FREELINE

Key Opinion Leader Event: FLT201 in Gaucher Disease

August 1, 2023

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Today’s agenda

Opening remarks and corporate overview

Michael Parini, Chief Executive Officer, Freeline Therapeutics

Gaucher disease overview

Dr. Reena Sharma, Consultant Adult Metabolic Medicine and Honorary Senior Lecturer, Salford Royal Hospital, UK

FLT201 overview

Dr. Pamela Foulds, Chief Medical Officer, Freeline Therapeutics

Q&A

Creating better gene therapies for chronic debilitating diseases

Potential first- and best-in-class lead program

Highly differentiated gene therapy candidate FLT201 for Gaucher disease type 1 in first-in-human clinical trial

Innovative research

Lead research program leverages same novel GCase variant as FLT201 for GBA1-linked Parkinson's disease

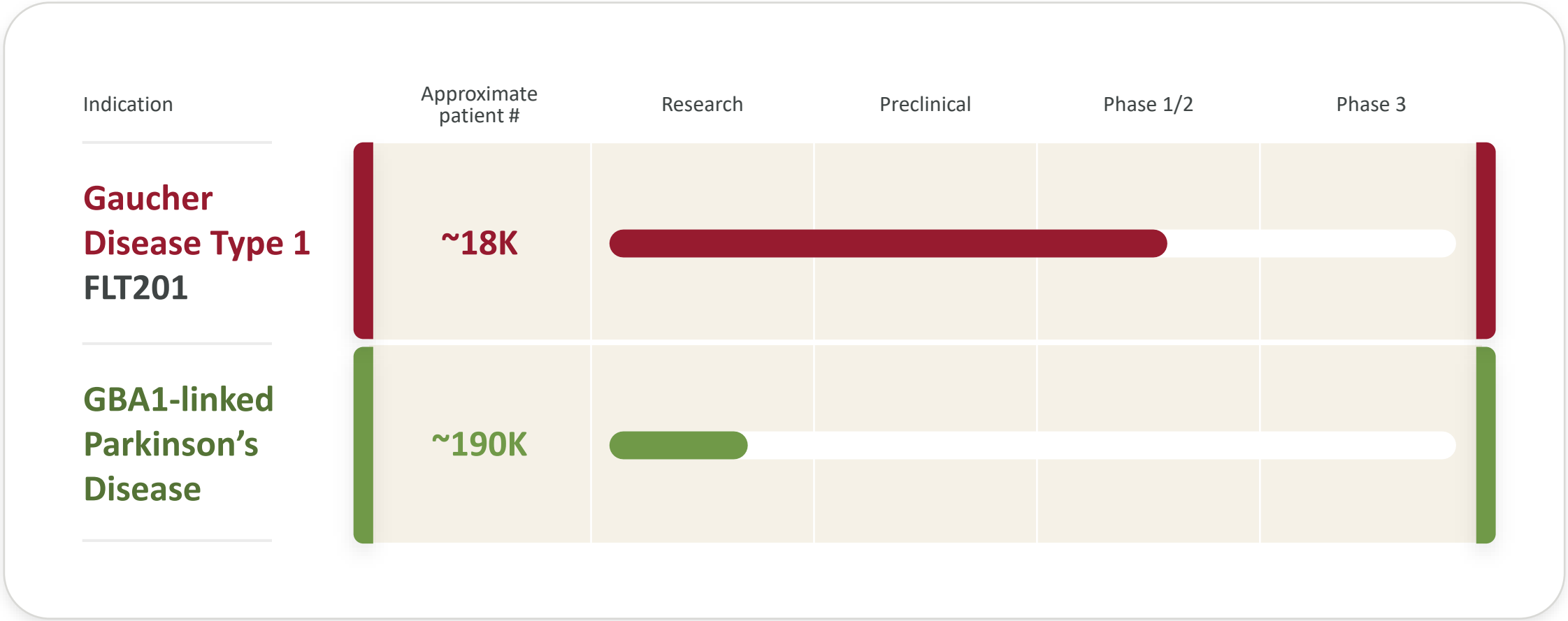
Near-term data readout in Gaucher disease

Initial safety and enzyme activity data for FLT201 expected in Q3 2023

Experienced management team

Seasoned leaders with the experience and expertise to drive progress and execution

Lead clinical program with first- and best-in-class potential with research extending innovation into larger disease area



Estimated patient numbers for Gaucher disease Type 1 are for US, UK, EU4 and Israel (Hematology. 2017 Mar;22(2):65-73. doi: 10.1080/10245332.2016.1240391; this figure represents the total theoretical genetic prevalence of the indication. The seroprevalence of antibodies against the AAVS3 capsid renders some patients ineligible for AAV gene therapy. We estimate approximately 60% would be eligible for AAVS3 gene therapy. Company estimate of patient numbers for GBA1-linked PD are for US, UK and EU4 .

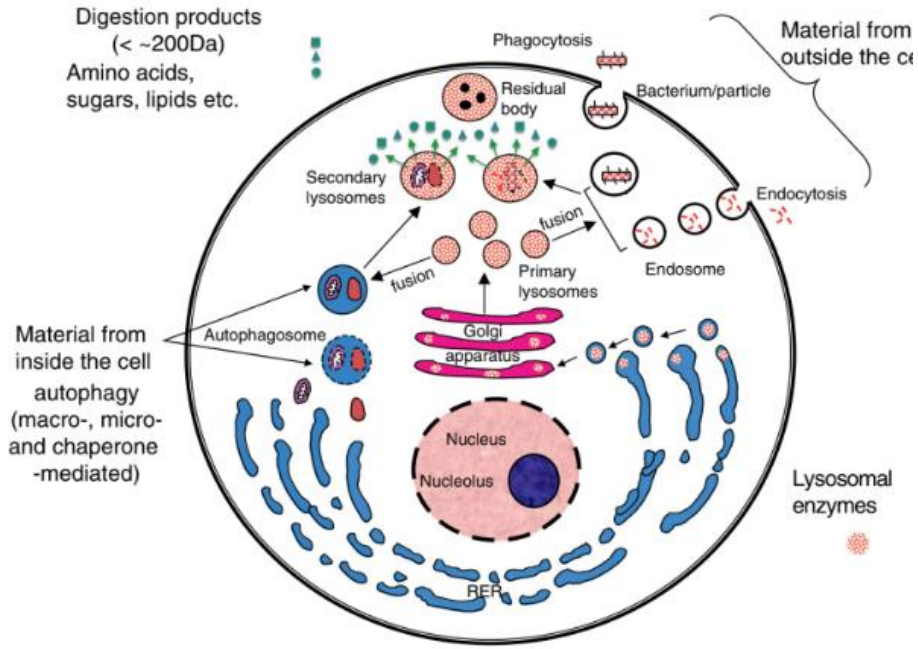
Overview of Gaucher Disease

Dr. Reena Sharma

Consultant Adult Metabolic Medicine and Honorary Senior Lecturer

Salford Royal Hospital, UK

Gaucher Disease is a Lysosomal Storage Disease

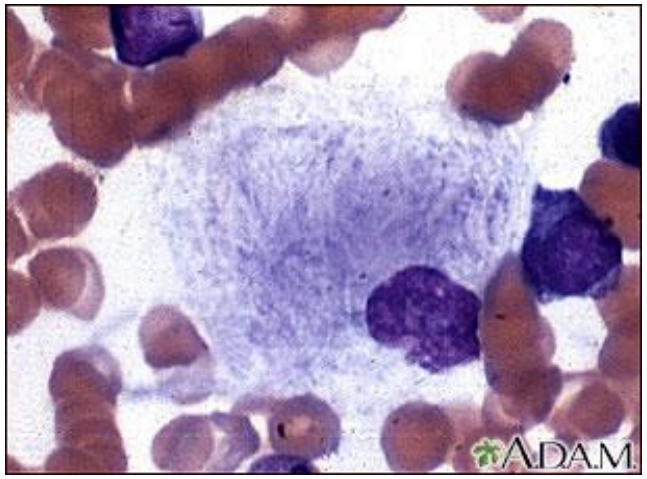


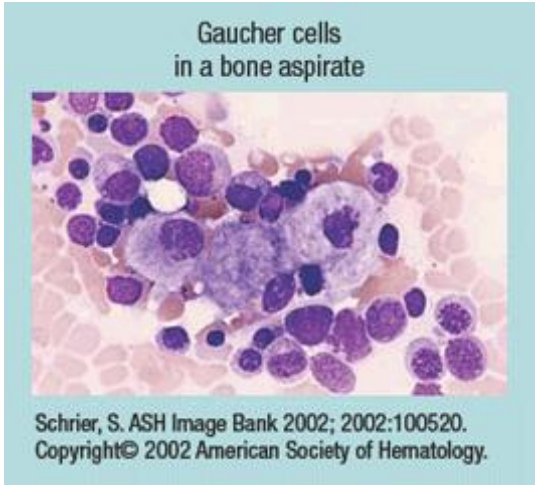
- Recycling centres in human cells
- Hydrolyses macromolecules in acidic medium
- Enzyme deficiency leads to accumulation of undigested macromolecules
- Transporter defects leads digested material not available for other cells and is sequestered within lysosomes
- Manifestations depends on which organs and tissues are most affected by the disruption

Lysosomal Storage Disorders: A Practical Guide. edited by Atul B. Mehta, Bryan Winchester

Progressive Disorder Affecting Multiple Organs and Systems

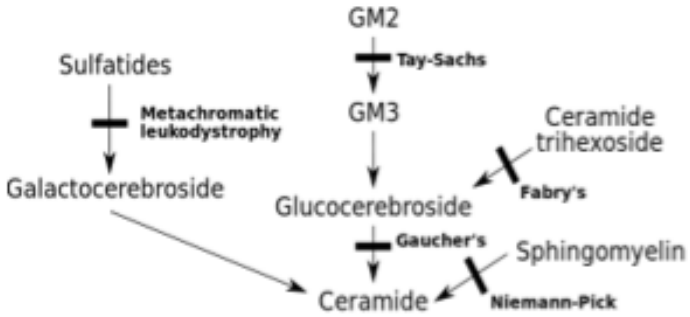
- First described in1882 by French medical student Phillipe Charles Ernest Gaucher
- Enlarged spleen and large cells were the first to be classed as Gaucher disease
- Gaucher cells: Glucosylceramide laden macrophages

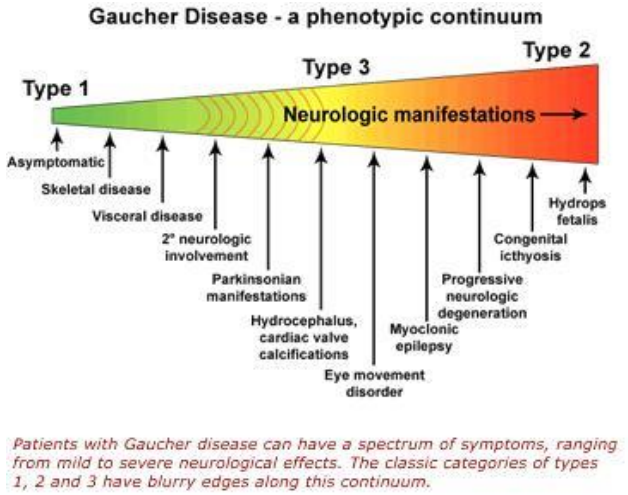




Aetiology and Pathogenesis of Gaucher Type 1

- GBA is causative gene: leads to deficiency of glucocerebrosidase (GCase) and buildup of cell lipid components glucosylceramide (Gb-1)/glucosylsphingosine (lyso-Gb1)
- In macrophage lysosomes: spleen/liver/bone/BM/lungs
- Produce inflammatory environment
- Gaucher disease Type 1 (non-neuronopathic form): most common type with variable symptoms and progression

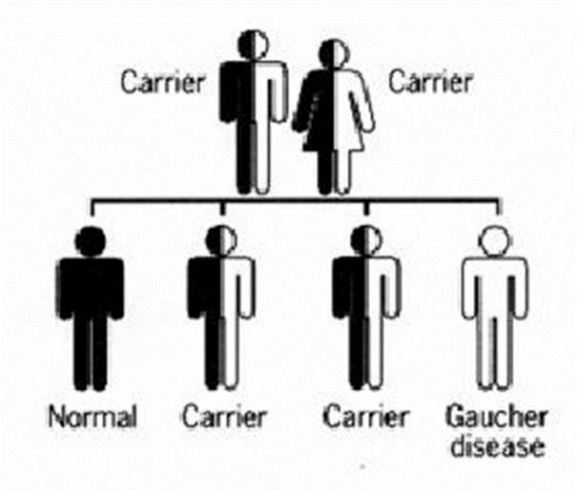




Patients with Gaucher disease can have a spectrum of symptoms, ranging from mild to severe neurological effects. The classic categories of types 1, 2 and 3 have blurry edges along this continuum.

Epidemiology

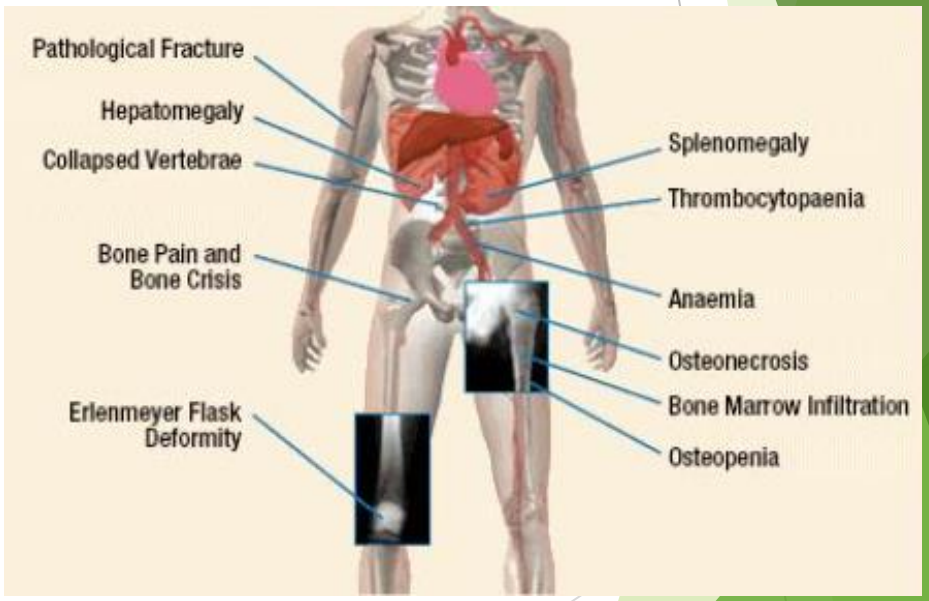
- Gaucher disease is one of the most common lysosomal storage disorders
- Type 1 is most common type of Gaucher disease in US, Europe and Israel
  - ~90 % of all Gaucher patients
- High incidence in Ashkenazi Jewish population
  - Carrier frequency is 1 in 12 and mutation frequency is 1 in 850
- In non-Jewish population, incidence is 1 in 40,000 -1 in 86,000 live births



Signs and Symptoms

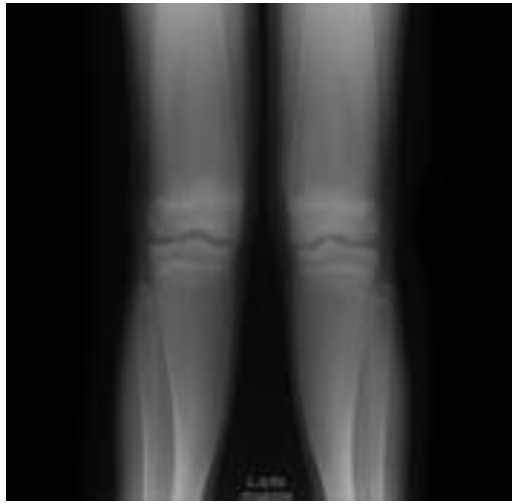
- Diagnosis can take years and multiple doctors; misdiagnosis is common
- Symptoms limit daily activities, personal/professional life and affect mental health

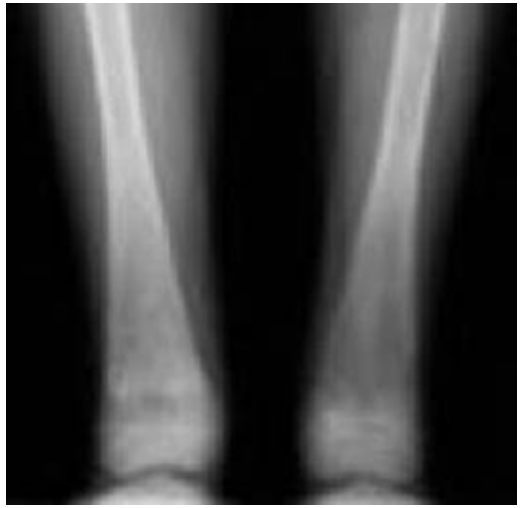
- Anaemia
- Thrombocytopenic
- Bleeding and bruising
- Hepatomegaly
- Splenomegaly and splenic infarcts
- Bone marrow infiltration
- Bone crisis
- Pathological fractures of vertebra and long bones
- Bone deformities
- Fatigue
- Pulmonary dysfunction
- Depression



Radiological Features

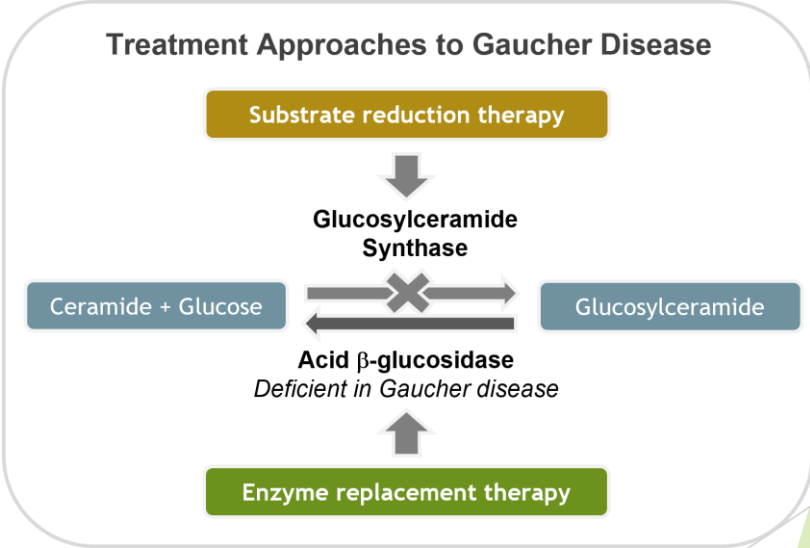
- Erlenmeyer flask bone deformity
- Bone marrow hyperplasia or infiltration leads to abnormal modeling





Current Treatment Landscape: ERT and SRT

- Enzyme replacement therapy (ERT)
  - First approval 1991; 3 therapies on the market today
  - Standard of care; paved the path for many other ERTs for various LSDs
  - Produced by recombinant DNA technology
  - Analogue of human enzyme GCase given IV every other week indefinitely
- Substrate reduction therapy (SRT)
  - 2 therapies on the market today
  - Reduces production of glucocerebroside
  - Oral therapy 2-3 times per day indefinitely
- Significantly changed the quality of life since ERT
- Splenectomy is rarely required now
- Improved haematological parameters and organomegaly; bone crisis reduced



How We Monitor Patients

- Biochemical and blood parameters
  - Haemoglobin
  - Platelets
  - Chitotriosidase
  - Angiotensin converting enzyme
  - GB1 (Glucosylsphingosine:lyso-Gb1)
  - Serum electrophoresis
  - Vitamin D
- Imaging
  - MRI abdomen and pelvis
  - DEXA scan
  - Skeletal survey if needed
- Clinical examination

What are the Unmet Needs

- Residual disease burden continues
- Biomarkers improve but rarely come to normal level
- Persistent radiological evidence of the disease
- Burden of long-term treatment as ERT or oral therapy
- Osteoporosis (reduced trabecular bone density score)
- Arthritis: Seropositive and seronegative arthritis
- Increased incidence of autoimmune disorders
- Risk of Myeloma and malignancies
- Risk of Parkinsonism

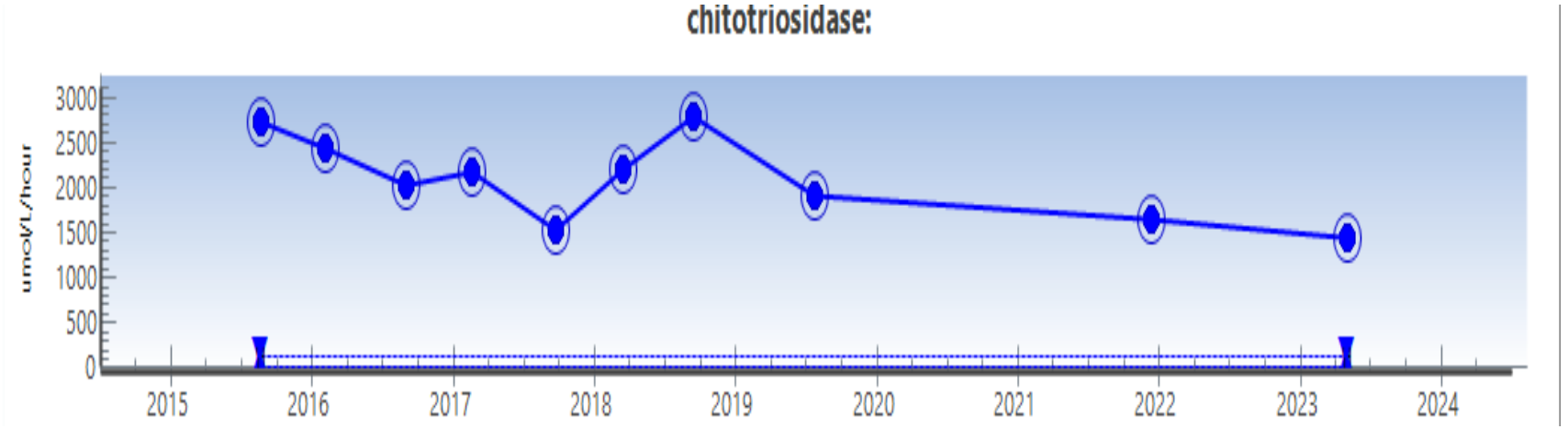
Many patients still have clinical manifestations after 10 years on ERT

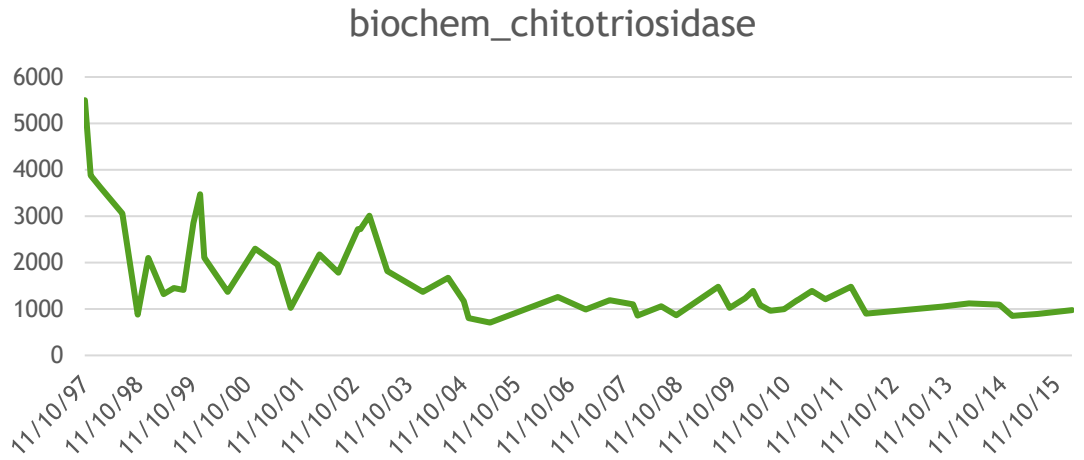
Rates in patients with symptom at baseline (%)	In those patients, rates after 10 years on ERT
Moderate/severe thrombocytopenia (76%)	28%
Anemia (43%)	12%
Moderate/severe splenomegaly (87%)	44%
Moderate/severe hepatomegaly (80%)	18%
Bone pain within last month (52%)	43%

Includes non-splenectomized patients only

Weinreb et al J Inherit Metab Dis (2013) 36:543–553

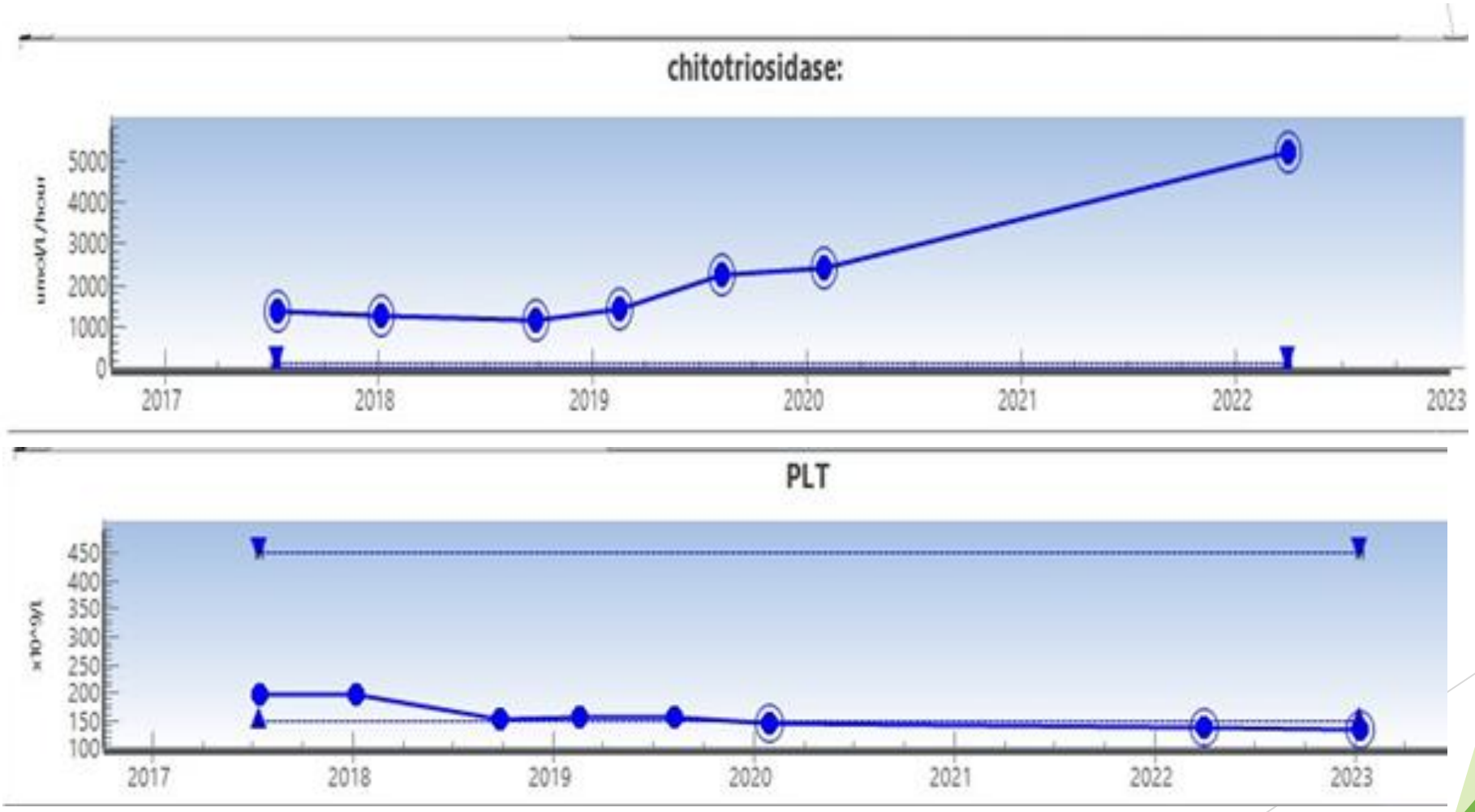
Persistent Elevated Chitotriosidase



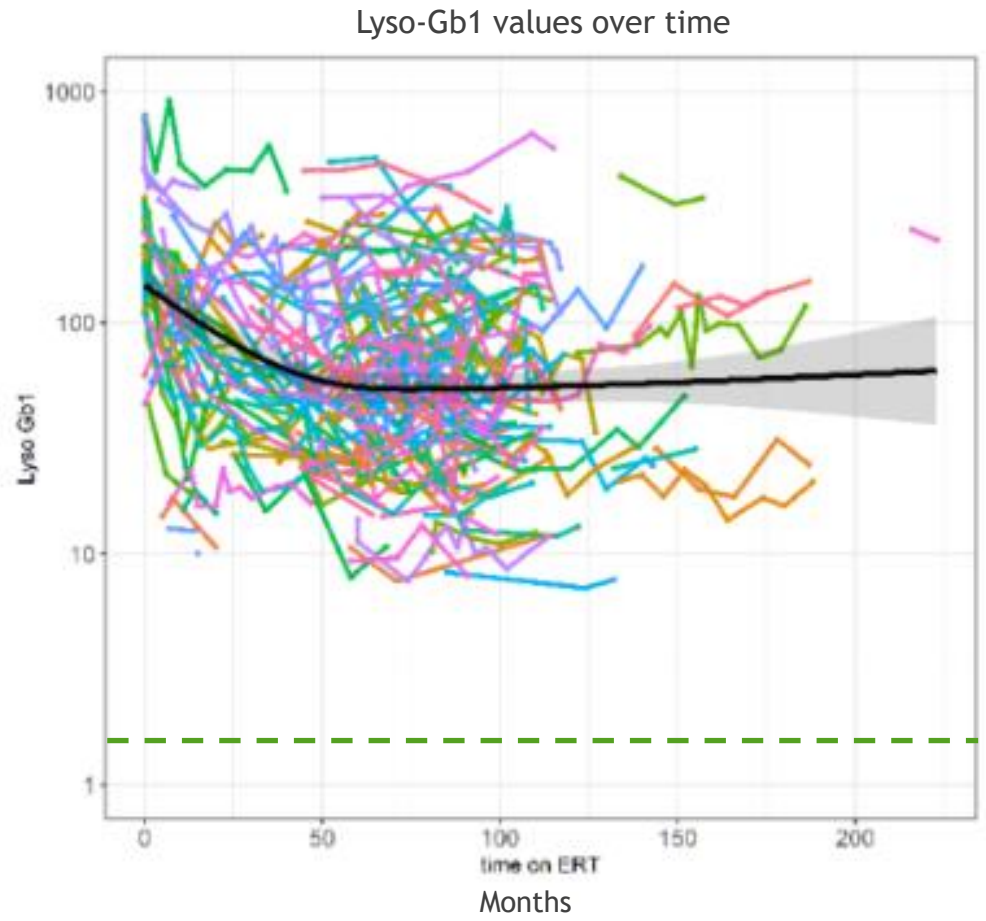


(reference range 4 -120 micromol/L/hr)

What Poor Compliance Looks Like



Lyso-Gb-1 levels improve with ERT but remain significantly elevated in some even years later



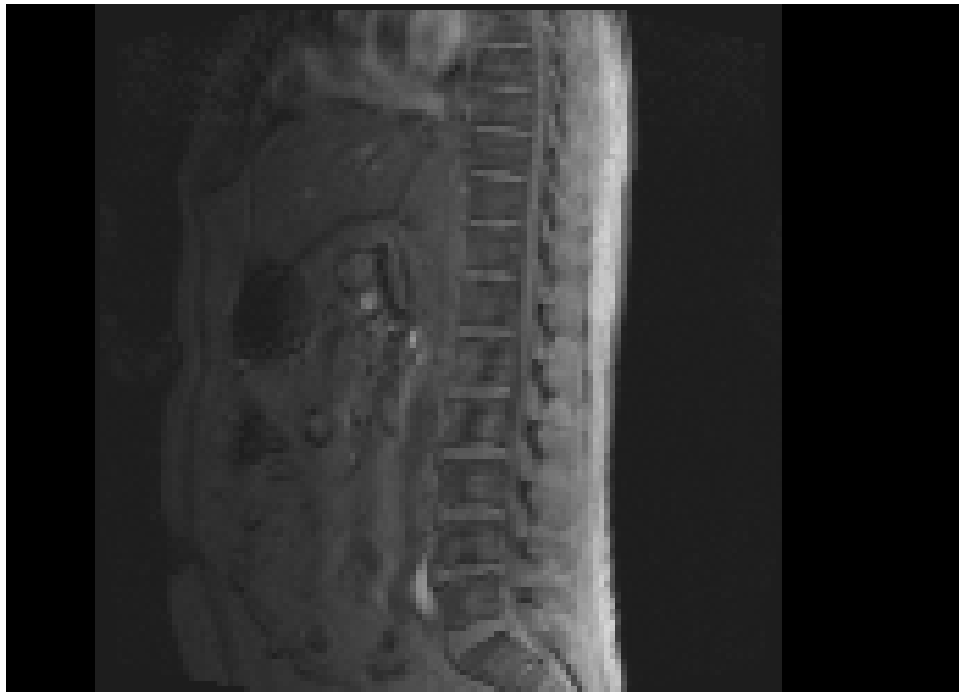
normal reference range for lyso-Gb_1 is ≤1.9 nmol/L

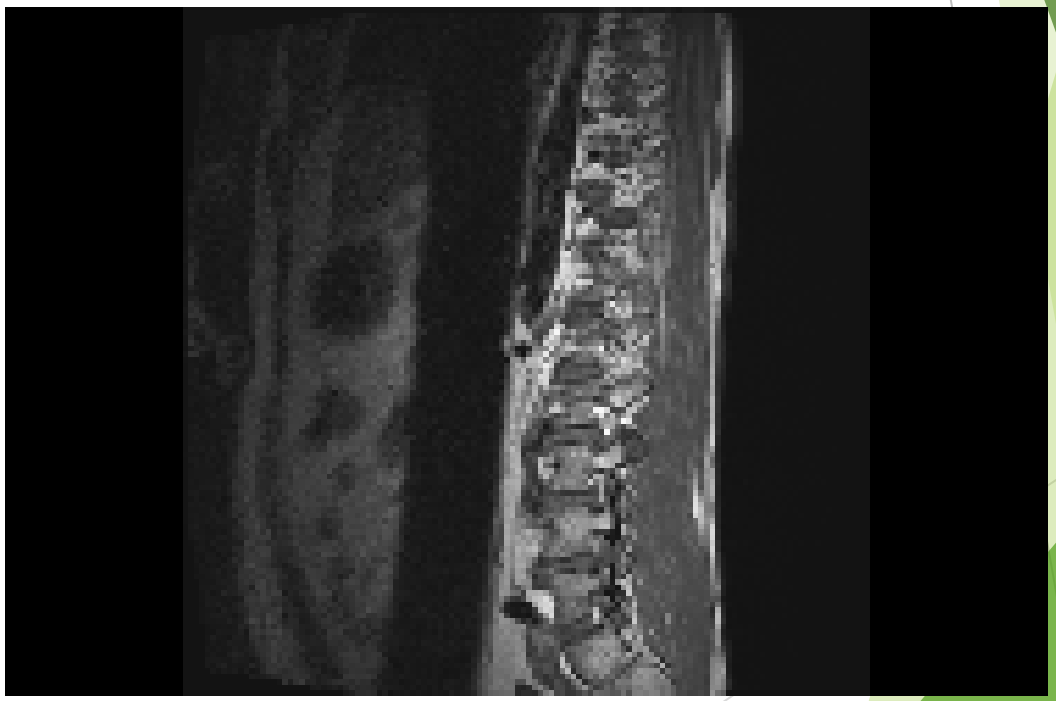
Dinur, et al Int. J. Mol. Sci. 2021, 22, 7699

Velaglucerase alfa (73 individuals, 407 measures). The black line and grey area indicate the unadjusted regression line (with 95% confidence interval (CI)).

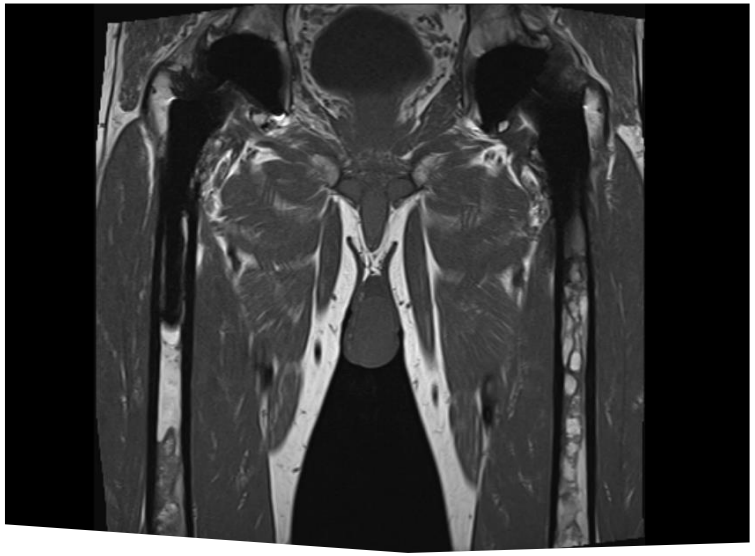
MRI of Spine

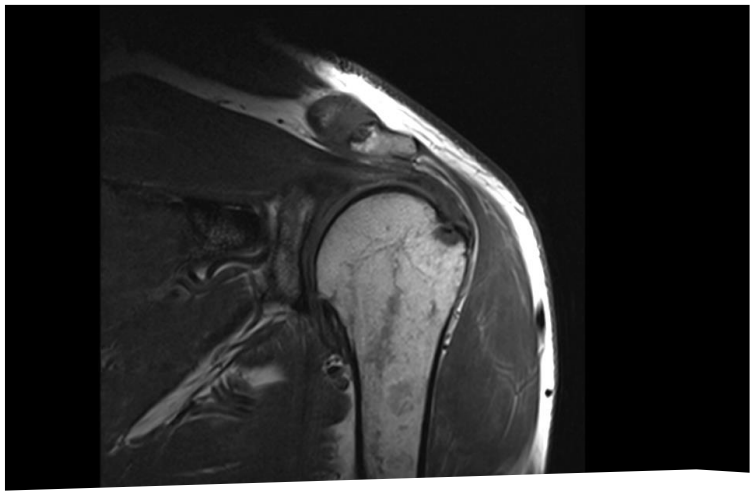
- Changes in MRI of spine in spite of being on ERT

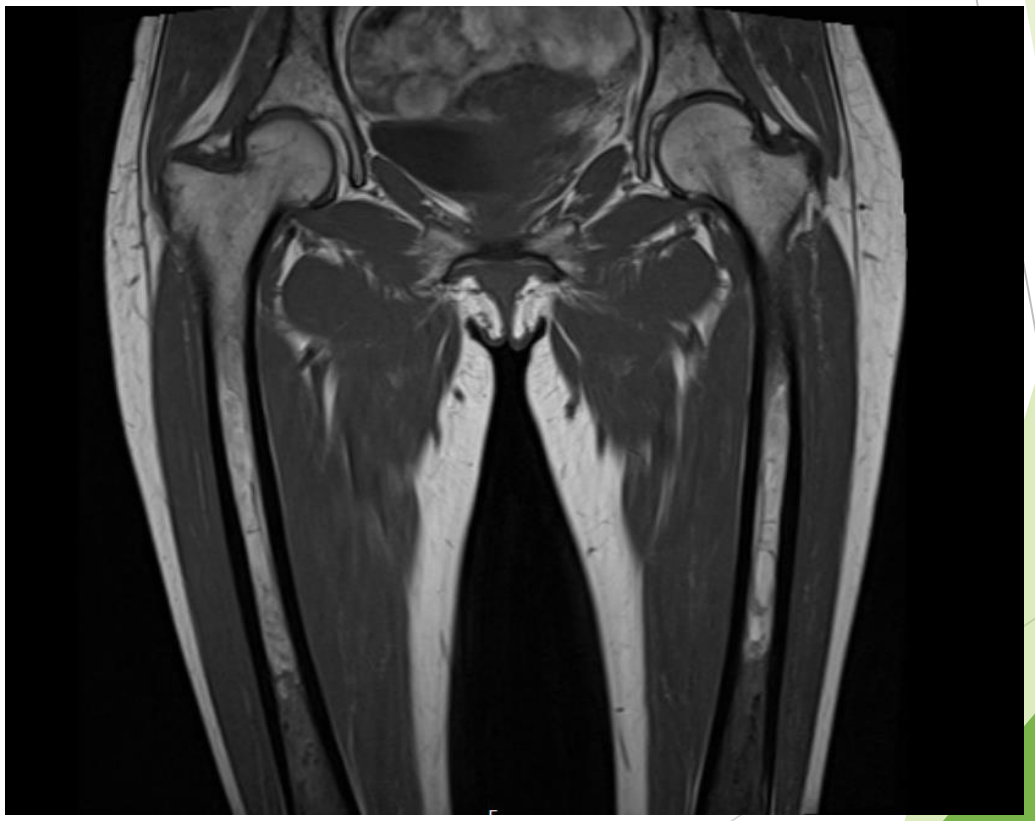




MRI of Femur, Pelvis, Humerus







Why Gaucher is a Good Candidate for Gene Therapy

- Well demarcated diagnostic tests and genetic screening
- Clear cause and effect of both missing enzyme and replacing enzyme
- Experience with ERT has shown that replacing missing enzyme is efficacious
- Many patients have symptoms as ERT wears off- good to have a continuous supply
- Ability to get higher levels of continuous enzyme than ERT ever can- may allow for better clinical outcomes
- Lifetime burden of current therapies lends itself to new alternatives
- Treatment centers in place, allowing for rapid education on new therapies and processes that may be specific to gene therapy
- Strong patient organisations with focus on clinical advancements, newborn screening, and payor coverage

What is the Hope for Gene Therapy

- Single dose- so no need to have lifelong ERT or SRT….I have patients in their 80s on ERT
- Further reduce burden of disease
- Have positive benefit on the bone disease burden
- Reduce osteoporosis and fractures
- Reduce the risk of malignancy and inflammatory arthritis
- Reduce the risk of having Parkinson's disease


Thank you
for
listening!

FLT201 for Gaucher Disease

Pamela Foulds, MD
Chief Medical Officer, Freeline Therapeutics

Potential first- and best-in-class gene therapy for Gaucher disease Type 1

HIGHLY DIFFERENTIATED

- Novel transgene encoding a rationally engineered longer-acting GCase variant
- Potential to penetrate deeper tissues that current therapies do not sufficiently reach
- Proprietary AAVS3 capsid delivers high and durable protein expression at low doses

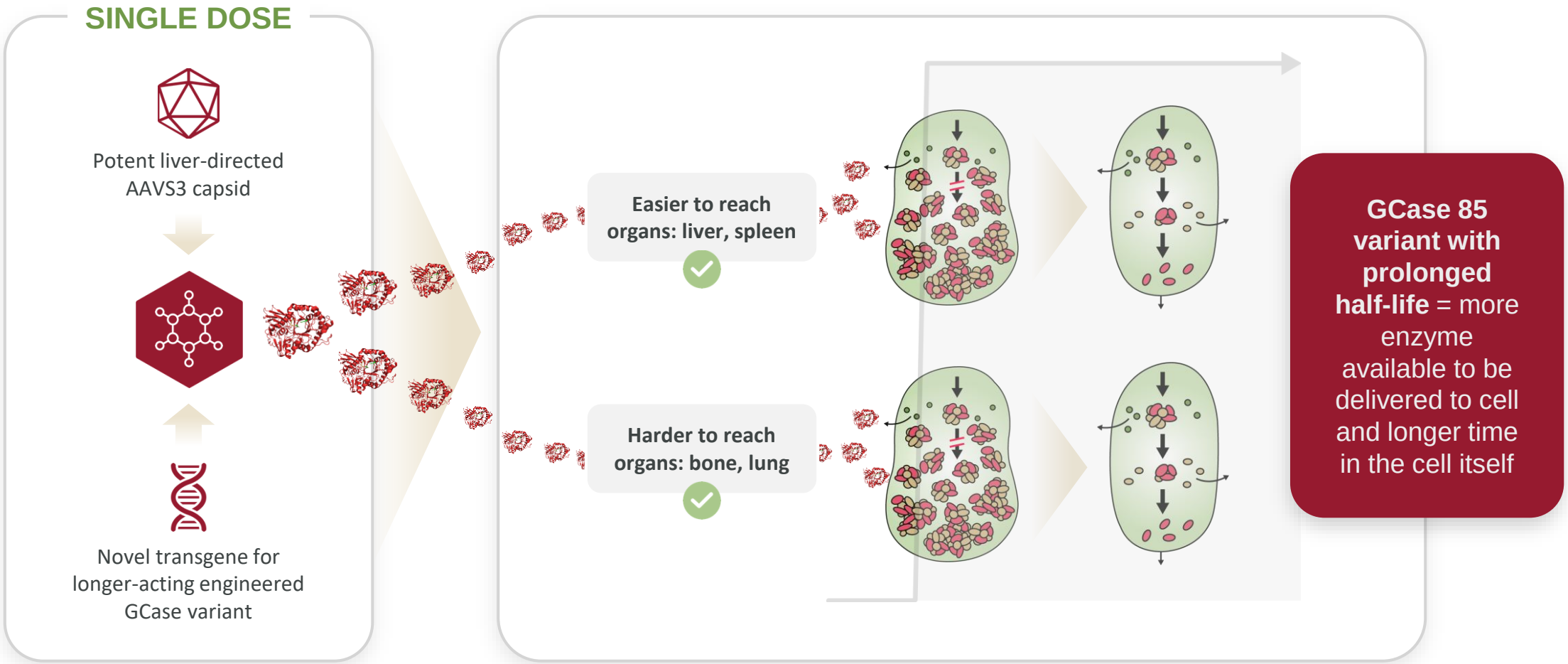
SIGNIFICANT MARKET OPPORTUNITY

- Most common type of Gaucher disease
- ~18k patients in US, UK, EU4 and Israel

ENCOURAGING DATA & NEAR-TERM CATALYST

- Robust preclinical data showing GCase uptake and substrate clearance in all disease-affected tissues
- Dosing underway in Phase 1/2 GALILEO-1 trial
- Initial data, including safety and enzyme activity, from first cohort expected in Q3 2023

FLT201 has potential to deliver continuous level of enzyme and penetrate deeper tissues that ERT does not reach



Our scientists engineered GCase variant with substantially longer half-life than wildtype

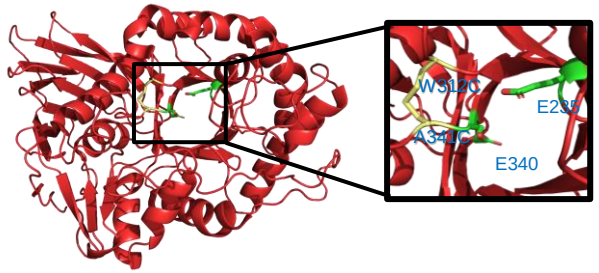
Key features of GCase variant

- 20-fold increase in half-life in lysosomal pH compared with wildtype (wt)
- Specific activity unchanged compared to wt GCase
- 6-10 fold increase in half-life compared to wt, enabling increased steady-state plasma levels *in vivo*

GCase variant 85 structure

Two internal amino acid substitutions

- Does not impinge on the active site
- Minimizes 3D structural change

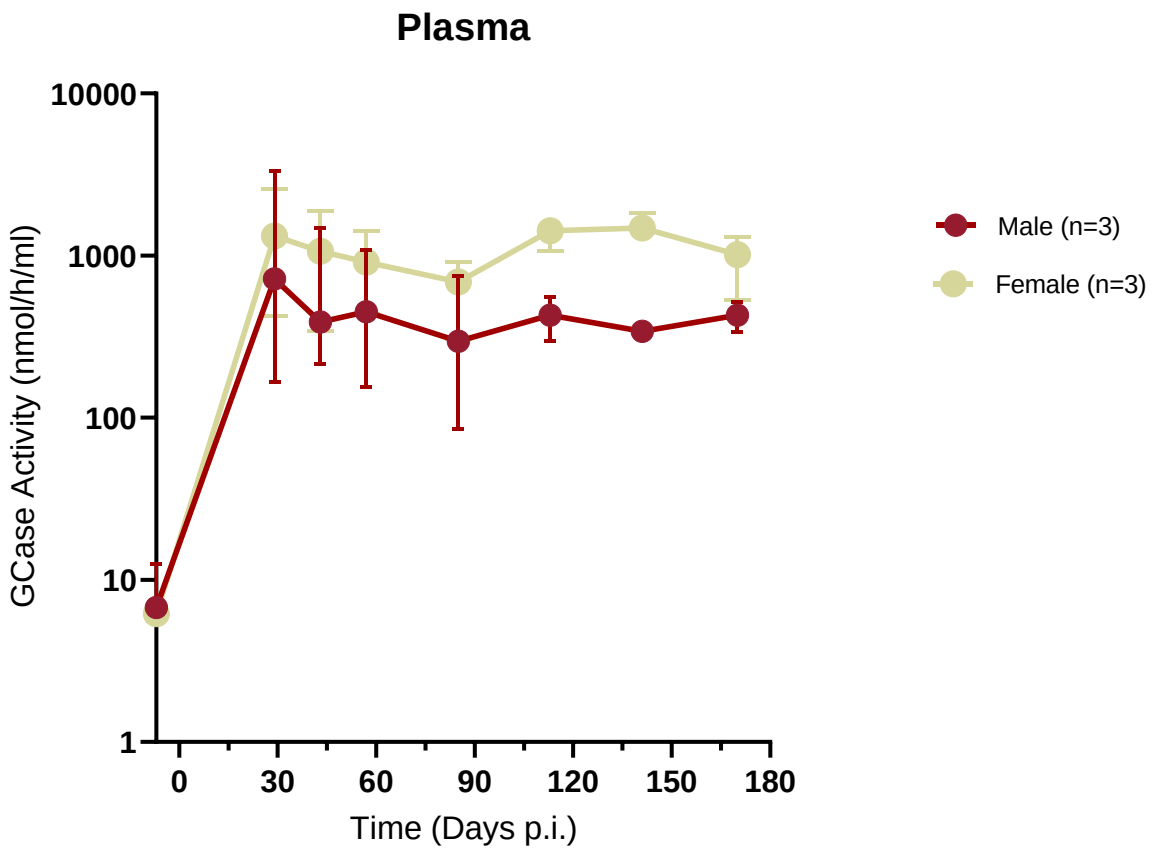


Biophysical properties of variant 85 and wildtype GCase

	Lysosomal pH	Human serum
	HALF-LIFE (MINUTES)	
WT GCase	388	24
Variant 85	>8,639	143
Improvement	**>21X**	**6X**

WORLD*Symposium* 2021: Fabrizio Comper et al. Generation of β-Glucocerebrosidase Variants with Increased Half-life in Human Plasma for Liver Directed AAV Gene Therapy Aimed at the Treatment of Type 1 Gaucher Disease

FLT201 showed high GCase expression in non-human primates

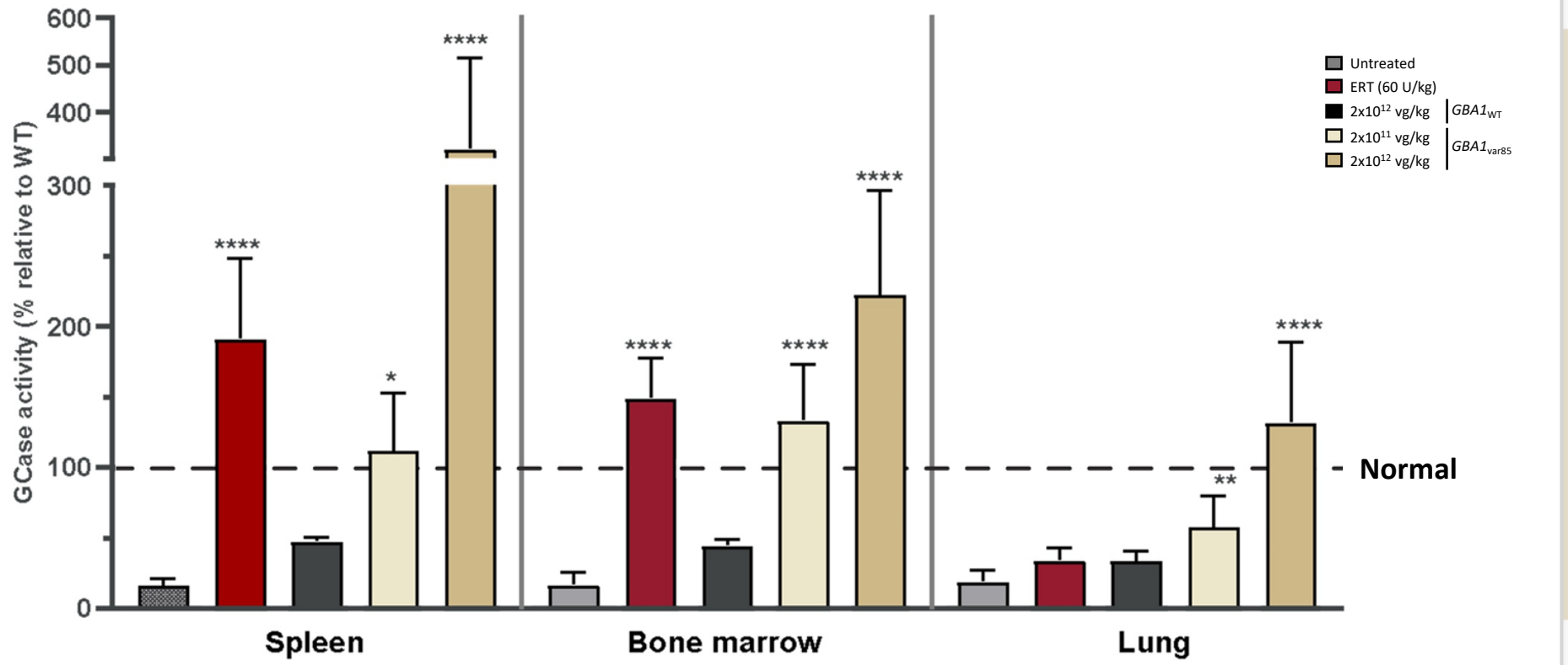


Achieved steady increases in GCase plasma levels

- A single injection of FLT201 was well tolerated
- Resulted in a rapid increase of GCase in plasma that was sustained for at least six months (trial ongoing)

Dose: FLT201 $2x10^{12}$ vg/kg (long-term observational trial)

FLT201 led to increased uptake in key tissues in Gaucher mice



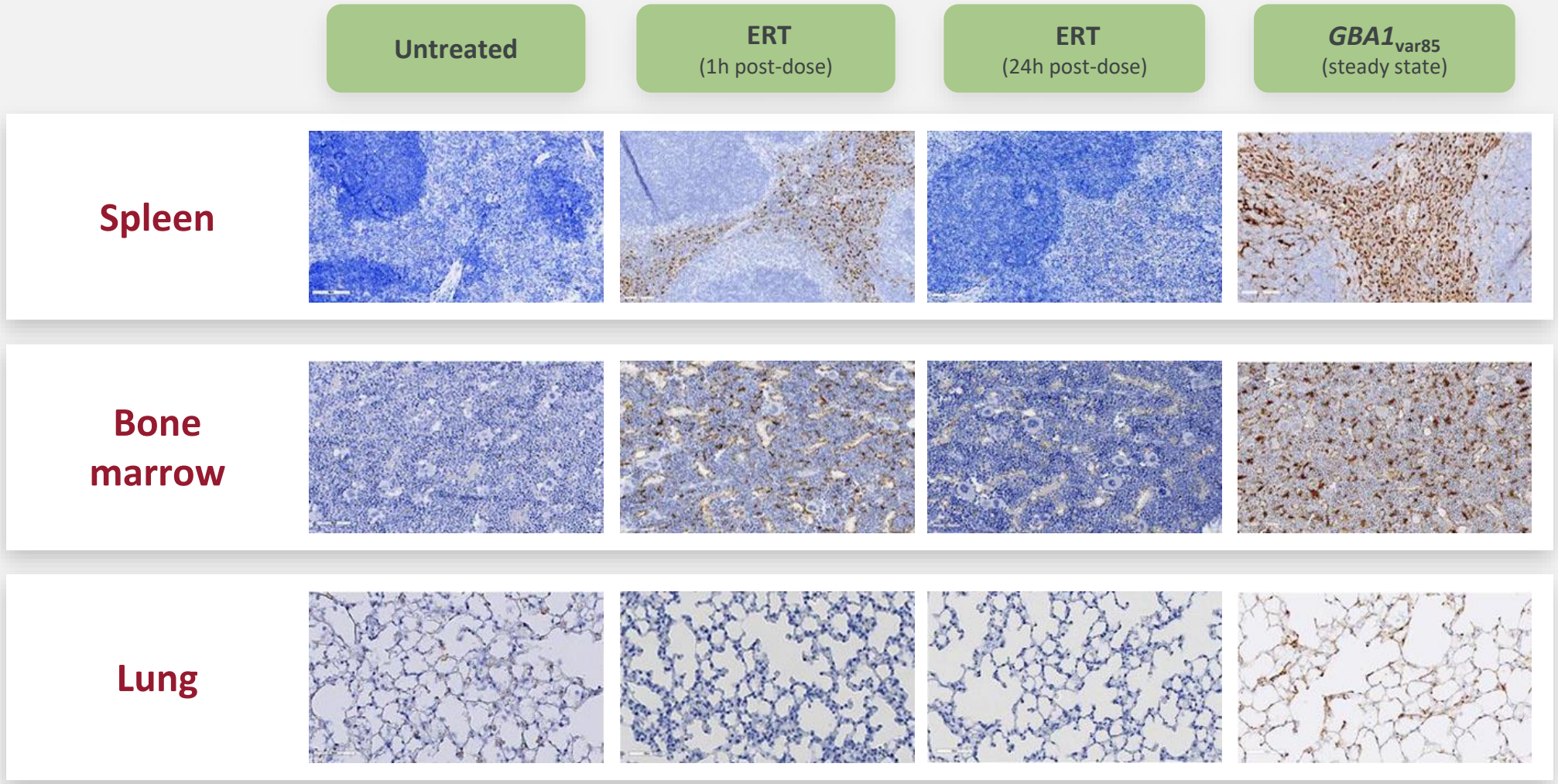
- Dose-dependent increases in GCase to above normal levels in key tissues
- Greater GCase uptake in key tissues than with ERT or with short half-life wildtype gene therapy

Data represented as mean ± SD. n= 9 to 16 per treatment group. * P≤0.05, ** P≤0.001, P≤0.001, **** P ≤ 0.0001, one-way ANOVA.

American Society of Gene & Cell Therapy 2021 Annual Meeting: Romuald Corbau et al. FLT201, a Novel Investigational AAV-Mediated Gene Therapy Candidate for Gaucher Disease Type 1
WORLD*Symposium* 2021: Romuald Corbau et al. FLT201: An AAV-Mediated Gene Therapy for Type 1 Gaucher Disease Designed to Target Difficult to Reach Tissues

ERT=Velaglucerase alfa

Enhanced and sustained GCase uptake observed in key tissues compared to ERT



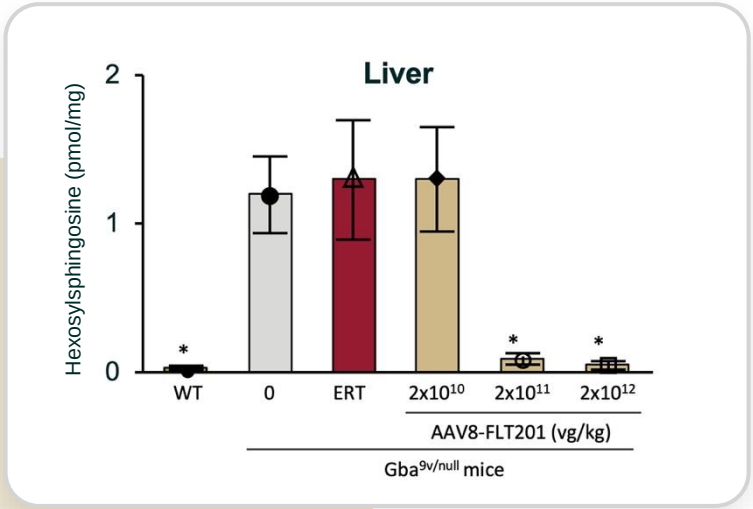
Anti-GCase – DAB, Haematoxylin counterstain, AAV8

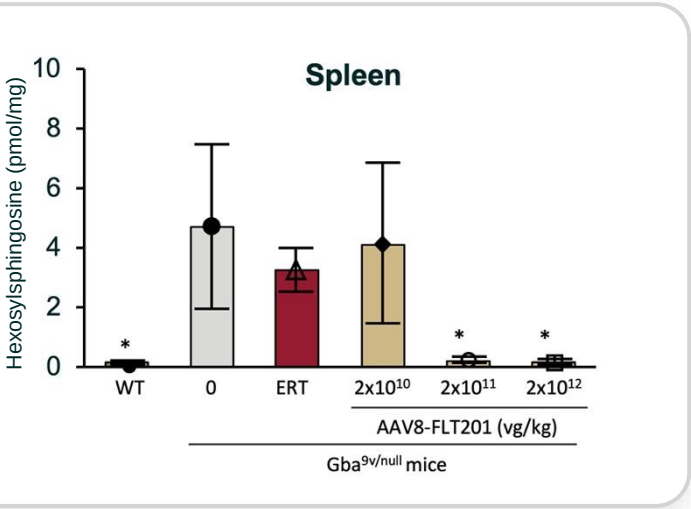
ERT=Velaglucerase alfa

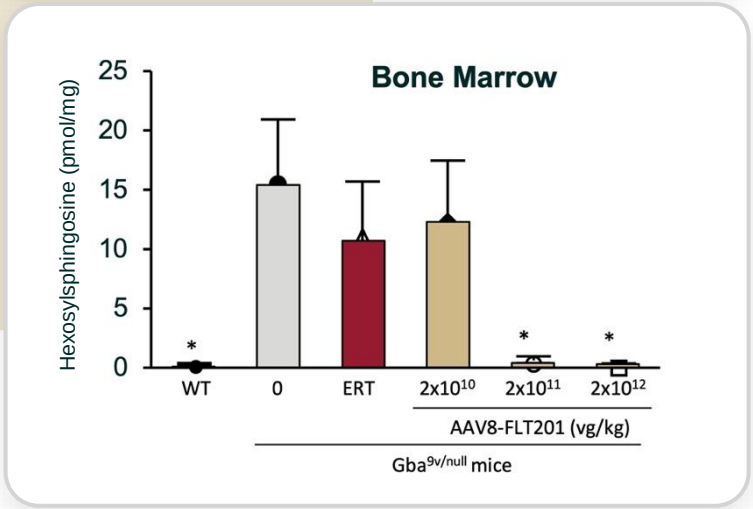
FLT201 cleared harmful substrate in key tissues in Gaucher mice

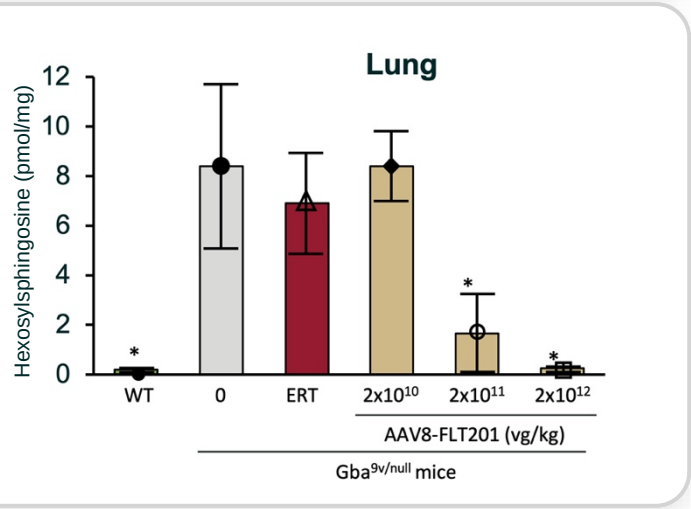
Robust substrate elimination

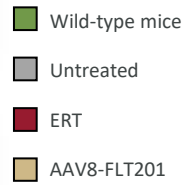
Dose-dependent reductions of lyso-Gb1 observed in all tissues analyzed, including bone marrow and lung









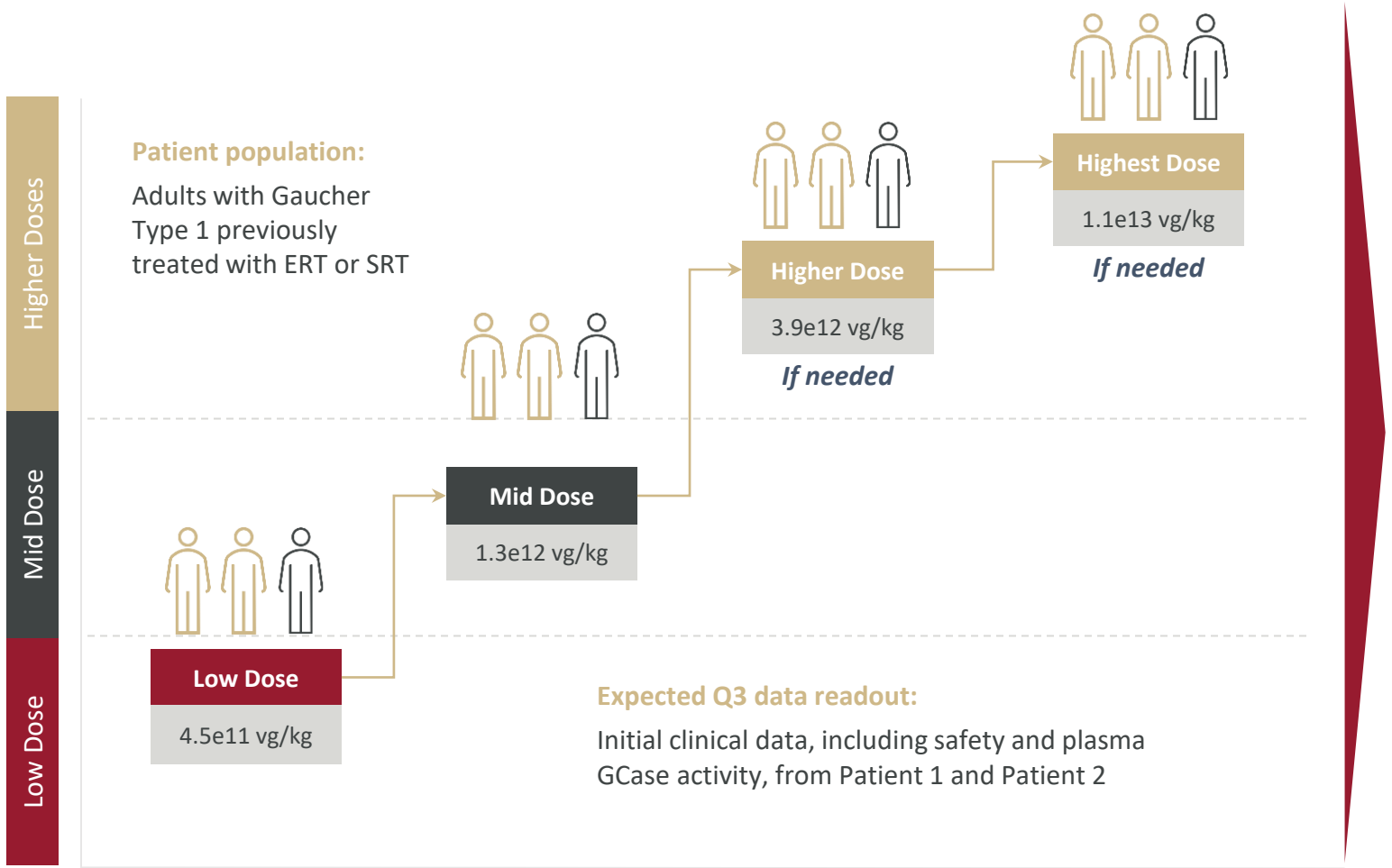


WORLD*Symposium* 2021: Romuald Corbau et al. FLT201: An AAV-Mediated Gene Therapy for Type 1 Gaucher Disease Designed to Target Difficult to Reach Tissues
AAV8-FLT201 = AAV8 pseudo-typed FLT201 genome. ERT = Velaglucerase alfa 60 U/kg biweekly (equivalent of the standard of care in humans). WT = wild-type mice. Evaluated 12 weeks post-injection. *p ≤ 0.0001.

GALILEO-1 Phase 1/2 dose-finding trial design

First-in-human, open-label, multicenter study; dosing underway in cohort 1

Establish a dose that delivers sustained increases in GCase activity to reduce substrate accumulation and improve clinical parameters



Planned to be dosed in each cohort

Flexibility to expand number of patients dosed

Trial protocol allows for testing up to four doses. Depending on dose response in the initial cohorts, we may not ultimately need to escalate to the later doses. The Data Monitoring Committee may recommend the next dose level at the next planned dose level, at same, higher or lower dose level based on emerging safety/tolerability, PK, PD and efficacy data. A 4-week stagger between patients is built into the protocol, with the exception of the US where there is an 8-week stagger prior to dosing a subsequent patient.

Freeline has the opportunity to change the course of disease for people with Gaucher disease Type 1

Serious ongoing unmet need

Current treatment provides incomplete responses, leading to continued symptoms and disease progression

Carry heavy life-long treatment burden

~18K people living with Gaucher disease type 1

Life-changing potential

FLT201 delivers rationally engineered GCase variant with longer half-life

Potential to reach deep tissues and improve outcomes

Highly differentiated from other gene therapy approaches

High protein expression at low doses provides potential safety and manufacturing advantages

Near-term value drivers

Dosing in first-in-human study underway

Initial safety and enzyme activity data from first cohort expected in Q3 2023

Focus and expertise to advance FLT201 in Gaucher disease

FREELINE

Q&A