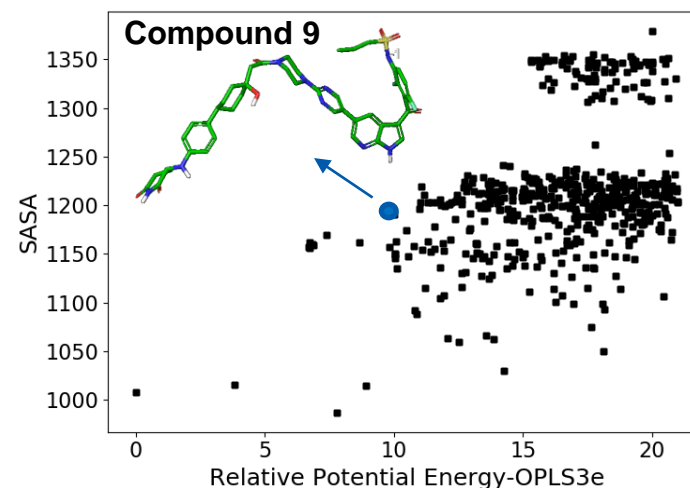
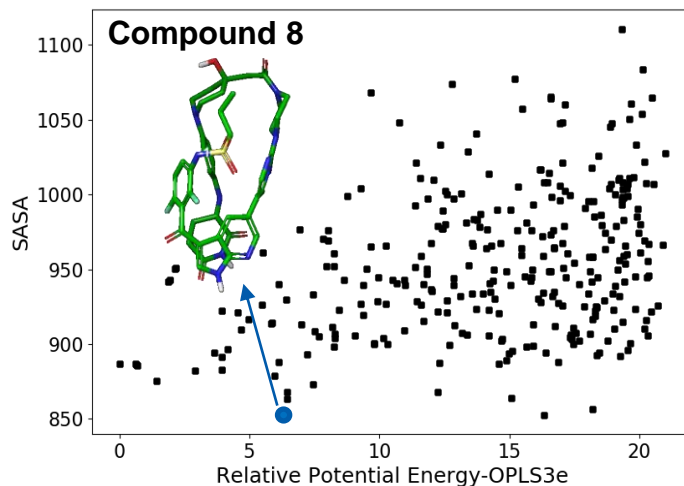
Hydrophobic Collapse Improved Oral Bioavailability



*Applied to
quinazolinone
series*



Solvent Accessible Surface Area (SASA) analysis

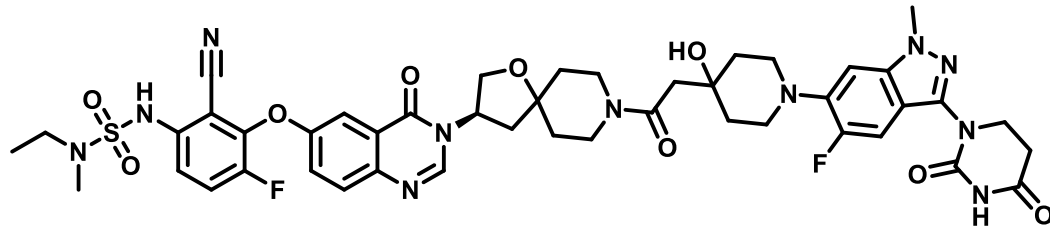


Hypothesis:

- In the context of the BRAF BiDAC degraders, piperidine N provided higher propensity for conformation collapse, resulting in lower SASA

tPSA, topological polar surface area

CFT1946 Displays A Balanced Preclinical Profile



CFT1946

BRAF-V600E DC₅₀ / E_{max} [24 h] **14 nM / 26%**

A375 NRAS^{Q61K} * pERK 1 h [nM] **42**

A375 NRAS^{Q61K} * GI₅₀ 96 h [nM] **150**

HepG2 GI₅₀ [μM] **>10**

CL_{obs} Mouse / Rat [mL/min/kg] **0.8 / 0.5**

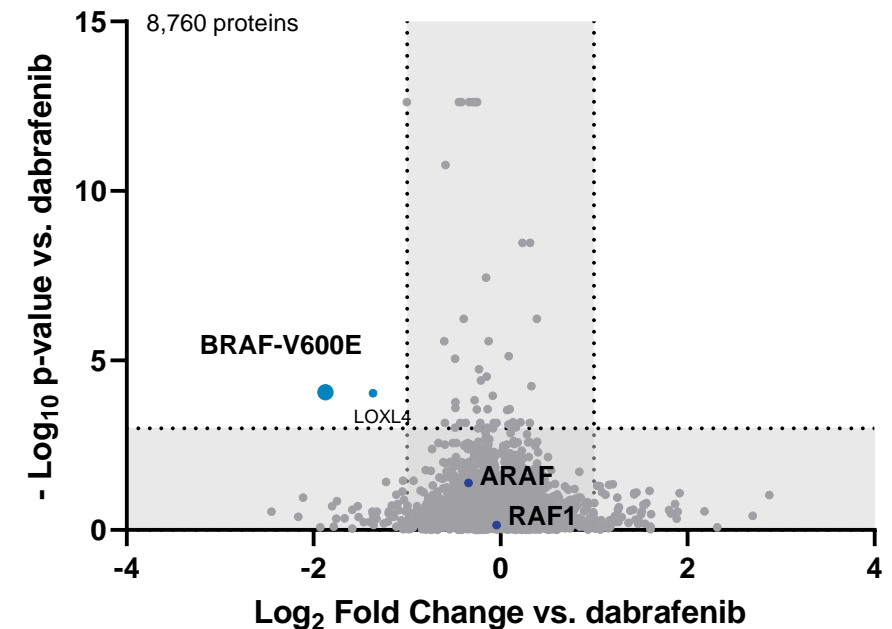
F % Mouse / Rat **89 / 89**

Degradation Selectivity **Exquisite for
BRAF-V600E**

* An engineered disease-relevant BRAF inhibitor resistant cell line

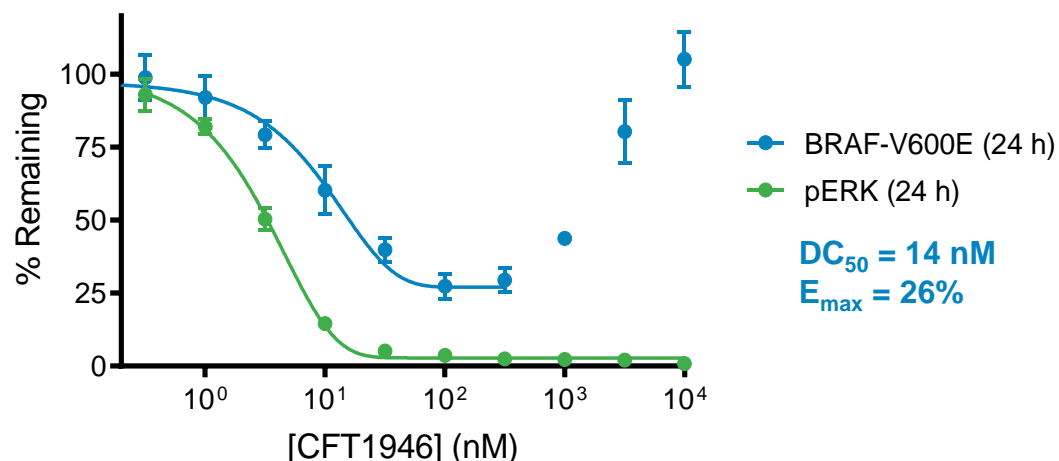
Proteome Profiling Demonstrates Selectivity of CFT1946 for BRAF-V600E

CFT1946 (300 nM, 24 h) in A375 Cells



CFT1946 is an On-Mechanism, CRBN-Based, BRAF-V600X BiDAC™ Degradator

CFT1946 Degrades BRAF-V600E in a Dose Dependent Manner

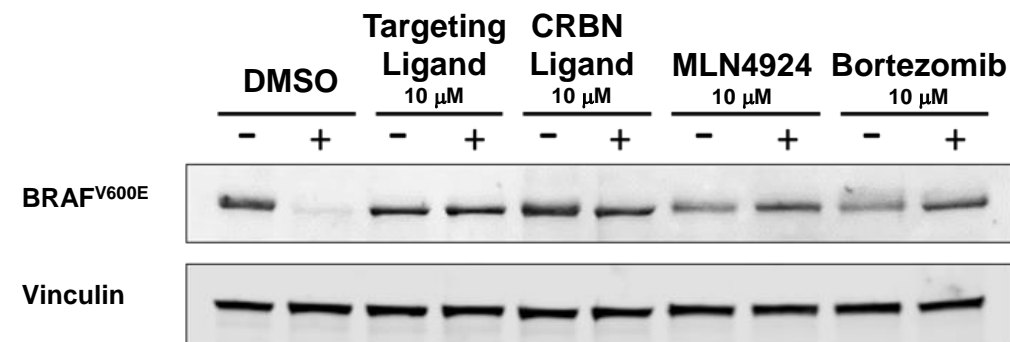


- HiBiT assay shows BRAF-V600E degradation with CFT1946 treatment in dose-dependent manner
- pERK loss aligns with loss of BRAF-V600E protein demonstrating MAPK pathway inhibition

HiBiT; high affinity bioluminescent tag; IMiD, immunomodulatory imide drug.
C4 Therapeutics data on file.

CFT1946 is an On-Mechanism BiDAC™ Degradator

CFT1946 (100 nM) in A375 cells @ 24 h

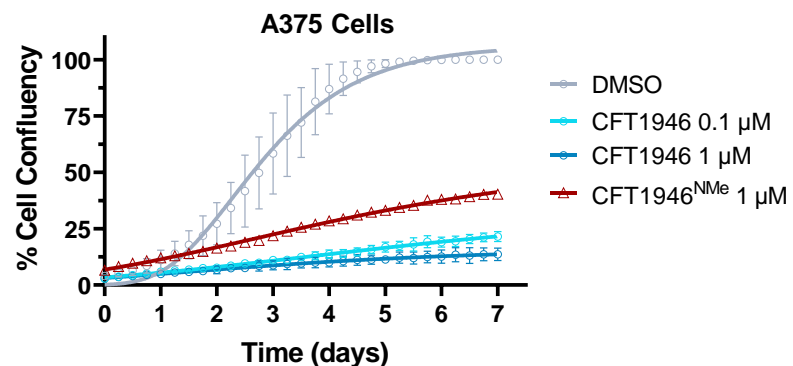
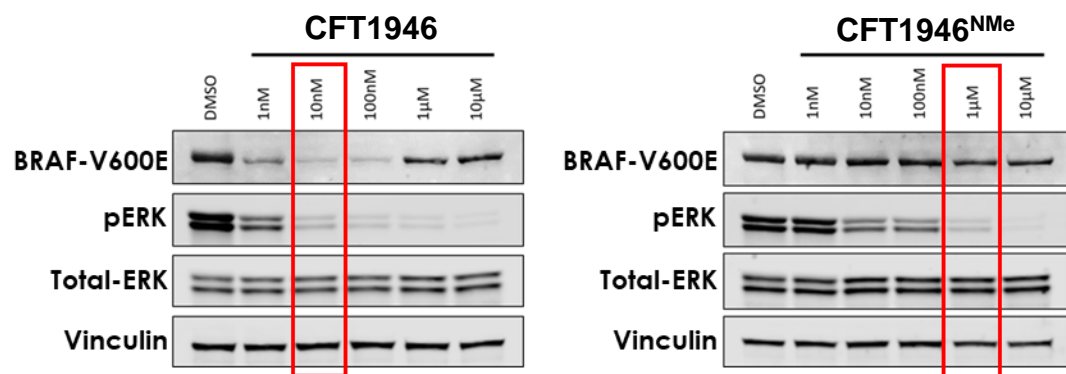


*note: +/- refers to presence or absence of 100 nM CFT1946

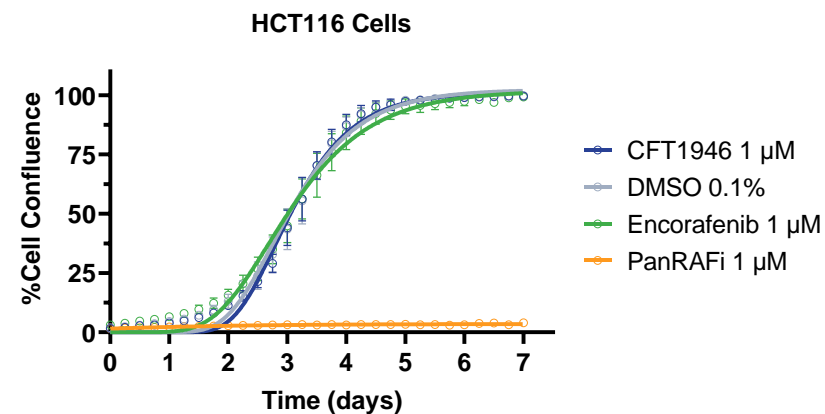
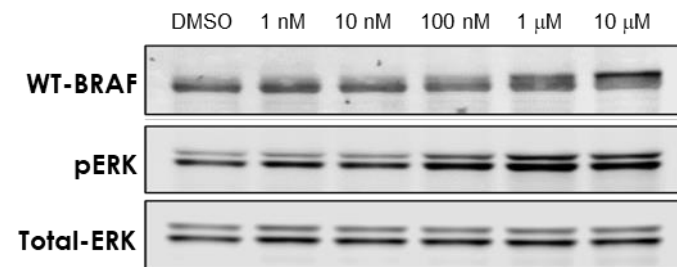
- BRAF-V600E degradation with CFT1946
- No BRAF-V600E degradation with ligand competition, CRBN ligand competition, inhibition of CUL4 E3 with MLN4924 or inhibition of the proteasome with bortezomib

CFT1946 Causes BRAF-V600E Degradation, Potent Inhibition of MAPK Signaling, & Loss of Viability in BRAF-V600E Cells but Not in WT-BRAF Cells

BRAF-V600E Degradation by CFT1946 Causes Loss of MAPK Signaling Superior to Inhibition Alone



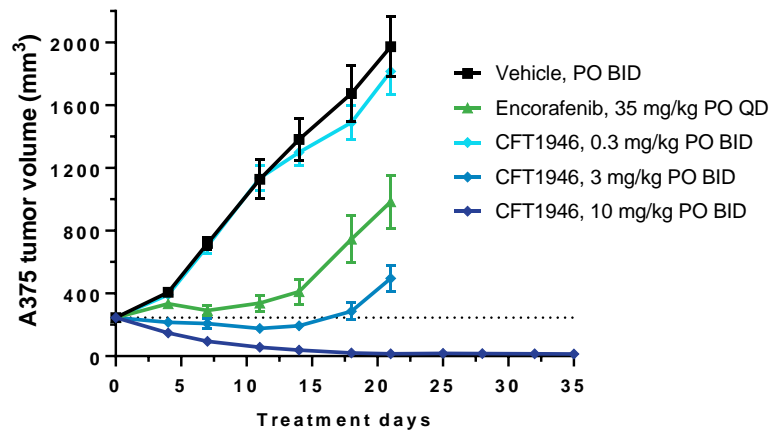
CFT1946 Treatment of WT-BRAF Cells Has No Effect on MAPK Pathway and Cell Growth



*note: CFT1946^{NMe} is a non-CRBN binding version of CFT1946; BRAF is BRAF-V600E MAPK, MAP kinase.
C4 Therapeutics data on file.

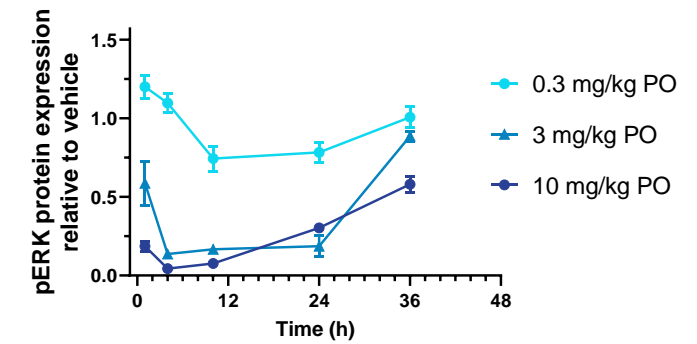
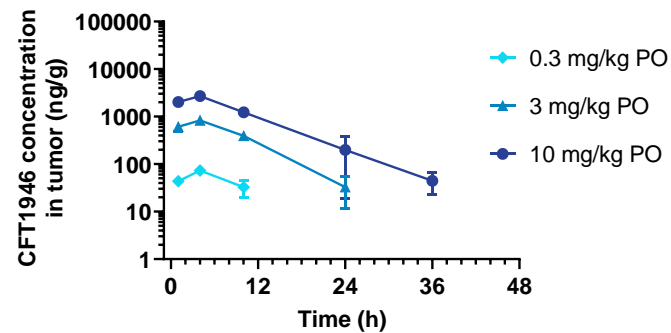
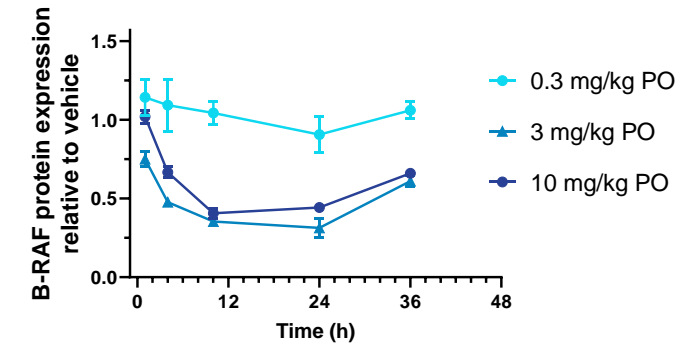
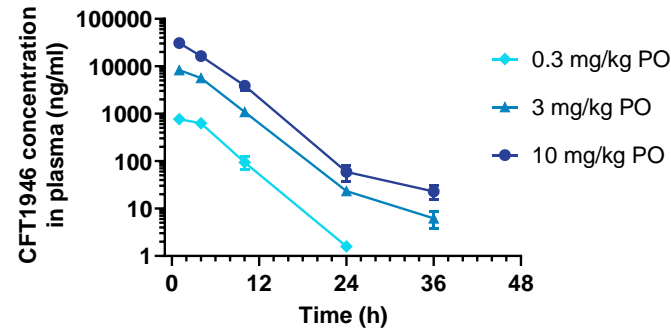
CFT1946 Induces Tumor Regression in the BRAF-V600E A375 Xenograft Mouse Model in Accordance with PK/PD Results

CFT1946 Treatment of A375 Cell Line *in vivo* Shows Dose-Dependent Tumor Regression Superior to Inhibitor



CFT1946 dose-response xenograft data demonstrates that 10 mg/kg BID dose results in sustained tumor regression and is the minimum efficacious dose

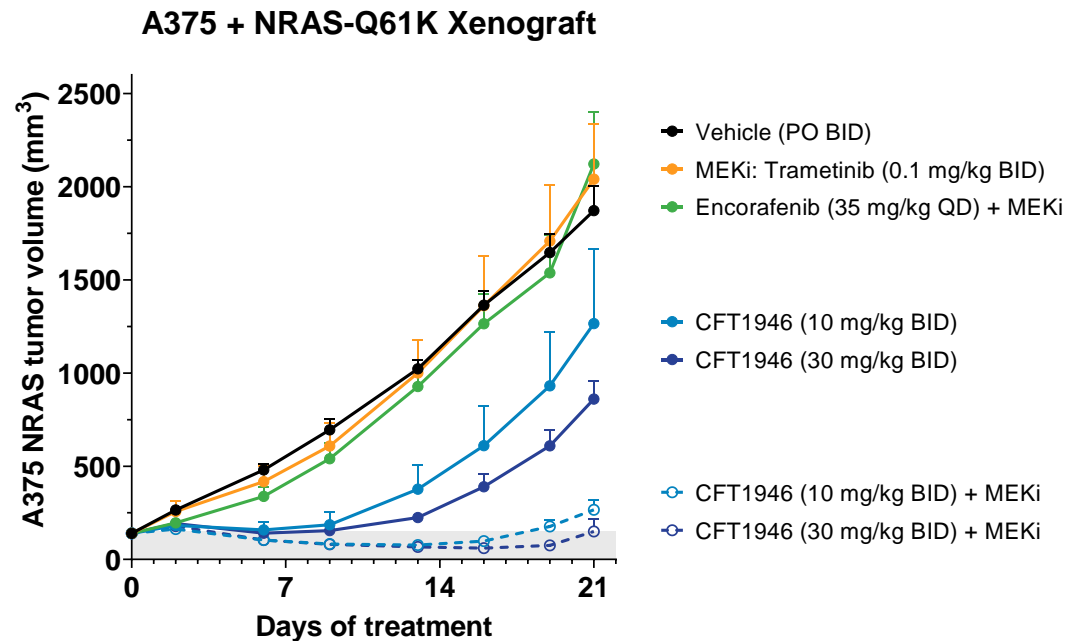
Dose Proportional PK and PD for CFT1946 Observed After Single Dose PO Treatment



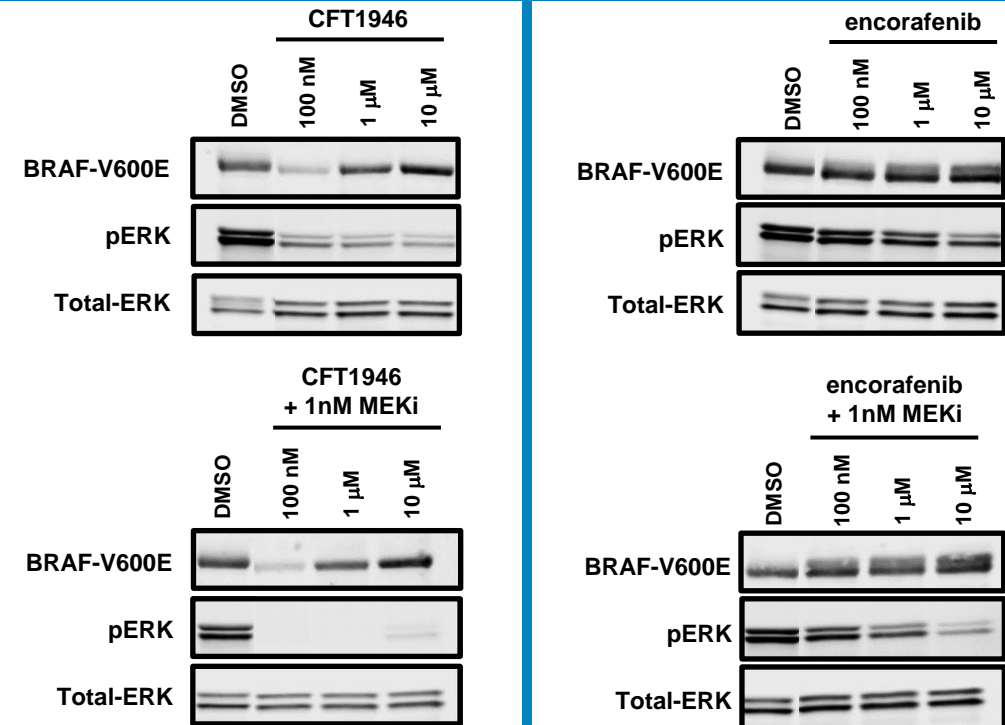
BID, twice a day; MAPK, MAP kinase; PO, by mouth; PK/PD, pharmacokinetics/pharmacodynamics; QD, once daily.
C4 Therapeutics data on file.

CFT1946 is Active in BRAF-V600E/NRAS-Q61K, a Model of Clinical Resistance to BRAF Inhibitors

Combination Treatment of BRAFi Resistant Xenograft Model with CFT1946 and MEKi Shows Tumor Growth Inhibition/Regression



CFT1946 as a Single Agent and in Combination with MEKi is Effective in MAPK Pathway Inhibition, Superior to BRAFi



BID, twice a day; BRAFi, BRAF inhibitor; MAPK, MAP kinase; MEKi, MEK inhibitor; PO, by mouth; PK/PD, pharmacokinetics/pharmacodynamics; QD, once daily. C4 Therapeutics data on file.



CFT1946 is a potent and mutant-selective BiDAC™ degrader of BRAF-V600E and superior to inhibitors in *in vitro* and *in vivo* models with BRAF-V600E–driven disease and in the escape mutant BRAF-V600E/NRAS-Q61K–driven model.



The medicinal chemistry path leading to CFT1946 demonstrates that it is possible to access catalytically efficient and orally bioavailable degraders through rational ligand and linker modifications.



Based on the preclinical profile, CFT1946 is currently being evaluated in a Phase 1 trial in patients with both BRAF-V600X–driven cancers and inhibitor-resistant BRAF-V600X–driven cancers.

Acknowledgments

Thank you to the C4T scientists & our CRO partners
across the globe who made this work possible