(PF427) ACU SMALL MOLECULE STING AGONISTS DEMONSTRATE TP53-INDEPENDENT ANTI-LEUKAEMIC ACTIVITY AND VENETOCLAX SYNERGY IN ACUTE MYELOID LEUKAEMIA

Topic: 03. Acute myeloid leukemia - Biology & translational research

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Background

Patients with acute myeloid leukaemia (AML) face a poor prognosis, particularly those with high-risk disease such as *TP53* mutations. Despite recent therapeutic advances, sustaining long-term remission remains a major challenge that must be met. Drug activators of the "stimulator of interferon genes" (STING) pathway have been explored for their potential as an immunotherapy in the solid tumour setting. We previously reported a novel cell-intrinsic mechanism of action of the STING agonist diABZi in AML (Cancer Cell 2024). STING activation induced apoptosis potently in AML cells, with diABZI demonstrating combinatorial activity with the pro-apoptotic BCL-2 inhibitor venetoclax (VEN) in AMLs with and without *TP53* mutation. Here we report the preclinical activity of a new series of systemically deliverable, synthetic non-cyclic dinucleotide small molecule STING agonists in AML.

Aims

To investigate the preclinical activity of ACU STING agonists (ACU2086 and ACU0943) against AML.

Methods

ACU STING agonists were provided by Aculeus Therapeutics. *In vitro* assays were performed using ACU2086 and *in vivo* experiments with the clinical grade compound ACU0943. VEN was purchased from Chemgood. Drug sensitivity assays were performed on human AML cell lines and primary patient samples. Cell death was assessed by PI/DAPI staining and flow cytometry. Cell proliferation was assessed by CellTitreGlow (CTG). For *in vivo* testing, MOLM-13 cells were transplanted into NOD-*scid*IL2Rgamma^{null} (NSG) mice. Mice received no treatment (control), VEN (50mg/kg orally 5 days/week), ACU0943 (1.5mg/kg IV biweekly), or combination VEN/ACU0943 for 2 weeks (n=5 per group). Leukaemic burden (%human CD45+ cells) in peripheral blood (PB), bone marrow (BM) and spleen was measured at end of therapy. Groups were compared using Welch's ANOVA with Dunnett's multiple comparison test.

Results

In vitro, ACU2086 monotherapy demonstrated activity in 6/7 AML cell lines tested (including 3/4 with *TP53* mutations). Using CRISPR-modified cell lines, cell death induced by ACU2086 was confirmed to be via STING dependent apoptosis, with killing blocked by knockout of either *STING* or *BAX* and *BAK*. In contrast, cell death was independent of TP53, with isogenic *TP53* knockout (KO) cells demonstrating equivalent sensitivity to wildtype (WT) cells. Although *BAX/BAK* KO cells were resistant to STING-induced cell death, CTG assays demonstrated their growth was inhibited by ACU2086. Growth arrest was also seen in *TP53/BAX/BAK* KO cells, suggesting the anti-proliferative effect of ACU2086 is also TP53 independent. Combination ACU2086 and VEN was additive to synergistic in MOLM-13 and OCI-AML3 cells (Bliss Synergy Scores 11.1 and 8.9, respectively). Across 14 primary samples tested, the average IC_{50} for VEN was >10 µM, 3.3µM (95% CI 2.2-5.1µM) for ACU2086 and 0.33µM (95% CI 0.26-0.41µM) for VEN/ACU2086 combination.
In vivo, mice treated with ACU0943 had approximately 10-fold lower blast burden in PB, BM and spleen compared with control and VEN treated mice (Figure 1). Combination VEN/ACU0943 was most effective, with further log-fold reduction in blast burden compared with ACU0943 alone, providing clear evidence of combinatorial activity.

Summary/Conclusion

ACU STING agonists exert potent cell-intrinsic, *TP53*-independent activity against AML, with efficacy enhanced by combination with VEN. These data highlight the potential of ACU STING agonists as a novel treatment for AML and provide preclinical evidence supporting a planned phase 1 clinical trial.

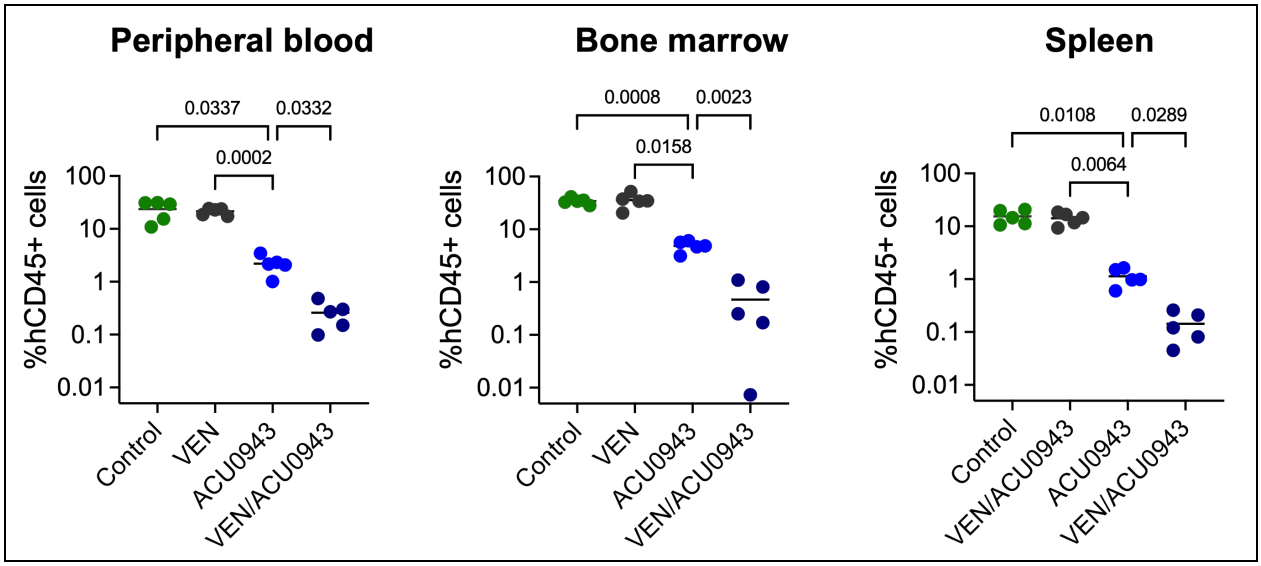

Figure 1