Gene Therapy

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Presenter Disclosure Information

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Gene therapy is not anymore science fiction

• Gene therapy trial listed under gene therapy in clinicaltrial.gov?
Gene therapy is not anymore science fiction

- Gene therapy trials listed under gene therapy in clinicaltrial.gov ? 589
  - Cancer
  - Rare diseases
How does gene therapy work?
Gene therapy for lipid disorders

1. Lipoprotein lipase deficiency
   • Human LPL [S447X, Alipogene Tiparvovec]
     – Improve post-prandial hypertriglyceridemia
     – ?reduce episodes of pancreatitis
     – Approved in the EU
     – Trial planned in US
Gene therapy for lipid disorders

2. Homozygous familial hypercholesterolemia (HoFH)
   • Human LDLR (NCT00891306)
Familial Hypercholesterolemia (FH)

- inherited disorder
- severe hypercholesterolemia with lifelong accumulation of plasma LDL-C
- premature CVD

Homozygous Familial Hypercholesterolemia

12 Y.O. female
LDL-C=780 mg/dL, xanthomas since age 3,
coronary heart disease, cardiac bypass
FH is Caused by Mutations in Genes Affecting LDL Receptor Functionality

- The impaired LDLR functionality leads to a decreased clearance of LDL particles from plasma

HeFH
LDL-C > 190 mg/dl

HoFH
LDL-C > 500 mg/dl

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Molecular causes of FH

**LDLR (chr 19p13):**
Primary familial hypercholesterolemia
OMIM: 143890

**APOB (chr 2p24):**
Fam. defective ApoB
OMIM: 144010

**PCSK9 (chr 1p32):**
Proprotein convertase subtilisin/kexin type 9
OMIM: 603776

**LDLRAP1 (chr 1p36):**
Autosomal recessive hypercholesterolemia
OMIM: 603813
Penn HoFH Cohort: 
LDLR Negative Subjects Have a More Severe Phenotype

<table>
<thead>
<tr>
<th></th>
<th>LDLR negative (n=18)</th>
<th>LDLR defective (n=16)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age (yr) at visit 1</strong></td>
<td>11.5 (3.3 - 29)</td>
<td>28.1 (3.3 - 44.6)</td>
<td>0.008</td>
</tr>
<tr>
<td><strong>Age at 1st xanthomas</strong></td>
<td>2.0 (0.25 - 4)</td>
<td>7.0 (1 - 15)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>Age (yr) at FH dx</strong></td>
<td>3.0 (0.5 - 7)</td>
<td>8.0 (2 - 17)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>TC (mg/dl) at dx</strong></td>
<td>895 (602-1260)</td>
<td>686 (519-900)</td>
<td>0.002</td>
</tr>
<tr>
<td><strong>Age (yr) at start of Rx</strong></td>
<td>5.0 (1.2 - 10)</td>
<td>16.0 (2 - 31)</td>
<td>0.001</td>
</tr>
<tr>
<td><strong>Age (yr) at CAD</strong></td>
<td>12.5 (6 - 16)</td>
<td>22.0 (16 - 37)</td>
<td>0.004</td>
</tr>
</tbody>
</table>

**Lipid profile at visit 1**

<table>
<thead>
<tr>
<th></th>
<th>Mean (SD)</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>TC (mg/dl)</strong></td>
<td>618 (238)</td>
<td>453 (162)</td>
<td>0.025</td>
</tr>
<tr>
<td><strong>LDL-C (mg/dl)</strong></td>
<td>535 (214)</td>
<td>393 (159)</td>
<td>0.040</td>
</tr>
<tr>
<td><strong>Apo B (mg/dl)</strong></td>
<td>388 (142)</td>
<td>293 (92)</td>
<td>0.032</td>
</tr>
</tbody>
</table>

Kolansky DM, Am J Cardiol 2008; 102:14-38-43  
Cuchel NLA 2016
New Lipid Lowering Drugs Affect LDL-C Levels by Inhibiting VLDL Secretion

mipomersen

lomitapide

B100

TG
Would liver-directed gene therapy of LDLR for HoFH work?
Liver transplantation normalizes LDL-C in patients with Homozygous Familial Hypercholesterolemia

Liver directed *ex vivo* gene therapy in patients with HoFH


<table>
<thead>
<tr>
<th>LDL cholesterol</th>
<th>Pre + statin</th>
<th>Pre - statin</th>
<th>Post - statin</th>
<th>Post + statin</th>
</tr>
</thead>
<tbody>
<tr>
<td>FH1</td>
<td>448 ± 30 (6)</td>
<td>482 ± 19 (7)</td>
<td>372 ± 35 (51)</td>
<td>380 ± 29 (17)</td>
</tr>
<tr>
<td>FH2</td>
<td>510 ± 24 (3)</td>
<td>516 ± 15 (5)</td>
<td>515 ± 43 (14)</td>
<td>477 ± 38 (56)</td>
</tr>
<tr>
<td>FH3</td>
<td>804 ± 49 (5)</td>
<td>792 ± 37 (6)</td>
<td>745 ± 82 (23)</td>
<td>803 ± 75 (5)</td>
</tr>
<tr>
<td>FH4</td>
<td>421 ± 44 (5)</td>
<td>530 ± 28 (5)</td>
<td>525 ± 55 (20)</td>
<td>448 ± 20 (7)</td>
</tr>
<tr>
<td>FH5</td>
<td>786 ± 103 (2)</td>
<td>737 ± 34 (12)</td>
<td>595 ± 41 (18)</td>
<td>664 ± 42</td>
</tr>
</tbody>
</table>


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Evolution of gene therapy strategies

- Adeno Associated Virus (AAV) family was discovered
- AAVs are much less immunogenic than vectors used before
- In vivo approach become possible
- Several AAVs are used in gene therapy for several genetic conditions
- AAV8 can produce efficient and prolonged gene transfer in the liver
AAV-LDLR administration reduces LDL-C levels and atherosclerotic burden in animal models
AAV8-LDLR administration reduces LDL-C levels in a humanized mouse model *

*humanized mouse model: LDLR-Apobec-DKO/hApoB-Tg mice

Kassim SH et al Human Gene Therapy. 2013, 24: 19-26
Mice transduced with hLDLR-L318D are resistant to PCSK9–mediated regulation

AAV8-mLDLR administration mediate regression of atherosclerotic lesions Ldlr-/-Apobec1-/-mice

High fat diet

Kassim et al 2010, Plos One
AAV8-based gene therapy is been used to treat Hemophilia B

Long-Term Safety and Efficacy of Factor IX Gene Therapy in Hemophilia B


NEJM 2014
AAV8-based gene therapy is been used to treat Hemophilia B

- **Efficacy:**
  - Sustained increased levels of FIX
  - Decreased use of replacement therapy

- **Safety**
  - Production of neutralizing antibodies
  - Transitory mild to moderate increased LFT
    - Possible T-cell activation with destruction of transduced hepatocytes
    - Steroid treatment provided to blunt immune response
FHGT002 - Phase I/II Clinical Trial in HoFH

• **Primary objectives:**
  To determine the **safety** of AAV-LDLR administration in patients with HoFH
  
  *AEs of special interest: LFTs*

• **Secondary objective:**
  To assess **efficacy**:
  
  LDL-C levels @ 3 months
  LDL catabolism @ 3 months

• **Exploratory objectives:**
• To assess **immune response** following vector administration
  
  HLA typing, NAb titer, T-cell response, Vector concentration

Clinicaltrials.gov NCT02651675
FHGT002 – Key inclusions criteria

1. Male or female ≥ 18 years of age.
2. **LDLR mutations** at both alleles in the setting of a clinical presentation consistent with HoFH.
3. **NAbs titer** <1:5
4. Stable concurrent allowed lipid lowering medications — statins, ezetimibe, bile acid sequestrants, PCSK9i
FHGT002 - Phase I/II Clinical Trial in HoFH

• Design:
  • Phase I/II open label dose escalation (3+3 design)

<table>
<thead>
<tr>
<th>Cohort</th>
<th>Dose (GC/kg)</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td>$2.5 \times 10^{12}$</td>
</tr>
<tr>
<td>2</td>
<td>$7.5 \times 10^{12}$</td>
</tr>
</tbody>
</table>

• Total subjects = 9 to 12
FHGT002 - Phase I/II Clinical Trial in HoFH

- Design

-2 to 2 Days

Weeks

-26 0 4 8 12 26 52

Kinetics  Inpatient  Kinetics

Screening stage  Vector administration stage  Follow up

Stop selected LLT

Selected LLTs can restart

Study timeline

Clinicaltrials.gov NCT02651675

Cuchel NLA 2016
Summary

- Treatment of rare disorders of lipid metabolism with gene therapy is becoming a possibility
- AAV-based gene therapy may be safe and effective
- Gene therapy for LDL deficiency and HoFH are currently been pursued
Acknowledgments

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  - RegenXBio