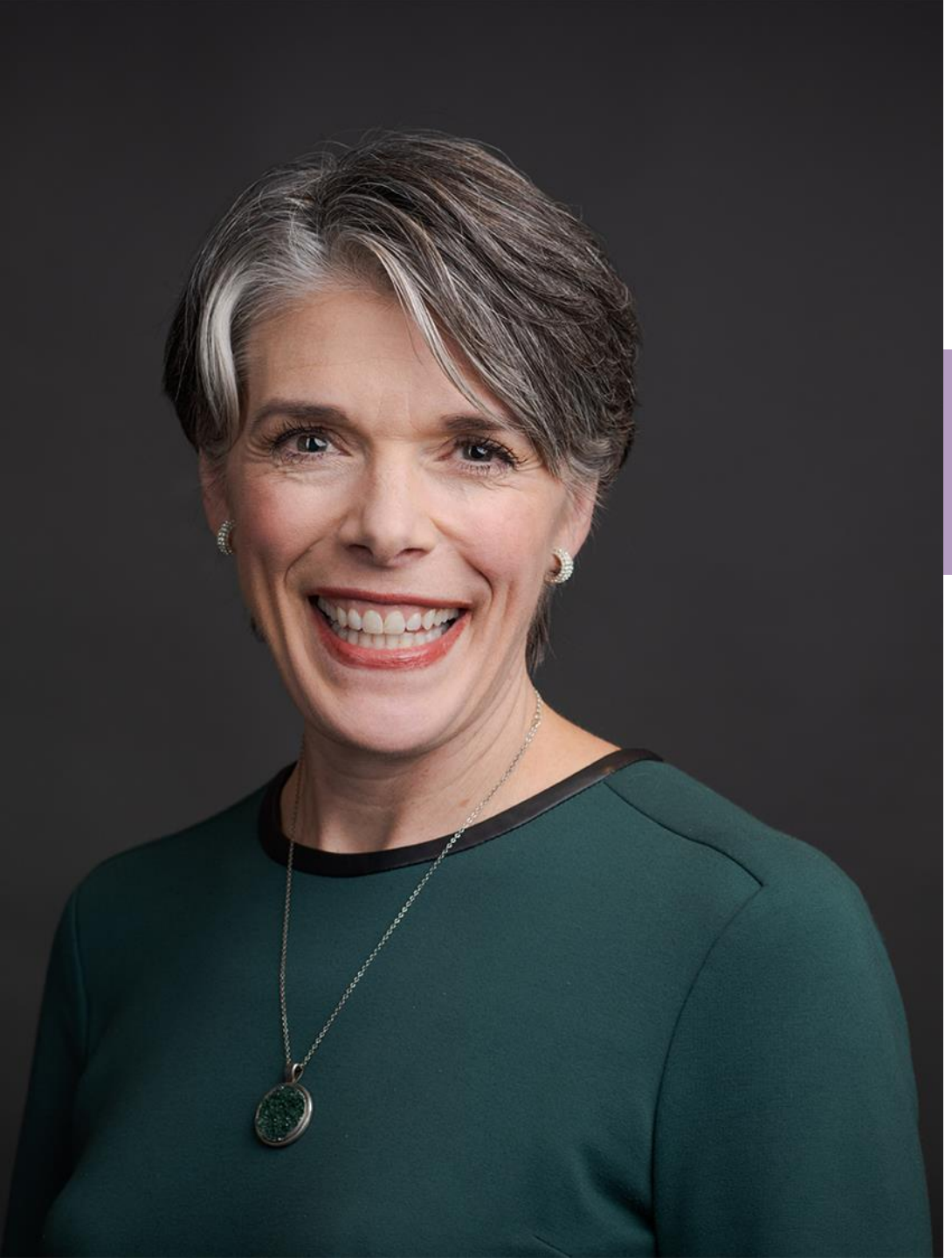
Primary Mitochondrial Diseases: A Rare Disease Virtual Workshop

May 22, 2025 | 10am – 3:30pm *(eastern)*

The public meeting will begin shortly

This activity is supported by the Food and Drug Administration (FDA) of the U.S. Department of Health and Human Services (HHS) as part of an award of $90,000 in federal funds (100% of the project). The contents are those of the author(s) and do not necessarily represent the official views of, nor an endorsement, by FDA, HHS, or the U.S. Government. For more information, please visit FDA.gov.

REAGAN-UDALL FOUNDATION FOR THE FDA



REAGAN-UDALL FOUNDATION FOR THE FDA

Welcome

Susan C. Winckler, RPh, Esq.

CEO, Reagan-Udall Foundation for the FDA

Housekeeping

REAGAN-UDALL FOUNDATION FOR THE FDA

Due to the meeting size, your microphone and video will remain off during the meeting

Please share your questions using the Zoom Q&A function

This public meeting is being recorded
The slides, transcript, and video will be available at www.ReaganUdall.org

Today's Agenda (Session 1)

REAGAN-UDALL FOUNDATION FOR THE FDA

10 am	**Welcome & Opening Remarks** Susan C. Winckler, RPh, Esq., Reagan-Udall Foundation for the FDA
10:05 am	**FDA Opening Remarks** Catherine Pilgrim-Grayson, MD, MPH, Center for Drug Evaluation and Research, FDA
10:10 am	**Setting the Stage: A Variety of Perspectives** Michio Hirano, MD, Columbia University Medical Center **Clinical Trial Design and Implementation: Patient Population Considerations** Anna Choe, MD, MPH, Center for Drug Evaluation and Research, FDA **Primary Mitochondrial Disease Drug Development** Reenie McCarthy, JD, Stealth Biotherapeutics
11:05 am	**Reactor Panel** Marni Falk, MD, Children's Hospital of Philadelphia & University of Pennsylvania Perelman School of Medicine Brian Tseng, MD, PhD, The POLG Foundation Philip Yeske, PHD, United Mitochondrial Disease Foundation
11:35 am	**10-minute Break**

Today's Agenda (Session 2)

REAGAN-UDALL FOUNDATION FOR THE FDA

11:45 am	**Selecting Patient-focused Outcomes & Statistical Considerations** Naomi Knoble, PhD, Center for Drug Evaluation and Research, FDA Yan Wang, PhD, Center for Drug Evaluation and Research, FDA
12:15 pm	**Presentation on Clinical Outcome Assessment (COA) example** Amel Karaa, MD, Massachusetts General Hospital & Harvard Medical School
12:30 pm	**Reactor Panel** Kasey Woleben, Cure Mito Foundation Zarazuela Zolkipli-Cunningham, MBChB, MRCP, Children's Hospital of Philadelphia & University of Pennsylvania Perelman School of Medicine
1 pm	**LUNCH**

Today's Agenda (Session 3)

REAGAN-UDALL FOUNDATION FOR THE FDA

1:30 pm	**Current Approaches, Challenges, and Opportunities** Magnus Hansson, MD, PhD, Abliva AB Chad Glasser, PharmD, MPH, Tisento Therapeutics **PLUS Q&A (20min)**
2:30 pm	**Using What We've Learned To Move Forward** Jason Colquitt, Across Healthcare Amel Karaa, MD, Massachusetts General Hospital & Harvard Medical School Kerry Jo Lee, MD, Center for Drug Evaluation and Research, FDA Sophia Zilber, Lived Experience Perspective
3:30 pm	**Closing Remarks & Adjourn**

REAGAN-UDALL FOUNDATION FOR THE FDA

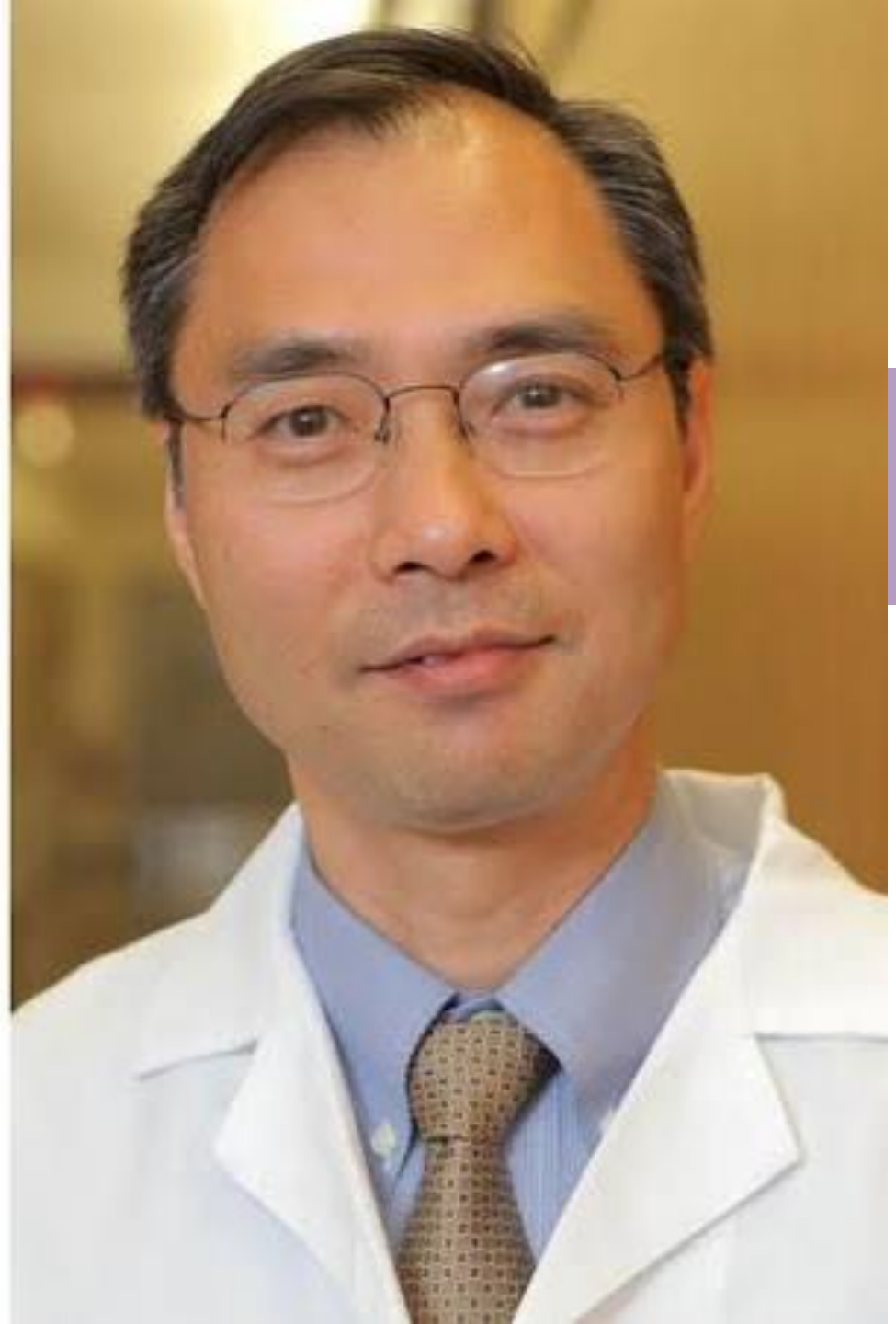
FDA Opening Remarks

Catherine Pilgrim-Grayson, MD, MPH

Director, Division of Rare Disease & Medical Genetics

Center for Drug Evaluation and Research

U.S. Food and Drug Administration



REAGAN-UDALL FOUNDATION FOR THE FDA

Setting the Stage: A Variety of Perspectives

Michio Hirano, MD

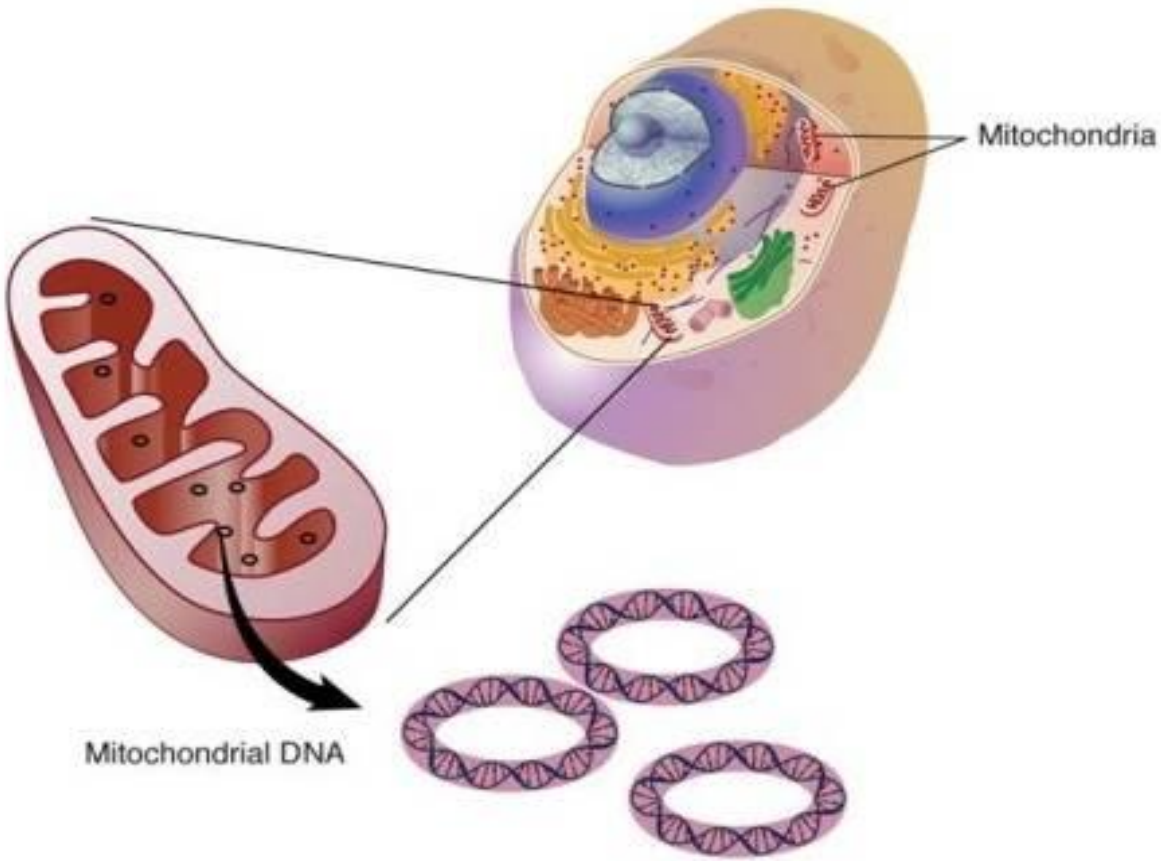
Columbia University Irving Medical Center

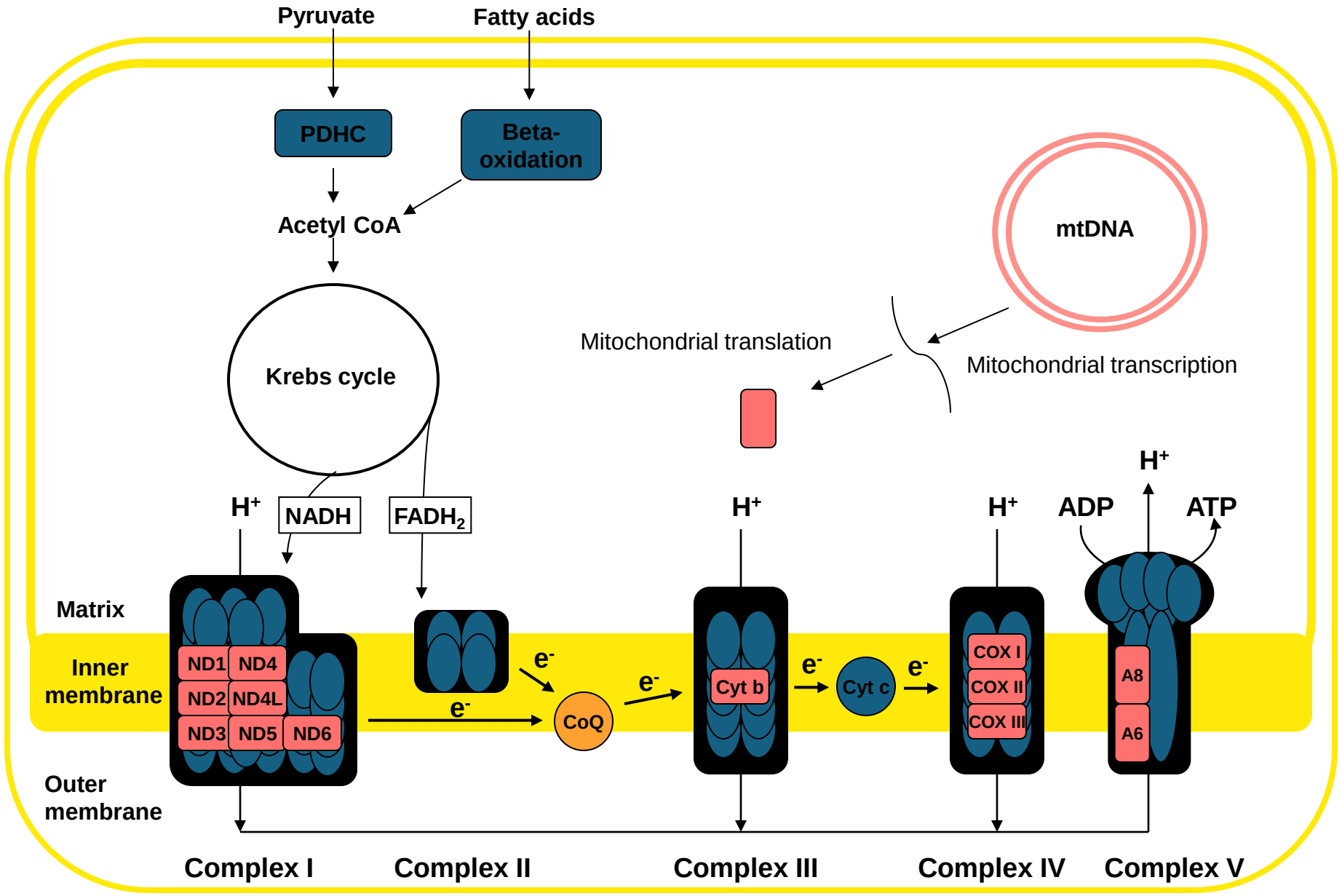
Setting the Stage:
A Variety of Perspectives

Primary Mitochondrial Diseases
A Virtual Rare Disease Workshop
Reagan-Udall Foundation for the FDA
May 22, 2025

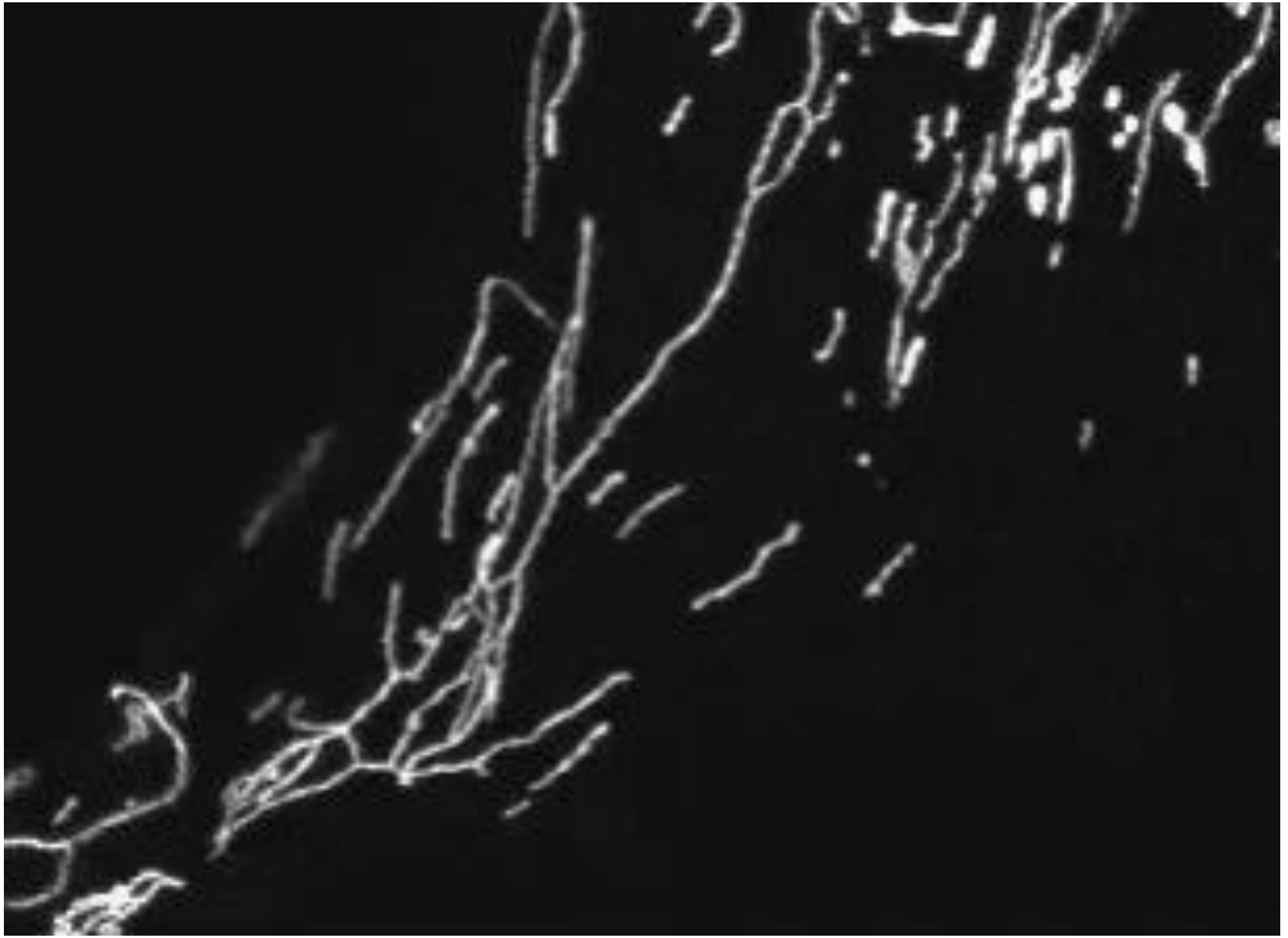
Michio Hirano, MD
Columbia University Irving Medical Center
New York, NY

Mitochondria are the powerhouses of the cell





	Complex I	Complex II	Complex III	Complex IV	Complex V
mtDNA-encoded	7	0	1	3	2
nDNA-encoded	39	4	10	10	12

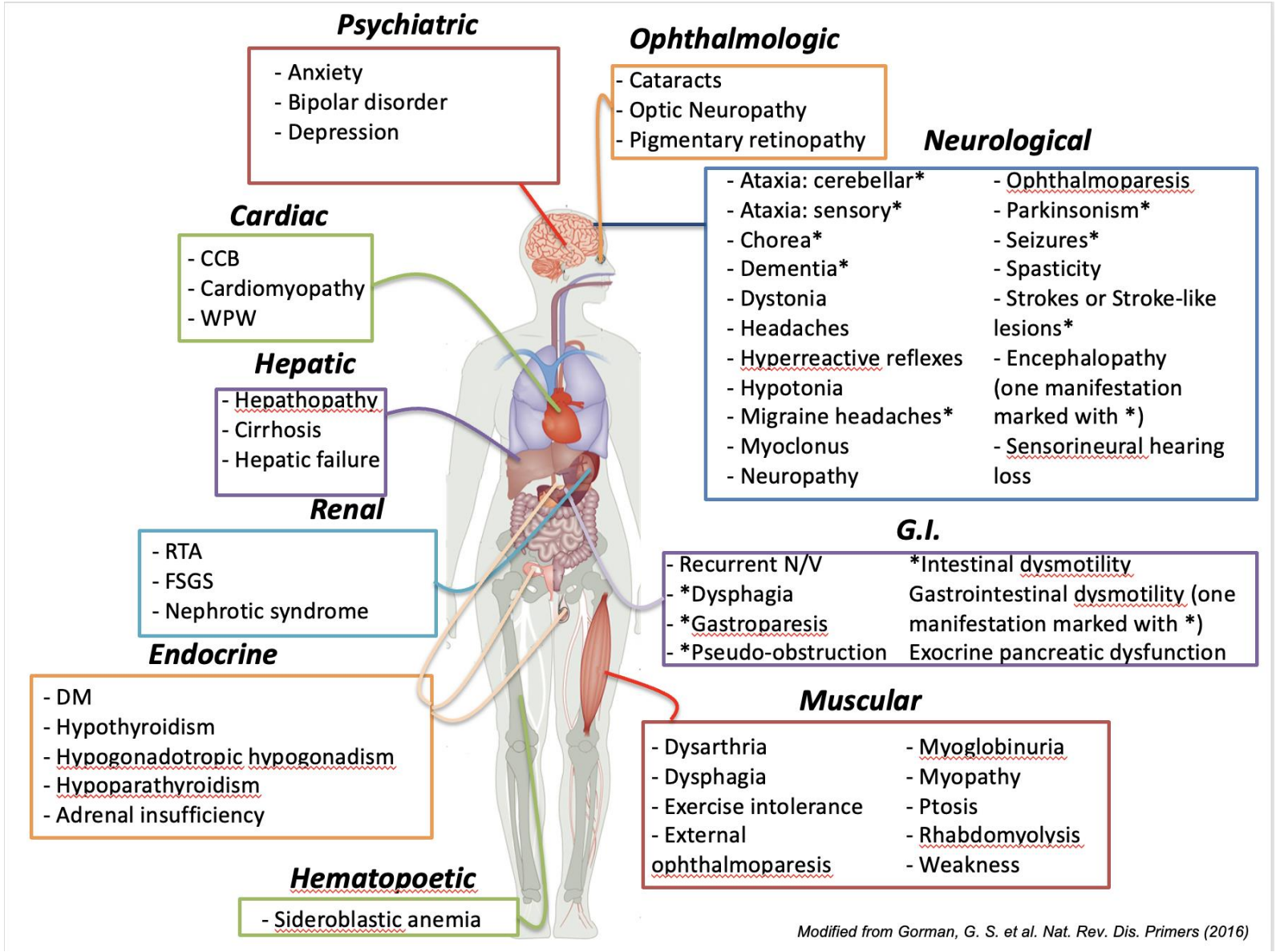


D. Chan Caltech

Mitochondrial diseases are complicated because:

- Mitochondria are required by virtually all cells in the body.
- Mitochondria perform multiple functions.
- Mitochondria are the products of two genomes: nuclear DNA and mitochondrial DNA (mtDNA).
- There are numerous mitochondrial diseases.

Clinical Manifestations of Mitochondrial Diseases



RESEARCH ARTICLE

Mitochondrial disease patient motivations and barriers to participate in clinical trials

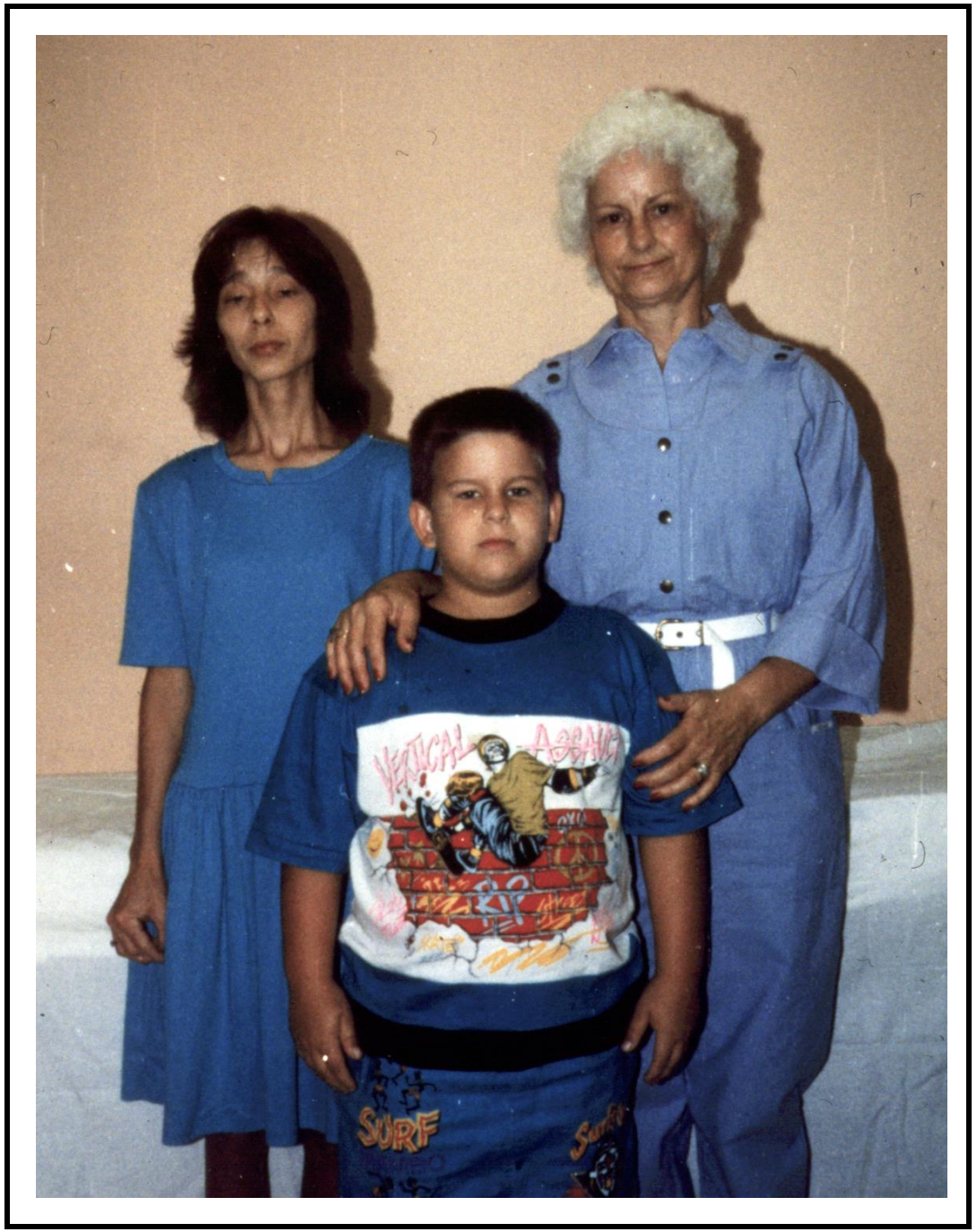
Zarazuela Zolkipli-Cunningham[1,2], Rui Xiao[3], Amy Stoddart[2,4,5], Elizabeth M. McCormick[2,5], Amy Holberts[6], Natalie Burrill[2], Shana McCormack[2,7,8], Lauren Williams[9], Xiaoyan Wang[9], John L. P. Thompson[9], Marni J. Falk[2,5,9]*

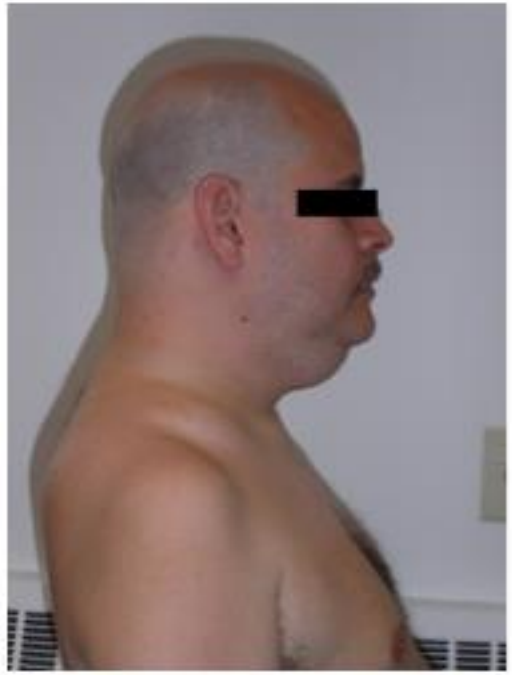
PLOS One 2018

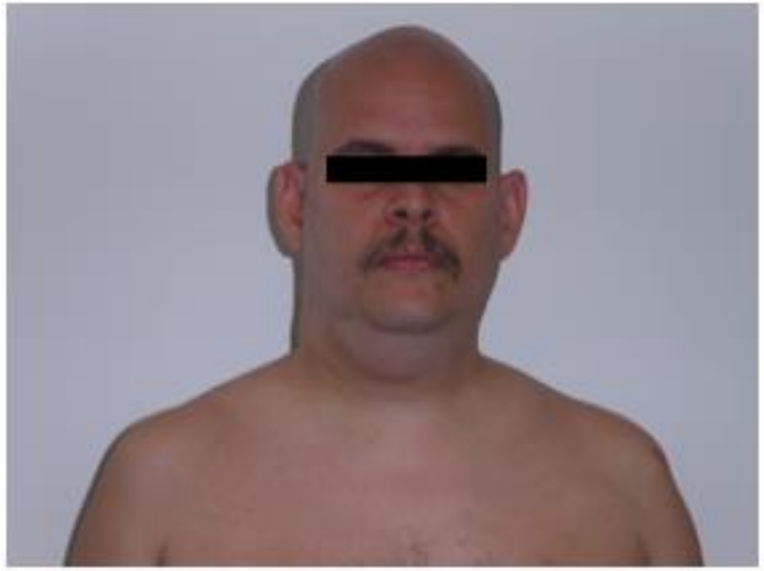
- Mitochondrial disease patients reported an average of 16 symptoms.

Kearns-Sayre syndrome (KSS)

- Progressive external ophthalmoplegia
- Pigmentary retinopathy
- Cardiac conduction block
- Myopathy

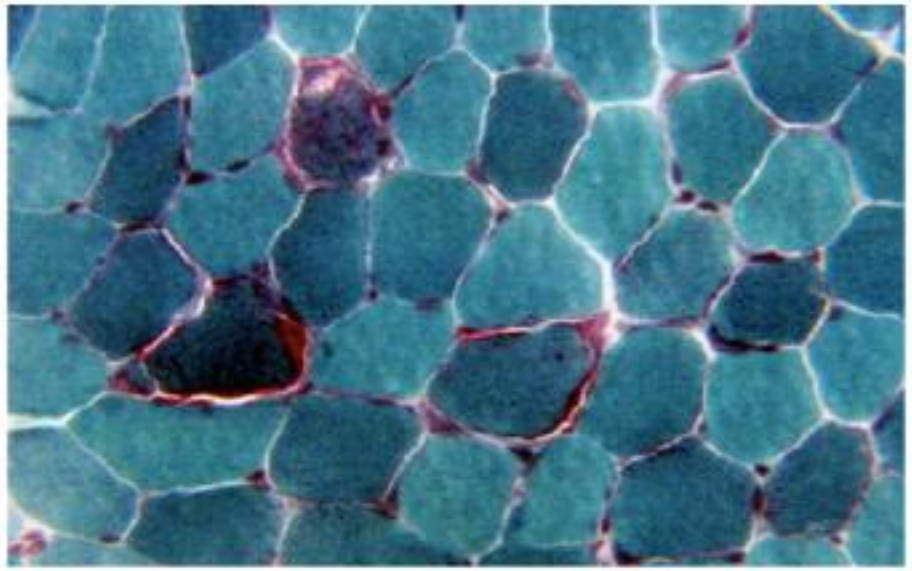






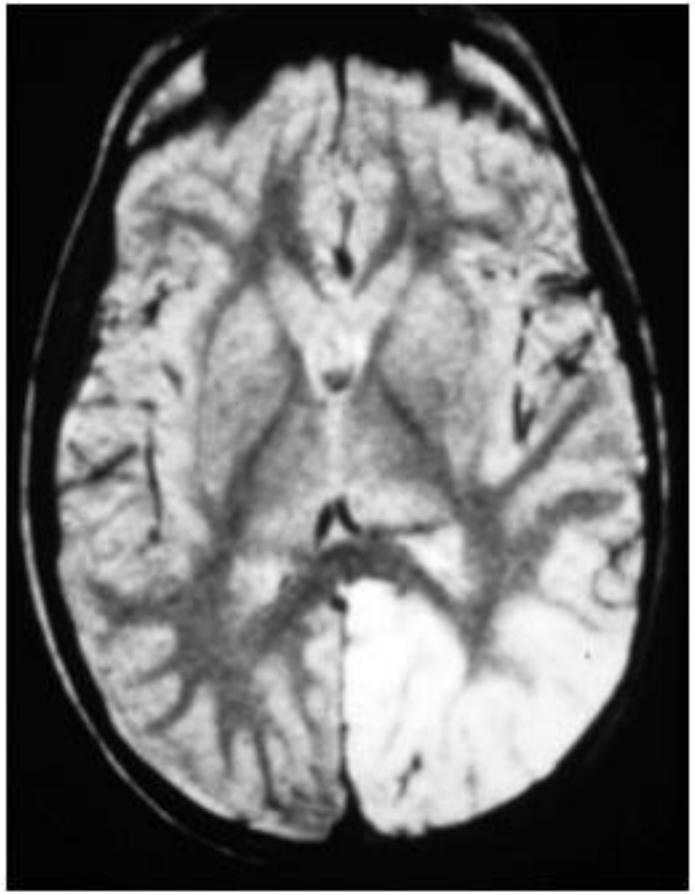
Myoclonus Epilepsy Ragged-Red Fibers (MERRF)

- Myoclonus epilepsy and ataxia
- Ragged-red fibers
- Other features: peripheral neuropathy, lipomas, short stature, hearing loss, and optic atrophy



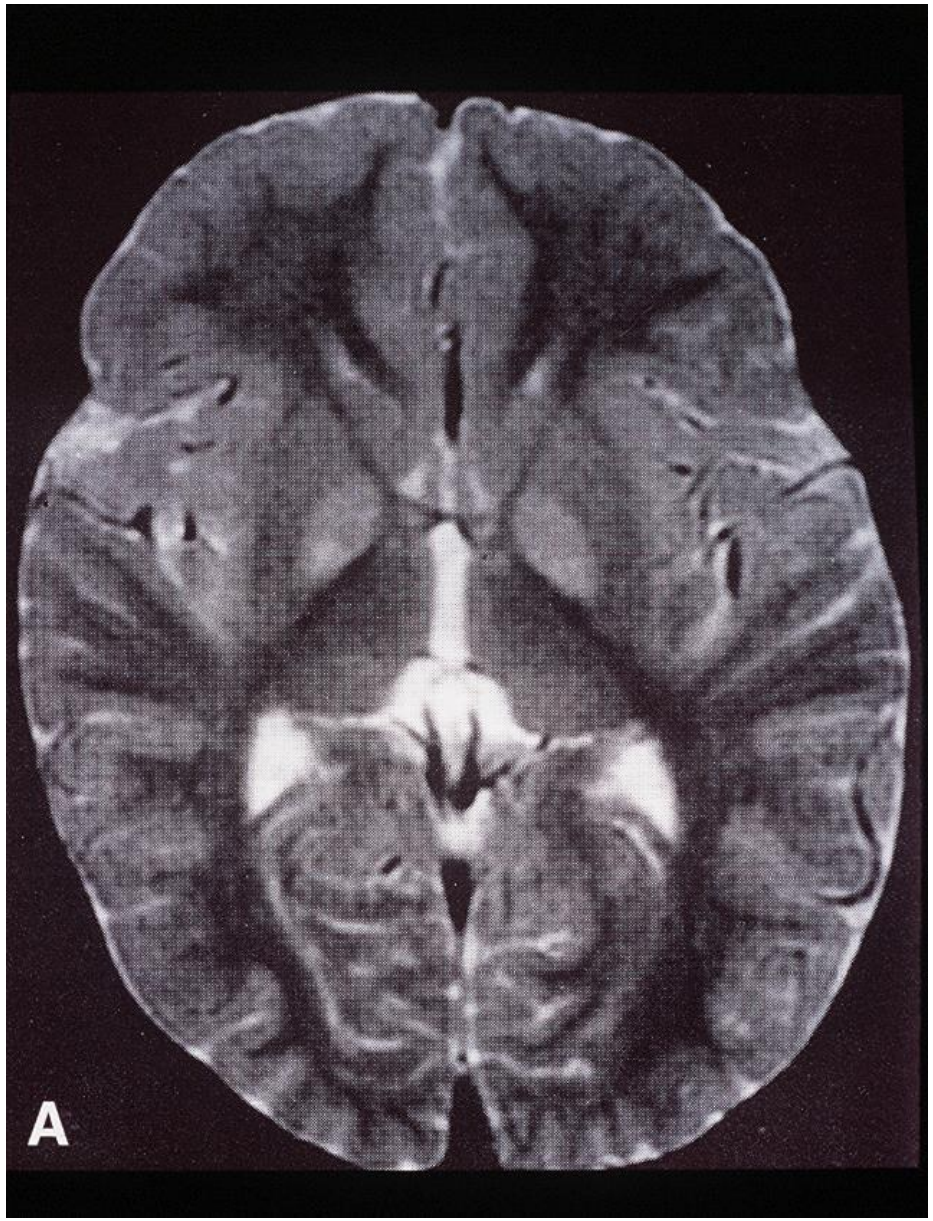
Mitochondrial Encephalopathy, Lactic Acidosis, and Stroke-like episodes (MELAS)

- **Stroke-like episodes at a young age**
- **Encephalopathy manifesting as seizures, dementia, or both**
- **Lactic acidosis, ragged-red fibers, or both**



T2-MRI

Leigh Syndrome



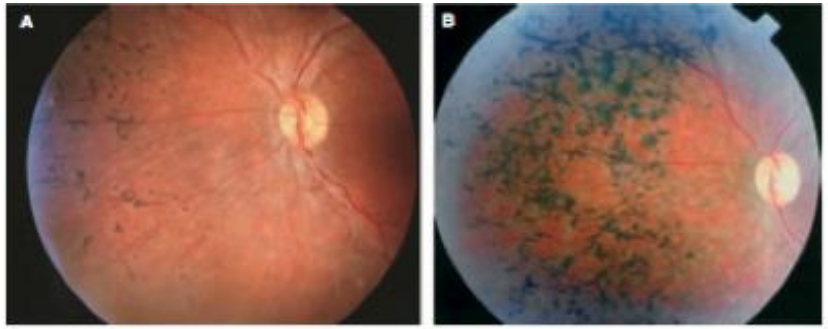
Subacute necrotizing encephalopathy affecting basal ganglia, brainstem, and sparing the mammillary bodies.

Typically begins in infancy with psychomotor regression or retardation.

Other manifestations include: hypotonia, feeding problems, respiratory abnormalities, vision and hearing loss, nystagmus, ataxia, and seizures.

Neuropathy Ataxia Retinitis Pigmentosa (NARP)

- Peripheral neuropathy
- Cerebellar ataxia
- Pigmentary retinopathy
- Maternal inheritance
- Lactic acidosis



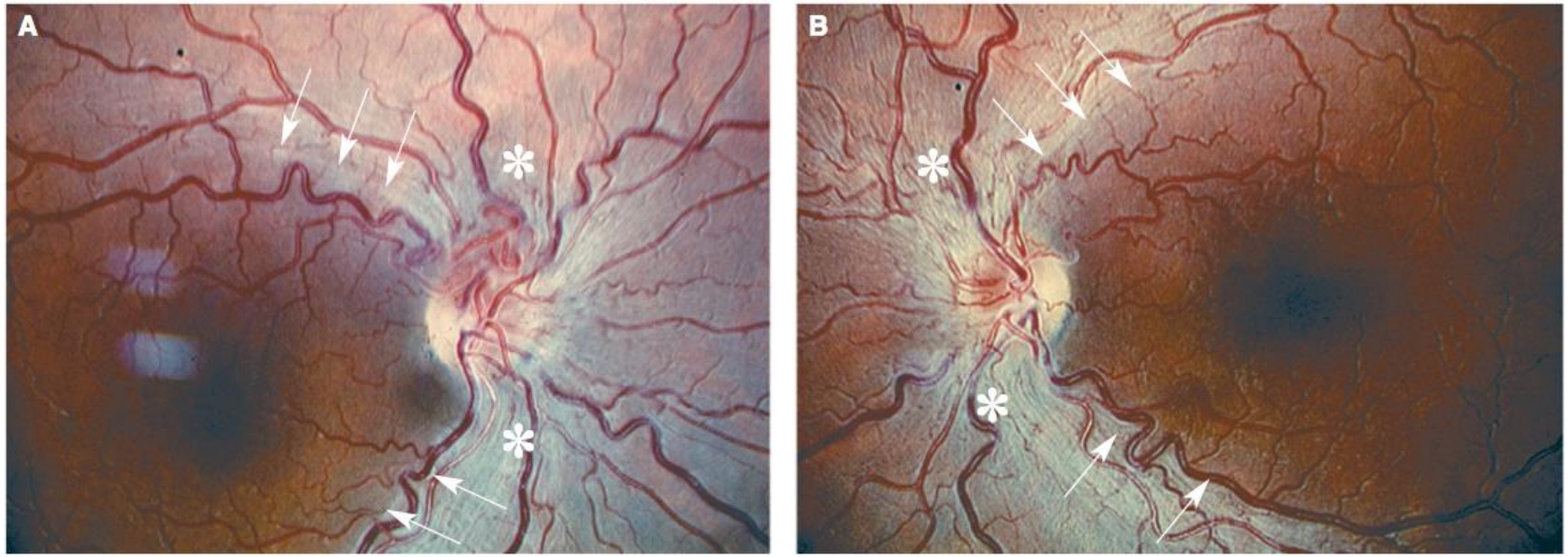
Carelli V, Barboni P, and Sadun AA. "Mitochondrial Ophthalmology" in Mitochondrial Medicine. 2006

Maternally Inherited Leigh Syndrome (MILS)

- Devastating encephalopathy in infancy or childhood
- Psychomotor regression
- Other features include: pigmentary retinopathy, seizures, ptosis, ophthalmoplegia, nystagmus, dystonia, tremor, pyramidal tract signs, ataxia, and impaired respiration.

Leber hereditary optic neuropathy (LHON)

- **Subacute to acute loss of vision**
- **Predominantly affects men (60-90%)**
- **Age at onset is usually 18-30**
- **Clinical features of optic neuropathy**
- **Peripapillary telangectasias are characteristic but not always present**



Carelli et al Mitochondrial Ophthalmology. In: Mitochondrial Medicine

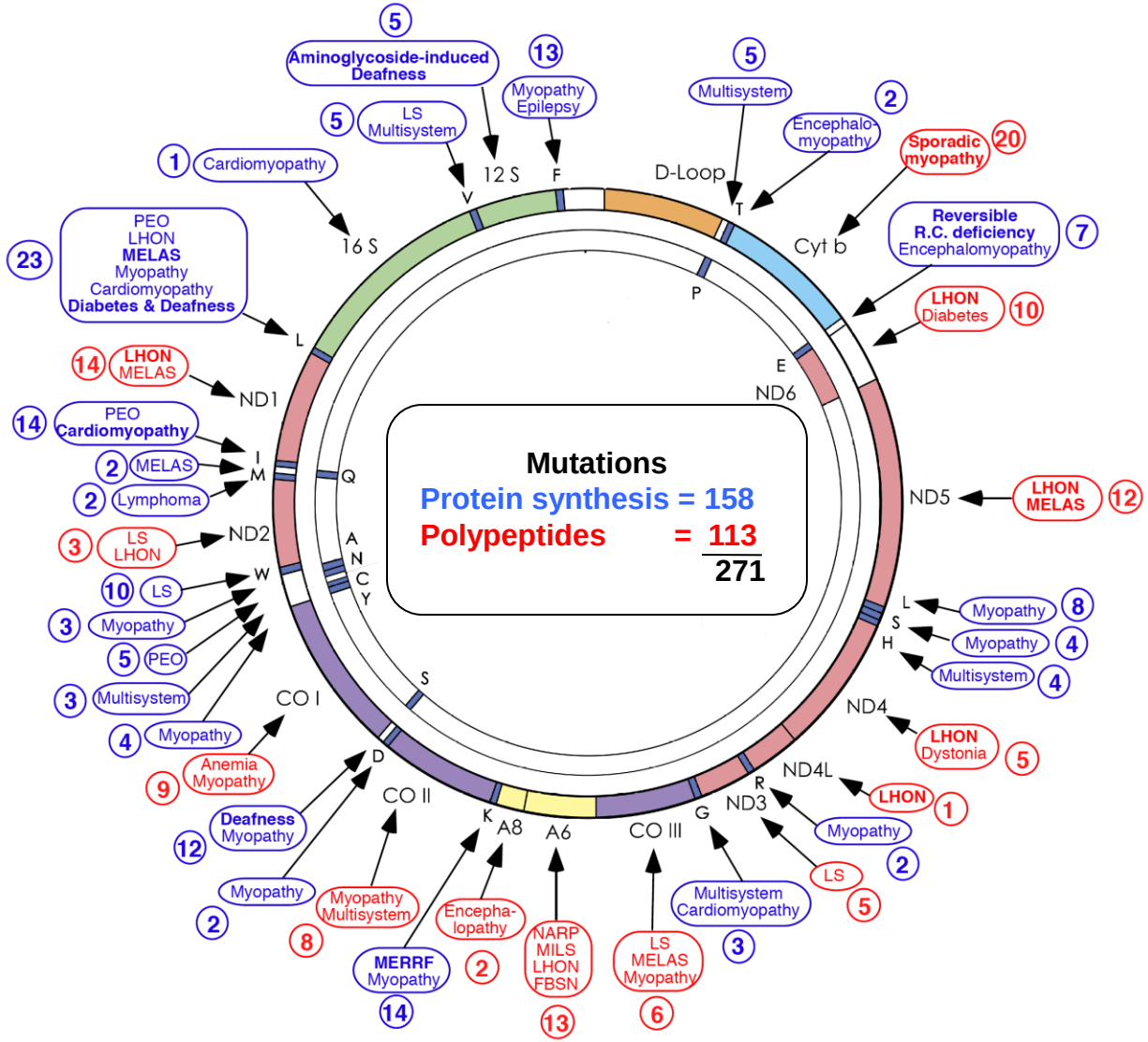
MAJOR SYNDROMES DUE TO mtDNA MUTATIONS

	KSS	MERRF	MELAS	NARP/ MILS	LHON
SEIZURES	-	+	+	+	-
ATAXIA	+	+	-	+	-
STROKE	-	-	+	-	-
MYOCLONUS	-	+	+	-	-
NEUROPATHY	+	+	+	+	-
PEO	+	-	+	-	-
OPTIC NEUROPATHY	-	+	-	-	+
RETINOPATHY	+	-	+	+	-
HEART BLOCK	+	-	-	-	-
WPW	-	-	+	-	+

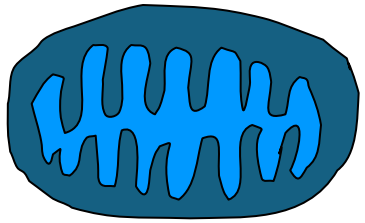
MAJOR SYNDROMES DUE TO mtDNA MUTATIONS

	KSS	MERRF	MELAS	NARP	LHON
SEIZURES	-	+	+	+	-
ATAXIA	+	+	-	+	-
STROKE	-	-	+	-	-
MYOCLONUS	-	+	+	-	-
NEUROPATHY	+	+	+	+	-
PEO	+	-	+	-	-
OPTIC NEUROPATHY	-	+	-	-	+
RETINOPATHY	+	-	+	+	-
HEART BLOCK	+	-	-	-	-
WPW	-	-	+	-	+
mtDNA mutation	Single deletion	m.8344A>G	m.3243A>G	m.8993T>G m.8993T>C	m..11778G>A m.3460G>A m.14484T>C

Mitochondrial morbidity map - 2025

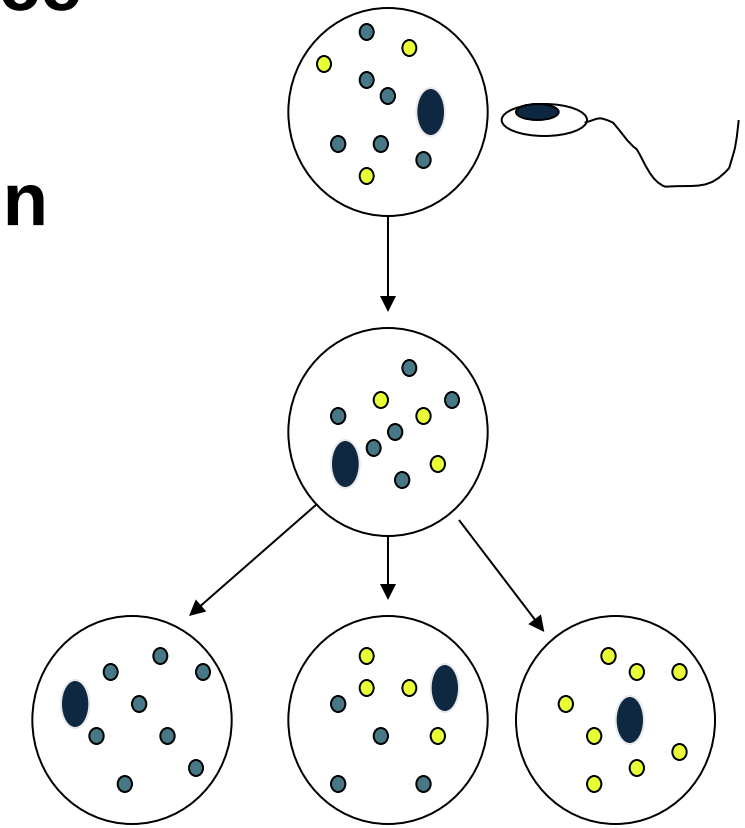


Courtesy of E.A. Schon



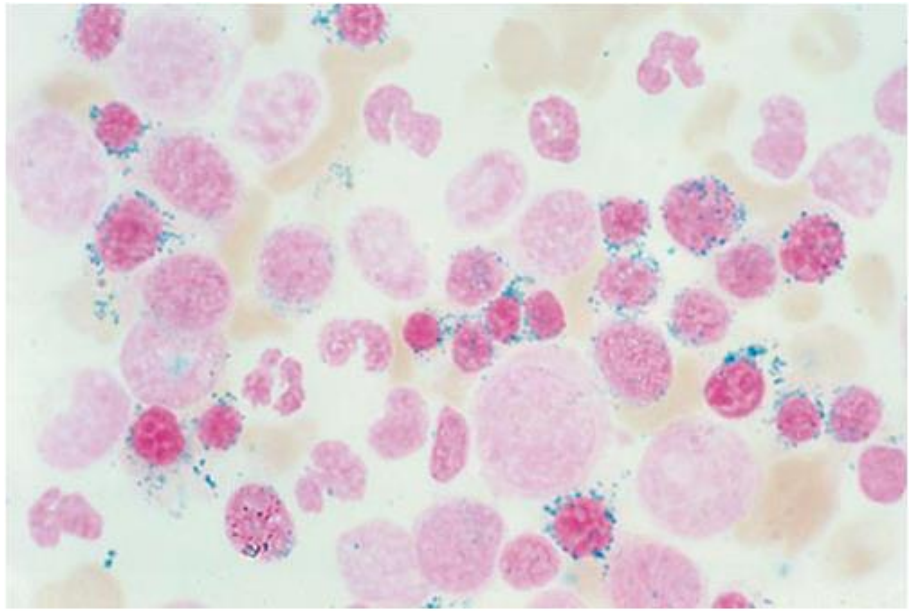
Mitochondrial DNA Rules

- **Maternal inheritance**
- **Heteroplasmy**
- **Mitotic Segregation**
- **Threshold Effect**



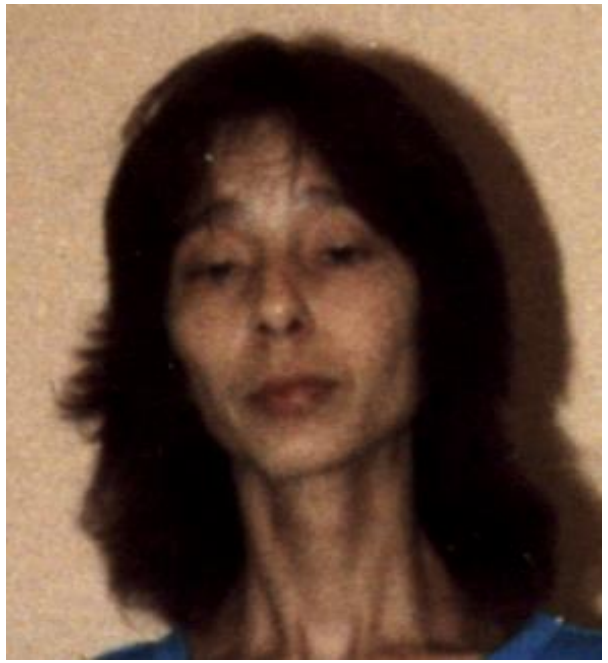
Kearns-Sayre syndrome and Pearson syndrome: Two phenotypes due to one genotype

Ring sideroblasts in sideroblastic anemia



Tefferi A, Li C. In *Atlas of Clinical Hematology.*
Edited by JO Armitage

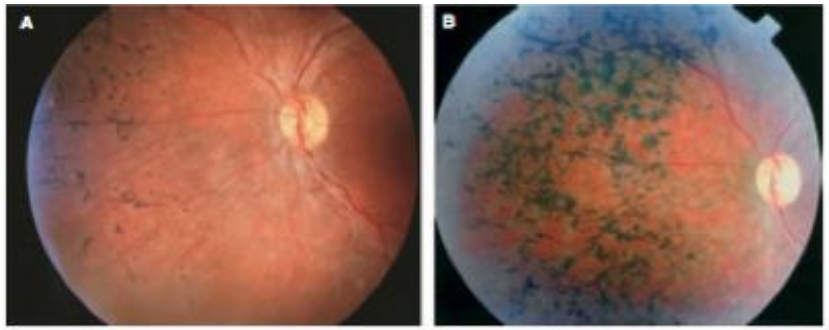
KSS



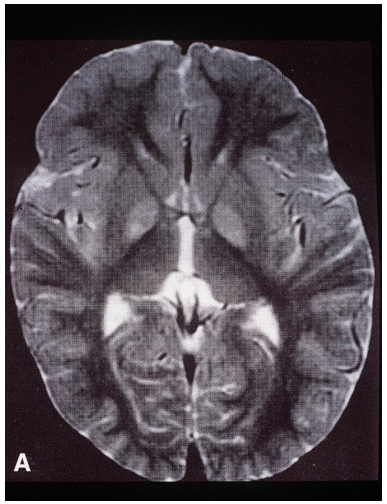
Heteroplasmy matters

m.8993T>G and m.8993T>C mtDNA mutations

Neuropathy Ataxia Retinitis Pigmentosa (NARP)



Maternally Inherited Leigh Syndrome (MILS)



>90% mutation load

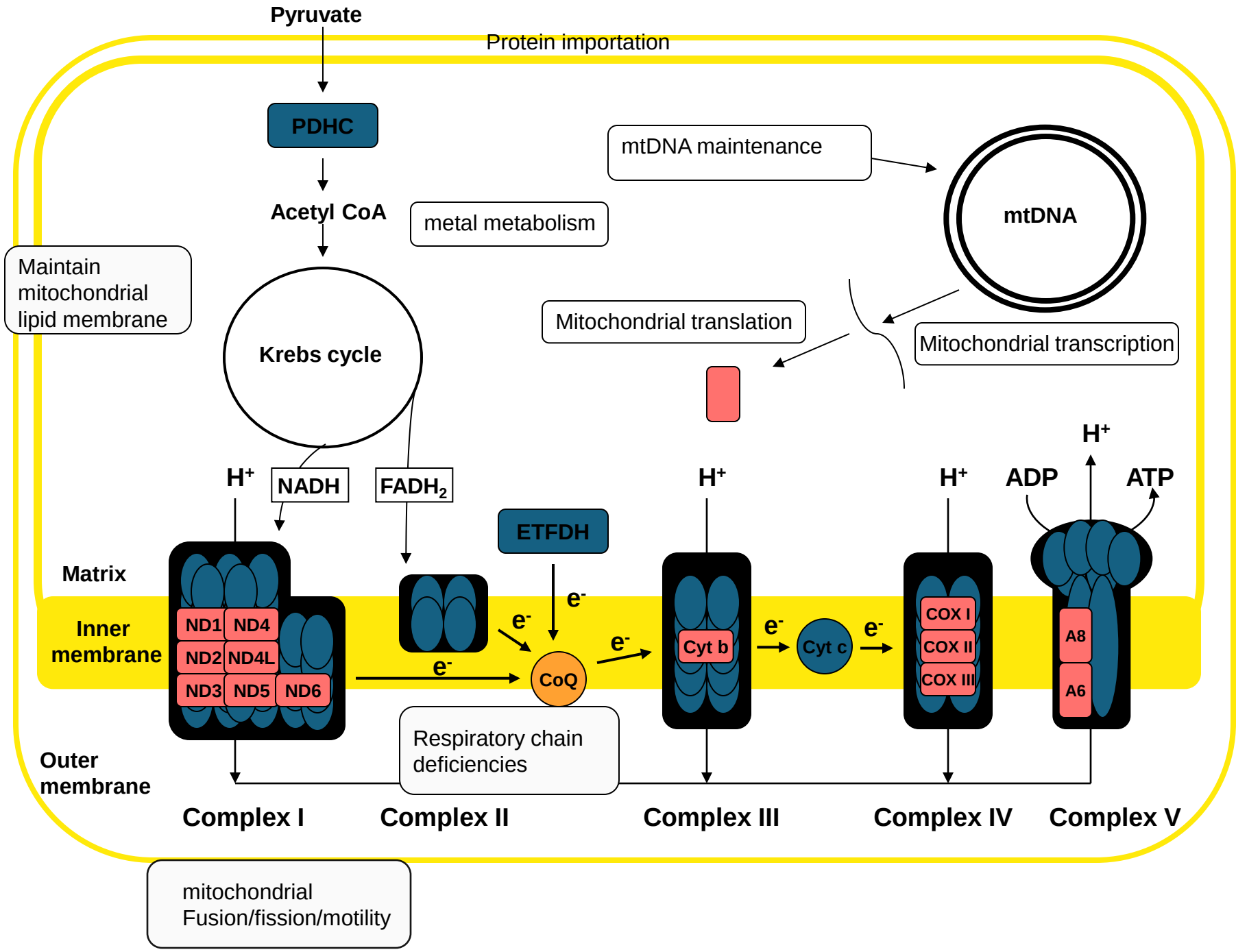
>250 nDNA mitochondrial disease genes and the number is expanding by about 1-2 per month

Table 1 | Products of genes that are known to be mutated in respiratory chain disorders grouped by pathway

Oxidative phosphorylation subunits	mtDNA maintenance and expression	Oxidative phosphorylation biogenesis and regulation	Nucleotide transport and synthesis	Membrane dynamics and composition
Nuclear encoded				
Complex I: NDUFA1, NDUFA2, NDUFA9, NDUFA10, NDUFA11, NDUFA12, NDUFB3, NDUFB9, NDUFS1, NDUFS2, NDUFS3, NDUFS4, NDUFS6, NDUFS7, NDUFS8, NDUFV1, NDUFV2 **Complex II**: SDHA, SDHB, SDHC, SDHD **Complex III**: UQCRB, UQCRQ **Complex IV**: COX4I2, COX6B1 **Complex V**: ATP5E	TWINKLE, MTFMT, GFM1, LRPPRC, MPV17, MRPS16, MRPS22, POLG, POLG2, TRMU, TSFM, TUFM, C12orf65, MTPAP, MRPL3, SARS2, YARS2, HARS2, MARS2, AARS2, RARS2, EARS2, DARS2, TACO1, MTO1, RMND1, PNPT1, PUS1	**Complex I**: NDUFAF1, NDUFAF2, NDUFAF3, NDUFAF4, NDUFAF5, NDUFAF6, ACAD9, FOXRED1, NUBPL **Complex II**: SDHAF1, SDHAF2 **Complex III**: BCS1L, HCCS, TTC19 **Complex IV**: COX10, COX15, ETHE1, FASTKD2, SCO1, SCO2, SURF1, COX14, COA5 **Complex V**: ATPAF2, TMEM70 **Fe–S**: ABCB7, FXN, ISCU, NFU1, BOLA3, GLRX5 **Other**: DNAJC19, GFER, HSPD1, SPG7, TIMM8A, AIFM1, AFG3L2	DGUOK, RRM2B, SLC25A3, ANT1, SUCLA2, SUCLG1, TK2, TYMP	ADCK3, AGK, COQ2, COQ6, COQ9, DRP1, MFN2, OPA1, PDSS1, PDSS2, TAZ, SERAC1
mtDNA encoded				
Complex I: ND1, ND2, ND3, ND4, ND4L, ND5, ND6 **Complex III**: CYTB **Complex IV**: COX1, COX2 **Complex V**: ATP6, ATP8	12S rRNA, tRNATyr, tRNATrp, tRNAVal, tRNAThr, tRNASer1, tRNASer2, tRNAArg, tRNAGln, tRNAPro, tRNAAsn, tRNAMet, tRNALeu1, tRNALeu2, tRNALys, tRNAIle, tRNAHis, tRNAGly, tRNAPhe, tRNAGlu, tRNAAsp, tRNACys, tRNAAla			

List of gene products was generated through synthesis of existing compilations of genes known to be mutated in respiratory-chain disease[47,48], as well as review of the literature.

Vafai and Mootha, Nature 2012


Pyruvate
Protein importation
PDHC
Acetyl CoA
metal metabolism
mtDNA maintenance
mtDNA
Maintain mitochondrial lipid membrane
Krebs cycle
Mitochondrial translation
Mitochondrial transcription
H+
NADH
FADH2
ETFDH
H+
H+
ADP
ATP
H+
Matrix
Inner membrane
ND1
ND4
ND2
ND4L
ND3
ND5
ND6
e-
e-
e-
CoQ
e-
Cyt b
e-
Cyt c
e-
COX I
COX II
COX III
A8
A6
Outer membrane
Respiratory chain deficiencies
Complex I
Complex II
Complex III
Complex IV
Complex V
mitochondrial Fusion/fission/motility

Genetic Classification of Mitochondrial Disorders

Box 2 | Nuclear gene defects in mitochondrial diseases and their function

Phospholipid metabolism
AGK, SERAC1 and TAZ

Metabolism of toxic compounds
HIBCH, ECHS1, ETHE1 and MPV17

Disulfide relay system
GFER

Iron–sulfur protein assembly
ISCU, BOLA3, NFU1 and IBA57

tRNA modification
MTO1, GTP3BP, TRMU, PUS1, MTFMT, TRIT1, TRNT1 and TRMT5

Aminoacyl-tRNA synthetases
AARS2, DARS2, EARS2, RARS2, YARS2, FARS2, HARS2, LARS2, VARS2, TARS2, IARS2, CARS2, PARS2, NARS2, KARS, GARS, SARS2 and MARS2

Release factors
C12orf65

Elongation factors
TUFM, TSFM and GFM1

Mitoribosomal proteins
MRPS16, MRPS22, MRPL3, MRP12 and MRPL44

mRNA processing
LRPPRC, TACO1, ELAC2, PNPT1, HSD17B10, MTPAP and PTCD1

Mitochondrial fusion and fission
OPA1 and MFN2

Deoxynucleotide triphosphate synthesis
DGUOK, TK2, TYMP, MGME1, SUCLG1, SUCLA2, RNASEH1, C10orf2, POLG, POLG2, DNA2 and RRM2B

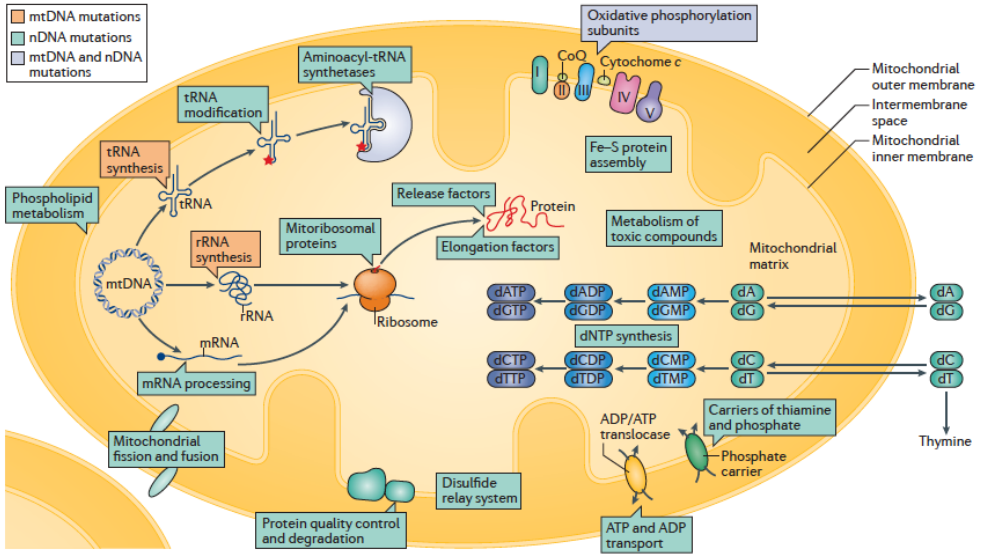
Solute carriers of thiamine and phosphate
SLC19A3, SLC25A3 and SLC25A19

Respiratory chain subunits

- Complex I: *NDUFS1, NDUFS2, NDUFS3, NDUFS4, NDUFS6, NDUFS7, NDUFS8, NDUFV1, NDUFV2, NDUFA1, NDUFA2, NDUFA9, NDUFA10, NDUFA11, NDUFA12, NDUFA13, NDUFAF2, NDUFAF6 and NDUFB11*
- Complex II: *SDHA, SDHB, SDHC, SDHD and SDHAF1*
- Complex III: *UQCRB, BCS1L, UQCRQ, UQCRC2, CYC1, TTC19, LYRM7, UQCC2 and UQCC3*
- Complex IV: *COA5, SURF1, COX10, COX14, COX15, COX20, COX6B1, FASTKD2, SCO1, SCO2, LRPPRC, TACO1 and PET100*
- Complex V: *ATPAF2, TMEM70, ATP5E and ATP5A1*
- Coenzyme Q10 deficiency: *PDSS1, PDSS2, COQ2, COQ4, COQ6, COQ8A, COQ8B and COQ9* (secondary defects: *ETFDH and APTX*)

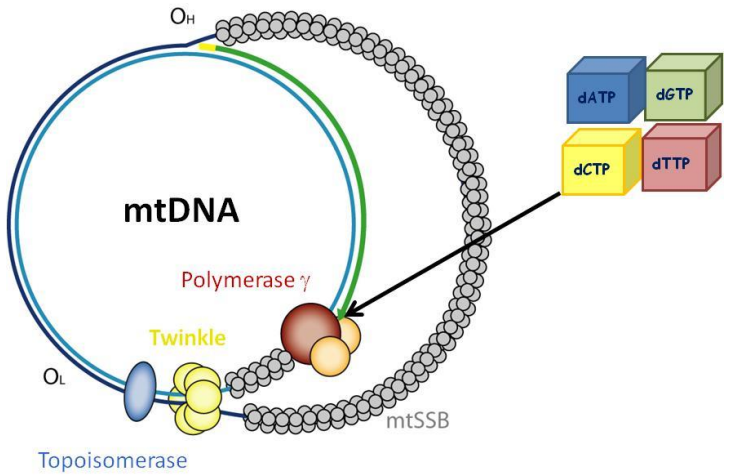
Protein quality control and degradation
FBXL4, AFG3L2 and SPG7

ATP and ADP transport
ANT1



Gorman et al *Nat Rev Dis Primers* 2016

36 genes linked to mtDNA maintenance disorders



mtDNA replication/repair (10 genes)

- ***POLG1***
- *POLG2*
- *TWNK*
- *SSBP1*
- *MGME1*
- *DNA2*
- *RNaseHI*
- *TFAM*
- *TOP3A*
- *LIG3*

Nucleoside/nucleotide metabolism (11 genes)

- *TYMP*
- *TK2*
- *DGUOK*
- *MPV17*
- *GMPR*
- *ABAT*
- *GUK1*
- *RRM1*
- *RRM2B*
- *SUCLA2*
- *SUCLG1*

Mitochondrial dynamics (6 genes)

OPA1
MFN2
SPG7
AFG3L2
MSTO1
MICOS13

Membrane Channels (3 genes)

SCL25A4
SLC25!21
SLC25A10

Other or unknown function (6 genes)

AGK
GFER
FBXL4
MRM2
C1QBP
GMPR

Lopez-Gomez et al *Neuromusc Disorders* 2022
Shintaku et al. *J Clin Invest* 2022
Hildago-Guttierez et al *Ann Neurol* in press

Phenotypic diversity of *POLG* mutations

- **Autosomal dominant or recessive PEO**
- **SANDO (sensory ataxic neuropathy, dysarthria, ophthalmoplegia)**
- **MIRAS (mitochondrial recessive ataxia syndrome)**
- **MEMSA (myoclonic epilepsy myopathy sensory ataxia)**
- **Alpers-Huttenlocher syndrome**
- **Parkinsonism with peripheral neuropathy**
- **Leigh syndrome**
- **Axonal CMT**
- **MNGIE-like disease**
- **MELAS-like disease**
- **MERRF-like disease**

Prevalence of Nuclear and Mitochondrial DNA Mutations Related to Adult Mitochondrial Disease

Gráinne S. Gorman, MRCP,[1,2] Andrew M. Schaefer, MRCP,[1,2] Yi Ng, MRCP,[1,2] Nicholas Gomez,[1,2] Emma L. Blakely, PhD,[1,2] Charlotte L. Alston, PhD,[1,2] Catherine Feeney,[1,2] Rita Horvath, PhD,[1,3] Patrick Yu-Wai-Man, PhD,[1,3] Patrick F. Chinnery, PhD,[1,3] Robert W. Taylor, PhD,[1,2] Douglass M. Turnbull, PhD,[1,2] and Robert McFarland, PhD[1,2]

Ann Neurol, 2015

mtDNA mutations ~1 in 5,000 people
Symptomatic nDNA mutations ~1/34,000

ARTICLE

Pathogenic Mitochondrial DNA Mutations Are Common in the General Population

Hannah R. Elliott,[1] David C. Samuels,[2] James A. Eden,[3] Caroline L. Relton,[3] and Patrick F. Chinnery[1,3,*]

~1 in 200 people carries a mtDNA mutation

Am J Hum Genet, 2008

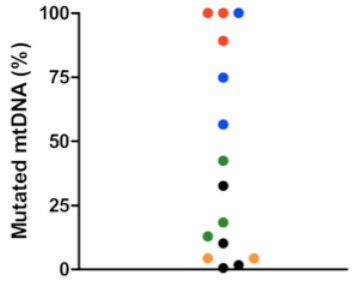


Figure 1. Percentage of Mutated mtDNA in the 15 Mutation-Positive Cases
Red: m.14484T→C; blue: m.11778G→A; green: m.3460G→A; black: m.3243A→G; orange: m.1555A→G.

Pathogenic mtDNA mutations are common in the general population

Number of FDA-approved drugs for primary mitochondrial diseases

0

Thank you for your attention

REAGAN-UDALL FOUNDATION FOR THE FDA

Clinical Trial Design and Implementation: Patient Population Considerations

Anna Choe, MD, MPH

Center for Drug Evaluation and Research

U.S. Food and Drug Administration

Clinical Trial Design: Population Considerations

Anna Choe, MD, MPH
Medical Officer
US FDA - Center for Drug Evaluation and Research
Division of Rare Diseases and Medical Genetics (DRDMG)

5/22/2025

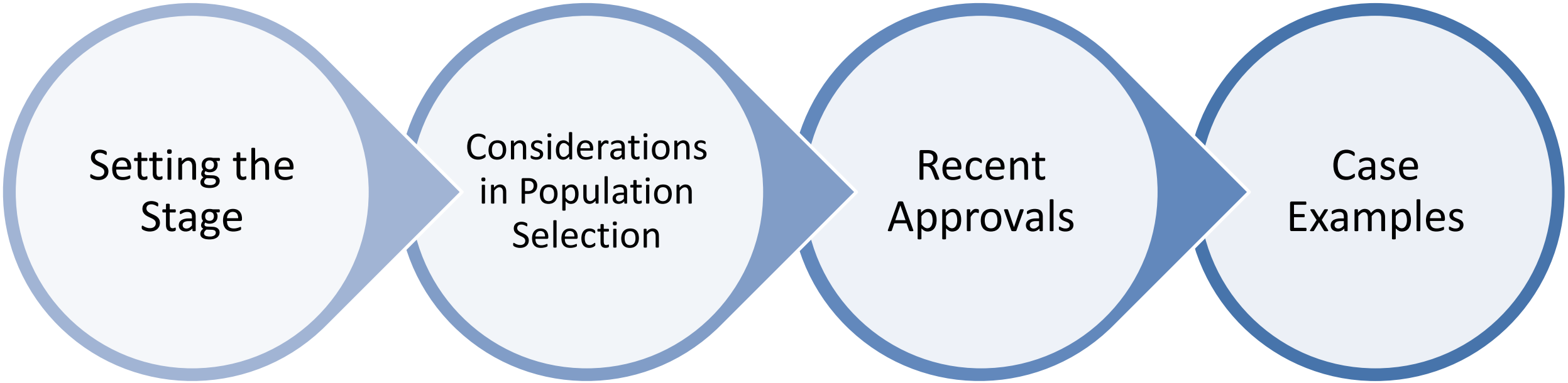
Disclaimer

- My own views and not an official FDA position.

- No financial interests to disclose.

- “Drug” for both drugs and biologics.

Roadmap


Setting the Stage
Considerations in Population Selection
Recent Approvals
Case Examples

To be approved for marketing, FDA must determine that the drug is safe and effective

- "effective" is codified in statute:
  - Demonstrates "substantial evidence that the drug will have the effect it purports or is represented to have under proposed labeled conditions of use" (21CFR 314.125, 21CFR 314.126)

- "safe" is not explicitly defined in statute or regulations
  - Because all drugs can have risks, the demonstration of safety is interpreted as a determination that drug's benefit outweighs its risks

Substantial Evidence of Effectiveness

- Effectiveness established “substantial evidence” (FD&C Act, 1962 amendments)
  - Minimum of 2 adequate and well-controlled studies, each persuasive on its own
- Complimentary statutory standard (FDAMA, 1997)
  - One adequate and well-controlled study and “confirmatory” evidence
- Adequate & Well-Controlled (AWC) Studies
  - Studies designed well enough to be able “to distinguish the effect of a drug from other influences, such as spontaneous change, placebo effect, or biased observation” (21 CFR 314.126)
  - Effect shown in AWC must be clinically meaningful

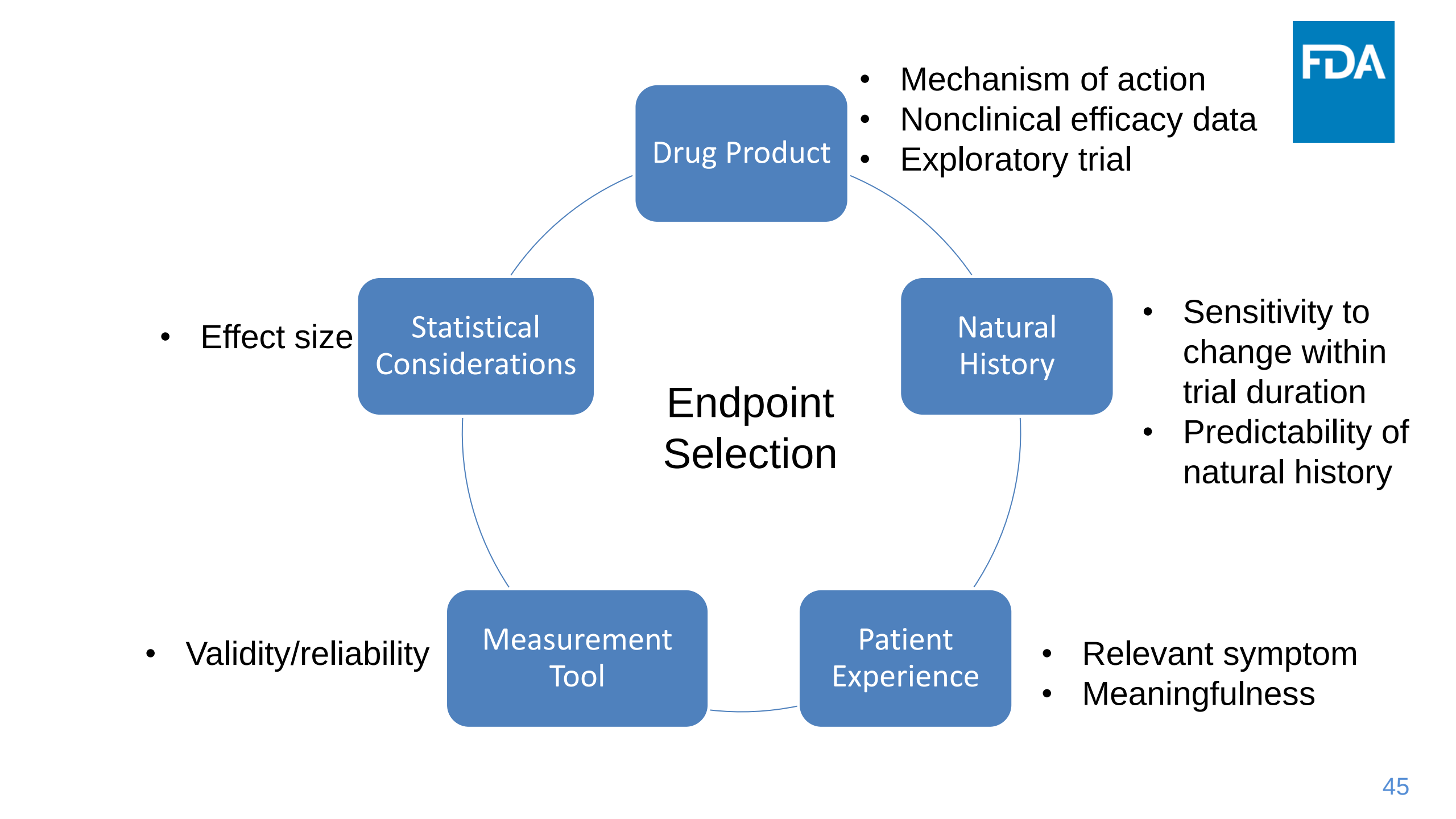
Adequate & Well-Controlled Studies

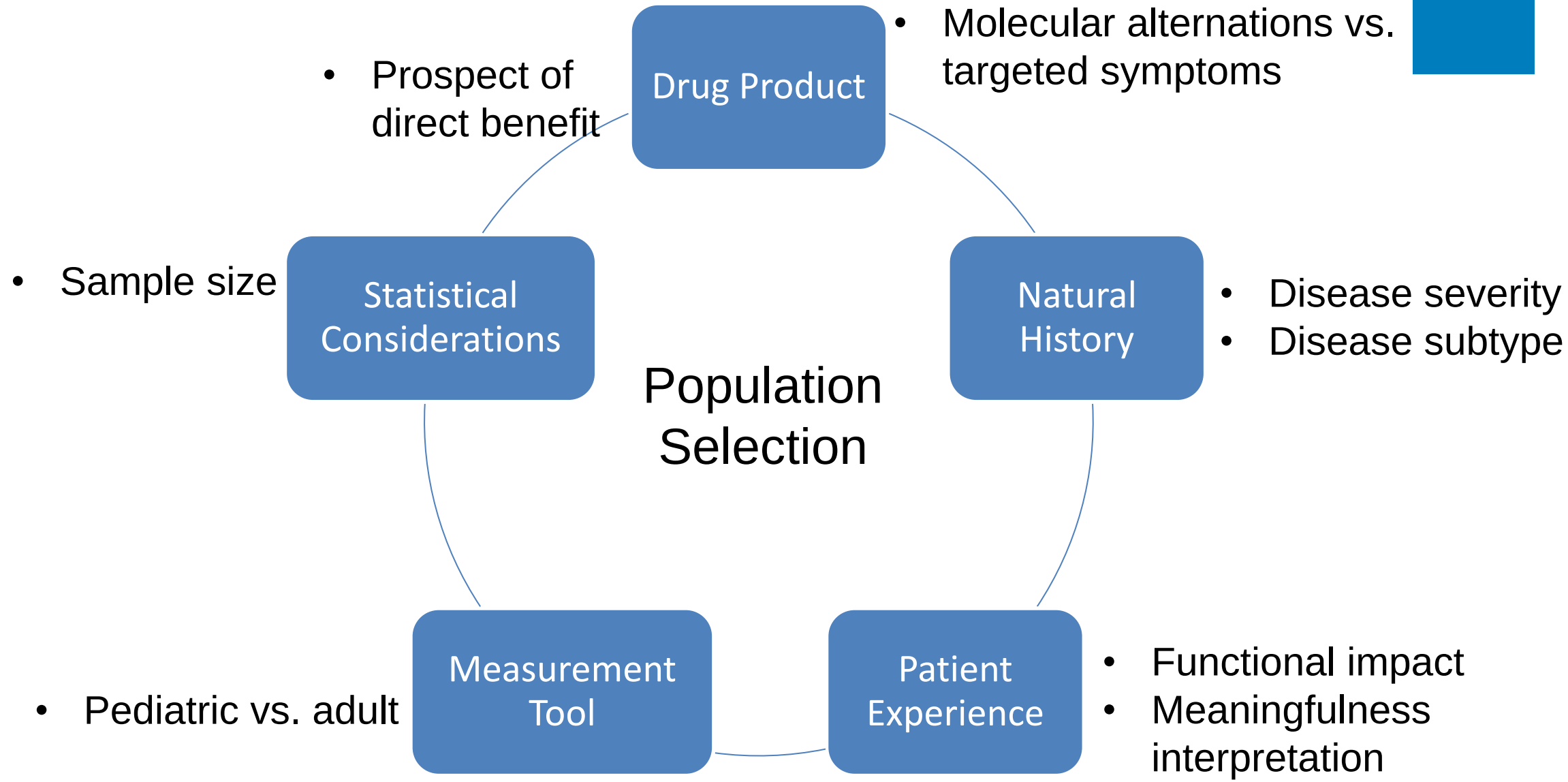
- A clear statement of objectives and methods of analysis
- A design which permits a valid comparison with a control
- Adequate assurance that subjects have the condition being studied
- Adequate measures to minimize bias in subject assignment to treatment group
- Adequate measures to minimize bias on the part of subjects, observers, and analysts of the data
- Well-defined and reliable methods to assess response, and
- Adequate analysis of the results of the study to assess the effect of the drug.

Primary Mitochondrial Diseases (PMD)

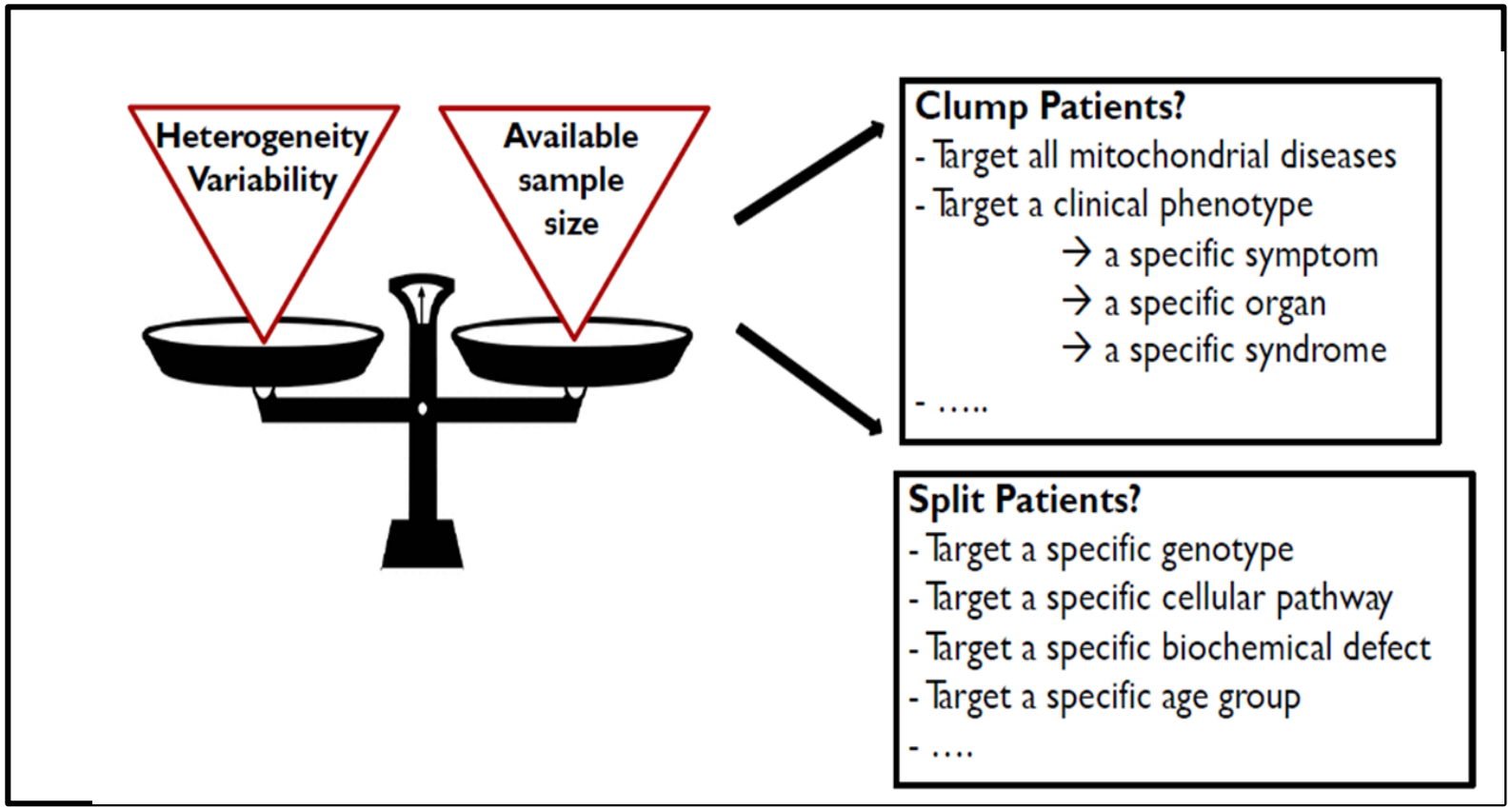
- Rare
- Complex genotype
- Multisystemic disease
- Heterogeneous/variable presentation
- Limitations of available natural history

- To optimize success of AWC in PMD, careful selection of endpoint and trial population is key


Drug Product
• Mechanism of action
• Nonclinical efficacy data
• Exploratory trial
Natural History
• Sensitivity to change within trial duration
• Predictability of natural history
Patient Experience
• Relevant symptom
• Meaningfulness
Measurement Tool
• Validity/reliability
Statistical Considerations
• Effect size
Endpoint Selection


Drug Product
• Molecular alternations vs. targeted symptoms
Natural History
• Disease severity
• Disease subtype
Patient Experience
• Functional impact
• Meaningfulness interpretation
Measurement Tool
• Pediatric vs. adult
Statistical Considerations
• Sample size
• Prospect of direct benefit
Population Selection

Heterogeneity vs. Sample Size



*

* Karaa, A. (2019, September 6). *Patient Population Selection and Consideration for Pediatric Patient Enrollment in Clinical Trials*. Developing Therapies for Primary Mitochondrial Diseases: Bridging the Gaps. https://www.fda.gov/news-events/fda-meetings-conferences-and-workshops/developing-therapies-primary-mitochondrial-diseases-bridging-gaps-09062019-09062019

Recent DRDMG Approvals

- levacetylleucine & arimoclomol approved in 2024
  - Niemann Pick C “The primary manifestations and rate of disease progression is heterogenous”
- cipaglucosidase alfa approved in 2023
  - Late-onset Pompe disease “slowly progressive, heterogeneous late-onset Pompe disease”
- pegunigalsidase alfa-iwxj approved in 2023
  - Fabry disease “The disease course and severity can vary as a function of the phenotype”
- velmanase alfa-tycv approved in 2023
  - Alpha-mannosidosis “symptoms, progression and severity vary widely”
- olipudase alfa-rpcp approved in 2022
  - Acid sphingomyelinase deficiency “The disease presentation and progression rate vary greatly in type A/B patients”

Recent DRDMG NME Approvals

- levacetylleucine & arimoclomol approved in 2024
  - Niemann Pick C "The primary manifestations and rate of dis… heterogenous"
- cipaglucosidase alfa approved in 2023
  - Late-onset Pompe disease "slowly progressive, heterogeneou… disease"
- pegunigalsidase alfa-iwxj approved in 2023
  - Fabry disease "The disease course and severity can vary as a function of the phenotype"
- velmanase alfa-tycv approved in 2023
  - Alpha-mannosidosis "symptoms, progression and severity vary widely"
- olipudase alfa-rpcp approved in 2022
  - Acid sphingomyelinase deficiency "The disease presentation and progression rate vary greatly in type A/B patients"

- 25-123 subjects in AWC
- 6/6 approvals based on RCT trials

Olipudase alfa (Xenpozyme)

- Substantial evidence of effectiveness: one adequate and well controlled clinical trial with confirmatory evidence
  - 52-wk randomized blinded placebo-controlled trial in 31 adults with ASMD (acid sphingomyelinase deficiency) showed a clinically meaningful and statistically significant improvement in lung function (diffusion capacity of the lungs for carbon monoxide (DLCO)), spleen/liver size

- Trial population: adults, ASMD type B, enriched based on primary endpoints

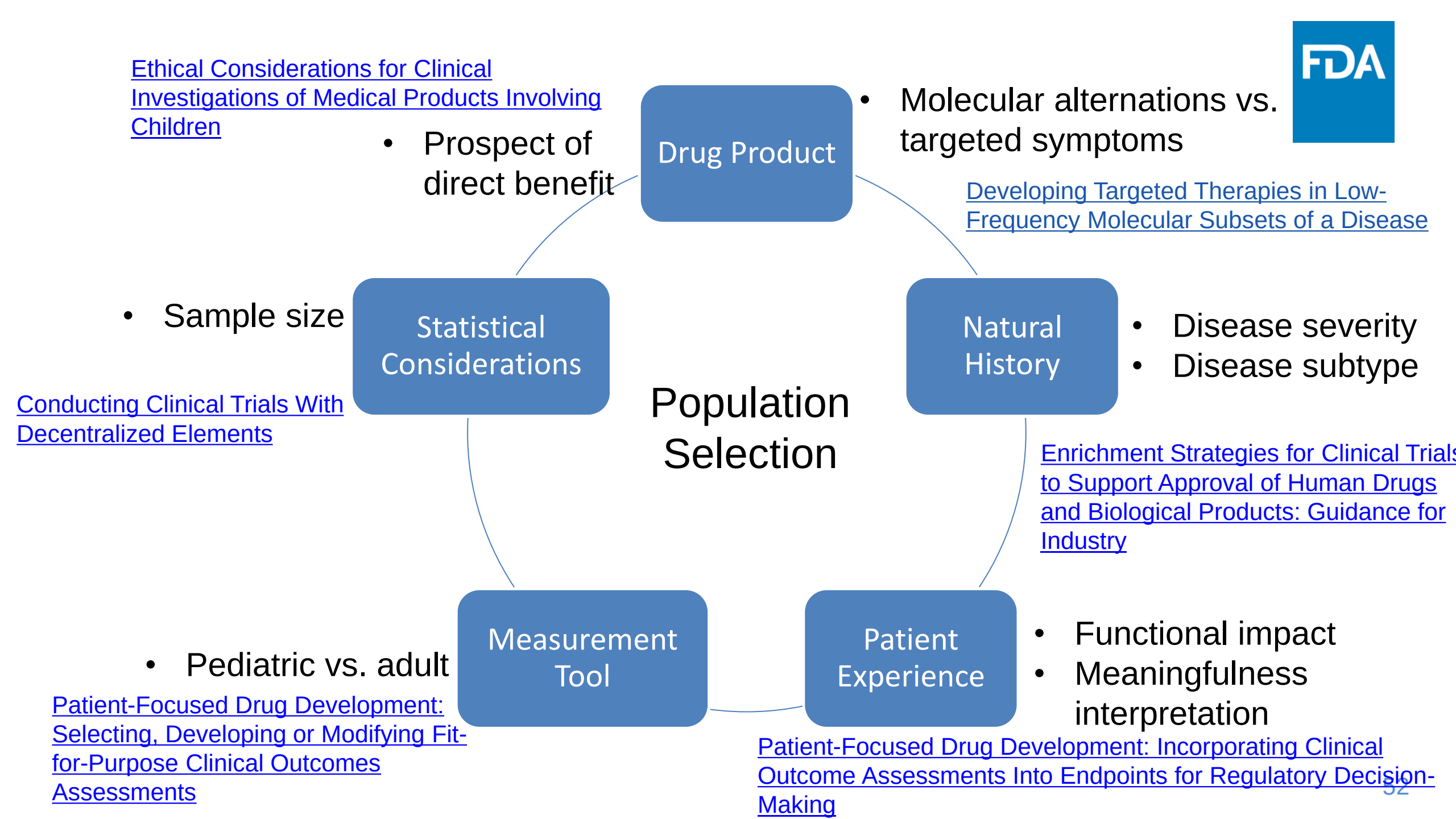
- Indication for treatment of non-central nervous system (CNS) manifestations of ASMD in pediatric and adult patients
  - Includes ASMD type A and pediatric patients given the mechanism of action, disease pathophysiology, and available clinical data

Levacetylleucine (Aqneursa)

- Substantial evidence of effectiveness: one adequate and well controlled clinical trial with confirmatory evidence
  - Multinational, randomized, double-blinded, placebo-controlled, crossover phase 3 trial in 60 subjects showed statistically significant improvement in functional Scale for Assessment and Rating of Ataxia (fSARA)

- Trial population: pediatric and adult patients with cerebellar ataxia from NPC, enriched based on primary endpoint

- Indication for treatment of neurological manifestations of Niemann-Pick disease type C (NPC) in adults and pediatric patients weighing ≥15 kg
  - Indicated for neurological manifestations given different aspects of pragmatic neurological function measured by fSARA


Ethical Considerations for Clinical Investigations of Medical Products Involving Children
• Prospect of direct benefit
Drug Product
• Molecular alternations vs. targeted symptoms
Developing Targeted Therapies in Low-Frequency Molecular Subsets of a Disease
• Sample size
Statistical Considerations
Natural History
• Disease severity
• Disease subtype
Conducting Clinical Trials With Decentralized Elements
Population Selection
Enrichment Strategies for Clinical Trials to Support Approval of Human Drugs and Biological Products: Guidance for Industry
• Pediatric vs. adult
Measurement Tool
Patient Experience
• Functional impact
• Meaningfulness interpretation
Patient-Focused Drug Development: Selecting, Developing or Modifying Fit-for-Purpose Clinical Outcomes Assessments
Patient-Focused Drug Development: Incorporating Clinical Outcome Assessments Into Endpoints for Regulatory Decision-Making

Take-Home Messages

1. Recent approvals in rare, heterogeneous diseases were based on smaller, RCTs.
2. Natural history, MOA, preclinical data, exploratory trial, patient experience, measurement tool, and sample size inform population selection.
3. PMD has unique challenges and opportunities for selection of endpoints and trial population in trial design.
4. Additional strategies can be considered to reduce variability and improve enrollment/retention.

Primary Mitochondrial Disease Drug Development

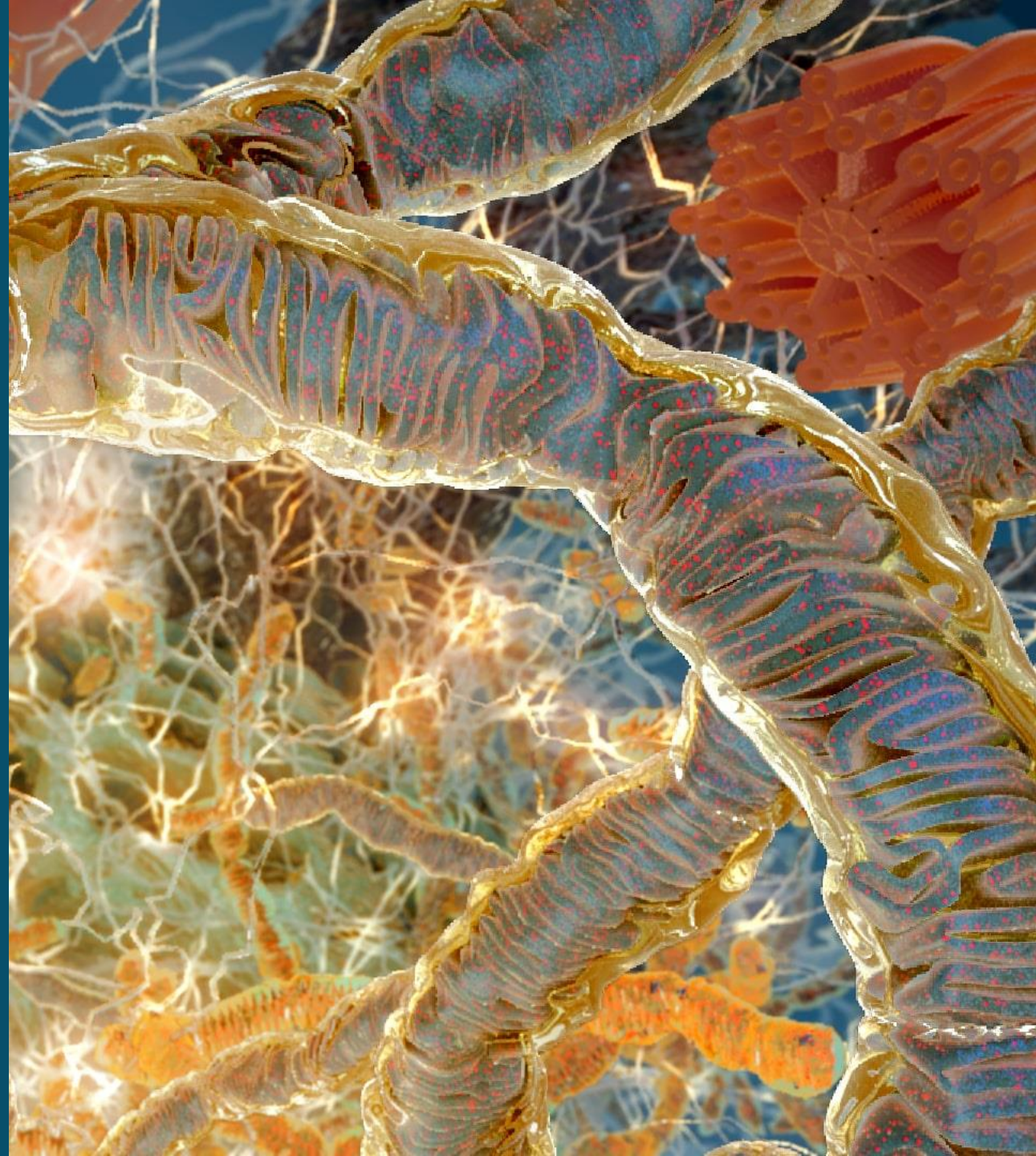
- **Reenie McCarthy, JD**
  Stealth BioTherapeutics

REAGAN-UDALL FOUNDATION FOR THE FDA

Stealth BIOTHERAPEUTICS

Trial Population Considerations: Basket? No basket?

Primary Mitochondrial Diseases Workshop
Reagan-Udall Foundation for the FDA
May 22, 2025



Forward-Looking Statements

We are an “emerging growth company” as defined under the Securities Act of 1933, as amended (the “Act”).

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Key Talking Points

1. Basket/No-basket trial design
   - Clinical disease presentation may influence decision whether to study single affected gene (“no basket”) or multiple pathogenic genetic variants (“basket”) in a clinical trial
   - Mechanism of action (MOA) & ADME properties of therapeutic agent also relevant to basket/no-basket decision
2. Trial enrichment strategies to reduce heterogeneity in basket design
   - Targeting common phenotypes across diverse genotypes is highly dependent on investigator rigor in trial recruitment, expert consensus on classifications & detailed natural history characterization
   - Multiple approaches including prescriptive parameters to normalize degree of impairment, reduce variability of genotypic spectrum & optimize identification of target organ systems have had limited practical success historically
   - Balancing basket trial populations by genotype may enable detection of signals in subgroups (as recommended by FDA)
- Conclusions & future considerations: patient reported outcome assessments (PROs) may be inherently well-suited to detect efficacy signals in patient populations with highly heterogenous clinical presentation

Pioneering Mitochondria-Targeted Therapies

Late-state pipeline of novel drug candidates targeting rare & age-related diseases

We have clinical experience with elamipretide, our lead investigational product, in:

Primary Mitochondrial Diseases

- Barth syndrome – NDA filed & under review by FDA
- Leber's hereditary optic neuropathy – signals in phase (P) 2 trial;[1] did not proceed to P3 due to concerns with interventional window (first year?) & con-med management (idebenone)
- Primary mitochondrial myopathy – signals in P1 & P2 trials;[2] P3 signal in subgroup with majority POLG-1 genotypic representation;[3] new P3 initiated

Secondary Mitochondrial Diseases

- Dry age-related macular degeneration – in P3 development

Elamipretide | Safety well-characterized | **>1,700 patients** exposed to once-daily SC injections | >400 patient years of exposure; **>7 years** exposure for some patients

Pipeline | **Bevemipretide**
SBT-255
SBT-580 series

1. Karanjia, Sadun, Elamipretide Topical Ophthalmic Solution for the Treatment of Subjects with Leber Hereditary Optic Neuropathy: A Randomized Trial, Clinical Trial, Ophthalmology 2024; 2. Karaa et al., Randomized dose-escalation trial of elamipretide in adults with primary mitochondrial myopathy, Neurology, 2018; Karaa et al., A randomized crossover trial of elamipretide in adults with primary mitochondrial myopathy, Journal of Cachexia, Sarcopenia and Muscle, 2020; 3. Karaa et al., Efficacy and Safety of Elamipretide in Individuals With Primary Mitochondrial Myopathy: The MMPOWER-3 Randomized Clinical Trial, Neurology, 2023; Karaa et al., Genotype-specific effects of elamipretide in patients with primary mitochondrial myopathy: a post hoc analysis of the MMPOWER-3 trial, Orphanet Journal of Rare Diseases, 2024; 4. Baseline demographics for NuPOWER trial; data analysis pending

To Basket or Not to Basket?

Influence of clinical disease presentation on trial design: Barth syndrome & LHON examples

Hornby *et al.*
Orphanet Journal of Rare Diseases (2022) 17:336
https://doi.org/10.1186/s13023-022-02469-5

Orphanet Journal of Rare Diseases

RESEARCH Open Access

Natural history comparison study to assess the efficacy of elamipretide in patients with Barth syndrome

Brittany Hornby[1], William Reid Thompson[2], Mohammed Almuqbil[3], Ryan Manuel[4], Anthony Abbruscato[5], Jim Carr[5] and Hilary J. Vernon[4*]

Abstract

Background: Natural history studies are increasingly recognized as having an important role in drug development for rare diseases. A phase 3, observational, retrospective, and non-interventional study was designed to establish a natural history control (NHC) cohort of patients with Barth syndrome (BTHS) to provide further analysis of the efficacy of elamipretide observed in an open-label extension (OLE) phase of the TAZPOWER trial, a clinical trial that tested the efficacy of 40 mg daily of elamipretide in patients with BTHS.

Methods: This was a retrospective, non-interventional study. A propensity score model was used to compare elamipretide-treated patients and NHCs. The analysis included 8 patients from the TAZPOWER OLE and 19 untreated NHCs (including 12 with serial echocardiographic assessments).

Results: For the 6-min walk test (6MWT, primary endpoint), the least squares (LS) mean difference between groups was 79.7 m ($P=0.0004$) at week 64 and 91.0 m ($P=0.0005$) at week 76 in favor of elamipretide. Significant improvements in muscle strength (secondary endpoint), as assessed by handheld dynamometry (HHD) were also observed with elamipretide, with LS mean differences of 40.8 Newtons at 64 weeks ($P=0.0002$) and 56.7 Newtons at 76 weeks ($P=0.0005$). Patients continuously treated with elamipretide also experienced statistically significant improvements in other secondary endpoints (i.e., 5 times sit-to-stand [5XSST], multi-domain responder index [MDRI]). The functional improvements were robust to sensitivity analyses. Left ventricular stroke volume increased from baseline in patients with elamipretide but decreased in NHCs.

Conclusions: Overall, the study established a NHC for use in assessing the efficacy of therapeutic interventions in patients with BTHS and the results suggest that elamipretide may improve natural history of BTHS at least in part by attenuating the natural decline in heart function and provide meaningful improvements in heart function and functional capacity in patients with BTHS compared to NHCs.

Highlights:

- A matched Natural History Control (NHC) was used to evaluate elamipretide in BTHS
- Elamipretide may improve natural history of BTHS by attenuating natural decline in heart function

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AMERICAN ACADEMY OF OPHTHALMOLOGY®

Elamipretide Topical Ophthalmic Solution for the Treatment of Subjects with Leber Hereditary Optic Neuropathy

A Randomized Trial

Rustum Karanjia, MD, PhD,[1,2,3,4] *Alfredo A. Sadun, MD, PhD*[1,2]

Purpose: This study aimed to assess the safety, tolerability, and potential efficacy of topical elamipretide in patients affected with Leber hereditary optic neuropathy (LHON).

Design: This phase II, prospective, randomized, vehicle-controlled, single-center clinical trial involved administration of elamipretide 1% topical ophthalmic solution to patients with LHON over a 52-week double-masked treatment period, followed by an open-label extension (OLE) for up to 108 additional weeks of treatment.

Participants: Twelve patients with LHON were included in this study. Patients aged 18 to 50 years with decreased vision for at least ≥ 1 year and ≤ 10 years, and a genetically confirmed diagnosis of m.11778G>A LHON were eligible for this trial.

Methods: For the first 52 weeks of the study, patients were randomized to 1 of 3 groups: elamipretide in both eyes or elamipretide in 1 eye (left eye and right eye were considered separate groups) and vehicle in the other eye, followed by an OLE in which both eyes were treated with elamipretide.

Main Outcome Measures: The primary outcome measure was assessment of adverse events (AEs) from the administration of topical elamipretide, and the primary efficacy end point was change in best-corrected visual acuity (BCVA). Secondary outcome measures included changes in color vision, visual field mean deviation, and electrophysiological outcomes.

Results: Elamipretide was well tolerated with the majority of AEs being mild to moderate and resolving spontaneously. The change from baseline in BCVA in elamipretide-treated eyes was not significantly different from the vehicle eyes at any time point. Six of 12 subjects met the criteria for clinically relevant benefit (CRB). In the post hoc analysis, change from baseline in mean deviation in the central visual field was significantly greater in elamipretide-treated eyes versus the vehicle eyes. Compared with baseline, both treatment groups showed improvement in color discrimination and contrast sensitivity in the OLE.

Conclusions: Elamipretide treatment was generally well tolerated, with no serious AEs reported. Although this study did not meet its primary BCVA efficacy end point, improvements across assessments on visual function during the OLE and the post hoc findings of the Humphrey automated visual field central region were encouraging and require further exploration.

Financial Disclosure(s): The author(s) have no proprietary or commercial interest in any materials discussed in this article. *Ophthalmology 2024;131:422-433 © 2023 by the American Academy of Ophthalmology. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).*

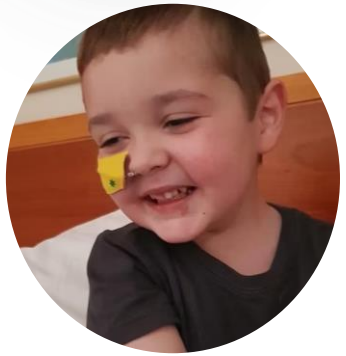
Supplemental material available at www.aaojournal.org.

Leber hereditary optic neuropathy (LHON) is the most common hereditary optic neuropathy, and it is characterized by subacute asynchronous severe bilateral loss of vision.[1-3] Unlike other hereditary optic neuropathies, not all subjects who carry a causative LHON variant develop vision loss, and some remain carriers of the variant. Conversion to the affected state is characterized by a painless loss of visual acuity and color vision and the development of a central scotoma. The central scotoma expands during the initial subacute phase, which lasts for the first 6 months. A nadir is typical and reached by approximately 8 months, at which point the visual acuity stabilizes and defines the end of the dynamic phase of disease onset. The chronic phase begins after this and is characterized by relative stability of all clinical metrics, followed by a slow decline in visual acuity over the ensuing decades.[4-6] The visual prognosis of

  https://doi.org/10.1016/j.ophtha.2023.10.033

Mechanism of Action/Disease Considerations in Trial Design

We see therapeutic concentrations of elamipretide in peripheral tissues, but not in the brain; the POLG example



"Liam the Lion"
ABC-Aus

Infantile/pediatric-onset POLG disorders including Alpers-Huttenlocher syndrome (AHS) & childhood myocerebrohepatopathy spectrum (MCHS) often involve damage to the brain & are characterized by seizures, liver failure, & developmental delays

Elamipretide is unlikely to impact central nervous system (CNS) sequelae

Prince Frederik
The POLG Foundation

Adolescent & early-adult-onset POLG disorders commonly lead to ataxia, peripheral neuropathy & seizures

Elamipretide may impact peripheral neuropathy; PRO may be more appropriate than 6MWT

Late onset disease is characterized by ptosis, PEO, peripheral neuropathy, ataxia & muscle weakness

Elamipretide may impact peripheral neuropathy & muscle weakness, but these may require different endpoints (PRO & 6MWT)

Stealth BIOTHERAPEUTICS

Hikmat et al., 2020

Primary Mitochondrial Myopathies (PMM)

2016 new clinical consensus

National Organization for Rare Disorders (NORD) first published a physician's guide to Primary Mitochondrial Myopathy (PMM) in 2016

*"Primary mitochondrial myopathies (PMM) are genetically defined disorders leading to defects of oxidative phosphorylation...**affecting predominantly, but not exclusively, skeletal muscle**. Thus, secondary involvement of mitochondria, frequently observed in other neuromuscular diseases (e.g., Duchenne muscular dystrophy) is not considered PMM. Moreover, **individuals with muscle disease symptoms but with other systems are affected (i.e., brain, liver, kidney, etc.) are not considered affected by PMM**, and they may fit into a more straightforward clinical syndrome like Kearns-Sayre syndrome, MELAS syndrome, etc."*[1]

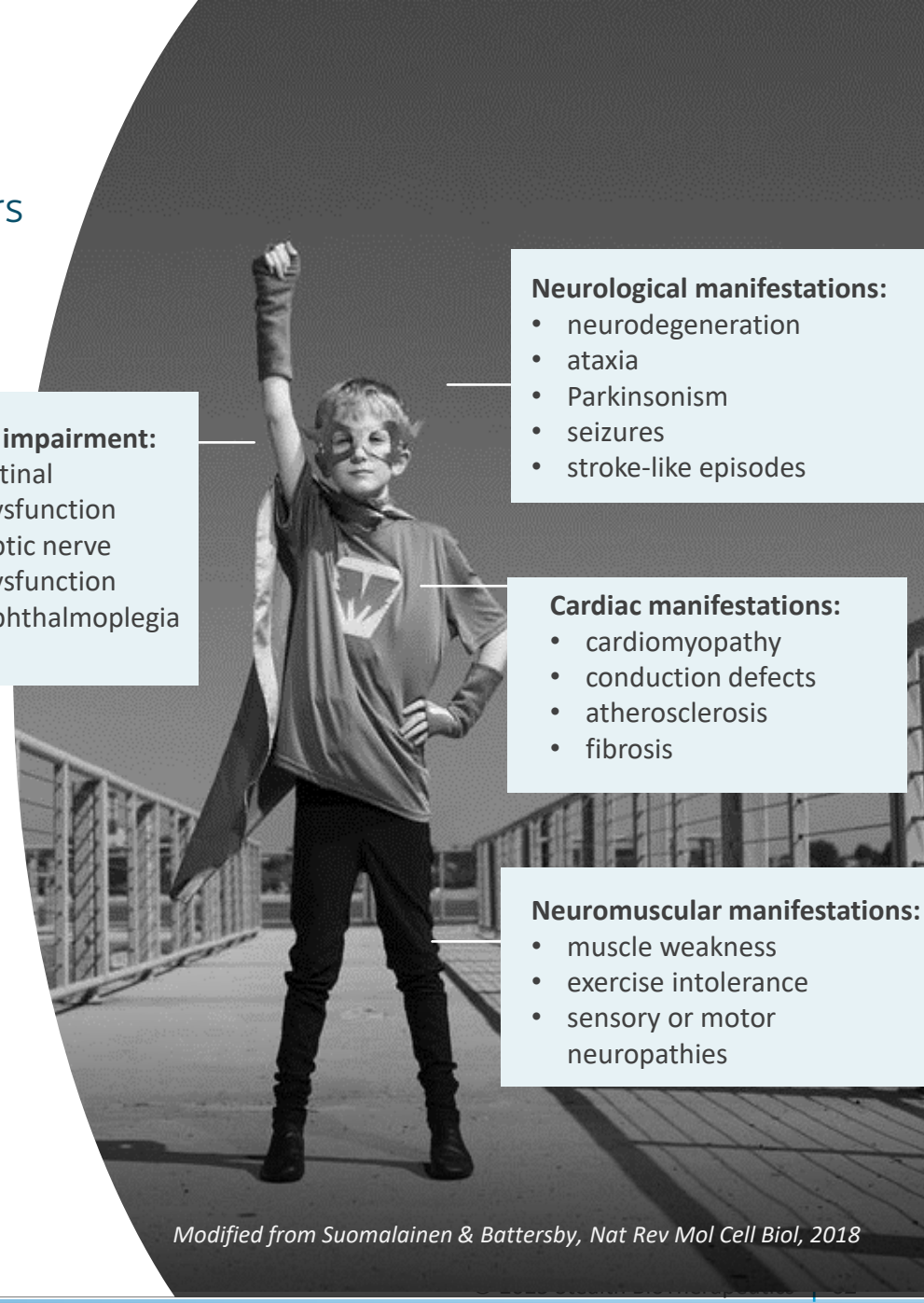
- Premised on a clinical recommendation to diagnose & treat primary mitochondrial diseases based upon clinical presentation rather than genetic etiology
- Opened the door to basket trial designs
- NOT accepted by European regulatory agency (EMA), which still requires that disease designations be based upon genetic etiology

1 https://rarediseases.org/rare-diseases/primary-mitochondrial-myopathies/

Primary Mitochondrial Myopathies

New consensus solved for some development challenges & introduced others

- ✓ Facilitated basket approach, alleviating patient identification & enrollment challenges of single-genotype trials (noting low prevalence of many genotypes)
- ✓ Informed functional endpoint selection, i.e., 6MWT (or variations), 5XSST, 3TUG, etc. given focus on predominantly myopathic disease presentation
- ❖ BUT –
  - Highly reliant on clinician identification of predominance of skeletal muscle related myopathy
  - Required enrichment via inclusion/exclusion criteria to reduce variability of myopathic disease presentation (for example, specifying degree of baseline impairment, requiring known comorbidities, etc.)
- ✓ **Allows detection of specific responsive genotypes (in our case, POLG-1) well-represented in a basket trial**



Modified from Suomalainen & Battersby, Nat Rev Mol Cell Biol, 2018

Primary Mitochondrial Myopathies

Development history – an overview

2014.....2015.....2016.....2017.....2018.....2019.....2020.....2021.....2022.....2023.....2024.....2025

elamipretide

RePOWER registry

MMPOWER
mtDNA & nDNA

MMPOWER-2
mtDNA & nDNA

MMPOWER-3
mtDNA & nDNA - stratified

NuPOWER
nDNA (majority POLG-1)

omaveloxolone

MOTOR
mtDNA & nDNA

mavodelpar

P 1/2
mtDNA

STRIDE
mtDNA

ASP0367

MOUNTAINSIDE
mtDNA & nDNA

Primary Mitochondrial Myopathies

The Genotype/Phenotype Debate

*Caused by nuclear (n) & mitochondrial (mt) DNA pathogenic variants; mtDNA heteroplasmy introduces variability; heterogeneous clinical impact on **brain, peripheral nervous & skeletal muscle systems***

POLG (nDNA) (largest group of nDNA)

- AHS affects **brain** (seizures) & liver
- MCHS affects **muscle, brain & nerves**
- Progressive External Ophthalmoplegia (PEO) affects **muscles** controlling eye movement
- Ataxia Neuropathy Spectrum Disorders causes **neurological problems** (**ataxia** & **neuropathy**)
- Mitochondrial Recessive Ataxia Syndrome (MIRAS) causes **ataxia** with severe epilepsy
- Sensory Ataxic Neuropathy, Dysarthria, & Ophthalmoparesis (SANDO) causes sensory **ataxia**, dysarthria & ophthalmoparesis
- **Myopathy; peripheral neuropathy**

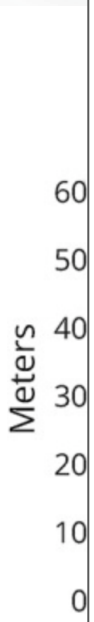
m.3243A>G (MT-TL1) (mtDNA) (largest group of mtDNA)

- Mitochondrial Encephalopathy Lactic Acidosis & Stroke-like episodes syndrome (MELAS) causes **neurological sequelae** (stroke-like episodes/ encephalopathy) & **myopathy**
- Maternally Inherited Deafness & Diabetes (MIDD) causes sensorineural hearing loss & diabetes
- PEO affects **muscles** controlling eye movement
- Leigh's syndrome affects the **brain** (developmental regression & seizures) & **muscle**
- **Myopathy**

Targeting skeletal muscle myopathy within & across diverse genotypes is highly dependent on clinician rigor in trial recruitment, expert consensus on classifications & detailed natural history characterization

Our Early Development Experience – MMPOWER P1/2

Dose-ranging; n=36 w/nDNA & mtDNA mutations; 5-day treatment period



ARTICLE OPEN ACCESS CLASS OF EVIDENCE

Randomized dose-escalation trial of elamipretide in adults with primary mitochondrial myopathy

Amel Karaa, MD, Richard Haas, MD, Amy Goldstein, MD, Jerry Vockley, MD, PhD, W. Douglas Weaver, MD, and Bruce H. Cohen, MD

Neurology® 2018;90:e1212-e1221. doi:10.1212/WNL.0000000000005255

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Abstract

Objective
To assess the safety and efficacy of elamipretide, an aromatic-cationic tetrapeptide that readily penetrates cell membranes and transiently localizes to the inner mitochondrial membrane where it associates with cardiolipin, in adults with primary mitochondrial myopathy (PMM).

Methods
A Study Investigating the Safety, Tolerability, and Efficacy of MTP-131 for the Treatment of Mitochondrial Myopathy (MMPOWER) was a phase I/II multicenter, randomized, double-blind, placebo-controlled trial of elamipretide in 36 participants with genetically confirmed PMM. Participants were randomized to intravenous elamipretide (0.01, 0.1, and 0.25 mg/kg/h or placebo for 2 hours in a dose-escalating sequence). The primary efficacy measure was the change in distance walked in the 6-minute walk test (6MWT) after 5 days of treatment. Other efficacy measures included changes in cardiopulmonary exercise testing parameters, in participant-reported symptoms, and in serum and urinary biomarkers. Safety, tolerability, and pharmacokinetics were also measured.

Results
Participants who received the highest dose of elamipretide walked a mean of 64.5 m farther at day 5 compared to a change of 20.4 m in the placebo group ($p = 0.053$). In addition, there was a dose-dependent increase in distance walked on the 6MWT with elamipretide treatment ($p = 0.014$). In a model that adjusted for additional covariates possibly affecting response, the adjusted change for the highest dose of elamipretide was 51.2 vs 3.0 m in the placebo group ($p = 0.0297$). No significant differences were observed in other efficacy and safety endpoints.

Conclusions
Elamipretide increased exercise performance after 5 days of treatment in patients with PMM without increased safety concerns. These findings, as well as additional functional and patient-reported measures, remain to be tested in larger trials with longer treatment periods to detect other potential therapeutic benefits in individuals affected by this condition.

Classification of evidence
This trial provides Class I evidence that for patients with PMM, elamipretide improved the distance walked on the 6MWT.

MORE ONLINE

→ **Class of Evidence**
Criteria for rating therapeutic and diagnostic studies
NPub.org/coe

CME Course
NPub.org/cmelist

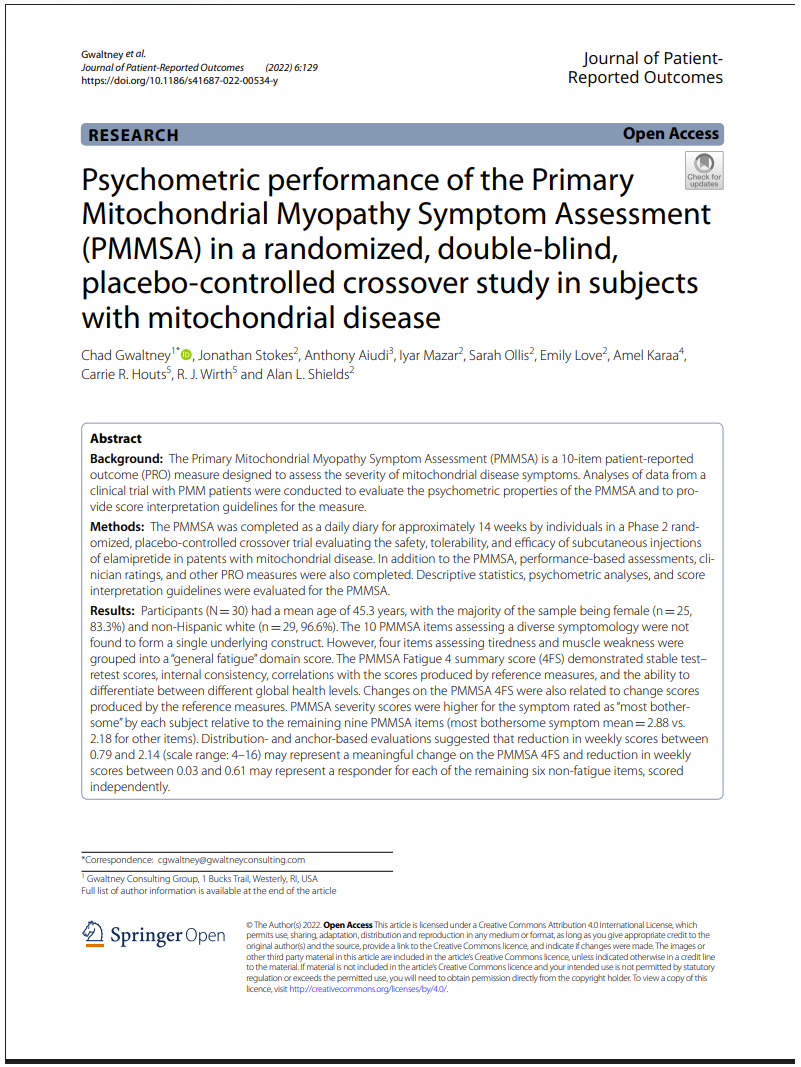
RELATED ARTICLE

Editorial
Remodel mitochondria and get energized
Page 633

- Improvement in 6MWT for subjects in high dose cohort (n=9) vs. placebo (n=9); dose-dependent increase in 6MWT with elamipretide
- No significant differences in other efficacy & safety parameters

Development of PMMSA & Most Bothersome Symptom

Developed & Validated to Assess Symptoms Most Problematic for Individual Patients



Gwaltney et al.
Journal of Patient-Reported Outcomes (2022) 6:129
https://doi.org/10.1186/s41687-022-00534-y

Journal of Patient-Reported Outcomes

RESEARCH Open Access

Psychometric performance of the Primary Mitochondrial Myopathy Symptom Assessment (PMMSA) in a randomized, double-blind, placebo-controlled crossover study in subjects with mitochondrial disease

Chad Gwaltney[1*], Jonathan Stokes[2], Anthony Aiudi[3], Iyar Mazar[2], Sarah Ollis[2], Emily Love[2], Amel Karaa[4], Carrie R. Houts[5], R. J. Wirth[5] and Alan L. Shields[2]

Abstract

Background: The Primary Mitochondrial Myopathy Symptom Assessment (PMMSA) is a 10-item patient-reported outcome (PRO) measure designed to assess the severity of mitochondrial disease symptoms. Analyses of data from a clinical trial with PMM patients were conducted to evaluate the psychometric properties of the PMMSA and to provide score interpretation guidelines for the measure.

Methods: The PMMSA was completed as a daily diary for approximately 14 weeks by individuals in a Phase 2 randomized, placebo-controlled crossover trial evaluating the safety, tolerability, and efficacy of subcutaneous injections of elamipretide in patents with mitochondrial disease. In addition to the PMMSA, performance-based assessments, clinician ratings, and other PRO measures were also completed. Descriptive statistics, psychometric analyses, and score interpretation guidelines were evaluated for the PMMSA.

Results: Participants (N = 30) had a mean age of 45.3 years, with the majority of the sample being female (n = 25, 83.3%) and non-Hispanic white (n = 29, 96.6%). The 10 PMMSA items assessing a diverse symptomology were not found to form a single underlying construct. However, four items assessing tiredness and muscle weakness were grouped into a "general fatigue" domain score. The PMMSA Fatigue 4 summary score (4FS) demonstrated stable test–retest scores, internal consistency, correlations with the scores produced by reference measures, and the ability to differentiate between different global health levels. Changes on the PMMSA 4FS were also related to change scores produced by the reference measures. PMMSA severity scores were higher for the symptom rated as "most bothersome" by each subject relative to the remaining nine PMMSA items (most bothersome symptom mean = 2.88 vs. 2.18 for other items). Distribution- and anchor-based evaluations suggested that reduction in weekly scores between 0.79 and 2.14 (scale range: 4–16) may represent a meaningful change on the PMMSA 4FS and reduction in weekly scores between 0.03 and 0.61 may represent a responder for each of the remaining six non-fatigue items, scored independently.

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Full list of author information is available at the end of the article

We developed & validated the Primary Mitochondrial Myopathy Symptom Assessment (PMMSA) PRO following FDA's patient focused drug development guidelines

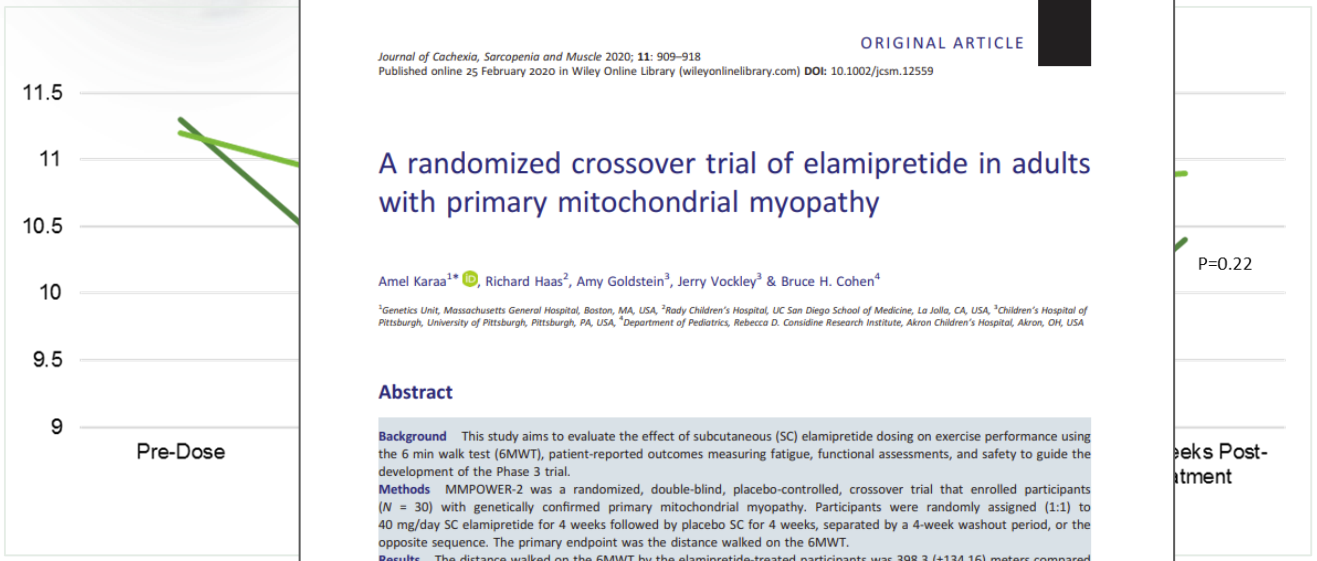
<u>10 Symptoms Identified Were Identified & Assessed via a Daily Diary</u>

- Tiredness at rest
- Tiredness during activities
- Muscle weakness at rest
- Muscle weakness during activities
- Balance problems
- Vision problems
- Abdominal discomfort
- Muscle pain
- Numbness
- Headache

Patients also identified their individual **"Most Bothersome Symptom"** from the PMMSA list, with changes on this patient-unique symptom tracked from baseline to end of treatment in our P 2 & P 3 clinical trials

Our Early Development Experience – MMPOWER-2 P2

n=30 MMPOWER subjects; 4-week treatment period; crossover design



ORIGINAL ARTICLE

Journal of Cachexia, Sarcopenia and Muscle 2020; **11**: 909–918
Published online 25 February 2020 in Wiley Online Library (wileyonlinelibrary.com) **DOI:** 10.1002/jcsm.12559

A randomized crossover trial of elamipretide in adults with primary mitochondrial myopathy

Amel Karaa[1]*, Richard Haas[2], Amy Goldstein[3], Jerry Vockley[3] & Bruce H. Cohen[4]

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Abstract

Background This study aims to evaluate the effect of subcutaneous (SC) elamipretide dosing on exercise performance using the 6 min walk test (6MWT), patient-reported outcomes measuring fatigue, functional assessments, and safety to guide the development of the Phase 3 trial.

Methods MMPOWER-2 was a randomized, double-blind, placebo-controlled, crossover trial that enrolled participants (N = 30) with genetically confirmed primary mitochondrial myopathy. Participants were randomly assigned (1:1) to 40 mg/day SC elamipretide for 4 weeks followed by placebo SC for 4 weeks, separated by a 4-week washout period, or the opposite sequence. The primary endpoint was the distance walked on the 6MWT.

Results The distance walked on the 6MWT by the elamipretide-treated participants was 398.3 (±134.16) meters compared with 378.5 (±125.10) meters in the placebo-treated group, a difference of 19.8 m (95% confidence interval, −2.8, 42.5; P = 0.0833). The results of the Primary Mitochondrial Myopathy Symptom Assessment Total Fatigue and Total Fatigue During Activities scores showed that participants treated with elamipretide reported less fatigue and muscle complaints compared with placebo (P = 0.0006 and P = 0.0018, respectively). Additionally, the Neuro-QoL Fatigue Short Form and Patient Global Assessment showed reductions in symptoms (P = 0.0115 and P = 0.0421, respectively). In this 4-week treatment period, no statistically significant change was observed in the Physician Global Assessment (P = 0.0636), the Triple Timed Up and Go (P = 0.8423) test, and wrist/hip accelerometry (P = 0.9345 and P = 0.7326, respectively). Injection site reactions were the most commonly reported adverse events with elamipretide (80%), the majority of which were mild. No serious adverse events or deaths were reported.

Conclusions Participants who received a short-course treatment of daily SC elamipretide for 4 weeks experienced a clinically meaningful change in the 6MWT, which did not achieve statistical significance as the primary endpoint of the study. Secondary endpoints were suggestive of an elamipretide treatment effect compared with placebo. Nominal statistically significant and clinically meaningful improvements were seen in patient-reported outcomes. The results of this trial provided an efficacy signal and data to support the initiation of MMPOWER-3, a 6-month long, Phase 3 treatment trial in patients with primary mitochondrial myopathy.

Keywords Myopathy; Primary mitochondrial disease; Elamipretide; Exercise intolerance; Primary mitochondrial myopathy; Crossover trial

Received: 26 January 2020; Revised: 4 February 2020; Accepted: 6 February 2020
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Email: akaraa@mgh.harvard.edu

- Improvement on 6MWT vs. placebo (trended, not significant)
- Reduced fatigue vs. placebo on PMMSA Total Fatigue scale (measured muscle weakness & tiredness at rest & during activities)
- Improvement in novel "**Most Bothersome Symptom**" on PMMSA (mean -0.3; p=0.011)
- Crossover trial design with some evidence of a carryover effect (albeit not significant)
- Injection site reactions (mostly mild) most common adverse event; no serious adverse events or deaths

Basket Approach in Late-stage Development

MMPOWER-3 P3 trial

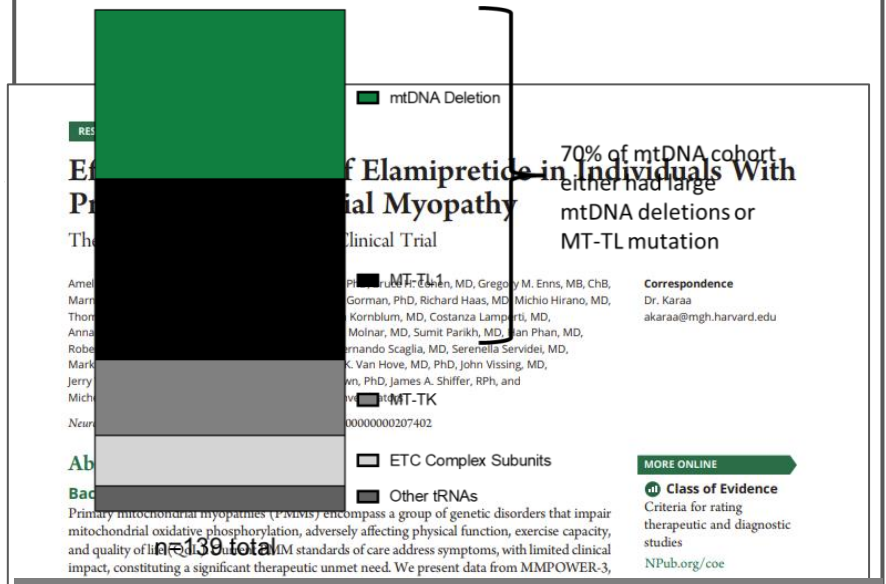
n=218 subjects w/nDNA & mtDNA mutations; 24-week treatment period

- 73% mtDNA mutations, 27% nDNA mutations
- Stratified by mtDNA:nDNA mutations to allow subgroup analyses:

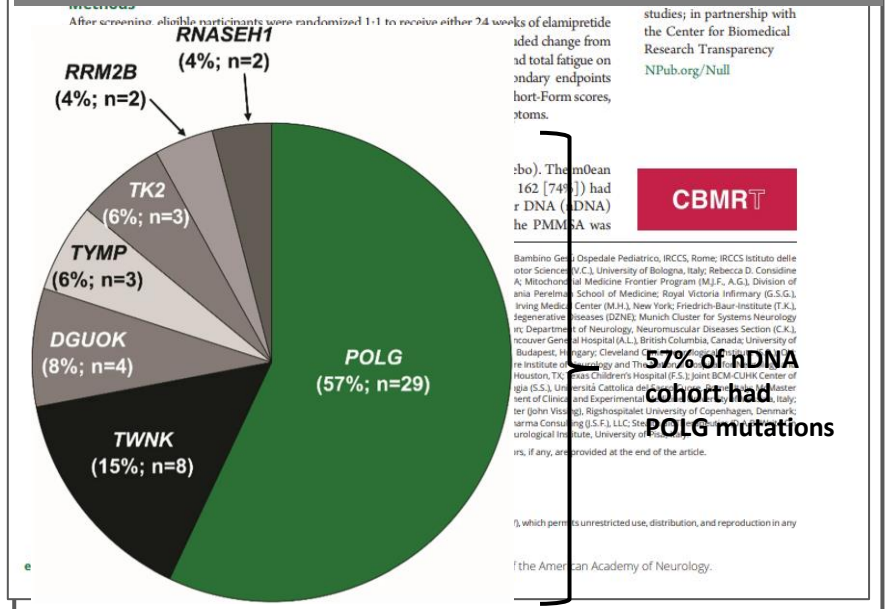
> *FDA: "We generally agree with your plan to enroll subjects…who have a clinical phenotype of…PMM affecting predominantly, but not exclusively, skeletal muscle. However, it is unclear to us if the various genetic mutations that you plan to study would lead to the same rate of clinical progression of the skeletal muscle weakness…* ***we suggest that you also attempt to balance the treatment arms with respect to the various mutations to be enrolled in your planned trial…*** *The extent to which any positive results…in a subset of patients with PMM…represented in your development program could be extrapolated to a broader range of patients with mutations that were not studied would be a review issue."*

- Adjudication committee confirmed pathogenic variants associated with myopathy; inclusion criteria defined min/max impairment

mtDNA cohort



nDNA cohort

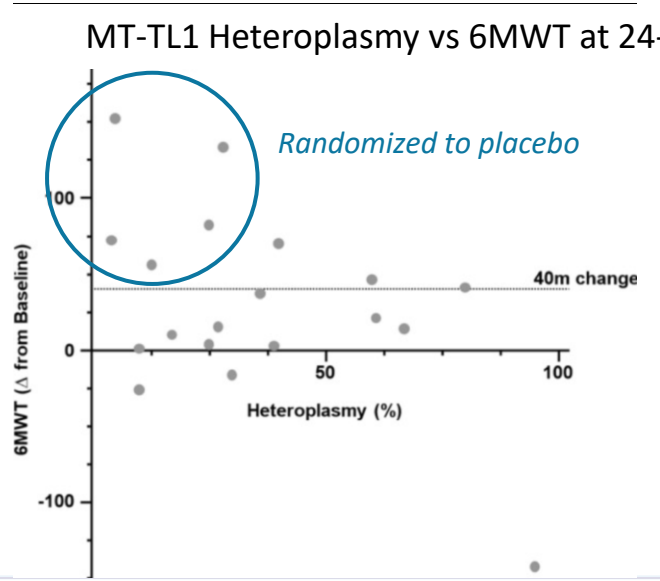


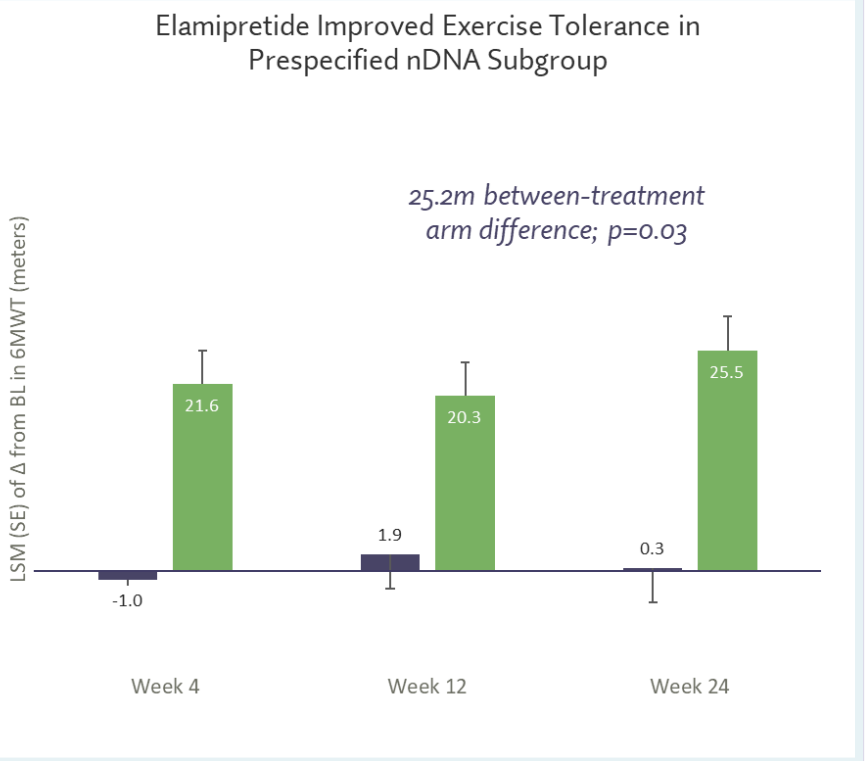
n=59 total

Challenges with Basket Approach

MMPOWER-3 trial failure & lessons learned

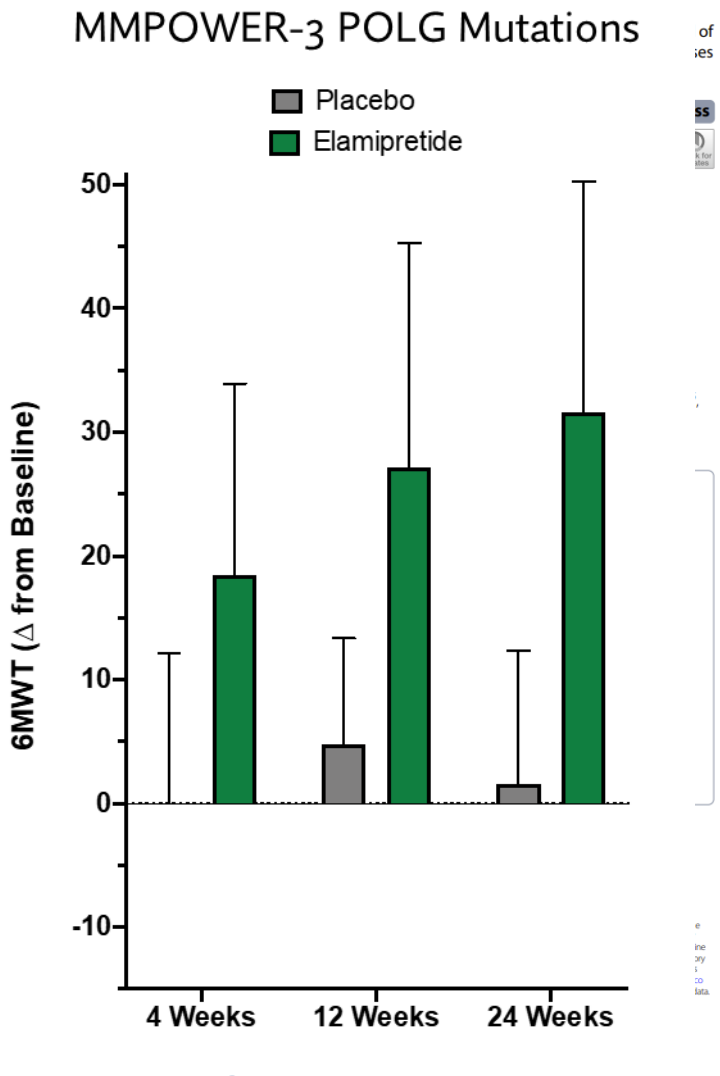
- No change observed between groups on 6MWT or fatigue (primary endpoints)
  - Large placebo response in patients with mtDNA mutations was associated with variable heteroplasmy across mtDNA cohort (imbalance favoring placebo); individuals with low heteroplasmy in MT-TL1 pathogenic variants (n=49) walked significantly further
  - **Patients with nDNA mutations randomized to elamipretide demonstrated nominally significant improvement on 6MWT vs placebo, with no mean change on placebo & a significant exposure-response relationship** *(post-hoc analysis of pre-specified subgroup)*
- No significant differences in other efficacy & safety parameters





Silver Lining: Subset of Responders Informed NuPOWER Trial

Premised primarily on POLG-1 signal, NuPOWER P3 enrolled subjects with nDNA mutations involving the replisome



- Most subjects in the nDNA cohort had replisome-related mutations, primarily POLG
  - POLG subjects demonstrated a nominally significant improvement in 6MWT distance on elamipretide, with de minimis mean placebo effect observed *(post hoc)*
- This informed the design of NuPOWER, a second P3 clinical trial, which enrolled n=102 patients with nDNA mutations (n=58 with POLG-1 mutations)
  - Primary endpoint 6MWT
  - Also assessing Patient Global Impression of Disease Severity (PGI-S) (secondary) & Most Bothersome Symptom on the PMMSA (exploratory)

NuPOWER Designed to Enrich for Myopathic Phenotype

POLG & other nDNA mutations can lead to ataxia, peripheral neuropathy & seizures, in addition to myopathy

Enrichments Implemented

nDNA mutations – • *Primary analysis on replisome-related mutations*	✓
Myopathic phenotype – • *Ocular muscle involvement (PEO) ensures myopathy (versus neurological) disease presentation* • *Specified impairment on 6MWT* • *Investigators counseled to avoid predominant ataxic & neuropathic disease presentations*	*TBD*
Exposure-response relationship – *Dose increased from 40mg SC to 60mg SC 1X daily*	✓
Duration – *12-month study duration leverages learnings from Barth syndrome program, where exercise tolerance peaked & was maintained after 9 mos. of therapy*	✓

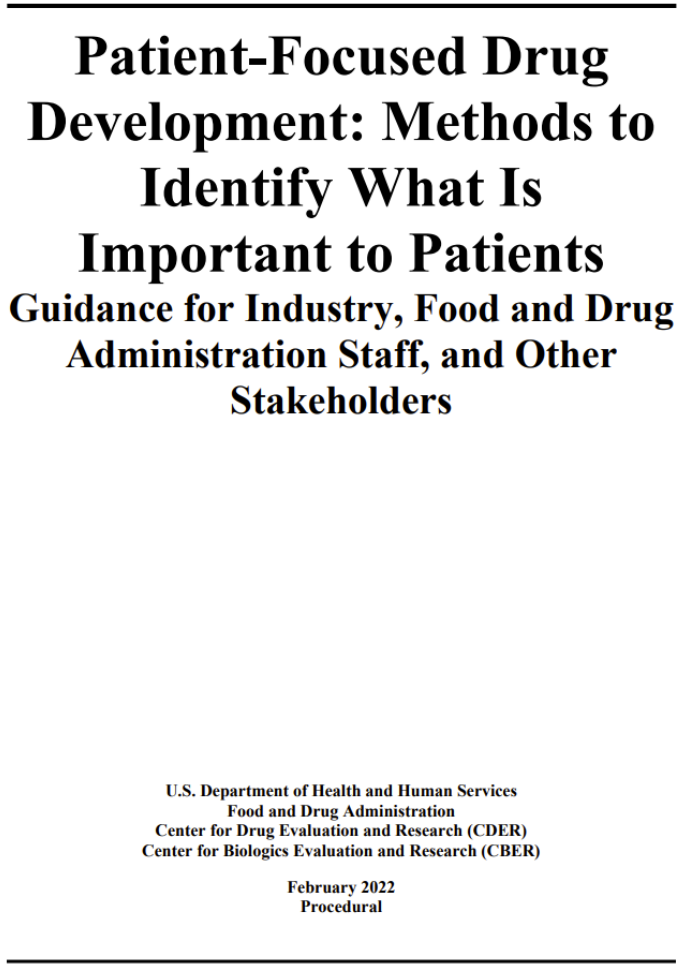
NUPOWER

Enrolled n=102 subjects with nDNA mutations

- Most (n=94) with replisome-related mutations; POLG-1 most common genotype (n=58)
- Success of **enrichment strategies to eliminate phenotypic variability** to be determined (TBD); baseline demographics show:
  - Most bothersome symptom variable, indicating differential disease burden with many subjects (>20%) identifying non-myopathic symptoms as "most bothersome"
  - Medical history confirmed inclusion of patients with more ataxic or neuropathic presentation
  - Variable age of onset (known to correspond to differential phenotypic presentation in POLG-1)
- Study readout expected H2 2025

Ramifications for Outcome Assessments

Can we customize endpoints to address high intra-patient heterogeneity in disease presentation?



Patient-Focused Drug Development: Methods to Identify What Is Important to Patients

Guidance for Industry, Food and Drug Administration Staff, and Other Stakeholders

U.S. Department of Health and Human Services
Food and Drug Administration
Center for Drug Evaluation and Research (CDER)
Center for Biologics Evaluation and Research (CBER)

February 2022
Procedural

- In Barth syndrome, we anticipated FDA guidance (!) by conducting semi-structured qualitative interviews of patients asking them to reflect on their trial experience
- Patients reported a variety of self-reported changes as being important to them, including:
  - Cessation of nighttime enuresis (bed-wetting) for several patients
  - Improvements in appetite for several patients
  - Improvements in time to recover from activities for several patients
- The commonality of some of these (e.g., nighttime enuresis) were unknown to the patient community prior to this exercise
- The “most bothersome symptom” was designed as an individually customized endpoint within a pre-specified menu of identified symptoms

Ramifications for Outcome Assessments

Can patient global impression scales capture perceived benefit despite variable phenotypic presentation?

Can most bothersome symptom inform functional assessments of interest?

Patient Global Impression of Disease Severity

- The PGI-S entails a single question whereby patients rate the severity of their disease symptoms
  - This enables patients to speak to overall severity of their own individual symptoms, which may be useful in a disease characterized by high inter-patient variability
- FDA has increasingly requested the inclusion of patient-reported GISs to aid in the determination of responder thresholds in assessing treatment efficacy, establish meaningfulness of within-patient changes, or to support the construct validity of other clinical outcome assessments[1]

Most Bothersome Symptom

- Allows personalized identification of most problematic symptom from menu of pre-specified choices
- Potential to inform functional outcome assessments to prioritize for each patient?
  - For example, if most bothersome symptom is muscle weakness or tiredness during activities, then 6MWT may be a relevant functional assessment
  - Alternatively, if balance is most bothersome, the SWAY balance application could be a relevant assessment

[1] Gnanasakthy et al., Value in Health, 2021

Take-aways

1. Basket/No-basket trial design should be informed by agent in development & clinical disease presentation
2. Reducing heterogeneity in basket trials depends on clinician rigor in recruitment, consensus recommendations & well-characterized natural history, all areas requiring further development. Historically, enrichment strategies have shown limited practical success. Stratification may help with interpretability of subgroups if heterogeneity drowns signals in basket ITT populations.
3. Alternatively (or additionally), consider endpoints that allow more individualized assessment of treatment benefit irrespective of phenotypic presentation, such as global impression scales or a most bothersome symptom construct.

Stealth BIOTHERAPEUTICS

Patients Are Waiting

"We don't know what is coming tomorrow. But in my wildest dream, we will grow old."

Quote from The POLG Foundation Film

Stealth BIOTHERAPEUTICS

BREAK

The meeting will resume at 11:45am ET

REAGAN-UDALL FOUNDATION FOR THE FDA

Selecting Patient-focused Outcomes & Statistical Considerations

Naomi Knoble, PhD

Center for Drug Evaluation and Research, FDA

Yan Wang, PhD

Center for Drug Evaluation and Research, FDA

REAGAN-UDALL FOUNDATION FOR THE FDA

Selecting Patient-Focused Outcomes and Statistical Considerations

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Statistical Reviewer
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Center for Drug Evaluation and Research

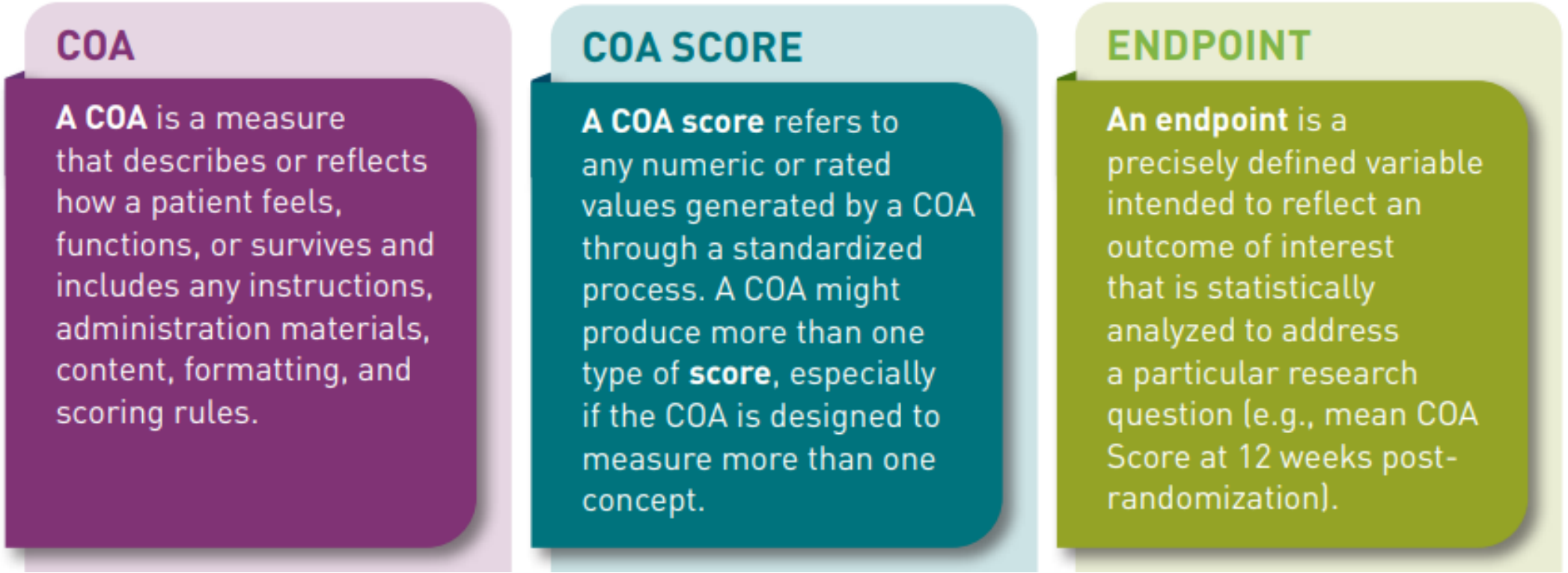
May 22, 2025

Disclaimer

- The views presented are the presenters' and do not reflect a position by the FDA.

- We have no conflicts of interest to disclose.

Clinical Outcome Assessments and Scores

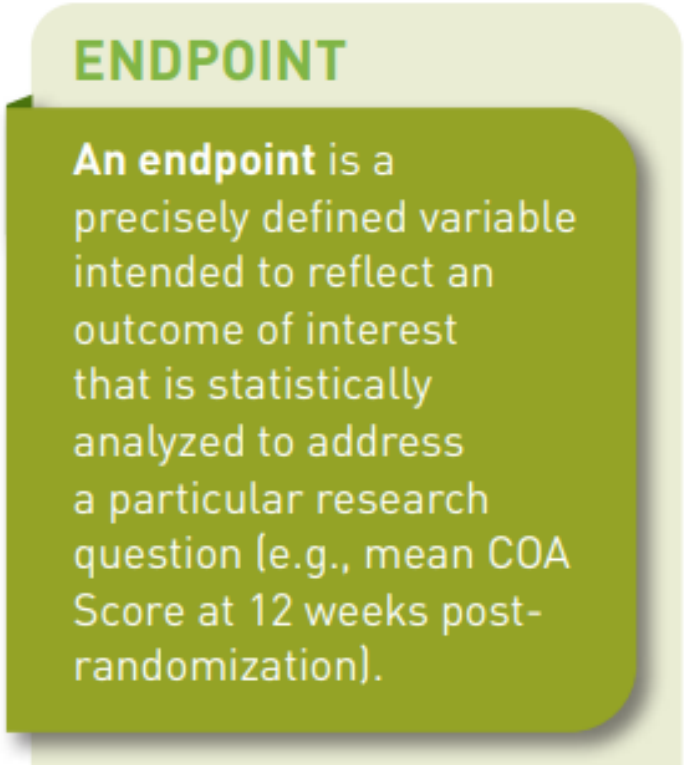


COA: Clinical Outcome Assessment; **See** *BEST Glossary* https://www.ncbi.nlm.nih.gov/books/NBK338448/

See PFDD draft guidance 3: Selecting, Developing, or Modifying Fit for Purpose COAs

Graphic from the PFDD guidance 3 snapshot: https://www.fda.gov/media/159516/download

Endpoint Strategies for Complex Conditions



- **One primary, multiple secondary endpoints**
  - Useful for one central disease feature most likely to be changed by the product MOA within the study duration
  - Secondary endpoints measure other important clinical features which may/may not be experienced by all patients
- **Multiple primary endpoints**
  - Useful when an improvement in at least one symptom/impact would be evidence of clinical benefit
- **Co-primary endpoints**
  - When clinical benefit can *only* be concluded if the drug/gene therapy has an effect on each of the endpoints
- **Multicomponent endpoint**
  - A single overall endpoint constructed from two or more COA scores

MOA: mechanism of action

FDA draft guidance (April, 2023). *Incorporating Clinical Outcome Assessments Into Endpoints for Regulatory Decision-Making.* https://www.fda.gov/regulatory-information/search-fda-guidance-documents/patient-focused-drug-development-incorporating-clinical-outcome-assessments-endpoints-regulatory

FDA (2022). *Multiple Endpoints in Clinical Trials*. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/multiple-endpoints-clinical-trials

Selecting Patient-Focused Outcomes

Engage

Listen to and integrate perspectives from patients, caregivers, clinical experts and others into measurement approach

Get Specific

Identify specific aspect(s) of target clinical outcomes

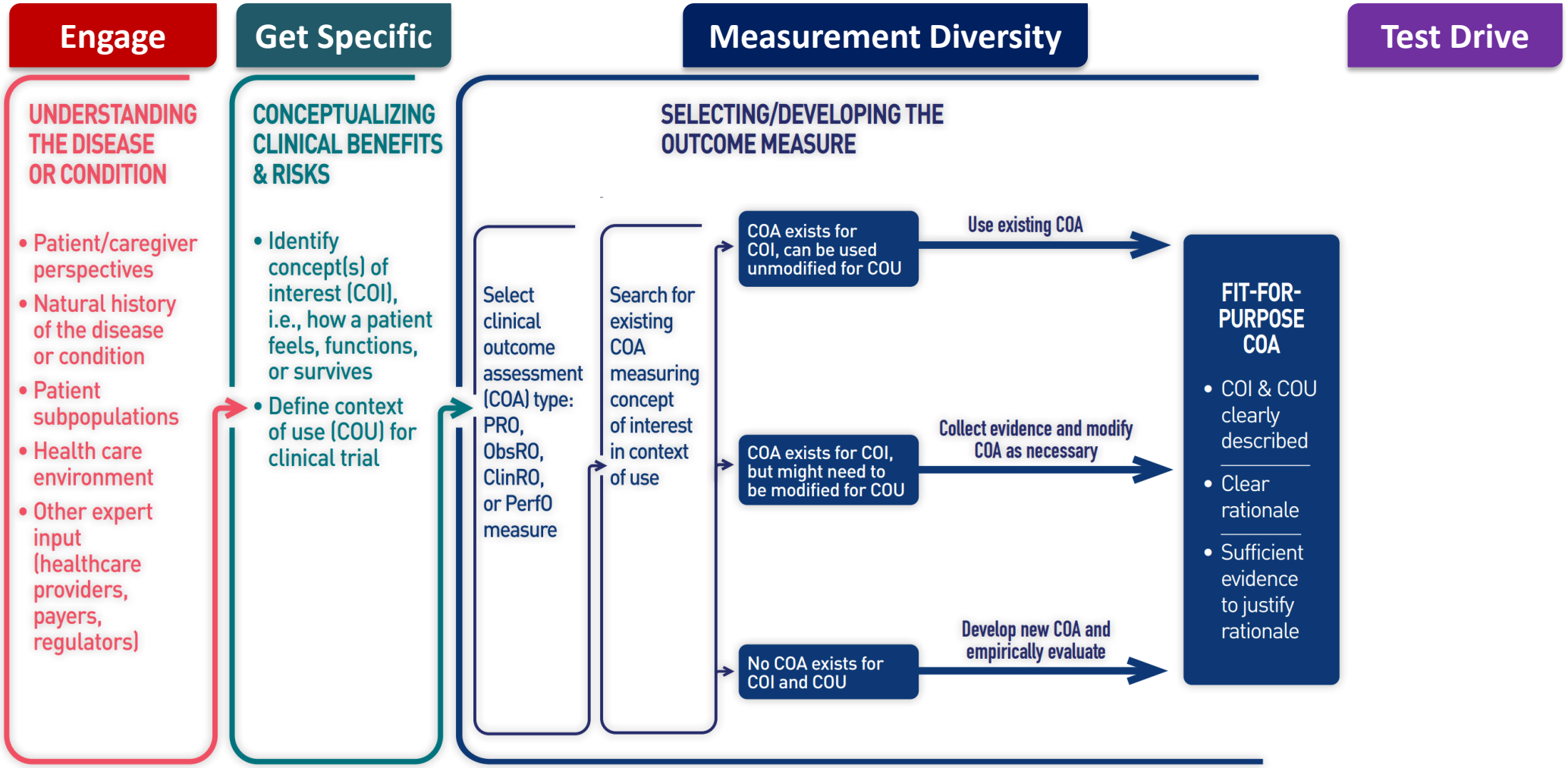
Measurement Diversity

Several types of COAs offer distinct sources of evidence within the endpoint hierarchy

Test Drive

Pilot test (aka “test drive”) measures to learn about strengths, limits, feasible implementation before pivotal trial

Roadmap to Patient-Focused Outcome Measurement



Concept of interest: The aspect of an individual's clinical, biological, physical, or functional state, or experience that the assessment is intended to measure.
See *BEST Glossary* https://www.ncbi.nlm.nih.gov/books/NBK338448/

See the draft guidance, *Selecting, Developing, or Modifying Fit-for-Purpose Clinical Outcome Assessments*
https://www.fda.gov/regulatory-information/search-fda-guidance-documents/patient-focused-drug-development-selecting-developing-or-modifying-fit-purpose-clinical-outcome

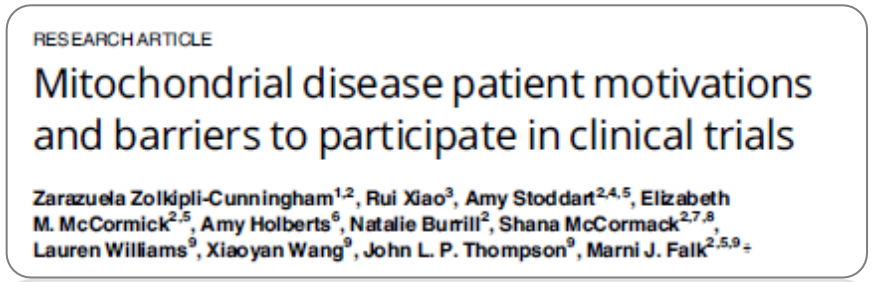
Patient-Focused Outcome Measurement:

Understanding the Disease or Condition

Engage

Listen to and integrate perspectives from patients, caregivers, clinical experts and others into measurement approach

- The mito community ***has*** engaged patient, caregiver, clinical expert, regulatory, and other perspectives



These are some examples of PMD community engagement and is not intended to reflect a preference by FDA for a publication, author(s), group, or any other preference.

Patient-Focused Outcome Measurement:
Conceptualizing Clinical Benefits & Risks

Get Specific

Identify specific aspect(s) of target clinical outcomes

Identify **specific aspects or attributes** of general symptoms likely to change in response to treatment within the trial duration

- *Duration*
- *Frequency*
- *Severity*
- *Worst experience*
- *Presence/absence*

For an applied example discussion of aspects of symptoms, see the Migraine Clinical Outcome Assessment System (MiCOAS) Measure Development Report available at https://vpghealth.com/micoas/

FDA draft guidance (June 2022). *Selecting, Developing, or Modifying Fit-for-Purpose Clinical Outcome Assessments*. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/patient-focused-drug-development-selecting-developing-or-modifying-fit-purpose-clinical-outcome

Patient-Focused Outcome Measurement:
Conceptualizing Clinical Benefits & Risks

Get Specific

Identify specific aspect(s) of target clinical outcomes

Daily Diary for Clinically Relevant Symptom Severity

- Reduction in monthly headache days or monthly migraine days
- Reduction in weekly mean daily maximal hunger score (e.g., Imcivree (setmelanotide)
- Responder defined as a patient achieving both the stool frequency and abdominal pain intensity responder criteria in the same week for a specified portion of the treatment duration (e.g., Linzess (linaclotide))
- 1-point reduction in worst weekly scratching score (e.g., Bylvay (odevixibat))

Advantages

- Patients as partners in patient-centered data collection
- Potentially sensitive COA-based endpoints

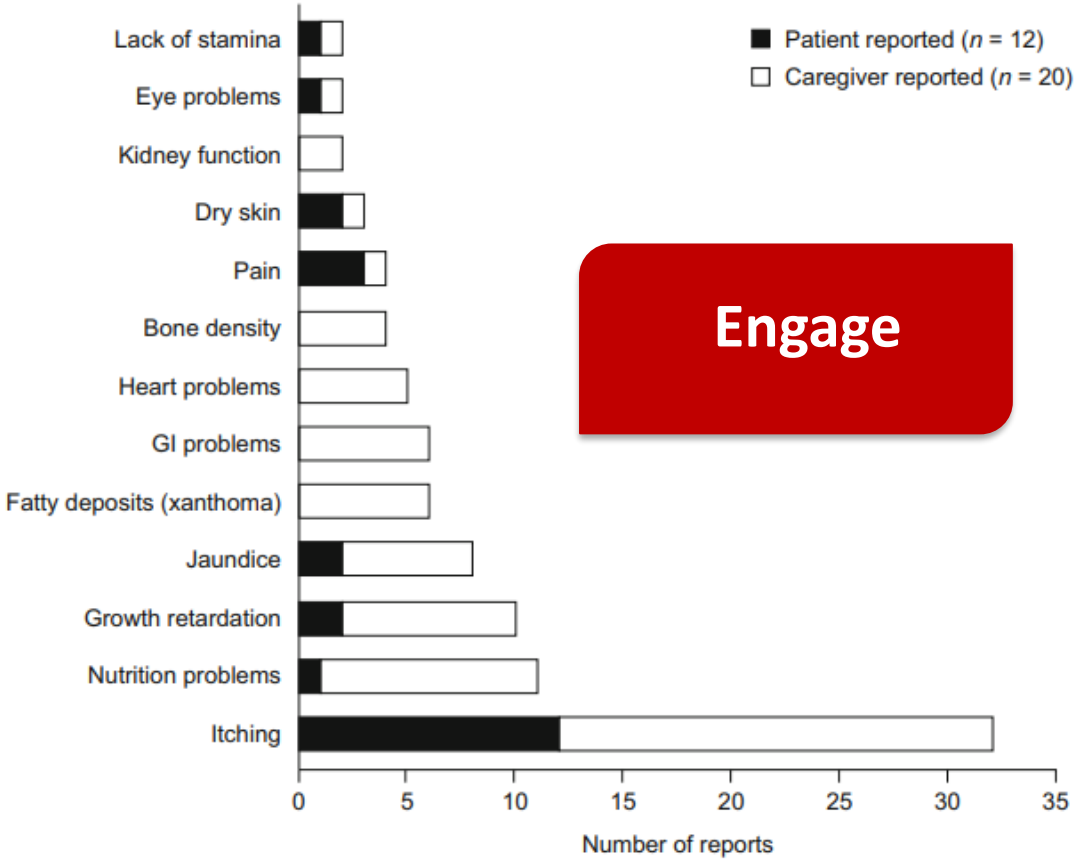
Disadvantages

- Patient burden
- Data integrity issues

These examples are not exhaustive and do not convey a preference by FDA for a specific measurement or medical product.

Patient-Focused Outcome Measuremen

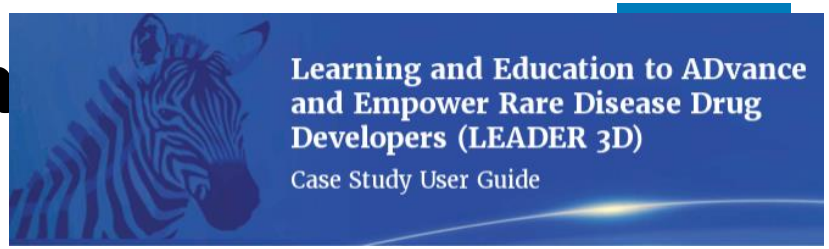
Conceptualizing Clinical Benefits & Risks



Source: Figure 3 Clinical features of Alagille Syndrome reported by caregivers and patients, from Kamath, B.M., Abetz-Webb, L., Kennedy, C. et al. Development of a Novel Tool to Assess the Impact of Itching in Pediatric Cholestasis. Patient 11, 69–82 (2018).

Engage

Get Specific



Bylvay (odevixibat)

Indication: Treatment of pruritus in patients 3-mo and older with PFIC

Study design: 24-week, randomized, double-blind, placebo-controlled trial

COA: Caregiver-reported daily diary of observed scratching (0 no scratching to 4 worst possible scratching)

Primary efficacy endpoint: Mean % of assessments that are ≤1 or at least 1-point drop from baseline of the patient's worst weekly average scratching scores

See FDA's Learning and Education to ADvance and Empower Rare Disease Drug Developers (LEADER 3D): https://www.fda.gov/media/186133/download?attachment

This example does not convey a preference by FDA for a specific measurement or medical product.

Patient-Focused Outcome Measurement

Selecting the Outcome Measure: Measurement Diversity

Measurement Diversity

Several types of COAs offer distinct sources of evidence within the endpoint hierarchy

- A "perfect" measurement of an outcome *may* exist; however, most measures have one or several limitations
  - Relying on one and only one COA to generate all clinical outcome evidence in a clinical trial risks that its limitations may undermine the evidence
- Diversify your measurement within the endpoint hierarchy to mitigate risks while considering:
  - Patient/caregiver participation burden
  - Ease of implementation for sites/staff/investigators
  - Best sources of evidence for the selected outcomes

Use the full endpoint hierarchy for an inclusive measurement strategy

Balance disadvantages of relying on one COA (or COA type) with patient participation burden

Consider exit interviews/surveys

Benjamin et al. Patient-Reported Outcome and Observer-Reported Outcome Assessment in Rare Disease Clinical Trials: An ISPOR COA Emerging Good Practices Task Force Report. Value Health. 2017 Jul-Aug;20(7):838-855; Walton MK, Powers JH 3rd, Hobart J, et al. Clinical Outcome Assessments: Conceptual Foundation-Report of the ISPOR Clinical Outcomes Assessment - Emerging Good Practices for Outcomes Research Task Force. *Value Health*. 2015;18(6):741-752. doi:10.1016/j.jval.2015.08.006; FDA guidance (2023). *Rare Diseases: Considerations for Development of Drugs and Biologic Products* https://www.fda.gov/regulatory-information/search-fda-guidance-documents/rare-diseases-considerations-development-drugs-and-biological-products; FDA guidance (2022). *Multiple Endpoints in Clinical Trials*. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/multiple-endpoints-clinical-trials

Patient-Focused Outcome Measurement

Selecting the Outcome Measure: Measurement Diversity

Measurement Diversity

Several types of COAs offer distinct sources of evidence within the endpoint hierarchy

Pombiliti (cipaglucosidase alfa)

Indication: Adults with late-onset Pompe disease (LOPD)

Study design: 52-week, randomized, double-blind, active-controlled trial

Measurement: 6MWT (PerfO), sitting FVC % predicted (SE), GSGC (ClinRO), MMTL (ClinRO), PROMIS physical function (PRO), PROMIS fatigue (PRO)

Endpoint approach: One primary, multiple secondary

Primary Efficacy Endpoint: Change from baseline in distance walked on 6MWT (PerfO)

Key Secondary Efficacy Endpoint: Mean change in sitting FVC % predicted from baseline to Week 52 (SE)

6MWT: 6-min walk test; **ClinRO**: clinician-reported outcome; **FVC**: forced vital capacity; **GSGC**: Gait, Stairs, Gowers maneuver, and Chair test; **MMTL**: Manual muscle testing lower extremities; **PRO**: patient-reported outcome; **PerfO**: performance outcome; **SE**: surrogate endpoint

https://www.accessdata.fda.gov/drugsatfda_docs/nda/2023/761204Orig1s000TOC.cfm

These examples are not exhaustive and do not convey a preference by FDA for a specific measurement or medical product.

“Test Drive” Measurement Approaches

Test Drive

Pilot test (aka “test drive”) measures to learn about strengths, limits, feasible implementation before pivotal trial

- **Conduct a pilot study (an observational study) of different types of assessments**
  - Generate information about how to implement assessments
  - Learn strengths and limitations of each COA
  - Reduces risks of failure due to measurement and implementation

- Pilot testing can be done in stages
- Helps ensure alignment with clinical trial design

Reviews for Approved Products are Public

Drugs@FDA: FDA-Approved Drugs

https://www.accessdata.fda.gov/scripts/cder/daf/index.cfm

Global Tests for Multiple Endpoints in Rare Disease Clinical Trials

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Challenges in Drug Development for PMD

- Rare, complex, multisystemic diseases
- Heterogeneous clinical manifestations
  - difficult to find single measure to support a primary endpoint
  - multiple endpoints or individualized endpoints may be needed to evaluate treatment effects
- Traditional trial designs and testing methods may have low statistical power to detect a treatment effect due to
  - very small patient populations for some disease subtypes
  - small or moderate treatment effect of an investigational product on a single endpoint

Novel trial designs and global tests may be used to overcome some of these challenges

Global Tests for Multiple Endpoints

- Main goals
  - Increase the statistical power of detecting a treatment effect
  - Describe treatment effects more comprehensively for diseases with heterogeneous clinical presentations where a single outcome measure does not suffice to fully capture the treatment effects
- Global tests have been studied for decades, and well-known tests include Rank-Sum, OLS, and GLS by O'Brien [1984]
- Ristl et al. [2019]: "*Methods for the analysis of multiple endpoints in small populations: A review*".

Trial 1: R, DB, AC, Superiority, 12-Month Trial in Patients with LOPD

For the primary endpoint, estimated treatment difference numerically favors the test product (cipaglucosidase alfa + miglustat), but it does not meet the pre-defined statistical significance level of 0.05.

	TP (n=85)	AC (n=38)	Treatment Difference in LS Means (95% CI)
6MWT (primary)	21 (42)	8 (41)	**14 (-1, 28)** **p > 0.10**
FVC% (secondary)	-1.1 (6.3)	-3.3 (5.0)	2.3 (0.0, 4.6) p < 0.05
Post-hoc Global Tests for 6MWT and FVC%			
NS-Sum	p < 0.03		
Test-Statistic-Sum	p < 0.01		
O'Brien Rank-Sum	p < 0.03		

TP: cipaglucosidase alfa + miglustat. AC: a non-U.S.-Approved alglucosidase alpha product. LOPD: late-onset Pompe disease.
6MWT: distance walked during a 6-minute walk test. FVC%: percent predicted forced vital capacity.
Mean (SD) of changes from baseline in 6MWT and FVC% at 12 months are presented for each treatment group.

For a new trial, should both endpoints be selected as primary endpoints and tested using a **global test** to evaluate the totality of treatment effects?

Global Tests: Combine Information from Multiple Endpoints

Combine outcome data at patient-level

- **O'Brien Rank-Sum:** based on the sum of the ranks of the outcome of each endpoint
- **NS-Sum:** based on the sum of the normalized scores of the outcome of each endpoint

Combine test statistics at endpoint-level

- **Test-Statistic-Sum:** based on the sum of the test statistics for treatment comparison for each endpoint.

Global Tests: Test a Global Null Hypothesis
"Drug has no effect on any of the multiple endpoints"

- When p-value < 0.05 (or other pre-defined significance level), reject the global null hypothesis and conclude that the drug has effect on at least one endpoint
- P-value should be presented and interpreted with descriptive summary statistics for the individual endpoints to elucidate the treatment effects
- "P-value < 0.05" may not necessarily indicate an overall benefit if discordant effects are observed
- No multiplicity issue, but does not provide inferences on individual endpoints

Apply to traditional tests for multi-component endpoints and composite endpoints

Traditional tests for multi-component endpoints are global tests

Trial 2: R, DB, PC, 12-month Trial in Patients with Nieman-Pick Disease (NPC)

Primary endpoint: change from baseline in **total score** of 4-domain NPC severity score

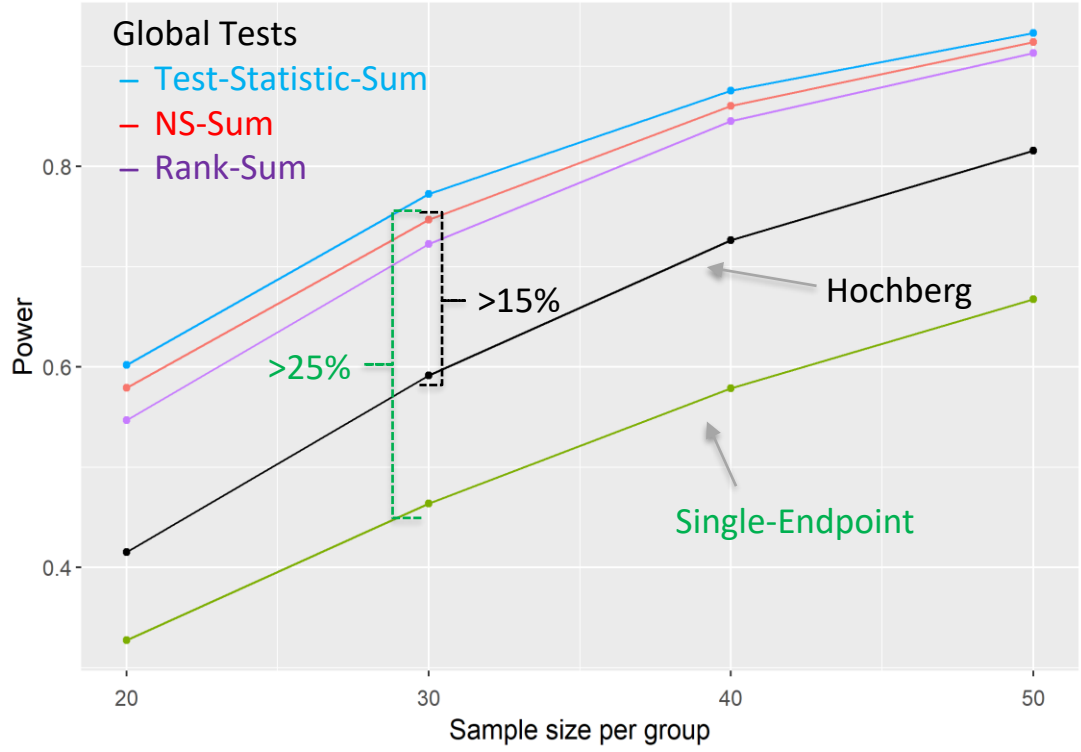
Domain	Mean (SD) Change from baseline at 12 Months		
	Placebo (N = 16)	Arimoclomol (N = 34)	Treatment Difference P-value
Ambulation	0.3 (0.9)	0.4 (0.7)	> 0.9
Fine Motor Skills	0.6 (1.3)	0.3 (0.9)	> 0.4
Speech	0.3 (0.8)	-0.1 (1.0)	> 0.3
Swallow	0.7 (1.1)	0.1 (1.1)	= 0.2
Total Score	1.9 (3.1)	0.7 (2.2)	**< 0.09**

Global Test →

- A multi-component endpoint can be viewed as a global test for the individual domain endpoints that combine the scores of individual domain endpoints at patient-level using the **total score** of the individual domain score.
- **Advantage of global tests:** no need to consider the "**total score**" as an endpoint, thus no need to validate it as an endpoint.

Power Comparison: Global Tests vs. Traditional Approaches

Global tests can be more powerful when a drug has effect on both endpoints (effect size of 0.5 for both endpoints)



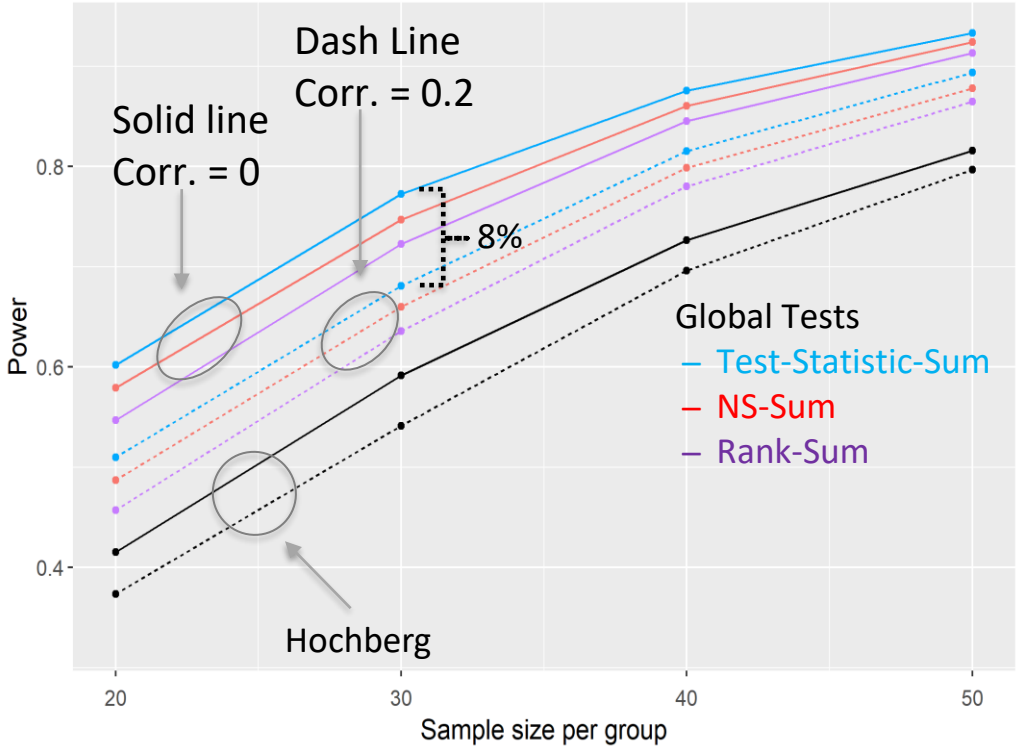
Two independent endpoints from a normal distribution with effect size of 0.5

Take-home message

Ideal scenario to use global tests is when a drug has similar treatment effect sizes on all endpoints.

Correlation Impacts Power

Correlation of 0.2 was observed between 6MWT and FVC% endpoints in trial 1 for LOPD



Power of global tests decreases when correlation of the endpoints increases

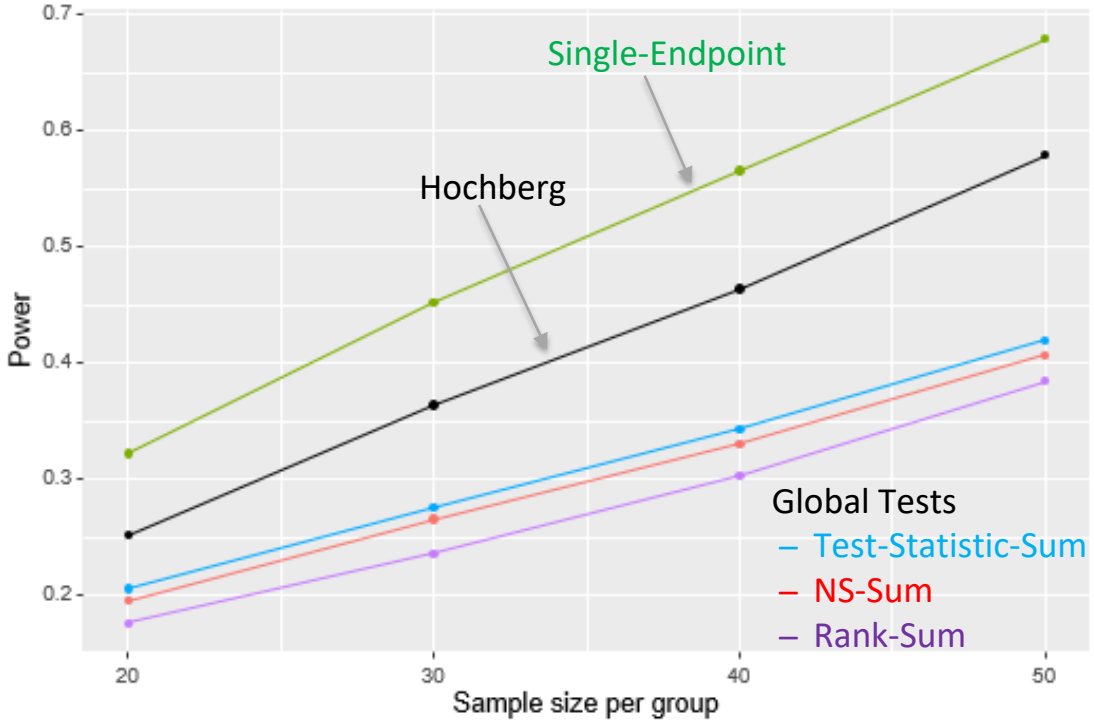
Take-home message

Ideally multiple endpoints should be selected to represent distinct clinical manifestations.

Two independent endpoints from a normal distribution with effect size of 0.5

Global tests are less powerful when a drug has an effect only on one endpoint

Effect size is 0.5 for one endpoint and 0 for the other endpoint



Take-home message

Do not include an endpoint as a primary endpoint when a drug is not expected to have an effect on this endpoint.

Global Tests

Can provide a broad efficacy assessment for **novel trials** that use different endpoints for different subsets of patients to accommodate patients' heterogeneous clinical presentations

Potential Novel Trial for PMD

Novel Trial: Patients have **different primary endpoints** depending on their symptoms

Population	Meeting inclusion criteria for symptoms of	Primary endpoint(s)
A	muscle weakness and chronic fatigue	6MWT and fatigue Score
B	muscle weakness only	6MWT only
C	chronic fatigue only	Fatigue Score only

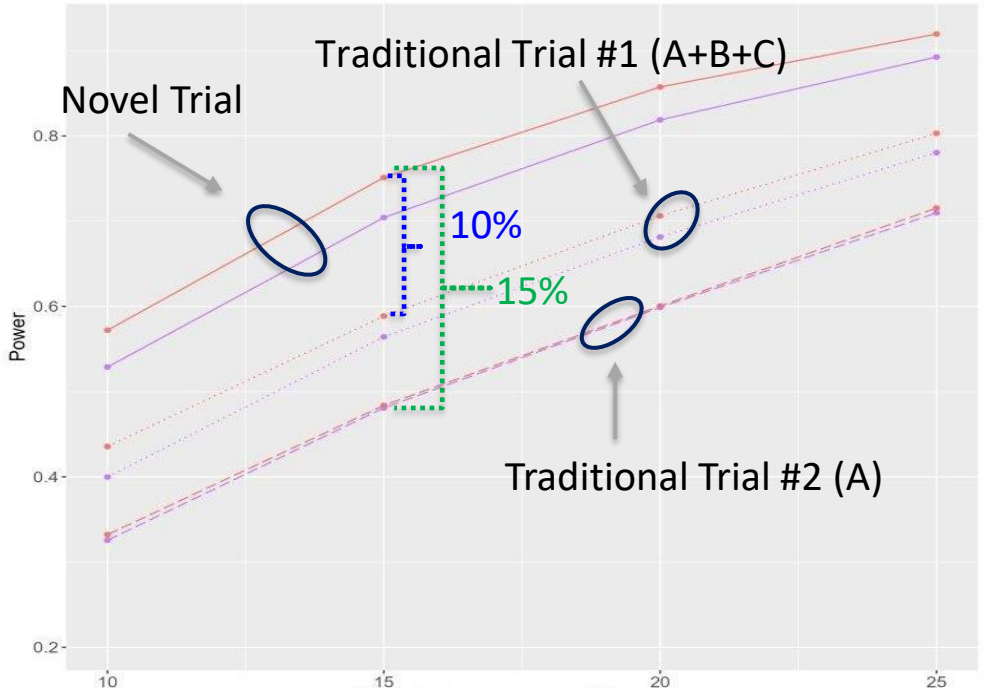
Traditional Trials: All patients have the **same primary endpoints**

Traditional Trial	Including patients from	Primary endpoints
1	Populations A, B, and C	6MWT and fatigue Score
2	Populations A only	

Potential Novel Trial Evaluated Using Global Tests

Simulation Study
Novel trial using global tests has higher power

- Power increased >10% compared to the Traditional Trial #1
- Power increased >15% compared to the Traditional Trial #2 that uses data from Population A only



Global Tests
- NS-Sum
- Rank-Sum

Two independent endpoints from a normal distribution with effect size = 0.5 for each of the primary endpoints in novel trial

Multi-domain Responder Index (MDRI) Approach Is a Global Test Approach Combining Data at Patient-Level

- Proposed by increasing number of sponsors in regulatory submissions
- Combine multiple endpoints using a responder **threshold for a minimal clinically important difference (MCID)** for each endpoint. There are multiple ways to combine.
- For each patient, **MDRI score** is the sum of the responder scores for the multiple endpoints
- Hypothesis testing is performed to compare the **means of the MDRI score** between treatment groups

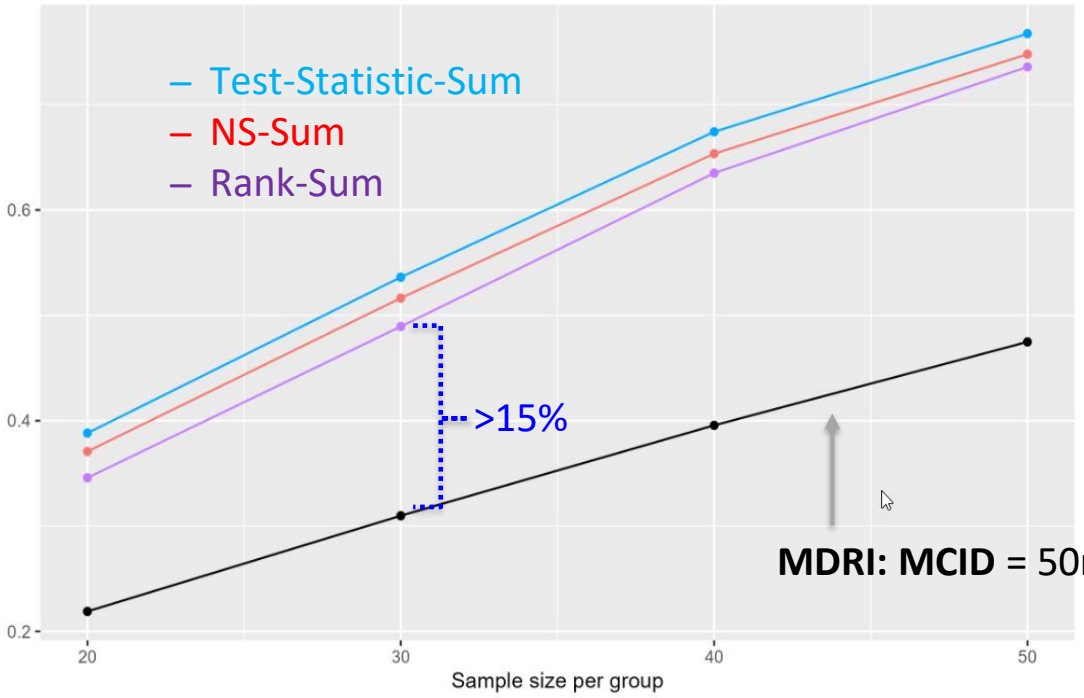
MDRI example construction and calculation

Endpoint	Change in 6MWT	Change in FVC%	MDRI Score
MCID Threshold	50 meters	5%	
Responder score	-1 | 0 | +1	-1 | 0 | +1	
Patient #1:			
Change observed	-15m	+5.8	
Responder Score	0	+1	+1
Patient #2:			
Change observed	+68m	+2.8	
Responder Score	+1	0	+1
Patient #3:			
Change observed	-55m	-11.0	
Responder Score	-1	-1	-2

MDRI Approach: Potential Issues

- May be difficult to reach consensus on MCID thresholds
- May have lower power than other global tests

Simulation Study
Based on the data for the 6MWT and FVC% endpoints in Trial 1 for LOPD



MDRI: MCID = 50m for 6MWT and 10% for FVC%

Summary

- Global tests can be used to provide a global inference on treatment effects in PMD trials enrolling patients with heterogenous clinical manifestations
  - May have higher power than traditional testing methods when the investigational product has effect on each of the multiple endpoints
  - Can provide a broad efficacy assessment for novel trials that use individualized endpoints

References

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2) FDA Briefing Document for the Advisory Committee Meeting for Arimoclomol, August 2, 2024: https://www.fda.gov/advisory-committees/advisory-committee-calendar/updated-public-participation-information-august-2-2024-meeting-genetic-metabolic-diseases-advisory#event-materials
3) O'Brien, P. C.: "Procedures for comparing samples with multiple endpoints". *Biometrics, Vol. 40, No. 4: 1079-1087, 1984*
4) Pocock J.S. et al.: "The Analysis of Multiple Endpoints in Clinical Trials". *Biometrics, Vol. 43, No. 3: 487-498, 1987*
5) Ristl R. et al.: "Methods for the analysis of multiple endpoints in small populations: A review". *Journal of Biopharmaceutical Statistics, 29:1, 1-29, 2018*
6) Tandon and Kakkis: "The multi-domain responder index: a novel analysis tool to capture a broader assessment of clinical benefit in heterogeneous complex rare diseases". *Orphanet Journal of Rare Diseases*. 2021,16:183. https://doi.org/10.1186/s13023-021-01805-5

Acknowledgement

Dr. Yared Gurmu performed the simulation studies and the global tests for the trial examples.

REAGAN-UDALL FOUNDATION FOR THE FDA

Presentation on Clinical Outcome Assessment (COA) example

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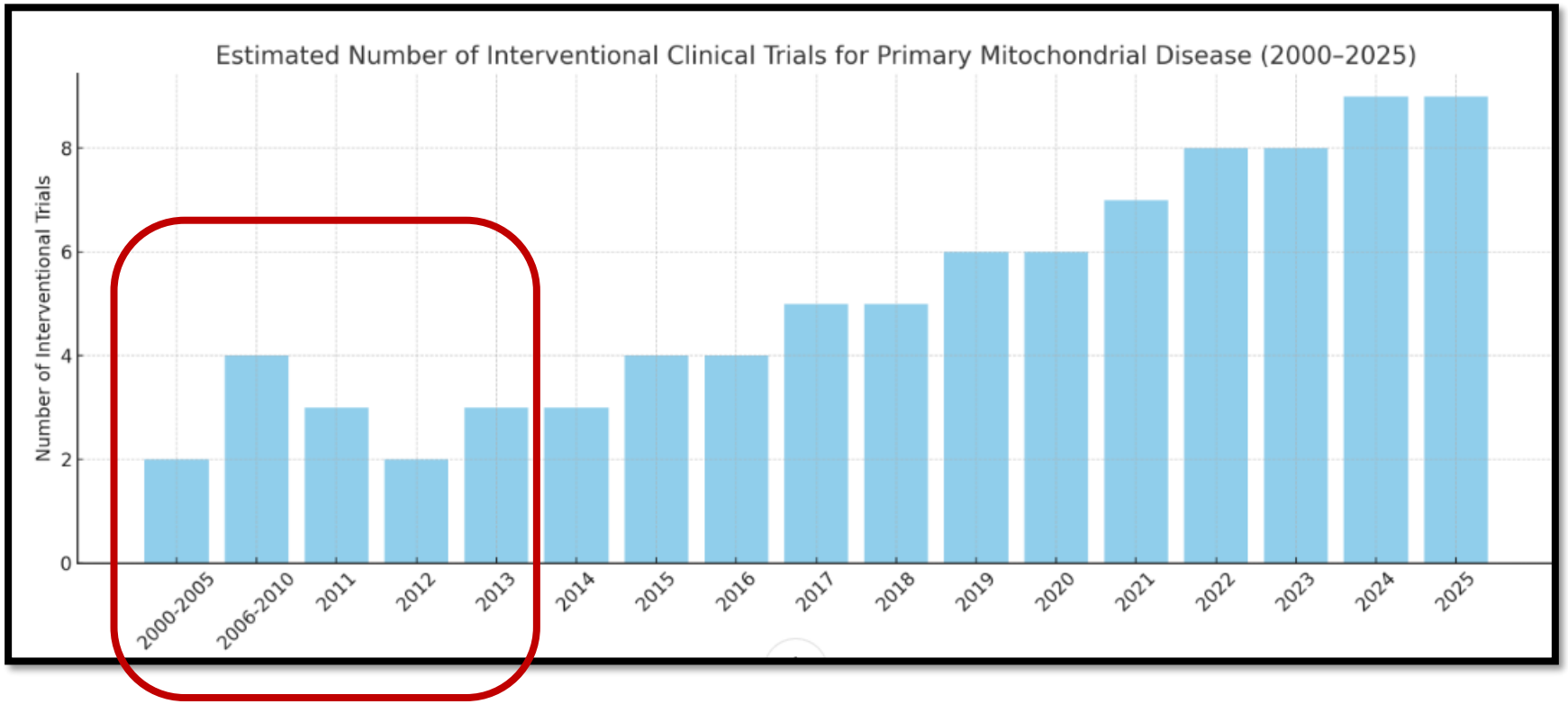
Clinical Outcomes Assessments: Lessons from the recent PMD clinical trials

AMEL KARAA, MD

PRIMARY MITOCHONDRIAL DISEASES



PRIMARY MITOCHONDRIAL DISEASES

Molecule	Trial Stage	Treatment period	Trial Population	Outcome measure	Results
Carnitine	Double blind placebo/controlled crossover design	8 weeks 4 weeks WO 8 weeks	≥14 yo MM with CPEO (Biopsy) (n= 12) Healthy volunteers (n=10)	PFT CPET Knee extension dynamometry Body composition	Improved weight
Resveratrol	Randomized, double-blind, cross-over and placebo-controlled.	8 weeks 4 weeks WO 8 weeks	≥18 yo PMM (n=11)	**(HR) during submaximal cycling exercise** VO2max during maximal exercise Perceived exertion, Lactate concentrations SF-36, FSS Muscle biochemistry	No change
Bezafibrate	Open label	6 weeks	≥18 yo PMM (m.3243A>G) (n= 6)	Cycle ergometer (submax Ex), 3TUG, Actigraphy FIS 31P-MRS, Muscle Biopsy	NA
Creatine	randomized, double blind, placebo-controlled, crossover trial	4 weeks 4 weeks WO 4 weeks	≥18 yo PMM (n= 11)	Neuromuscular symptom score Hammersmith motor ability score MRC score	Did not meet endpoints
Niacin	Open label	4-10 months	≥18 yo CPEO (n=5)	Body composition 6MWT, PFT, Metabolomic	Improved body composition, strength

CLINICAL OUTCOMES ASSESSMENTS

Outcome measure: is a tool used to assess a patient's status.
→ measured variable (e.g. fatigue score)

Endpoint: a targeted outcome of a clinical trial that is statistically analyzed to help determine the efficacy and safety of the therapy being studied

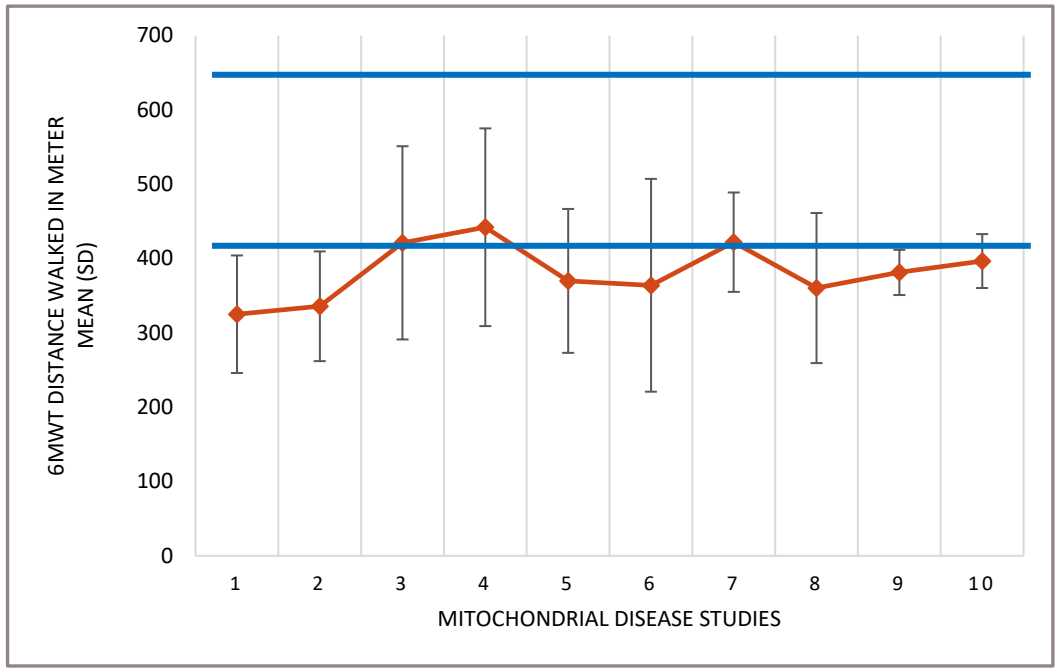
→ change from baseline to X weeks in mean fatigue score

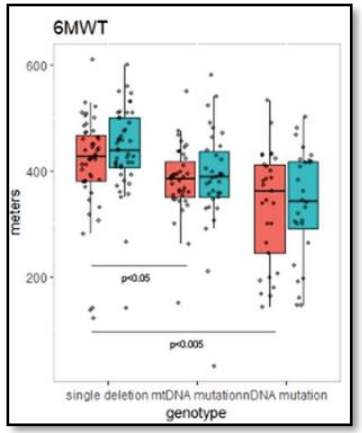
Clinical Trials in Primary Mitochondrial Myopathy

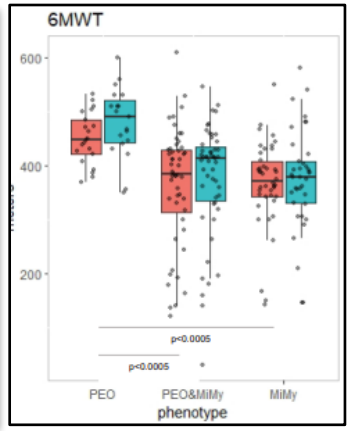
Trial phase	Design	Primary / Secondary Outcomes	Endpoint Met?
Phase 2, MOTOR trial: omaveloxolone in mitochondrial myopathy NCT02255422	Randomized, double-blind, placebo-controlled (parallel)	Primary: Peak exercise workload; Secondary: 6MWT distance, respiratory/biochemical markers	No, peak exercise (p=0.77), 6MWT (p=0.38), significant submax lactate decrease (p<0.05) at 12 weeks
MMPOWER-3: Phase 3, Elamipretide in primary mitochondrial myopathy NCT03323749	Randomized, double-blind, placebo-controlled	Primary: 6MWTdistance; PMMSA total fatigue, NeuroQoL, PGA	No, 6MWT change −3.2 m (p=0.69), PMMSA fatigue Δ −0.07 (p=0.37).
TAZPOWER: Elamipretide in Barth syndrome (NCT03098797)	Randomized, Double-Blind, Placebo-Controlled, Crossover & Open-Label Extension	6MWT and the Barth Syndrome Symptom Assessment (BTHS-SA) scale	Blinded phase: no significant difference. Open-label phase: improvements in 6MWT (P=0.02) and BTHS-SA fatigue score (P=0.03).
Phase 2, RCT of acipimox in mitochondrial myopathy EudraCT 2018-002721-29	Randomized, double-blind, placebo-controlled, adaptive design	Primary: ↑muscle ATP content; Secondary: exercise tolerance (cycle ergometry), symptom scores, metabolites	No (no significant ATP increase)
Phase 2 Treatment With Combination Pyrimidine Nucleosides in Patients With TK2 Deficiency	Open label & compassionate use	Safety, motor function, PFTs, growth, QoL, CGI, PGI	Improvement or disease stabilization across motor, respiratory, and feeding domains

The 6 Minute Walk Test (6MWT)

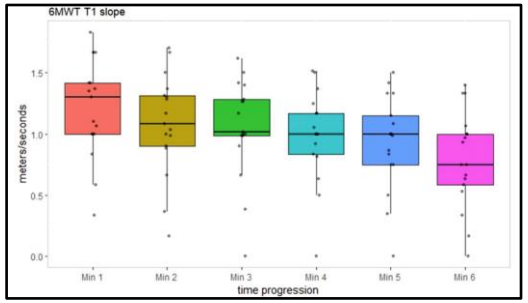
Standardized submaximal exercise test measuring the distance an individual can walk in 6 minutes, reflecting integrated cardiopulmonary and muscular function.

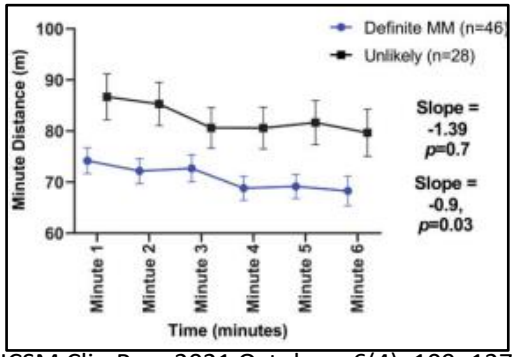






Journal of Neurology (2022) 269:6555–6565





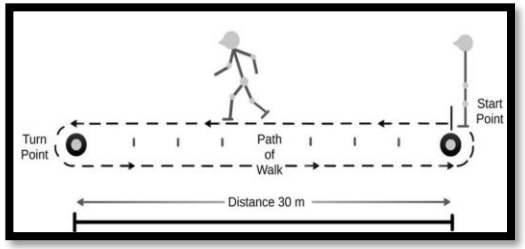
JCSM Clin Rep. 2021 October ; 6(4): 109–127

The 6 Minute Walk Test (6MWT)

Validity:
- Baseline mean 6MWT distance
- 6MWT scores correlations

Reliability:
- Significant test-retest variability
(>408 m showed more variability)
- Limited short-term responsiveness in PMD

Applicability (Adult vs Pediatric):
- Appropriate for ambulatory patients



Thesis: Benavent-Caballer, Vicent 2016

Regulatory perspective:

- Guidance
- Familiariaty as an established motor endpoint.
- Patient-centricity (do changes translate into real benefit)

The 6 Minute Walk Test (6MWT)

Use in PMD (pros)

- Has face and construct validity
- Widely used in PMM trials
  Simple and well-validated in other diseases

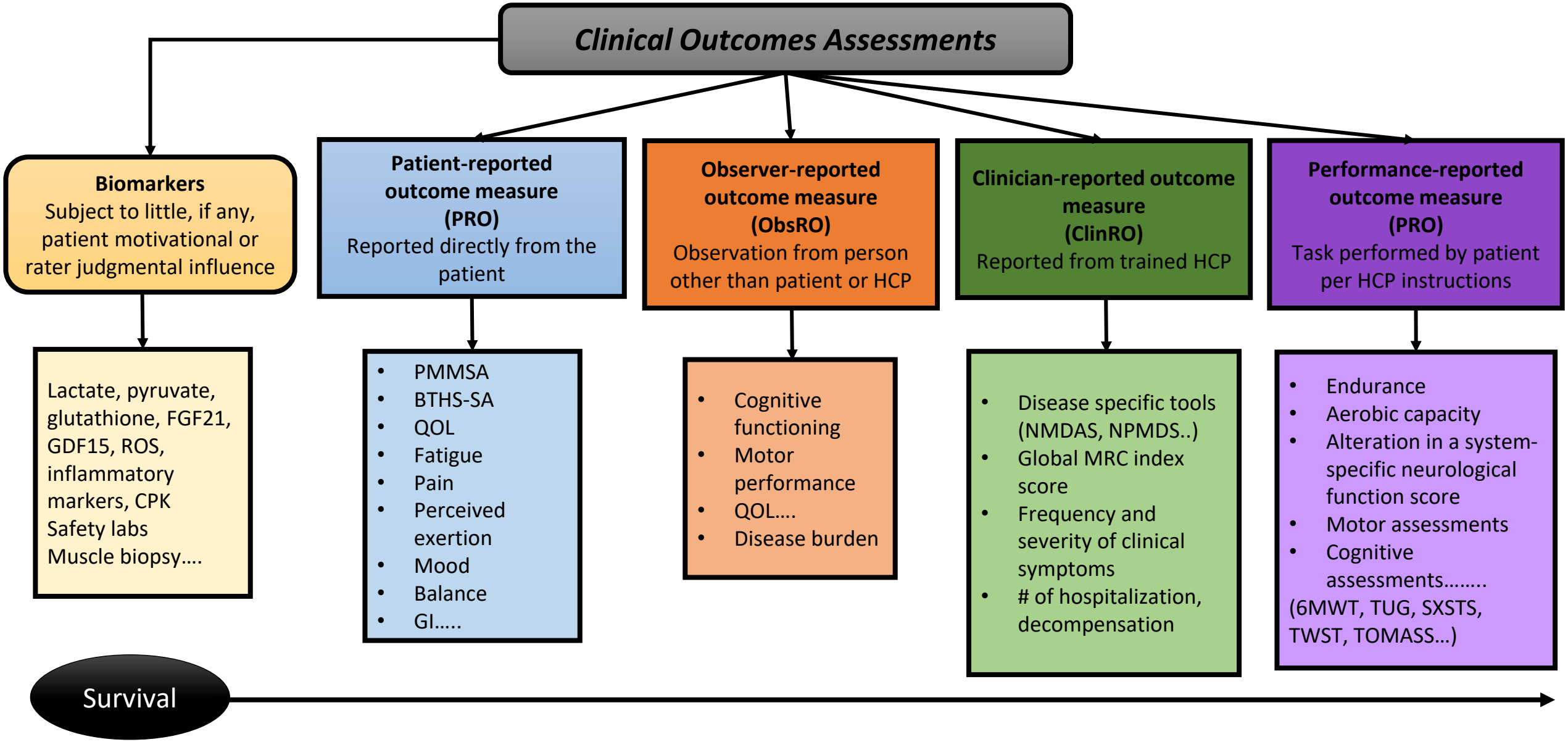
→ How about the 12MWT?

Clinical Trials in Primary Mitochondrial Diseases

Trial phase	Design	Primary / Secondary Outcomes	Endpoint Met?
Phase 2 RP103-MITO-001 Safety/tolerability/efficacy in inherited mitochondrial disease NCT02023866	Open-label, dose-escalation	Primary: Safety, tolerability; Secondary: metabolic biomarkers (e.g. glutathione)	Unknown (study terminated)
Phase 2b EPI-743 in Leigh Syndrome (NCT01721733)	Randomized, Placebo-Controlled, Double-Blind	Neuromuscular function, respiratory function, disease severity, morbidity & mortality, biomarkers	No
Phase 2/3 Vatiquinone trial (MIT-E) for mitochondrial epilepsy NCT04378075	Randomized, Double-Blind, Placebo-Controlled	Seizure frequency, disease-related hospitalizations, status epilepticus occurrences	No
Sonlicromanol (KH176) Phase 2b in m.3243A>G (MELAS/MIDD) (NCT02909400)	Randomized crossover, double-blind	Primary: Attention (Cogstate IDN); Secondary: fatigue, balance, pain, QoL	No, fatigue, balance and pain scores improved over 1-year treatment.

Clinical Trials in Primary Mitochondrial Diseases

Title NCT# (Trial phase)	Intervention	Design	Primary / Secondary Outcomes	Endpoint Met?	Key Results & Conclusions
GS010 (Lenadogene) Phase 3 LHON (RESCUE) NCT02652767 (Phase 3)	Lenadogene nolparvovec (AAV2 gene therapy) vs sham injection	Randomized, sham-controlled, double-masked	Primary: Change in best-corrected visual acuity (BCVA)	Yes	Gene therapy eye showed sustained vision gains. Five-year follow-up showed bilateral improvement in BCVA and good safety. Participants treated ≤6 mo had earlier gains; improvements persisted.
GS010 (Lenadogene) Phase 3 LHON (REVERSE) NCT02652780 (Phase 3)	Lenadogene nolparvovec vs sham	Randomized, sham-controlled	Primary: BCVA	Yes	Similar design to RESCUE. Long-term data again show durable bilateral BCVA gains with lenadogene vs sham. Supports persistent benefit of single-dose gene therapy.
RHODOS: RCT of idebenone in LHON EudraCT 2006-002679-42 (Phase 3)	Idebenone 900 mg/day vs placebo	Randomized, double-blind, placebo-controlled	Primary: Change in BCVA at 6 months	Yes (subgroup)	Overall trial missed significance, but subgroup analysis suggested higher rates of vision improvement in idebenone vs placebo, especially if treated early. Long-term follow-up (RHODOS-OFU) showed persistent benefit in treated patients.


Clinical Outcomes Assessments
Biomarkers
Subject to little, if any, patient motivational or rater judgmental influence
Patient-reported outcome measure (PRO)
Reported directly from the patient
Observer-reported outcome measure (ObsRO)
Observation from person other than patient or HCP
Clinician-reported outcome measure (ClinRO)
Reported from trained HCP
Performance-reported outcome measure (PRO)
Task performed by patient per HCP instructions
Lactate, pyruvate, glutathione, FGF21, GDF15, ROS, inflammatory markers, CPK
Safety labs
Muscle biopsy….
PMMSA
BTHS-SA
QOL
Fatigue
Pain
Perceived exertion
Mood
Balance
GI…..
Cognitive functioning
Motor performance
QOL….
Disease burden
Disease specific tools (NMDAS, NPMDS..)
Global MRC index score
Frequency and severity of clinical symptoms
# of hospitalization, decompensation
Endurance
Aerobic capacity
Alteration in a system-specific neurological function score
Motor assessments
Cognitive assessments……..
(6MWT, TUG, SXSTS, TWST, TOMASS…)
Survival

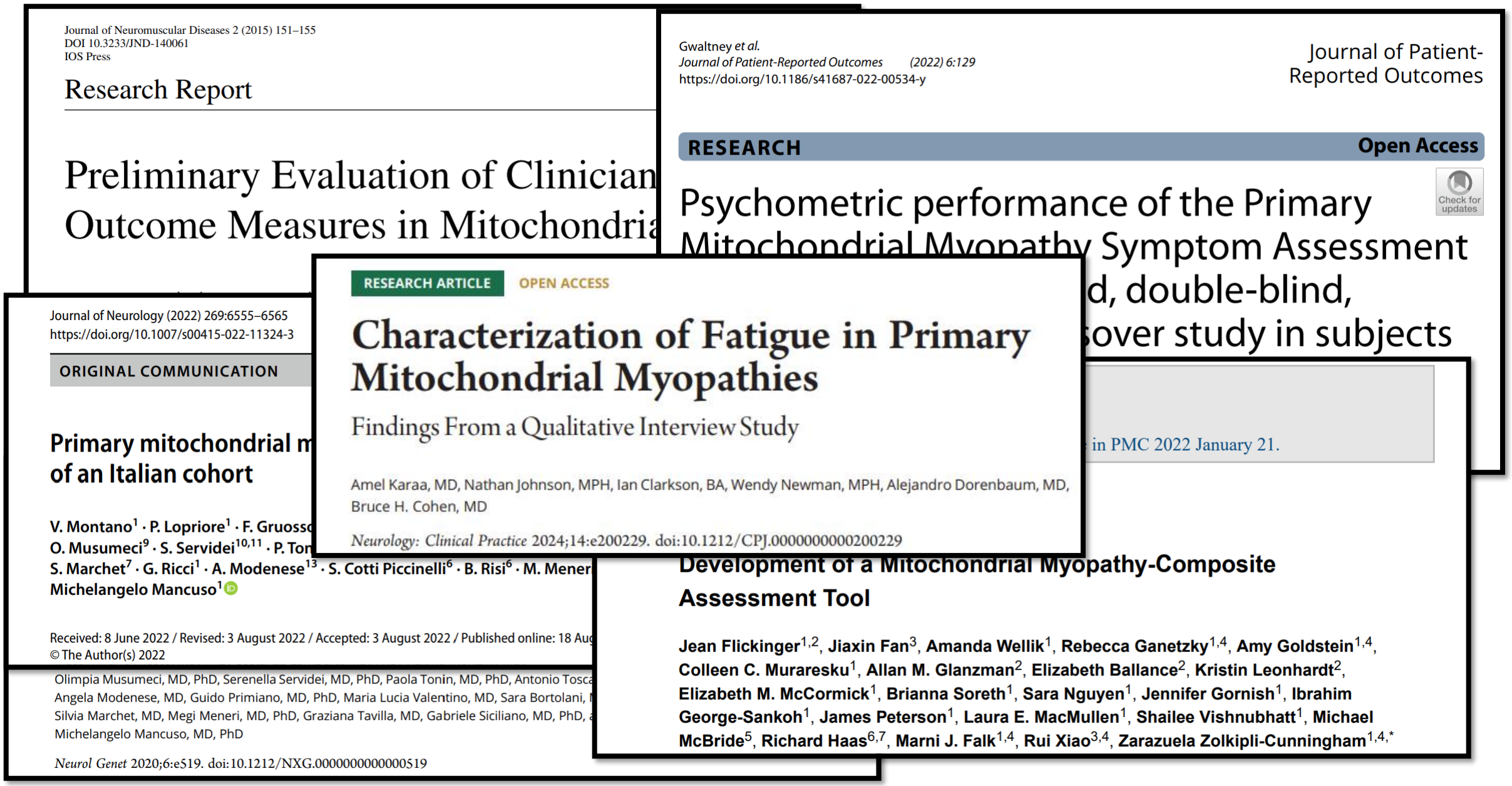
Outcome Type	Use in PMD (pros)	Use in PMD (cons)
PROMs (Fatigue/QoL) Fatigue Severity Scale SF-36, NeuroQoL PGI, PGC…	Reflect patient-perceived symptoms FDA values patient-reported benefit.	Highly subjective, placebo-prone, and may not correlate with objective measures (sometimes fatigue improves despite no 6MWT change).

LESSONS LEARNED: COA

- Need for validation in specific disease subgroups (genotype and/or phenotype specific)
- S
- I
(I
- Should not be too specialized & needing very specific expertise (may not be practical for all trial settings)

Multimodal Approach:

No single outcome measure is universally superior.
→ Role for composite scoring and GST approaches?



Journal of Neuromuscular Diseases 2 (2015) 151–155
DOI 10.3233/JND-140061
IOS Press

Research Report

Preliminary Evaluation of Clinician
Outcome Measures in Mitochondria

Gwaltney *et al.*
Journal of Patient-Reported Outcomes (2022) 6:129
https://doi.org/10.1186/s41687-022-00534-y

Journal of Patient-Reported Outcomes

RESEARCH Open Access

Psychometric performance of the Primary Mitochondrial Myopathy Symptom Assessment
d, double-blind,
over study in subjects

Journal of Neurology (2022) 269:6555–6565
https://doi.org/10.1007/s00415-022-11324-3

ORIGINAL COMMUNICATION

Primary mitochondrial m
of an Italian cohort

V. Montano[1] · P. Lopriore[1] · F. Gruosso
O. Musumeci[9] · S. Servidei[10,11] · P. Ton
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RESEARCH ARTICLE OPEN ACCESS

Characterization of Fatigue in Primary Mitochondrial Myopathies

Findings From a Qualitative Interview Study

Amel Karaa, MD, Nathan Johnson, MPH, Ian Clarkson, BA, Wendy Newman, MPH, Alejandro Dorenbaum, MD, Bruce H. Cohen, MD

Neurology: Clinical Practice 2024;14:e200229. doi:10.1212/CPJ.0000000000200229

in PMC 2022 January 21.

Development of a Mitochondrial Myopathy-Composite Assessment Tool

Jean Flickinger[1,2], Jiaxin Fan[3], Amanda Wellik[1], Rebecca Ganetzky[1,4], Amy Goldstein[1,4], Colleen C. Muraresku[1], Allan M. Glanzman[2], Elizabeth Ballance[2], Kristin Leonhardt[2], Elizabeth M. McCormick[1], Brianna Soreth[1], Sara Nguyen[1], Jennifer Gornish[1], Ibrahim George-Sankoh[1], James Peterson[1], Laura E. MacMullen[1], Shailee Vishnubhatt[1], Michael McBride[5], Richard Haas[6,7], Marni J. Falk[1,4], Rui Xiao[3,4], Zarazuela Zolkipli-Cunningham[1,4,*]

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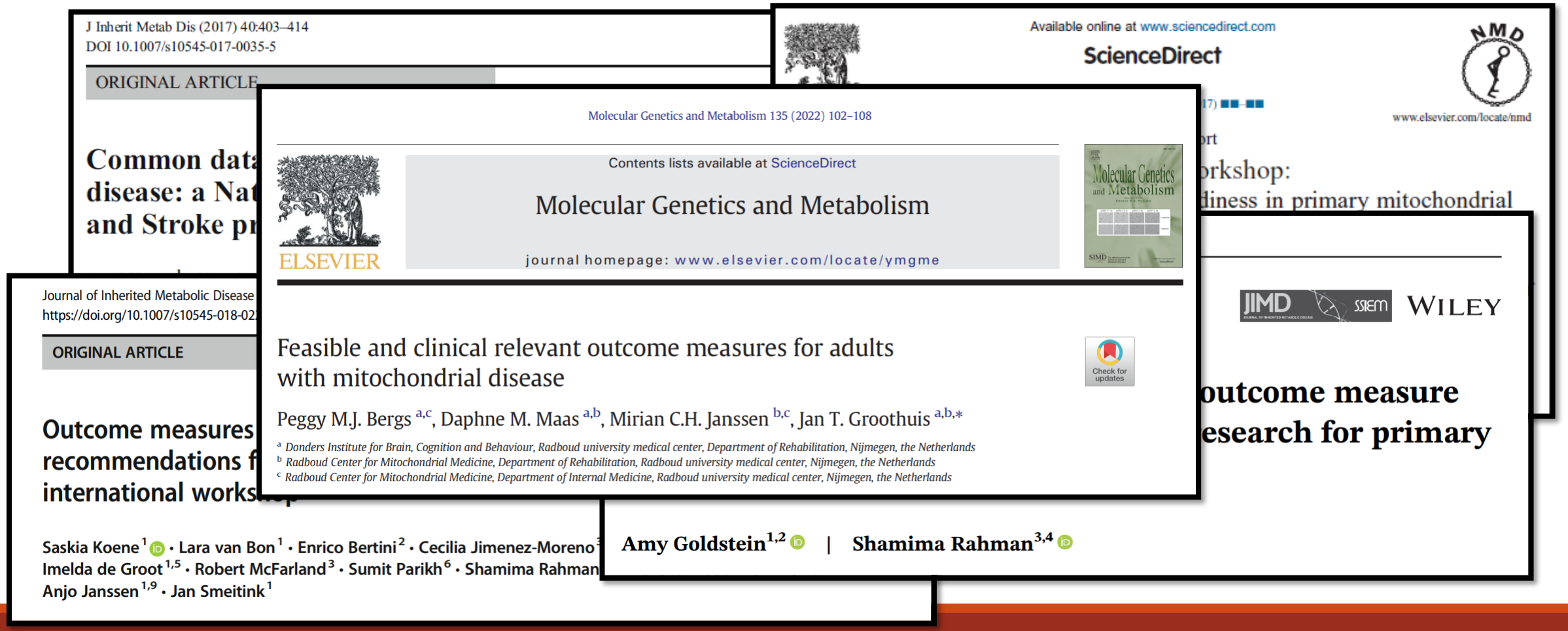
Silvia Marchet, MD, Megi Meneri, MD, PhD, Graziana Tavilla, MD, Gabriele Siciliano, MD, PhD,
Michelangelo Mancuso, MD, PhD

Neurol Genet 2020;6:e519. doi:10.1212/NXG.0000000000000519

LESSONS LEARNED:TRIALS

- What does the drug really do?
- How long does it take for a change to happen?
- Are we treating long enough to observe a change?
- How do we minimize the large placebo effect?
- Open label studies may not be representative of true effect

LOOKING TO THE FUTURE



LOOKING TO THE FUTURE

all
s)

Thank you

For any questions:

Amel Karaa, MD

Massachusetts General Hospital
Harvard Medical School

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Lunch

The meeting will resume at 1:30pm ET

REAGAN-UDALL FOUNDATION FOR THE FDA

Current Approaches, Challenges, and Opportunities

- **Magnus Hansson, MD, PhD**
  Abliva AB

- **Chad Glasser, PharmD, MPH**
  Tisento Therapeutics

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Pharming®

Targeting the powerhouse of cells to improve the lives of primary mitochondrial disease patients

Magnus Hansson, MD, PhD
Executive Medical Director,
KL1333 Global Program Lead

Disclaimer

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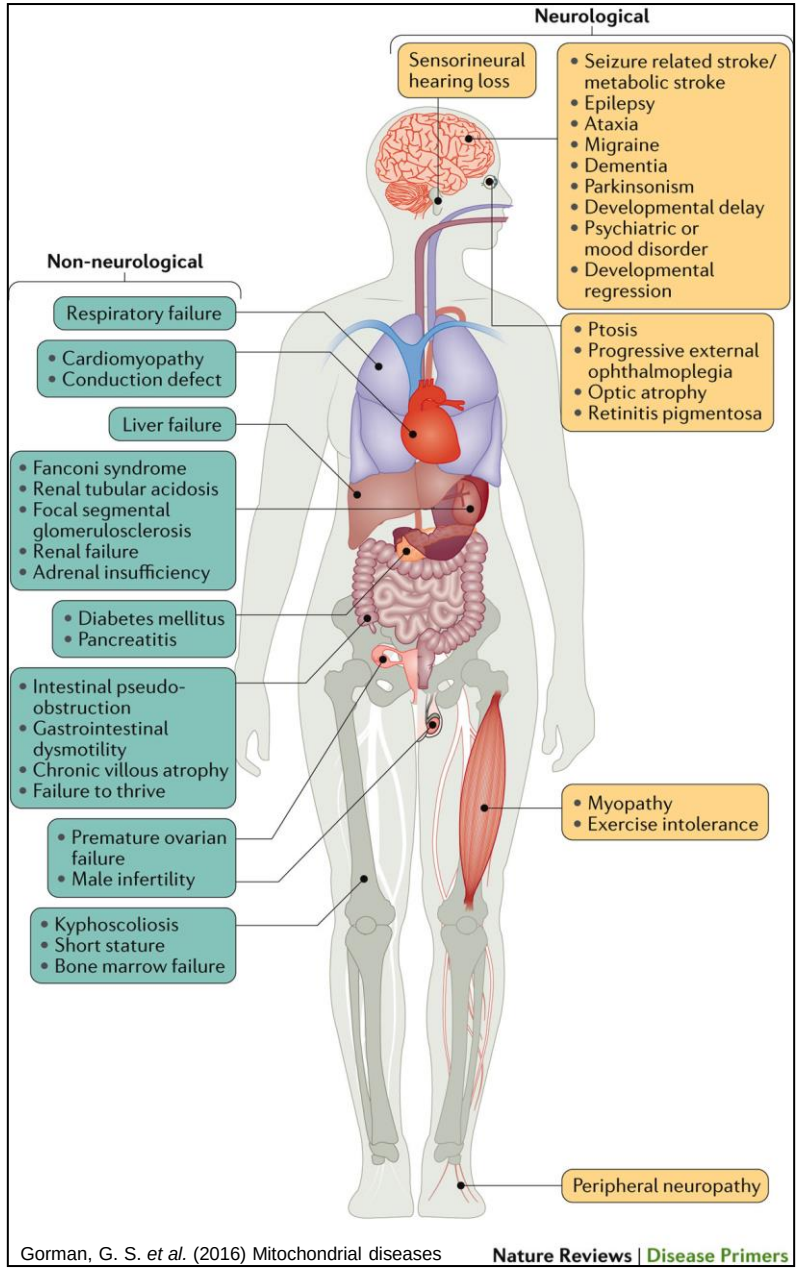
FALCON study

Evaluating safety and efficacy of KL1333 in adult patients with primary mitochondrial disease

- Ongoing placebo-controlled study with registrational design
- Study focus on adult patients with multisystemic disease caused by pathogenic point mutations or large deletion of the mitochondrial genome (mtDNA)



The challenges and opportunities of mitochondrial disease

- Powering almost every cell in our bodies, mitochondrial disease can present with a multitude of symptoms

- No approved therapies
  - Challenge: no regulatory precedent
  - Opportunity: meaningful impact of a high unmet need

What matters to patients?

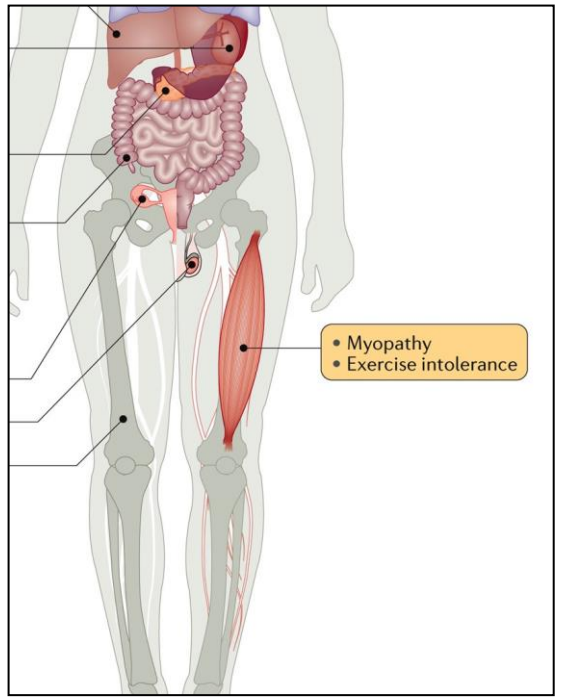
- Q: Which abilities or symptoms would you rank as most important for a possible drug treatment today?
  1. **Reduction in chronic fatigue (68%)**
  2. **Reduction in muscle weakness (57%)**

https://www.umdf.org/wp-content/uploads/2019/12/UMDF-EL-PFDD-Meeting-VOP-Report_v2019-12-03.pdf

Clinical trial program to evaluate KL1333

Can we assess the abilities and symptoms that the patients prioritize?

"I started missing work because I was too exhausted to even get up to use the rest room."



"I am afraid of what the progressive muscle weakness will do to me, not being able to care for myself is a concern."

From the Voice of the patient report, UMDF 2019

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Fatigue is common in mitochondrial disease

Multiple studies demonstrate consistent findings

Study of 132 patients in a specialist outpatient clinic in the UK found that:

- "Fatigue was common… with 64% of patients reporting excessive symptomatic fatigue" [1]

Study at 10 national centers in the US found that:

- "fatigue is very common amongst patients with PMD, with 71-100% of patients reporting fatigue" [2]

➢In both studies, fatigue correlated with overall disease burden and severity (NMDAS total score), but not with myopathy scores

1. Gorman et al. Neuromuscular Disorders 25 (2015) 563–566
2. Parikh et al. Neuromuscular Disorders 29 (2019) 895–902

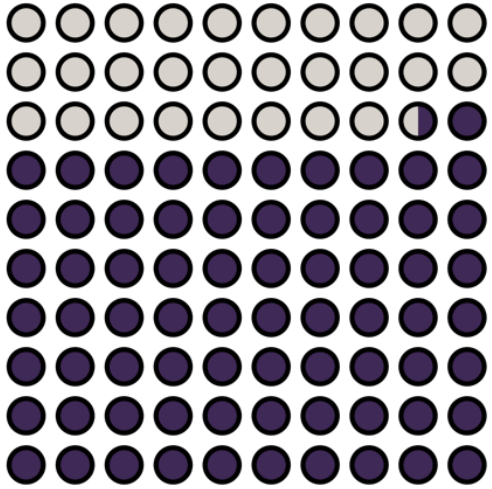
Myopathy is common in mitochondrial disease

High proportion of patients with myopathy confirmed in natural history registry

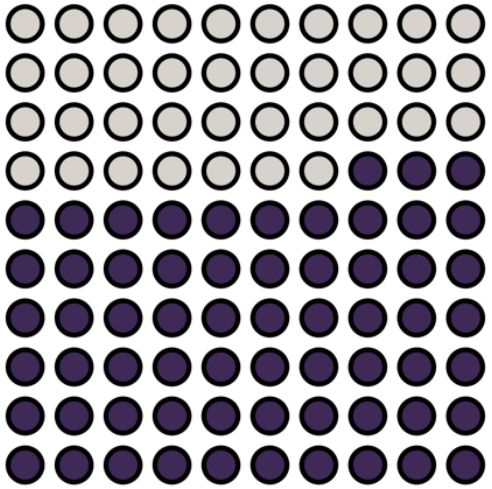
- Study mitochondrial disease registry in the UK (MitoCohort):

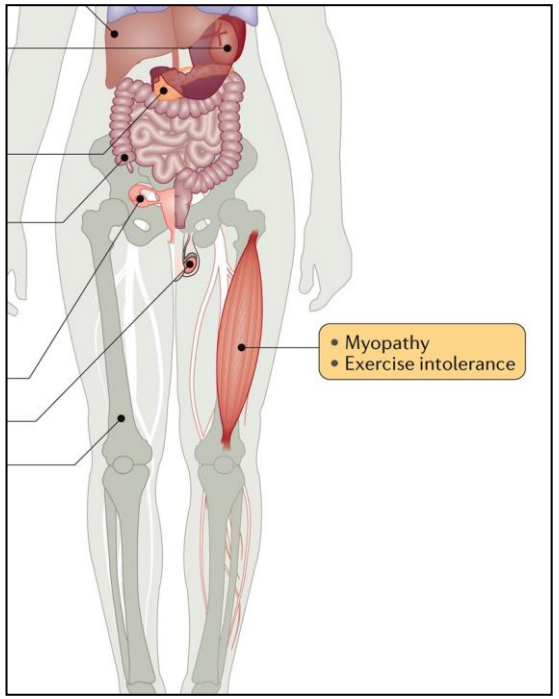
  (>500 adults with active entries over the past 3 years)

Exercise Intolerance (72%)



Proximal muscle weakness (63%)





Collaboration with the Wellcome Centre for Mitochondrial Research, Newcastle University, UK

Early test of the Clinical Outcome Assessment strategy

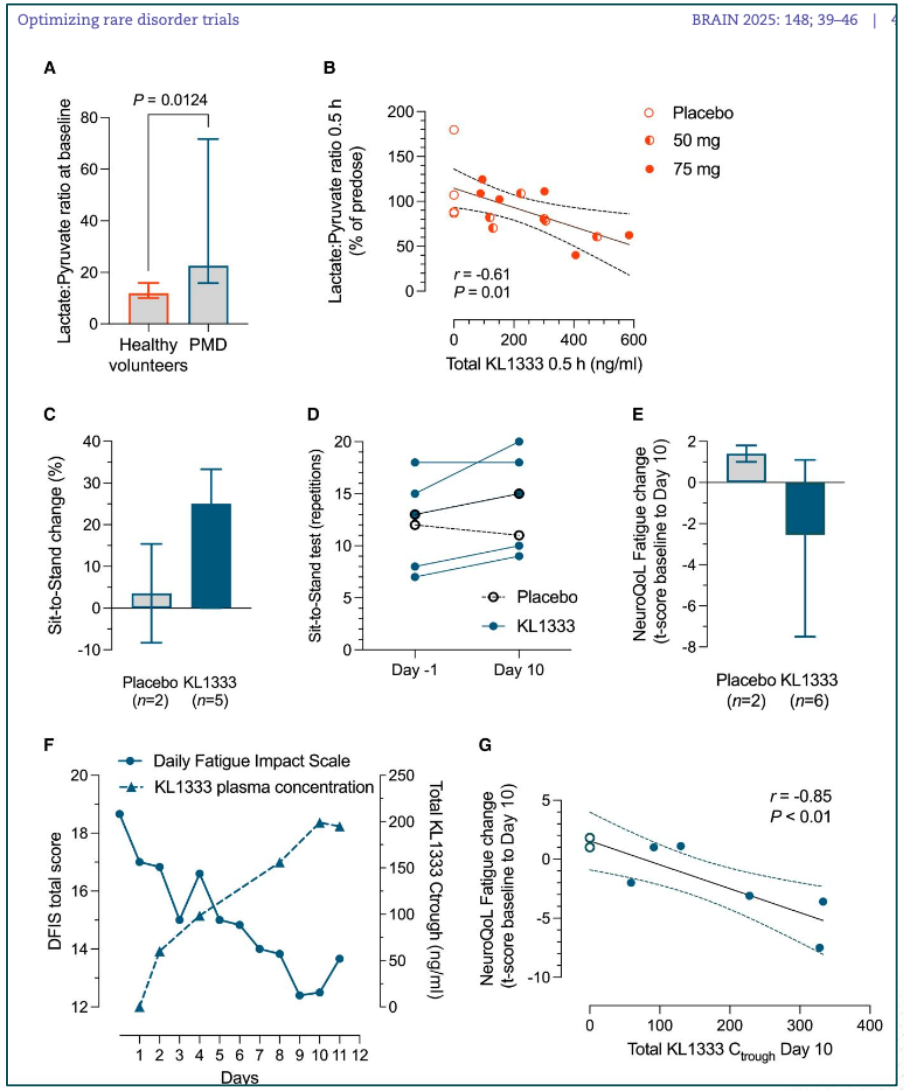
Phase 1b randomized controlled study

https://doi.org/10.1093/brain/awae308 BRAIN 2025: 148; 39–46 | 39

BRAIN REPORT

Optimizing rare disorder trials: a phase 1a/1b randomized study of KL1333 in adults with mitochondrial disease

Chiara Pizzamiglio,[1,2,†] Renae J. Stefanetti,[3,4,5,†] Robert McFarland,[3,4,5] Naomi Thomas,[3,4,5] George Ransley,[6] Matilda Hugerth,[7] Alvar Grönberg,[7] Sonia Simon Serrano,[7,8] Eskil Elmér,[7,8] Michael G. Hanna,[1,2] Magnus J. Hansson,[7,8] Gráinne S. Gorman[3,4,5,‡] and Robert D. S. Pitceathly[1,2,‡]



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Patient interview study to enable a primary endpoint for fatigue

Development of a fit-for-purpose Clinical Outcome Assessment for mitochondrial disease

Patient interviews

1. Concept elicitation

2. Cognitive interview

Mapping of fatigue questions

Existing short forms and item banks (PROMIS®)

Selection of final fatigue items

PROMIS® Fatigue Mitochondrial Disease Short Form

ABLIVA
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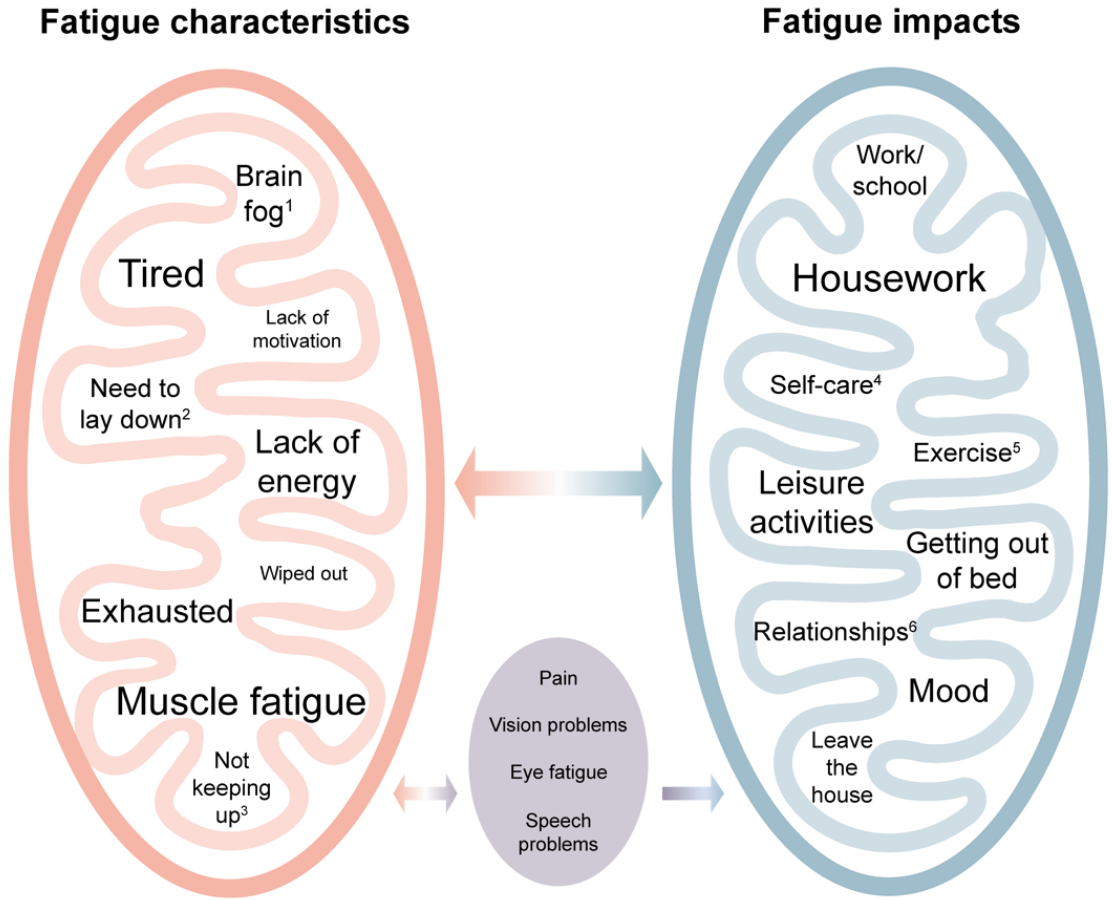
Concept elicitation interviews

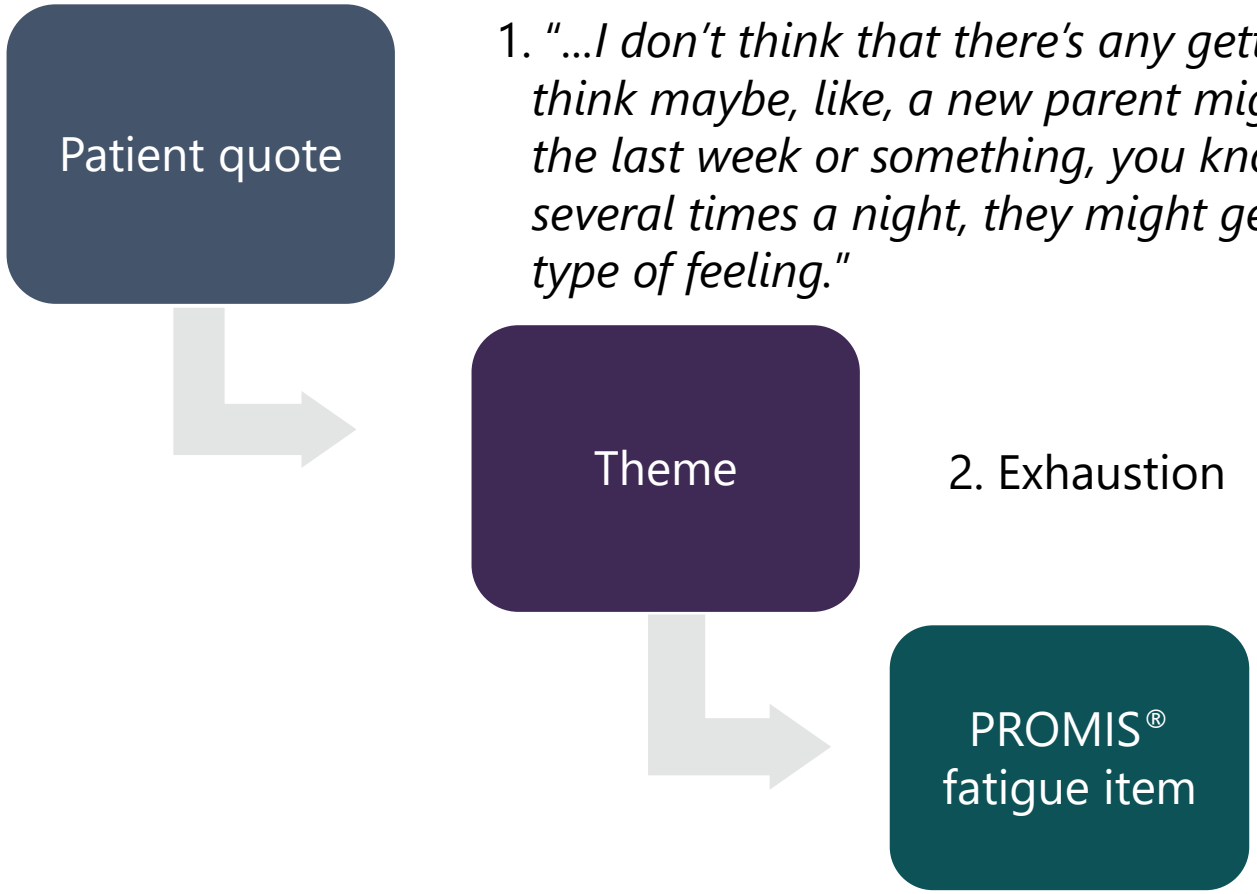
Fatigue-related themes and intersection with other PMD symptoms



Fatigue characteristics and impacts on daily life reported by at least 50% of study participants.

The level of endorsement (frequency of reporting) is proportional to the font size of the respective theme

Concept Elicitation and Mapping of Fatigue Questions



Cognitive interviews

Examples of PROMIS fatigue item performance in Cognitive Interviews

Fatigue item	Interpreted correctly	Easy to understand or answer	Relevant to fatigue experience	Able to select a response option
FATIMP30 **How often were you too tired to think clearly?**	100%	92%	100%	100%
FATIMP26 **How often were you too tired to socialize with your family?**	100%	75%	67%	75%

PROMIS® Fatigue Mitochondrial Disease Short Form

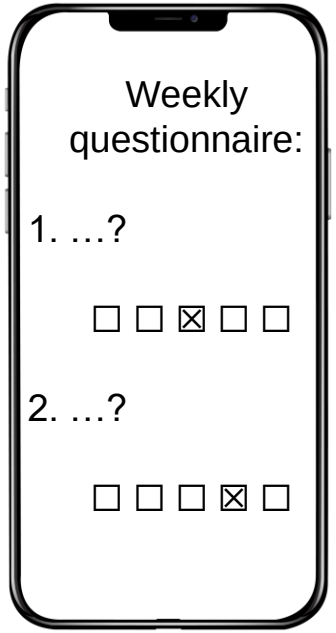
In the past 7 days…	Never	Rarely	Sometimes	Often	Always
How often did you feel tired?	☐	☐	☐	☐	☐
How often did you run out of energy?	☐	☐	☐	☐	☐
How often did you experience extreme exhaustion?	☐	☐	☐	☐	☐
How often were you too tired to think clearly?	☐	☐	☐	☐	☐
How often were you too tired to do your household chores?	☐	☐	☐	☐	☐
How often were you too tired to enjoy life?	☐	☐	☐	☐	☐
How often were you too tired to leave the house?	☐	☐	☐	☐	☐
How often were you too tired to take a bath or shower?	☐	☐	☐	☐	☐
How often did you have to push yourself to get things done because of your fatigue?	☐	☐	☐	☐	☐

Content validity study manuscript submitted for publication. Preprint available at https://papers.ssrn.com/sol3/papers.cfm?abstract_id=5043346

The Clinical Outcome Assessment for fatigue is tested in the participant's home environment to reflect impact on daily life

ePRO Endpoint

PROMIS® Fatigue Mitochondrial Disease Short Form



- ePRO questionnaire is triggered before study visits and every week
- Answered in participant's usual home environment rather than after travelling to the study clinic

FALCON study analysis approach

Two alternative primary endpoints* evaluates changes in fatigue and muscle function

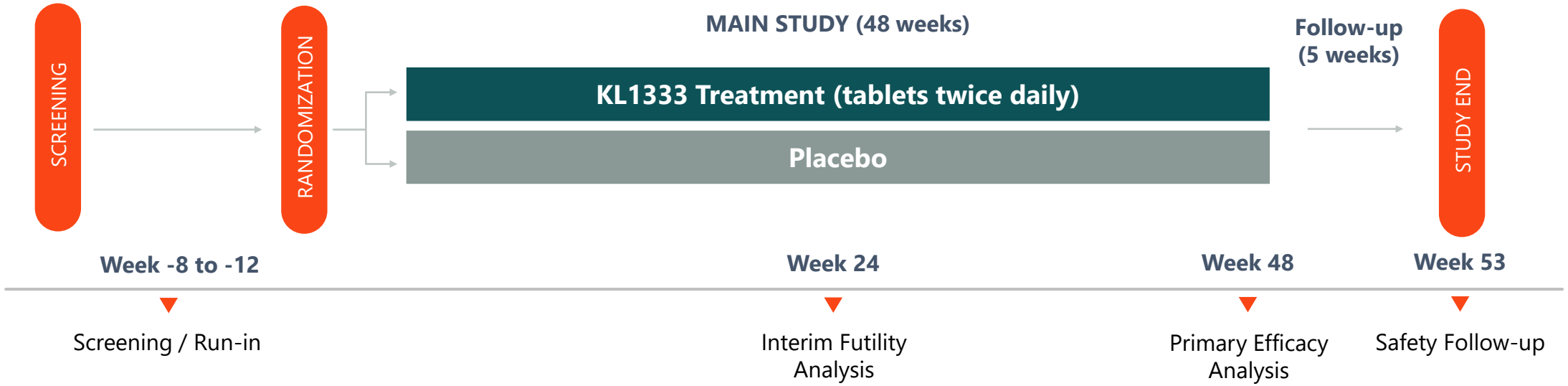
(1) PROMIS® Fatigue Mitochondrial Disease Short Form

In the past 7 days…	Never	Rarely	Some times	Often	Always
How often did you experience extreme exhaustion?	☐	☐	☐	☐	☐
How often did you run out of energy?	☐	☐	☐	☐	☐
How often did you feel tired?	☐	☐	☐	☐	☐
How often were you too tired to enjoy life?	☐	☐	☐	☐	☐
How often did you have to push yourself to get things done because of your fatigue?	☐	☐	☐	☐	☐
How often were you too tired to do your household chores?	☐	☐	☐	☐	☐
How often were you too tired to take a bath or shower?	☐	☐	☐	☐	☐
How often were you too tired to leave the house?	☐	☐	☐	☐	☐
How often were you too tired to think clearly?	☐	☐	☐	☐	☐

(2) 30 second Sit-to-Stand (30s STS) test:

*Study is positive if either of the two endpoints, or both, demonstrate benefit

FALCON study design



PARTICIPANTS: Adult Primary Mitochondrial Disease patients (mtDNA mutations*) with myopathy and fatigue

DESIGN: Randomized, double-blind, parallel-group, placebo-controlled (40% placebo, 60% active)

ALTERNATIVE PRIMARY ENDPOINTS: PROMIS® Fatigue Mitochondrial Disease Short Form, 30 Second Sit-to-Stand test

*Including MIDD-MELAS-m.3243A>G associated spectrum disease, single large scale mtDNA deletion associated KSS-CPEO spectrum disorders, MERRF, and other multisystemic mitochondrial DNA-related disease.;

Conclusions

KL1333 for Primary Mitochondrial Disease

- The KL1333 development program shows that it is feasible to study both fatigue and myopathy in an interventional trial for mitochondrial disease
  - Both symptoms are common in adults with mitochondrial disease
- Collaboration between stakeholders (FDA, advocacy, KOLs and industry) enabled the successful development of a mitochondrial disease-specific fatigue COA
  - Content validity established for the PROMIS® Fatigue Mitochondrial Disease Short Form

➢This has enabled a patient centric study with two primary objectives and endpoints reflecting the highest ranked needs from the voice of the patients

Thank you!

Tisento's Approach to Overcoming Drug Development Challenges in MELAS

Chad Glasser PharmD MPH

Sr. Director of Clinical Research

PMD Virtual Public Workshop
Hosted by Reagan-Udall Foundation for the FDA
May 22nd, 2025

About Tisento Therapeutics

Tisento Therapeutics is a clinical-stage biopharmaceutical company focused on developing novel medicines to treat diseases with significant unmet medical needs, beginning with **MELAS** and other genetic mitochondrial diseases

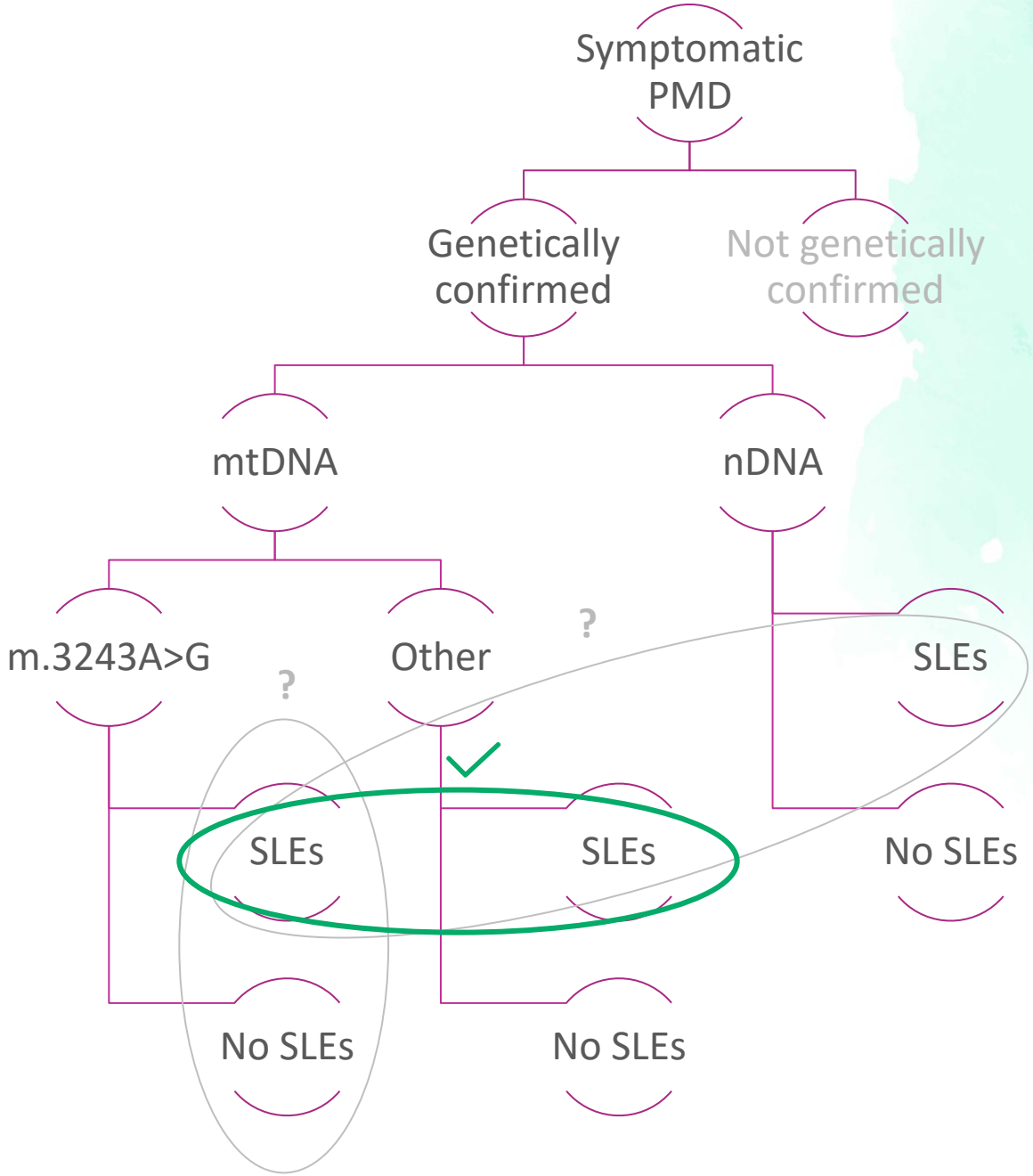
- **Our focus:** Zagociguat, oral, once-daily, first-in-class, brain-penetrant soluble guanylate cyclase (sGC) stimulator in Phase 2b development for **MELAS**
- **Internal team: 12** employees
- **Headquarters: Cambridge**, MA

PRIZM
MELAS CLINICAL TRIAL

Tisento™ THERAPEUTICS

Challenge #1: Defining MELAS

- **Why this matters** → Changing disease definitions greatly impair ability to utilize prior research and to estimate prevalence and clinical trial feasibility
- Varying definitions of MELAS used over the past 30+ years
  - For example, the m.3243A>G pathogenic variant has historically been equated to MELAS
- MELAS is now defined by a history of SLE(s)
  - Though the definition of a SLE also varies



SLE = stroke-like episode

Challenges and Phase 2b PRIZM study approach

PRIZM
MELAS CLINICAL TRIAL

Partner with regulators, patient advocacy, and KOLs

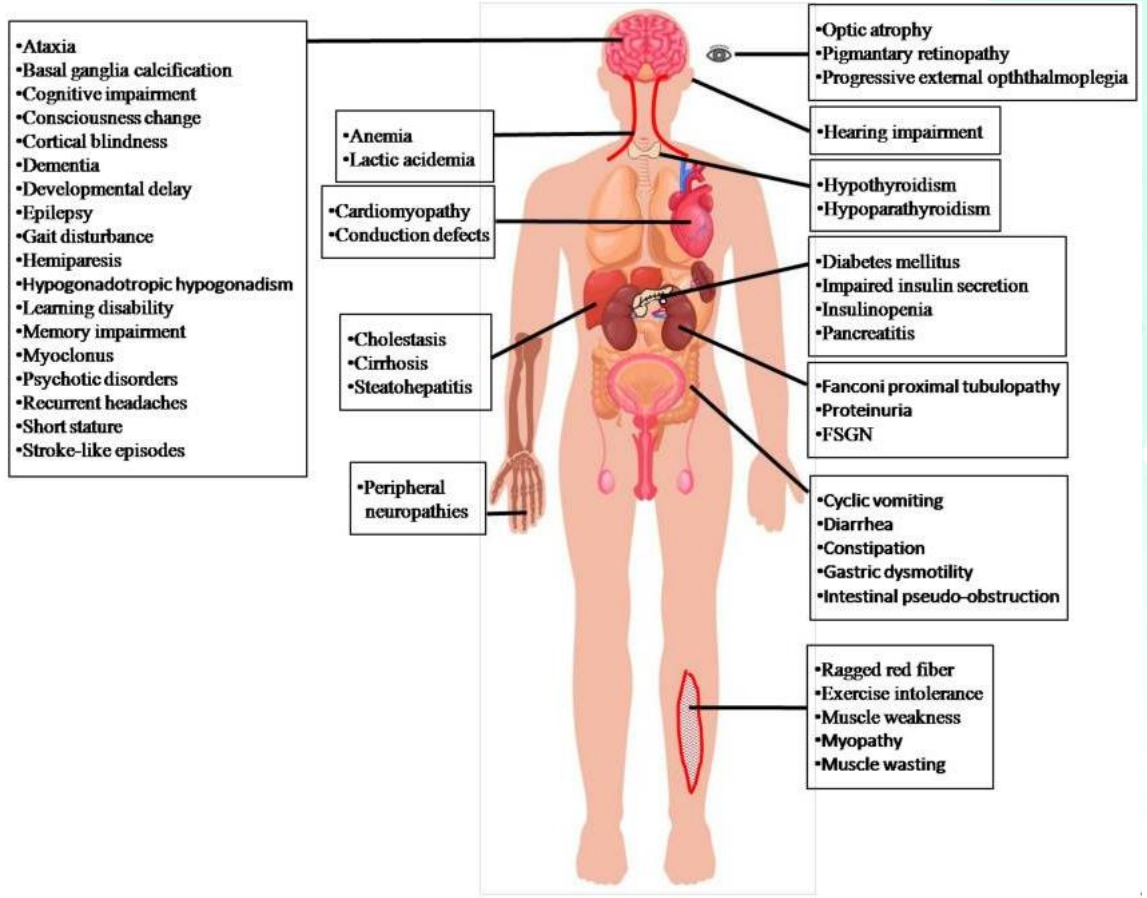
#1: Defining MELAS

Tisento™ THERAPEUTICS

KOL= Key opinion leader

Challenge #2: Characterizing the MELAS disease experience

- Complex, multi-system, heterogeneous
- Symptomatology, severity, and progression vary across patients
- Major events, such as SLEs, are relatively infrequent and difficult to predict
- Understanding the patient experience can help focus development efforts
  - "What are the most frequent and bothersome symptoms that impact daily life in the majority of patients?"



Tisento™ THERAPEUTICS

Fan HC, Lee HF, Yue CT, Chi CS. Clinical Characteristics of Mitochondrial Encephalomyopathy, Lactic Acidosis, and Stroke-Like Episodes. Life (Basel). 2021 Oct 20;11(11):1111.

Challenges and Phase 2b PRIZM study approach

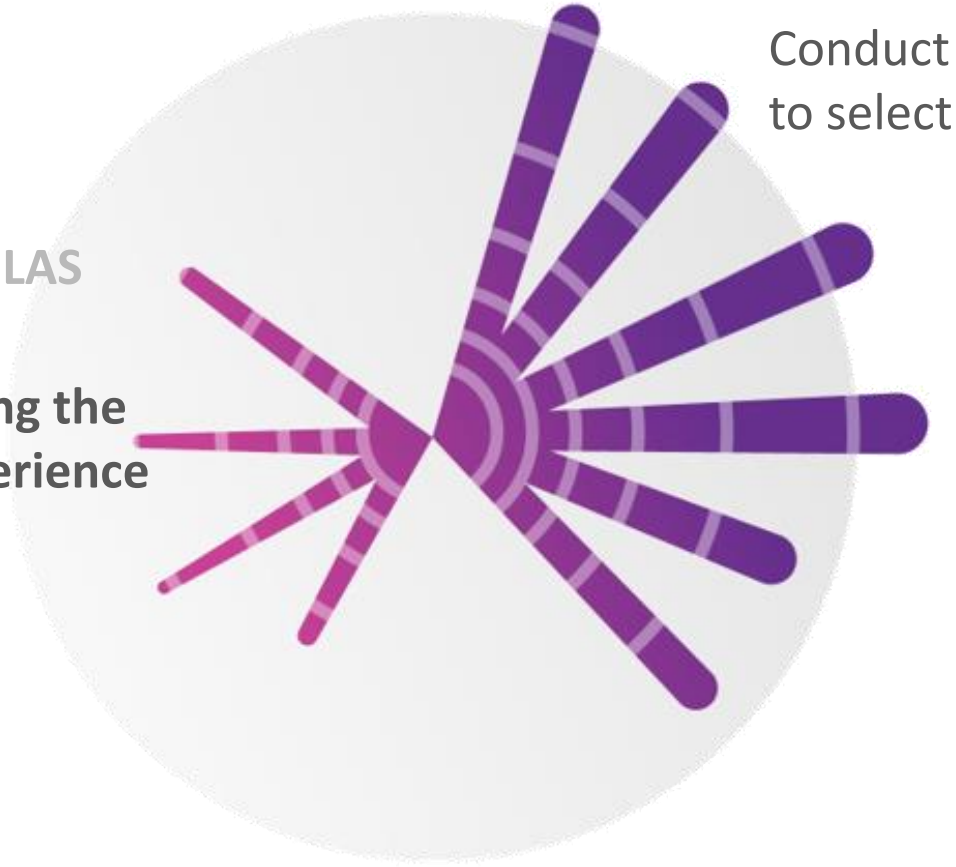
PRIZM
MELAS CLINICAL TRIAL

Partner with regulators, patient advocacy, and KOLs

Conduct a **qualitative patient/expert MELAS interview study** to select outcome measures and build endpoint model

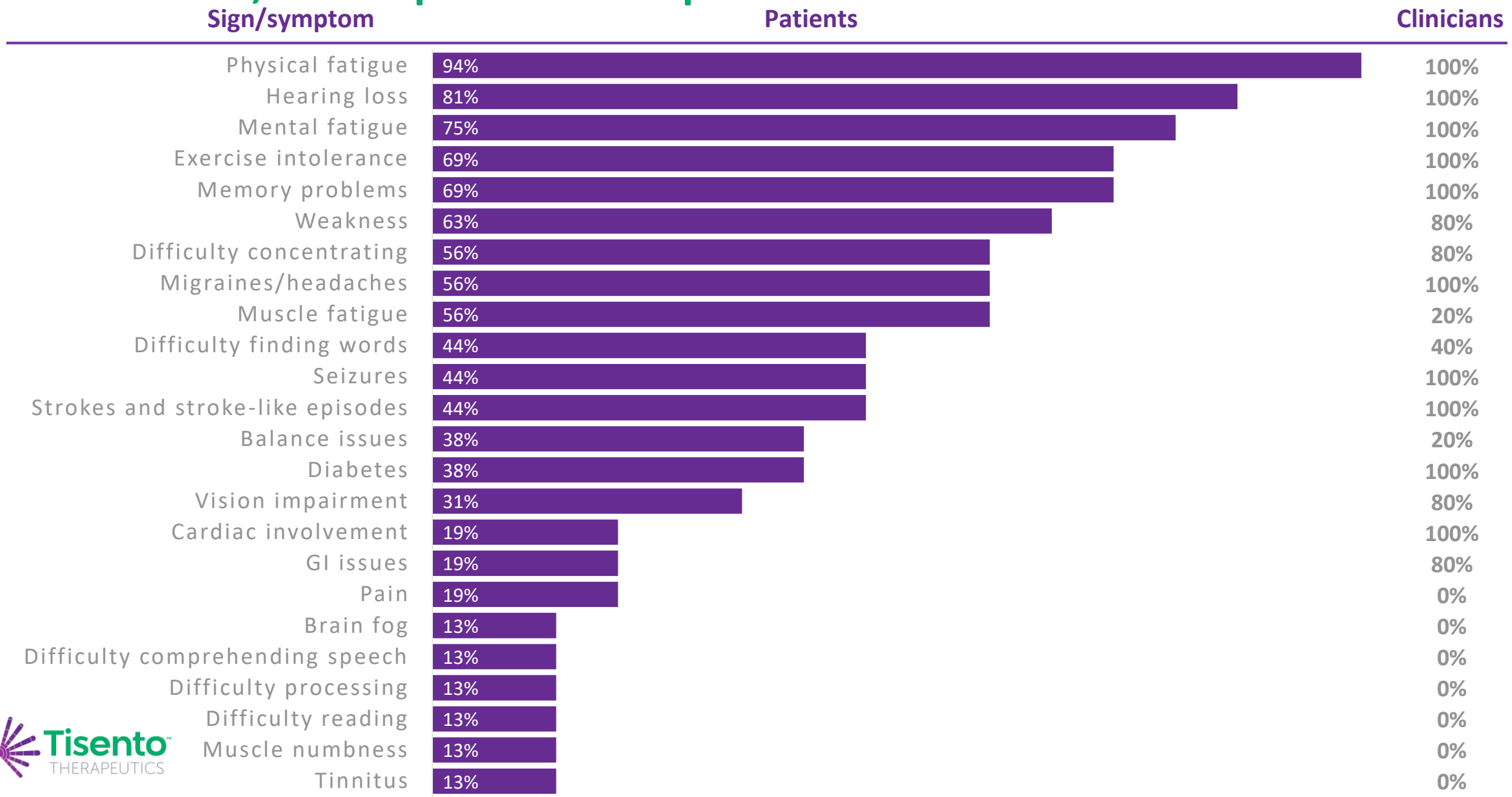
#1: Defining MELAS

#2: Characterizing the disease experience



KOL= Key opinion leader

Symptoms related to fatigue and cognitive impairment are frequent, bothersome, and important to improve



Tisento THERAPEUTICS

Challenge #3: Assessing treatment benefit

- There is no one-size-fits-all, sensitive, validated outcome measure able to assess treatment effects in MELAS
- There are no established or widely accepted biomarkers of disease
- PROs and PerfOs aimed at assessing the important and more frequent neurological manifestations of MELAS are needed
  - Previous trials that enrolled patients with MELAS incorporated outcome measures focused more on myopathy symptoms (e.g., 6MWT)
- Establishing adequate statistical power in rare disease remains a challenge

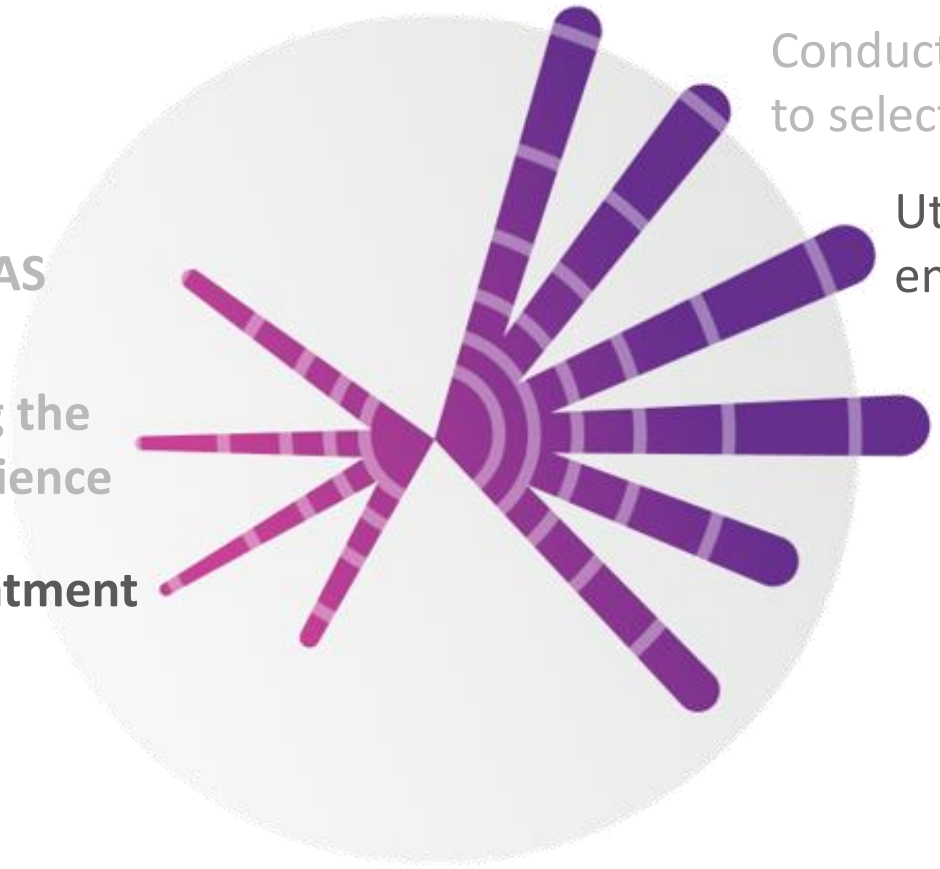
Challenges and Phase 2b PRIZM study approach

PRIZM
MELAS CLINICAL TRIAL

#1: Defining MELAS

#2: Characterizing the disease experience

#3: Assessing treatment benefit



Partner with regulators, patient advocacy, and KOLs

Conduct a **qualitative patient/expert MELAS interview study** to select outcome measures and build endpoint model

Utilize a **crossover design** with a **GST** in the primary endpoint to increase power and address heterogeneity

Collect data **weekly** via fit-for-purpose **PRO *and* PerfO** measures focused on **fatigue** and **cognition**

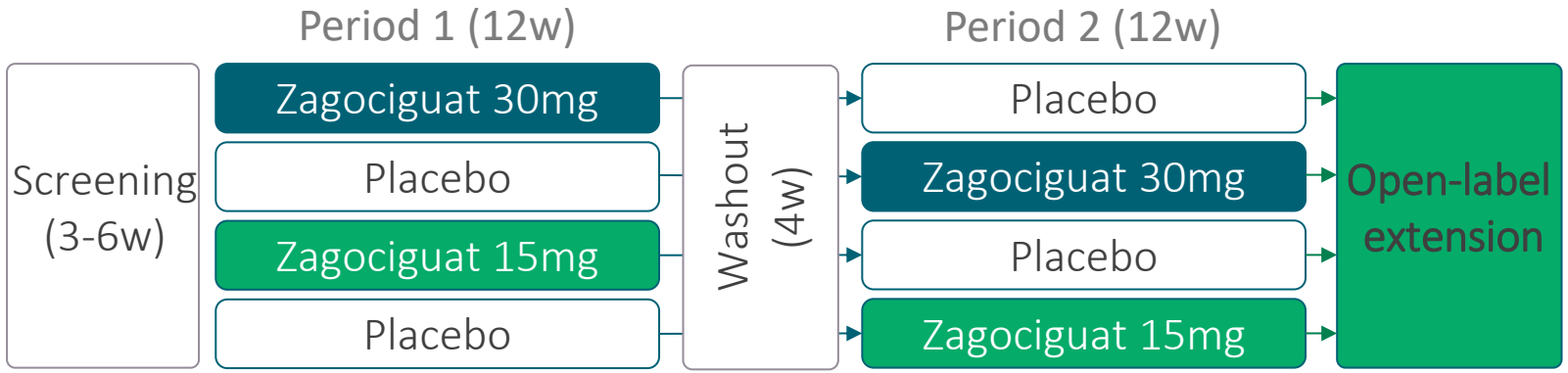
Tisento™ THERAPEUTICS

KOL= Key opinion leader; PRO= patient reported outcome; PerfO= Performance outcome, GST=Global statistical test

PRIZM study design

PRIZM
MELAS CLINICAL TRIAL

A Phase 2b randomized, double-blind, placebo-controlled crossover study investigating the efficacy and safety of zagociguat in participants with MELAS



Total Sample Size = ~44 participants

Global Study: 25 sites in US, Canada, UK, Italy, Germany, Australia

Tisento THERAPEUTICS

PRIZM study design

PRIZM
MELAS CLINICAL TRIAL

A Phase 2b randomized, double-blind, placebo-controlled crossover study investigating the efficacy and safety of zagociguat in participants with MELAS

Key eligibility criteria: ≥ 18 yrs of age with a mtDNA mutation and history of stroke-like episodes/lesions

Primary Objectives and Endpoints:

- Efficacy: to evaluate the effects of zagociguat on **fatigue** and **cognition**
  - PROMIS Fatigue MELAS Short Form scores
  - Groton Maze Learning Test scores (executive function)
  - International Digit Symbol Substitution Test scores (processing speed)

  *These 3 endpoints will be combined using a **global statistical test** (GST)*

- Safety: To evaluate **safety** and **tolerability** of zagociguat

Tisento™ THERAPEUTICS

Challenge #4: Enrolling a MELAS-focused clinical trial

- Patient selection and homogeneity are key to robust clinical trial design but further limit the already small patient pool
- Timelines and enrollment at each site are difficult to predict due to multiple factors:
  - No precedent or similar MELAS trials for comparison
  - Regulatory environment (FDA, EMA, CTIS, local IRBs)
  - Site start-up delays (site resources, contract/budget processes, training, scheduling)
  - Only a select number of sites with mitochondrial disease expertise, primarily academic institutions
  - Complicated patient population
- Patients are still hesitant to participate in clinical research

Challenges and Phase 2b PRIZM study approach

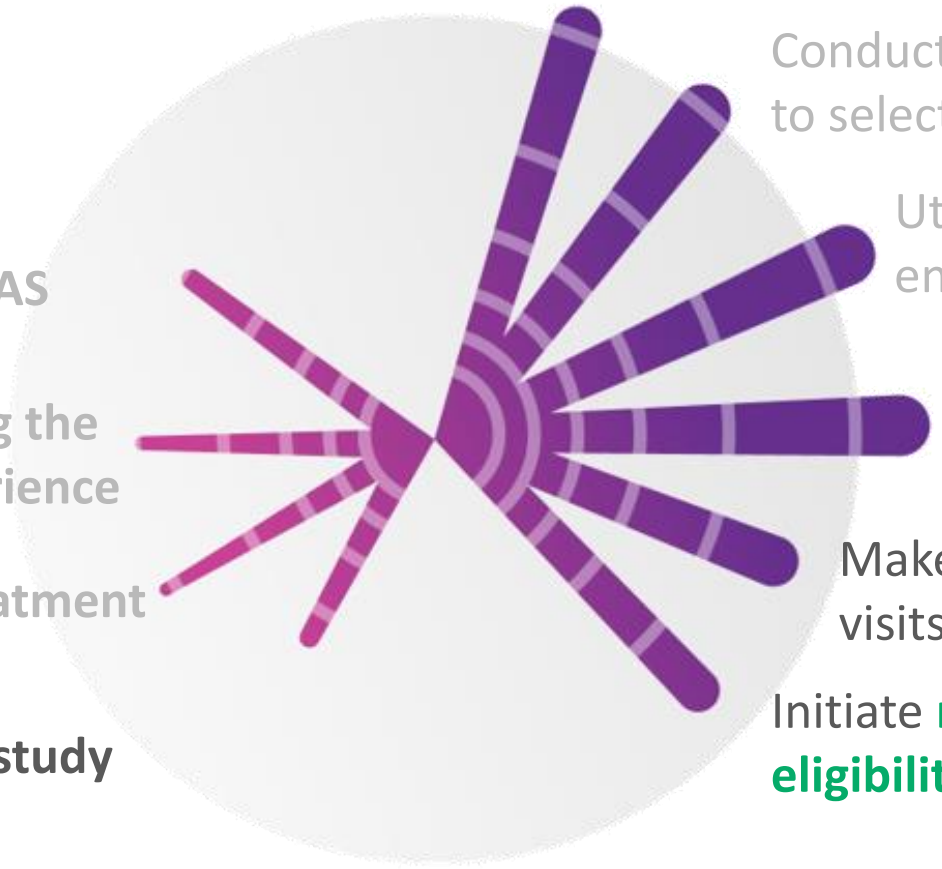
PRIZM
MELAS CLINICAL TRIAL

#1: Defining MELAS

#2: Characterizing the disease experience

#3: Assessing treatment benefit

#4: Enrolling the study



Partner with regulators, patient advocacy, and KOLs

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Utilize **crossover design** with a **GST** in the primary endpoint to increase power and address heterogeneity

Collect data **weekly** via fit-for-purpose **PROs *and* PerfOs** focused on **fatigue** and **cognition**

Make study **more attractive** by incorporating at-home visits, travel reimbursements, and access to active drug

Initiate **more sites**, **build relationships** with sites, **reassess eligibility criteria** based on screening data, and wait

Tisento™ THERAPEUTICS

KOL= Key opinion leader; PRO= patient reported outcome; PerfO= Performance outcome, GST=Global statistical test

Challenges and Phase 2b PRIZM study approach

PRIZM
MELAS CLINICAL TRIAL

#1: Defining MELAS

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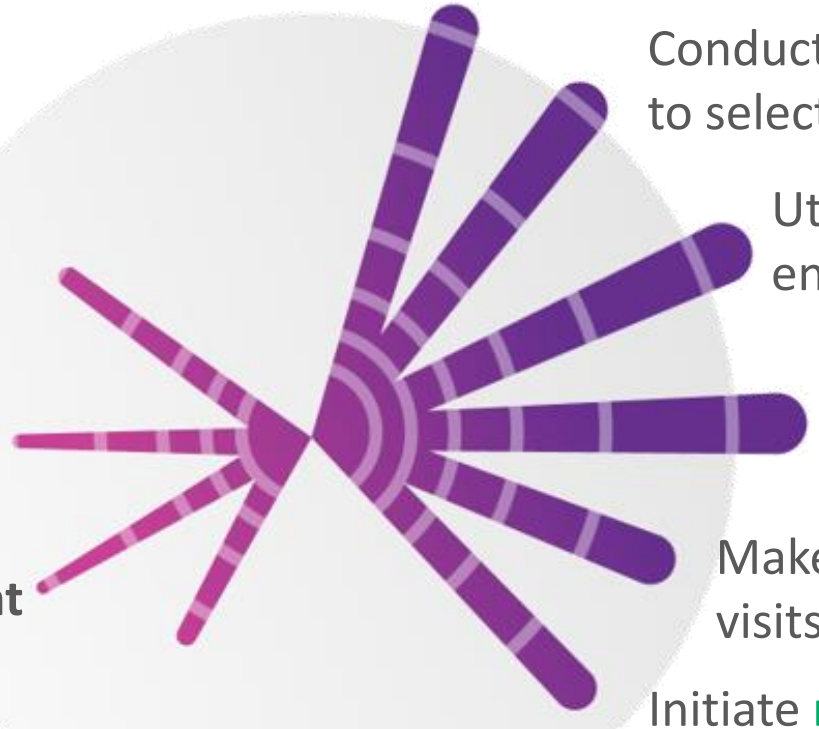

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Tisento™ THERAPEUTICS

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THERAPEUTICS

Using What We've Learned To Move Forward

REAGAN-UDALL FOUNDATION FOR THE FDA

Jason Colquitt
Across Healthcare

Amel Karaa, MD
Massachusetts General Hospital & Harvard Medical School

Kerry Jo Lee, MD
Center for Drug Evaluation and Research, FDA

Sophia Zilber
Lived Experience Perspective

Primary Mitochondrial Diseases: A Virtual Rare Disease Workshop

May 22, 2025 | 10am – 3:30pm *(eastern)*

Thank You for Joining Us!

Meeting materials will be posted on our website: www.reaganudall.org

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