Predictive Genomics and Precision Cardiovascular Care

Jacques Genest MD

Cardiovascular Health Across the Lifespan
McGill University Health Center

ACC Rockies 15 March 2017 Banff, Ab
Disclosure J. Genest MD 2017

Advisory Board, Speaker’s Bureau, Consultant, Grants, Clinical Trials

- Merck *
- Pfizer
- Novartis
- AMGEN *
- Cerenis*

- Sanofi/Regeneron *
- Lilly*
- Aegerion
- RengenXBio

CADTH, CDR
INESSS
EAS, IAS

Stock ownership: none;
Off label use: none
* Scientific Advisory

Relevant disclosure: IMPROVE-IT, CANTOS, CAPREE steering Committees; REVEAL, ACCELERATE, AMG145, Sanofi, TANGO, Lilly Clinical Trials.
DSMB: RE-Energize trial
Predictive Genomics and Precision Cardiovascular Care

- Genetics in Cardiovascular Diseases
- Tier-1 Genetic testing: FH
- FHCanada Registry
- Novel therapies
Monogenic Disorders
Genetic Testing in Cardiovascular Diseases

1. Monogenic Disorders

- Dilated cardiomyopathies
- Hypertrophic cardiomyopathies
- Arrhythmogenic cardiomyopathies: Long QT syndromes, ARVD
- Familial Hypercholesterolemia
- Structural Defects (Marfan, etc.)
Genetic Testing in Cardiovascular Diseases

2. Pharmacogenomics and Complex traits

- Pharmacogenomics (warfarin, clopidogrel, beta-blockers, statin intolerance, etc…)
- Complex traits (BP, Diabetes, Dyslipidemias)
- Epigenetics

… Not quite ready for prime time.
Gene Structure: exons
DNA Sequencing

Cost per Raw Megabase of DNA Sequence

Moore’s Law

National Human Genome Research Institute

genome.gov/sequencingcosts

Genomics: Drinking from the firehose
Phenotype (BP, Glucose, LDL-C, WBC...)

Prevalence (Log scale)

10^1 10^6

Evidence-Based Medicine vs. Precision and Personalized Medicine

Evidence-Based Med.

Precision-Based Med.

99.99%

95%
Genetic testing in CVD

- When is genetic testing recommended for cardiovascular diseases?
- Lack of agreement on genetic panel (selected mutations – genechip vs. Exome-wide sequencing)
  - Cost a major hurdle
  - Bioinformatics
  - Expert interpretation
- Hereditary Breast and Ovarian Cancer Syndrome (HBOC) – *BRCA1* or *BRCA2* genes;
- Lynch syndrome (LS)
- Familial hypercholesterolemia (FH)

FH has been recognized as a tier 1 genetic disorder by the Centers for Disease Control Office of Public Health Genomics in the United States, meaning that sufficient evidence for health benefit exists to implement case finding via family history–based screening, cascade screening, or other strategies.

Gidding S et al. circulation 2015;132:2167
Franz Hals (1683)

Portrait of a sixty year old woman holding a book
Does Genetic Data Change Clinical Decision-Making?

- In the case of Familial Hypercholesterolemia: Yes
  - Allows precision medicine
  - Warrants cascade screening
Diagnostic Yield and Clinical Utility of Sequencing Familial Hypercholesterolemia Genes in Patients With Severe Hypercholesterolemia

Amit V. Khera, MD, Hong-Hee Won, PhD, Gina M. Pelosi, PhD, Kim S. Lawson, MS, Traci M. Bartz, MS

Xuan Deng, MPhil
Alexander G. B Akihiro Nomur
L. Adrienne C Heribert Schun
James G. Wilson
Diego Ardissino

HUMAN GENETICS

Genetic identification of familial hypercholesterolemia within a single U.S. health care system

Low-density Lipoprotein Cholesterol, Familial Hypercholesterolemia Mutation Status, and Risk for Coronary Artery Disease

Amit V. Khera, Hong-Hee Won, Gina M. Peloso, Sekar Kathiresan, on behalf of investigators from the Myocardial Infarction Genetics and CHARGE Consortia
**Clinical Importance:** CAD Risk is Substantially Higher in FH Mutation Carriers with LDL ≥ 190

<table>
<thead>
<tr>
<th></th>
<th>OR for CAD (95%CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>LDL ≥ 190 mg/dl</td>
<td></td>
</tr>
<tr>
<td>FH Mutation – (N = 1,264)</td>
<td></td>
</tr>
<tr>
<td>FH Mutation + (N = 73)</td>
<td></td>
</tr>
<tr>
<td>LDL &lt; 130 &amp; FH Mutation –</td>
<td>Reference</td>
</tr>
</tbody>
</table>

*Logistic Regression* in Myocardial Infarction Genetics Consortium Studies

*Covariates:* Gender, Study, 5 principal components of ancestry
Clinical Importance: For a Given Observed LDL, FH Mutation Carriers are at Increased Coronary Risk
Genetic identification of familial hypercholesterolemia within a single U.S. health care system

FH Variants Are Associated with CAD

Fig. 2. FH variants are associated with increased risk of CAD.  

Science 2016;354:6319
GWAS, Risk Factors, Genetics

Personalized Medicine for High Risk
### Association Between the Chromosome 9p21 Locus and Angiographic CAD Burden: A Collaborative Meta-Analysis

<table>
<thead>
<tr>
<th>Cohort</th>
<th>Ethnicity</th>
<th>size</th>
<th>OR (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CATHGEN</td>
<td>European ancestry</td>
<td>3021</td>
<td>1.22 (1.10, 1.36)</td>
</tr>
<tr>
<td>CDCS</td>
<td>European ancestry</td>
<td>678</td>
<td>0.92 (0.73, 1.16)</td>
</tr>
<tr>
<td>China</td>
<td>Han Chinese</td>
<td>1165</td>
<td>1.31 (1.10, 1.55)</td>
</tr>
<tr>
<td>Cleveland GB</td>
<td>European ancestry</td>
<td>2471</td>
<td>1.05 (0.93, 1.19)</td>
</tr>
<tr>
<td>EmCB</td>
<td>European ancestry</td>
<td>2357</td>
<td>1.16 (1.01, 1.34)</td>
</tr>
<tr>
<td>Feldkirch/Austria</td>
<td>European ancestry</td>
<td>914</td>
<td>0.88 (0.72, 1.06)</td>
</tr>
<tr>
<td>IHCS (replication set)</td>
<td>European ancestry</td>
<td>1014</td>
<td>1.08 (0.90, 1.28)</td>
</tr>
<tr>
<td>IHCS (sample set)</td>
<td>European ancestry</td>
<td>1748</td>
<td>1.11 (0.97, 1.28)</td>
</tr>
<tr>
<td>Japan</td>
<td>Japanese</td>
<td>596</td>
<td>1.03 (0.81, 1.30)</td>
</tr>
<tr>
<td>Korea</td>
<td>Korean</td>
<td>522</td>
<td>0.98 (0.77, 1.25)</td>
</tr>
<tr>
<td>MEDSTAR</td>
<td>European ancestry</td>
<td>824</td>
<td>1.19 (0.98, 1.45)</td>
</tr>
<tr>
<td>Munich/Germany</td>
<td>European ancestry</td>
<td>2028</td>
<td>1.07 (0.91, 1.26)</td>
</tr>
<tr>
<td>OHGS</td>
<td>European ancestry</td>
<td>1714</td>
<td>1.36 (1.18, 1.58)</td>
</tr>
<tr>
<td>PENNCATH</td>
<td>European ancestry</td>
<td>841</td>
<td>1.02 (0.82, 1.26)</td>
</tr>
<tr>
<td>SAS</td>
<td>European ancestry</td>
<td>1094</td>
<td>1.02 (0.86, 1.21)</td>
</tr>
<tr>
<td>D+L Overall</td>
<td></td>
<td></td>
<td>1.10 (1.04, 1.17)</td>
</tr>
<tr>
<td>Bayesian Overall</td>
<td></td>
<td></td>
<td>1.11 (1.07, 1.17)</td>
</tr>
</tbody>
</table>

**NOTE:** Weights are from random effects analysis

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**J Am Coll Cardiol. 2013;61(9):957-970**
Risk Factors and risk of MI

- Smoking
- Diabetes
- Hypertension
- Abd. Obesity
- Psychol index
- Fruits/Veg
- Exercise (-)
- Alcohol (-)
- Apo B / Apo AI

Yusuf S et al. INTERHEART Lancet 2004;364:937-952
Clinical Importance: For a Given Observed LDL, FH Mutation Carriers are at Increased Coronary Risk

- Mean LDL: 203 mg/dl
- Mean LDL: 205 mg/dl

Odds Ratio for Coronary Artery Disease (95%CI):

- No: 5.2 (4.4–6.2)
- Yes: 17.0 (5.3–77.9)

10-20x ↑ risk
Implications for Clinical Medicine

Routine Lipid Testing

Sequencing FH Genes

Differential Treatment

FH Mutation Negative

FH Mutation Positive

Lifestyle Changes
Early Pharmacotherapy
Cascade Screening

35 M → LDL 167 → Sequencing FH Genes → Differential Treatment

Routine Lipid Testing

LDL 167
New Therapies: PCSK9

- Inhibiting PCSK9
- Small molecules
- Monoclonal antibodies
- Silencing RNA
- PCSK9 vaccines
A: LDL-R pathway in absence of PCSK9

B: Intracellular PCSK9 route

C: Extracellular PCSK9 route

Mature PCSK9 Degradation
PCSK9 Directly Binds to the LDLR

Kwon et al. 2008. PNAS. 105:1820
Antibody technology has evolved over past decades

Red = mouse
Blue = human

Fully Mouse
1st generation
Highly Immunogenic

Chimeric
2nd generation
e.g. Abciximab
Can be time-consuming to create

Humanized
3rd generation
e.g. Bococizumab
Still very immunogenic

“Fully” Human
4th generation
e.g. Evolocumab and Alirocumab
Can be time-consuming to create

Nomenclature: Prefix (Pharma) C (Cardiovascular) UMAB
Anti-drug antibodies (ADA): the challenge

**Immunogenicity:**
- The potential for an antigen to induce an immune response
- Immunogenicity against therapeutic proteins that are not in the normal human repertoire is a normal immune response.
- Reaction to neo-antigens
  - Proteins are non-human
  - Fusion proteins create new epitopes
  - Unusual glycosylation
## PCSK9 Outcome Trials

<table>
<thead>
<tr>
<th>Trial</th>
<th>Alirocumab</th>
<th>Evolocumab</th>
<th>Bococizumab</th>
</tr>
</thead>
<tbody>
<tr>
<td>ODYSSEY Outcomes</td>
<td><strong>18,600</strong></td>
<td><strong>27,500</strong></td>
<td><strong>17,000</strong></td>
</tr>
<tr>
<td>(secondary prevention)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FOURIER</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(secondary prevention)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SPIRE1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(secondary prevention)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SPIRE2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(primary prevention)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

| No of patients | 18,600 | 27,500 | 17,000 | 10,000 |

<table>
<thead>
<tr>
<th>Dosage</th>
<th>s/c, Q2W</th>
<th>s/c, Q2W or Q4W</th>
<th>s/c, Q2W</th>
<th>s/c, Q2W</th>
</tr>
</thead>
</table>

|------------------|----------|-----------------|----------|----------|

<table>
<thead>
<tr>
<th>Expected End date</th>
<th>Mar 2018</th>
<th><strong>This Friday!</strong></th>
<th>Aug 2017</th>
<th>Aug 2017</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Primary endpoint</th>
<th>CHD death</th>
<th>non-fatal MI</th>
<th>fatal and non-fatal ischemic stroke</th>
<th>high risk UA requiring hospitalization</th>
<th>CV death</th>
<th>MI</th>
<th>Stroke</th>
<th>hospitalization for UA</th>
<th>coronary revascularization</th>
<th>CV death</th>
<th>non-fatal MI</th>
<th>non-fatal stroke</th>
<th>hospitalization for UA needing urgent revascularization</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Duration</th>
<th>Up to Month 64</th>
<th>Up to 5 years</th>
<th>Up to Month 60</th>
<th>Up to Month 60</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Population</th>
<th>Patients 4 to 52 wks post ACS</th>
<th>History of clinically evident CVD: MI, stroke or symptomatic PAD and ≥1 major RF or ≥2 minor RFs</th>
<th>High risk patients</th>
<th>High risk subjects</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>LDL-C ≥70 (1.8)</td>
<td>LDL-C ≥70 (1.8) or LDL-C ≥100 (2.6) or LDL-C ≥100 (2.6)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

46,100 patients
Mission, Vision & Goal

The MISSION of the Canadian FH Registry is to bring together a multi-disciplinary group of physicians, basic and clinical researchers to improve the delivery of care to patients with severe lipoprotein disorders, especially FH, and to foster collaborative research.

Our VISION is to create a Canada-wide network of academic clinics, integrating lipid specialists, endocrinologists and cardiologists to treat patients with the highest standard of care and to create a collaborative research environment. Using a “hub and spoke” model, the registry will be extended in various communities to link primary care physicians with provincial academic centers.

The GOALS are to improve care to patients with FH and to reduce cardiovascular disease in this population at high risk.
**ClinicalTrials.gov**
A service of the U.S. National Institutes of Health

[Try our beta test site](www.FHCanada.net)

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Home > Find Studies > Search Results > Study Record Detail

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**Familial Hypercholesterolemia Canada / Hypercholesterolemie Familiale Canada (FHCanada)**

This study is currently recruiting participants. (see Contacts and Locations)

Verified June 2014 by McGill University Health Center

Sponsor:
McGill University Health Center

Collaborators:

ClinicalTrials.gov Identifier:
NCT02009345

First received: November 21, 2013
Last updated: June 20, 2014
Last verified: June 2014
History of Changes
Number of participating sites in Canada

11 other sites extra-Québec that will submit to REB soon
2932 patients in the database so far.
**Chicoutimi**

<table>
<thead>
<tr>
<th>DB1/E21 Database Dx</th>
<th>Genotyped patients n=1207; Canadian definition</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Definite FH</td>
<td>Probable FH</td>
</tr>
<tr>
<td>C646Y</td>
<td>7</td>
<td>1</td>
</tr>
<tr>
<td>D&gt;15kb</td>
<td>235</td>
<td>13</td>
</tr>
<tr>
<td>E207K</td>
<td>14</td>
<td>0</td>
</tr>
<tr>
<td>R329X</td>
<td>14</td>
<td>0</td>
</tr>
<tr>
<td>W66G</td>
<td>579</td>
<td>72</td>
</tr>
<tr>
<td>Y468X</td>
<td>8</td>
<td>0</td>
</tr>
<tr>
<td>W66G/D&gt;15kb</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>None of the above*</td>
<td>57</td>
<td>194</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>916</strong></td>
<td><strong>280</strong></td>
</tr>
</tbody>
</table>

*: LDLR, PCSK9, APOB and LDLRAP1 genes were sequenced when LDL-C was > 8,0 mmol/L
LDL cholesterol burden in individuals with or without familial hypercholesterolaemia as a function of the age of initiation of statin therapy.

Nordestgaard B G et al. Eur Heart J 2013;eurheartj.eht273

© The Author 2013. Published by Oxford University Press on behalf of the European Society of Cardiology.
Other initiatives: Canadian Definition of FH

http://www.circl.ubc.ca
LDL-C 95% Percentile

Comparison in 95th percentile of LDL between males and females cohort: GDML lab data

n=3,336,046 patients
*LDL-C ≥ 5.0 mmol/L (>40 yo)

+ 

DNA Mutation
Xanthomas
LDL >8.5 mmol/l

Yes
Definite FH

No
1st degree relative with ↑ LDL-C
Or ASCVD or 1st degree relative with early onset ACVD

Yes
Probable FH (Consider DNA testing)

No
Hypercholesterolemia (Consider DNA testing)
### CardioRisk Calculator (TM)

#### Family History

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td>Male</td>
</tr>
<tr>
<td>Age</td>
<td>50</td>
</tr>
<tr>
<td>CAD atherosclerosis</td>
<td>No</td>
</tr>
<tr>
<td>Non-CAD atherosclerosis</td>
<td>No</td>
</tr>
<tr>
<td>Drug Treatment for Elevated LDL-C</td>
<td>Yes</td>
</tr>
<tr>
<td>Current Statin</td>
<td>Atorvastatin</td>
</tr>
<tr>
<td>Avg Daily Statin Dosage</td>
<td>80mg</td>
</tr>
<tr>
<td>Current Ezetimibe</td>
<td>10mg</td>
</tr>
<tr>
<td>Resin</td>
<td></td>
</tr>
<tr>
<td>PCSK9</td>
<td></td>
</tr>
<tr>
<td>Tendon xanthomata</td>
<td>No</td>
</tr>
<tr>
<td>Arcus cornealis below the age of 45yo</td>
<td>No</td>
</tr>
<tr>
<td>Known DNA mutation in FH related gene(s)</td>
<td>No</td>
</tr>
<tr>
<td>Known Baseline/Untreated LDL-C</td>
<td></td>
</tr>
<tr>
<td>Calculate Current LDL-C</td>
<td></td>
</tr>
<tr>
<td>Current/Treated LDL-C</td>
<td>3.05 mmol/L</td>
</tr>
<tr>
<td>Lp(a) if known</td>
<td></td>
</tr>
</tbody>
</table>

#### Other initiatives: Canadian Definition of FH

- CardioRisk Calculator (TM)
- FHCanada
  - Familial Hypercholesterolemia

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*Image of the CardioRisk Calculator interface showing input fields for various parameters related to family history and clinical information.*
Other initiatives: Canadian Definition of FH

Imputed LDL-C calculation:
- Imputed Baseline/Untreated LDL-C: 7.78 mmol/L (abnormal)

Save and add to patient’s file:
- Save button

Other diagnostic information:
- Canadian Criteria for HeFH:
  - Probable Clinical Familial Hypercholesterolemia (Consider DNA testing)
  - Imputed Baseline/Untreated LDL-C >= 5.0 mmol/L
  - Family History of premature CVD (1st Male relative < 55 yo)

- Simon Broome Register criteria for HeFH:
  - Possible diagnosis for Clinical Familial Hypercholesterolemia
  - Imputed Baseline/Untreated LDL-C > 4.9 mmol/L
  - Family History of premature CVD (1st Male relative < 55 yo)

- Dutch Lipid Clinic Network Criteria for HeFH:
  - Probable Clinical Familial Hypercholesterolemia
  - Imputed Baseline/Untreated LDL-C >= 6.50 mmol/L
  - Family History of premature CVD (1st Male relative < 55 yo)
Predictive Genomics and Precision Cardiovascular Care

- Genetics in Cardiovascular Diseases
- Tier-1 Genetic testing: FH
- FHCanada Registry
- Novel therapies
ORION-1
Inclisiran inhibits PCSK9 synthesis by RNA interference
Planned interim analysis of a multi-center randomized controlled dose-finding trial

Kausik K Ray, Ulf Landmesser, Lawrence A Leiter, David Kallend, Peter Wijngaard
Robert Dufour, Timothy Hall, Mahir Karakas, Traci Turner, Frank LJ Visseren,
R Scott Wright, and John JP Kastelein

On behalf of the ORION-1 investigators
Antisense phosphorothioate oligonucleotide of 15-30 nucleotides wherein all of the backbone linkages are modified by adding a sulfur at the non-bridging oxygen (phosphorothioate) and a stretch of at least 10 consecutive nucleotides remain unmodified (deoxy sugars) and the remaining nucleotides contain an O´-methyl O´-ethyl substitution at the 2´ position (MOE).
Oligonucleotide Therapeutics — A New Class of Cholesterol-Lowering Drugs

Anastasia Khvorova, Ph.D.

A Chemical Configuration

3-Passenger-strand bioconjugate
Triantennary GalNAc

Backbone modifications
Phosphorothioate

2-Ribose modifications

2'-Deoxy
2'-Fluoro
2'-O-Methyl

Guide Strand
Passenger Strand

Khvorova A. NEJM 2017;376:4
PCSK9 synthesis inhibition via RNA interference

Inclisiran harnesses a natural catalytic process

- Synthetic double strand 21-23mer oligonucleotide
- 3x GalNAc at sense 3’ end enables hepatic-specific uptake via ASGP receptor
- Chemically modified to prevent RNAse degradation
- Dicer separates antisense strand – and incorporates it into RISC
- RISC degrades PCSK9 mRNA catalytically to halt PCSK9 protein synthesis in the liver

RISC - RNA induced silencing complex
**B Mechanism of Action**

- PCSK9 down-regulates LDL receptors, increasing serum LDL cholesterol levels.
- Up-regulation of LDL receptors, decreasing serum LDL cholesterol levels.
- Asialoglycoprotein receptor (ASGPR).
- Normal clearance of LDL cholesterol.

1. PCSK9 production and secretion.
2. LDL receptor degradation.
3. PCSK9 mRNA degradation.
4. PCSK9 mRNA complex cleaves PCSK9 mRNA.
5. LDL receptor production and secretion.

- siRNA loaded into RISC complex.
- RISC guide strand.
- RISC siRNA complex cleaves PCSK9 mRNA.
- Passenger siRNA strand removed.

DNA, Nucleus.
RNA-Targeted Antisense Drugs Block the Translation of PCSK9 Protein

Efficacy of one dose of inclisiran up to day 90
Significant, durable PCSK9 and LDL-C lowering

**PCSK9**

- Mean percent change (±95% CI)
- Days from first injection
- p-value for all comparisons to placebo < 0.0001

**LDL-C**

- Mean percent change (±95% CI)
- Days from first injection
- p-value for all comparisons to placebo < 0.0001

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- **Placebo (N=124)**
- **100mg (N=59)**
- **200mg (N=121)**
- **300mg (N=120)**
- **500mg (N=64)**
A: LDL-R pathway in absence of PCSK9

B: Intracellular PCSK9 route

C: Extracellular PCSK9 route
A Cholesterol-Lowering VLP Vaccine that Targets PCSK9

Erin Crosseya,1,2, Marcelo J. A. Amarb,1,2, Maureen Sampsonb, Julianne Peabodya, John T. Schillerc, Bryce Chackeriana,1,2,* and Alan T. Remaleyb,2,*

aDepartment of Molecular Genetics and Microbiology, University of New Mexico, MSC08-4660, Albuquerque, NM 87131, USA

bLipoprotein Metabolism Section, Cardio-Pulmonary Branch, National Heart, Lung and Blood Institute, National Institutes of Health, Building 10 – 2C433, 10 Center Drive, MSC 1666, Bethesda, MD 20892, USA

cLaboratory of Cellular Oncology, National Cancer Institute, National Institutes of Health, Bethesda, MD 20892, USA
Conclusions

- LDL-C (and Remnant cholesterol) is causal in atherosclerosis
- Lowering non-HDL-C with statins decreases cardiovascular events, CV deaths and total mortality
- Non-statin drugs may provide additional benefit
- PCSK9 inhibitors lower LDL-C. They are indicated in FH and ASCVD patients not at goal. Cost is a major limitation
HDL Stories

Torcetrapib NEJM 2007
ABCA1 Mendelian Randomization 2008
ACCORD NEJM 2010
AIM-HIGH May 2011
Dalcetrapib May 2012
HDL Mendelian Randomization 2012
HPS-2 THRIVE Dec 2012
Evacetrapib 2015
ApoAl Milano 2016