CFT7455, a novel IKZF1/3 degrader, enhances the anti-myeloma activity of monoclonal and bispecific antibodies by augmenting immune responses

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Introduction

Immunomodulatory imine drugs (IIDs), such as pomalidomide (POM) and lenalidomide (LEN) are widely used in all treatment phases of multiple myeloma (MM)[1]. IIDs degrade the MM survival transcription factors Ikaros (IKZF1) and Aiolos (IKZF3)[2]. IKZF1 and IKZF3 are essential for differentiation of B and T lymphocytes and viability of tumor cells. We previously described CFT7455, an orally bioavailable MonoDAC™ degrader designed to be highly potent and selective against IKZF1 and IKZF3. CFT7455 greatly increases target degradation potency and catalytic activity compared to approved IIDs. This translates into improved anti-tumor responses in preclinical models of IID-sensitive, and IID-resistant MM and non-Hodgkin lymphomas (NHL)[3-4].

Beyond direct anti-myeloma activity, IIDs also induce proinflammatory immune responses. Numerous preclinical studies have demonstrated that IIDs affect T-cells by promoting activation and proliferation, inducing proinflammatory cytokines, such as IL2 and IFNγ, and enhancing CD8+ T-cell effector activity[5-6]. Similar IID-mediated immunomodulatory effects have also been observed on NK cells[5-6]. Initial clinical data from our current CFT7455 Phase 1 dose escalation study (NCT04756726) in MM and NHL patients show CFT7455 is well tolerated, demonstrates anti-myeloma activity, and displays evidence of immunomodulatory effects.

Antibody-based therapies, such as monoclonal antibodies (mAbs) like daratumumab (targeting anti-CD38) and bispecific T-cell engagers (BiTEs) like teclistamab (targeting BCMA), have dramatically changed the MM therapeutic landscape[7]. These therapies lead to enhanced interactions between MM cells and immune effector cells such as T and NK cells. IKZF1/3 degraders may be particularly beneficial when used in combination with antibody therapies because they have the potential to amplify the anti-MM immune responses that they generate. In this study, we investigated the effects of CFT7455 on immune cell activation and observed that clinically relevant concentrations of CFT7455 enhanced the anti-MM activity of daratumumab and teclistamab in *in vitro* models of antibody dependent cellular cytotoxicity (ADCC) and T-cell dependent cellular cytotoxicity (TDCC), respectively. In addition, we found that activation of T-cells by CFT7455 is consistent with clinical data generated in our Phase 1 dose escalation study. Taken together, these data provide a compelling rationale for further investigation of CFT7455 in combination with antibody-based therapies for the treatment of MM.

Rationale for combination of CFT7455 with antibody-based therapies

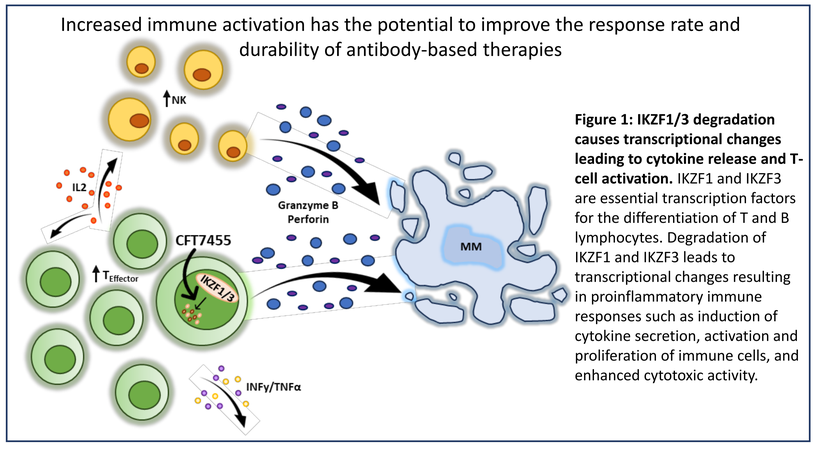

Figure 1: IKZF1/3 degradation causes transcriptional changes leading to cytokine release and T-cell activation. IKZF1 and IKZF3 are essential transcription factors for the differentiation of T and B lymphocytes. Degradation of IKZF1 and IKZF3 leads to transcriptional changes resulting in proinflammatory immune responses such as induction of cytokine secretion, activation and proliferation of immune cells, and enhanced cytotoxic activity.

References

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CFT7455 results in deep degradation of Ikaros (IKZF1) and Aiolos (IKZF3) in purified CD3+ T-cells

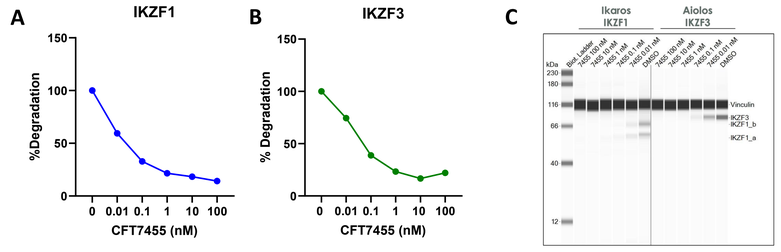

Figure 2: *In vitro* degradation of IKZF1 (A) and IKZF3 (B) by CFT7455 in purified CD3+ T-cells. Pan T cells were purified from a leukopak using the EasySep™ Human T Cell Isolation Kit and treated with CFT7455 for 48 hours. Graphs represent quantitation of protein levels from whole cell lysates and were analyzed by JESS Simple Western™ system using Ikaros and Aiolos specific antibodies (C).

CFT7455 activates T-cell proliferation at clinically relevant concentrations

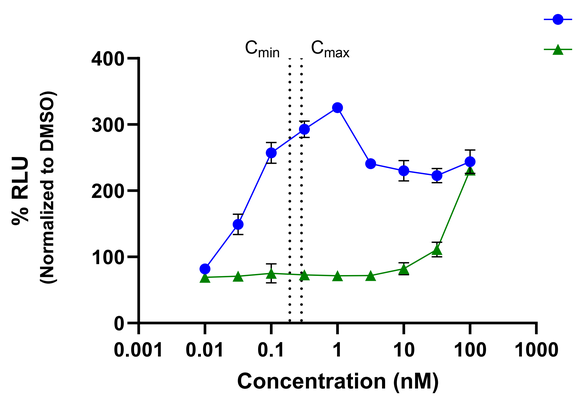

Figure 3: CT7455 demonstrates immunostimulatory effects on purified CD3+ T-cells. Purified CD3+ T-cells were treated with CFT7455 or POM (half log-fold serial dilutions) and proliferative activity was measured by Cell-TiterGlo® after 6 days of treatment. Dashed lines indicate C_{min}/C_{max} for expected human exposure at 50 micrograms CFT7455 daily.

CFT7455 induces cytokine secretion and release of cytotoxic effector proteins

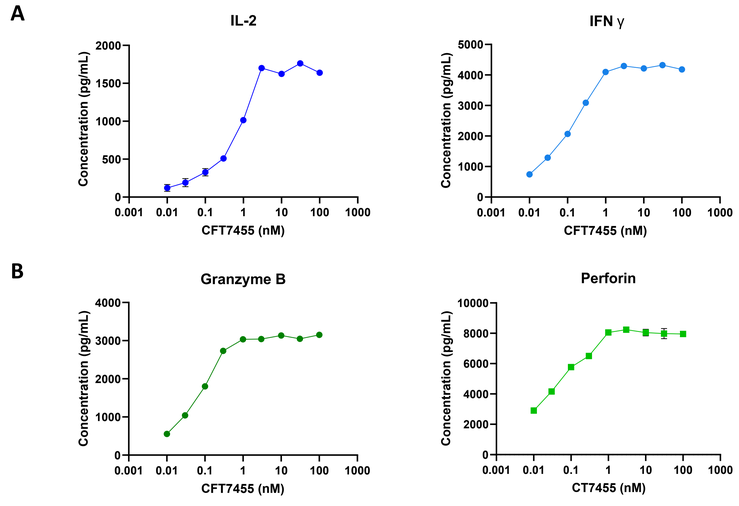

Figure 4: CT7455 stimulates secretion of proinflammatory cytokines and cytotoxic effector proteins in a dose-dependent manner. Supernatants from purified CD3+ T-cells treated with CFT7455 for 6 days were evaluated for secretion of cytokines (A) and release of cytotoxic effector proteins (B) using Meso Scale Discovery (MSD) platform.

CFT7455 enhances the activity of daratumumab in antibody dependent cellular cytotoxicity (ADCC) assays

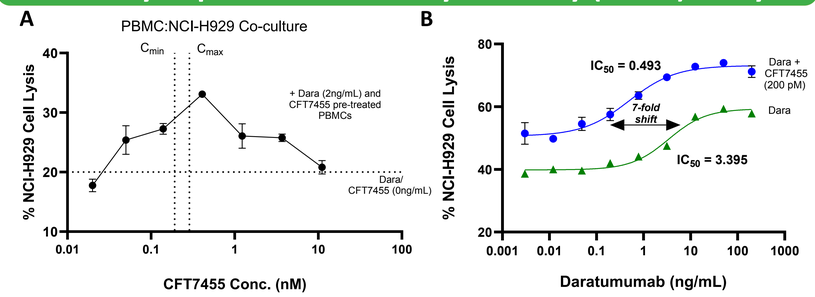

Figure 5: Pretreatment of PBMCs with CFT7455 leads to enhanced NCI-H929 myeloma cell lysis induced by daratumumab (Dara). PBMCs (effector cells) were pre-treated with CFT7455 for 6 days prior to ADCC assay. Following pretreatment, PBMCs and MM target cells were co-cultured (E:T ratio, 10:1) for 6 hours in the presence of Dara (2 ng/mL) and varying concentrations of CFT7455. Cell lysis was measured by flow cytometry. Optimal assay conditions for E:T ratio and antibody concentration were determined in a prior pilot study (A). The effective dose of Dara in ADCC assays is reduced by the presence of CFT7455 (200 pM) (B). Dashed lines indicate C_{min}/C_{max} for expected human exposure at 50 micrograms CFT7455 daily. Abbreviation: PBMCs –Peripheral blood mononuclear cells.

CFT7455 enhances the activity of teclistamab in T-cell dependent cellular cytotoxicity (TDCC) assay

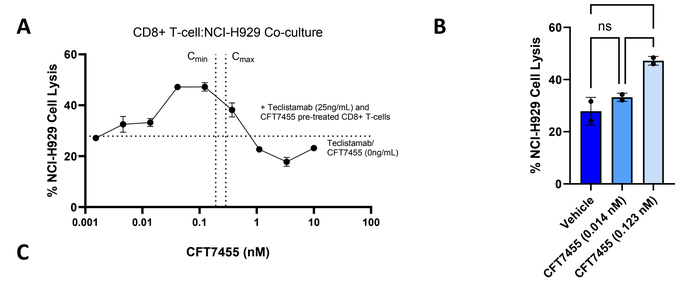

Figure 6: CFT7455 synergizes with teclistamab to augment NCI-H929 MM tumor cell lysis in TDCC assays. CD8+ T-cells were isolated from PBMCs pretreated with CFT7455 for 6 days. Following pretreatment, CD8+ (effector) cells were co-cultured with MM target cells (E:T, 2:1) in the presence of teclistamab (25 ng/mL) and varying concentrations of CFT7455 for 24 hours. Cell lysis was measured by flow cytometry. Optimal assay conditions for E:T ratio and antibody concentration were determined in a prior pilot study (A). Statistical analysis of the % cell lysis at CFT7455 concentrations representing C_{min}/C_{max} (B). The increase in CFT7455-dependent TDCC activity is more pronounced at day 6 versus day 3 at a representative dose of 0.123 nM (C). Dashed lines indicate C_{min}/C_{max} for expected human exposure at 50 micrograms CFT7455 daily. Abbreviation: PBMCs –Peripheral blood mononuclear cells.

Summary

CFT7455, a MonoDAC™ degrader, designed to achieve potent target degradation and catalytic activity against its targets, IZKF1 and IKZF3, is currently in Phase 1 dose escalation studies in MM and NHL. Initial clinical data demonstrates CFT7455 exhibits anti-myeloma activity and displays evidence of immunomodulatory effects consistent with preclinical *in vitro* data reported here.

- **Clinically relevant concentrations of CFT7455 demonstrate immunomodulatory effects in an *in vitro* setting including activation of T-cells, secretion of proinflammatory cytokines, and release of cytotoxic effector proteins from purified CD3+ T cells.**
- **The enhanced activity of CFT7455-treated effector cells leads to improved anti-MM responses to daratumumab (anti-CD-38 mAb) and teclistamab (BCMA BiTE) in ADCC and TDCC assays, respectively.**
- **The ability of CFT7455 to increase T-cell activation provides a strong rationale for combination with mAbs and BiTEs for the treatment of MM.**

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