Genes, and the genetic background of centronuclear myopathy

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A few words about Strasbourg

IGBMC: a major center for biomedical research
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Congenital myopathies
Better: structural myopathies - no fiber degeneration

- Affect children and adults in all population
- Significant burden for families
- Mostly progressive muscle weakness
- Specific structural anomalies on biopsy
Congenital myopathies

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- Significant burden for families
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Centronuclear myopathy (CNM)

Age of onset + severity depend on gene

<table>
<thead>
<tr>
<th>Gene</th>
<th>Age of Onset</th>
<th>Severity</th>
</tr>
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<tbody>
<tr>
<td>MTM1</td>
<td>birth</td>
<td>severe</td>
</tr>
<tr>
<td>RYR1</td>
<td>birth</td>
<td>moderate</td>
</tr>
<tr>
<td>BIN1</td>
<td>child/adulthood</td>
<td>moderate/mild</td>
</tr>
<tr>
<td>DNM2</td>
<td>child/adulthood</td>
<td>moderate/mild</td>
</tr>
</tbody>
</table>
Science is (a bit) like football. Complementary expertise within a team.

defense

Mégane

Johann

Xavière

Sarah

genetics

which gene?
Science is (a bit) like football. Complementary expertise within a team.

**defense**
- Mégane
- Johann
- Sarah

**midfield**
- Christine
- Xavière
- Sarah
- Jocelyn
- Roberto

**genetics**
- which gene?

**cell biology**
- what happens in the cell?
Science is (a bit) like football
complementary expertise within a team

Knowing the genetic cause is the first step towards a therapy

genetics
which gene?
cell biology
what happens in the cell?
therapies
how to treat?
Science is (a bit) like football
complementary expertise within a team

Knowing the genetic cause is the first step towards a therapy

- Genetics: Which gene?
- Cell Biology: What happens in the cell?
- Therapies: How to treat?
Chromosomes: compacted DNA

Genetic cause = mutation on DNA

Chromosomes are ranked by size

Each chromosome twice:
- 1 from mother
- 1 from father

Find the mutation:
Read the DNA
A book....but a rather big one
Harry Potter novels are very small compared to DNA

1 000 000 letters
A book....but a rather big one
Harry Potter novels are very small compared to DNA

1 000 000 letters

DNA: 6 000 000 000 letters
= Hogwarts library
A book....but a rather big one
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Misspelling in one book, one chapter, one sentence, one word
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Harry Potter novels are very small compared to DNA

1 000 000 letters

DNA: 6 000 000 000 letters
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Misspelling in one book, one chapter, one sentence, one word

How to find the misspelling (=mutation)?

How do we know that a word is misspelled?
2 millionen pages as this one
What is needed to read DNA?
And what to interpret

A sequencer
A computer scientist
What is needed to read DNA?
And what to interpret

A sequencer

A computer scientist
What is needed to read DNA?
And what to interpret

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A computer scientist
2-step analysis to find the genetic cause
First panel, then exome

Challenge: 20 000 genes, 700 misspellings of unknown significance
2-step analysis to find the genetic cause
First panel, then exome

Challenge: 20,000 genes, 700 misspellings of unknown significance
Not all genes play a role in muscle

→ First read 200 myopathy genes = panel
2-step analysis to find the genetic cause
First panel, then exome

Challenge: 20 000 genes, 700 misspellings of unknown significance
Not all genes play a role in muscle

→ First read 200 myopathy genes = panel

ABHD5 ACADVL ACTA1 ACTG1 ACTN1 ACTN2 ACVR1 ACYP2 AFG3L2 AGL AGRN AMPD1 ANO5
ASAH1 ATP2A1 B3GALNT2 BAG3 BGN BICD2 BIN1 CACNA1A CACNA1S CALM3 CAPN3 CASQ1
CAV3 CCDC78 CHKB CFL2 CHAT CHKB CHRNA1 CHRND CHRNE CHRNG CLCN1 CNBP CNTN1
COL6A1 COL6A2 COL6A3 COLQ CPT2 CRYAB DAG1 DES DMD DMPK DNAJB2 DNAJB5 DNAJB6
DNM2 DOK7 DPAGT1 DSE DYNC1H1 DYSF ELAC2 EMD ENO1 ENO3 EPG5 ERBB3 ETFA ETFB
ETFDH FHL1 FKBP14 FKRP FKTN FLNC GAA GBE1 GFPT1 GH1 GNE GYG1 GYS1 H6PD HINT1
HRAS HSPG2 ISCU ISPD ITGA7 KBTBD13 KCNA1 KCNE3 KCNJ10 KCNJ18 KIF1B KLHL40
KLHL41 KLHL9 KRT19 KRT8 LAMA1 LAMA2 LAMB2 LAMC1 LAMC2 LAMP2 LARGE LDB3 LDHA
LMOD3 LMNA LPIN1 MAGEE1 MAMLD1 MATR3 MEGF10 MGME1 MSTN MTM1 MTMR14 MUSK
MYBPC1 MYBPC2 MYBPC3 MYBPH MYF6 MYH1 MYH14 MYH2 MYH3 MYH7 MYH8 MYL1 MYOM2
MYOT NEB NOS1 OBSCN OBSL1 OPTN PABPN1 PFKM PGAM2 PGK1 PGM1 PGM5 PHKA1 PHKB
PLEC PLEKHG5 PNPLA2 POLG POMGNT1 POMK POMT1 POMT2 PRKAG2 PTRF PUS1 PYGM
RAPSN RYR1 SCN4A SECISBP2 SEPN1 SMCHD1 SGCA SGCB SGCD SGCE SGCG SGCGZ SIL1
SLC22A5 SLC25A20 SLC25A4 SLC33A1 SNTB1 SNTB2 SNTG1 SNTG2 SOS1 SPEG STAC3 STIM1
SVIL SYNE1 SYNE2 TAZ TCAP TIA1 TMEM43 TMEM5 TNNI2 TNNT1 TNNT3 TPM2 TPM3
TRAPPCC11 TRIM32 TRIM37 TRIM63 TRPV4 TTN TULP3 UBA1 UTRN VCP VRK1 YARS2 ZAK
2-step analysis to find the genetic cause

First panel, then exome

**Challenge:** 20 000 genes, 700 misspellings of unknown significance

Not all genes play a role in muscle

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CNM genes
Panel solves > 60% of cases
Still many patients without molecular diagnosis

There must be some unknown genes
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Next step: read all 20 000 genes (= exome)
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Next step: read all 20 000 genes (= exome)

Myocapture: research program on 1000 exomes

gene found, confirmation ongoing
gene found and confirmed

candidate gene

.....no candidate...
Panel solves > 60% of cases
Still many patients without molecular diagnosis

There must be some unknown genes

Next step: read all 20 000 genes (= exome)

Myocapture: research program on 1000 exomes

3 main reasons

- Approach doesn’t cover all genes
- Mutations in regulators regions (20%)
- .....no candidate...

47,7% candidate gene
22,2% gene found and confirmed
15,8% gene found, confirmation ongoing
14,3% gene found and confirmed ongoing
Panel solves > 60% of cases
Still many patients without molecular diagnosis

There must be some unknown genes

Next step: read all 20 000 genes (= exome)

**Myocapture:** research program on 1000 exomes

3 main reasons
- Approach doesn’t cover all genes
- Mutations in regulators regions (20%)
- Not enough family members

Only patient: 25% < patient + parents: 50% < patients + parents + others: 75%
Biopsy still needed?
Or is genetic testing sufficient?
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My very clear answer: it depends
Biopsy still needed?
Or is genetic testing sufficient?
My very clear answer: it depends

Case 1: patient with clinical presentation of CNM
→ panel sequencing (straightforward, rapid results)
→ mutation in MTM1

Solved - no biopsy required
Biopsy still needed?  
Or is genetic testing sufficient?  

My very clear answer: it depends

Case 1: patient with clinical presentation of CNM  
→ panel sequencing (straightforward, rapid results)  
→ mutation in MTM1  

Solved - no biopsy required

Case 2: patient with clinical presentation of CNM  
→ panel sequencing: no mutation in CNM genes  
→ exome sequencing: mutation in new gene  

Full characterization necessary - biopsy required  
(time-consuming experiments)

Can indicate novel therapeutic targets  
→ great help for many patients
Nuclei during muscle development

muscle cells → fusion → myotube → maturatio → muscle fiber
Nuclei during muscle development

muscle cells → fusion → myotube → maturatio → muscle fiber

section
Nuclei during muscle development

"myotubular myopathy"

muscle fibers look like myotubes before maturation
Nuclei during muscle development

- Muscle cells → fusion → myotube → maturation → muscle fiber

T-Tubules (MTM1, BIN1, DNM2 involved)

"Myotubular myopathy"
Muscle fibers look like myotubes before maturation

MTM1, BIN1, DNM2, RYR1, TTN involved in muscle structure

Structure determines function
Nuclei during muscle development

- **Muscle cells** → **Fusion** → **Myotube** → **Maturatio** → **Muscle fiber**

- **Calcium channel** (RYR1)

- **T-Tubules** (MTM1, BIN1, DNM2 involved)

- **Muscle fibers** look like myotubes before maturation

- **Calcium store**

- **“Myotubular myopathy”**

- **MTM1, BIN1, DNM2, RYR1, TTN** involved in muscle structure

- **Structure determines function**
Nuclei during muscle development

Muscle cells → fusion → myotube → maturation → muscle fiber

- Calcium channel (RYR1)
- Filaments (TTN involved)
- T-Tubules (MTM1, BIN1, DNM2 involved)
- Calcium store

“myotubular myopathy”
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- calcium channel (RYR1)

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Calcium store

“myotubular myopathy”

muscle fibers look like myotubes before maturation

MTM1, BIN1, DNM2, RYR1, TTN involved in muscle structure

Structure determines function
850 MTM/ZNM-Patienten weltweit
Worldwide 850 MTM/CNM patients

30 in TTN (2013)

→ In vielen Ländern vermutlich unterdiagnostiziert

175 Patienten ohne Molekulardiagnose

• Keine adäquate genetische Beratung
  No appropriate genetic counseling
• Keine Pränatal-Diagnose
  No prenatal diagnosis
• Keine Prognose
  No prognosis
• Keine Entwicklung von Therapien
  No development of therapies