Teaching Course 1

Mitochondrial diseases for beginners (Level 1)

Mitochondrial diseases of the brain

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Mitochondrial diseases of the brain

Thomas Klopstock

Disclosures

Research support, speaker honoraria, travel costs and/or consulting fees from ApoPharma Inc., CoA Therapeutics, Retrophin Inc., GenSight Biologics and Santhera Pharmaceuticals
Mitochondrial diseases of the brain

MELAS - Mitochondriale Enzephalomyopathy, Lactic Acidosis and stroke-like episodes

<table>
<thead>
<tr>
<th>Frequency</th>
<th>Manifestations</th>
<th>Frequency</th>
<th>Manifestations</th>
</tr>
</thead>
<tbody>
<tr>
<td>≥90%</td>
<td>• Stroke-like episodes</td>
<td>25%-49%</td>
<td>• Basal ganglia calcification</td>
</tr>
<tr>
<td></td>
<td>• Dementia</td>
<td></td>
<td>• Myoclonus</td>
</tr>
<tr>
<td></td>
<td>• Epilepsy</td>
<td></td>
<td>• Ataxia</td>
</tr>
<tr>
<td></td>
<td>• Lactic acidemia</td>
<td></td>
<td>• Episodic altered consciousness</td>
</tr>
<tr>
<td></td>
<td>• RRF on muscle biopsy</td>
<td></td>
<td>• Gait disturbance</td>
</tr>
<tr>
<td>7%-89%</td>
<td>• Hemiparesis</td>
<td>&lt;25%</td>
<td>• Depression</td>
</tr>
<tr>
<td></td>
<td>• Cortical vision loss</td>
<td></td>
<td>• Anxiety</td>
</tr>
<tr>
<td></td>
<td>• Recurrent headaches</td>
<td></td>
<td>• Psychotic disorders</td>
</tr>
<tr>
<td></td>
<td>• Hearing impairment</td>
<td></td>
<td>• Diabetes mellitus (type 1 or 2)</td>
</tr>
<tr>
<td></td>
<td>• Muscle weakness</td>
<td></td>
<td></td>
</tr>
<tr>
<td>50%-74%</td>
<td>• Peripheral neuropathy</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Learning disability</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Memory impairment</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Recurrent vomiting</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Short stature</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

El-Hattab et al, 2018

El-Hattab et al, 2016
The phenotypical spectrum of the m.3243A>G mutation

- Hypacusis: 49%
- Respiratory...: 47%
- Migraine: 44%
- Exercise intolerance: 43%
- Glucose Intolerance: 36%
- Depression: 35%
- GI Symptoms: 29%
- Ataxia: 29%
- No Encephalopathy: 29%
- Muscle Weakness: 26%
- Epilepsy: 25%
- Ptosis: 21%
- SLE: 18%
- 0% 10% 20% 30% 40% 50% 60% 70%

mtDNA mutation mostly m.3243A>G

MELAS - Imaging

- cortical pattern
- independent of vascular territories
- occipital, temporal > parietal >> frontal

• own patient, 29 yrs
• Pauli et al, 2013
• Sharfstein et al, 1999 „A herpes not so simplex“
• Geraets et al, 2013
MELAS - Imaging

Dynamic Lesion

Stroke-like episodes and lesions
• may recover completely or incompletely
• but after multiple episodes predominantly occipital atrophy

Pathophysiological hypothesis

- Neuronal energy deficiency
- Neuronale Hyperexcitability
- Epileptic Activity
- Cortical Edema
- Neuronal Loss
### Stroke-like lesions beyond MELAS

- **Alpers-Syndrom**
  with mutations in the mitochondrial Polymerase gamma gene (POLG)

- **SCAE (spinocerebellar ataxia and epilepsy)**
  with mutations in the mitochondrial Polymerase gamma gene (POLG)

### Diagnostics of MELAS resp. its associated mutations

- "ragged red fibers" in muscle biopsy
- Urine > Blood
Therapy of MELAS

Ng et al., in preparation Consensus-based Guidance for The Management of Mitochondrial Stroke-like Episodes
We do not advocate the use of L-arginine in the treatment of stroke-like episodes. There is no robust scientific evidence. RCT needed.

### Antiepileptic Tx

**Acute i.v.:**
- Levetiracetam
- Phenytoin
- Phenobarbital
- Lacosamide
- Midazolam

**Later p.o.:**
- Levetiracetam
- Topiramate
- Lamotrigine
- Valproate

### Table: KHENERGY Study

<table>
<thead>
<tr>
<th>Autor</th>
<th>Jahr</th>
<th>Medikament</th>
<th>Patienten</th>
<th>Design</th>
<th>Effekt</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tamopolsky</td>
<td>1997</td>
<td>Kreatin</td>
<td>6 MELAS</td>
<td>crossover 21+21</td>
<td>Laktat</td>
</tr>
<tr>
<td>Glover</td>
<td>2010</td>
<td>Coenzym Q</td>
<td>15 MELAS</td>
<td>crossover 60-60</td>
<td>Laktat</td>
</tr>
<tr>
<td>Kaufmann</td>
<td>2006</td>
<td>Dichloracetat</td>
<td>30 MELAS</td>
<td>crossover 90-90</td>
<td>MRS Gehirn</td>
</tr>
</tbody>
</table>

The KHENERGY Study: Safety and Efficacy of KH176 in Mitochondrial m.3243A>G Spectrum Disorders

Gorman et al., 2016

Mitochondrial diseases of the brain

- MELAS
- MERRF
- Leigh sy.
- LHON
MERRF - myoclonic epilepsy and ragged-red fibres

- a multisystemic mitochondrial disease that is characterised by myoclonus, seizures, cerebellar ataxia, and mitochondrial myopathy with ragged-red fibres.
- 80–90% of cases caused by the m.8344A>G mutation of the mtDNA

The phenotypical spectrum of the m.8344A>G mutation

German mitoNET cohort N = 34; black bars
Italian MITOCON cohort N = 34; grey bars
**Diagnostics of MERRF resp. its associated mutations**

![Diagram of mitochondrial DNA with markers and arrows pointing to "ragged red fibers" in muscle biopsy]

**Mitochondrial diseases of the brain**

![Neurological symptoms and signs related to mitochondrial diseases with images of MELAS, MERRF, Leigh syndrome, and LHON]

*Gorman et al., 2016*
Leigh syndrome: infantile subacute necrotizing encephalomyelopathy

- fatal disorder of early childhood
- showing psychomotor regression, movement disorders and brain stem dysfunction
- symmetrical lesions in basal ganglia and brain stem in imaging and pathologically
- Caused by many different mitochondrial defects, both mtDNA- and nuclear-encoded

<table>
<thead>
<tr>
<th>Biochemistry</th>
<th>Genetics</th>
</tr>
</thead>
<tbody>
<tr>
<td>Complex I</td>
<td>nuclear and mtDNA subunits</td>
</tr>
<tr>
<td>Complex II</td>
<td>nuclear subunits SDHA</td>
</tr>
<tr>
<td>Complex III</td>
<td>nuclear assembly factor</td>
</tr>
<tr>
<td>Complex IV</td>
<td>nuclear assembly factors</td>
</tr>
<tr>
<td>Complex V</td>
<td>mtDNA</td>
</tr>
<tr>
<td>PDHC</td>
<td>X-chromosomal subunit</td>
</tr>
</tbody>
</table>
### Leigh syndrome diagnostics

**Phenotype**
- maternal
- X-ch.
- de novo point mutation
- mtDNA depletion

**Gene panel**
- ATP8B1
- PDHA1
- SLC25A1
- SUCLA2
- SDHA
- SURF1
- SDHAF1
- SURF2
- SDHBD
- TMT1
- TMEM107

### MRI imaging patterns in mitochondrial diseases

**Two pathognomic patterns**
- Cortical hyperintensity (stroke-like lesion)
  eg in MELAS, POLG
- Hyperintensity basal ganglia and brain stem
  eg in Leigh syndrome

**Two unspezific patterns**
- Leukenzephalopathie
  eg in MNGIE, KSS
- Cerebral atrophy
  eg in CPEO, KSS
Mitochondrial diseases of the brain

- mtDNA mutation relative frequencies:
  - ~90% m.11778, m.3460, m.14484
  - Most frequent: ~70% m.11778G>A
    - Exception!!! ~90% m.14484T>C in patients of French-Canadian descent (due to founder event)

- Minimum prevalence:
  - 1 in 31,000: North of the UK
  - 1 in 39,000: Netherlands
  - 1 in 50,000: Finland

- Estimated as the most frequent mitochondrial disease

Leber’s Hereditary Optic Neuropathy (LHON)

- Estimated as the most frequent mitochondrial disease
- Minimum prevalence:
  - 1 in 31,000: North of the UK
  - 1 in 39,000: Netherlands
  - 1 in 50,000: Finland
- mtDNA mutation relative frequencies:
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Leber’s Hereditary Optic Neuropathy (LHON)

- Predominantly affects young adult males
- M>F
- Age at onset: range 4 – 82 yrs
  - peak of onset: 2nd and 3rd decades
  - early-onset LHON (< 12yrs)
  - late-onset LHON (> 50 yrs)

LHON: Clinical manifestations

- Painless acute/ subacute progressive vision loss
- Sequential affection of 1st and 2nd eye (after a median of 6-8 weeks) (~75%)
  - or
  - both eyes affected from onset (~25%)
- Progression in days/weeks/months, then stability
**LHON: Clinical manifestations**

- Fundoscopy: Papilledema in the acute phase, later optic atrophy, temporal predominant
- Visual fields: Bilateral central scotoma
- OCT: Reduced temporal circumpapillary RNFL thickness

**LHON: Pathogenesis & „window of opportunity“**

<table>
<thead>
<tr>
<th>Asymptomatic phase²</th>
<th>Subacute phase²</th>
<th>Dynamic phase²</th>
<th>Chronic phase²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Primary mtDNA mutation³</td>
<td>Secondary etiologic factors¹</td>
<td>Mitochondrial dysfunction</td>
<td>Reduced RGC function⁴</td>
</tr>
<tr>
<td>Loss of RGCs⁵</td>
<td>Loss of vision</td>
<td>Terminal vision loss</td>
<td></td>
</tr>
</tbody>
</table>

- In the acute/subacute phase, RGCs are inactive but are still viable³,⁴
- In some patients, this is reversible and vision may be recovered³,⁴

**References**

**LHON: Natural history & goals of therapy**

- Clinically Relevant Recovery (CRR)
- Clinically Relevant Stabilization (CRS)

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**RHODOS (Rescue Of Hereditary Optic Disease Outpatient Study)**

- double-blind, randomized, placebo-controlled, parallel group trial
- 85 LHON patients in 3 centers (Munich, Newcastle, Montreal)
- largest trial to date in an mtDNA-associated disease
Expanded Access Program (EAP)

established in late 2011 following an increasing number of unsolicited requests from physicians for access to the drug

Idebenone 300 mg orally 3 times daily

patients

\( n = 87^a \)

(from 38 sites)

Month 0

Month 60

Expanded Access Program (EAP)

LHON patients

\( n = 87^a \)

(from 38 sites)

Month 0

Month 60

Idebenone 300 mg orally 3 times daily

Catarino et al, submitted
EAP-CRR: Time to first recovery

Total patients with CRR=41 patients (100%)

Treatment duration of at least 18-24 months is needed to maximize the probability of observing an initial CRR

EAP: Average magnitude of VA recovery

Mean effect size of ~6 lines in responders after 16 month

Off - chart

n=34/69 responders with CRR at last assessment

Average visual acuity at nadir and last observation in patients CRR by mutation

In patients with recovery in both eyes, the eye with the best recovery is reported
Idebenone development program in LHON

LHON – Gene therapy

- Unmet need at least for idebenone non-responders
- The eye as an ideal organ for gene therapy
  - Immune-privileged, closed system
  - Intravitreal injections introduce genetic material close to target cells
  - Slow turnover of retinal cells support long-term expression of transduced genes
- AAV vector has proven safety and efficacy for transduction of retinal cells
- BUT: the mutation is not in a nuclear gene but in the mitochondrial DNA
  → allotypic expression
LHON – Gene therapy „from bench to bedside“

- GS010 restores respiratory chain complex I in patients fibroblasts (Boremal et al. 2008)
- GS010 prevents optic atrophy and visual loss in LHON rats (Cremers-Thies et al. 2018)
- Phase 1 trial demonstrates safety, tolerability and trends of efficacy (Vigò et al. 2019)
- Phase 3 trials

**Different patient inclusion criteria**

**Same design**

One eye randomized to GS010, other eye sham injection

**Same endpoints at Week 48**

**Primary**
- Mean difference change from baseline ETDRS letters, this treated eyes vs. sham treated eyes (Snellen VA used for statistical analysis at Week 48)

**Secondary**
- SD-OCT, visual field, color and contrast tests
- Reproducibility analysis:
  - Gain from baseline of 15 or more ETDRS letters
  - Snellen acuity better = or better than 20/200
  - Treated vs. sham eyes BCVA for best-seeking and worst-seeking eyes

LHON – Gene therapy - Results

- well tolerated, frequent intraocular inflammation responsive to conventional treatment and without sequelae
- change in retinal ganglion cell macular volume from baseline to week 72
  - Treated eyes: no loss
  - Untreated eyes: $-0.044 \text{mm}^3$ $p=0.0090$ (ANCOVA analysis)
- change in thickness of the papillo-macular bundle from baseline to week 72
  - Treated eyes: $-1.6 \text{ \mu m}$
  - Untreated eyes: $-3.6 \text{ \mu m}$ $p=0.0362$ (ANCOVA analysis)
- clinically meaningful improvement of +15 ETDRS letters in treated eyes but similar in untreated eyes

[Graph showing changes in visual outcomes over time]
Mitochondrial diseases of the brain

- MELAS
- MERRF
- Leigh sy.
- LHON

Gorman et al, 2016

Networks

mitoNET
German Network for mitochondrial disorders

<table>
<thead>
<tr>
<th>No.</th>
<th>Principal Investigator</th>
<th>Institution</th>
<th>Title of Subproject</th>
<th>Function in the consortium</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Dr. Klopstock</td>
<td>LMU München</td>
<td>Coordination of the consortium</td>
<td>Coordination, Monitoring, Processing of results</td>
</tr>
<tr>
<td>2</td>
<td>Dr. Proksch</td>
<td>TU München</td>
<td>mitoGENE</td>
<td>Molecular diagnostics by whole genome and RNA sequencing</td>
</tr>
<tr>
<td>3</td>
<td>Dr. Kremser</td>
<td>TU München</td>
<td>mitoVAUD</td>
<td>Validation platform for variants of uncertain significance by functional complementation</td>
</tr>
<tr>
<td>4</td>
<td>Dr. Pfeiffer</td>
<td>Univ. Greifswald</td>
<td>mitoMETABO</td>
<td>Biomarker discovery for disease, progression and treatment by metabolomics</td>
</tr>
<tr>
<td>5</td>
<td>Dr. Dittrich</td>
<td>Univ. of Frankfurt</td>
<td>mitoPROT</td>
<td>Investigate molecular pathomechanisms and treatment effects by proteomics and compakosmics</td>
</tr>
<tr>
<td>6</td>
<td>Dr. Geppert</td>
<td>LMU Munich</td>
<td>mitoREGISTRY</td>
<td>Clinical registry (cross-sectional and longitudinal)</td>
</tr>
<tr>
<td>7</td>
<td>Dr. Meister</td>
<td>LMU Munich</td>
<td>mitoYEAR</td>
<td>eHealth project to evaluate utility of wearable activity monitor as possible new endpoints for future clinical trials</td>
</tr>
<tr>
<td>8</td>
<td>Dr. Klopstock</td>
<td>LMU Munich</td>
<td>mitoSAMPLE</td>
<td>To collect biological materials and make them available for mitochondrial research</td>
</tr>
</tbody>
</table>

Antragstellung für die Fördermaßnahme Translationsoriентierte Verbundvorhaben im Bereich der seltenen Erkrankungen

29/06/2019
Global Networks

Mitochondrial Disorders: from a world-wide registry to medical genomics, toward molecular mechanisms and new therapies

Ludwig-Maximilians-Universität München
Dept. of Neurology
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Anna Baur-Ulatowska
Aimut Bischoff
Ira Brandstetter
Boriana Büchner
Claudia Catarino
Ivan Karin
Florentine Radelfahr
Claudia Stendel
Oskar Mikazans

Dept. of Ophthalmology
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Siegfried Priglinger
Günther Rudolph

Technical University of Munich
Dept. of Human Genetics
Thomas Mettling
Holger Prokisch

International collaborations

University of Bologna
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Valerio Carelli
Chiara La Morgia

University of Cambridge
Patrick Chinnery
Rita Horvath
Patrick Yu-Wai-Man

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Michelangelo Mancuso

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Tomas Honzik
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University of Newcastle
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Istituto Nazionale Neurologico, Milano
Valeria Tiranti