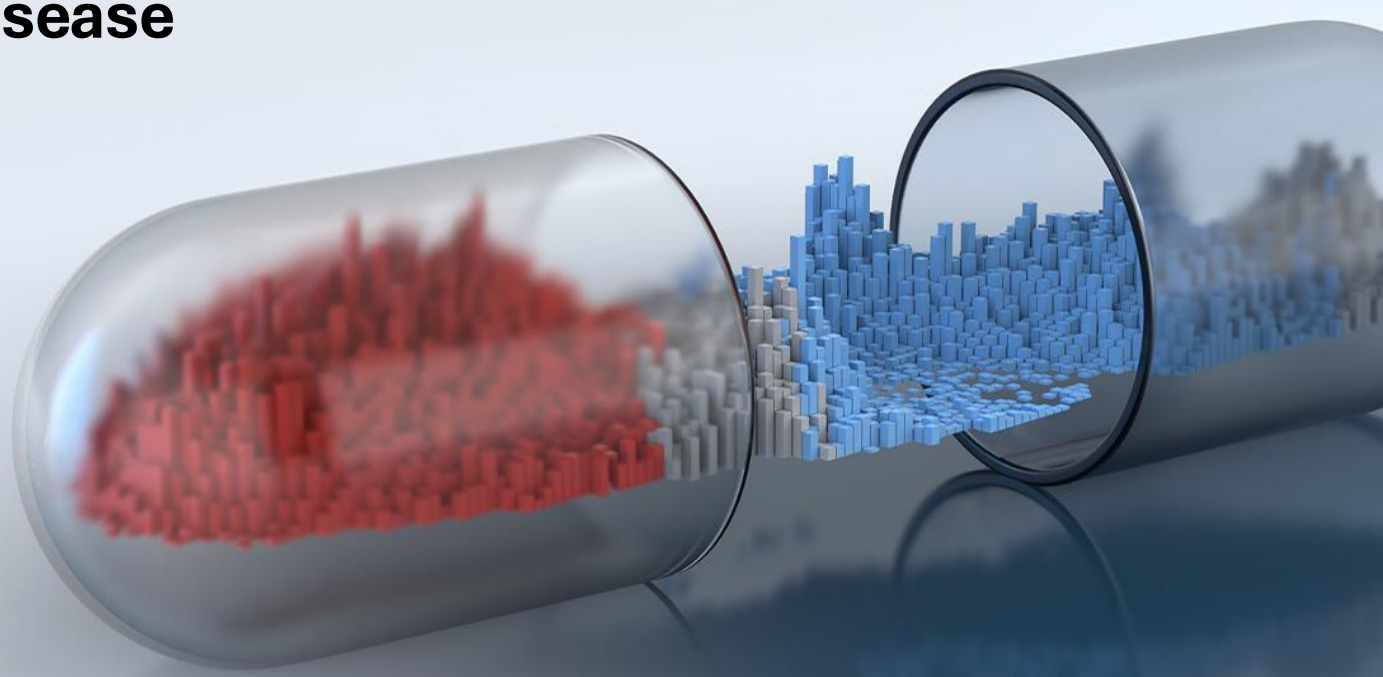
CLY-124 - a first-in-class DCN1 inhibitor that partially suppresses CUL3 neddylation and induces fetal hemoglobin is a new potential treatment for sickle cell disease



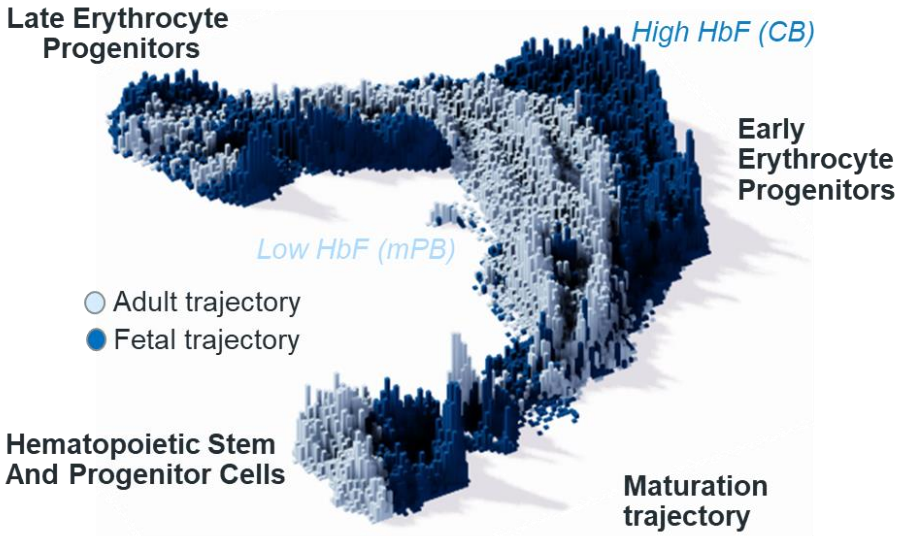
Cameron Trenor, MD

67th ASH Annual Meeting - Orlando

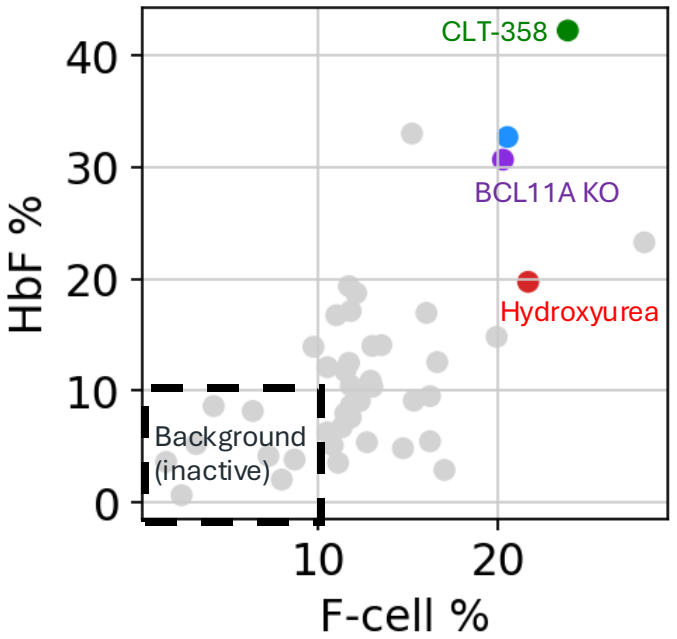
December 8, 2025

Transcriptomic analysis of fetal and adult erythropoiesis uncovered non-cytotoxic pathways for high HbF induction

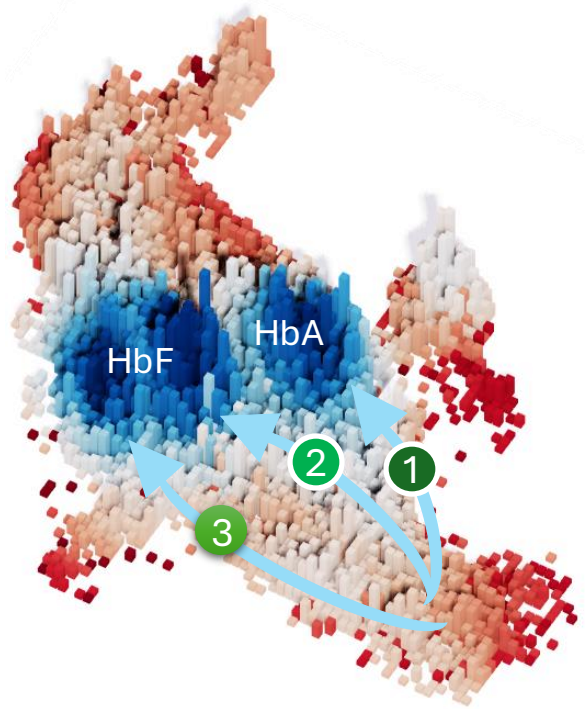
Systematic Transcriptomic analysis of Fetal and Adult Erythropoiesis



Validation of prediction by fetal hemoglobin induction in HSC derived erythroid cells



Predicted compounds induce HbF distinct from hydroxyurea

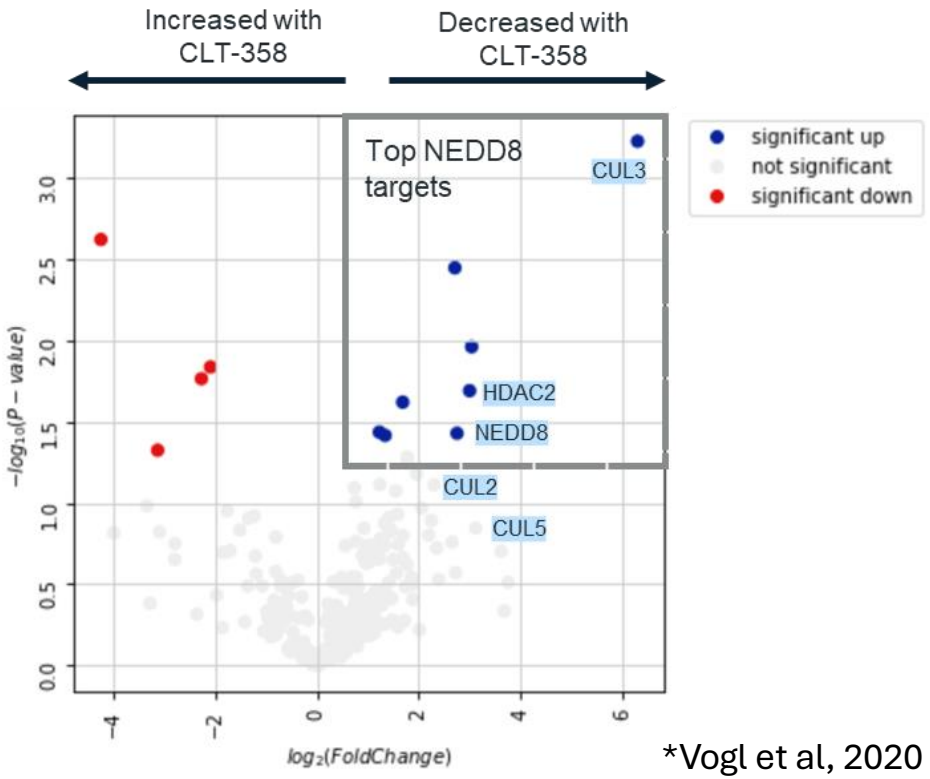


1 *Untreated*
2 *CLT-358* & BCL11A KO*
3 *Hydroxyurea*

*CLT-358 = MLN4924

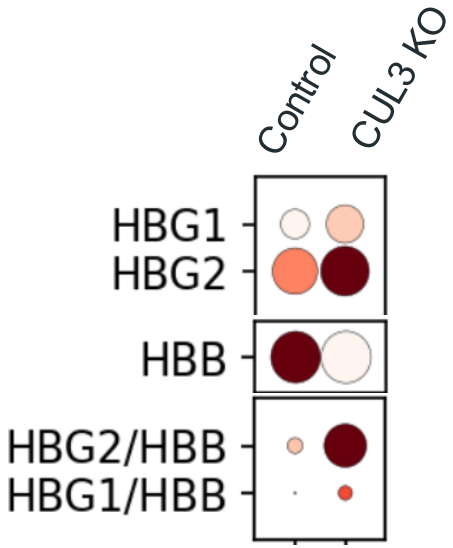
CLT-358 blocks CUL3 NEDDylation leading to globin switching

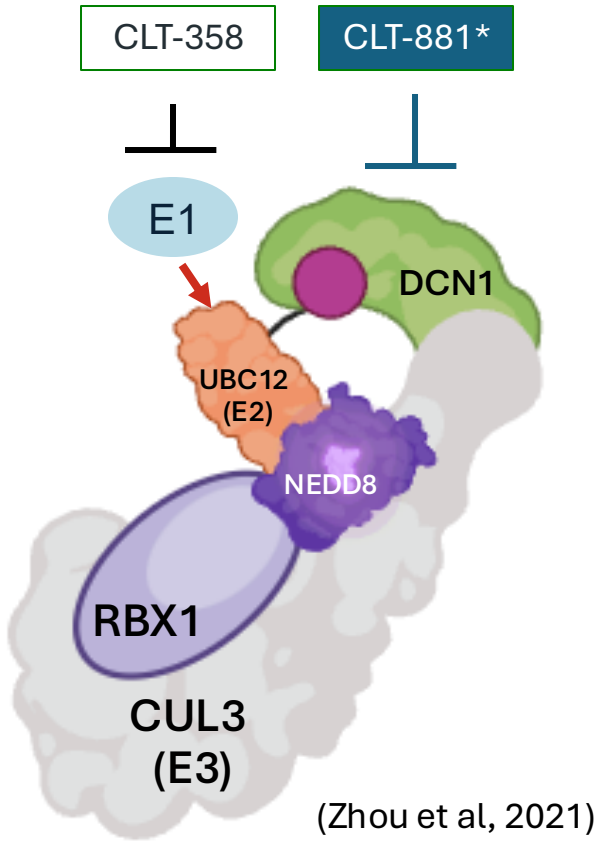
Differentially Neddylated proteins using proteomics analysis*



*Vogl et al, 2020

CUL3 CRISPR KO induces high HBG and HBG/HBB Ratio





(Zhou et al, 2021)

- CUL3 KO CD34 cells demonstrated cytotoxicity and skewing to non-erythroid lineages
- Homozygous CUL3 KO mice are embryonic lethal and conditional KO mice have CNS and CV defects
- No CUL3 inhibitors in clinical development

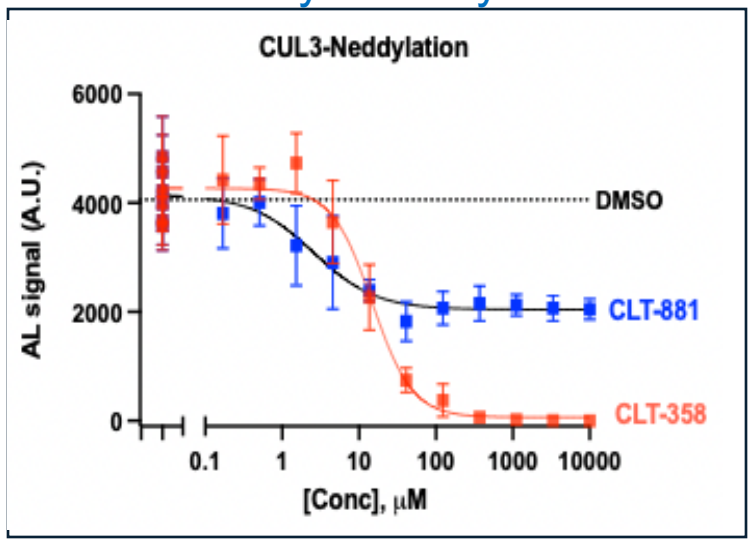
*CLT-881= specific DCN1 inhibitor tool compound

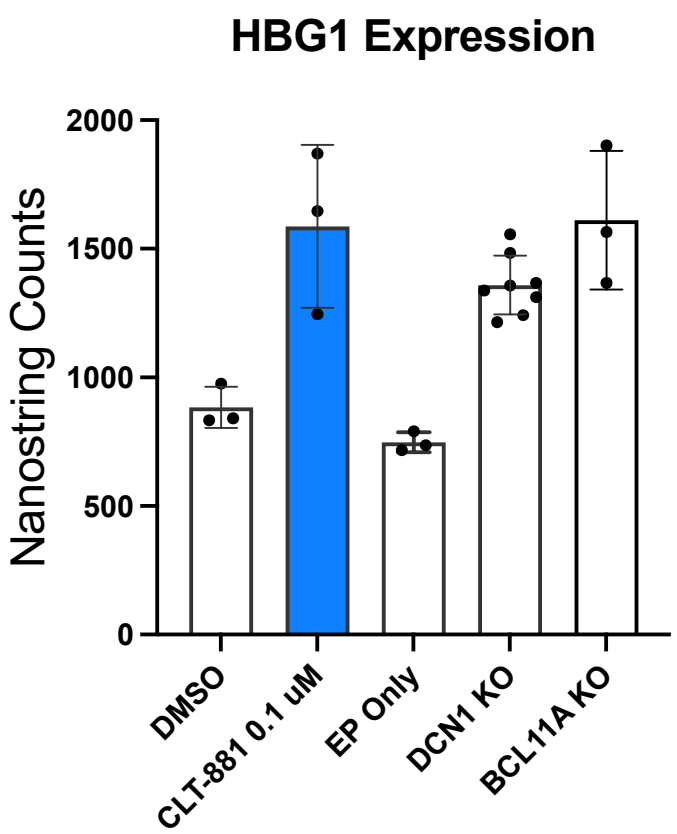
DCN1 inhibition partially suppresses CUL3 NEDDylation and induces fetal hemoglobin

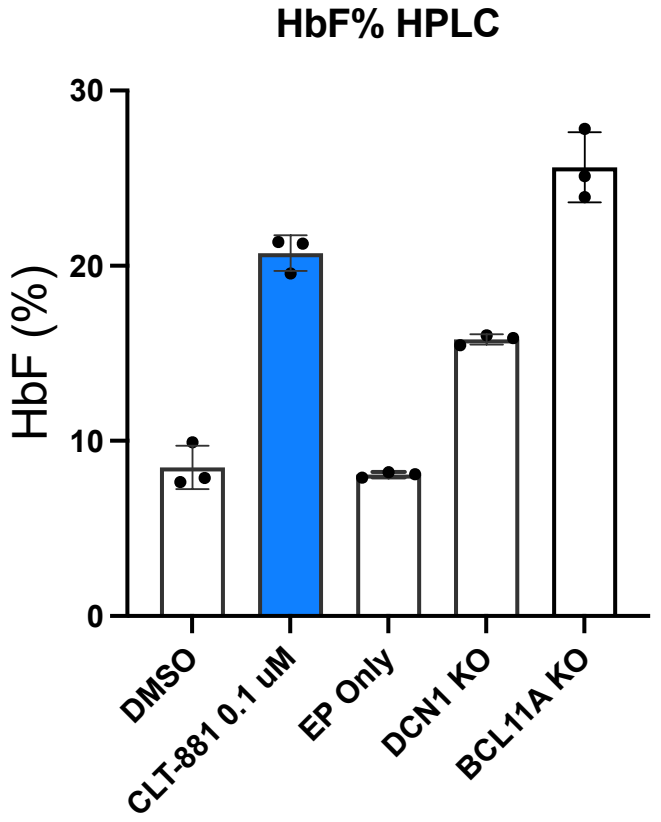
DCN1 inhibition partially suppresses CUL3 NEDDylation

CLT-881 and DCN1 KO Upregulates HBG1 Expression and induces HbF in hCD34 erythroblasts

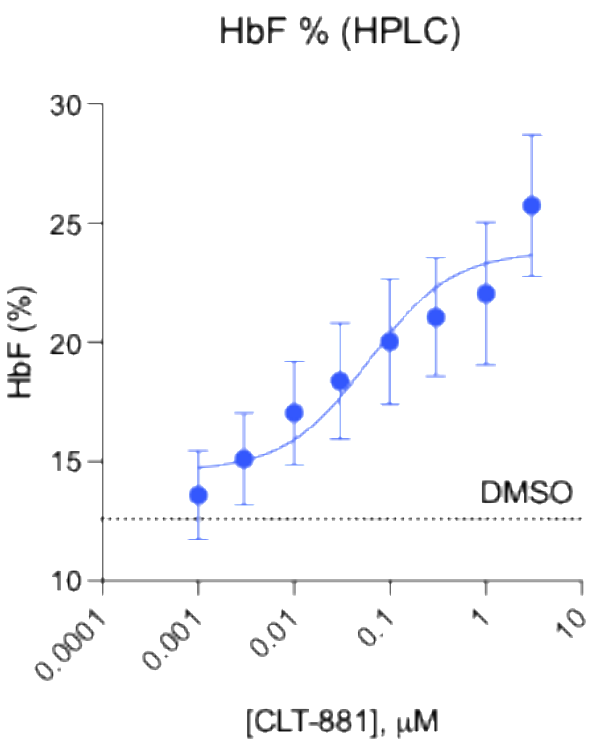
AlphaLISA shows **~60% reduction** in Cul3 NEDDylation by CLT-881

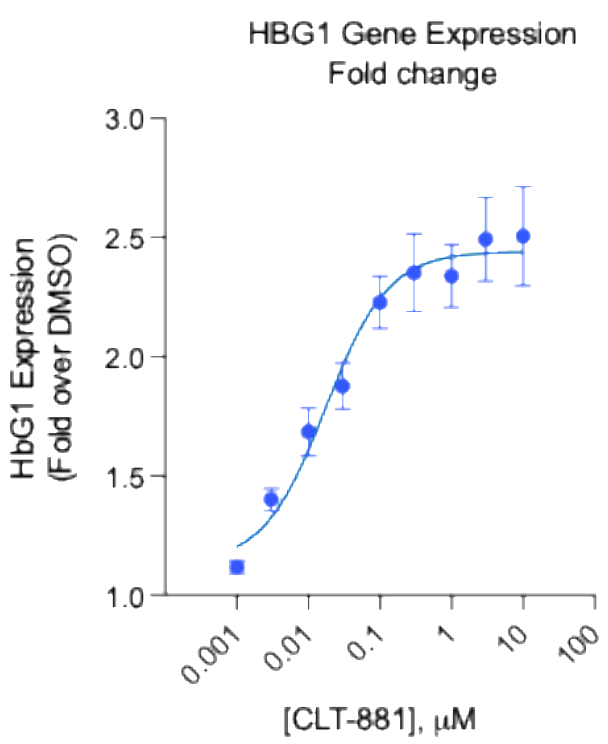


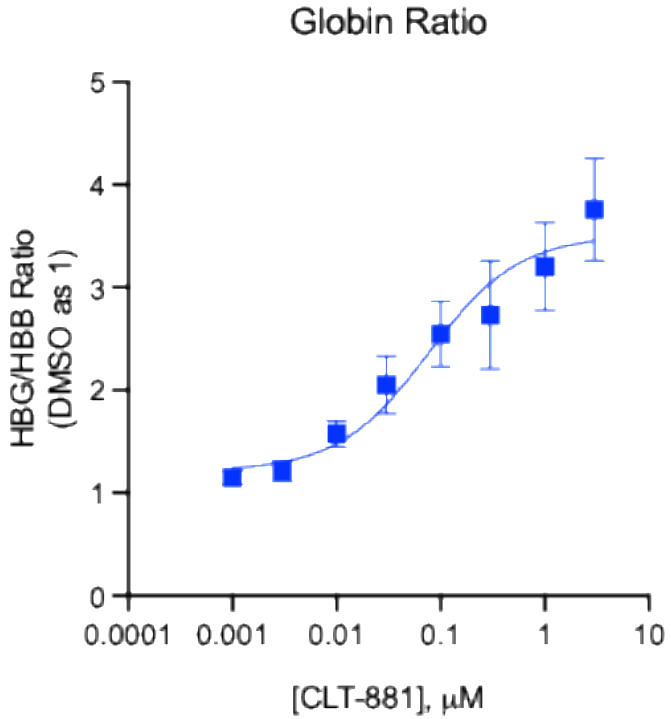




Dose-responsive HbF induction via DCN1 inhibition in vitro through globin switching







Up to 25% HbF

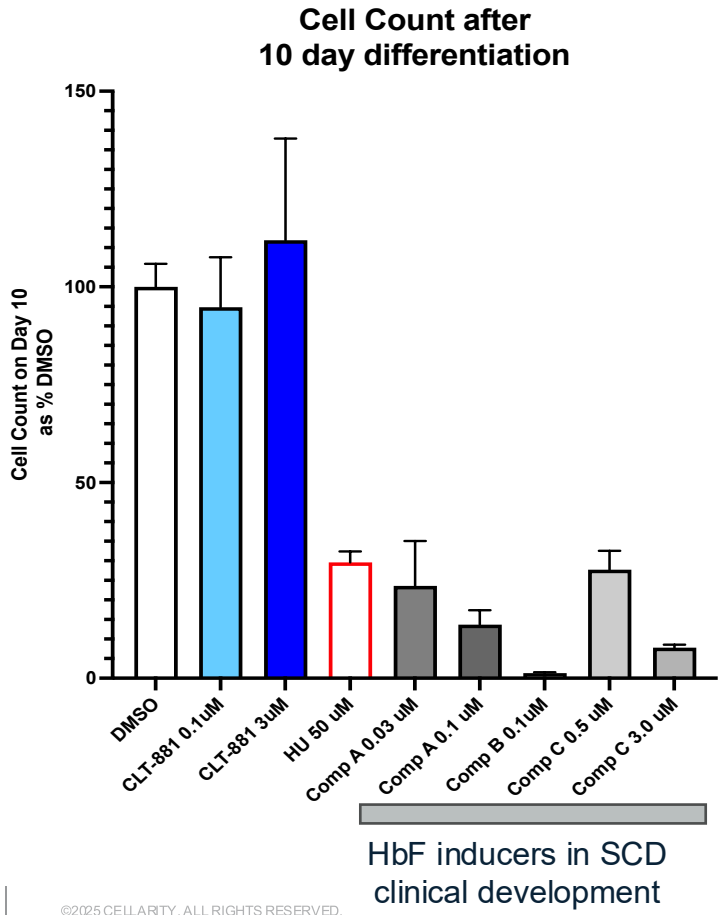
2.5-fold HBG1/2 increased expressions (EC_{50} = 17.5nM)

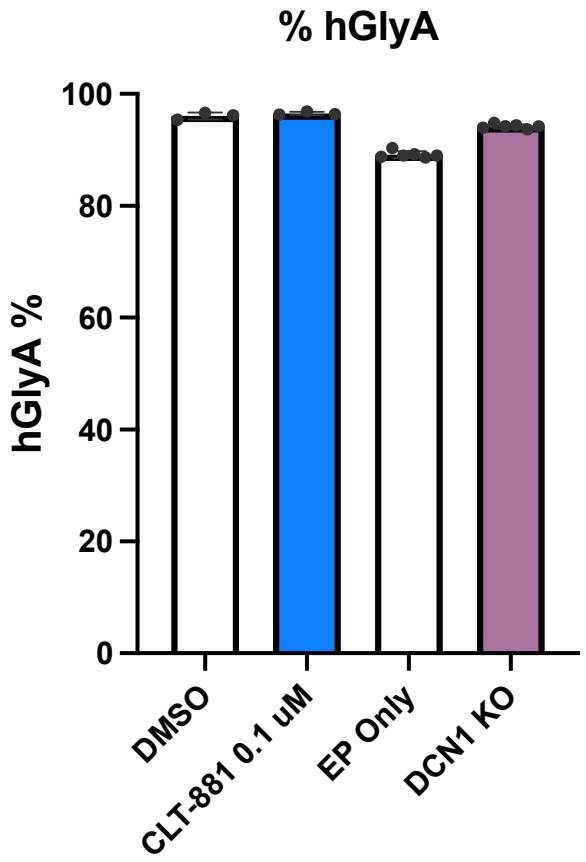
3-fold increase in globin gene ratio

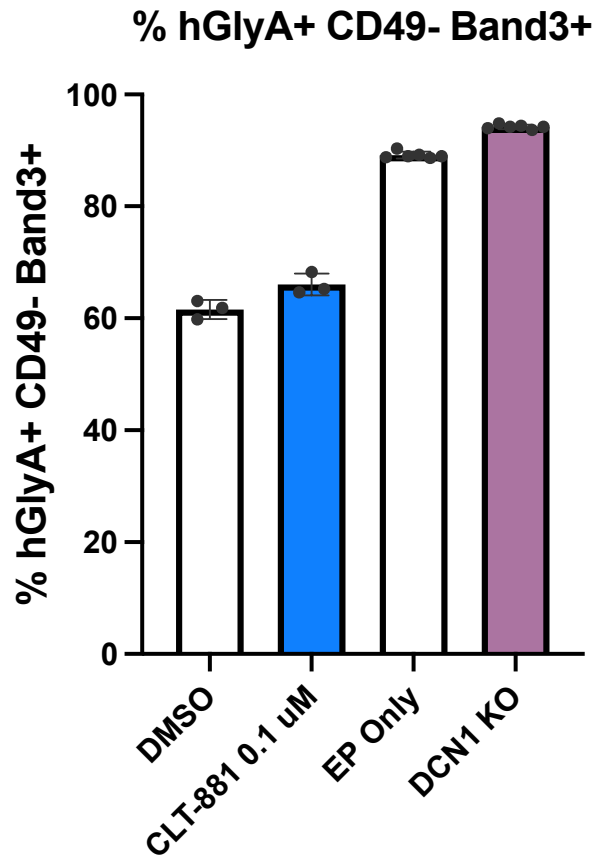
DCN1 inhibition is not cytotoxic and does not disrupt erythropoiesis

CLT-881 does not affect cell viability unlike HU and other HBF inducers

CLT-881 and DCN1 KO do not impair erythroid differentiation and maturation





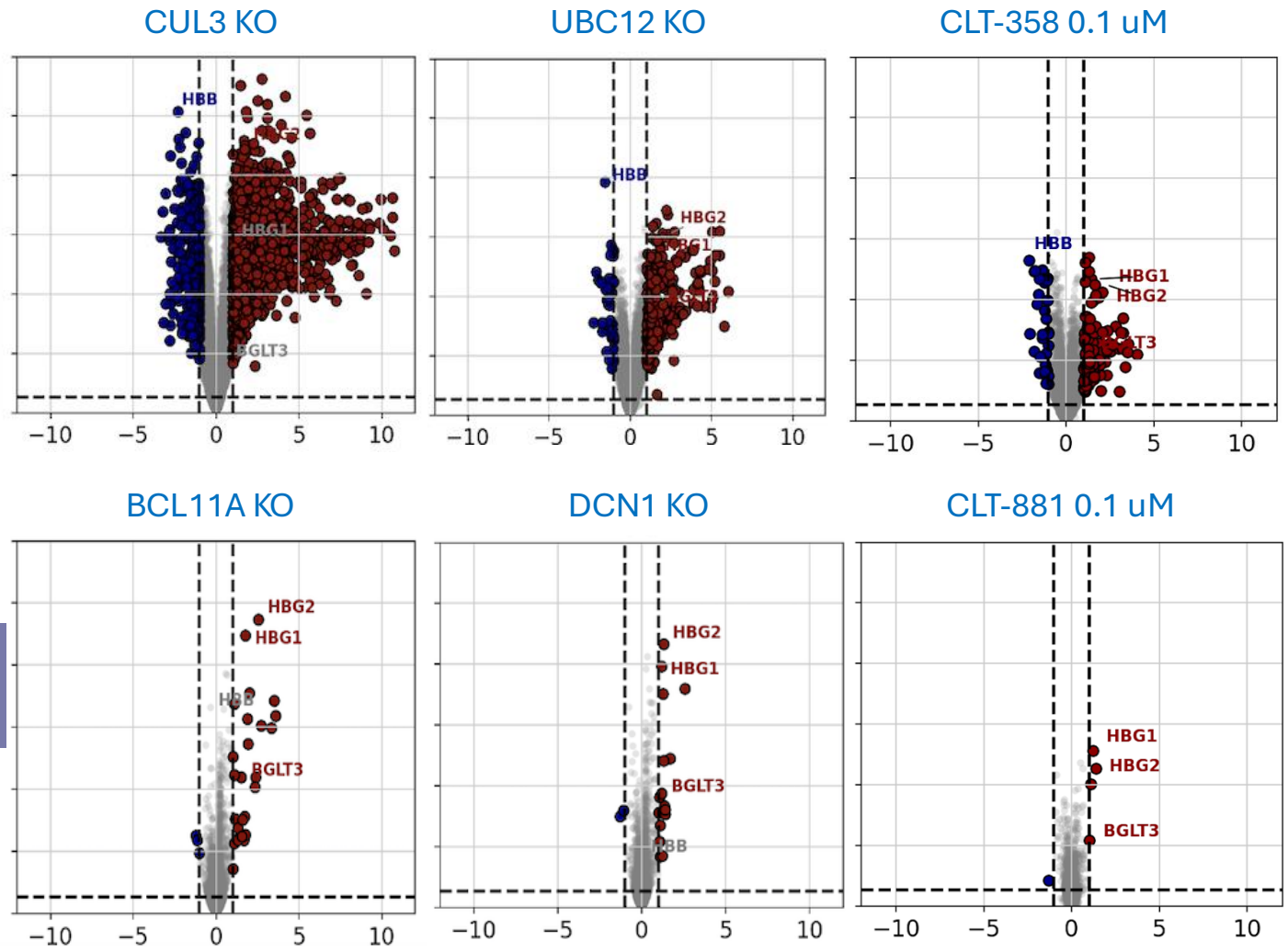


Cellarity

Transcriptomic selectivity of globin switching evident in DCN1 knockout and DCN1 inhibition

Pan-Neddylation Inhibition

Globin switching mechanisms



- Pan-Neddylation inhibition regulates 100s – 1000s of gene transcripts
- Like the enhancer editing of BCL11A, DCN1 inhibition has a relatively narrow transcriptomic profile

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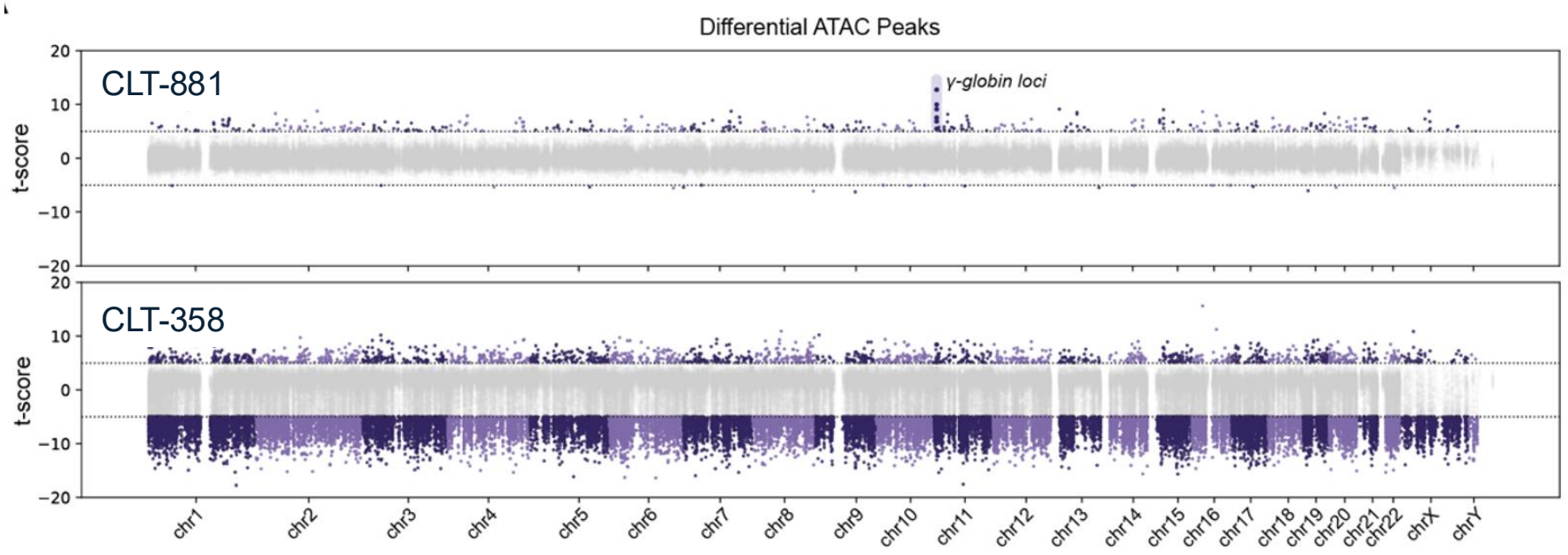
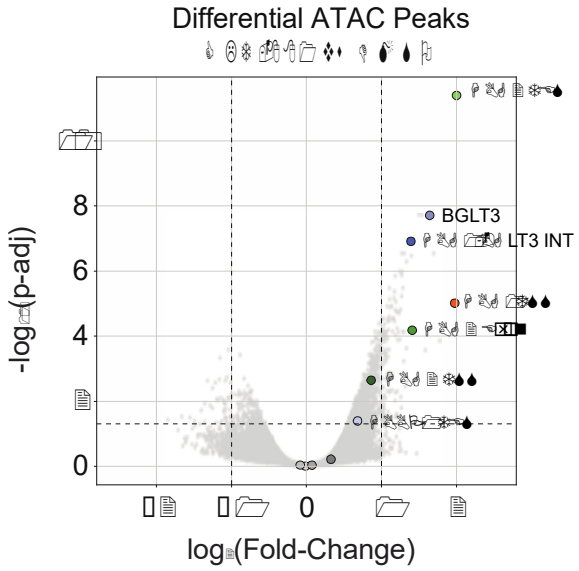
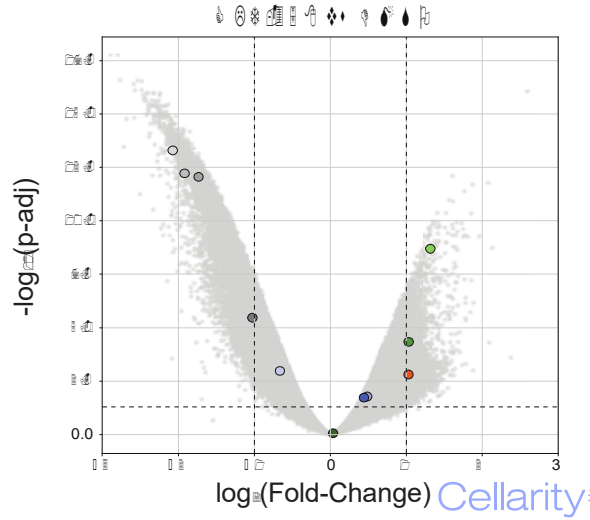
Genome-wide ATAC-seq after DCN1 inhibition reveals specific chromatin accessibility at the g-globin locus and avoids global chromatin accessibility







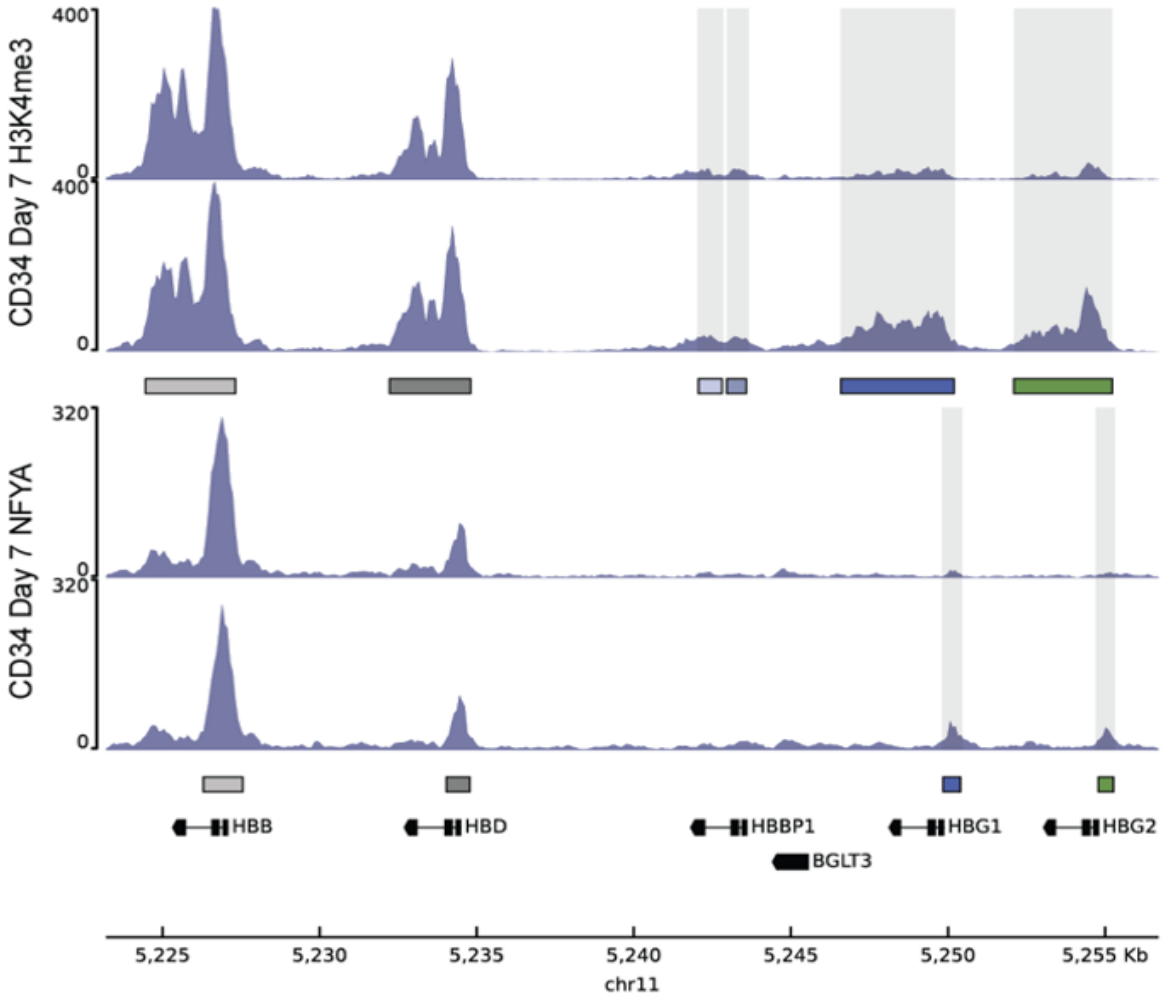
DCN1 inhibition increases chromatin accessibility specifically at the HBG1/2 loci

In addition, Micro Capture-C demonstrates gain of chromatin interactions between the LCR and BGLT3/HBBP1 region consistent with globin switching

Global genome profiling reveals selective enrichment of H3K4me3 and NFYA binding at the HBG1/2 genes

H3K4me3 Methylation

NFYA Occupancy

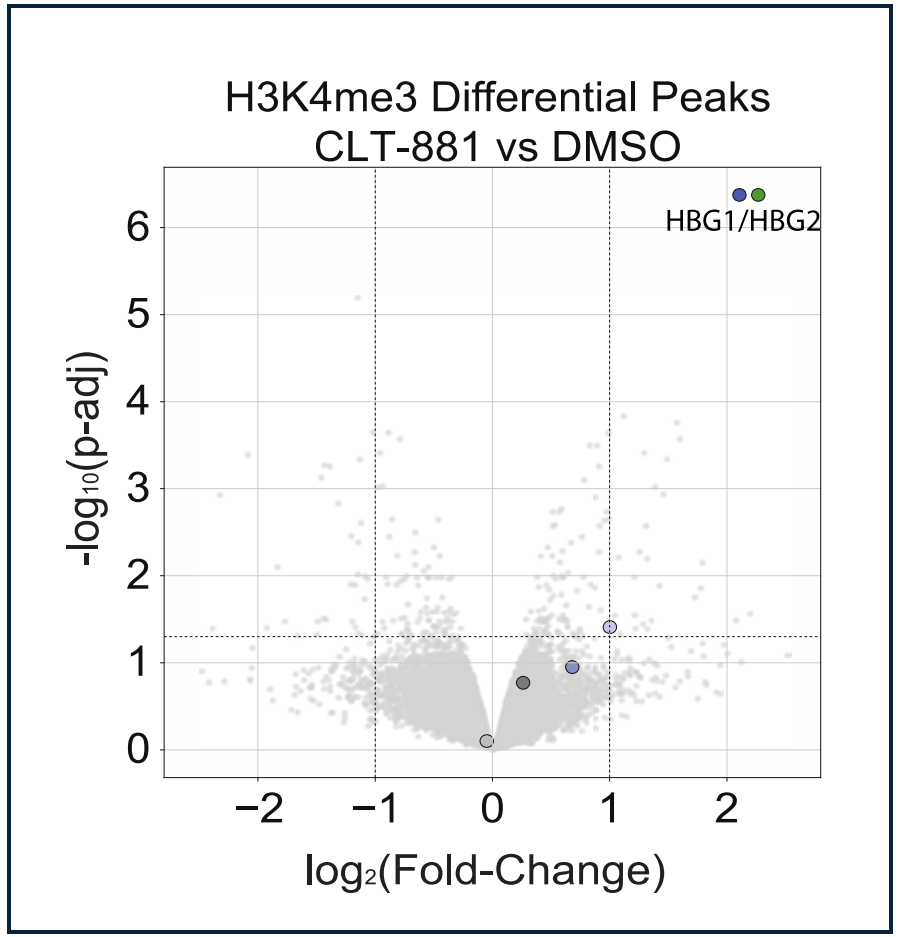


DMSO

CLT-881

DMSO

CLT-881



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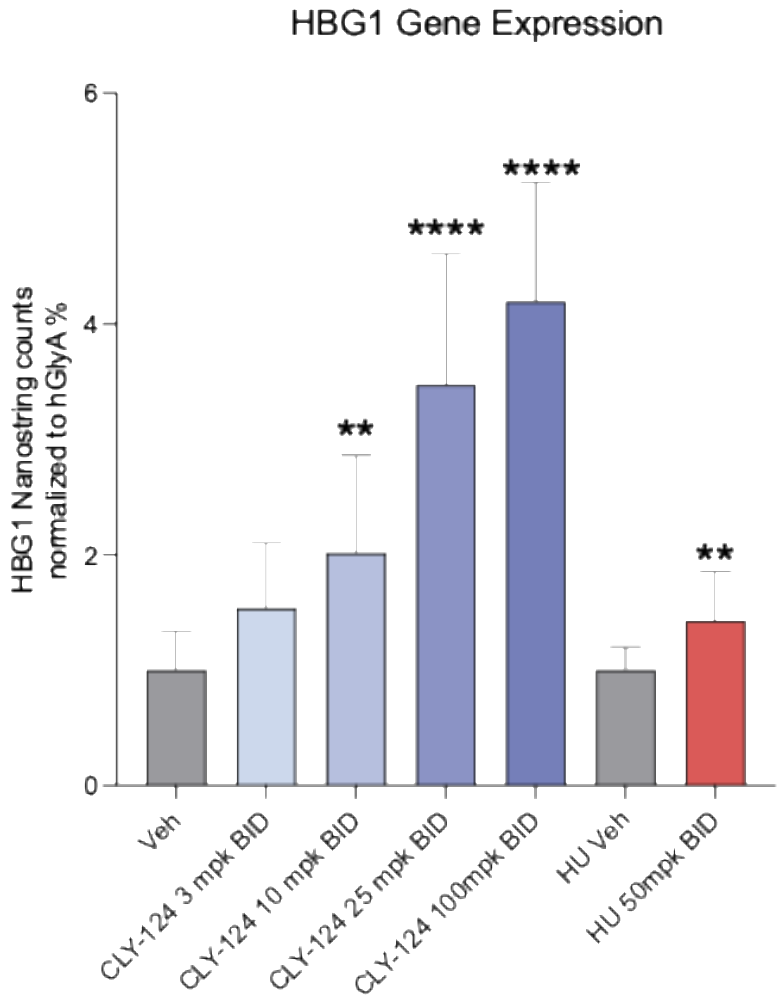
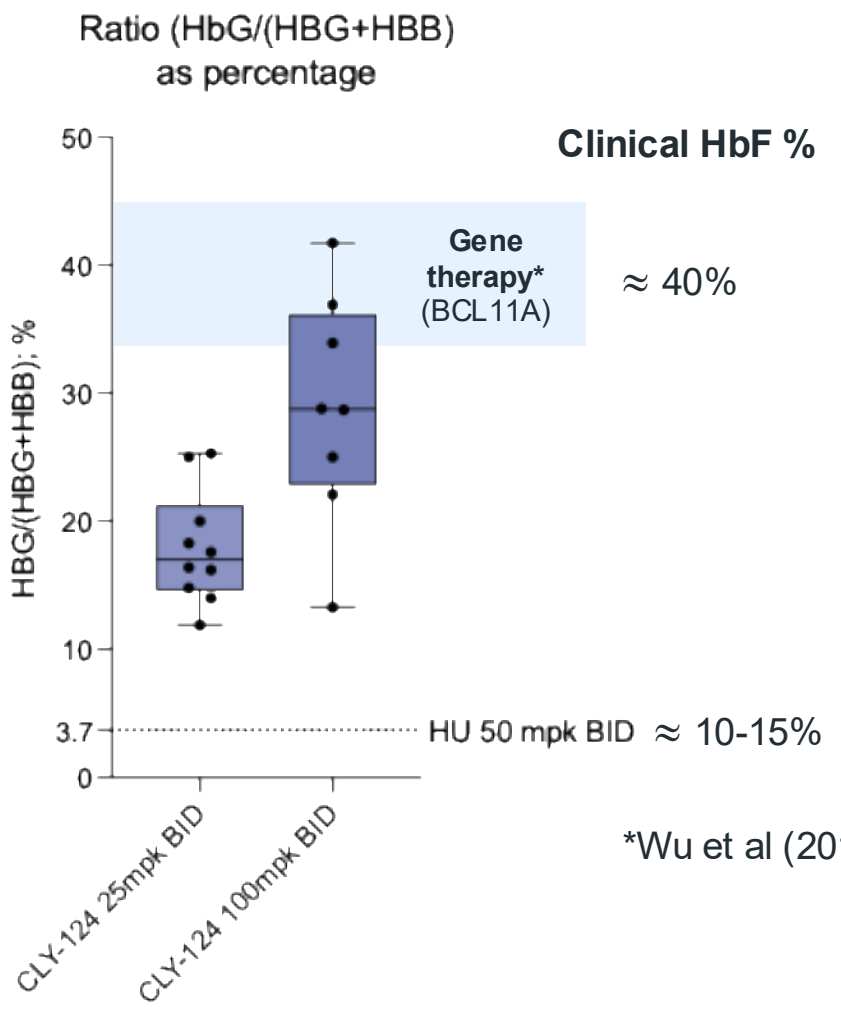
CLY-124 in vivo Efficacy in Humanized NBSGW mice



Dose-responsive increase in HGB1
Superiority to HU



Clinical HbF %

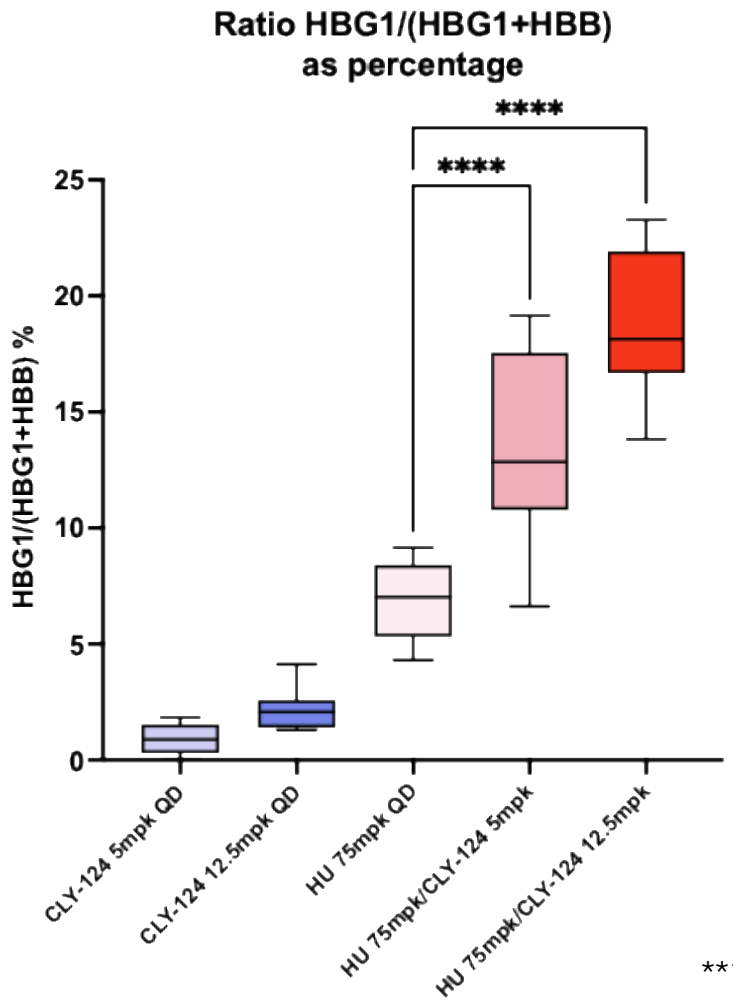
≈ 40%

≈ 10-15%

*Wu et al (2019); Qin et al (2022)

Globin gene ratios superior to HU and approaching gene therapy levels

CLY-124 is synergistic with HU in vivo



**** = Statistical significance

- ✓ NBSGW mice engrafted with human CD34+ cells receiving low doses of CLY-124 in combination with HU (75mpk)
- ✓ CLY124 + HU achieves greater HbF than either monotherapy alone
- ✓ Synergy proves that CLY-124 is mechanistically distinct from hydroxyurea
- ✓ In phase 1 study, lower dose cohorts may show HbF increase
- ✓ Derisked early clinical development with current HU patients

Cellarity

Summary

- NEDDylation and its regulation through DCN1 is a novel mechanism of HbF induction
- DCN1 inhibition demonstrates specificity for globin gene regulation not seen in pan-neddylation inhibitors
- DCN1 inhibition leads to globin switching without stress erythropoiesis or cytotoxicity
- CLY-124 is a specific and potent DCN1 inhibitor with dose-dependent HbF induction in vivo and synergy with HU
- GLP studies of CLY-124 do not show cytopenias, major organ dysfunction or genotoxicity
- CLY-124 is being studied in a phase 1 trial in healthy adults and patients with SCD

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Corey Hayford

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