DETERMINING OPTIMAL ENDPOINTS FOR GENE THERAPY IN SICKLE CELL DISEASE

Julie Kanter, MD
Director, Adult Sickle Cell Program
Co-Director Lifespan Sickle Cell Research Center
Associate Professor
Hematology/Oncology
University of Alabama at Birmingham
DISCLOSURE/CONFLICT OF INTEREST

- **Consultancy**: Guide point Global, GLG, Imara, Novartis, Editas
- **Honorarium**: Terumo, Bluebird Bio, Novartis, Global Blood Therapeutics
- **Honorarium**: Medscape, Rockpointe, Peervoice, Axis
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- **Steering Committee**: Novartis, Astrazeneca
- **Membership on a Scientific Advisory Committee**: Astrazeneca, BPL, Editas, Novartis, Modus, Sangamo

**Discussion of off-label drug use**: N/A
OUTLINE:

- Sickle Cell Disease: Definitions and Pathophysiology
- Current Therapies in Sickle Cell Disease
- Challenges and Barriers
- Defining the endpoints for gene therapy in treating sickle cell disease
SICKLE CELL DISEASE
Glutamate (glu), a negatively charged amino acid, is replaced by valine (val), which has no charge.
The result of a single point mutation is a significant change in hemoglobin structure which leads to an entire disease.

Note: The Sickle hemoglobin image is drawn at 50% of the size of the Normal hemoglobin.
MOLECULAR PATHOPHYSIOLOGY OF SCD

A single gene mutation is responsible for hemoglobin S.

There is significant phenotypic diversity not accounted for by hemoglobin genotypes.

Other genetic polymorphisms and differences in gene product function contribute to the complexity of phenotypic expression.

Although clinical patterns exist based on genotype, each individual with SCD has a unique clinical course.
PAIN: THE HALLMARK FEATURE OF SCD

• Hallmark of disease
• Primary reason people seek care
• Ischemic
• Secondary to vaso-occlusion
• Ubiquitous
• Present throughout life
SCD AND MORTALITY IN THE US

- Childhood survival 96%-98% for all genotypes
- In 2014, most deaths (66%) occurred at ages 25-54 years
- More recent surveillance data from Georgia and California showed mean age at death was 43 years for women, 41 years for men

COMPLICATIONS IN SCD

- Neurocognitive dysfunction
  - Meningitis
  - Stroke
- Indirect hyperbilirubinaemia
  - Sickle hepatopathy
- Gallstones
- Albuminuria
  - Isosthenuria
  - Substantial kidney injury
  - Papillary necrosis
- Delayed puberty
  - Erectile dysfunction
  - Priapism
- Avascular necrosis
- Bone marrow infarction
  - Osteomyelitis
- Retinopathy
  - Post-hypHEMA glaucoma
  - Retinal infarction
- Pulmonary hypertension
  - Acute chest syndrome
  - Acute pain event
- Cardiomegaly
  - Diastolic heart failure
- Anaemia
  - Leukocytosis
- Septicaemia
- Functional asplenia
- Splenic infarction
  - Splenic sequestration
- Complications of pregnancy
  - Skin ulcers
  - Chronic pain

CURRENT THERAPIES FOR SCD
DISEASE MODIFYING OPTIONS

• Hydroxyurea
  • First approved therapy
  • Modifies the course of SCD
  • Decreases the frequency of pain crisis for sickle cell anemia
  • Not universally accepted

• L-Glutamine
  • Anti-inflammatory

• Blood Transfusions
  • Improve stroke risk
  • Decrease recurrent acute chest syndrome in several studies
  • Multiple unwanted complications

• Palliative management
  • Pain management, opioids
STEM CELL TRANSPLANT

- Bone Marrow (STEM CELL) Transplant is the only cure for sickle cell disease at this time.
- Results of many studies show that transplants from matched related donors offer approximately an 85% chance of cure.
- Risk-vs-benefit considerations for BMT in adult patients with SCD are more complex than those in pediatric patients due to increased organ damage.
- Early studies are optimistic for improved outcomes and improved quality of life.
CHALLENGES IN BONE MARROW TRANSPLANT FOR EVERYONE WITH SCD

- Not everyone has a “great” match
- There is still a risk of graft-versus-host disease
- Immune suppressive medication can be long-term
- Risk of late-rejection
GENE THERAPY FOR SCD: CIRCUMVENTS THE NEED FOR FINDING MATCHED DONORS
TWO MAIN TYPES OF GENE THERAPY

• Gene Addition Therapy
  • ADD A NEW GENE
  • Don’t remove or change any of the existing genes

• Gene Editing
  • Edit a gene that is in the body
  • Sometimes—also add a new gene (HDR)
DEFINING ENDPOINTS IN SCD
ENDPOINTS IN PREVIOUS SCD THERAPEUTIC TRIAL

• Vaso-occlusive crisis/Pain severity

PROBLEM: Pain is subjective, has multiple causes, and can be difficult to differentiate nociceptive/ischemic from chronic pain

• Biologic endpoints
  • Transcranial doppler velocity
  • Proteinuria: marker of renal dysfunction
  • TR jet velocity
  • Pulmonary hypertension
  • DLCO

• Predictors of disease severity ≠ disease modifiers
  • Fetal hemoglobin
  • Total hemoglobin
  • White blood cell count
ENDPOINTS FOR GENE THERAPY FOR SCD
GENE THERAPY ENDPOINTS

- Outcomes of stem cell transplant in SCD demonstrate that sufficient engraftment of donor stem cells leads to curative therapy
  - Resolution of vaso-occlusive pain crisis
  - Decreased/absent risk of stroke
  - Stabilization of end organ dysfunction
- Outcomes achieved through a sufficient myeloid engraftment to yield stable hemoglobin production
- Mixed chimerism is acceptable as long as the resulting normal hemoglobin A >> HbS
HOW DO WE MEASURE GENE THERAPY

- Vector copy number (VCN) per cell: Average number of gene therapy letters delivered to a sample of blood stem cells

- Percentage of stem cells transduced with the vector: Percentage of blood stems cells which have received a gene therapy letter

- Cell dose: Amount of patient’s own blood stem cells returned to the patient after gene therapy letter was delivered
HOW ELSE TO MEASURE EFFECT?

Exploratory assay allows for single-cell resolution of Hb expression to assess pancellularity of HbA^{T87Q}

- Exploratory assay: Single red blood cell western blot with anti-β^{S} or anti-β^{A}/β^{A-T87Q} antibodies

<table>
<thead>
<tr>
<th>RBCs with normal adult Hb β^{A}/β^{A}</th>
<th>RBCs from β^{S}/β^{S} SCD patient</th>
<th>RBCs from sickle cell trait β^{S}/β^{A}</th>
<th>RBCs from β^{S}/β^{S} SCD patient on transfusions</th>
<th>RBCs from LentiGlobin treated β^{S}/β^{S} SCD patient</th>
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<tbody>
<tr>
<td>β^{A} only RBC</td>
<td>β^{S} only RBC</td>
<td>β^{S}/β^{A} RBC</td>
<td>β^{S}/β^{S} RBC containing β^{A-T87Q}</td>
<td>β^{S}/β^{S} RBC containing β^{A-T87Q}</td>
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Proportion of RBCs with HbS and/or HbA/HbA^{T87Q}

Data as of 7 March 2019
REFINEMENTS TO MANUFACTURING AND CELL HARVEST IMPROVED DRUG PRODUCT CHARACTERISTICS

**Vector copy number**

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**% Transduced cells**

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**CD34+ cell dose**

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*Group A shown as median (min – max); † Number of DP exceeds number of patients since some patients were harvested or mobilized more than once; ‡ 1 Group B DP lot was made using original manufacturing process, while the other 3 DP lots were using refined manufacturing process

BM, bone marrow; DP, drug product; HSC, hematopoietic stem cell; PB, peripheral blood; VCN, vector copy number
Peripheral blood VCN and HbAT87Q over time

Hb, hemoglobin; VCN, vector copy number

Data as of September 14, 2018
HOW ELSE TO MEASURE EFFECT?

- The new or edited gene has to make the “healthy” hemoglobin (protein)
- We have to know HOW MUCH hemoglobin the new gene is making?
- How much hemoglobin is stable?
- How is the new hemoglobin packaged?
GENE THERAPY ENDPOINTS

- We can translate stem cell transplant endpoints to gene therapy
- Sufficient VCN and transduction efficiency (or editing efficiency) to result in normal hemoglobin A>>HbS
- Pancellular expression is necessary
- Resolution (or near resolution) of hemolysis
- Rheologic properties equal to those of individuals with HbAS (sickle cell trait)
- Outcomes in studies with production of HbF are less defined due to the lack of a similar biologic model
- Long term:
  - Eventual resolution of vaso-occlusive pain
  - Decrease in stroke risk
  - Stabilization of organ dysfunction
SAFETY CONCERNS IN GENE THERAPY

- Insertional oncogenesis
- Random insertion
- Lack of sustainable gene to protein production
- Off target effects
- Difficulties and expense of identifying those off target effects
- Novel mutations
- Chemotherapy and secondary complications
- Lack of engraftment

Stopping points in all therapies must be defined

When is poor protein expression grounds for stopping ongoing investigation
WHAT IS THE FUTURE OF GENE THERAPY

• We have to define success
• We have to ensure successful therapy is approved by the FDA
• We have to monitor LONG TERM to see if these therapies give LONG TERM DISEASE MANAGEMENT/CURE
• We have to work to figure out how to make this type of therapy available, affordable and universal?