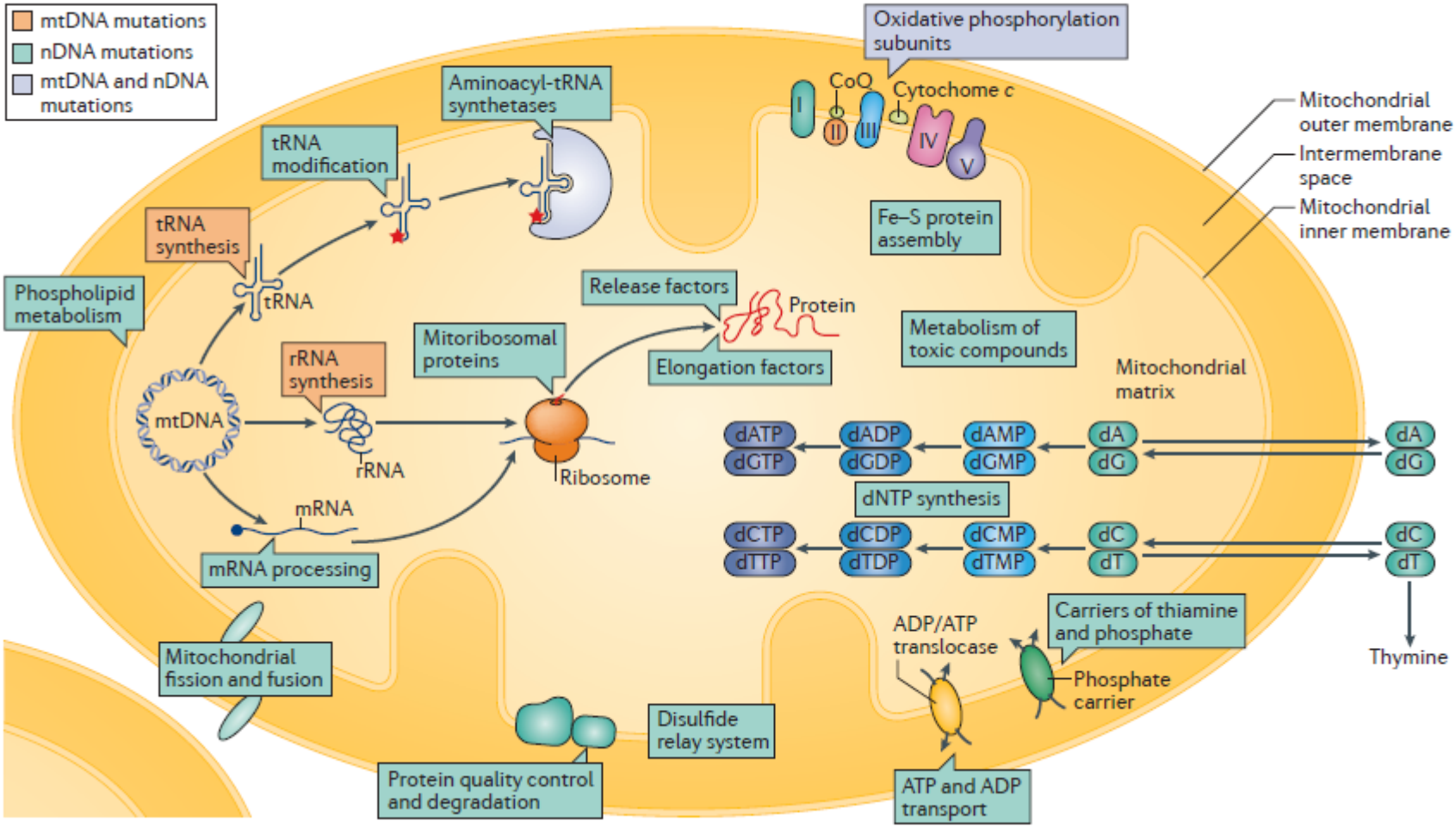
Malattie Mitocondriali

Fenotipi clinici

Michelangelo Mancuso, MD, PhD
University of Pisa

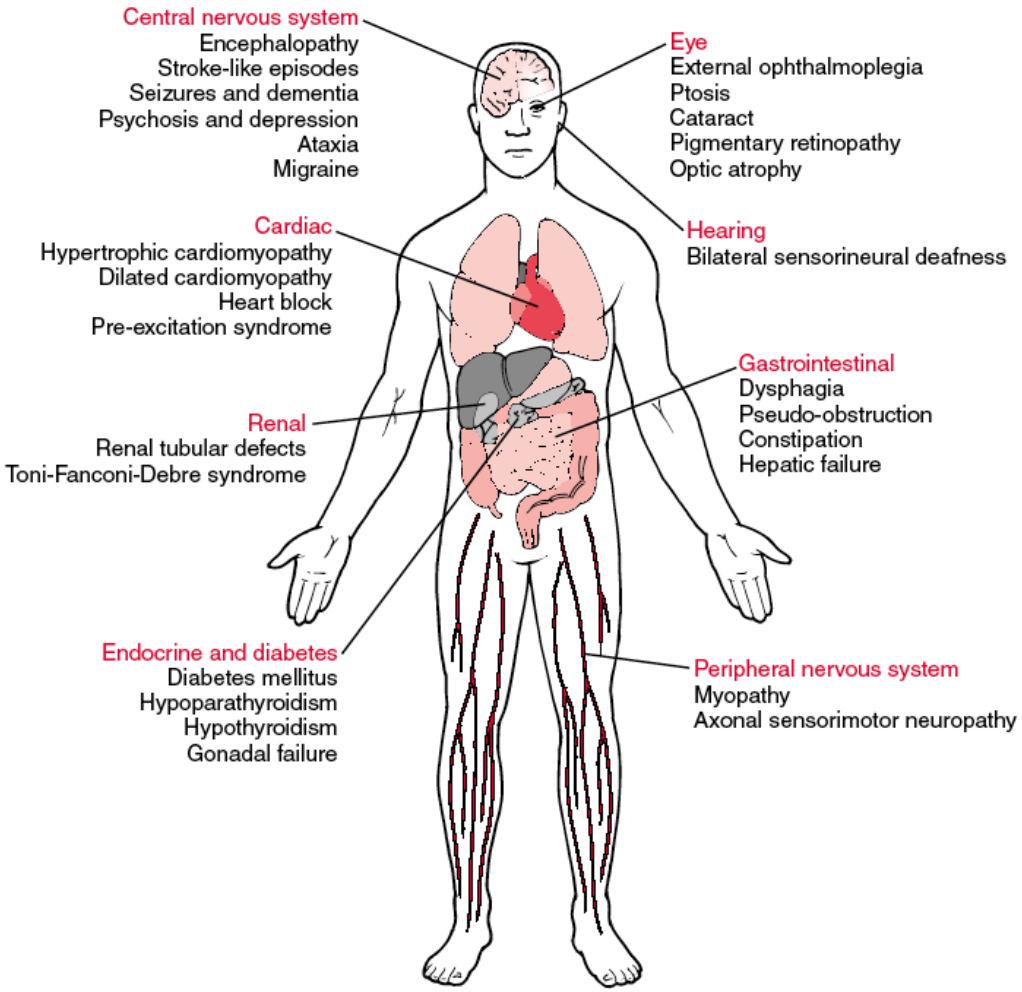

mtDNA mutations
nDNA mutations
mtDNA and nDNA mutations
Oxidative phosphorylation subunits
CoQ
Cytochome c
I
II
III
IV
V
Aminoacyl-tRNA synthetases
tRNA modification
tRNA synthesis
tRNA
Phospholipid metabolism
Release factors
Protein
Elongation factors
Mitoribosomal proteins
rRNA synthesis
mtDNA
rRNA
Ribosome
mRNA
mRNA processing
Fe–S protein assembly
Metabolism of toxic compounds
Mitochondrial matrix
Mitochondrial outer membrane
Intermembrane space
Mitochondrial inner membrane
dATP
dGTP
dADP
dGDP
dAMP
dGMP
dA
dG
dNTP synthesis
dCTP
dTTP
dCDP
dTDP
dCMP
dTMP
dC
dT
Thymine
ADP/ATP translocase
Carriers of thiamine and phosphate
Phosphate carrier
Mitochondrial fission and fusion
Disulfide relay system
Protein quality control and degradation
ATP and ADP transport

GENETIC CLASSIFICATION

- **Defects of mtDNA**
  - Mutations in protein synthesis genes
    - tRNA, rRNA, rearrangements
  - Mutations in protein-coding genes
    - Multisystemic (LHON, NARP/MILS)
    - Tissue-specific

- **Defects of nDNA**
  - Mutations in respiratory chain subunits
    - Complex I, Complex II
  - Mutations in ancillary proteins
    - Complex IV, Complex III
  - Defects of intergenomic signaling
    - AR-PEO with multiple Δ-mtDNA
    - mtDNA depletion
  - Defects of the lipid milieu
    - Barth syndrome
  - Defects of motility/fusion/fission
  - Defects of tRNA-synthetases

MITOCHONDRIAL DISORDERS



PRIMARY MITOCHONDRIAL MYOPATHIES

genetically defined disorders leading to defects of oxidative phosphorylation affecting predominantly, but not exclusively, skeletal muscle (see below for methodology). Secondary involvement of mitochondria, frequently observed in multiple neuromuscular diseases (i.e. inclusion body myositis, Duchenne muscular dystrophy, Kennedy disease) are not considered PMM

Workshop report

International Workshop:
Outcome measures and clinical trial readiness in primary mitochondrial myopathies in children and adults. Consensus recommendations.
Rome, Italy, 16–18 November 2016

Michelangelo Mancuso [a,*], Robert McFarland [b], Thomas Klopstock [c], Michio Hirano [d] on behalf of the consortium on Trial Readiness in Mitochondrial Myopathies [1]

[a] Department of Experimental and Clinical Medicine, Neurological Institute, University of Pisa, Italy
[b] Wellcome Trust Centre for Mitochondrial Research, Institute of Genetic Medicine, Department of Physiology and Functional Genomics NE1 3BZ, Newcastle University, Newcastle upon Tyne, UK
[c] Friedrich-Baur-Institut an der Neurologischen Klinik und Poliklinik, LMU München, Ziemssenstr. 1a, 80336 München, Federal Republic of Germany
[d] Department of Neurology, H. Houston Merritt Neuromuscular Research Center, Columbia University Medical Center, New York, NY, USA

Received 26 June 2017

PMM: clinical presentation

- Fatigue (defined as an overwhelming sense of tiredness, lack of energy and feeling of exhaustion)
- Exercise Intolerance
- Pain/Myalgia
- Weakness
- Wasting
- Dysphagia
- Spasms
- Myoglobinuria, triggered by exercise (cyt b or CoQ10 deficiency)
- Ptosis
- ophtalmoparesis

Italian Network of Mitochondrial Diseases

1800 Patients

	Patients (1330 tot with full phenotype described)	Percentage
Ptosis/ophthalmoparesis	636	47,8
Muscle weakness	488	36,7
Hearing loss	330	24,8
Exercise intolerance	279	21
Optic neuropathy	241	18,1
Muscle wasting	233	17,5
Cerebellar ataxia	198	14,8
Cognitive involvement	189	14,2
Hypotonia	180	13,5
Neuropathy	163	12,2
Swallowing impairment	162	12,1
Epileptic seizures	152	11,4
Muscle pain	152	11,4
Diabetes	121	9
Pyramidal involvement	115	8,6
Respiratory impairment	115	8,6
Cardiomyopathy	106	7,9
Migraine	102	7,6
Retinopathy	92	6,9
Gastroint. dysmotility	69	5,1

PEO

- The commonest phenotype of PMM, observed in about two-thirds of all cases of PMM
- Bilateral eyelid ptosis, often the presenting symptom, associated with a compensatory frontalis muscle contraction and, in severe cases, tilting of the head.
- Ptosis is accompanied by a slowly progressive, usually symmetrical limitation of eye movement (ophthalmoplegia) in all directions of gaze
- Diplopia is sometimes reported by the patients. Intrinsic ocular muscles are not involved.

PEO- II

- PEO is often associated with other signs of skeletal muscle involvement, typically slow progressive axial and proximal limb weakness affecting predominantly the hip and shoulder girdle muscles often with muscle wasting.
- Muscle weakness may also cause difficulty swallowing (dysphagia) and respiratory failure.
- Distal muscle weakness may be present but rarely seen early in the disease.
- From a genetic point of view, PEO may be autosomal dominant or recessive (due to nDNA mutations), sporadic (due to single large-scale deletion of mtDNA), or maternally inherited (due to mtDNA mutation).

Available online at www.sciencedirect.com
SciVerse ScienceDirect
Neuromuscular Disorders 22 (2012) S226–S229
www.elsevier.com/locate/nmd

Fatigue and exercise intolerance in mitochondrial diseases. Literature revision and experience of the Italian Network of mitochondrial diseases

M. Mancuso [a,*], C. Angelini [b], E. Bertini [c], V. Carelli [d], G.P. Comi [m], C. Minetti [f], M. Moggio [e], T. Mongini [g], S. Servidei [h], P. Tonin [i], A. Toscano [j], G. Uziel [k], M. Zeviani [l], G. Siciliano [a], The Nation-wide Italian Collaborative Network of Mitochondrial Diseases

20% Italian cohort

Table 1
Genotype-based approach. The patients have been divided in two groups, with and without exercise intolerance. For more details, see text. n.s., not significant difference. LHON: Leber hereditary optic neuropathy.

	Exercise intolerance: No ($n = 878$)	Exercise intolerance: Yes (222)	*P*
mtDNA A3243G mutation	62 (7.1%)	33 (14.9%)	**<0.0005**
mtDNA A8344G mutation	27 (3.1%)	9 (4.1%)	n.s.
mtDNA T8993C	19 (2.2%)	1 (0.5%)	n.s.
mtDNA LHON mutations	98 (11.2%)	1 (0.5%)	**<0.0001**
OPA1 mutations	85 (9.7%)	1 (0.5%)	**<0.0001**
POLG mutations	33 (3.8%)	8 (3.6%)	n.s.

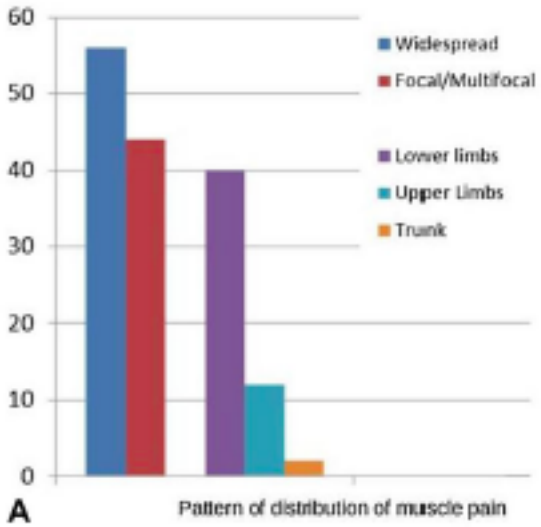
Journal of Neurology
https://doi.org/10.1007/s00415-019-09219-x

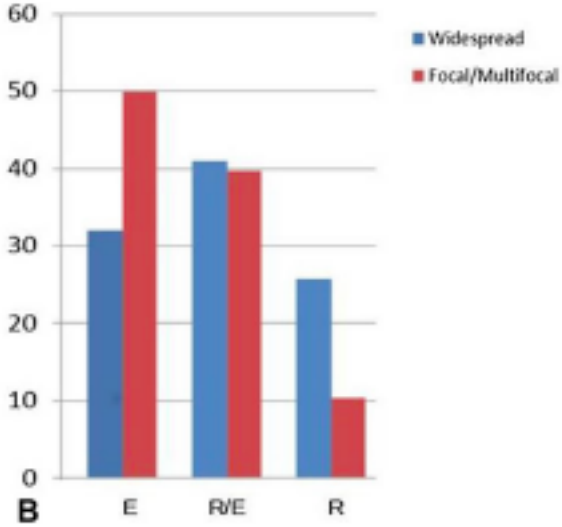
ORIGINAL COMMUNICATION

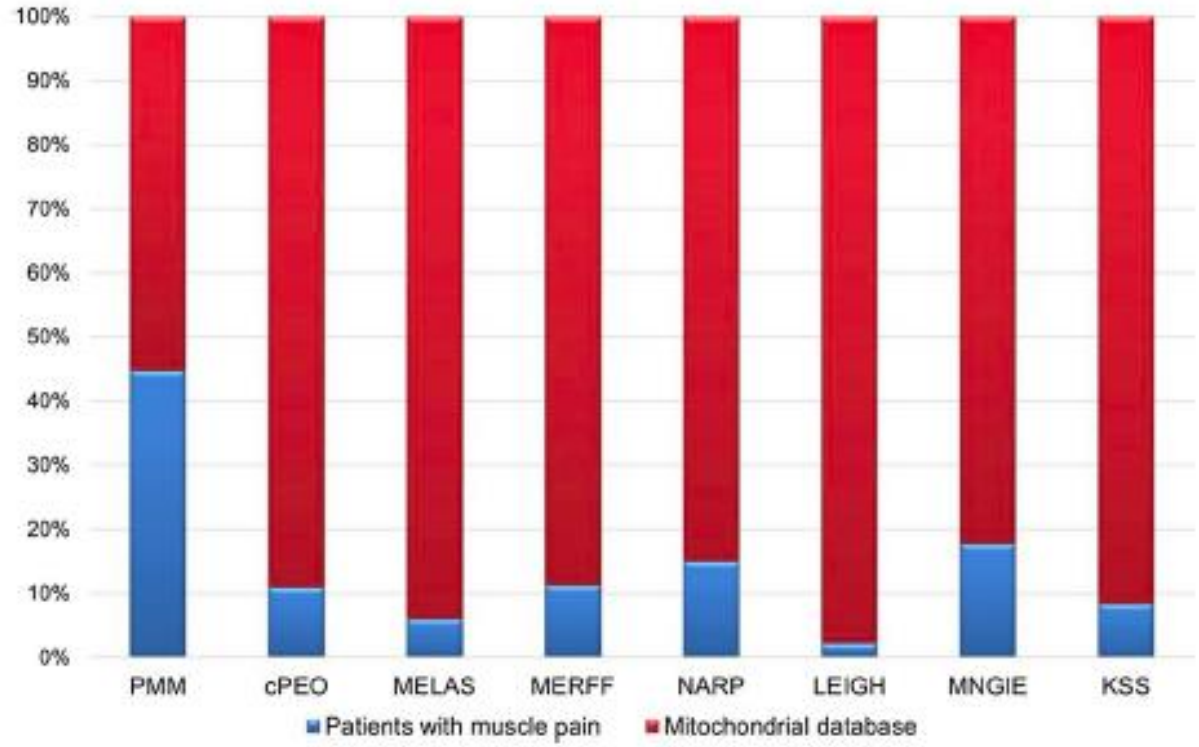
Muscle pain in mitochondrial d network

Massimiliano Filosto[1] · Stefano Cotti Piccinel
Olimpia Musumeci[5] · Paola Tonin[6] · Filippo Mar
Liliana Vercelli[3] · Anna Rubegni[7] · Anna Galvag
Antonio Toscano[5] · Alessandro Padovani[1] · Gab

Receive







Kearns-Sayre Syndrome

Clinical:	Onset before age 20 Ophthalmoplegia and ptosis Pigmentary retinopathy Heart conduction block Cerebellar ataxia Hearing loss Diabetes
Biochemistry:	Lactic acidosis Respiratory chain deficiency CSF protein >100 mg/dl
Morphology:	Ragged-red fibers (COX-negative)
Genetics:	Sporadic large-scale mtDNA deletions Most common mutation deletes 4,977 bp

- **KSS**

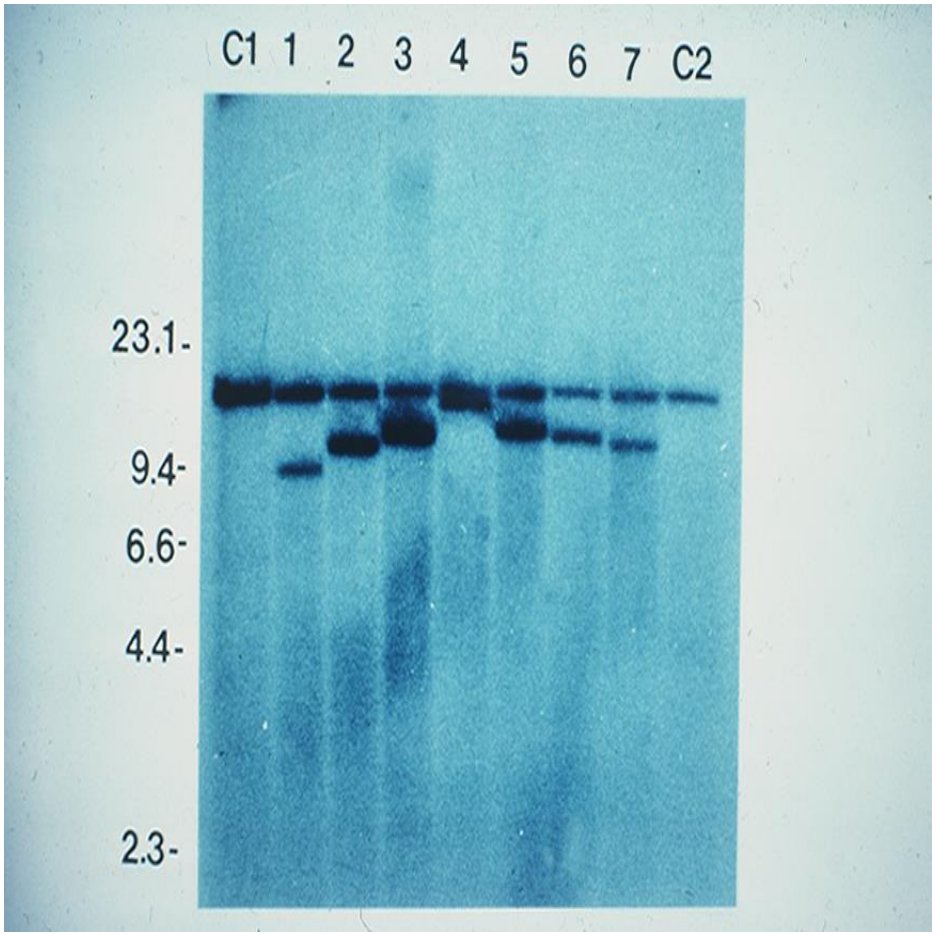
  mtDNA single deletion

- **CPEO**

  Ptosis
  Ophthalmoplegia
  Proximal weakness

- **Pearson syndrome**

  Sideroblastic anemia
  Exocrine pancreas dysfunction



J Neurol
DOI 10.1007/s00415-015-7710-y

ORIGINAL COMMUNICATION

Redefining phenotypes associated with mitochondrial DNA single deletion

Michelangelo Mancuso[1] · Daniele Orsucci[1] · Corrado Angelini[2] · Enrico Bertini[3] · Valerio Carelli[4] · Giacomo Pietro Comi[5] · Maria Alice Donati[6] · Antonio Federico[7] · Carlo Minetti[8] · Maurizio Moggio[9] · Tiziana Mongini[10] · Filippo Maria Santorelli[11] · Serenella Servidei[12] · Paola Tonin[13] · Antonio Toscano[14] · Claudio Bruno[8] · Luca Bello[2] · Elena Caldarazzo Ienco[1] · Elena Cardaioli[7] · Michela Catteruccia[3] · Paola Da Pozzo[7] · Massimiliano Filosto[17] · Costanza Lamperti[16] · Isabella Moroni[15] · Olimpia Musumeci[14] · Elena Pegoraro[2] · Dario Ronchi[5] · Donato Sauchelli[12] · Mauro Scarpelli[13] · Monica Sciacco[9] · Maria Lucia Valentino[4] · Liliana Vercelli[10] · Massimo Zeviani[16] · Gabriele Siciliano[1]

RESULTS: Kearns-Sayre syndrome (KSS)

Progressive external ophthalmoplegia plus:

- **pigmentary retinopathy**
- onset before age 20

Plus at least one of:

- cerebellar ataxia
- cardiac conduction block
- CSF protein > 0.1 g/L

Neuromuscular Disorders 22 (2012)

With these criteria: 15 subjects with KSS (6.6%)

M/F 0.88, age at onset 9.4 ± 4.8 years,

last control 29.4 ± 18.0 years, died 2/15 (13.3%)

Table 1 Clinical features of our patients with mitochondrial DN single deletion ($N = 228$)

	Onset (22.6 ± 14.6 years)	Last evaluation (41.3 ± 18.8 years
Eyelid ptosis	190 (83.3 %)	210 (92.1 %)
Ophthalmoparesis	95 (41.7 %)	192 (84.2 %)
Muscle weakness	29 (12.7 %)	106 (46.5 %)
Exercise intolerance	16 (7.0 %)	45 (19.7 %)
Hearing loss	16 (7.0 %)	42 (18.4 %)
Muscle wasting	5 (2.2 %)	41 (18.0 %)
Swallowing impairment	12 (5.3 %)	34 (14.9 %)
Increased CK	5 (2.2 %)	34 (14.9 %)
Ataxia	11 (4.8 %)	28 (12.3 %)
Retinopathy	12 (5.3 %)	24 (10.5 %)
Failure to thrive/short stature	11 (4.8 %)	22 (9.6 %)
Diabetes mellitus	2 (0.9 %)	20 (8.8 %)
Hypotonia	3 (1.3 %)	18 (7.9 %)
Muscle pain	2 (0.9 %)	13 (5.7 %)
Cardiac conduction defects	3 (1.3 %)	12 (5.3 %)
Increased liver enzymes	1 (0.4 %)	12 (5.3 %)
Anemia	6 (2.6 %)	11 (4.8 %)
Respiratory impairment	2 (0.9 %)	11 (4.8 %)
Neuropathy	3 (1.3 %)	10 (4.4 %)
Migraine	3 (1.3 %)	10 (4.4 %)
Cognitive involvement	4 (1.8 %)	8 (3.5 %)
Tremor	2 (0.9 %)	7 (3.1 %)
Psychiatric involvement	–	7 (3.1 %)
Cardiomyopathy	–	6 (2.6 %)
Hypothyroidism	–	6 (2.6 %)

Among the rare clinical features (1–2.5 %) were: optic neuropathy vomiting (2.2 %); pyramidal signs, cataract, pancytopenia, kidne involvement, gastrointestinal dysmotility (1.8 %); myoglobinuri microcephaly, consciousness disturbance and generalized seizure (1.3 %)

Table 6 New criteria defining KSS spectrum and PEO in patients with single deletion

KSS spectrum
Ptosis and/or ophthalmoparesis due to an mtDNA single large-scale deletion and at least one of the following features
Retinopathy
Ataxia
Cardiac conduction defects
Hearing loss
Failure to thrive/short stature
Cognitive involvement
Tremor
Cardiomyopathy
PEO
Ptosis and/or ophthalmoparesis due to a mtDNA single large-scale deletion not fulfilling the new "KSS spectrum" criteria or criteria for Pearson syndrome

With the new clinical definition, we were able to classify almost all (97%) our single-deletion patients:

- 62.7% PEO (141/22), vs 54.6 NMD 2012
- 31.6% KSS (71/225), vs 6.6 NMD 2012
- 2.7% Pearson (6/225), NMD 2.7

"New" KSS: multisystem involvement, more severe muscular impairment (weakness and wasting), MRI frequently abnormal (white matter, brainstem, basal nuclei), mean age at onset 21 years, worst prognosis.

"New" single-deletion PEO: prominent myopathic involvement, MRI frequently normal, mean age at onset 27 years, better prognosis.

MELAS

MITOCHONDRIAL ENCEPHALOPATHY, LACTIC ACIDOSIS, AND **STROKE-LIKE** EPISODES

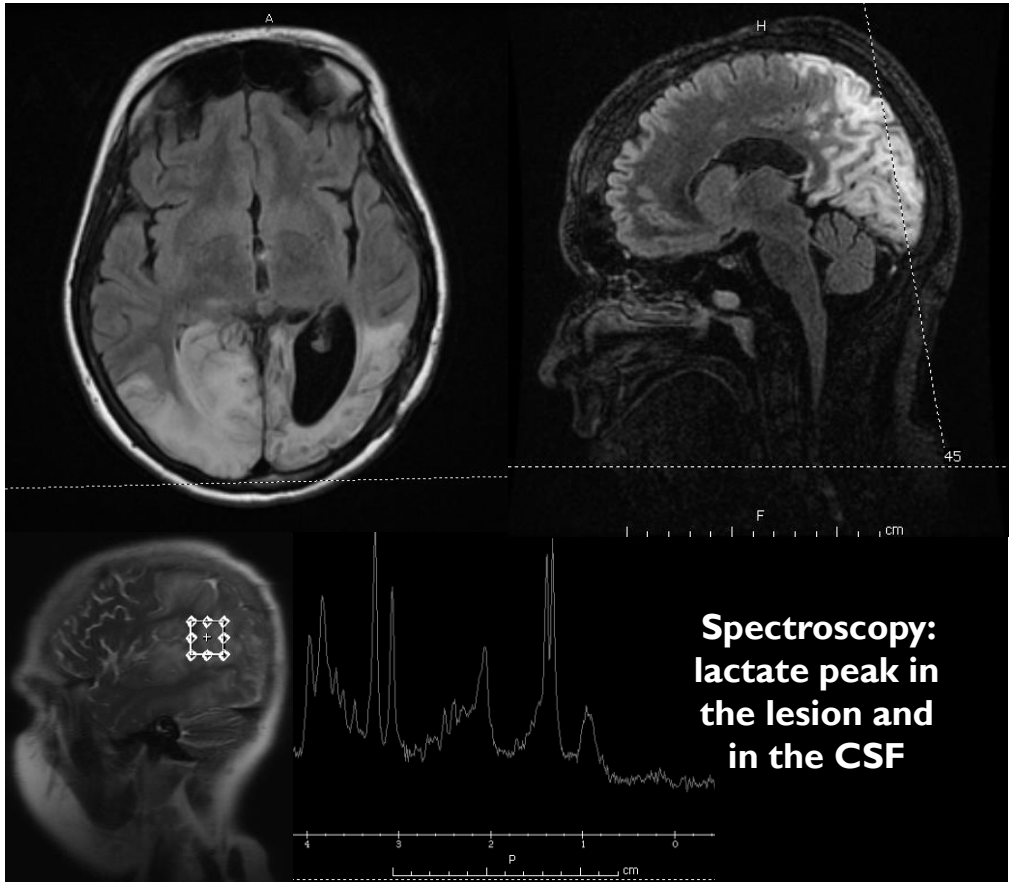
- Acute episodes can present at any age with neurological and/or psychiatric symptoms typically associated with cortical/subcortical MRI changes and EEG abnormalities.
- Stroke-like: **metabolic** stroke driven by SEIZURE activity !
- Recurrent stroke-like episodes: mostly posterior lesions
- **Mutation m.3243A>G tRNA Leu gene** but also other mt mutations and *POLG*

MELAS clinical features

• Stroke-like episodes	***100%***
• Focal or generalized seizures	*85-96%*
• Migraine-like headaches	*77-92%*
• Dementia	*65-90%*
• Mitochondrial myopathy	*87-89%*
• Short stature	*55-95%*
• Hypertrophic cardiomyopathy	*7-18%*
• **PEO**	***13%***
• **Diabetes**	***21%***
• Hearing loss	*27-75%*
• **Family history consistent**	***20-86%***
• **Lactic acidosis**	***94-97%***

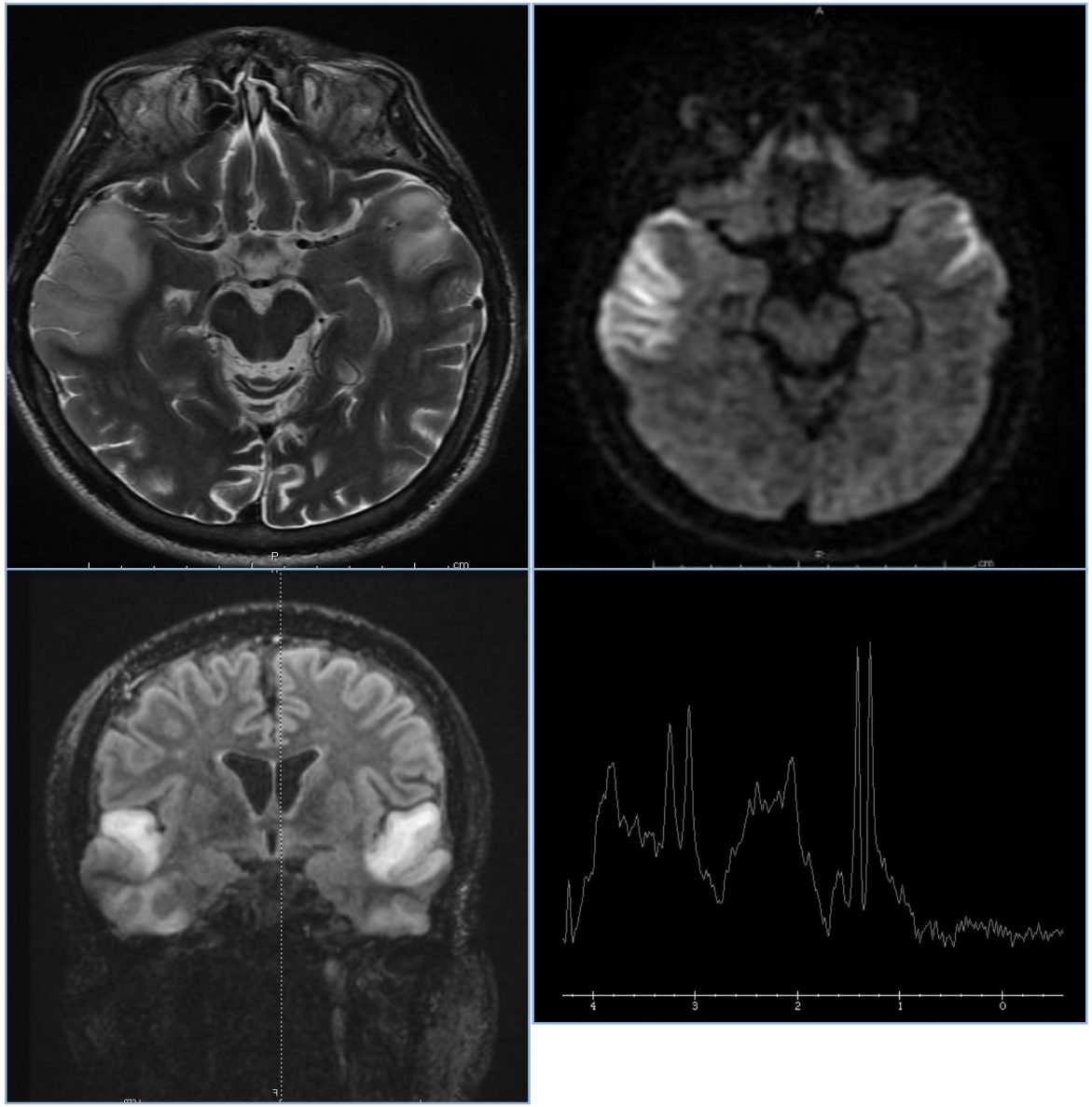
Thambisetty 2002

Age 20: Migraine, cortical blindness and status epilepticus partial



m.15092G>A cyt b p.G116S

MELAS: stroke-like episodes → **Atipical case**



♂

45 years

ER: episodes of **confusion** and **headache** in last 3 weeks, two **generalised seizures** followed by **coma**

family history: negative for neuromuscular or neurodegenerative disorders
Medical history: **hearing loss**

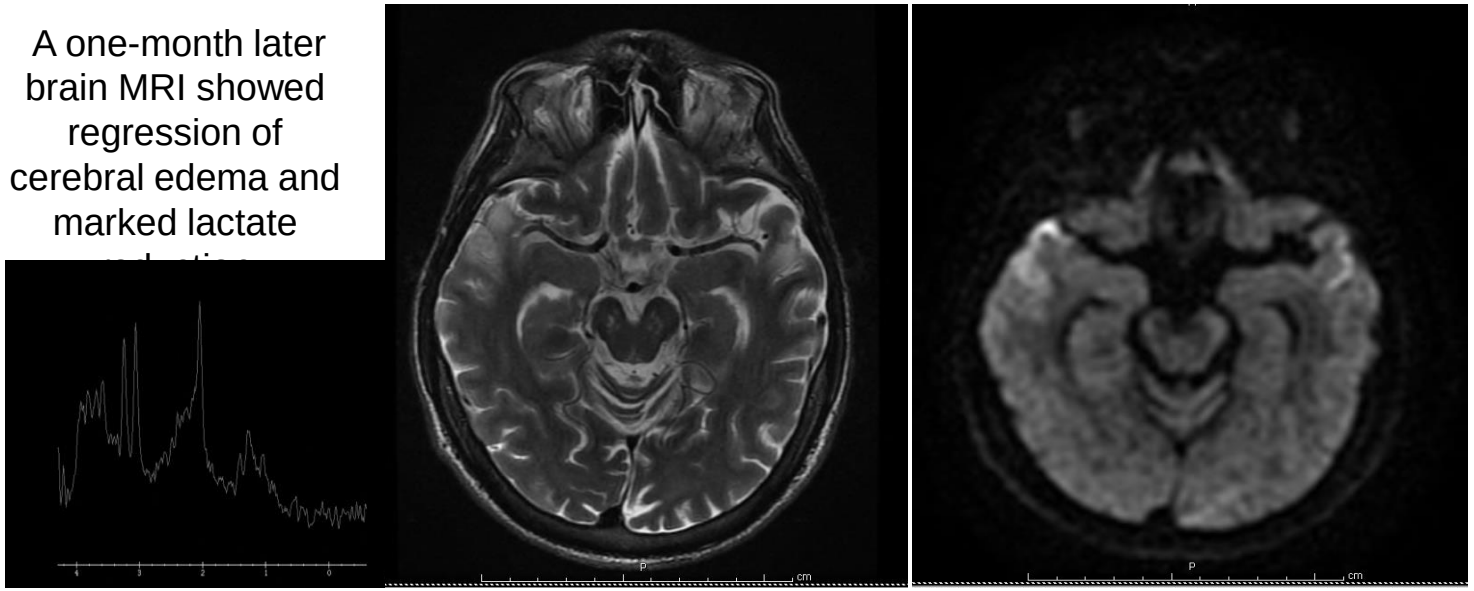
Brain MRI showed temporal lobes T2 hyperintensity with diffusion restriction and contrast uptake and bilateral temporal lobes T1 hyperintensity. Proton spectroscopy showed a lactate peak with reduction of N-Acetyl-Aspartate.

CSF presented increased proteins, glucose and **lactate** but not white cells. Increased **lactate** was also present in serum.

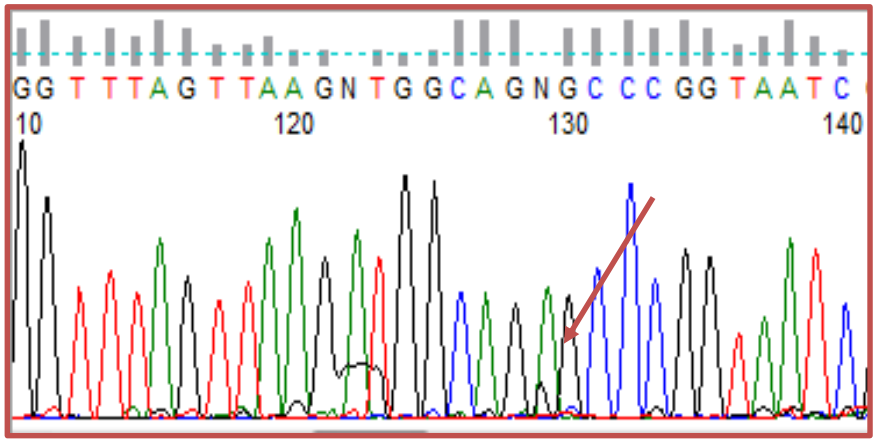
LEV, BDZ carnitine and 600 mgs of coenzyme Q10
NO ARGININE!!!

Rapid clinical improvement (GCS 13) and regression of the lactic acidosis

A one-month later brain MRI showed regression of cerebral edema and marked lactate



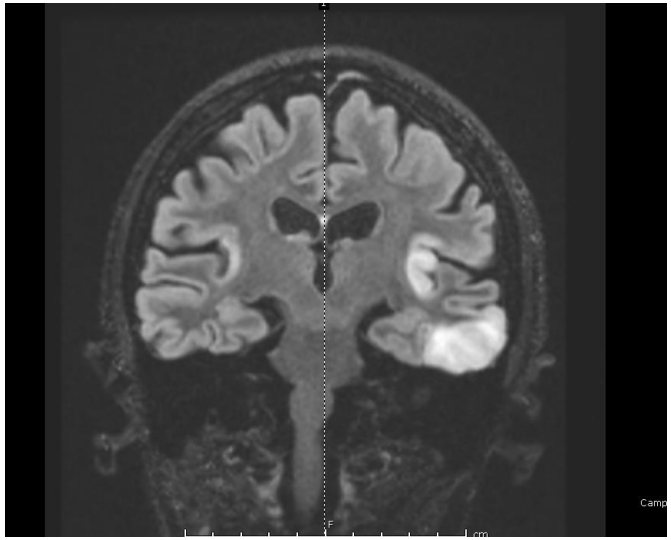
Genetic testing showed the m.3243A> G mtDNA mutation in urine

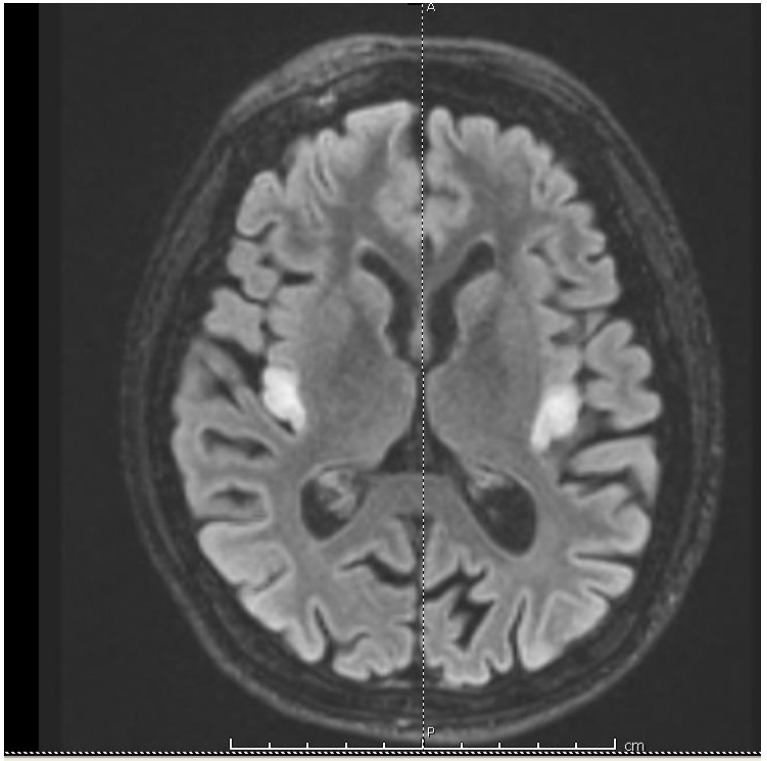


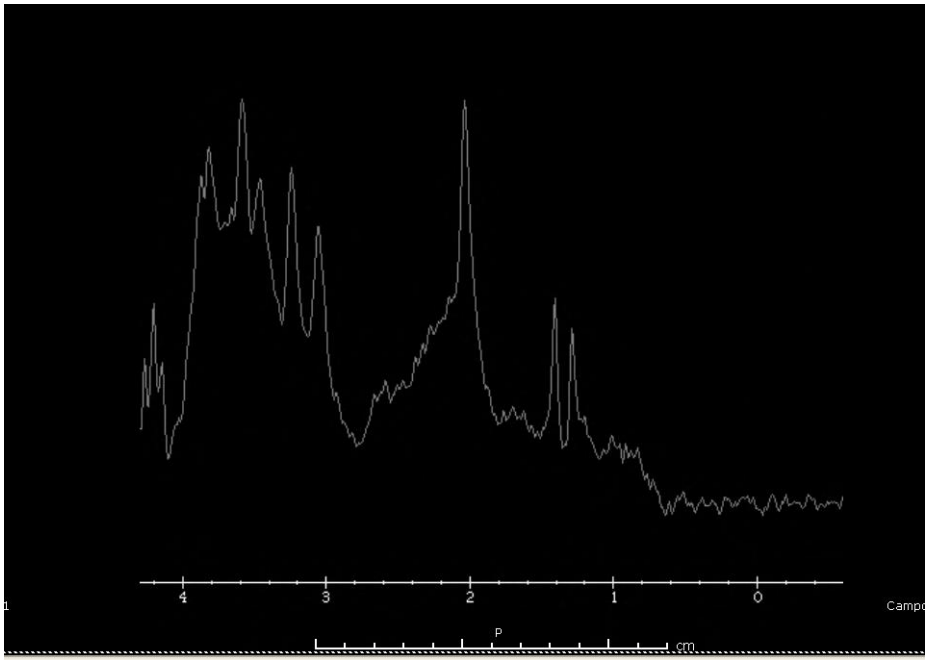
Case II

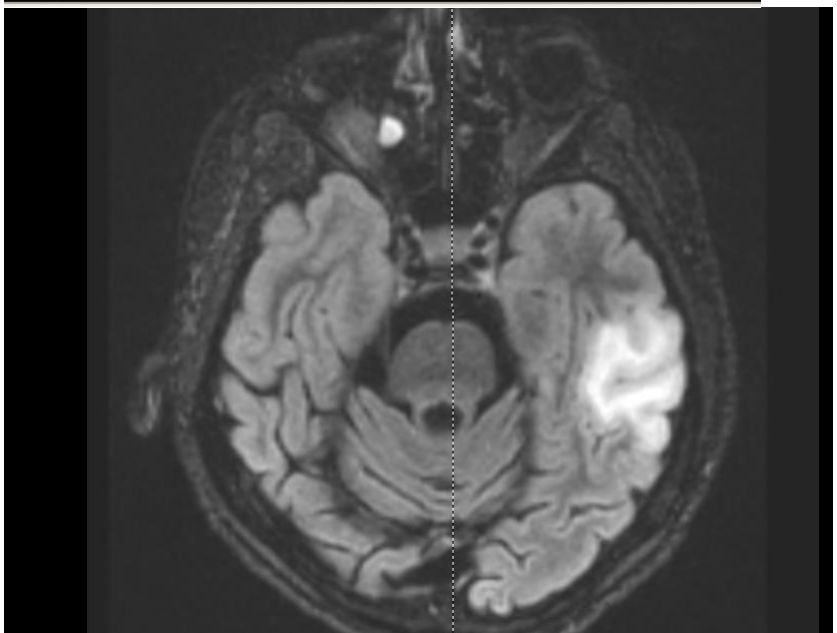
A 41-year-old woman –teacher- complained of subacute afasia, followed by one focal motor seizure

Clinical and family history both negative




A
P
cm

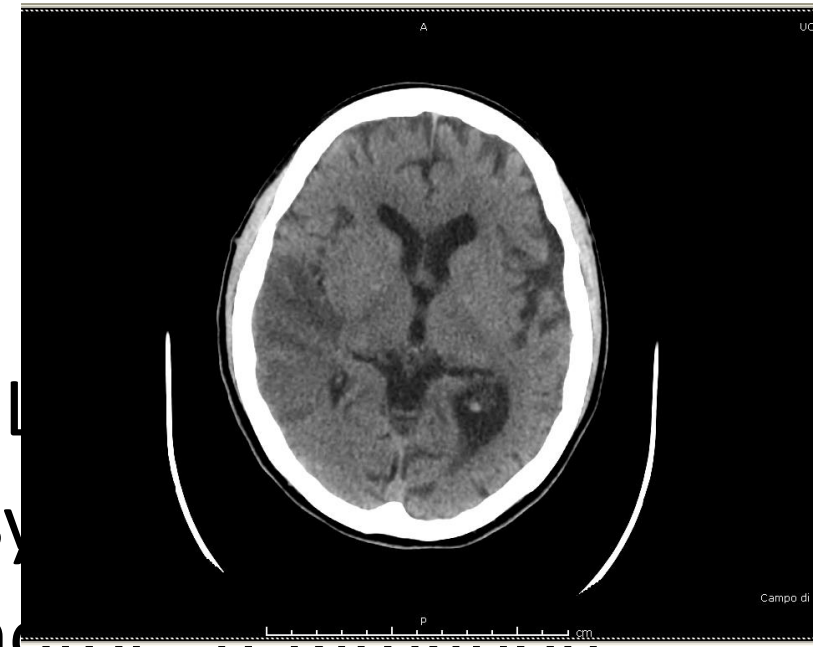

4
3
2
1
0
P
cm



Case II

m.3243 in urine
Tp: l-arg, coq10, carnitene, riboflavine
and delorazepam, aloperidol, cbz

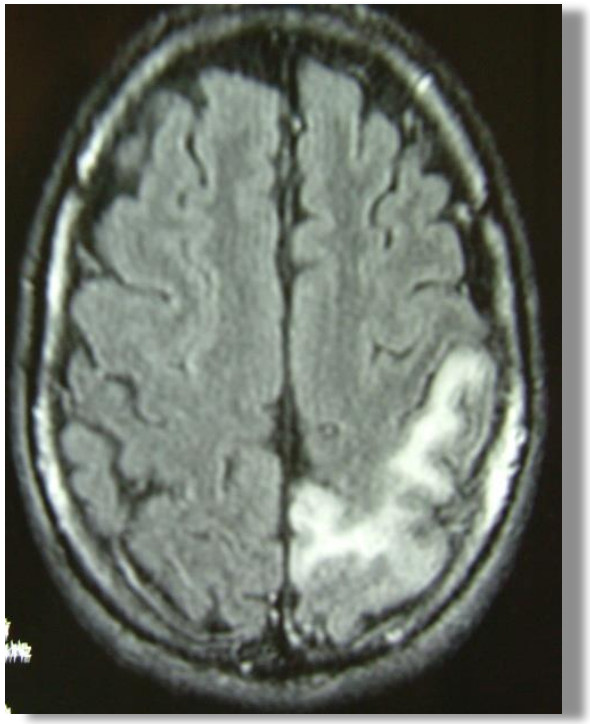
Four months after..



- **Acute negative symptoms**. L
  ADL, hypersonnia, negative sy
  depressed mood, psychomot retardation,
  refuse of food and liquid, bedridden
- Diagnosis: catatonia
- Tp: aloperidol, BDZ (lorazepam, delorazepam, diazepam,), promazine

Case III

- 48-yrs
- Acute aphasia and partial motor seizures
- Mild eyelid ptosis
- Previous medical history negative



m.3243 in urine

Tp: l-arg, coq10, LEV, cbz

One year after…..

- **Acute visual hallucination**
- ER-> ophtalmologist (sic) -> discharged at home
- Few days after also confusion……

EEG POSTERIOR STATUS EPILEPTICUS

take home message

- **Visual hallucination** very typical but: WHO KNOWS??
- Stroke like episode driven by SEIZURE activity -> potentially treatable??

MANAGEMENT

There is **no specific consensus approach for treating individuals with MELAS syndrome**.

All patients suspected to be suffering a stroke-like episode due to underlying mitochondrial disease
should be discussed or referred to a mitochondrial disease specialist in the acute setting.

Monitoring for the development of arrhythmia

Gastroparesis and small bowel intestinal pseudo-obstruction

MERRF (myoclonic epilepsy with RRF)

Clinical: Onset in childhood
Myoclonus
Epilepsy
Ataxia
Neuropathy
Multiple lipomas

Biochemistry: Lactic acidosis
Respiratory chain deficiency

Morphology: Ragged-red fibers (COX-negative)

Genetics: Maternally-inherited mtDNA point mutations
Most common mutation in $tRNA^{Lys}$ at nt-8344

Neurology® 2013;80:1-6

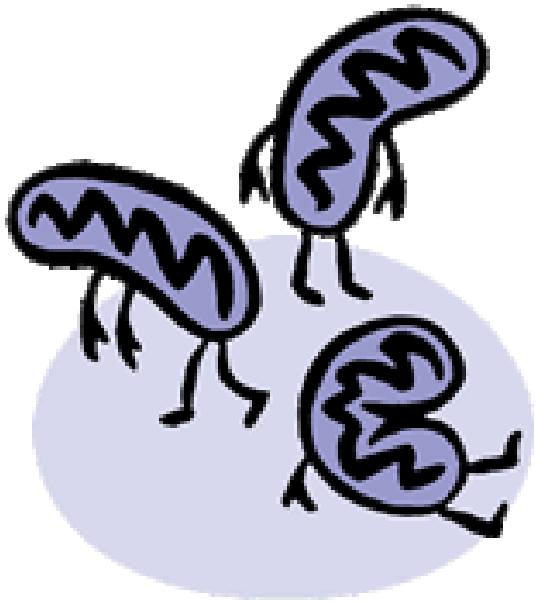
Table e-2. Clinical features of the 8344A>G carriers.

	Our cohort (n = 39)	Literature revision (n = 282)	Total (n = 39 + 282 = 321)
Muscle weakness	20 (51.3%)	118 (41.8%)	138 (43.0%)
Increased CK	15 (38.5%)	30 (10.6%)	45 (14.0%)
Exercise intolerance	15 (38.5%)	40 (14.2%)	55 (17.1%)
Seizures	12 (30.8%)	94 (33.3%)	106 (33.0%)
Hearing loss	12 (30.8%)	87 (30.9%)	99 (30.8%)
Multiple lipomatosis	11 (28.2%)	50 (17.7%)	61 (18.9%)
Ptosis/ophthalmoparesis	10 (25.6%)	19 (6.7%)	29 (9.0%)
Ataxia	8 (20.5%)	129 (45.7%)	137 (42.7%)
Myoclonus	8 (20.5%)	134 (47.5%)	142 (44.2%)
Muscle wasting	7 (17.9%)	17 (6.0%)	24 (7.5%)
Muscle pain	6 (15.4%)	13 (4.6%)	19 (5.9%)
Arrhytmia	6 (15.4%)	7 (2.5%)	13 (4.0%)
Neuropathy	5 (12.8%)	52 (18.4%)	57 (17.8%)
Cardiomyopathy	4 (10.3%)	20 (7.1%)	24 (7.5%)
Diabetes	4 (10.3%)	16 (5.7%)	20 (6.2%)
Cognitive involvement	3 (7.7%)	64 (22.7%)	67 (20.9%)
Migraine	3 (7.7%)	14 (5.0%)	17 (5.3%)
Swallowing impairment	2 (5.1%)	8 (2.8%)	10 (3.1%)
Hypothyroidism	2 (5.1%)	2 (0.7%)	4 (1.2%)

THE MITOCHONDRIAL DISORDER NOT TO MISS: WHICH ARE THE RED FLAGS?

Neurologist in trouble..

- 100s of different mtDNA-related diseases
- 100s of different nDNA-related diseases
- Even in individuals with the same mutation, there are different symptoms
- Change over time
- Challenging to diagnose
- Challenging to treat



Diagnostic approach

The diagnostic process **is no different from that employed for other diseases** and includes patient and family history, physical and neurologic examination, routine and special laboratory tests, exercise physiology, muscle biopsy for morphology and biochemistry, and molecular genetic screening

You see Watson, but you do not observe

Diagnosis: assessing involvement

- Brain MRI
- EEG
- Sleep Study
- PFTs
- Echocardiogram
- EKG
- Abdominal Ultrasound
- Swallow Evaluation
- Nutrition Assessment
- Developmental Assessment
- Vision Test
- Ophthalmologic Examination
- Hearing Test

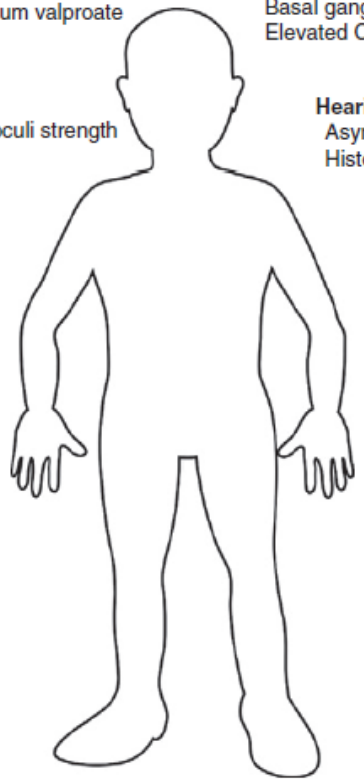
- Labs:
  - Liver Function Tests
  - Fasting Serum Glucose
  - Ammonia
  - CK
  - Amino Acids (alanine, citrulline..)
  - Lactic Acid
  - Free/Total Carnitine
  - Organic aciduria
  - Blood anemia…

Family history

A family history must be taken meticulously, with special attention to minimal and **apparently unspecific** signs in the maternal lineage, including short stature, diabetes, migraine, hearing loss, exercise intolerance and psychiatric disorders (depression, BP, autism, schizophrenia)

The little things are infinitely the most important

Clinical symptoms and signs



Seizures:
Acute de novo onset status epilepticus
Focal/ multi-focal myoclonus
Triggered by physiological decompensation
Well-controlled inter-ictal periods
Exacerbated by sodium valproate

CNS involvement:
Young-onset focal neurological deficit, typically associated with a clinical prodrome, focal seizures/ myoclonus, status epilepticus
Grey-matter affected that does not follow vascular territory and potentially reversible
Basal ganglia calcification
Elevated CSF lactate

Eye manifestations:
Ptosis
Asymmetric onset
Frontalis overactivity with preserved orbicularis oculi strength
Slow progression, little diurnal variation
Progressive external ophthalmoplegia
Retinal pigmentary changes
Perimacular in distribution
vision not affected in general

Hearing loss:
Asymmetric, young-onset sensorineural hearing loss
History of partial recovery at some point

Gastrointestinal dysmotility:
Severe constipation, can fluctuate with diarrhoea
Intermittent presentations with pseudo-obstruction

Diabetes:
Occur in the young and non-overweight
Less liable to diabetic retinopathy/ neuropathy

Myopathy:
Symmetrical proximal involvement
Can have significant myalgia

Neuropathy:
Subclinical involvement
Axonal or demyelinating or a sensory ganglionopathy

As a general hint, **the apparently unrelated involvement of two or more tissues** should suggest the possibility of mitochondrial disease, including the cases where the family history is unremarkable

LABORATORY TESTS

- Labs:
  - LFTs
  - CPK
  - Fasting Serum Glucose
  - Ammonia
  - Amino Acids
  - Lactic Acid
  - Free/Total Carnitine
  - UOA
  - Sideroblastic Anemia
  - FGF21, GDF15

When hypothesize a mitochondrial disorder?

Mitochondrial disorders in neurology are either underdiagnosed : “what is this bizarre syndrome?” or overdiagnosed: “this syndrome is so bizarre that it must be mitochondrial”

↓

but

↓

It is a mistake to confound strangeness with mystery. The most commonplace crime is often the most mysterious because it presents no new or special features from which deductions may be drawn. **The strange details, far from making the case more difficult, have really had the effect of making it less so**.”

Details
Mild signs-symptoms
Multidisciplinary approach
Observe (ie lipomas)!
Associations (i.e.)
-NSHL&DM
-myoclonus&ataxia
-PEO&Parkinsonism
-liver f. &encephalopathy

Deep inside
Lab tests (lactate,aa..)
Radiology

BEYOND NEUROLOGY

-cardiopathy
-liver imp.
-diabetes
-lactic acidosis

SNC

-seizures & myoclonus
-ataxia
-cognitive imp.
-stroke like ep.
-mov. Disorders
-optic atrophy
-NSHL
-psycomotor imp.

NEUROMUSCULAR

-PEO
-Exercise intolerance
-Weakness, fatigue, ex.int.
-wasting
-dysphagia
-numbness
-paresthesia