Response Evaluation In Neurofibromatosis Schwannomatosis
INTERNATIONAL COLLABORATION

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Gene Therapy Approaches for the Leukodystrophies

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REINS winter meeting lecture

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Response Evaluation In Neurofibromatosis Schwannomatosis
INTERNATIONAL COLLABORATION

Disclosures

- PI of ex vivo lentiviral gene therapy trial in cerebral adrenoleukodystrophy sponsored by bluebird bio
- Site-PI of Minoryx trial of leriglitazone for adrenomyeloneuropathy
- Consultant to Autobahn, Poxel, Takeda, Therapeutics, SwanBio Therapeutics, UpToDate and Taysha Gene Therapies
- Founder of SwanBio Therapeutics

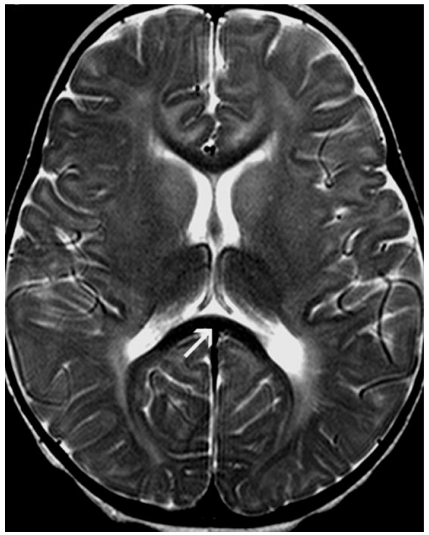
Definition of Leukodystrophies

- hereditary
- impair normal brain
- affect brain myelin throughout life
- commonly fatal

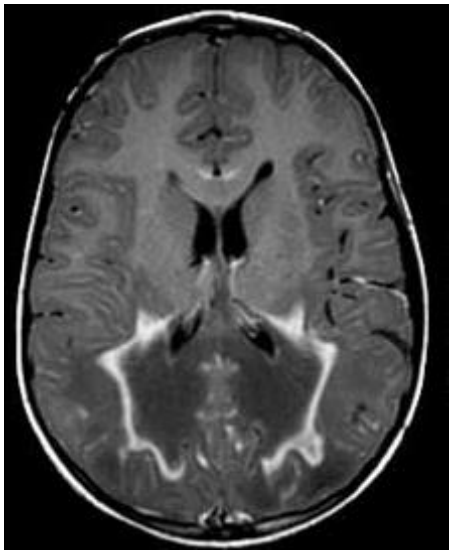
progressive: cognitive deterioration
neuropsychiatric difficulties (substance abuse not uncommon)
pyramidal and cerebellar abnormalities
visual abnormalities

dementia and death within a few years

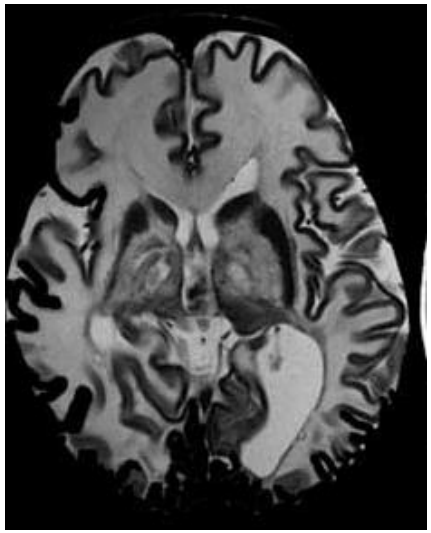
Hypomyelination (GM2)



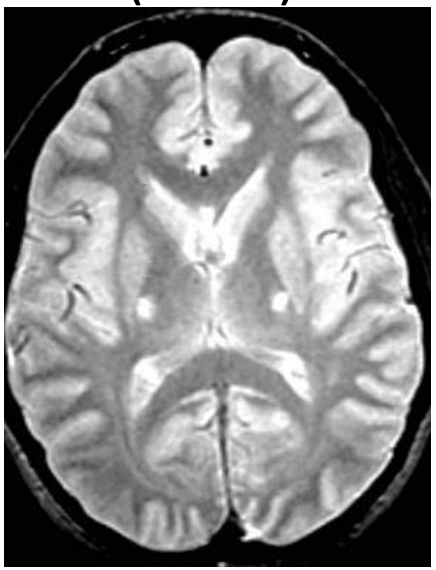
Inflammatory demyelination (CALD)



Spongiform Encephalopathy (Canavan)



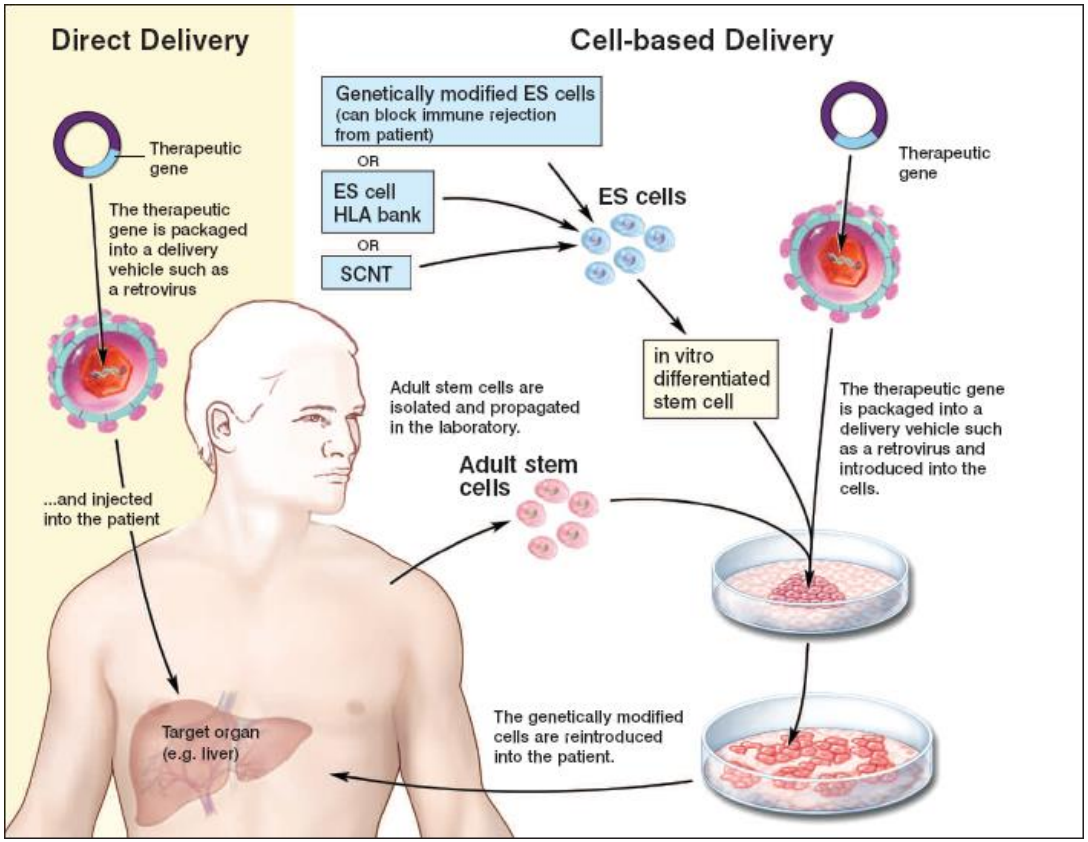
Hereditary Spastic Paraplegia (AMN)



Approaches to gene therapy

In vivo
gene therapy

AAV-mediated



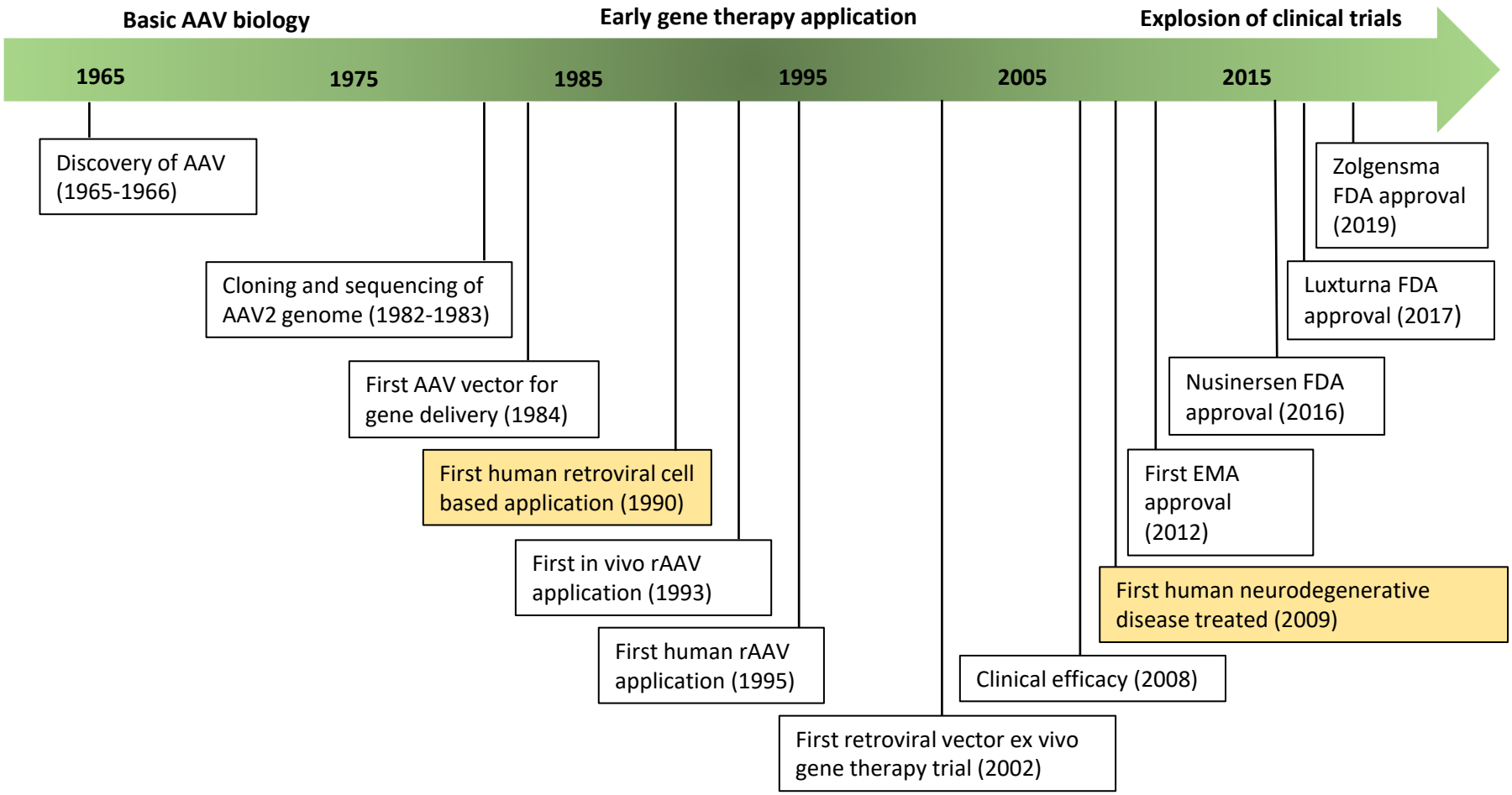
Ex vivo
gene therapy

Lentivirus-mediated

Antisense Oligonucleotides (ASOs)

Nanoparticles, lipoparticles, gene editing

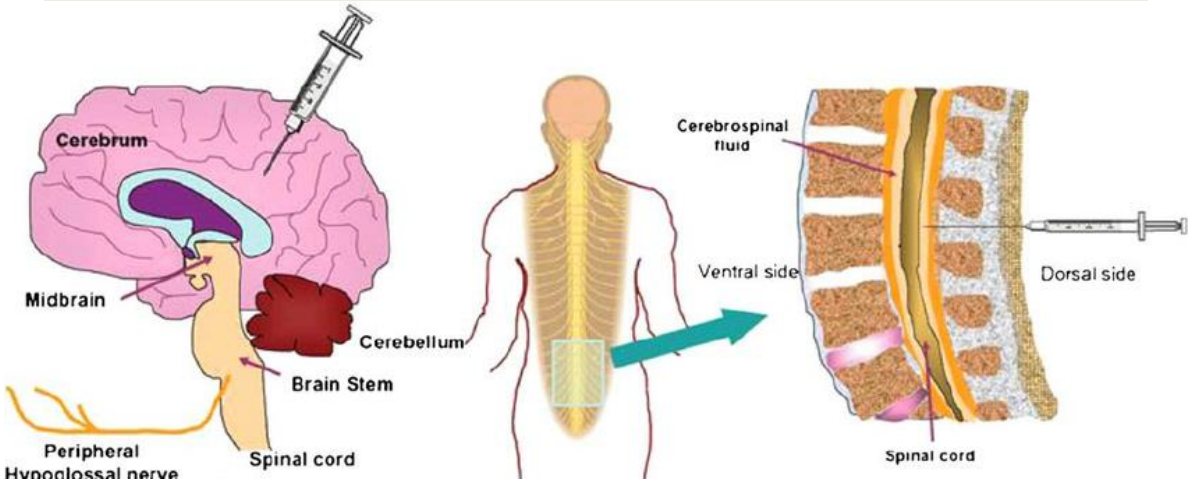
History of Gene Therapy

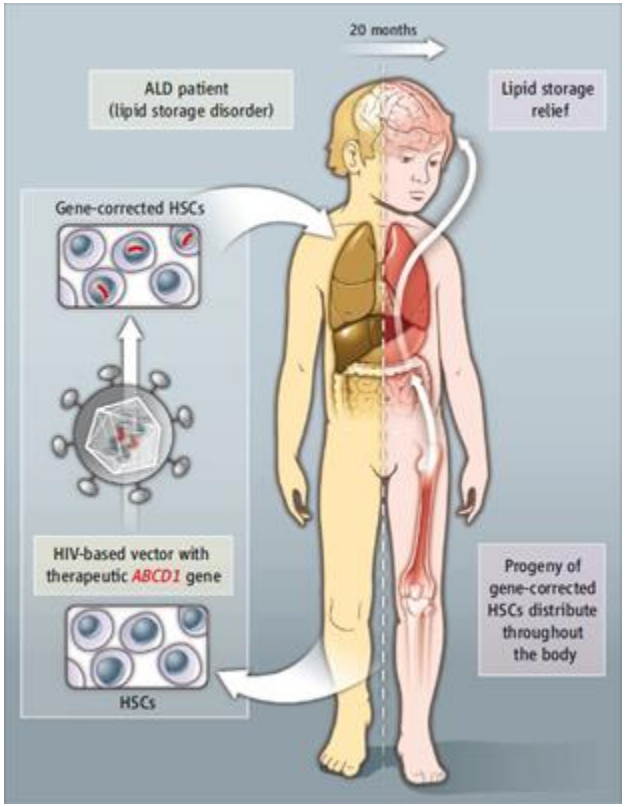


Adapted from "Adeno-associated virus vector as a platform for gene therapy delivery" by D. Wang et al., 2019

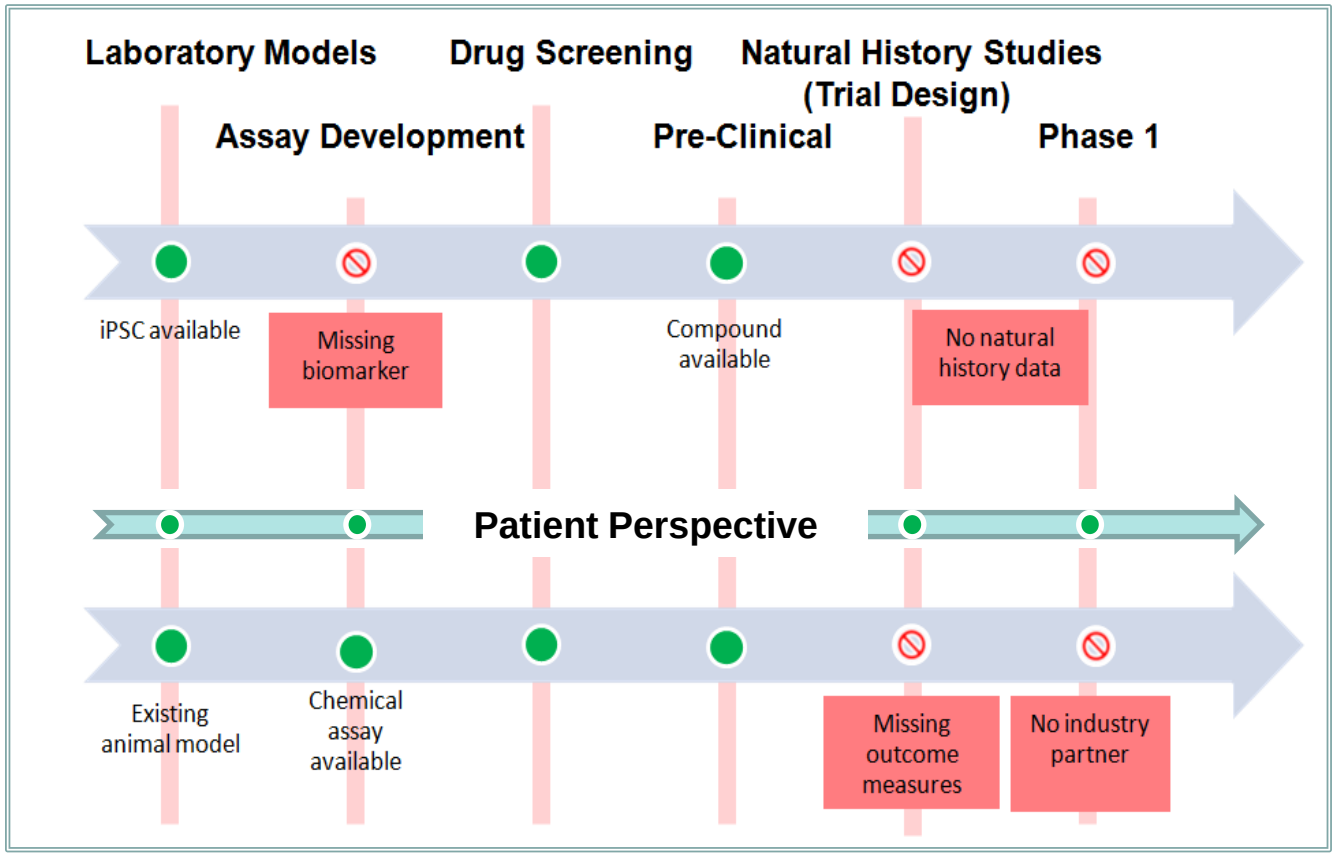
IN VIVO versus EX VIVO delivery for CNS disease

Adeno&associated,virus,(AAV)	Lentiviral (LVs)
• 4.7kbnon)enveloped$single)stranded$ **DNA** parvoviruses	• 8kbenveloped$single)stranded$**RNA** viruses



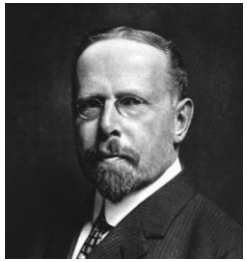


Center for Rare Neurological Diseases addresses gaps

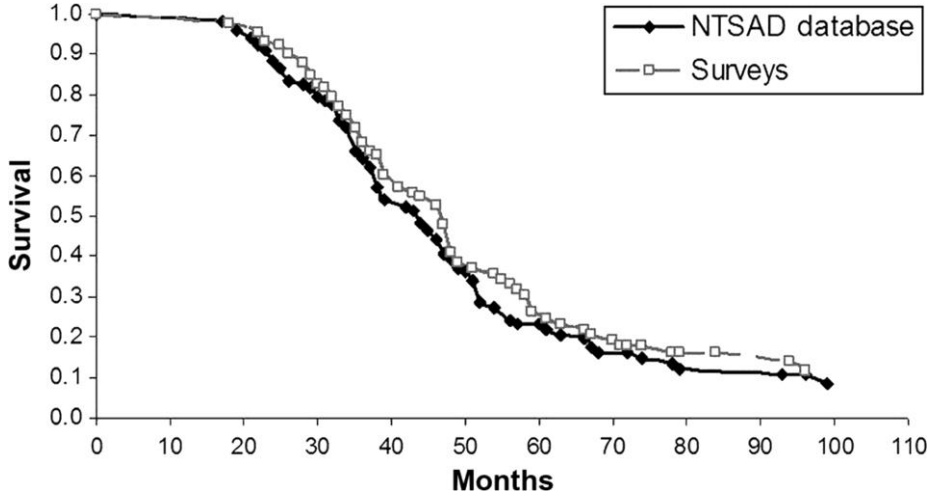


Infantile Tay Sachs and Sandhoff Disease (GM2)

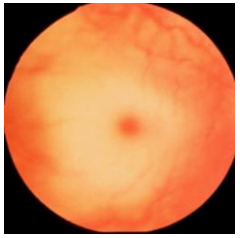
Warren Tay 1881

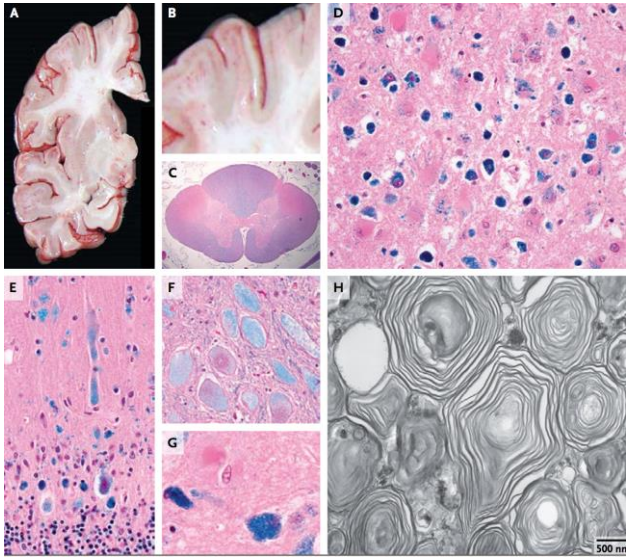


Bernard Sachs 1887

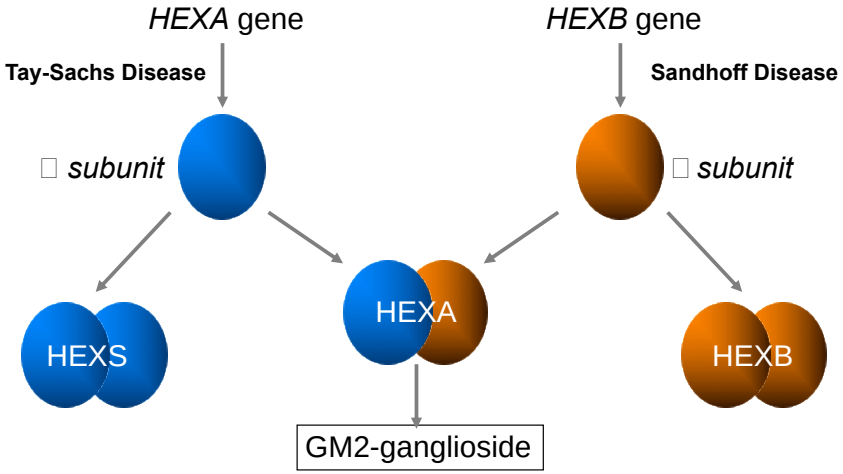


Bley et al, Pediatrics 2011



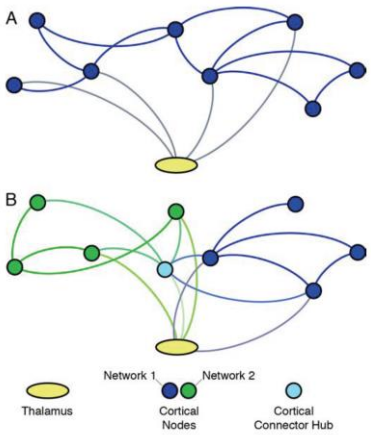


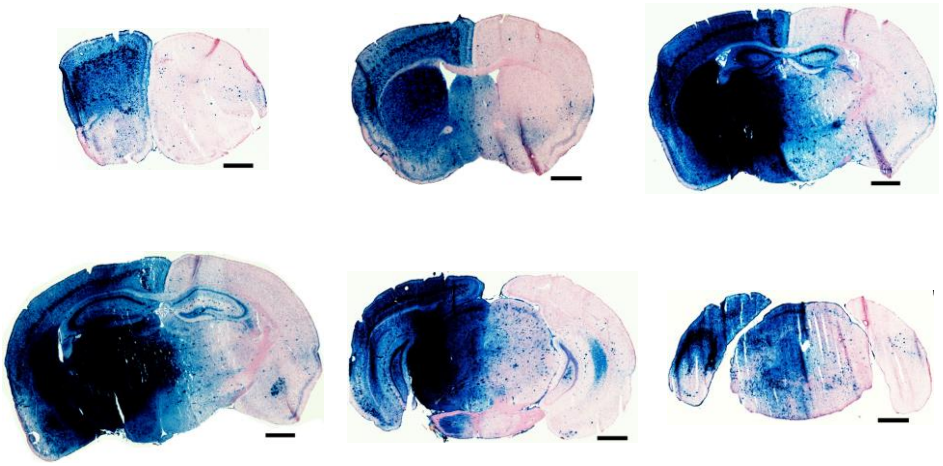
Krishnamoorthy et al, NEJM 2014

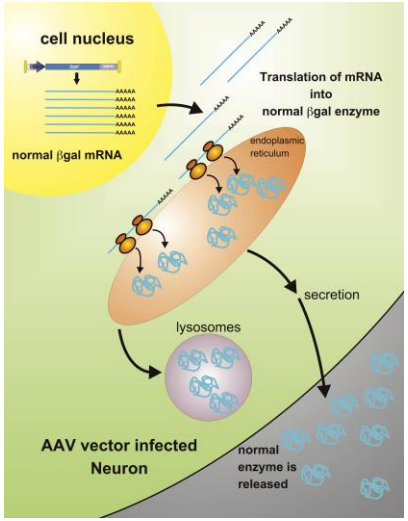


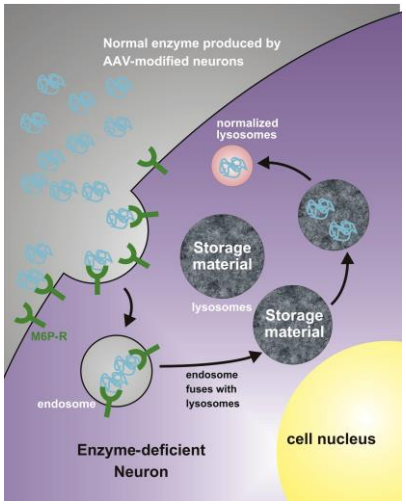
Rationale for thalamic AAV-GM2 delivery (AAVrh8-HEXA and AAVrh8-HEXB)

Esteves









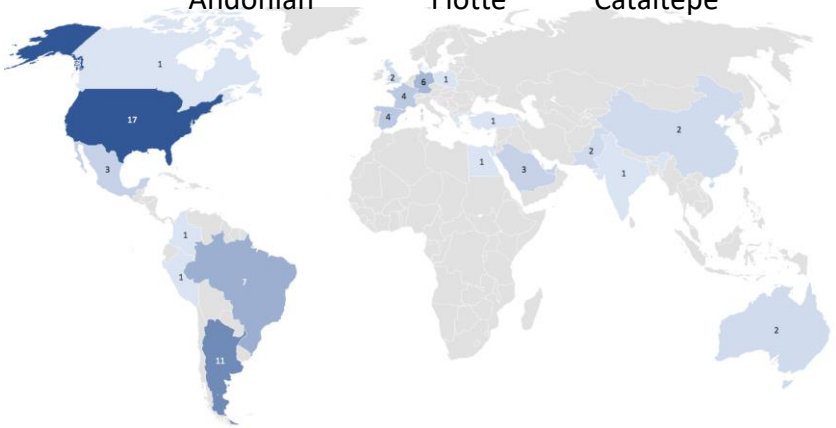
Human thalamus is integrative hub for functional brain networks; potential for lysosomal enzymes to achieve cross correction of neighboring cells

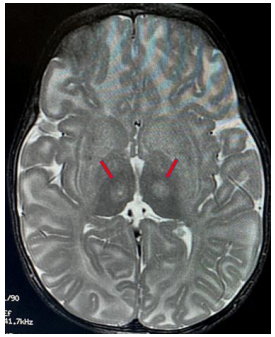
Worldwide recruitment for AAV-GM2 delivery to the brain and spinal cord

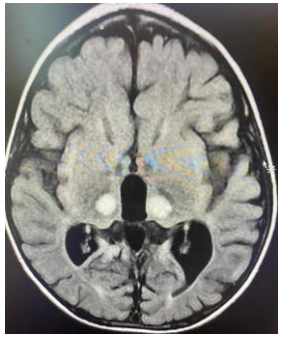
Andonian

Flotte

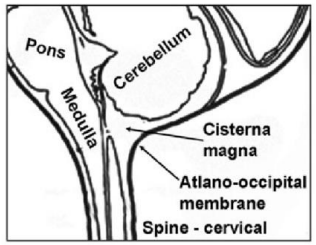
Cataltepe

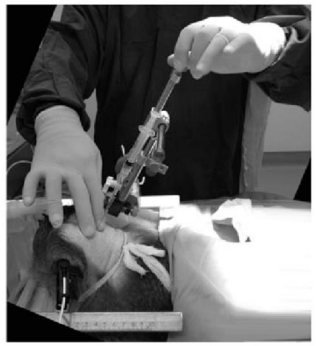


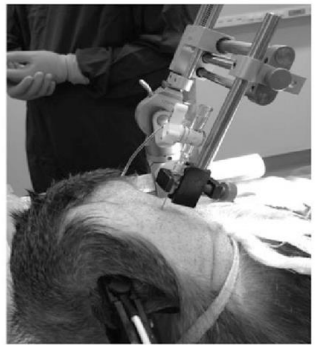


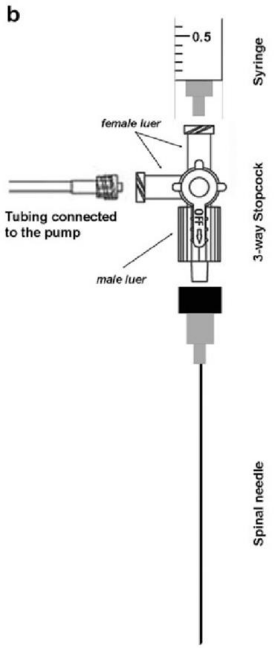


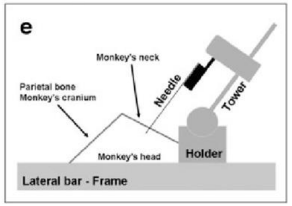
Patient 1 (SD) Patient 2 (TSD)



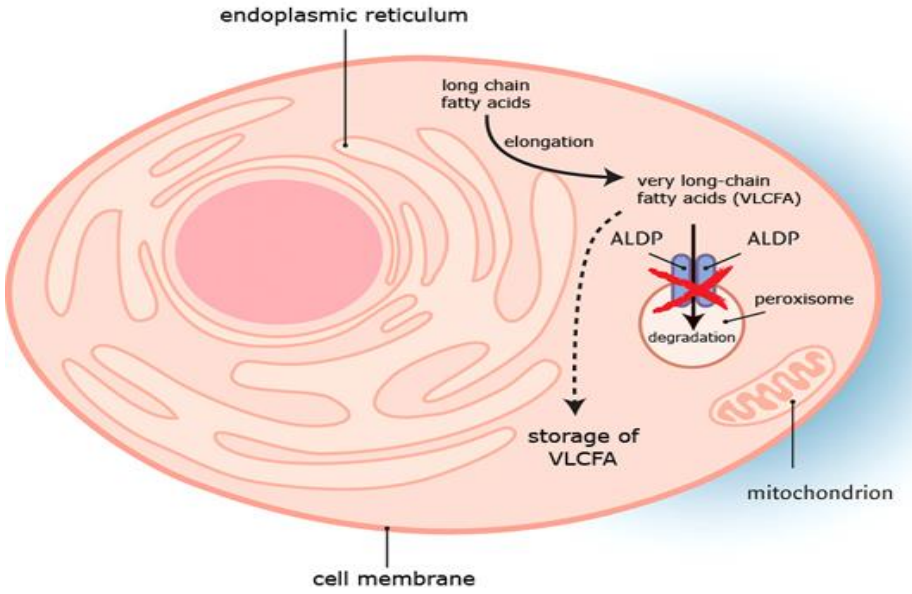








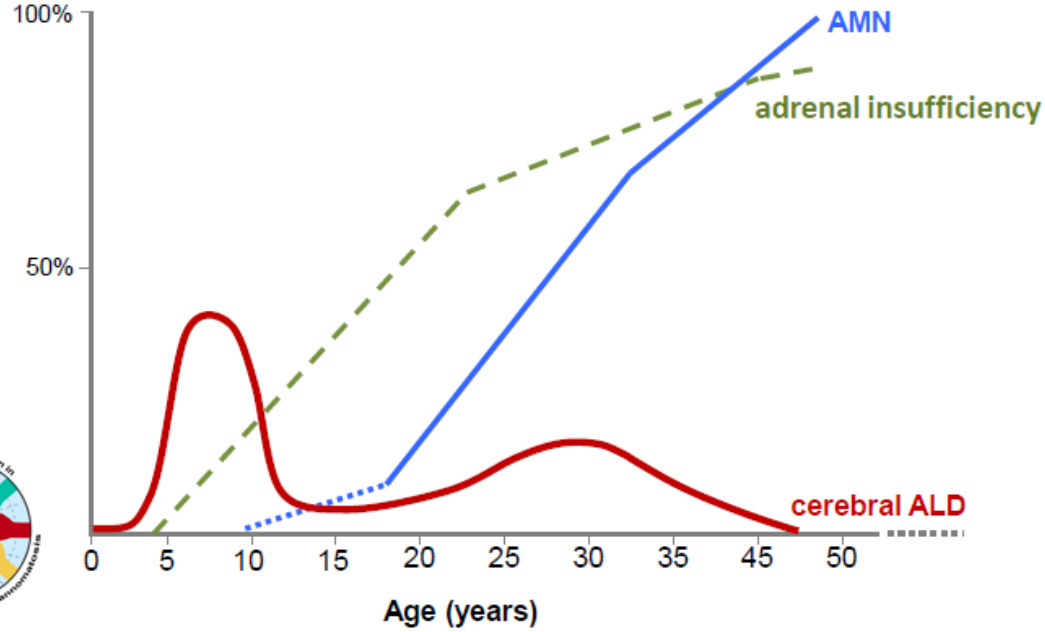
X-linked Adrenoleukodystrophy (X-ALD)

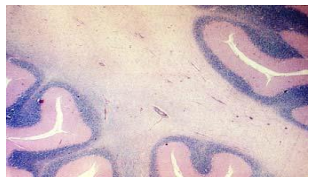


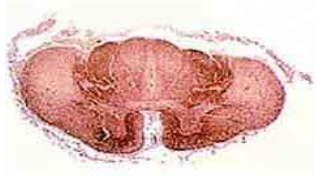
Adrenal dysfunction

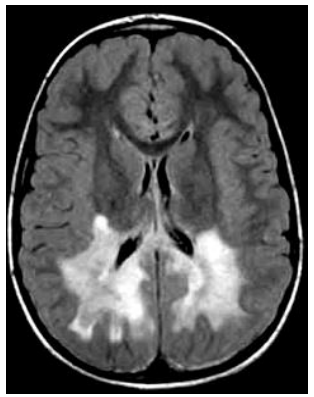
Acute brain inflammatory demyelination

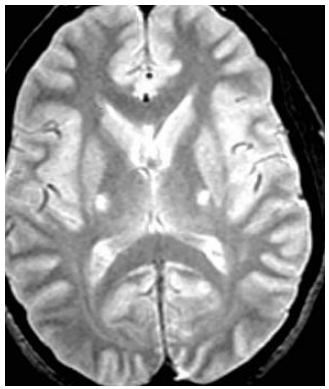
Chronic spinal cord axonal degeneration





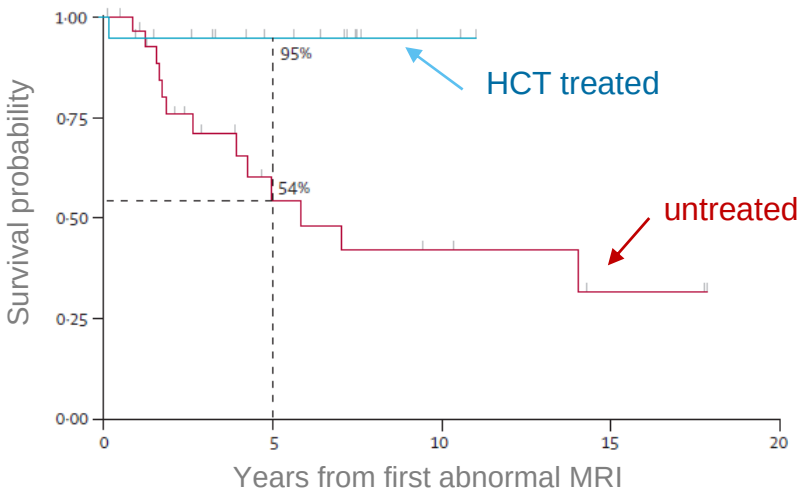






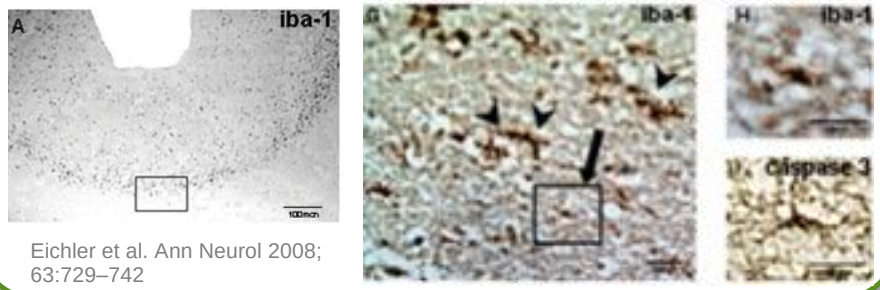
Allogeneic bone marrow transplant (BMT) to treat a cerebral adrenoleukodystrophy (ALD)

BMT with donor cells dramatically improves survival



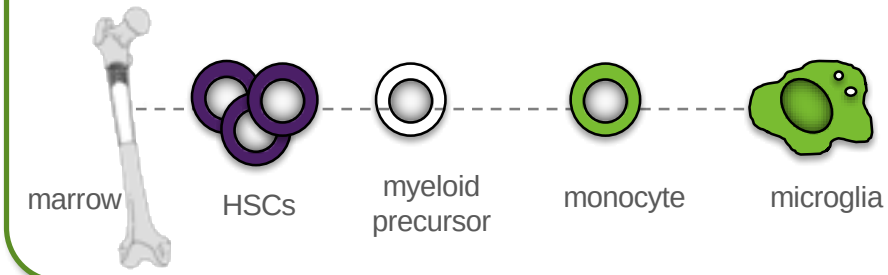
Mahmood et al. Lancet Neurol 2007; 6: 687–92

Microglial apoptosis in perilesional white matter of CALD

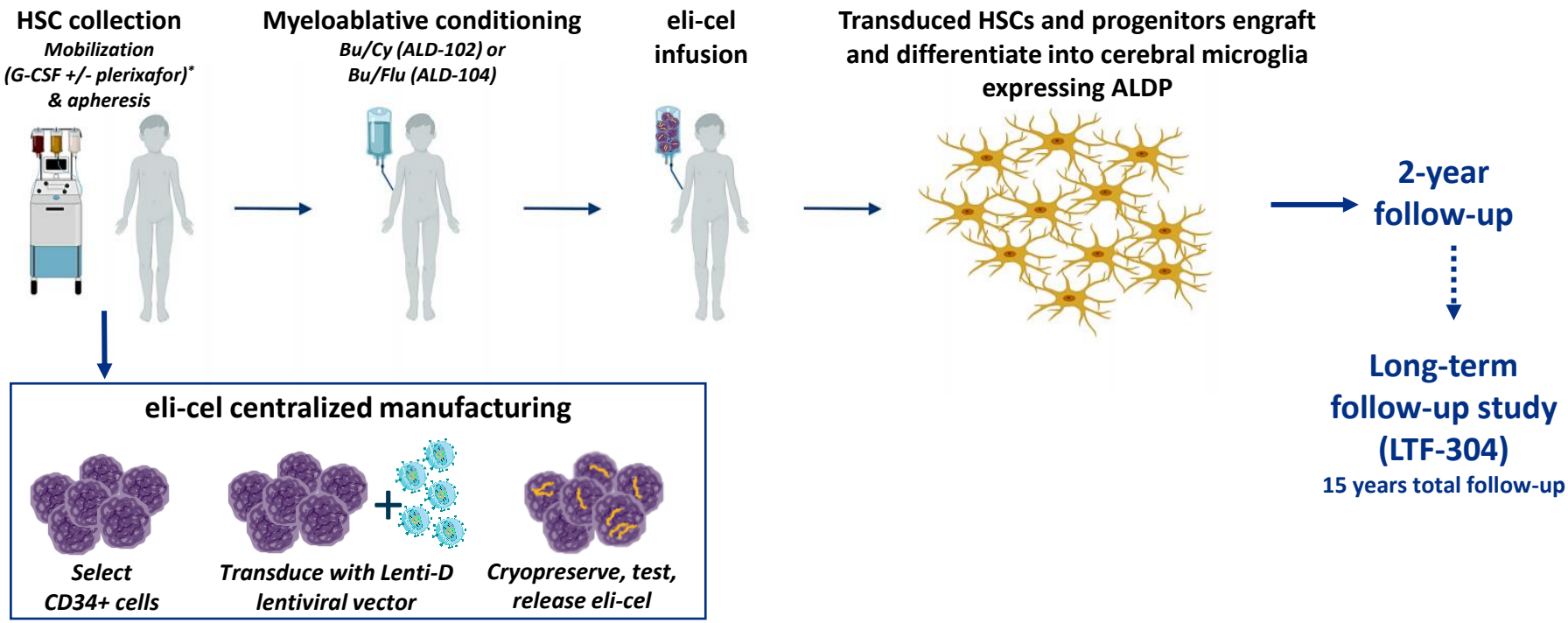


Eichler et al. Ann Neurol 2008; 63:729–742

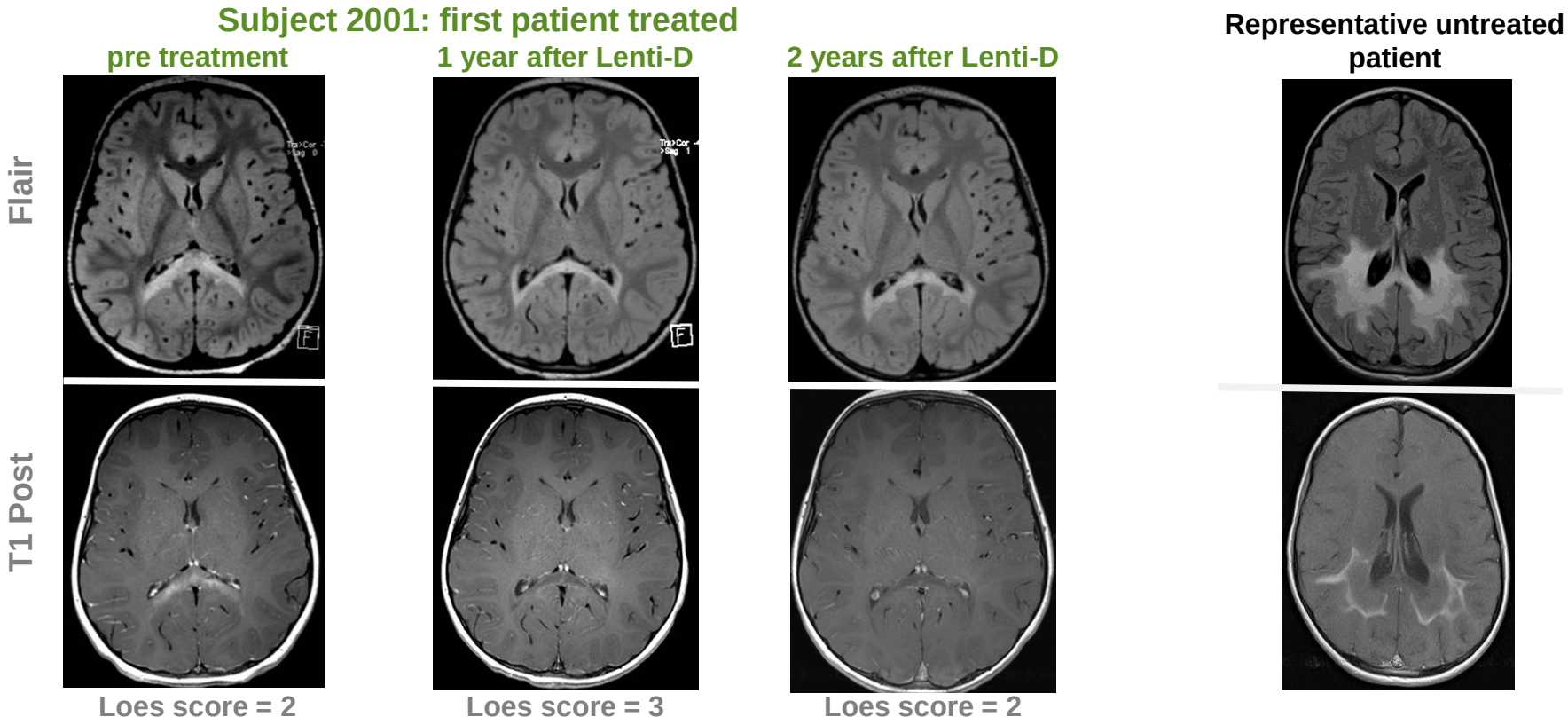
Bone-marrow derived monocytes enter CNS, differentiate into microglia expressing normal ALDP



Elivaldogene autotemcel (eli-cel) Gene Therapy: 2 Single-Arm Clinical Trials for Cerebral ALD



Neuroimaging outcomes demonstrate halting of cerebral ALD progression after Lenti-D treatment



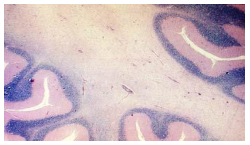
Ex vivo gene therapy for X-ALD

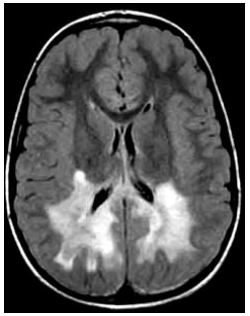
Milestones:
First trials of single gene addition in cerebral ALD
Encouraging efficacy data (stabilization)
Reassuring safety profile (no engraftment issues / no GvHD) but MDS

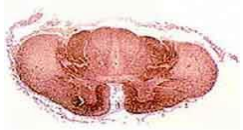
Limitations:
Delays in engraftment “time = brain”
Adverse events consistent with myeloablation

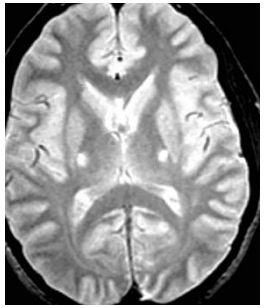
HSCT X-ALD patients also develop AMN (Van Geel et al, 2015)
AMN requires broad delivery to the entire spinal cord/peripheral nerve

Cerebral ALD (CALD)







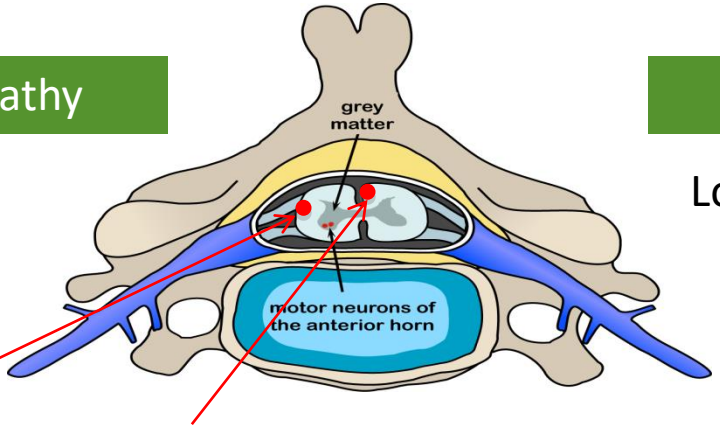


Adrenomyeloneuropathy (AMN)

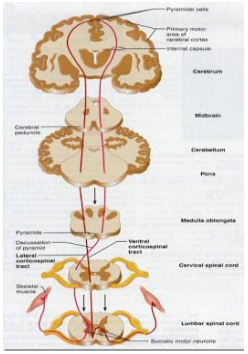
AAV to target neurodegeneration in the spinal cord

Adrenomyeloneuropathy

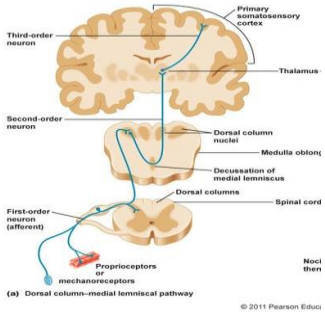
Dorsal root ganglion
Astrocytes
Microglia
Endothelial cells



Corticospinal tract



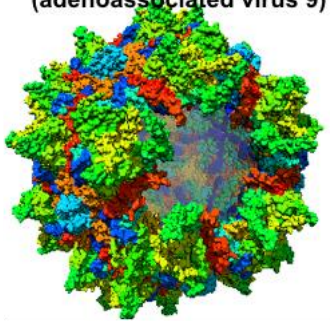
Dorsal columns

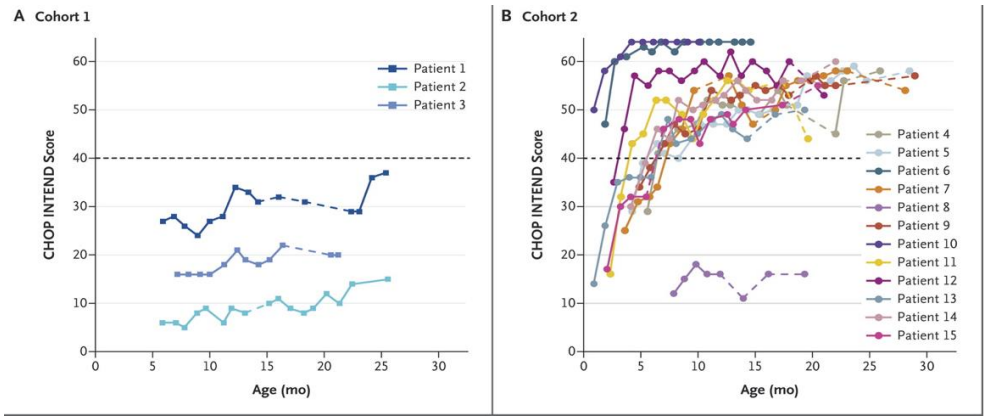


Spinal Muscular Atrophy

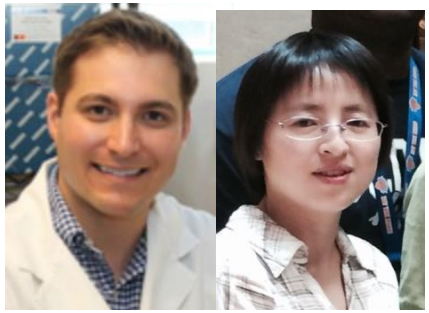
Lower motor neuron

AAV9
(adenoassociated virus 9)





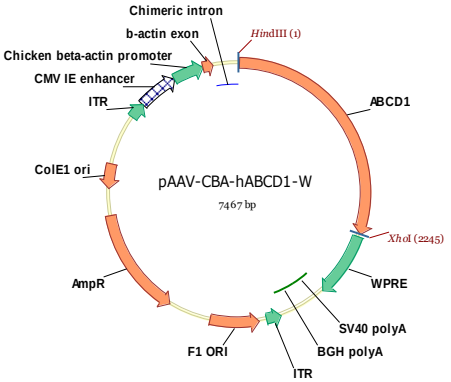
Spinal cord can be targeted by AAV9-mediated gene delivery



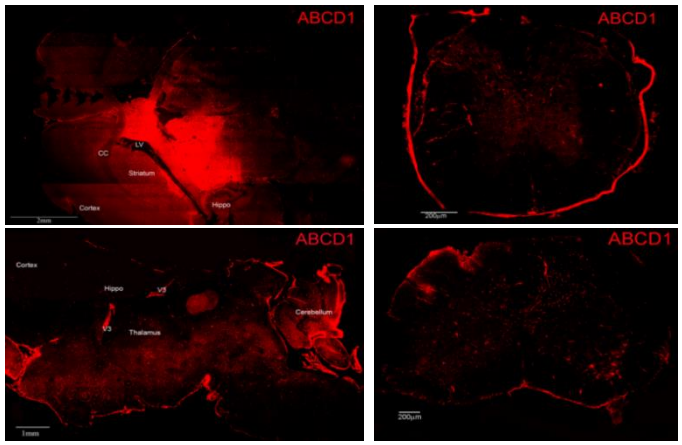
Maguire Gong

ICV

IV

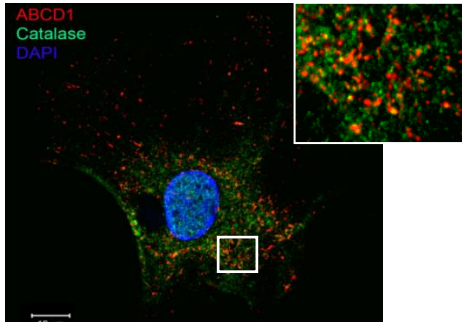


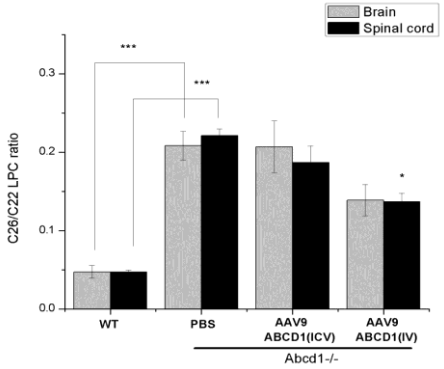
Brain Spinal cord



Gong et al. 2015 Molecular Therapy

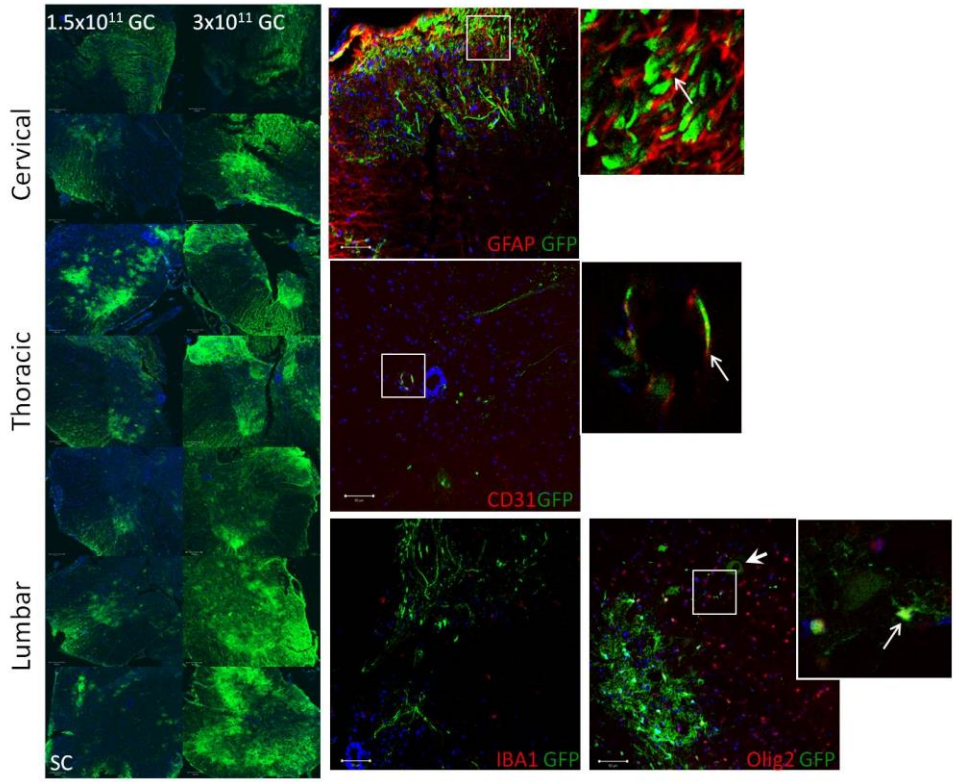
Peroxisome Localization

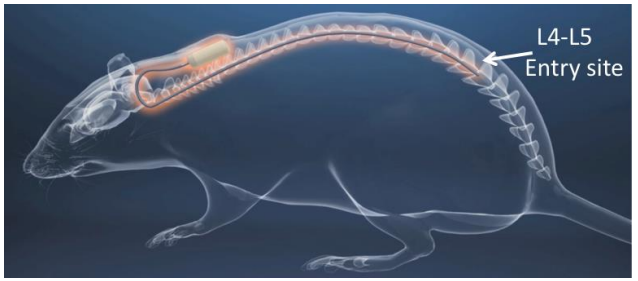


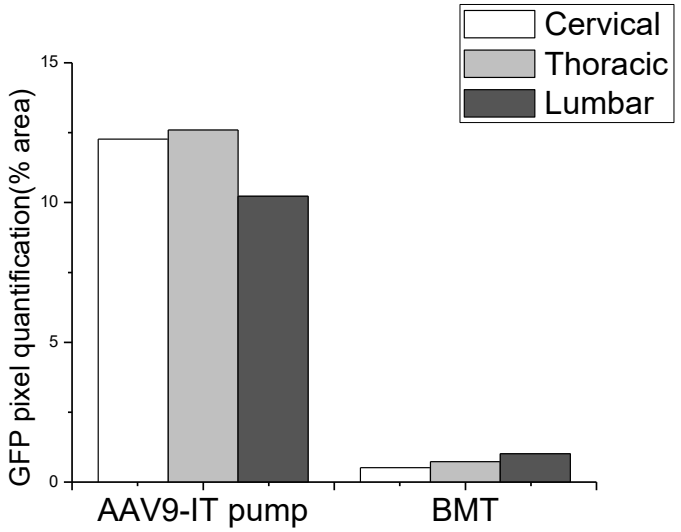


- ABCD1 delivered to CNS via both ICV and IV
- In vitro, into correct intracellular compartment (peroxisome)
- Lead to functional VLCFA degradation

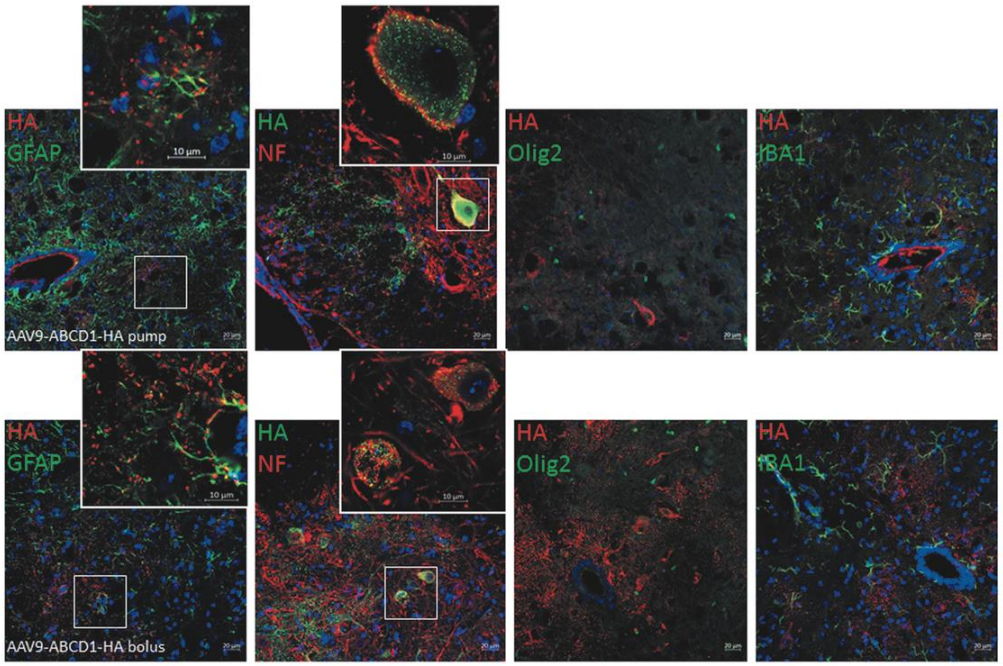
Intrathecal osmotic pump of AAV9 delivers more GFP to the spinal cord than bone marrow transplantation



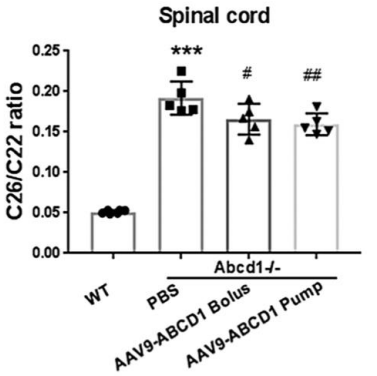


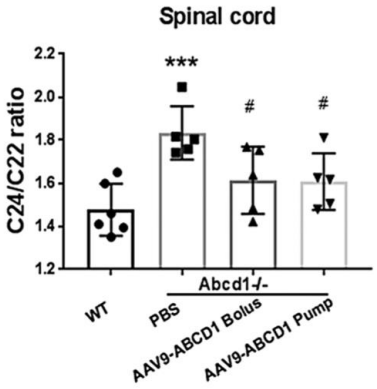


hABCD1 protein expression after intrathecal pump delivery for mouse model of adrenomyeloneuropathy (AMN)



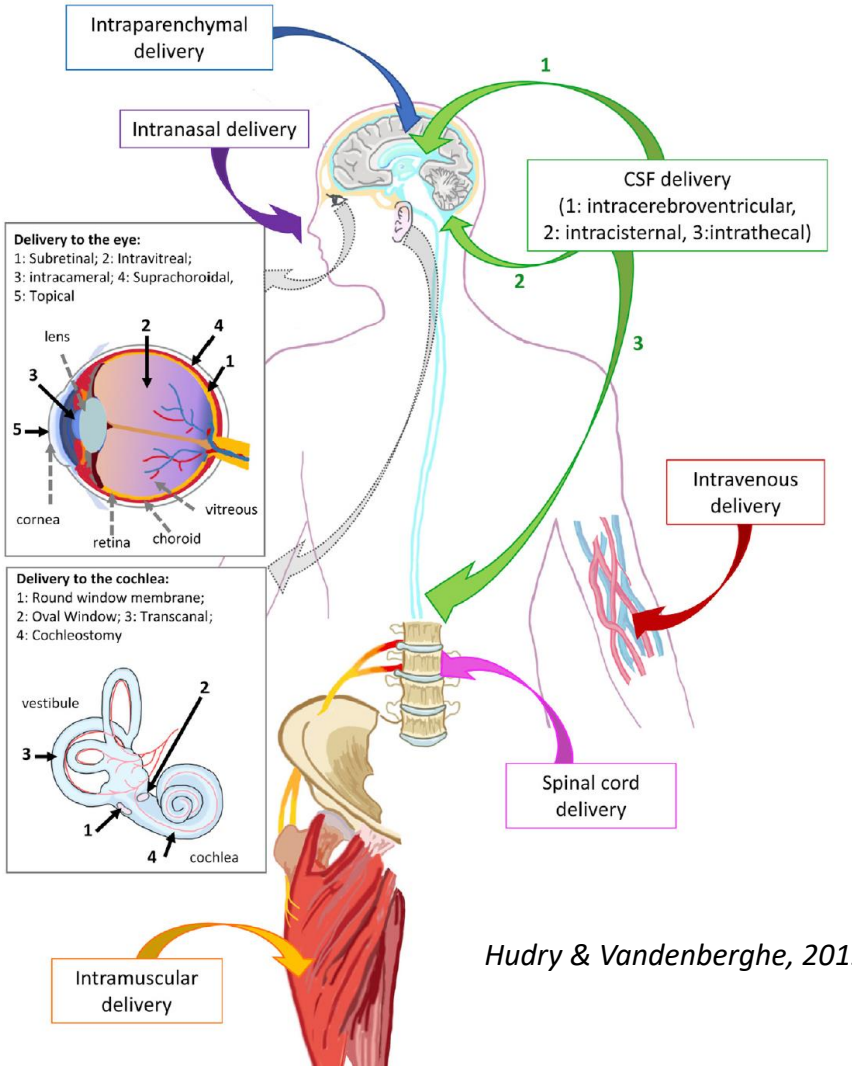
Gong et al, Hum Gene Ther 2018





In Vivo Routes of Delivery to Nervous System in MGH trials

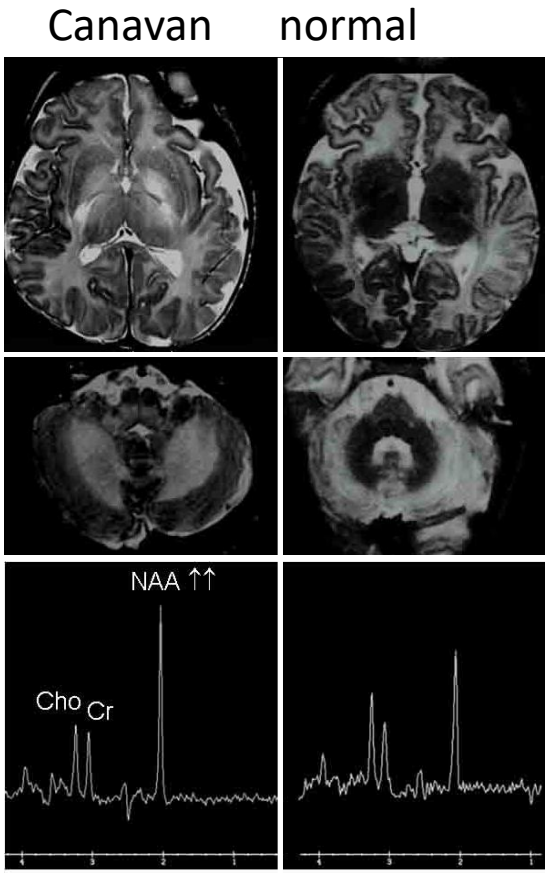
- Bilateral intrathalamic
- Intraventricular
- Lumbar intrathecal
- Intraventricular
- Intravenous



Hudry & Vandenberghe, 2019

Canavan Disease (CD)

- increasing head size
- not irritable
- poor head control, hypotonia
- nystagmus
- motor delay: unable to sit up
  but some can reach for objects

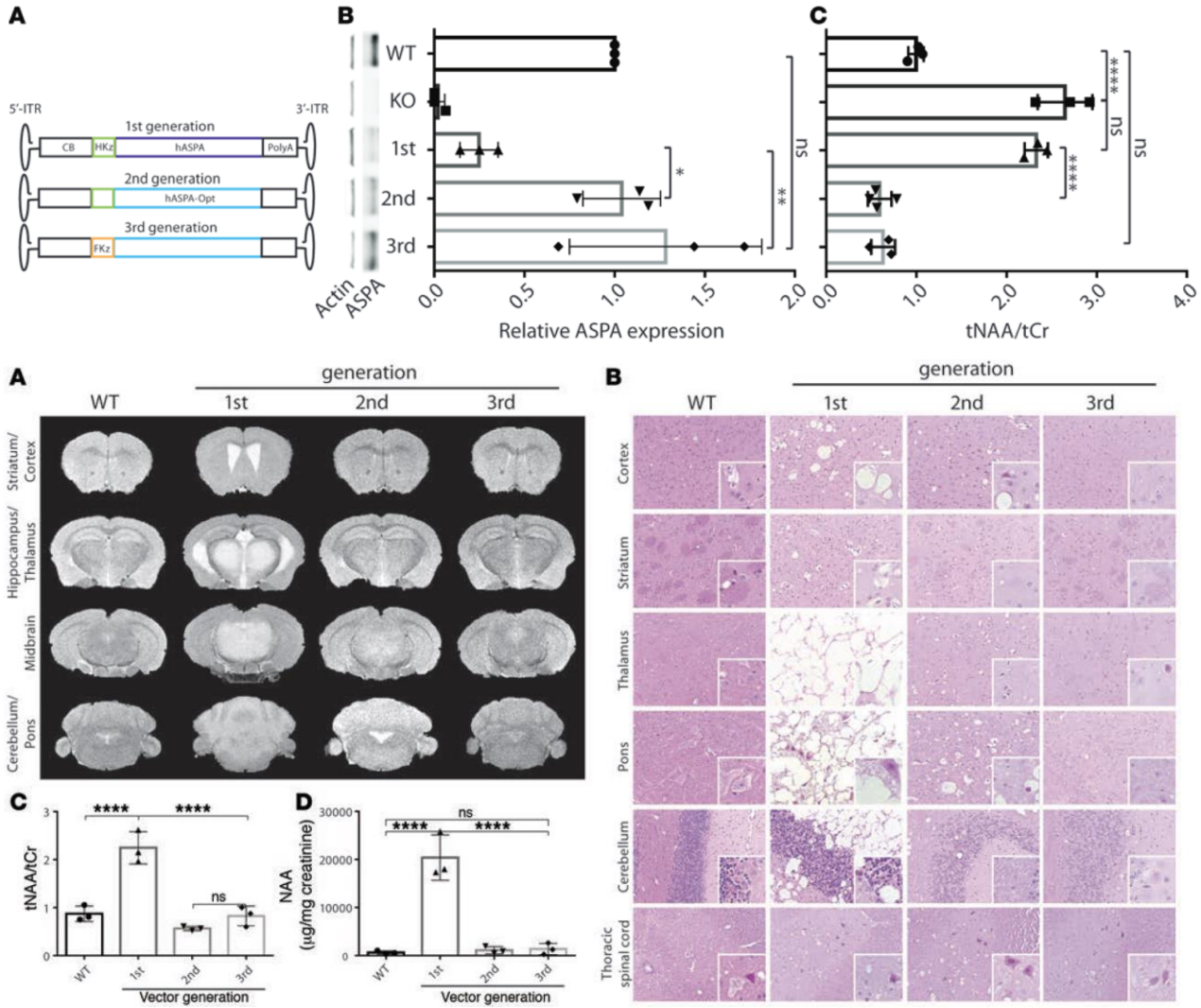


autosomal recessive, mutations

in aspartoacylase gene leading to

N-acetylaspartate (NAA) accumulation

Intravenous AAV9-ASPA rescues mouse model of Canavan disease

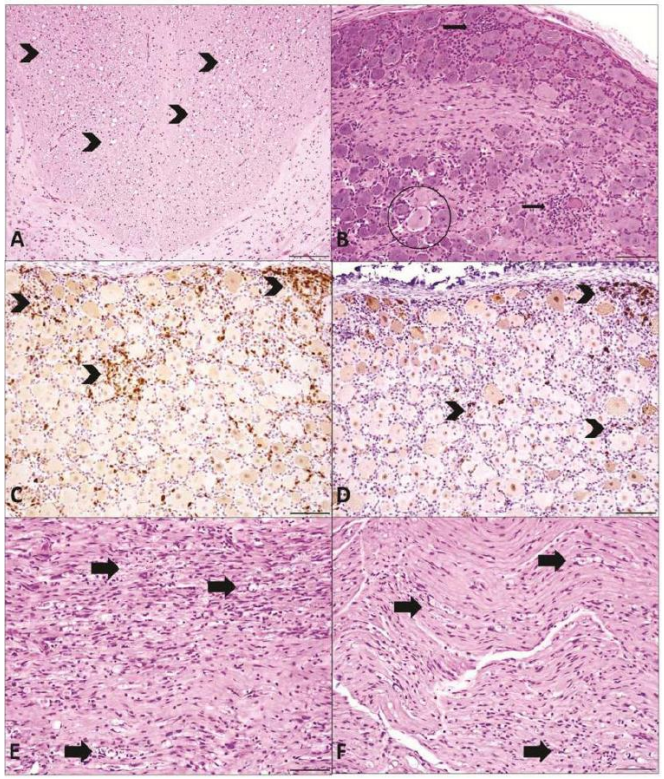


Gessler et al, JCI Insight 2017

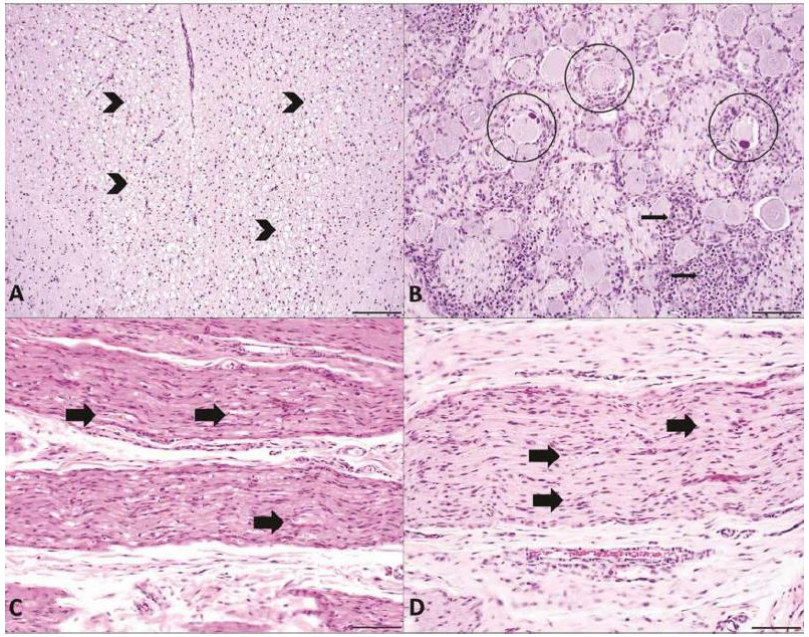
Potential Toxicities

- In vivo AAV-mediated gene therapy
- Ex vivo lentiviral gene therapy

Toxicity of high dose AAV9 variant expressing human SMN in NHP and piglets



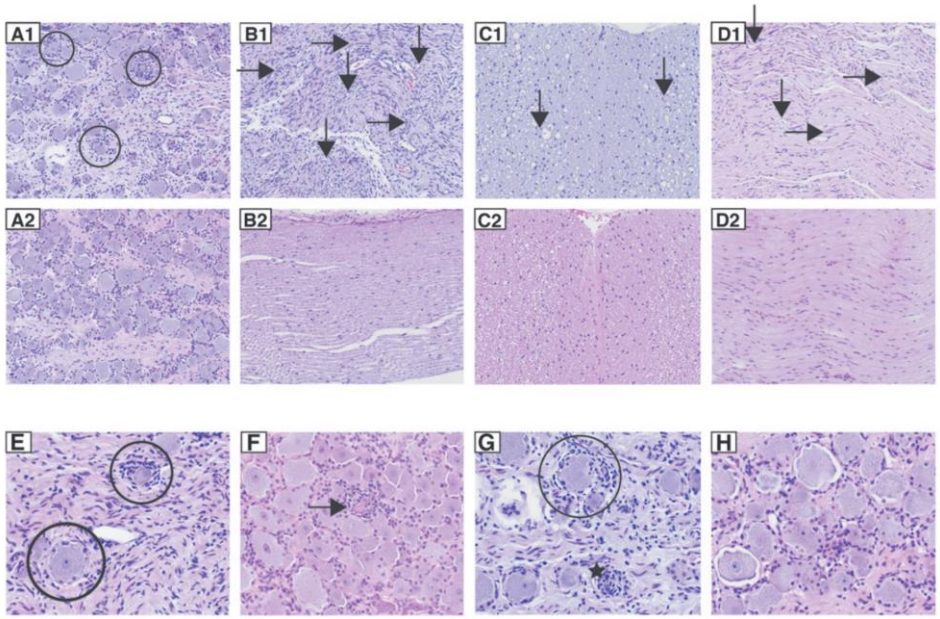
Liver histopathologic findings

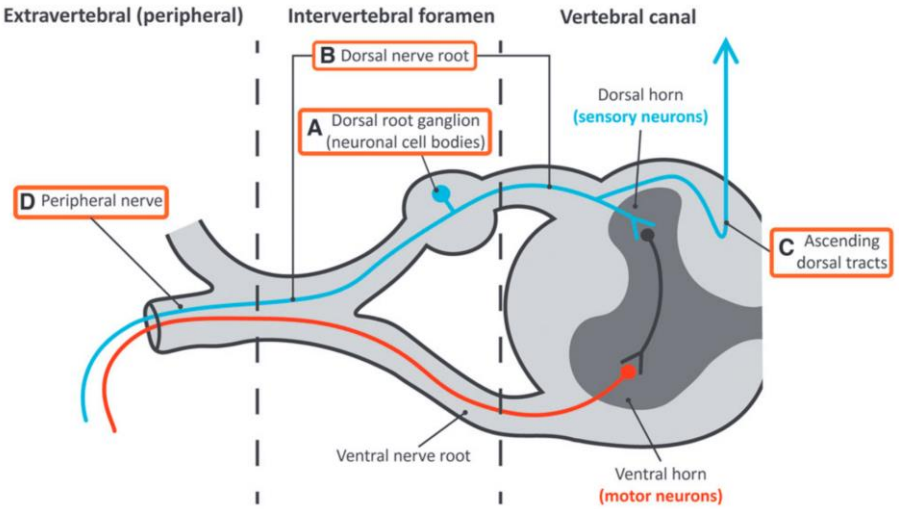


Dorsal root neuron ganglion degeneration

Hinderer et al, Hum Gene Ther 2018

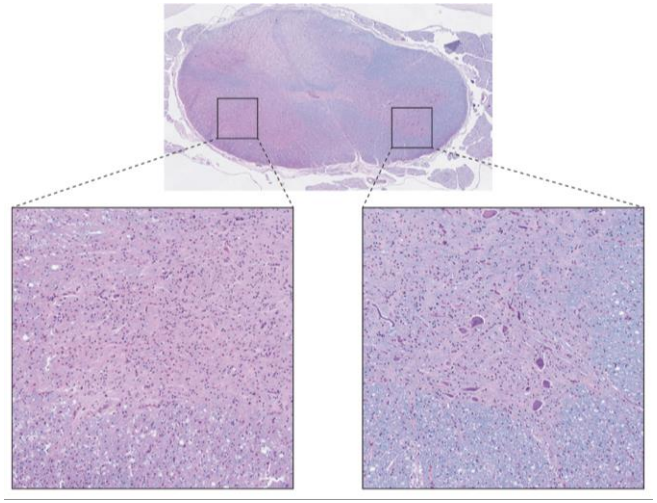
AAV-induced dorsal root ganglion pathology in NHP

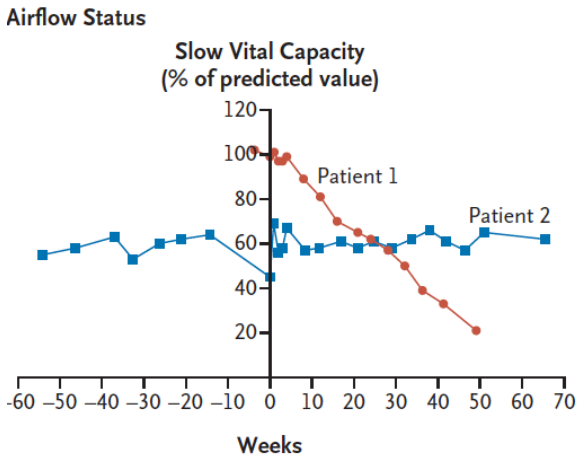


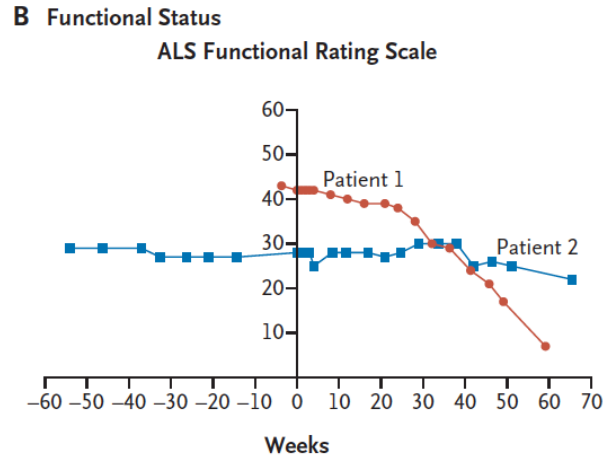


Important to separate from disease related DRG pathology

Immunomodulation makes a difference in SMA and ALS, alleviates potential DRG toxicity







Mueller et al, NEJM 2020

intrathecal infusion of an adeno-associated virus rh10 containing anti-SOD1 microRNA (AAV-miR-SOD1), patient 1 without and patient 2 with immunosuppression

Potential toxicities – ex vivo lentiviral HSC gene therapy

- myeloablation related events
  - transient nausea, vomiting, infections, fever,

- insertional mutagenesis
  - in retroviral gene therapy trials: leukemia (eg SCID - severe combined immune deficiency)
  - recent events in lentiviral gene therapy: myelodysplastic syndrome

Conclusion

1. Encouraging **efficacy** data of first GT trial in ALD
   - Lenti-D gene therapy may offer an alternative to allo-HSCT in patients with early cerebral disease, particularly for patients with no matched sibling donor
2. Specific **phenotypes** within an individual leukodystrophy require **different approaches** (ex vivo versus in vivo GT)
3. The **timing of intervention** is critical:
   - When early inflammation visible on brain MRI
   - Before lesion too extensive

Conclusion

1. Within each phenotype the **target structures and cells are critical:**
   - In brain disease, microglial pathology is prominent and correction of myeloid cells contributes to brain health.
   - In spinal cord disease, no disruption of the BBB is present and AAV-mediated gene transfer via intrathecal osmotic pump leads to widespread expression across spinal cord and dorsal root ganglia.
2. Success of gene therapy may depend on understanding of **network hubs such as the thalamus and dorsal root ganglia,** allowing biodistribution and connectivity
3. Route and overall approach to gene therapy **delivery** makes a difference – **technologies evolving** and need to be **matched to biology**

MGB Neurogenetics and Gene Therapy Fellowship

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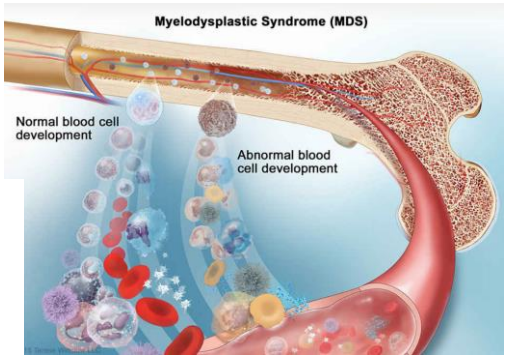
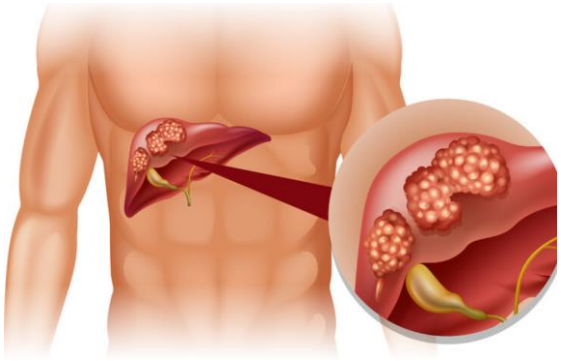
Program Director:
Dr. Florian Eichler

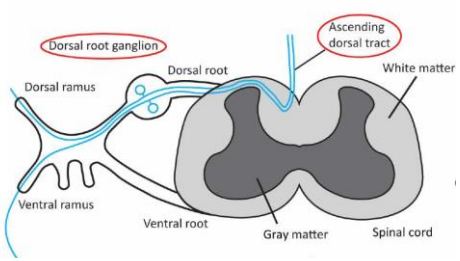
Associate Program Director:
Dr. Vikram Khurana

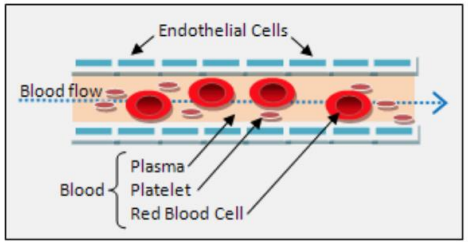
Training on genomic analysis and approaches to gene therapy (ex vivo and in vivo gene therapy, ASO trials), with broad exposure to technology development, regulatory and safety aspects and trial execution

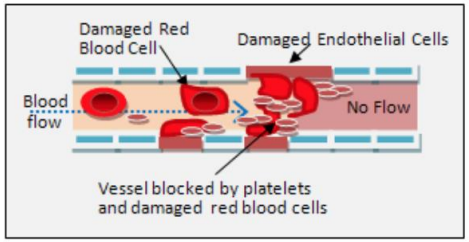
Understanding and Managing Gene Therapy in Neurology











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