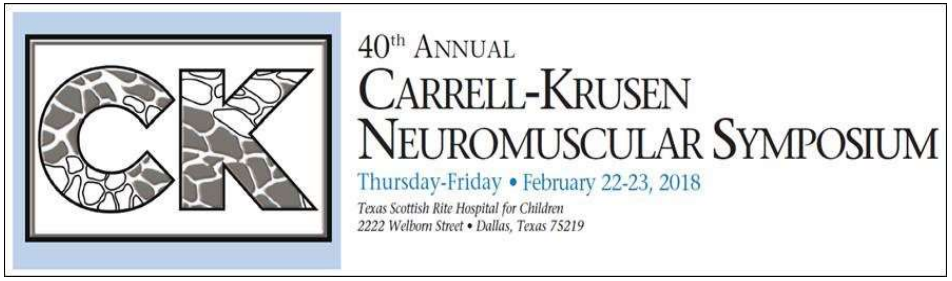


UCLA

A girl with developmental delay and a high CK

Mimi S. Lee, MD, Nishant Tiwari, MD, Emmanuelle Tiongson, MD,

Perry B. Shieh MD PhD.

Neuromuscular medicine fellow, UCLA

Department of Neurology, Department of Childrens' Hospital Los Angeles Neuropathology

Chief Complaint

- 4 yo Russia-born Armenian girl
  - learning disability
  - motor delay

Developmental stages

Born at full term via NSVD

@ 7-8 months, she sat up unsupported

@ 18 months, she began to walk

@ 3-4 yo, she started running

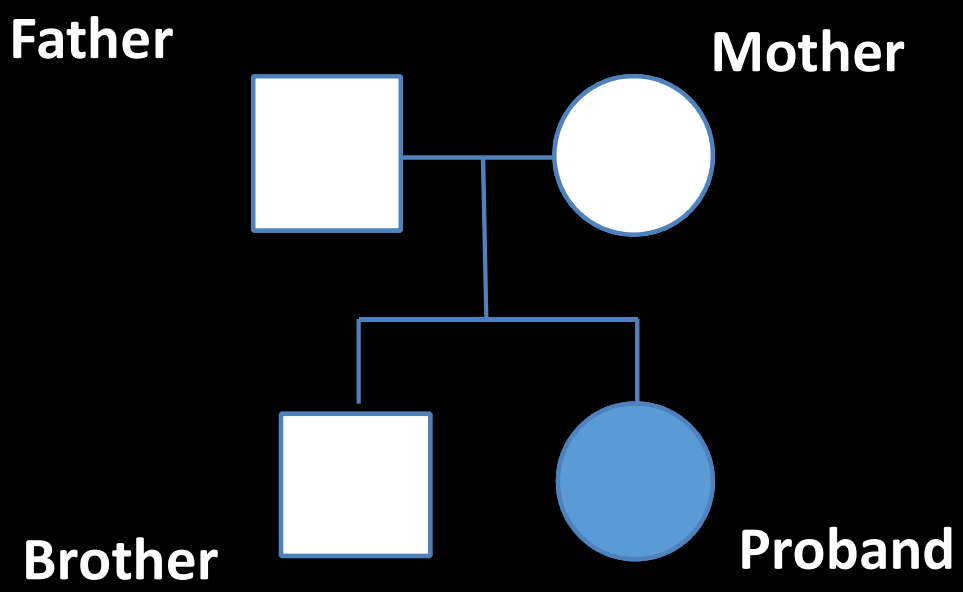
Review of Systems

- Pertinent positives:
  - learning disability
  - muscle weakness
  - hypotonia
  - frequent falls

Review of Systems

- Pertinent negatives:
  - diplopia
  - ptosis
  - vison loss
  - contracture
  - chest pain/palpitation
  - rashes or birthmarks

Family history



Previous work up in Moscow, Russia

- At age of 4
  - 3/2007: negative for Angelman Syndrome
  - 1/2007: negative examination for deletion in regions of chromosome 17, Smith-Magenis Syndrome
  - chromosome analysis: 46, XY, het 15
  - Serum examination: **NO** evidence of any hereditary aminoacidopathies, organic acidurias and defects of mitochondrial beta-oxidation

Previous work up

At age of 9

- 3/2011: Negative for deletion of exon 7 and 8 of the SMN gene

- 7/28/2011
  - CK: **2840**
  - Aldolase: **36.7**
  - Lactate/Pyruvate ratio: **19.27**

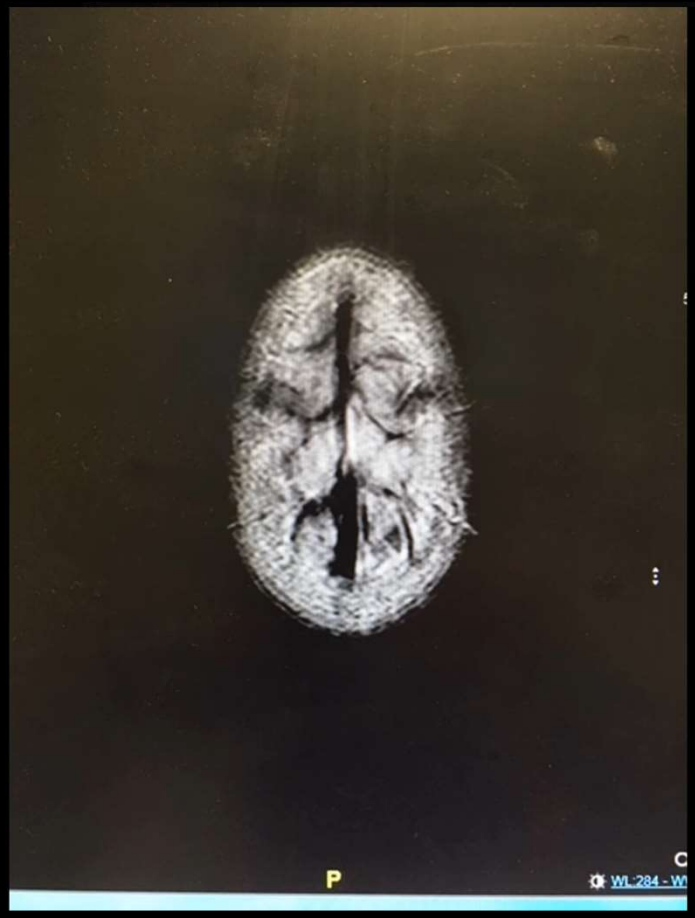
EEG at age of 9

- The main rhythm **does not meet the norm** for the given age group (N – 9Hz and higher).
  - Sleep is superficial
  - Marked diffuse changes of the cortex BEA.
  - Malfunction of the midline structures of the **basal-diencephalic level**
  - Complexes of sharp-slow wave type moderately above the background, diffuse (on the right and left), predominantly in the **left temporal-parietal** region are recorded.

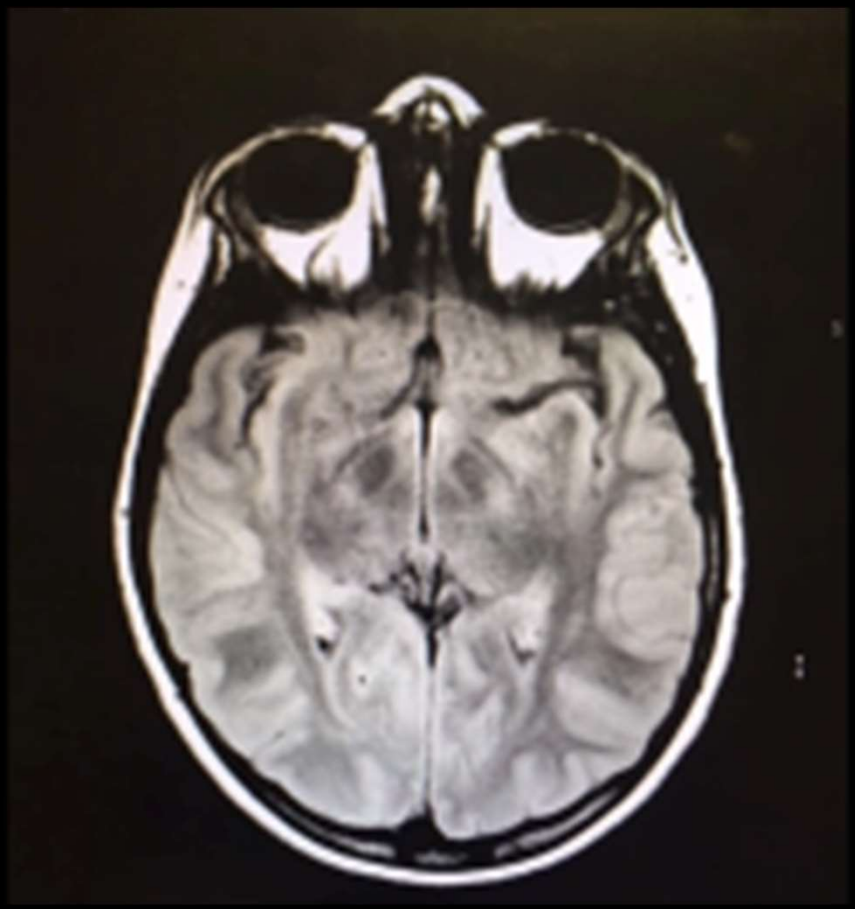

Children's
Hospital
LOS ANGELES SM
We Treat Kids Better

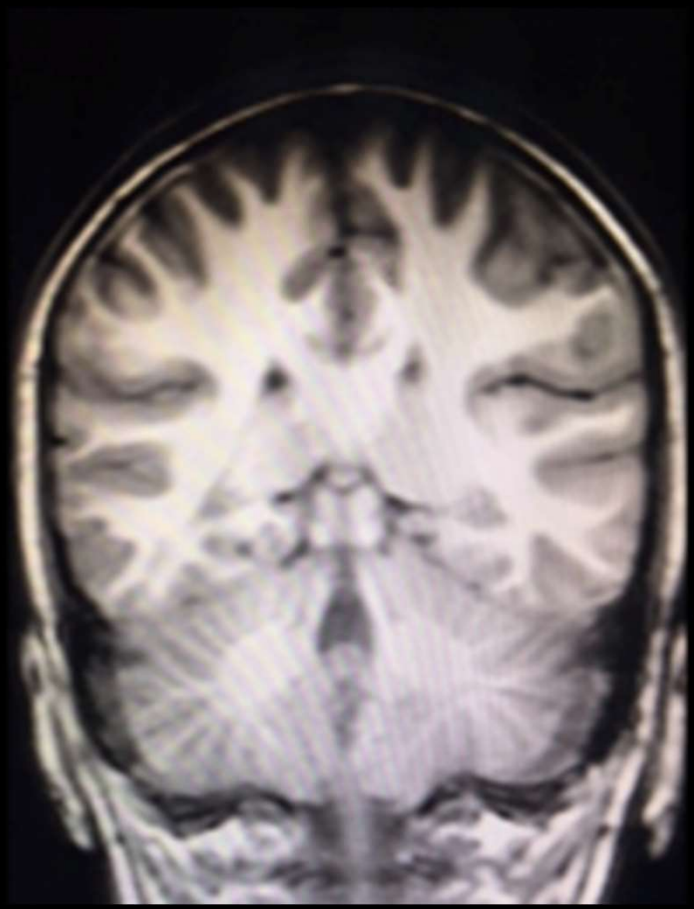
Imaging/Studies:

- 5/3/2011 MRI Pelvis/Femur: Nonspecific mild muscular atrophy
- 5/20/2011 Echo: Normal ventricular size and function
- 5/25/2011 MRI Brain: Normal non-contrast and contrast MRI of the brain
- 6/9/2011 Left quadriceps muscle biopsy: muscular dystrophy, type undetermined

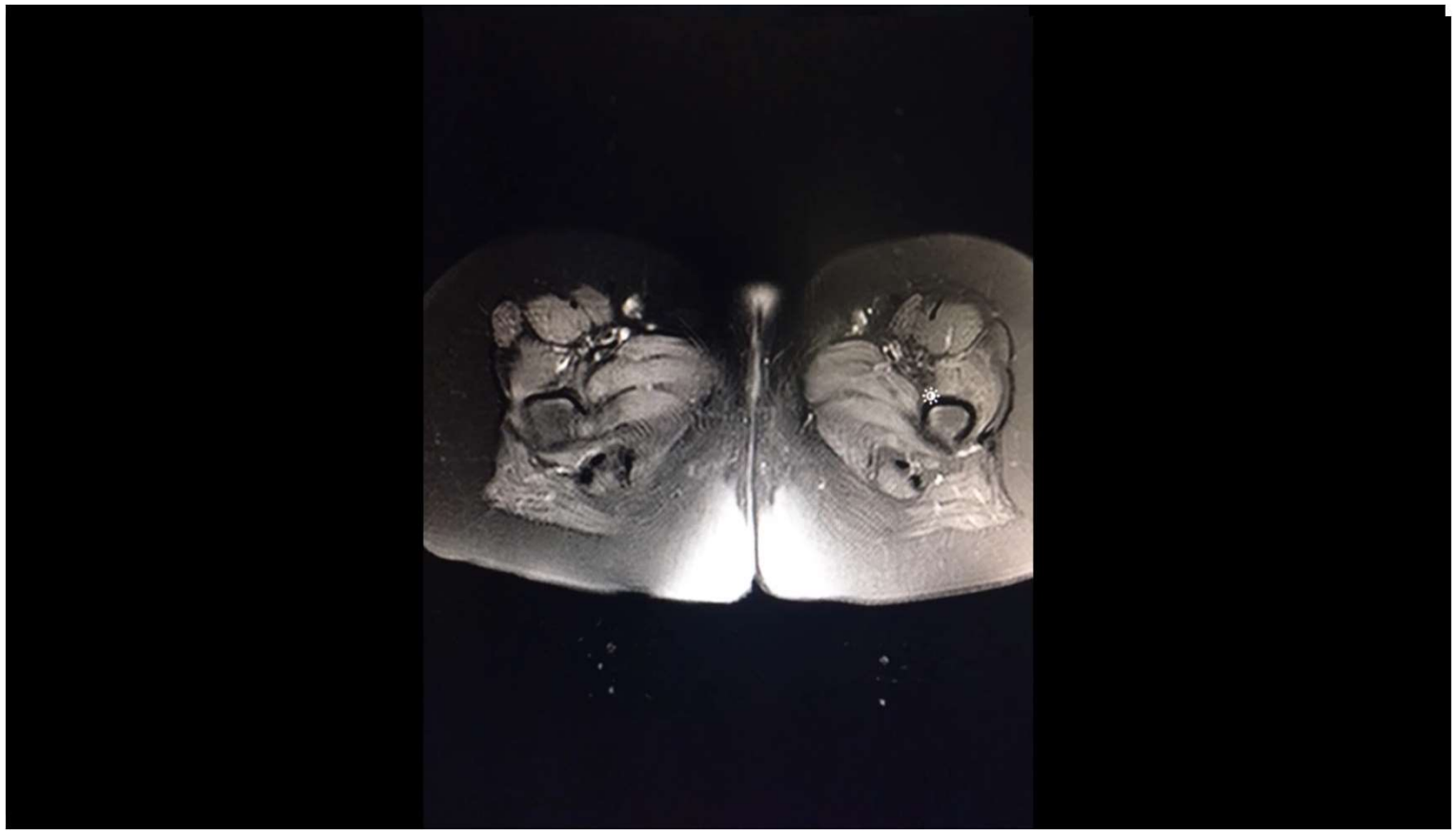

P
WL:284 - W

MRI brain @ 9 y.o.

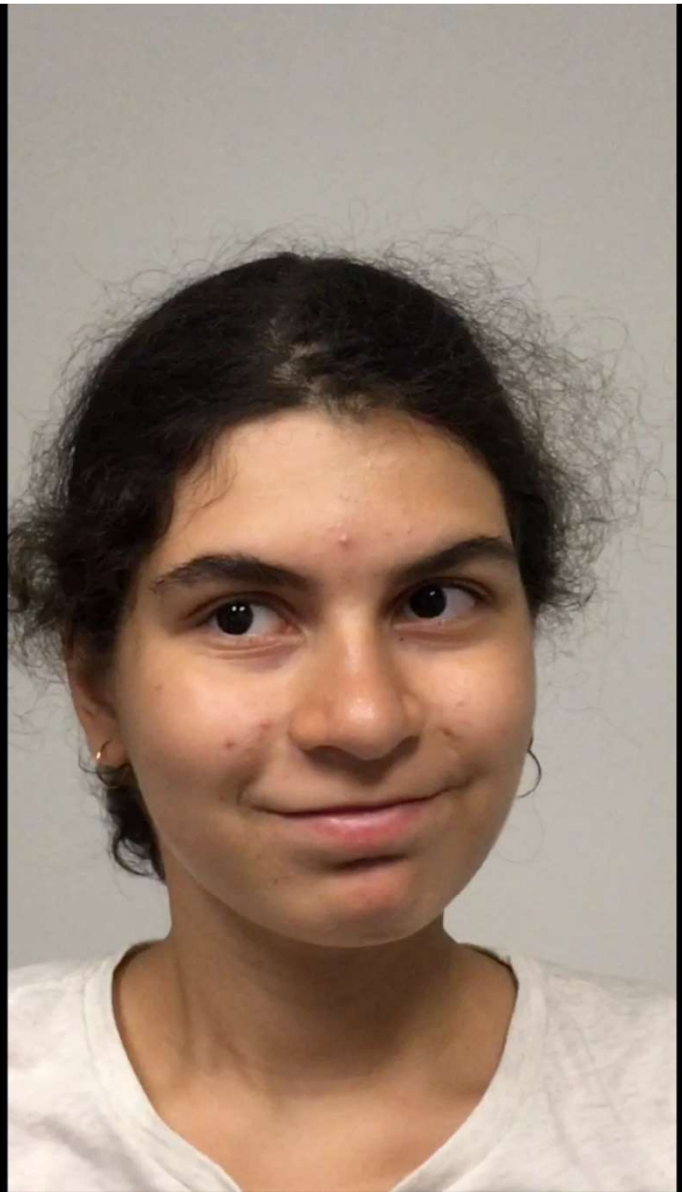




MRI of proximal muscles @ 9 y.o.



Referred to UCLA …



Physical Examination

- General: Well-appearing, alert, active child. Appears smaller than stated age
- HEENT: normal
- Lungs: CTAB
- Heart: RRR, 1+/6 systolic murmur
- Abdomen: NABS
- Extremities: No cyanosis, clubbing or edema
- Skin: No birthmarks or skin lesions

Neurological examination

- MS: AAO x 3
- Language/Speech:
  - Follows command
  - Only answers simple questions in Armenian
  - Avoid eye contacts
- Cranial nerves 2-12: normal

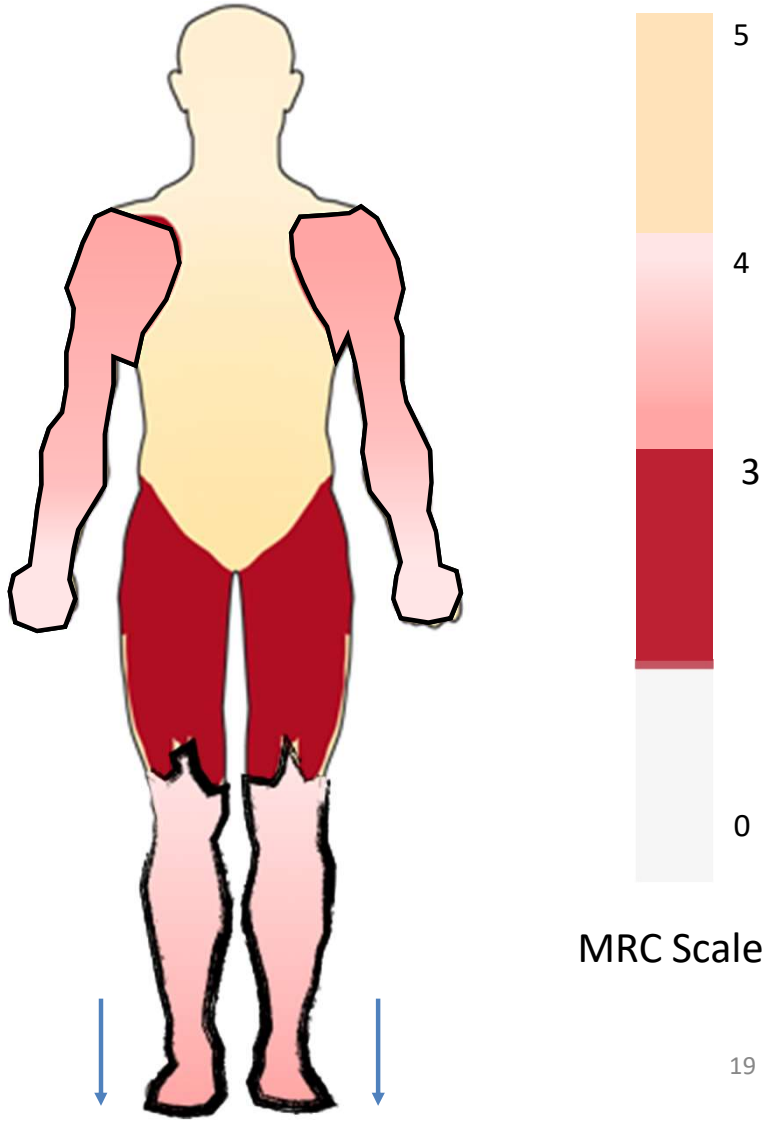
Motor: MRC 4 BUE and MRC 3 BLE .
No obvious calf hypertrophy.

Sensory: intact

Coordination: FNF intact

Reflexes: 1+ and symmetric at biceps, triceps, brachioradialis, patella; 2+ and symmetric at Achilles; toes down-going.

Gait: wide based with slight toe walking.
+ Gower's sign.



Summary

9 y.o. girl

- Developmental delay
- Short stature
- Lower extremity weakness
- HyperCKemia
- Abnormal MRI pelvis.

Impression

A Muscle and Brain condition

DDx:

- ? DMD/Becker
- ? Dystroglycanopathy (LGMD 2I)
- ? Myotonic dystrophy
- ? Others

EMG

EMG

Side	Muscle	Nerve	Root	Ins Act	Fibs/PSW	Fasc	Other	Amp	Dur	Poly	Recrt
Right	AntTibialis	Dp Br Peron	L4-5	Nml	None	None	None	Nml	Nml	Nml	Nml

Results:

Because of patient discomfort, testing was limited to EMG of one muscle, the right anterior tibialis, which was normal.

Impression:

This is a normal study. There is no electrodiagnostic evidence of muscle membrane instability in the right anterior tibialis muscle.

Laboratory studies

- CK **2840, 2343**
- Aldolase **36.7**
- Myotonic dystrophy 1 and 2 – negative
- Lactic acid 2.12
- Pyruvic acid 0.11

Impression

A disease of muscle and brain

DDx:

- ? DMD/Becker
- ? Dystroglycanopathy (LGMD 2I)
- ? Myotonic dystrophy
- **? Others**

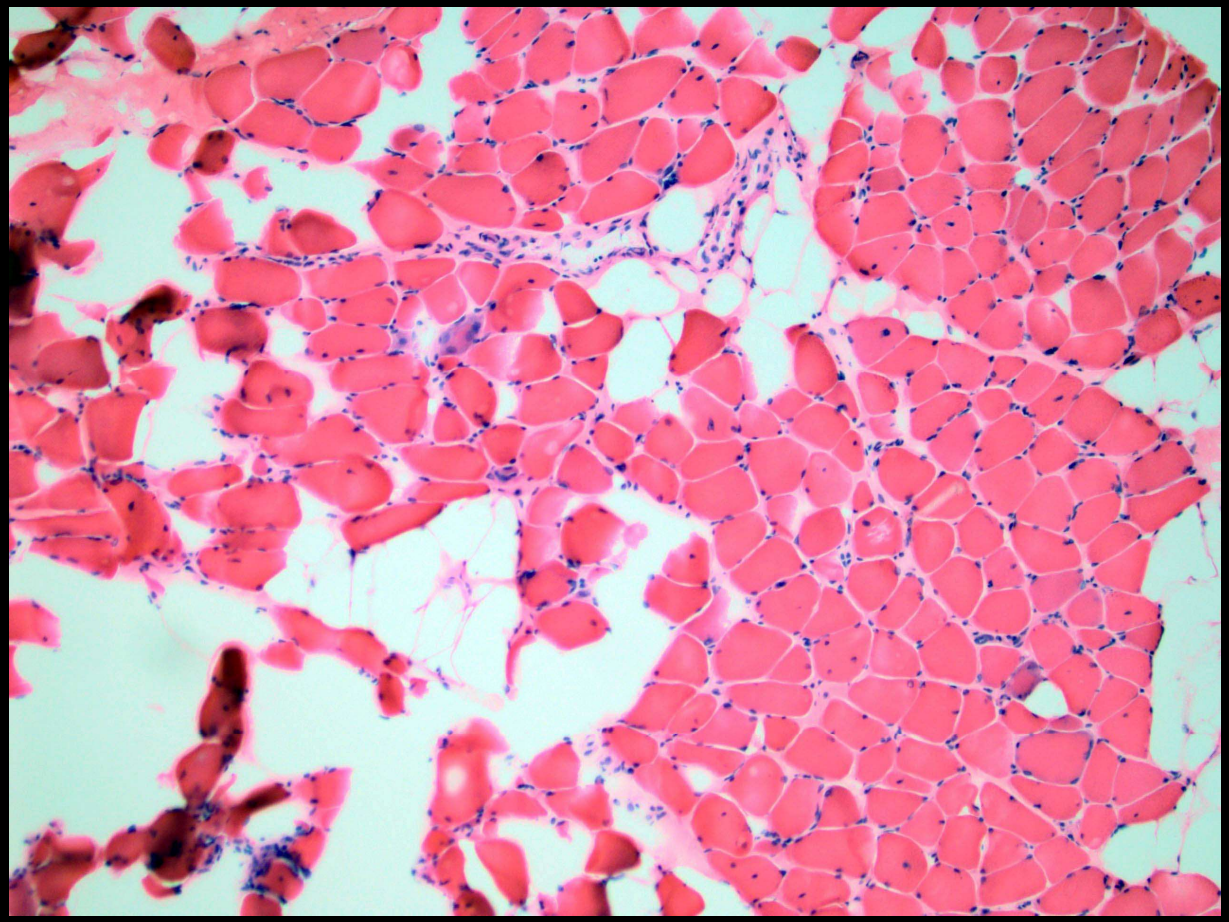
Targeted Genetic Testing

Variance of unknown significance

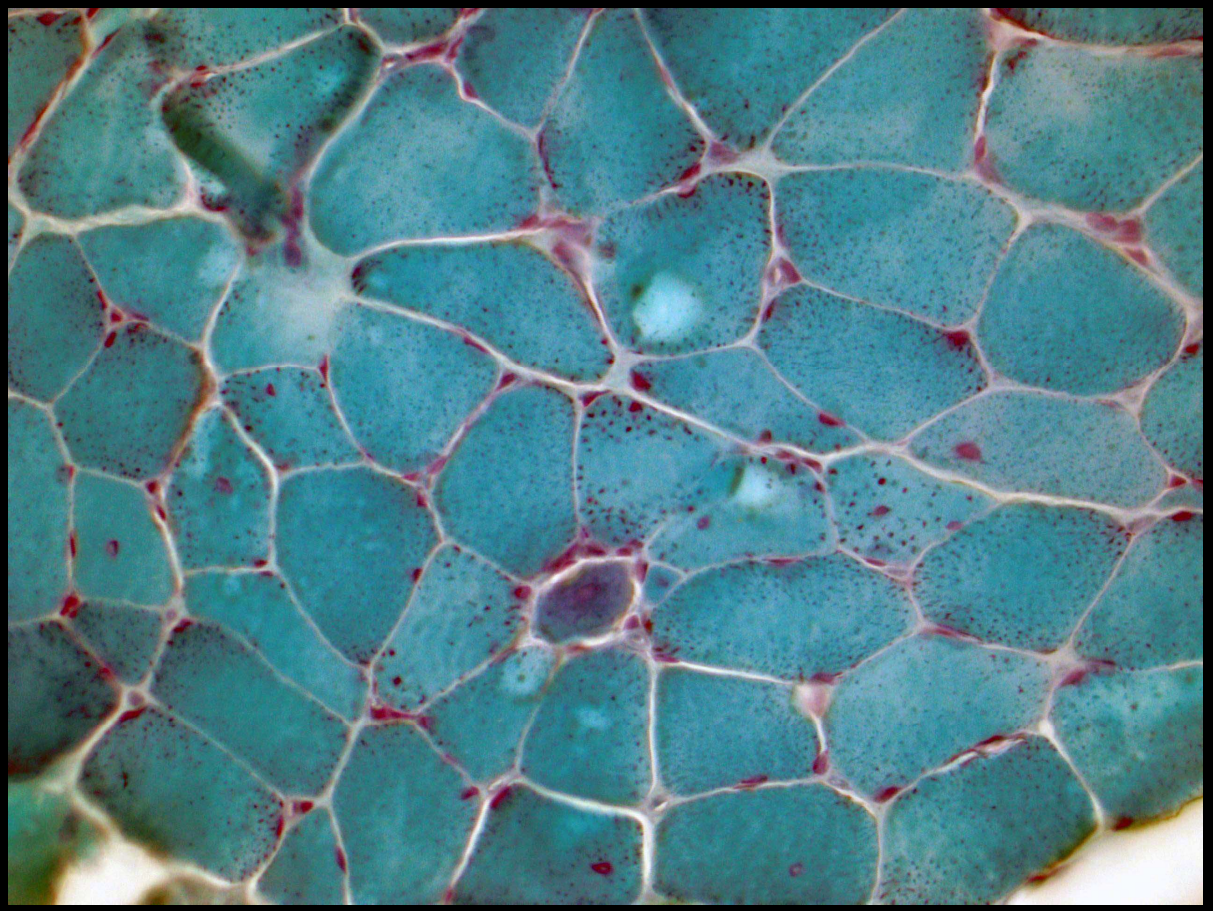
LMNA CCDS_1129.1 All Coding Exons (Exons 1-12) Sequenced

Exon	DNA Sequence Variation	Effect	Reference
10	c.1698 C>T, Heterozygous	p.His566His	rs4641

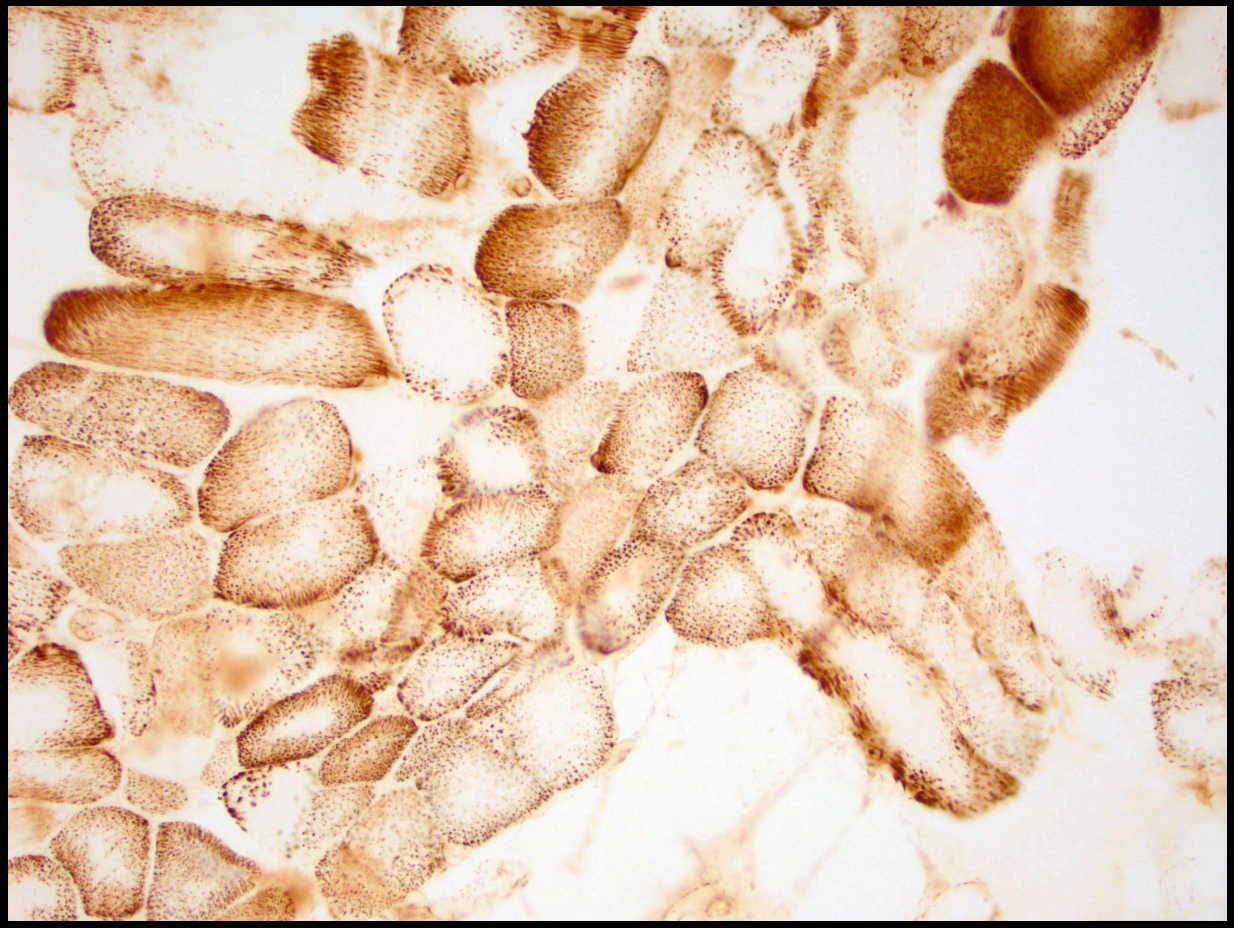
Muscle biopsy requested for review at UCLA



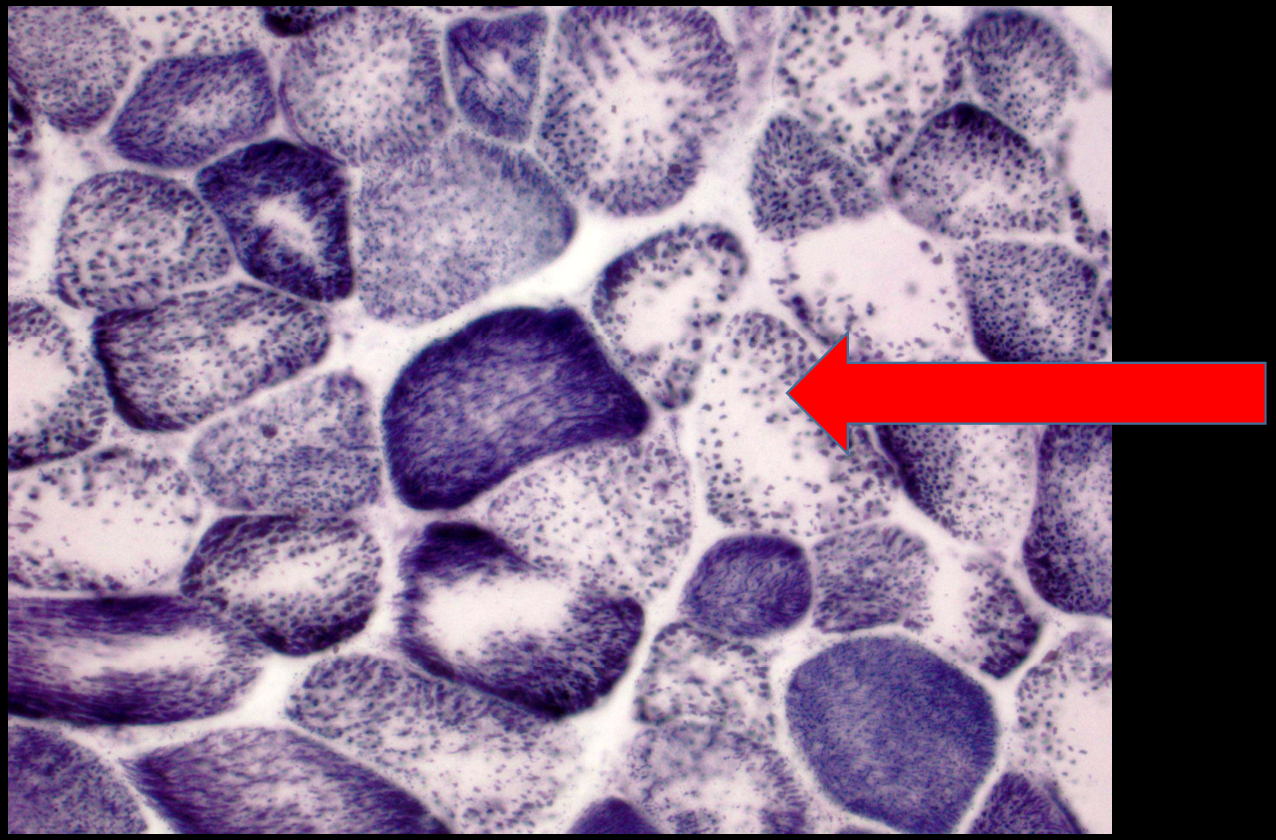
Trichrome



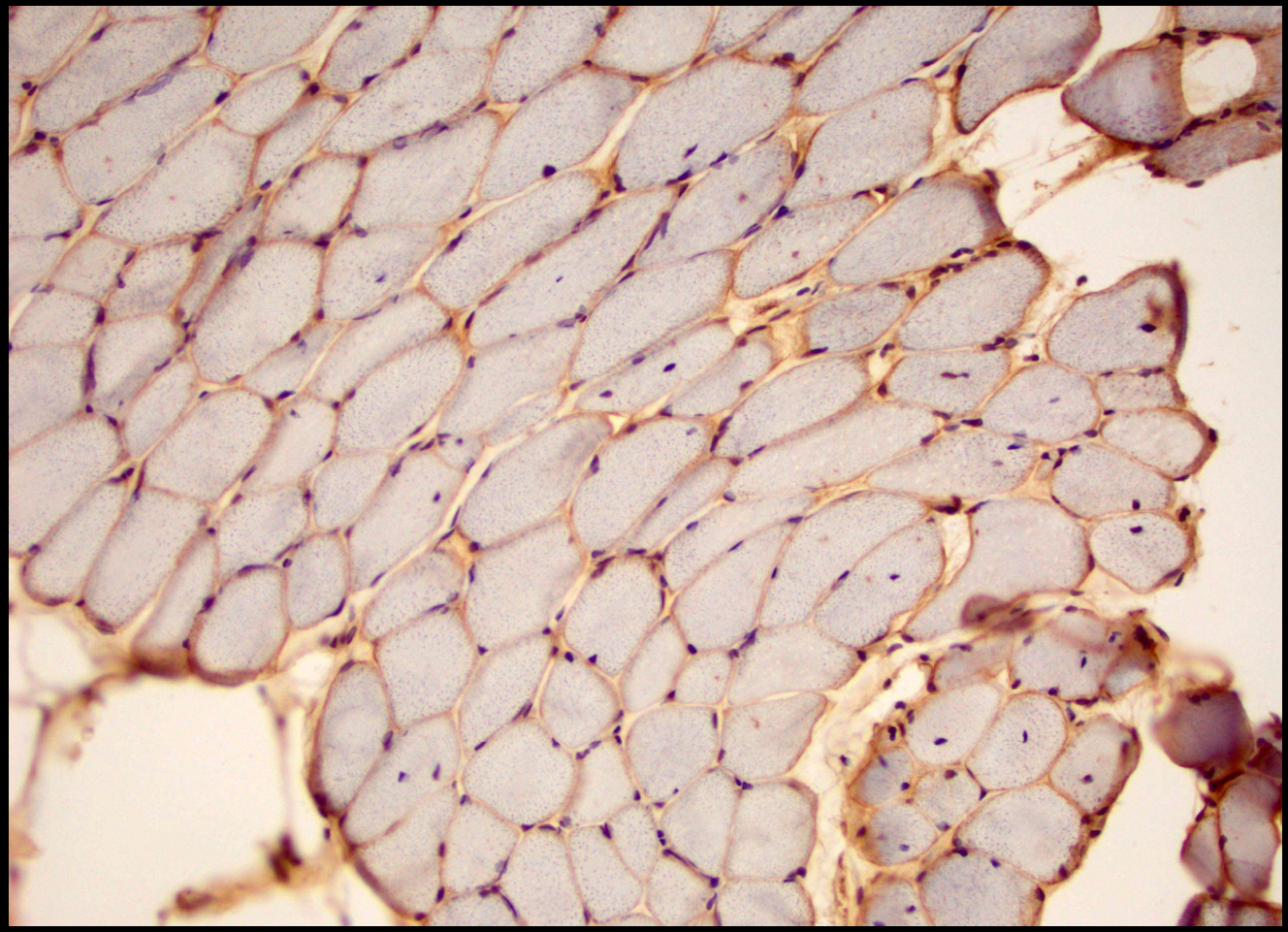
COX



NADH



Dystrophin staining



Selected immunohistochemistry

- Alpha Sarcoglycan Positive
- Beta Sarcoglycan Positive
- Gamma Sarcoglycan Positive
- Alpha Dystroglycan Positive

Differential Diagnosis

? DMD/Becker
? Dystroglycanopathy (LGMD 2I)
? Myotonic dystrophy
? Others

Others …

- Bethlem congenital muscular dystrophy
- congenital muscular dystrophy type 1A (MDC1A; merosin-deficient CMD)
- congenital muscular dystrophy type 1B (MDC1B)
- congenital muscular dystrophy type 1C (MDC1C)
- congenital muscular dystrophy type 1D (MDC1D)
- congenital muscular dystrophy with integrin deficiency
- Fukuyama congenital muscular dystrophy
- LMNA-related disorders
- muscle-eye-brain disease
- rigid spine muscular dystrophy (RSMD1)
- SEPN1-related disorders
- SYNE1-related disorder
- Ullrich congenital muscular dystrophy
- Walker-Warburg syndrome

And OTHERS …

Summary

9 y.o. girl with

- development delay
- proximal muscle weakness
- eccentrically located giant granules with central pallor
- mitochondrial involvement

Suspicious for Mitochondrial myopathy

To confirm our suspicion

- Requested EM which was done in Children's hospital
- Sent out Whole exome sequencing


EMS11-101-10
1 mm

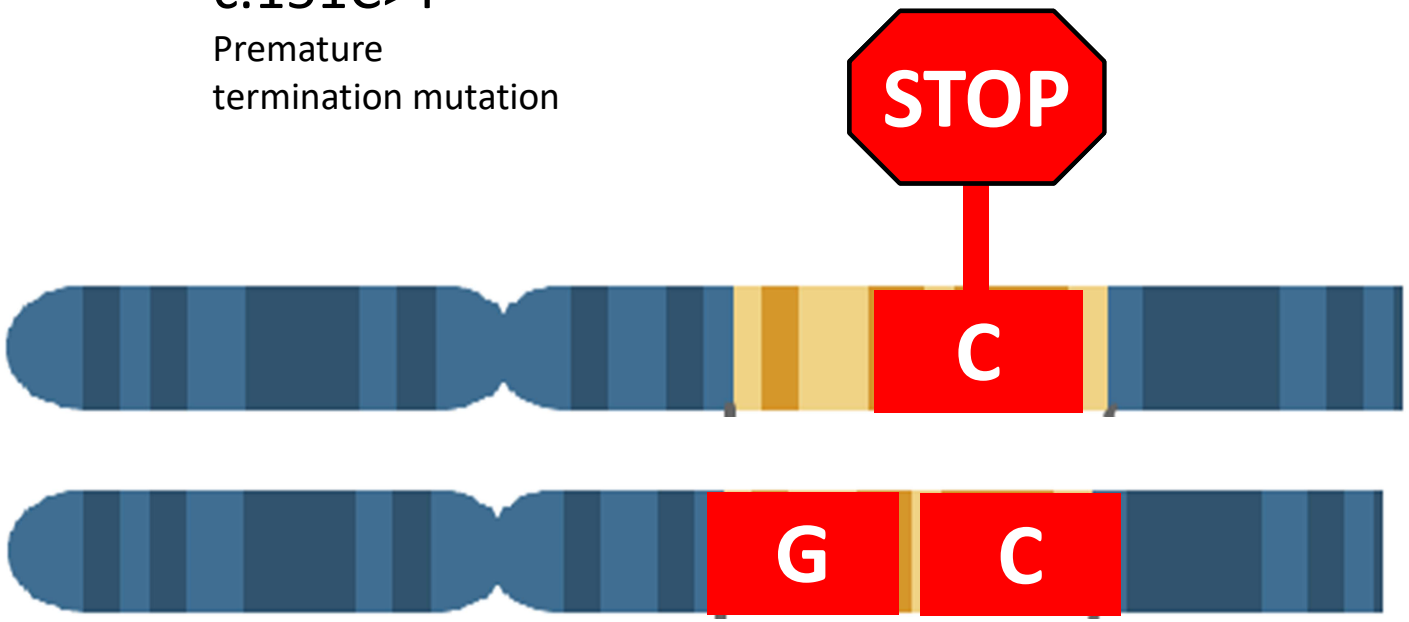
Whole Exome Sequencing

- Three heterozygous variants were identified in CHKB gene
  - c.151C>T
  - c.847G>A
  - c.902C>T

Choline Kinase Beta (CHKB)

c.151C>T

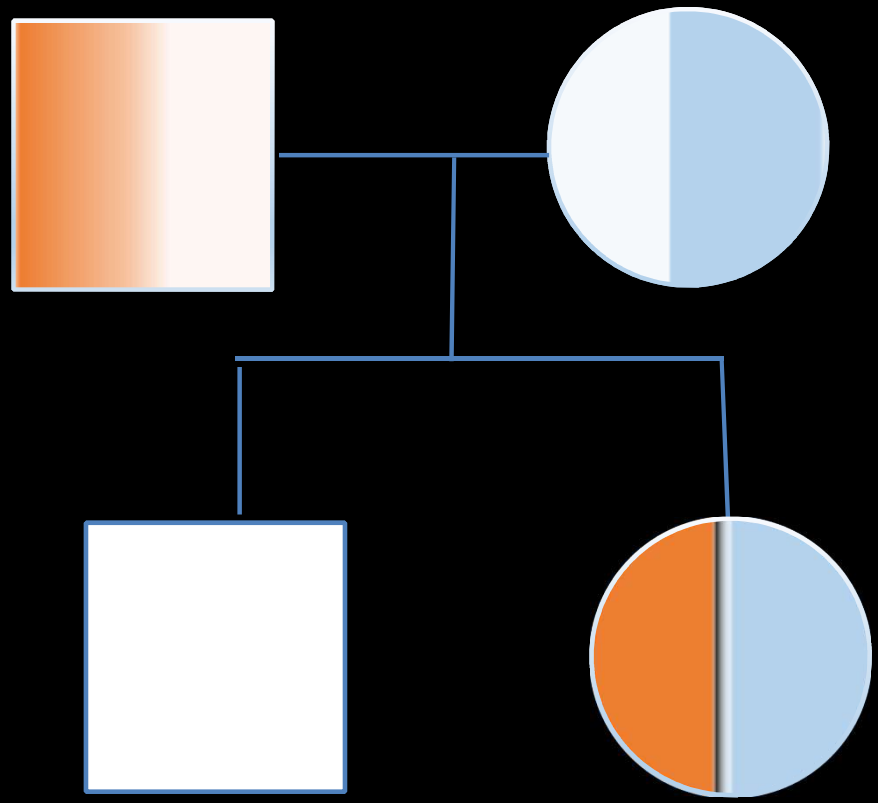
Premature
termination mutation



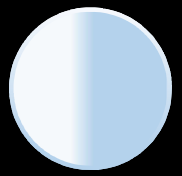
c.847G>A (AR)

c.902C>T ? pathogenic

Trio Next Generation Sequencing



Father c.847G>A



Mother c.151C>T

Choline Kinase Beta Muscular Dystrophy

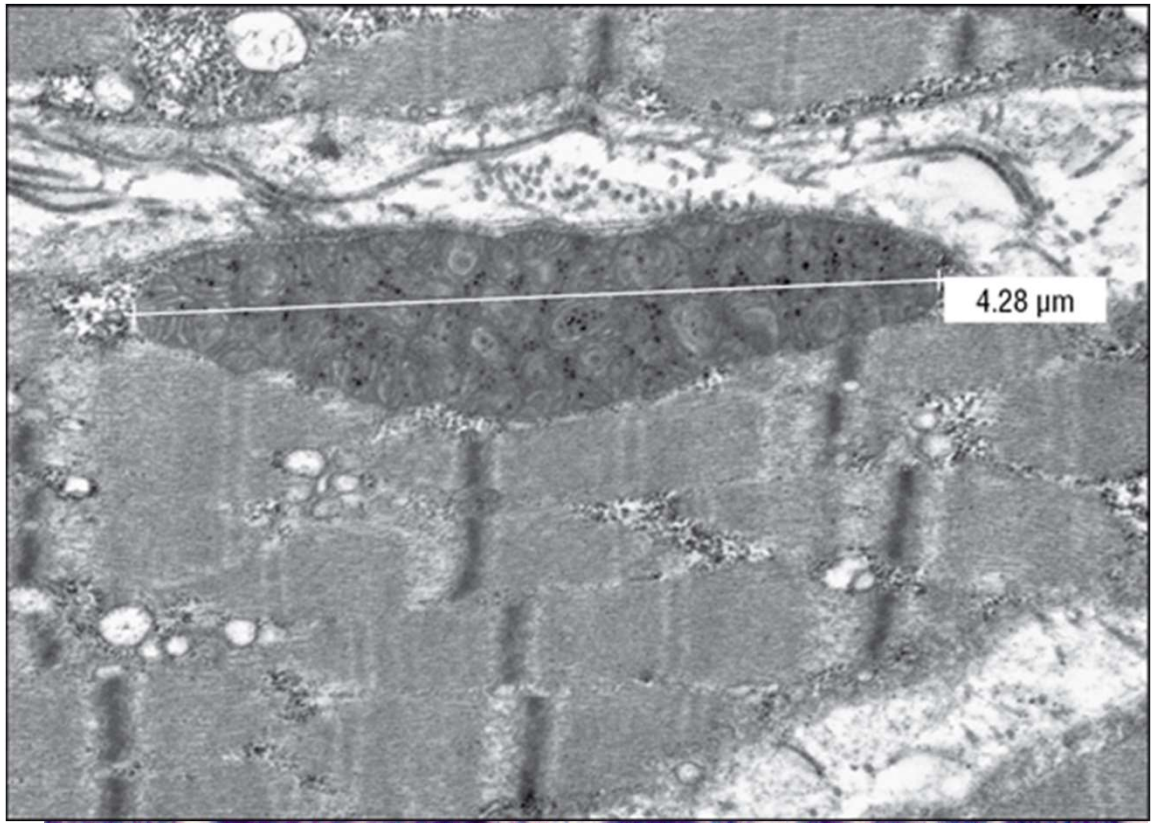
- First described In 1964 and 1966
  - Shy and Gonatas and Shy et al described children with myopathy and giant mitochondria and dubbed this condition "**megaconial myopathy**."
- AR, loss of function mutation
- Found on Cx 22
- CK elevated

Choline Kinase Beta Muscular Dystrophy

- Most characteristic phenotypic features
  - **intellectually delay (delay of speech and language and autism spectrum disorder)**
  - **progressive proximal muscle weakness**,
  - **approximately 50% of reported patients have cardiac involvement**

Unique **Histological** Features

- Gigantic mitochondria
  - **Megaconial**
- Peripheral placement
- Central devoid of organelles



Genetic confirmation

Nishino et al group published first genetic mutation responsible for this unique megaconial muscular dystrophy

- The similarities of these clinical and morphological features with those of a spontaneous mutant mouse harboring a loss-of-function mutation in the choline kinase beta gene (*Chkb*)

Choline kinase beta Muscular Dystrophy

- They found deleterious mutations in all patients and defined the molecular basis of this congenital megaconial muscular dystrophy

- Stereotypic phenotypes
  - Language
  - ? Autism
  - proximal muscle weakness

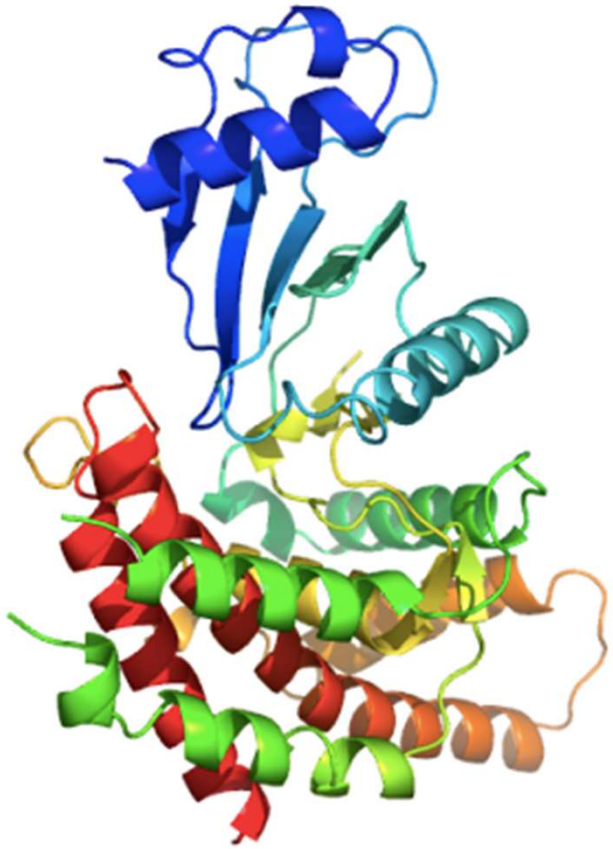
Table 1 Characteristics of the patients with CHKB gene mutations

Patients	1	2	3	4	5	6	7	8
Age at presentation	4 years 1 month	14 months	6 years 2 months	38 days	12 months	17 years	3 years	4 years
Gender	M	M	M	F	M	F	F	F
Current age	12 years 6 months	Died at age 2 years 1 month	Died at age 7 years 4 months	8 years 2 months	18 years 10 months	23 years 8 months	23 years 3 months	9 years 2 months
Presenting symptom	Ichthyosis, ID (Birth)	Motor developmental delay (14 months)	Proximal weakness ID Hyperactivity Ichthyosis	Foot deformity, intrauterine cardiac defect (PDA) Microcephaly (3 months)	ID, proximal weakness	Developmental delay, hyperactivity, delay in language, hand biting (2 years)	Delay in walking	Delay in language and walking
Age at independent walking	2 years 1 month	Not able to walk	3 years	Not able to walk at age 21 months	24 months	3 years	3 years	Not able to walk at age 6 years 6 months
Language skills	No words at age 4 years 6 months	Single words at age 15 months, no increase in number of words by 21 months of age	Two-word sentences at the age of 5 years	No words at age 14 months	Single words: 3–4 years, mild ID at age 12 years	Single words: 3 years	Single words: 5 years	Simple 2-word sentences: 6 years 6 months
Consanguinity/family history	2nd degree/N	1st degree/ Y	1st degree/ Y	1st degree/ N	1st degree/ N	1st degree/ N	1st degree/N	N/N
Seizures	N	N	N	N	N	Y (3.5 years, generalized tonic seizure)	Y (generalized tonic seizures with an onset at 9 years of age)	N
Autistic features/ behavioral problems	Severe, aggressive, anxious behaviors, stereotypical hand movements, head banging, self mutilation	N	Hyperactivity	N	N	Severe behavioral problems, stereotypical hand biting, short eye contact	Stereotypical head movements, echolalia, autistic features, aggressive behavior, crying attacks, sleep disorder	N
Skin findings/age at recognition	Ichthyosis, exfoliative desquamative diffuse lesions, Skin biopsy: ichthyosis congenita/Birth	N	Ichthyosis/4 years	Hirsutism (face and genital region)/ 8 months	N	Diffuse ichthyosis (forehead, face, body)/ 17 years	Ichthyosis (neck and trunk desquamation)/ 11 years	Ichthyosis (at birth, became prominent at age 12 months, recognized at the age of 4 years), diffuse, around umbilical region desquamation and xserosis
Dysmorphic features	Atypical face, prominent nasal bridge and ears, distal laxity	N	N	High arched palate, facial hypomimia, distal laxity	N	N	N	Hypertelorism, low set ears, long face
ID	Severe ID	NA	Moderate ID	NA	Mild ID	Severe ID	Severe ID	Moderate ID
Serum CK level (U/L) Normal≤300 U/L	3X	Normal	Normal	Mildly elevated	3X	9X	N-3X	Mildly elevated
Cardiac evaluation	ECHO normal (6 years), decreased left	ECHO: dilated CMP (21 months)	ECHO normal (6 years 2 months)	Fetal and neonatal ECHO: PDA	ECHO: normal (14 years)	ECHO: normal (17 years)	ECHO: Secundum ASD (3 years, 17 years)	ECHO: normal (7 years)

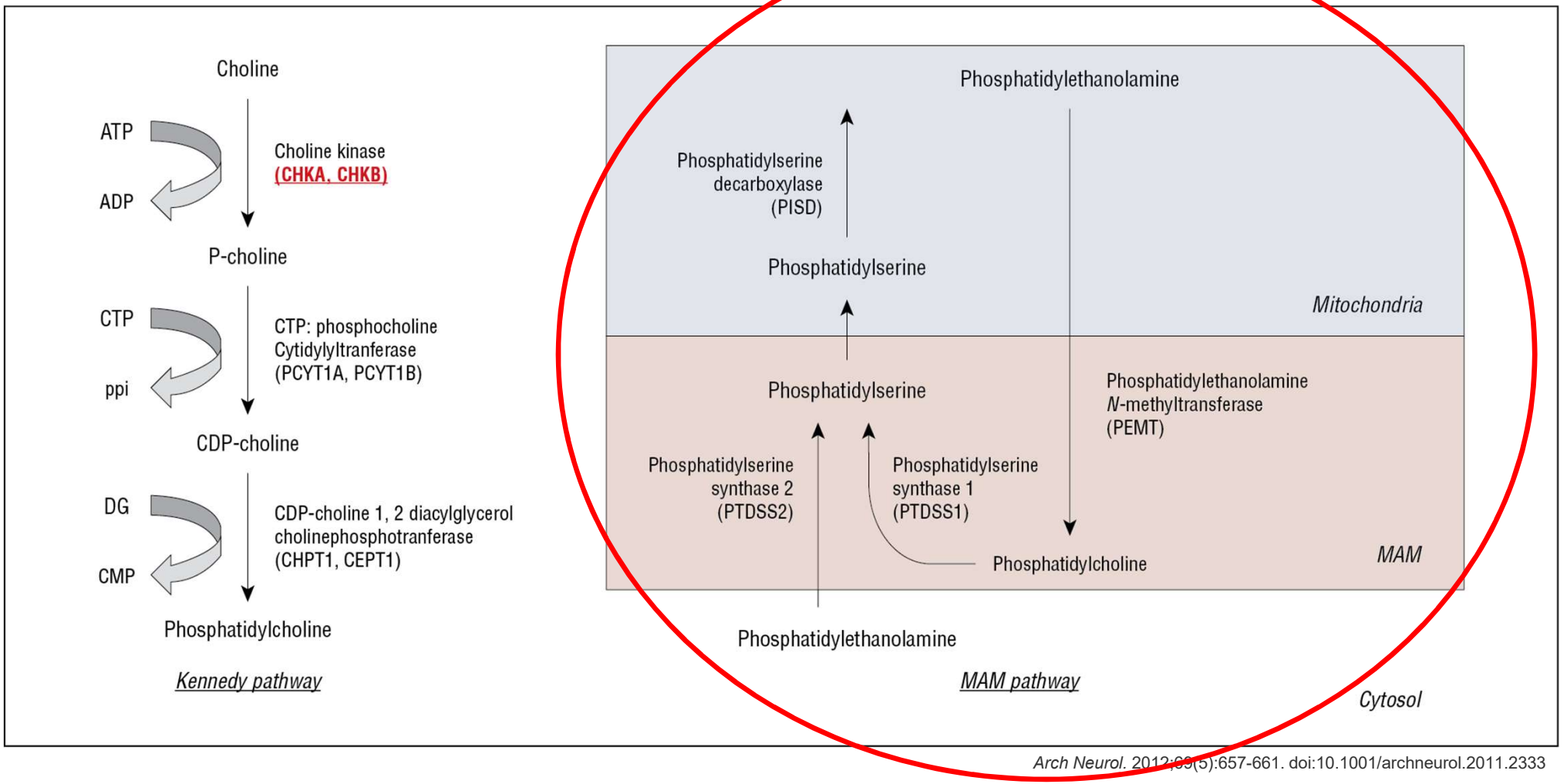
Background

- Choline kinase (CK) was discovered in 1953.

- Two genes encode choline kinase, *Chka* and *Chkb*, and 3 isoforms of the enzyme have been identified — CKα-1, CKα-2, and CKβ — and the active form of CK is a hetero- or homo-dimer.



Phospholipid Metabolism Pathways



Arch Neurol. 2012;69(5):657-661. doi:10.1001/archneurol.2011.2333

Choline kinase beta function

- Defect in choline kinase beta activity in muscle, and different distribution of choline kinase isoforms in muscle and brain describe varying degrees of phenotype in the mouse model and patients (Wu et al 2010; Mitsuhashi et al 2011a, b)

- In addition, mitochondrial dysfunction in muscle is described both in mutant mice and patients with *CHKB* mutations (Mitsuhashi et al 2011a, b; Gutierrez Rios et al 2012; Castro-Gago et al 2014)

Proposed pathogenesis

- The relationship between phospholipid abnormality and mitochondrial dysfunction could be explained by
  - Upregulation of a second alternative pathway

    - MAM dysfunction leading to altered mitochondrial dynamics, resulting in increased size and intracellular displacement of mitochondria

Proposed pathogensis

- Defect in choline kinase beta activity in muscle, and different distribution of choline kinase isoforms in **muscle** and **brain** describe varying degrees of phenotype in the mouse model and patients

- In addition, mitochondrial dysfunction in muscle is described both in mutant mice and patients with *CHKB* mutations

Alternative pathogenesis

CHKB, which is located on chromosome 22q13.33, is immediately upstream of *CPT1B* (carnitine palmitoyltransferase 1B)

- a key lipid transport enzyme located in the mitochondrial outer membrane
- Downstream effect
    - mitochondria resulting from alterations in overall fatty acid metabolism due to the effects on *CPT1B*, ultimately affecting mitochondrial respiratory chain activity.

Conclusion

- Muscle and brain disease
- Role of choline kinase beta gene is important for both brain and muscle development
- Two possible pathogenesis has been proposed including MAM pathway and CPT1B (down stream effect)

References

- *Gengshu Wu, Dennis E. Vance.* Choline kinase and its functionThis paper is one of a selection of papers published in this special issue entitled “Second International Symposium on Recent Advances in Basic, Clinical, and Social Medicine” and has undergone the Journal's usual peer review process.
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- Mitsuhashi S1, Hatakeyama H, Karahashi M, Koumura T, Nonaka I, Hayashi YK, Noguchi S, Sher RB, Nakagawa Y, Manfredi G, Goto Y, Cox GA, Nishino I.Muscle choline kinase beta defect causes mitochondrial dysfunction and increased mitophagy. Hum Mol Genet. 2011 Oct 1;20(19):3841-51. doi: 10.1093/hmg/ddr305. Epub 2011 Jul 12.
- Mitsuhashi S, Hatakeyama H, Karahashi M, Koumura T, Nonaka I, Hayashi YK, Noguchi S, Sher RB, Nakagawa Y, Manfredi G, Goto Y, Cox GA, Nishino I. Muscle choline kinase beta defect causes mitochondrial dysfunction and increased mitophagy. Hum Mol Genet. 2011 Oct 1;20(19):3841-51. doi: 10.1093/hmg/ddr305. Epub 2011 Jul 12.

Proposed mechanism

- It has been shown that this hindlimb muscular dystrophy is due to decreased biosynthesis of phosphatidylcholine and increased catabolism of phosphatidylcholine in the hindlimbs, but not the forelimbs, of mice

- In the mid 1950s it was clear that choline kinase was important for the biosynthesis of phosphatidylcholine, but no one predicted a role for choline kinase in muscular dystrophy, bone deformities, or cancer.

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Seizures	N	N	N	N	N	Y (3.5 years, generalized tonic seizure)	Y (generalized tonic seizures with an onset at 9 years of age)	N
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