Introduction to Clinical Research Boot Camp 2019

ITHS | Institute of Translational Health Sciences

ACCELERATING RESEARCH. IMPROVING HEALTH.

Welcome!

Tuesday, July 30-Wednesday, July 31

8:00am-4:30pm

UW Husky Union Building

Lyceum/250/145

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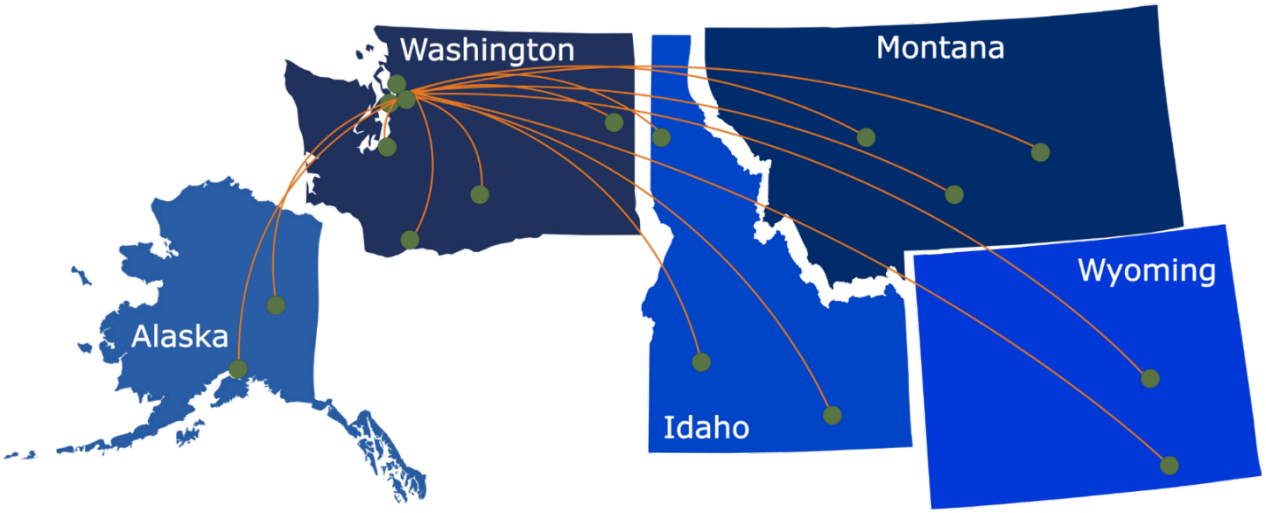

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What We Offer:

1. **Research Support Services:** Members gain access the different research services, resources, and tools offered by ITHS, including the ITHS Research Navigator.

2. **Community Engagement:** Members can connect with regional and community based practice networks

3. **Education & Training:** Members can access a variety of workforce development and mentoring programs and apply for formal training programs.

4. **Funding:** Members can apply for local and national pilot grants and other funding opportunities. ITHS also offers letters of support for grant submissions.

Contact our **Research Navigator**

- **Project Consultation**
- **Strategic Direction**
- **Resources and Networking**

Melissa D. Vaught, Ph.D.
ithsnav@uw.edu
206.616.3875

Introduction to Clinical Research Boot Camp 2019

Keynote

Working toward a cure in Hemophilia;

Progress in Gene Therapy

Presented by Barbara Konkle, MD

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Working Toward a Cure in Hemophilia: Progress in Gene Therapy

Barbara A. Konkle, M.D.
Chief Scientific Officer
Associate Director, Washington Center for Bleeding Disorders
Director, Hemostasis, Platelet Immunology and Genomics Laboratory
Bloodworks Northwest
Professor of Medicine/Hematology
University of Washington
Seattle, WA USA

UNIVERSITY of WASHINGTON

ITHS 7.30.19

Bloodworks Northwest

Disclosures

Shareholder	No relevant conflicts of interest to disclose
Grant / Research Support	Octapharma, Pfizer, Spark, Takeda/Shire, Uniqure, Sanofi
Consultant	BioMarin, Pfizer, Roche/Genentech, Sanofi
Employee	No relevant conflicts of interest to disclose
Paid Instructor	No relevant conflicts of interest to disclose
Speaker bureau	No relevant conflicts of interest to disclose
Other	No relevant conflicts of interest to disclose

Outline

- Brief history of gene therapy
  - Advances and setbacks
- Hemophilia as a target for gene therapy
- Ethical issues in gene therapy research/commercialization

Gene Therapy

- Definition: *Products that mediate their effects by transcription and/or translation of transferred genetic material and/or by integrating into the host genome and that are administered as nucleic acids, viruses or genetically engineered microorganisms*[1]
- Approaches:[2]
  - Somatic gene therapy
    - Change is not passed along to the next generation
    - Current approved approach
  - Germline gene therapy
    - Therapeutic or modified gene will be passed on to next generation[3]

Gene-edited babies: What went wrong and what could go wrong

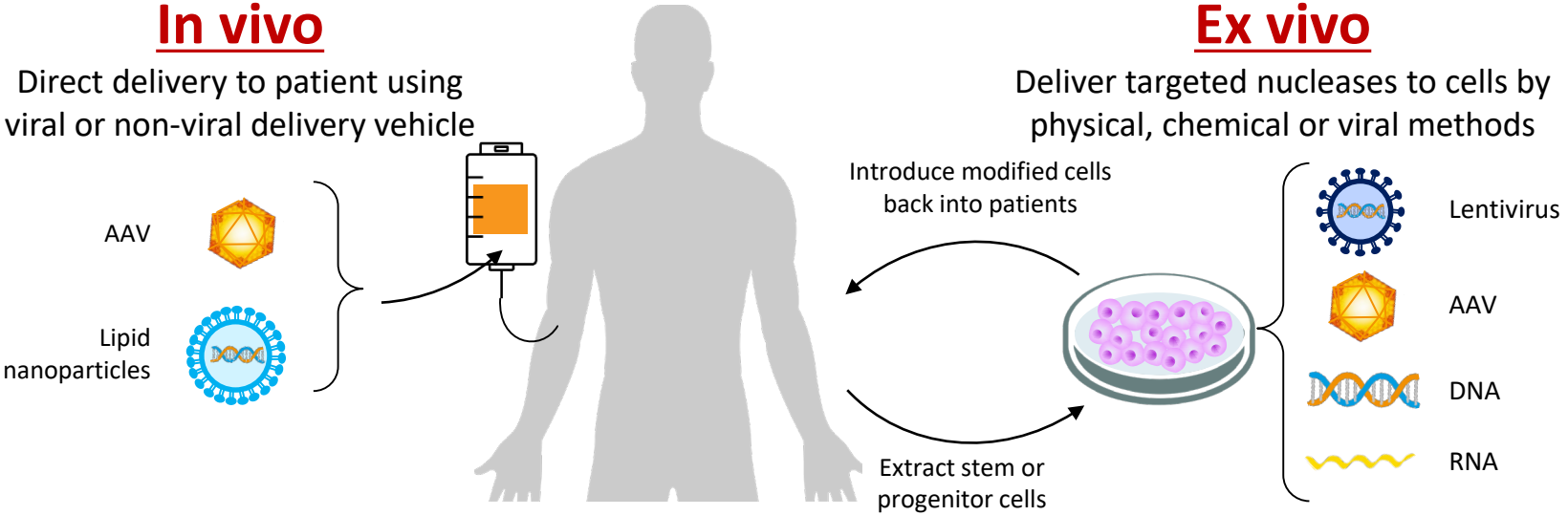
Haoyi Wang, Hui Yang

Published: April 30, 2019 • https://doi.org/10.1371/journal.pbio.3000224

PLOS BIOLOGY FIFTEENTH ANNIVERSARY

1. US FDA. https://www.fda.gov/media/81682/download (Accessed June 2019). 2. Wirth T, et al. *Gene* 2013;525:162. 3. Wang H, Yang H. *PLoS Biol* 2019;30;17(4):e3000224.

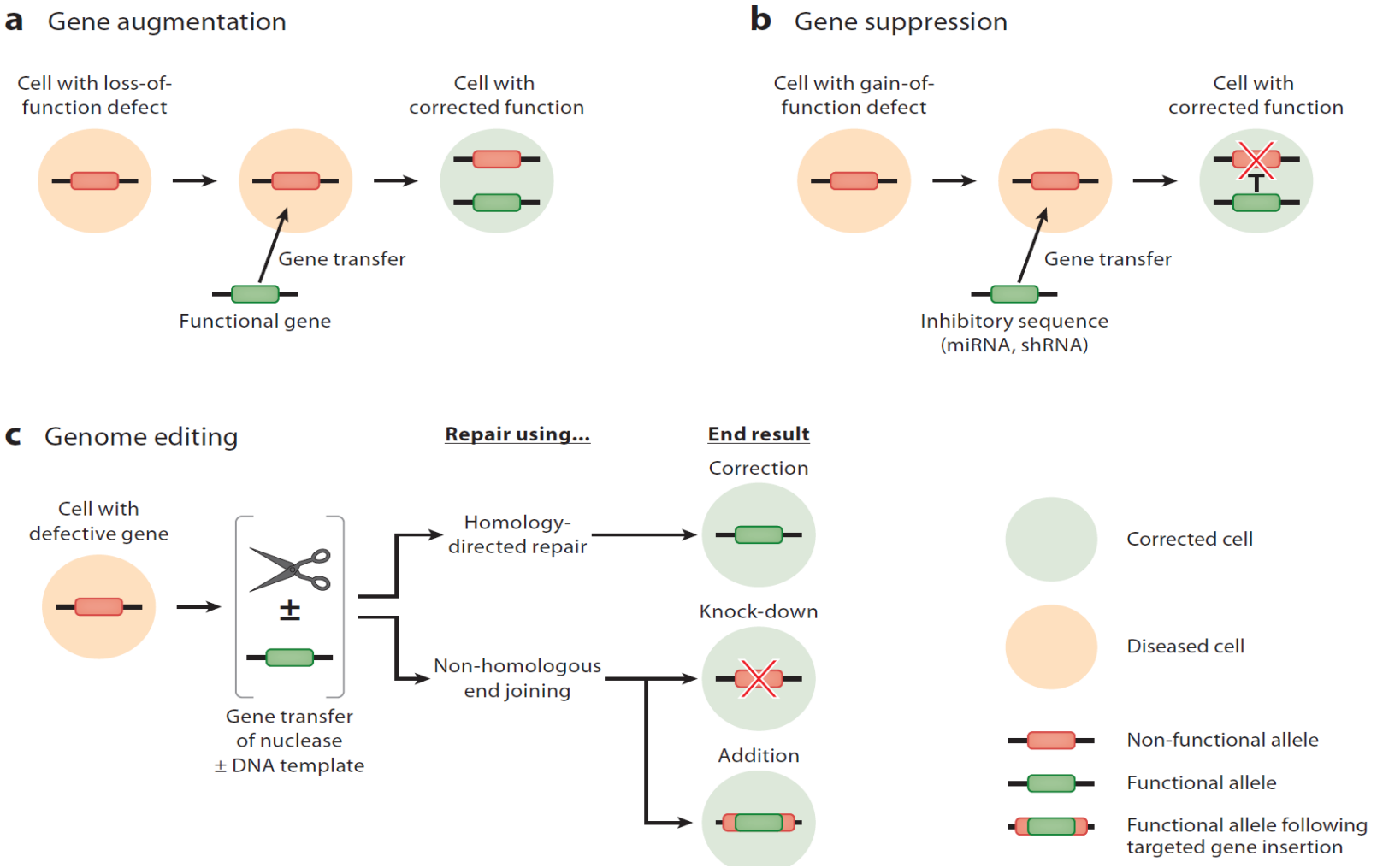
Approaches to Gene Therapy



- Common therapeutic strategies[1]
  - Lentivirus for *ex vivo* gene transfer into hematopoietic and other stem cells[2,3]
  - AAV for *in vivo* transfer into postmitotic tissues[2,4]

Image adapted from US FDA – What is gene therapy.[1]
AAV: Adeno-associated virus.
1. US FDA. https://www.fda.gov/vaccines-blood-biologics/cellular-gene-therapy-products/what-gene-therapy (Accessed June 2019). 2. Mingozzi F, High KA. *Nat Rev Genet* 2011:12:341. 3. Milone MC, O'Doherty U. *Leukemia* 2018;32:1529. 4. Colella P, et al. *Mol Ther Methods Clin Dev* 2018;8:87.

Approaches to Gene Therapy - 2



Anguela and High. Ann Rev Med. 2019;70:273-88.

Milestones in Gene Therapy

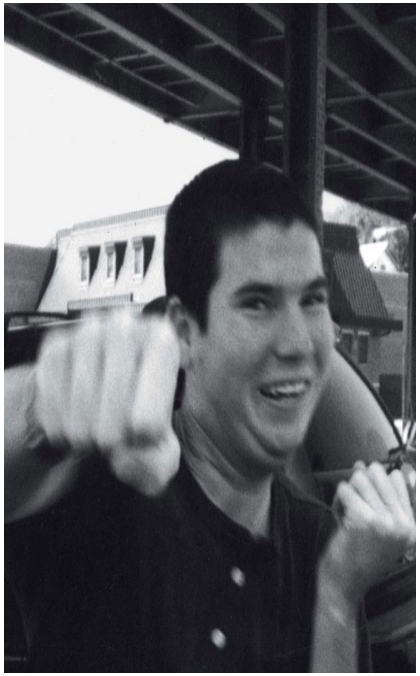
- Early studies with advances, but also setbacks
- First therapeutic ex-vivo gene therapy in 1990s
  - X-linked severe combined immune deficiency (SCID)
    - First generation γ-retroviral vectors with gene expressed under the control of viral regulatory elements
    - Positive response, however 5/20 developed leukemia due to insertional mutagenesis
  - Adenosine deaminase deficiency (ADA-SCID)
    - Retroviral transfer of ADA gene into HSCs
    - Early partial response, now with efficacy comparable to enzyme replacement
    - Approved by EMA in 2016
    - No leukemia
- Lentiviral vectors thought to be less genotoxic than retroviral vectors
  - Vectors under clinical development without viral regulatory elements

ADA-SCID: Adenosine deaminase severe combined immunodeficiency; EMA: European Medicines Agency; US FDA: U.S. Food and Drug Administration.

1. Wirth T, et al. *Gene* 2013;525:162. 2. Mingozzi F, High KA. *Nat Reviews: Genetics* 2011;12:341. 3. Anguela XM, High KA. *Annu Rev Med* 2019;70:273.

Major Setback in Gene Therapy in 1999

- Death of Jesse Gelsinger from adenoviral-mediated gene therapy for partial ornithine transcarbamylase deficiency
  - Major systemic reaction
  - Death from multi-organ failure
- Issues raised
  - Did subject meet inclusion criteria?
    - Milder disease
  - Conflict of interests
    - Involvement of investigator who developed vector in clinical trial
  - Did they underplay potential immune response?



JG 3 months before death

1. Wirth T, et al. *Gene* 2013;525:162. 2. Mingozzi F, High KA. *Nat Reviews: Genetics* 2011;12:341. 3. Anguela XM, High KA. *Annu Rev Med* 2019;70:273.

Continued Progress in Gene Therapy

- Steady progress in 21st century resulting in drug approvals
  - 2012, EMA approves first gene therapy Alipogene tiparvovec, for lipoprotein lipase deficiency
  - 2018, US FDA and EMA approve Voretigene neparvovec for RPE65 mutation-associated retinal dystrophy


SCIENTIFIC AMERICAN

The Most Expensive U.S. Medicine Now Has an Official Sticker Price

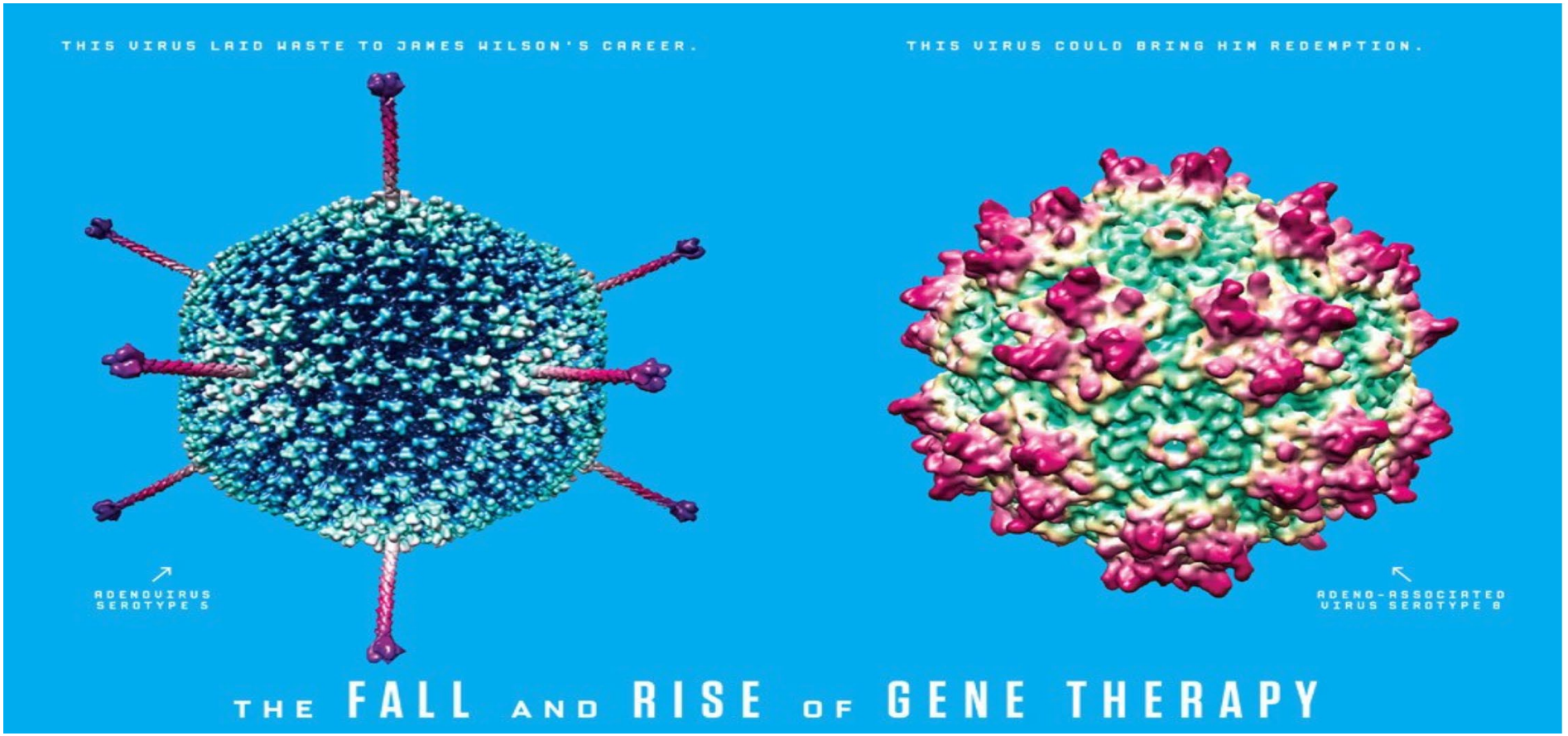
This gene therapy for vision loss will initially cost $850,000

- On June 9, 2019:
  - 3985 gene therapy studies on ClinicalTrials.gov

1. Mingozzi F, High KA. *Nat Reviews: Genetics* 2011;12:341. 2. Anguela XM, High KA. *Annu Rev Med* 2019;70:273.
3. https://clinicaltrials.gov/ct2/results?cond=&term=gene+therapy&cntry=&state=&city=&dist= (Accessed June 2019).

CARL ZIMMER SCIENCE 08.13.13 06:30 AM

GENE THERAPY EMERGES FROM DISGRACE TO BE THE NEXT BIG THING, AGAIN



AAV-Mediated *in-vivo* Gene Therapy

- Most common approach for *in vivo* gene transfer into post-mitotic tissues
- Can be targeted with tissue-specific regulatory elements
- Native virus is not known to cause disease and virus is replication defective
- Mostly non-integrating

1. Mingozzi F, High KA. *Nat Rev Genet* 2011:12:341. 2. Colella P, et al, Molec Ther Method Clin Develop 2018;8:87.

Gene Therapy for Hemophilia

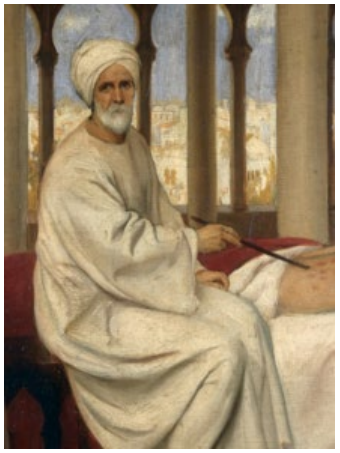
- Recognised early as good target
  - Single gene disorder[1]
  - Wide range of levels can produce therapeutic effect without safety concerns for factor activity[1]

- Early trials confirmed
  - Factor VIII and IX can be synthesized and undergo post-translational modification in cells that are not the normal site of production[2–4]
  - Functional factor activity can be secreted into the blood stream[2–4]

1. Lheriteau E, et al. *Blood Rev* 2015;29(5):321–8. 2. Murphy SL, High KA. *Br J Haematol* 2008;140:479–87. 3. Nathwani AC, et al. *N Engl J Med* 2011;365:2357–65.
4. Nathwani AC, et al. *N Engl J Med* 2014;137(21):1994–2004.

History of Hemophilia

- Talmud – 2nd century
  - Recognition of bleeding with circumcision
- Al-Zahrawi, renowned 10th-11th century Arab physician
  - Described families with hemorrhagic disorder in males
- John Otto, physician in Philadelphia, USA
  - In 1803, published a description of X-linked bleeding disorder.
- Queen Victoria – 19th century
  - Descendants spread hemophilia through Europe

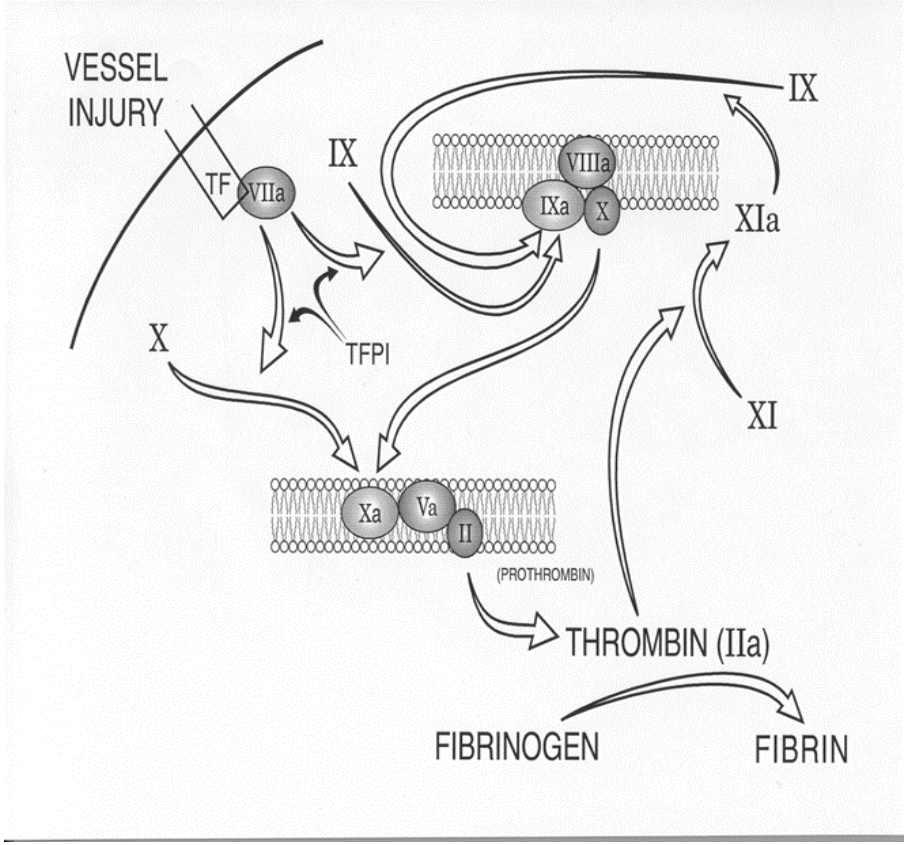


Hemophilia: Recognition

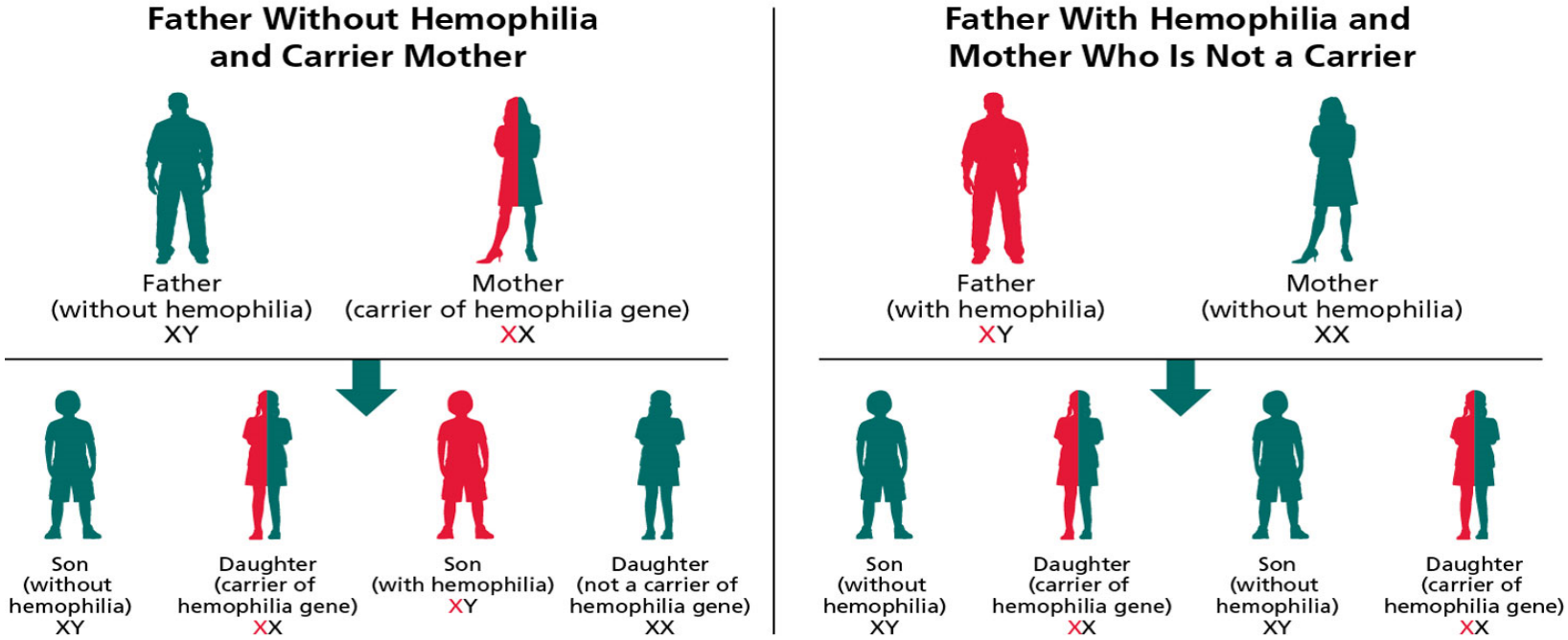
- Worldwide: At least 1/5000 male births
- New mutation rate ~ 30%
  - Thus hemophilia seen in all racial groups
  - First presentation may be bleeding symptoms in a female genetic carrier
- Hemophilia A - ~ 80% of cases
- Hemophilia B - ~ 20% of cases
- Presentation and diagnostic approach the same with A and B
  - Overall hemophilia B may be milder, but not useful on an individual patient level

Hemophilia: Pathophysiology

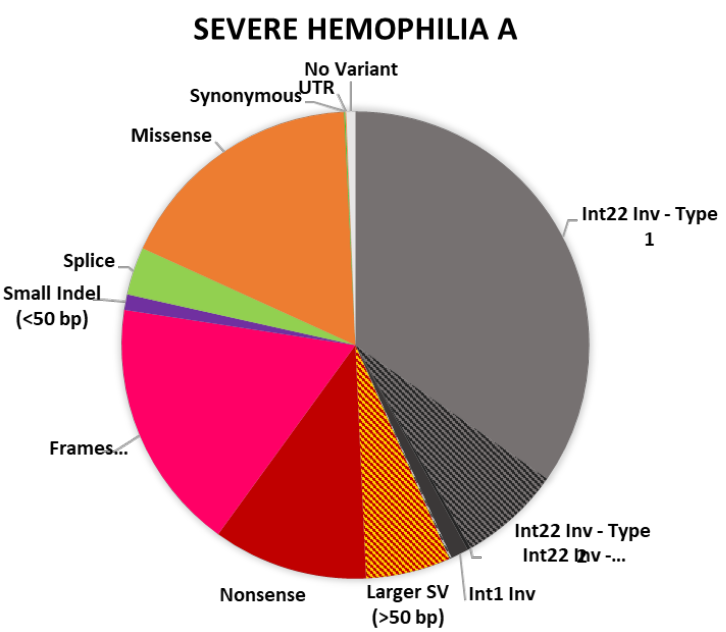
- FVIII accelerates the rate of FX activation by FIXa, eventually leading to the generation of thrombin (FIIa) and subsequent formation of the fibrin clot
- Deficiency of either FVIII or FIX predisposes to spontaneous and trauma-induced hemorrhage

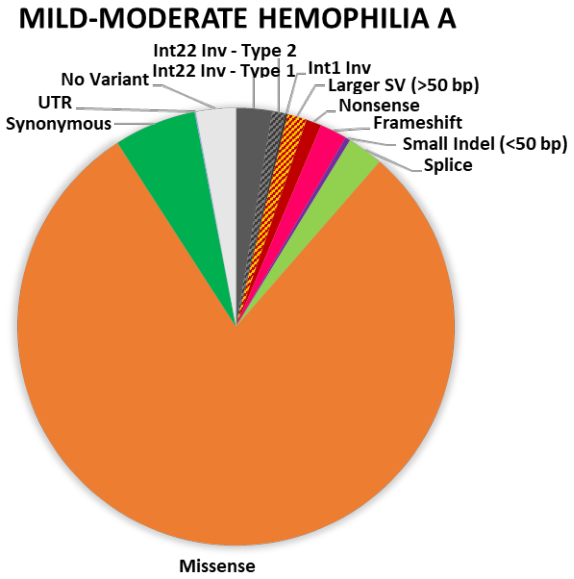


Inheritance of Hemophilia



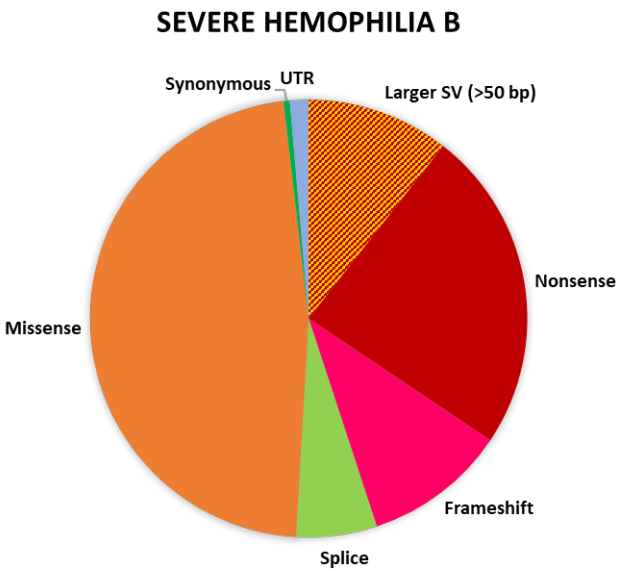
Genetics of Hemophilia A

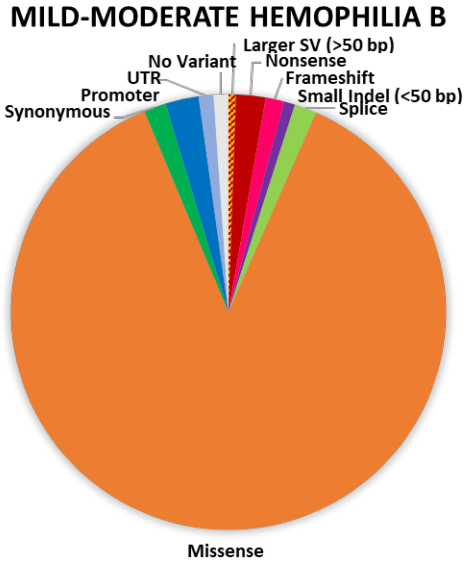




Johnsen JM, et al. *Blood Advances* 2017;1:824-834

Genetics of Hemophilia B





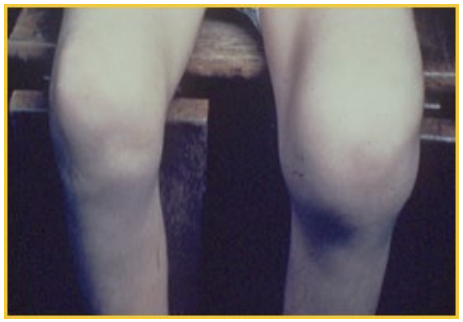
Johnsen JM, et al. *Blood Advances* 2017;1:8

Presentation of Hemophilia

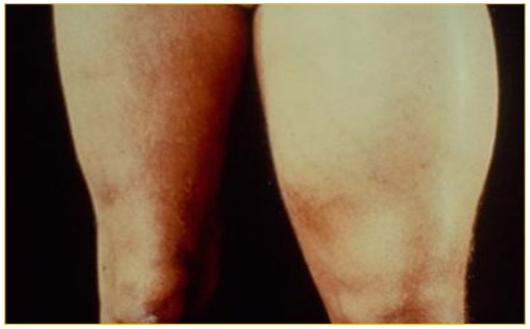
- Average onset of clinical symptoms
  - Severe: 1.5 years (many will present at birth)
  - Moderate: 3 years
  - Mild: 5 years
- Initial presentation:
  - Early postnatal procedures
  - With intramuscular injections
  - With dental eruptions/loss/tongue biting
  - Spontaneous hemarthroses after onset of walking

Sites of Bleeding

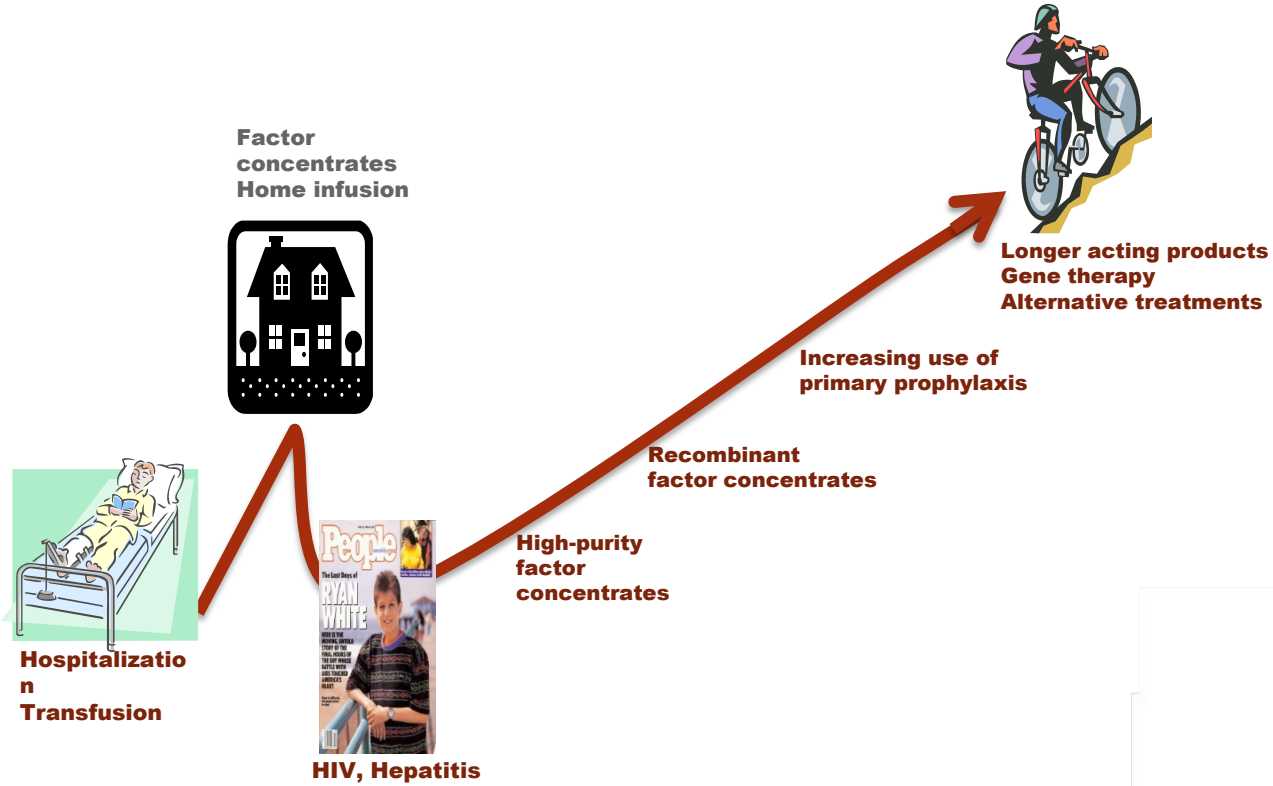
- Common
  - Mucous membrane
  - Soft tissue
  - Muscle
  - Joints (hemarthroses)



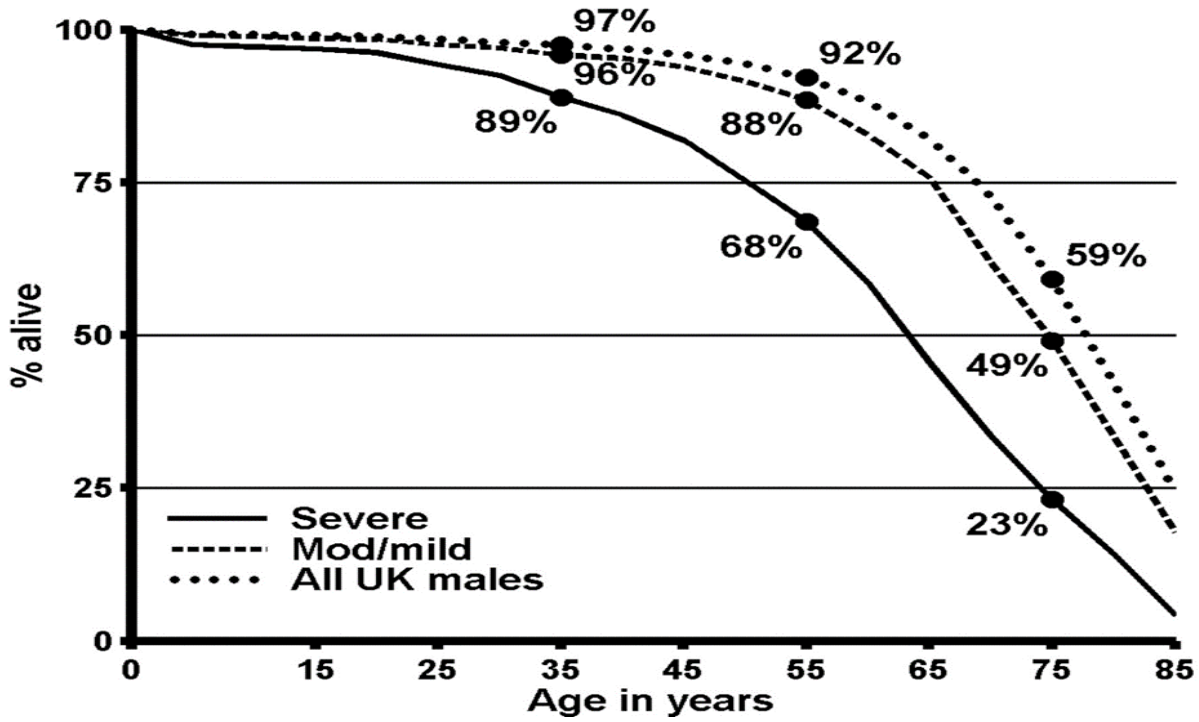
- Life-threatening
  - Central nervous system
  - Head
  - Neck and throat
  - Gastrointestinal
  - Retroperitoneal



Advances in Hemophilia Care: The Past Six Decades



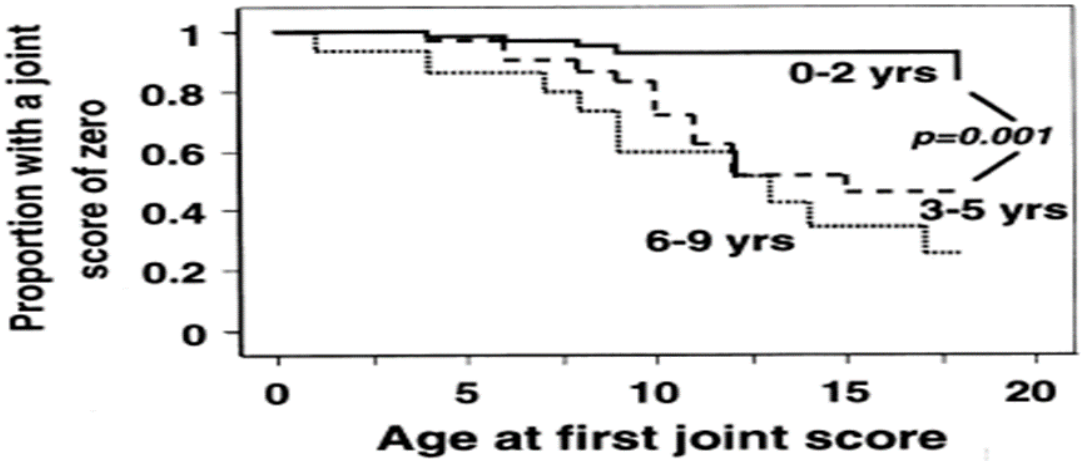
Effective therapy normalizes life expectancy



Darby et al, 2007

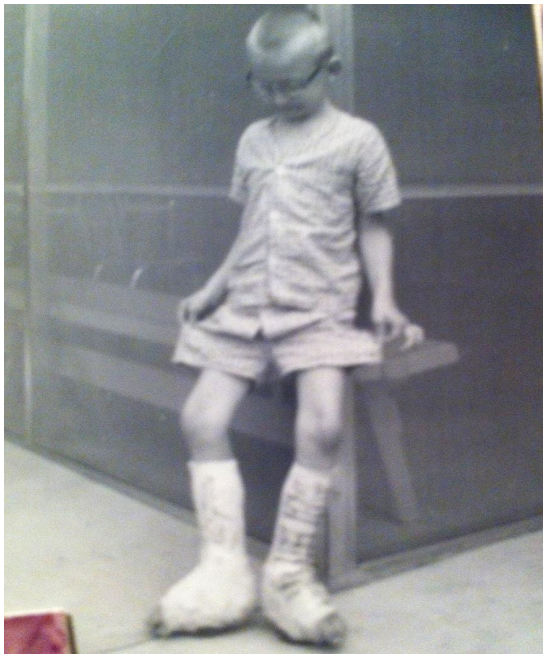
Joint Disease: Prevent by Primary Prophylaxis

- Prevents recurrent bleeding and chronic arthropathy
- Starting at an earlier age improves long-term outcomes
- Secondary prophy slows, but may not prevent, ongoing joint damage
- Low-dose primary prophylaxis can provide joint protection



Astermark J et al. *Br J Haematol.* 1999;105:1109-1113; Van den Berg HM et al. *Haemophilia.* 2006;12(suppl 3):159-168; Manco-Johnson MJ et al. *N Engl J Med*. 2007;357:535-544; Eshghi P et al. Clin Appl Thromb Hemost. 2018;24:513.; Wu RH, et al. Expert Rev Hematol. 2017;10:995.

Goal in Hemophilia Care



WFH
WORLD FEDERATION OF HEMOPHILIA
FÉDÉRATION MONDIALE DE L'HÉMOPHILIE
FEDERACIÓN MUNDIAL DE HEMOFILIA

WBDR
WFH World Bleeding Disorders Registry

Why gene therapy for hemophilia ?

- Factor therapy is very labor intensive and expensive
  - Breakthrough bleeding still occurs
- ~30% of patients with severe hemophilia A develop neutralizing antibodies (inhibitors) to treatment
- To date, alternative therapies do not normalize hemostasis
- Concern about treatment availability
- Patient desire to be cured of disease
- Most of the world without treatment

For prophylaxis with FVIII: Infusions every other day to twice weekly

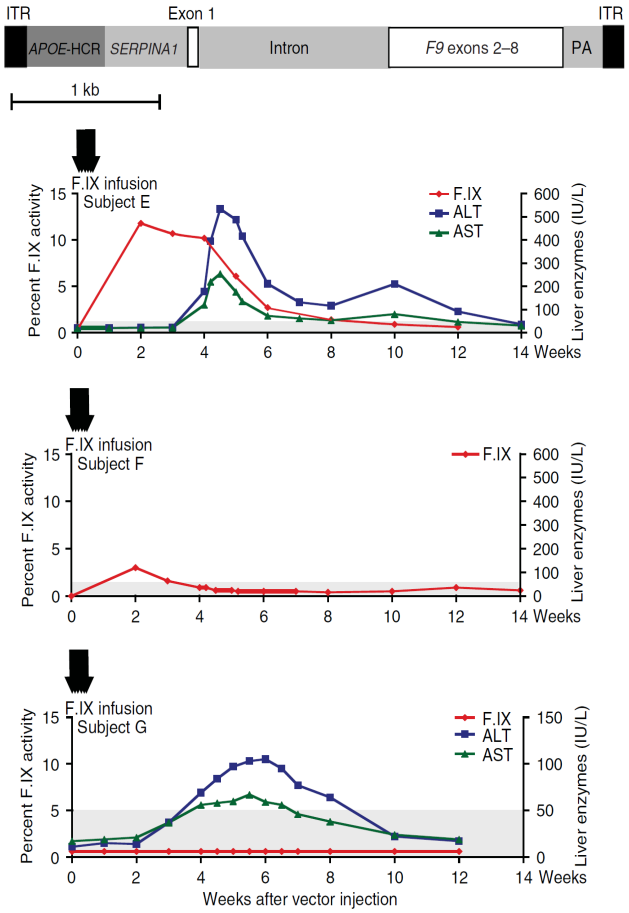
Gene Therapy Approaches in Hemophilia

Approach	Comments
Ex vivo F8 transfected fibroblast	• Implanted 100–400 million cells in peritoneal cavity • Small, transient increase in FVIII in 4/6 subjects
MoMLV-BDD-F8 IV	• Some evidence of vector in PBMCs • At most, small transient increases in FVIII
Adenovirus-F8	• Phase I trial stopped for inflammatory response in subject
Lentivirus	• In preclinical studies • Integrating vector, but risk of insertional mutagenesis decreased with improved vector design • Potential for use in liver-directed therapy in children • Ex vivo and in vivo HSC transduction to result in FVIII expression in megakaryocytes and platelets
AAV	• Vector used in current human trials • Wild-type virus is non-pathogenic • Predominantly non-integrating • Loss in dividing cells • Used for targeted integration into albumin locus

AAV: Adeno-associated virus; BDD: B-domain deleted; HSC: Hematopoietic stem cell; IV: Intravenous; MoMLV: Moloney murine leukemia virus; PBMC: Peripheral blood mononuclear cell.
Roth DA, et al. *N Engl J Med* 2001;344:1735. Powell JS, et al. *Blood* 2003;102:2038. Kelley et al. *Haemophilia* 2002;8:261-267. Evens H, et al. *Haemophilia* 2018;24(Suppl 6):50. Shi Q. *Molec Ther Methods Clin Dev* 2018;9:100. George L. *Blood Adv* 2017;1:2591.

AAV-Mediated Therapy in Hemophilia

- 1st in human
  - Intramuscular injection of *F9* construct into muscle[1]
    - Very low systemic expression with multiple muscles injected
      - Persistent expression in muscle[2]
- 1st liver infusion (AAV2-*F9*; CHOP/Stanford)[3]
  - Expression in high dose (2×10^{12}) subject
    - But unexpected hepatic inflammation and loss of transgene
      - Viral capsid T-cell immune response
  - Subject at same dose with anti-AAV2 antibodies
    - Limited expression
    - Study not continued

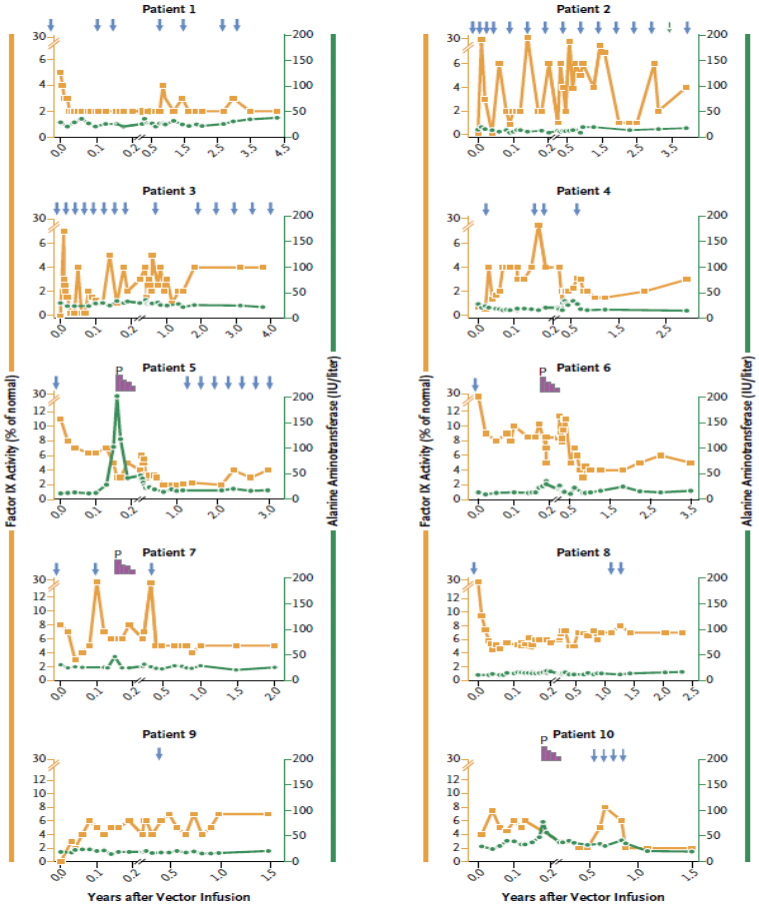


This slide contains information about a product that has not been approved by the Therapeutic Goods Administration.
Image from Manno *et al*.[3]
AAV: Adeno-associated virus; ALT: Alanine aminotransferase; AST: Aspartate aminotransferase.
1. Kay MA, et al. *Nat Genet* 2000;24:257. 2. Buchlis G, et al. *Blood* 2012;119:3038.3. Manno CS, et al. *Nat Med* 2006;12:342.

First study with long-term expression

Subsequent haemophilia B trial (St. Jude/UCL)

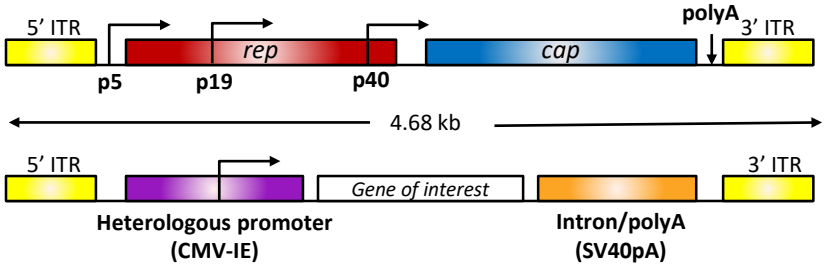
- Persistent FIX activity reported to date
  - Marked decrease in factor consumption
- Loss of transgene associated with transaminitis responsive to steroid therapy
- Study in long-term follow up



This slide contains information about a product that has not been approved by the Therapeutic Goods Administration.
Image from Nathwani *et al*.[2]

1. Nathwani AC, et al. *N Engl J Med* 2014;371:21. 2. Nathwani AC, et al. *Hematol Oncol Clin N Am* 2017;31:853.

Optimizing AAV Vectors



- Decrease in empty capsids
- Use of different AAV serotypes
- Optimization of liver-specific promoter/regulatory regions
- Codon optimization of *F8* and *F9* expression cassettes
- Use of optimized B-domain deleted *F8*
  - Size to allow optimal use of AAV
- Increase specific activity of *F9* insert through use of Padua variant (R338L)

Mingozzi F, High KA. *Nat Rev Genet* 2011;12:341. Evens H, et al. *Haemophilia* 2018;24(Suppl 6):50. George L. *Blood Adv* 2017; 1:2591. Pierce GF, Iorio A. *Haemophilia* 2018;24(Suppl.6):60. Colella P, et al. *Mol Ther Methods Clin Dev* 2018;8:87.

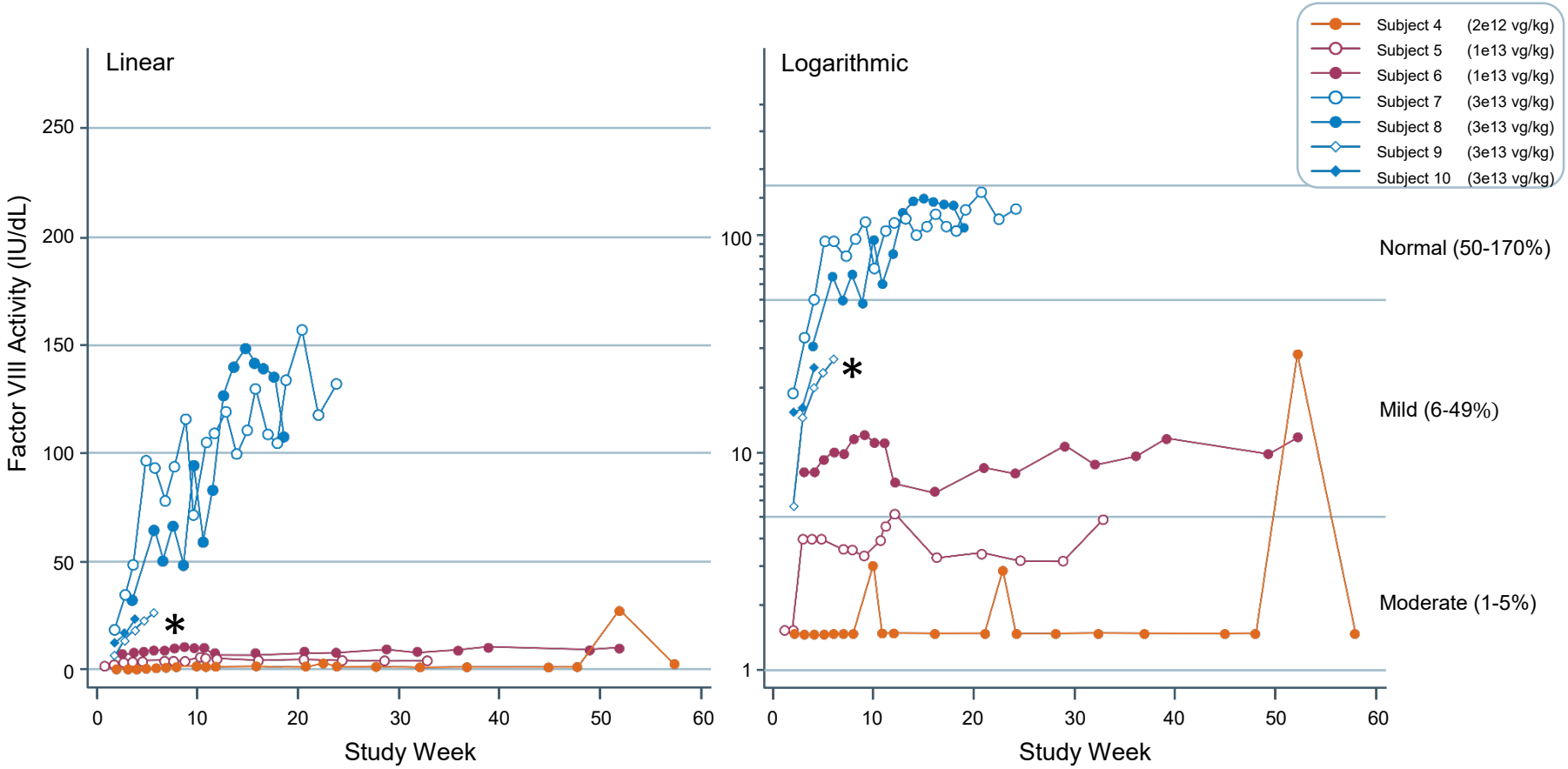
Successes in Haemophilia Gene Therapy

- Haemophilia B gene therapy may provide stable FIX levels for >8 years
- Now, both for haemophilia A and haemophilia B initial responses are being achieved to within or near normal factor levels
- Minimal short-term toxicity to date
- Patients with marked decrease in bleeding and use of factor replacement therapy
- Patients report feeling normal

This slide contains information about a product that has not been approved by the Therapeutic Goods Administration.

Nathwani AC, et al. *N Engl J Med*. 2014;371:21. https://hemophilianewstoday.com/2019/04/03/sb-525-shows-promise-hemophilia-a-phase-1-2-trial (Accessed June 2019)

Sangamo Phase I/II Trial: Factor VIII activity



* Subsequent to the data cut used for the ISTH presentation, Subject 9 attained normal levels at week 7

Questions in Haemophilia Gene Therapy

- Why is there such variability in expression between subjects?
  - Role of vector capsid, vector production, host immune repertoire, transgene construct, etc?
- Which factor assay methodology is relevant to bleeding risk?
  - One stage versus chromogenic
- Will factor activity levels be sustained?
  - Will that be different for haemophilia A and B?
  - Does site of vector transfection make a difference?

- If not sustained, will re-dosing be feasible?
- Can manufacturing be scaled up for commercialization?
- When will approaches allow gene therapy in young children and other populations, not included today?
- Can we modulate known and unknown risks of therapy?
- What will it cost and how will it be paid for?

Risks with AAV Gene Therapy

Some knowns

- Short-term liver toxicity
- Development of anti-AAV antibodies
- Wide inter-individual expression
  - Partially explained by anti-capsid T-cell immune response

Some unknowns

- Long-term liver toxicity
  - Impact of prior HCV infection unknown
- Risk of insertional mutagenesis
  - AAV integration estimated at 0.1–1%
    - Becomes real risk with current number of viral genomes infused
- Germline transfer
  - Animal models do not demonstrate AAV-infection of germ cells
  - In human studies vector has cleared from semen

AAV: Adeno-associated vrisu; HCV: Hepatitis C virus.

Pierce GF, Iorio A. *Haemophilia* 2018;24(Suppl.6):60. Nathwani AC, et al. *Hematol Oncol Clin N Am* 2017;31:853. Colella P, et al. *Mol Ther Methods Clin Devel* 2018;8:87. Perrin GQ, et al. *Blood* 2019;133:407.

Looking to the Future: My View

- Gene therapy will successfully decrease bleeding and factor consumption
- Some patients may not need factor infusion post-gene therapy
- Sustainability may depend on vector, achieved level and site expressed
- There will be gradual uptake in the community
- New approaches, including new vectors, will allow treatment and re-treatment of children and other patient groups
- An option for low-resource countries

Speaker's personal opinion.

That being said….we proceed with caution

- Ethical Issues
  - Consent for potential long-term unknown risks
  - Many patients excited about possibility of cure
    - How to be sure patient understands risks
    - Consent is a process
      - Current trials with initial observation period before vector infusion
  - What risks are acceptable when standard of care is very good?
  - In current trials with AAV
    - No or loss of response prevents re-dosing
      - In hemophilia can revert to prior therapy
  - How will price influence access?