CRISPR-Cas9 gene editing for Sickle Cell disease + β-Thalassemia

Most common genetic disease worldwide

- Transfusion-dependent β-Thalassemia - 60k/yr
- Sickle Cell Disease - 300k/yr

New diagnoses

TDT + SCD = impact β chain

- TDT: reduced or absent β chain
- SCD: HBB G6 mutation

Fetal Hemoglobin (α2 γ2) Adult hemoglobin

Globin gene synthesis

Onset of symptoms of TDT + SCD

* What happens to a patient who co-inherits hereditary persistence of fetal hemoglobin into adulthood with TDT or SCD?
BCL11A = Terminal rep. of HbF (δ) gene expression

Erythroid-specific Enhancer
(Turns on around birth)

BCL11A = part of the developmental switch required to turn off Hbδ and turn on HbB.

What happens if you reduce BCL11A for δ?
- δ levels do what? → what effect on TDT/SCD?

1) Preclinical
10 Healthy Donors

Harvest Bone Marrow
Purify CD34^+ cells (HSPCs)

CRISPR-Cas9
Targeting of BCL11A enhancer

Engraftment of HSPCs?

Targeting Efficiency?

Δ in γ? Hbδ

5' off Target
1/113 on Target

BCL11A
GATA

Erythroid Enhancer

1 2 3 4 5
1) Do infused cells persist?
   - @ infusion: 65,990 of CTX001 cells injected
     -> 46,190
     -> 64,390
     - 18 months: 62,990 nucleated peripheral blood cells

2) HBF levels: 0.3 g/dl -> 13.1 g/dl (TDT) Replaced HBA
   -> 9.19 g HBF -> 46% (2-fold Hb levels)

3) Symptoms:
   - CTX01 infusion
     - ~ 42 blood transfusions for TDT over 24 yrs
       - Transfusions = post-transplant
       - Every 3 weeks
   - 26.5 months

4) Why not KDR: BC1414A?

5) What about disease? Can't take cells out
   -> must target gene itself + fix mutation