The Discovery and Characterization of CFT7455:
A Potent and Selective Degrader of IKZF1/3 for the Treatment of Relapsed/Refractory Multiple Myeloma


C4 Therapeutics, Inc
Watertown, MA USA
James A. Henderson, PhD

- I have the following financial relationships to disclose:
  - Stockholder in: C4 Therapeutics
  - Employee of: C4 Therapeutics
- I will not discuss off label use and/or investigational use in my presentation.
• IKZF1/3 are transcription factors required for cancer cell growth and survival in multiple myeloma (MM)
• IKZF1/3 degrading IMiDs are widely used in MM treatment (lenalidomide, pomalidomide)
• Relapsed/refractory MM remains a high unmet medical need

Goal: Develop an IKZF1/3 **Monofunctional Degradation Activating Compound** (MonoDAC) with these properties:

- Class-leading catalytic activity to enable potent, rapid, and deep target degradation
- High binding affinity to overcome IMiD resistance
- Selective to reduce off-target liabilities
- Pharmacologic profile that enables sustained IKZF1/3 degradation

CFT7455: Potent Small Molecule IKZF1/3 Degrader with Enhanced Catalytic & Pharmacologic Properties

CRBN, cereblon; CUL4, cullin 4; DDB1, DNA damage-binding protein 1; IKZF1/3, Ikaros family zinc finger proteins 1 and 3; IMiD, immunomodulatory imide drug; monoDAC, monofunctional degradation activating compound; MM, multiple myeloma; RBX1, ring box protein 1; Ub, ubiquitin.
IKZF1/3 Degrader Lead Derived from MonoDAC Library Hit

**Potent Hit from MonoDAC Library**

**Compound 1**

IKZF1 DC$_{50}$, $E_{\text{max}}$ @ 6 hr = 16 nM, 16% (HiBiT H929)

**Improvement Using SBDD**

**Compound 2**

6 hr = 1 nM, 13%

**PK & In Vivo Screening**

**Compound 3**

6 hr = 0.6 nM, 12%  
1.5 hr = 4.2 nM, 25%  
Goal >10x Increase

**Screening Efficacy in H929 Xenografts**

10 Compounds Dosed QD PO at 5-20X GI50 (free)

IKZF1/3, Ikaros family zinc finger proteins 1 and 3; HiBiT; high affinity bioluminescent tag; monoDAC, monofunctional degradation activating compound; PK, pharmacokinetics; SBDD, structure-based drug design.
Need for Speed: Structural Biology Highlights Areas for Chemistry Exploration

- The monoDAC degrader binds to CRBN and modulates the surface to accommodate an interaction with neosubstrate.

CRBN, cereblon; IKZF1/3, Ikaros family zinc finger proteins 1 and 3; monoDAC, monofunctional degradation activating compound; PDB: 6h0f, pomalidomide CRBN complex bound to IKZF1(ZF2).

The monoDAC degrader binds to CRBN and modulates the surface to accommodate an interaction with neosubstrate.

The second zinc finger of IKZF1 lands on top of the CRBN-monoDAC degrader complex.

The β-hairpin glycine interaction with the monoDAC is critical for IKZF1/3 degradation.

CRBN, cereblon; IKZF1/3, Ikaros family zinc finger proteins 1 and 3; monoDAC, monofunctional degradation activating compound; PDB: 6h0f, pomalidomide CRBN complex bound to IKZF1(ZF2).


 Compound 3
PDB: 6h0f
CRBN X-Ray Structures Inspire the Design of the Tricyclic Core

Benzoimidazolone
C4T unpublished
1.17 Å

Pomalidomide
PDB 6h0f

PDB: 4ci3
C4T unpublished

CRBN, cereblon; FP, fluorescence polarization; IKZF1, Ikaros family zinc finger protein 1.
CRBN X-Ray Structures Inspire the Design of the Tricyclic Core

Benzoimidazolone
C4T unpublished
1.17 Å

Pomalidomide
PDB 6h0f

Overlay with Compound 4

Compound 4
CRBN FP $K_D = 830 \text{ nM}$

Pomalidomide
$K_D = 1600 \text{ nM}$

CRBN, cereblon; FP, fluorescence polarization; IKZF1, Ikaros family zinc finger protein 1.
CRBN X-Ray Structures Inspire the Design of the Tricyclic Core

Benzoimidazolone
C4T unpublished
1.17 Å

Pomalidomide
PDB 6h0f

Benzoisoindolinone
C4T unpublished
1.06 Å

CRBN, cereblon; FP, fluorescence polarization; IKZF1, Ikaros family zinc finger protein 1.

Compound 4
CRBN FP $K_D = 830$ nM

Pomalidomide
$K_D = 1600$ nM

Compound 5
$K_D = 34$ nM

Overlay with Compound 4

50-fold affinity increase
Exploring CRBN Interactions with the Potent Tricyclic Core

<table>
<thead>
<tr>
<th>Tri-Trp Pocket Interactions</th>
<th>Increased Hydrophobic Contacts with CRBN</th>
<th>+δ Aromatic C-H Interactions with Backbone Carbonyl</th>
</tr>
</thead>
<tbody>
<tr>
<td>Asn</td>
<td>Pro</td>
<td>Glu</td>
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<tr>
<td></td>
<td>Trp</td>
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From First Generation Lead to CFT7455

<table>
<thead>
<tr>
<th></th>
<th>Compound 3</th>
<th>Compound 6</th>
<th>CFT7455</th>
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</thead>
<tbody>
<tr>
<td>CRBN IC&lt;sub&gt;50&lt;/sub&gt; (293T NanoBRET)</td>
<td>9 nM</td>
<td>0.3 nM</td>
<td>0.4 nM</td>
</tr>
<tr>
<td>IKZF1 DC&lt;sub&gt;50&lt;/sub&gt;, E&lt;sub&gt;max&lt;/sub&gt; (1.5 hr)</td>
<td>4.2 nM, 25%</td>
<td>0.3 nM, 22%</td>
<td>0.17 nM, 20%</td>
</tr>
<tr>
<td>H929 HiBiT</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>H929 IC&lt;sub&gt;50&lt;/sub&gt; (96 hr)</td>
<td>2.3 nM</td>
<td>0.009 nM</td>
<td>0.07 nM</td>
</tr>
<tr>
<td>PPB mouse/human (% bound)</td>
<td>94.3 / 96.2</td>
<td>97.2 / 98.6</td>
<td>93.4 / 94.6</td>
</tr>
<tr>
<td>Mouse Vd&lt;sub&gt;ss&lt;/sub&gt;, T1/2, %F</td>
<td>6.2 L/kg, 1.7 h, 9%</td>
<td>2.9 L/kg, 1.3 h, 23%</td>
<td>5.6 L/kg, 2.0 h, 48%</td>
</tr>
</tbody>
</table>

CRBN, cereblon; IKZF1/3, Ikaros family zinc finger proteins 1 and 3; monoDAC, monofunctional degradation activating compound; PPB, plasma protein binding

C4 Therapeutics data on file.
CFT7455: Potent, Rapid and Selective Degradation of IKZF1/3

CFT7455

IKZF1/3 Degradation, H929 (4 hr)

GPE in RPMI-8226, 10 nM (4 hr)

GPE, global proteomics experiment; IKZF1/3, Ikaros family zinc finger proteins 1 and 3.
C4 Therapeutics data on file.
High Catalytic Activity of CFT7455 Improves Anti-Cancer Activity in H929 MM Cells

**Binding Affinity (FP)**

- Pomalidomide
- CC-220
- CC-92480
- CFT7455

**Degradation Potency**

- Pomalidomide
- CC-220
- CC-92480
- CFT7455

**MM Cell Viability**

- Pomalidomide
- CFT7455
- CC-92480
- CC-220

Catalytic activity enhancement resulted in >1000-fold improvement in potency vs. Pom*

CFT7455 Demonstrates High Potency in MM Cell Lines and Xenografts

CFT7455 Has Broad Antiproliferative Activity

*Open symbol indicates highest tested concentration at which growth was not inhibited by more than 50%

MM, multiple myeloma, QD, once daily.
C4 Therapeutics data on file.

CFT7455 Promotes Durable Tumor Regression

Vehicle (QD, PO)
CFT7455 (30 µg/kg, QD)
CFT7455 (10 µg/kg, QD)
CFT7455 (3 µg/kg, QD)
Pomalidomide (3000 µg/kg, QD)
CC-92480 (1000 µg/kg, QD)
CC-92480 (300 µg/kg, QD)
CFT7455 Efficacy Attributed to Durable Tumor PK and IKZF3 PD in NCI-H929 MM Model

- **Linear, Durable Tumor PK**
  - CC-92480 is not detectable after 4 hours
  - CFT7455 displays linear and durable tumor PK translating into deep IKZF3 degradation and regression in MM xenograft models

- **48 hr Degradation Kinetics**
  - Percent of IKZF3 remaining/normalized to GAPDH in NCI-H929 Tumors

- **IKZF3 PD Associated with Efficacy**
  - NCI-H929 tumor volume (mm$^3$)
  - Vehicle (QD, PO)
  - Pomalidomide (3000 µg/kg, QD)

IKZF3, Ikaros family zinc finger protein 3; MM, multiple myeloma; PD, pharmacodynamics; PK, pharmacokinetics; QD, once daily.

C4 Therapeutics data on file.
CFT7455 is Efficacious in MM Models Resistant or Insensitive to IMiDs

Reduction in CRBN Expression with Chronic IMiD Dosing

CFT7455 Retains Activity in Len- & Pom-Resistant MM Cells

CFT7455 Promotes Regression in Tumors Insensitive to Pom

CRBN, cereblon; IKZF1/3, Ikaros family zinc finger proteins 1 and 3; IMiDs, immunomodulatory imide drug; Len, lenalidomide; PO, orally; Pom, pomalidomide; QD, once daily. C4 Therapeutics data on file.
Discovery efforts were aimed at identifying an IKZF1/3 degrader with class-leading activity.
Structure-based design and in vivo screening were employed to discover CFT7455.

In vitro data with CFT7455 demonstrated:
- High CRBN binding affinity ($K_D = 0.9 \text{ nM}$)
- Rapid, selective, and deep degradation of IKZF1/3 that is associated with apoptosis
- Broad, potent antiproliferative activity in a panel of MM cell lines

In vivo MM models treated with CFT7455 demonstrated:
- Regression in the treatment-naïve H929 MM tumor models at doses $\geq 10 \mu\text{g/kg/day}$
- Durable antitumor responses consistent with long-lived pharmacodynamic activity
- Single-agent efficacy in models unresponsive to approved IMiDs

A Phase 1/2 clinical trial to assess the safety and tolerability of CFT7455 in patients with R/R MM or non-Hodgkin’s lymphoma (NCT04756726) is ongoing and early clinical data is presented in poster #CT186.
Thank you to the C4T scientists & our CRO partners who made this work possible