CRISPR-Cas9 gene therapy for Sickle Cell Disease and β-Thalassemia

1) Most common monogenic diseases worldwide
   a) Transfusion-dependent β-Thalassemia: 60k/yr
   b) Sickle Cell Disease: 300k/yr

2) Genetic causes
   a) Mutations in hemoglobin β chain

   b) Hemoglobin: \( \text{HHb} \)

   1. TDT - reduced or absent β chain of hemoglobin
   2. SCD - \( \text{HBB C}6^v \) mutation (protective against malaria)

   oxygenation
   deoxygenation
   polymerization
   sickle cell

   Patients homozygous for \( \text{Hbb}^o \) TDT or \( \text{HBB C}6^v \)

   hemolytic anemia, painful vasculitic events

   Severe symptoms

   Phenotype is modifiable: why?
1) HbF > HbS
- Suppress SCD/TDT disease of type

Bcl11A

- Enhancer
- Enhances HbF production

A) Bcl11A = part of fetal switch

B) What happens if you reduce Bcl11A in erythrocytes?

Preclinical
- Harvest bone marrow
- Purity HPCs

CRISPR
- Targeting
- Enhancer
- Assess
- Engraftment

C2 guide RNA
- Off-target?

Healthy donors

Efficiency?
Patient 3 TDT

Collect Bone marrow

CRISPR Targeting of BCL11A

CTX001 cells (Therapy)

CRISPR-edited HSPCs

Do infused cells produce HbF?

Myeloblastosis

Infusion of CTX001 cells

Patient Timeline:

42 blood transfusions for TDT over 2.4 yrs (1x/3 weeks)

Blood transfusions post transplant

Freq. of BCL11A editing in nucleated blood cells

How know/figure this out?