

Hur ser muskler med kontrakturer hos barn med cerebral pares ut på insidan?

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Enheten för Neuropediatrik
Institutionen för Kvinnor och Barns Hälsa
Karolinska Institutet

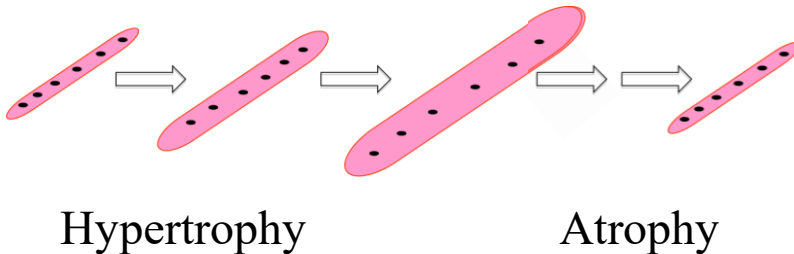
Agenda

- Kort intro om skelettmuskulatur organisation
- Muskulaturen vid Cerebral pares
 - Hur ser muskeln ut i mikroskopet
 - Vad vet vi om molekylära processer

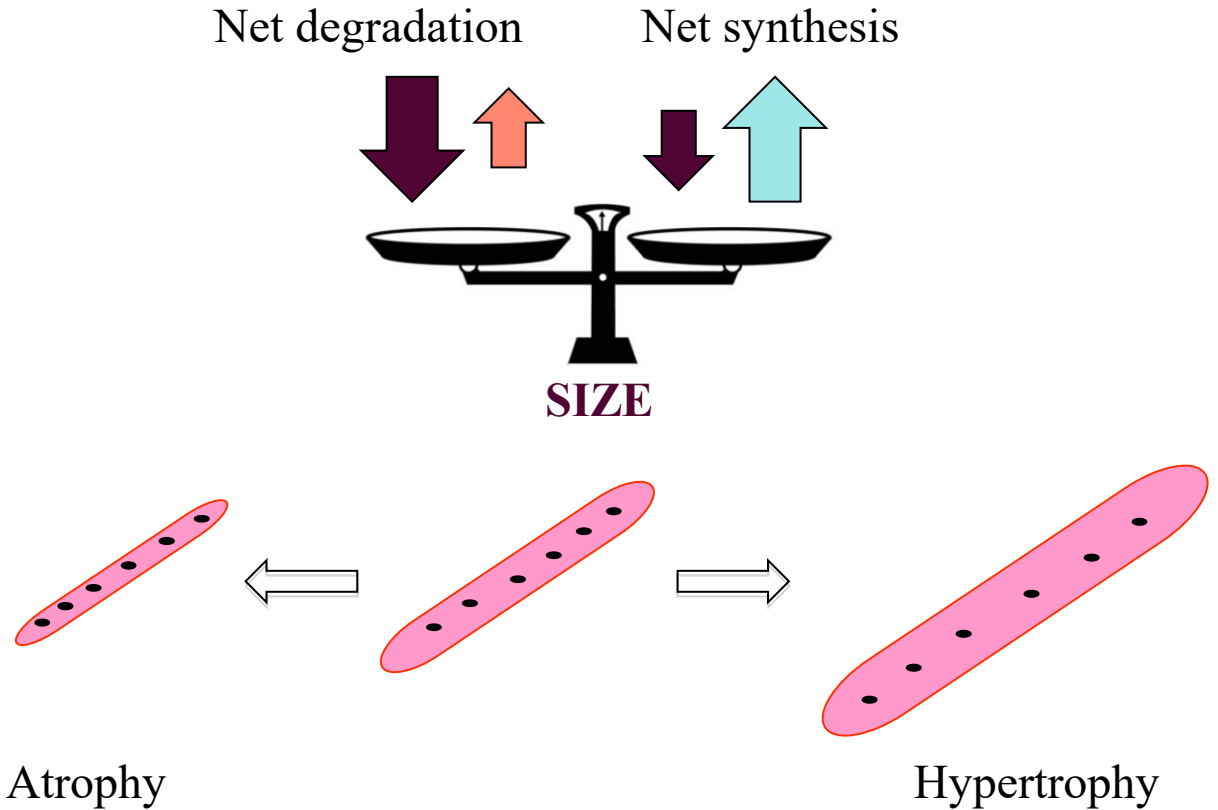
Ferdinand von Walden

Skeletal muscle

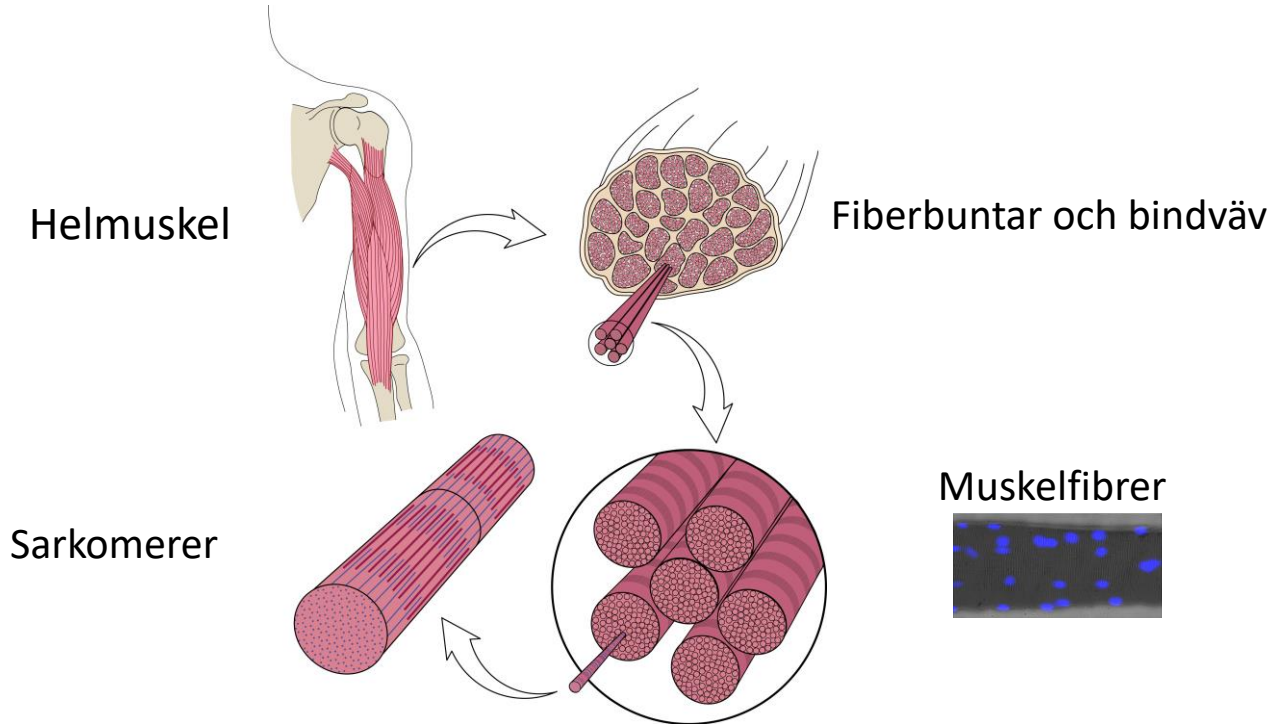
- Roughly 40% of body mass
- Important for:
 - Locomotion
 - Proper glucose homeostasis
 - Protein repository
- Responsive to increased – decreased use



Protein turnover in skeletal muscle dictates muscle mass



Skelettmuskelnns struktur

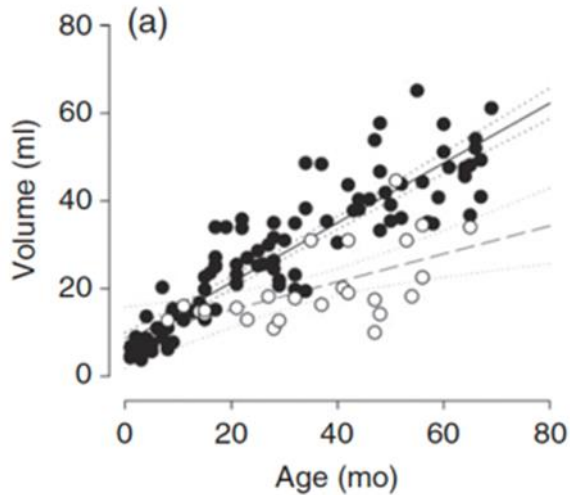


Vad utmärker musklerna hos barn med CP?

- Tunna och styva
- Kontrakturer är mycket vanligt
- Varför kontrakturer uppstår är okänt
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- Färre ribosomer (proteinfabrik)
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- Mer bindväv
- Färre mitokondrier (kraftverk)

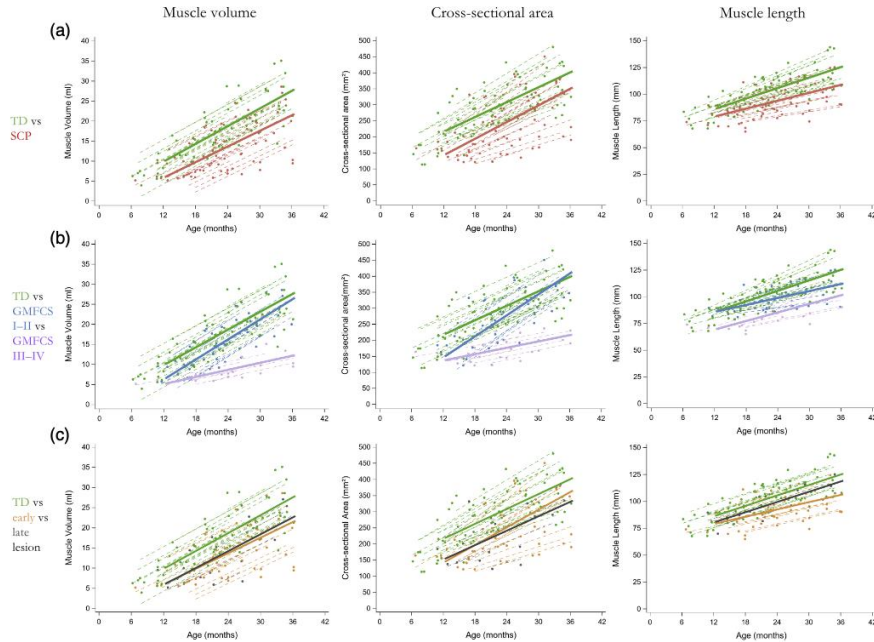


Muskerna växer inte som förväntat



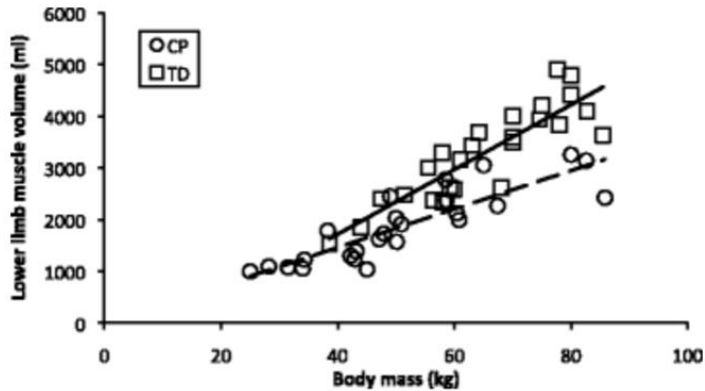
Herskind et al 2015.
Dev Med

Musklerna växer inte som förväntat



De Beukelar et al
2023. Dev Med

Musklerna växer inte som förväntat



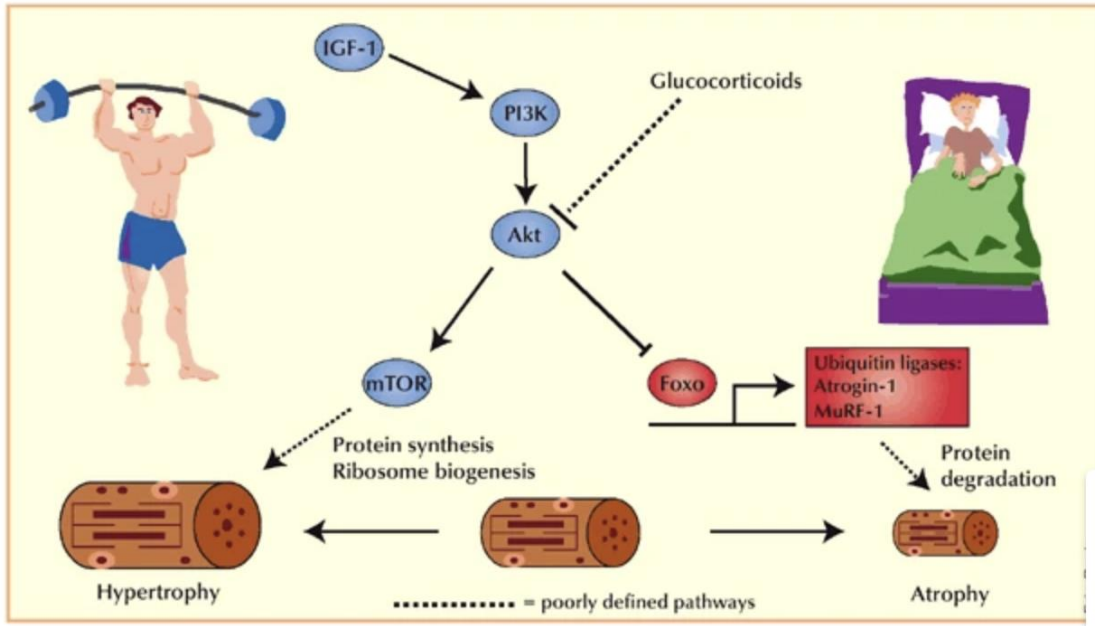
Noble et al 2017.
BMC Neurol

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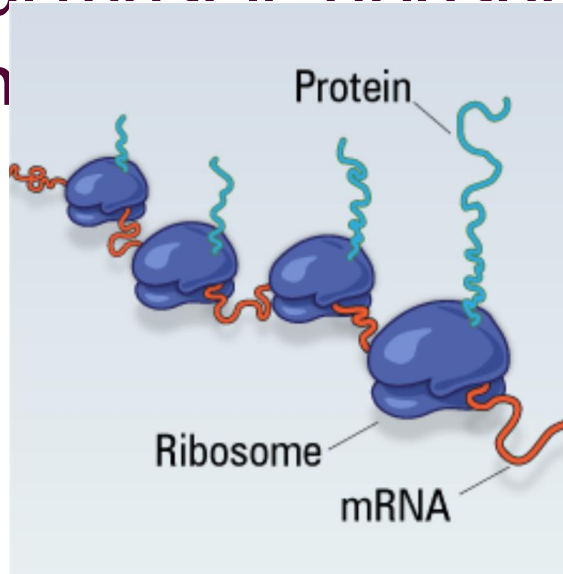


Påverkan på



Ribosomer och muskelns stamceller

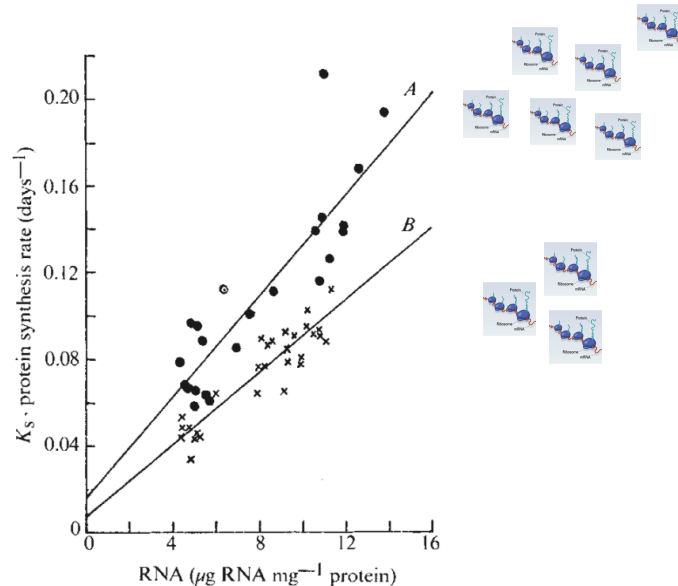
Protein synthesis rate depends on skeletal muscle ribosome content



www.broadinstitute.com

Ribosomal RNA (rRNA) approx. 80% of total RNA

Protein synthesis rate depends on skeletal muscle ribosome content

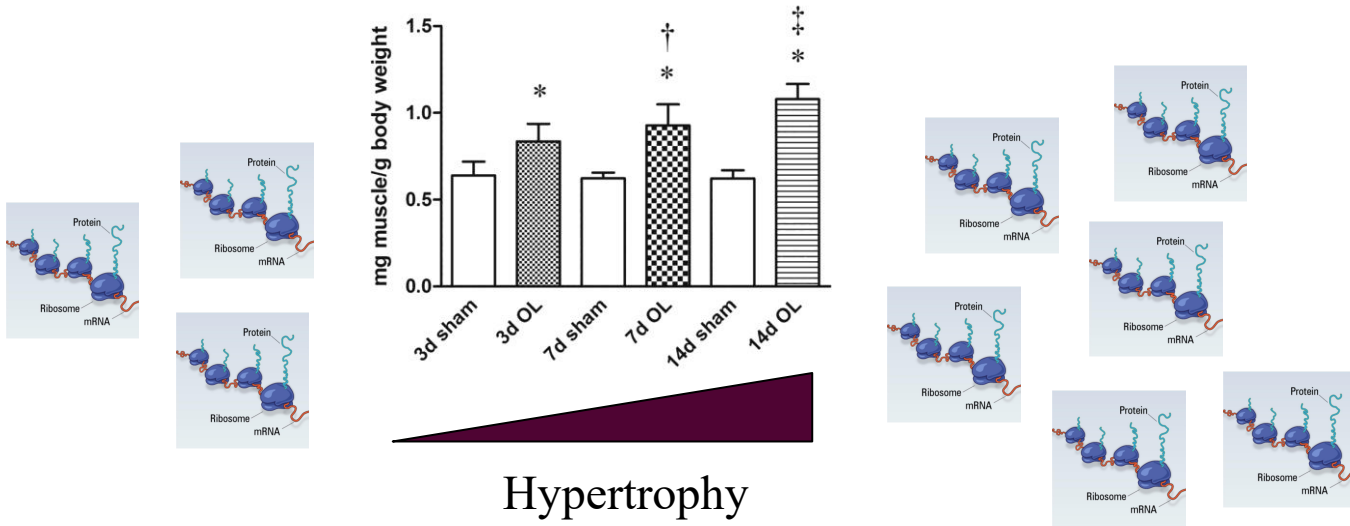


Millward et al. Nature 1973

Synergist ablation procedure – rodent model of skeletal muscle hypertro

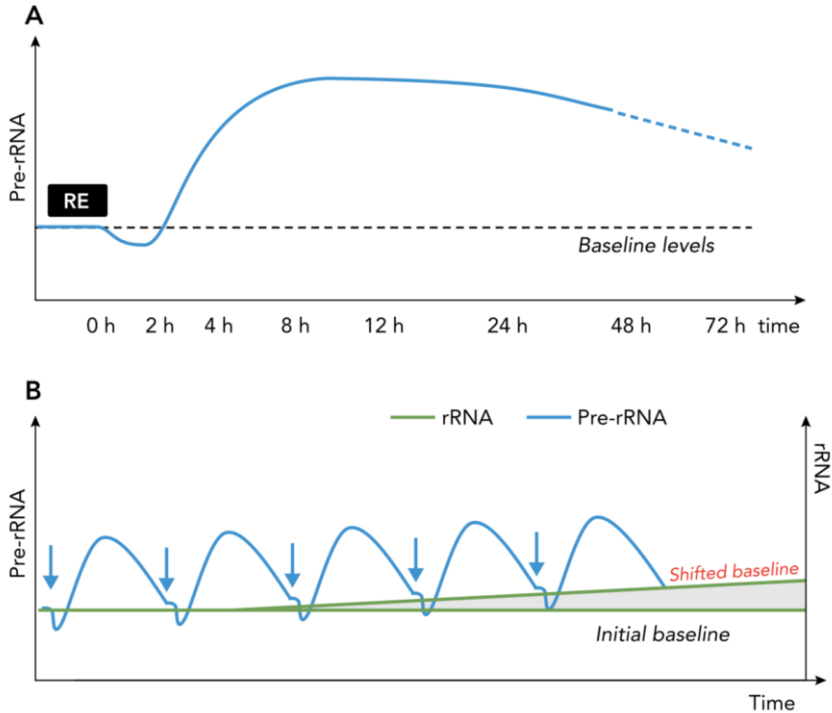


Ribosome production starts early during the growth process



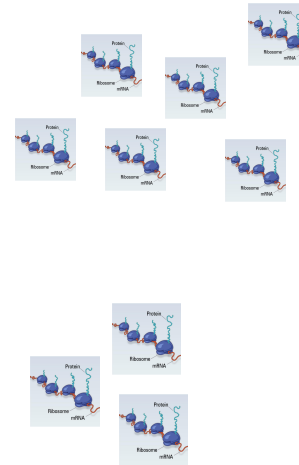
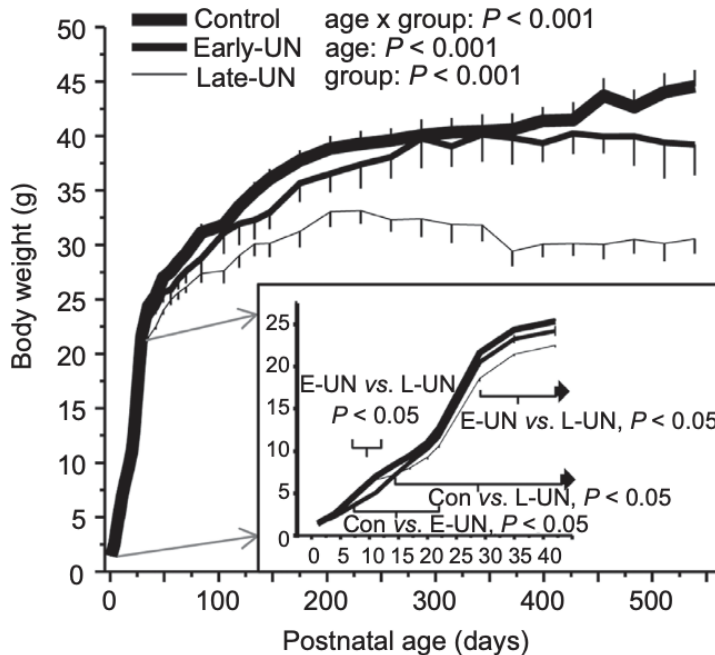
Von Walden et al 2012, Am J Physiol

Protein synthesis rate depends on skeletal muscle ribosome content



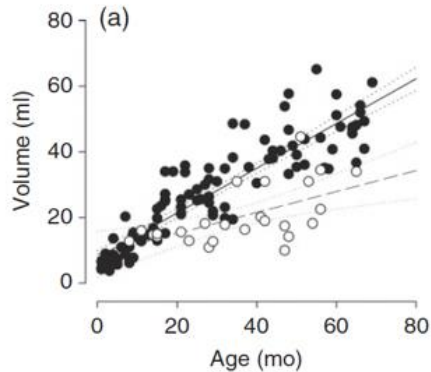
Figueiredo & McCarthy, *Physiology* 2019

Malnutrition during early life lowers adult body weight

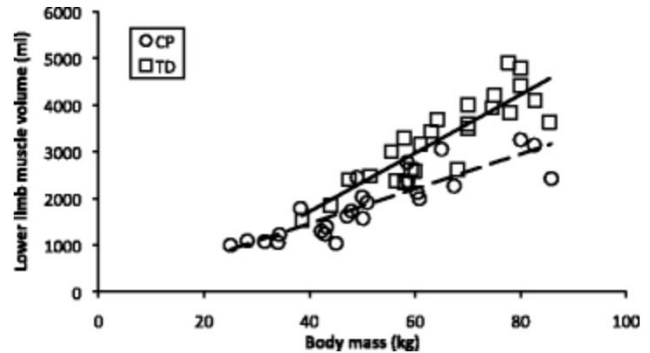


Ribosome content dictates the ability for the muscle to catch up

Are there too few ribosomes in CP muscle?

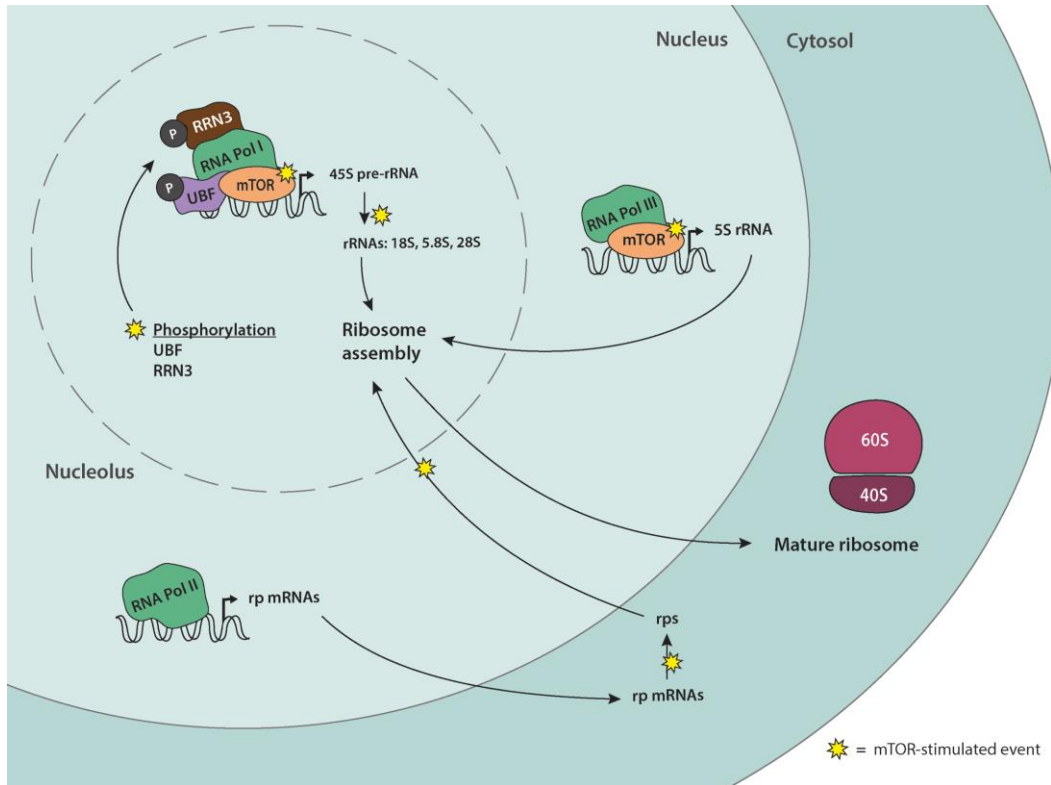


Herskind et al 2015. Dev Med

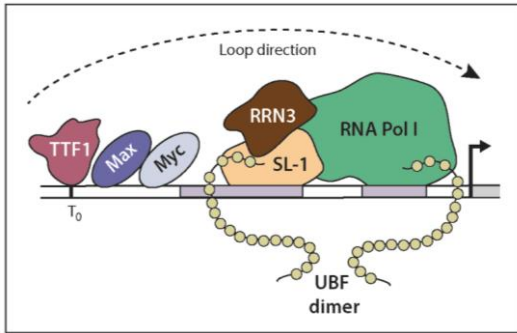


Noble et al 2017. BMC Neurol

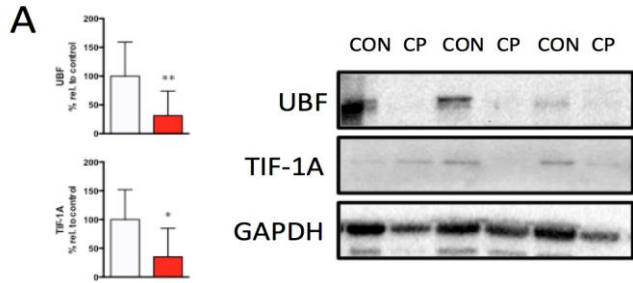
Ribosome biogenesis – the process of making new ribosomes



Suppressed ribosome biogenesis in skeletal muscle of children with cerebral palsy

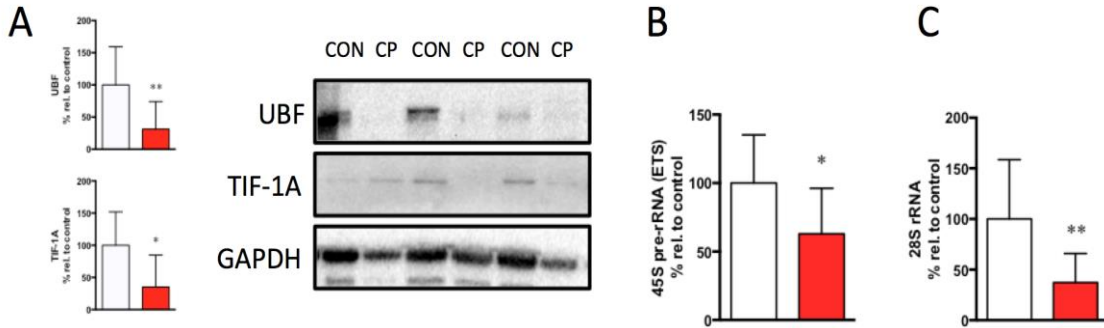


Pre-initiation complex at the rDNA promoter



Skeletal muscle ribosome biogenesis is impaired in cerebral palsy (A) Protein levels of transcription factors UBF and TIF-1A are significantly reduced in cerebral palsy muscle (red bars) as compared to healthy controls (white bars).

Suppressed ribosome biogenesis in skeletal muscle of children with cerebral palsy



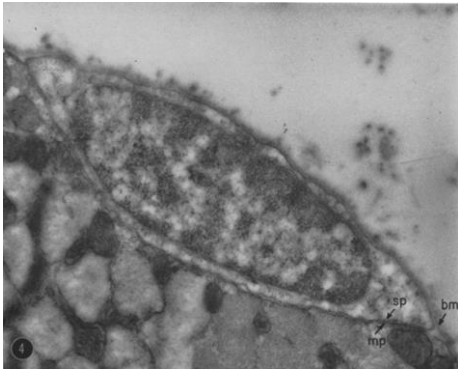
Skeletal muscle ribosome biogenesis is impaired in cerebral palsy (A) Levels of UBF and TIF-1A (B) Abundance of 45S pre-rRNA and (C) 28S rRNA levels in cerebral palsy muscle (red bars) as compared to healthy controls (white bars).

Vad utmärker musklerna hos barn med CP?

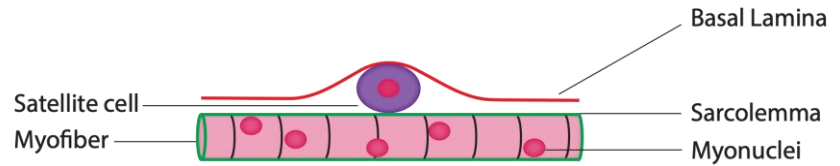
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Muscle Stem Cells

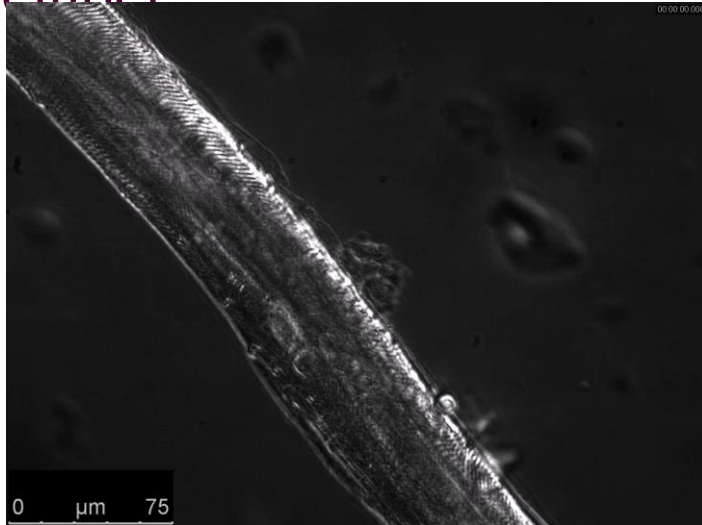


Mauro 1961



Egner et al 2016 Dayanidhi and Lieber 2014

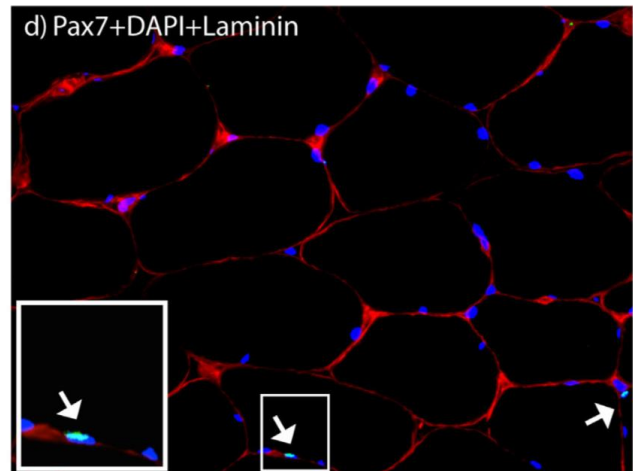
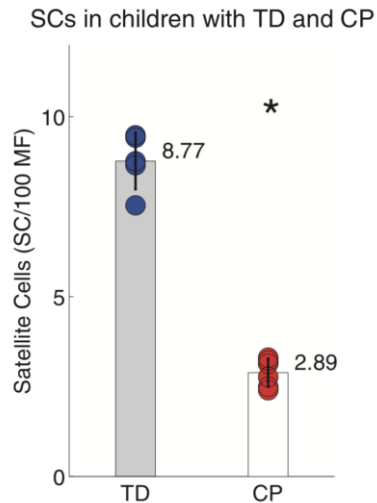
Aktiverad satellitcell på en muskelfiber



Fewer satellite cells in CP

Reduced Satellite Cell Number In Situ in Muscular Contractures From Children With Cerebral Palsy

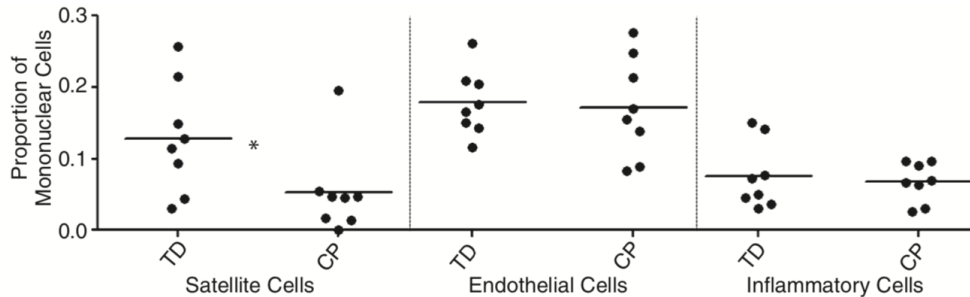
Sudarshan Dayanidhi,^{1,2} Peter B. Dykstra,¹ Vera Lyubasyuk,¹ Bryon R. McKay,³ Henry G. Chambers,^{1,4} Richard L. Lieber^{1,2,5}



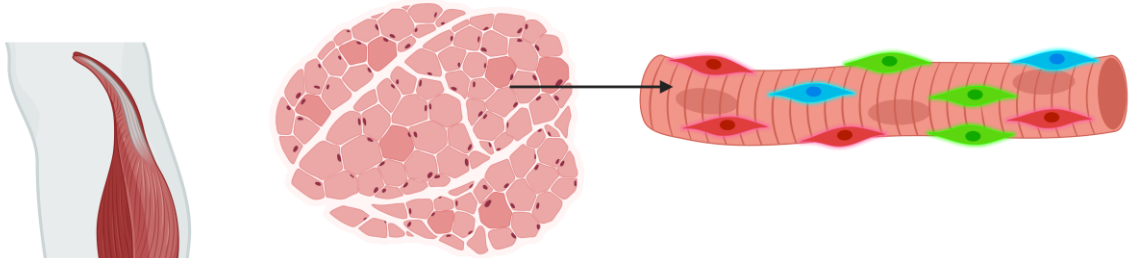
Fewer satellite cells in CP

Reduced satellite cell population may lead to contractures in children with cerebral palsy

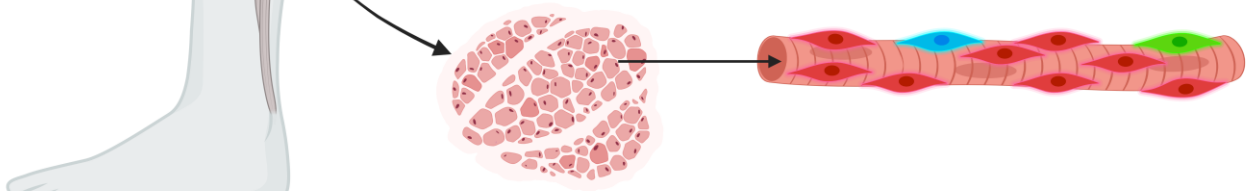
LUCAS R SMITH¹ | HENRY G CHAMBERS^{2,3} | RICHARD L LIEBER^{1,2}



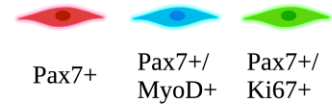
CP Non-Contractured Muscle



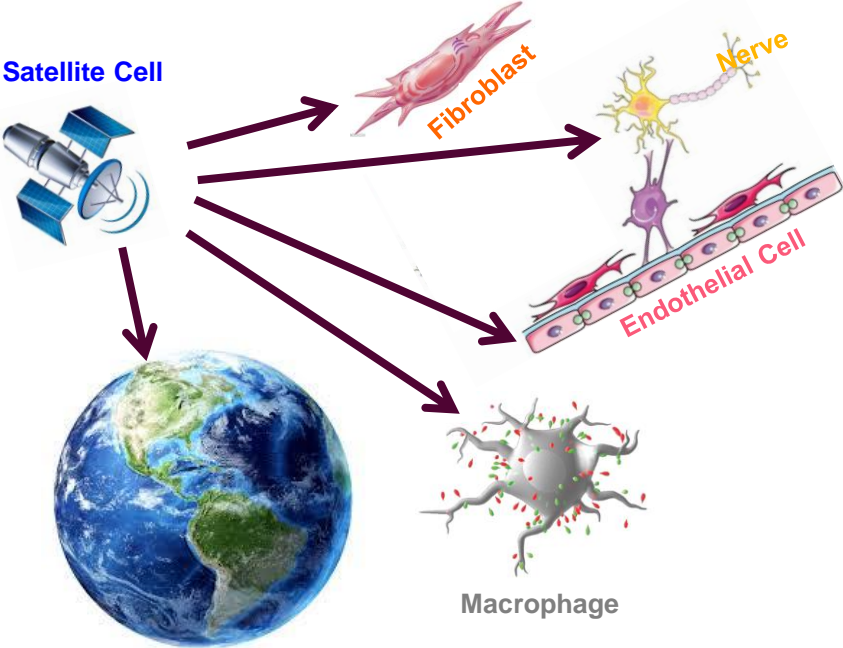
Resident muscle stem cells (MuSC) in muscle contractures have reduced myogenic characteristics



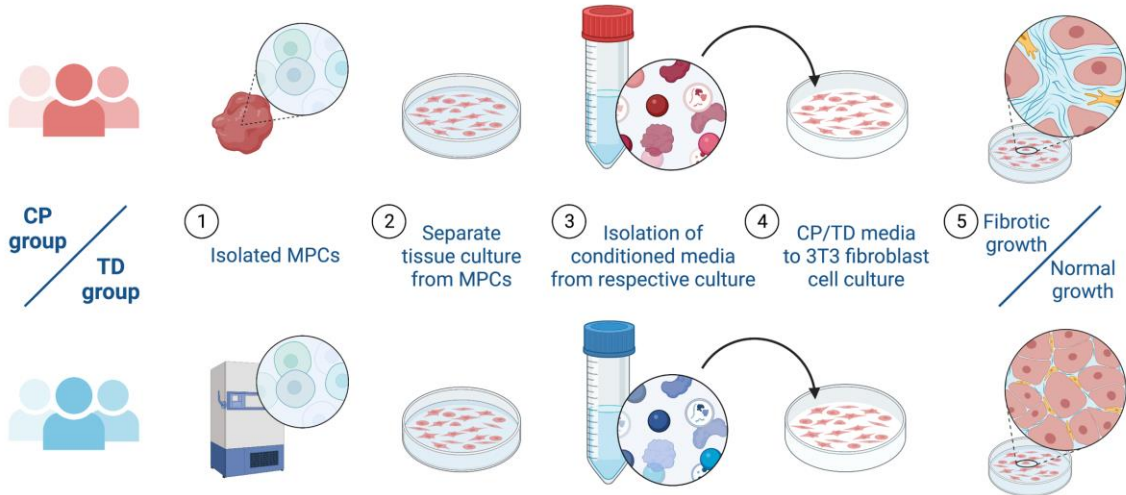
CP Contractured Muscle



“Satellite” Cell – A Name With A Deeper Meaning?



Pågående projekt för att se hur stamcellerna påverkar kringliggande celler

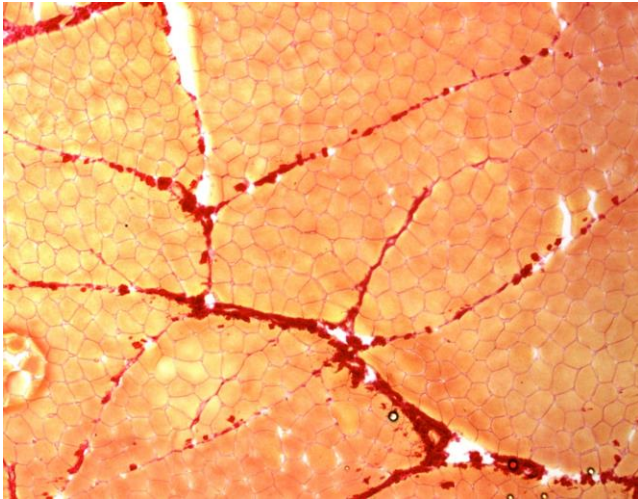


Vad utmärker musklerna hos barn med CP?

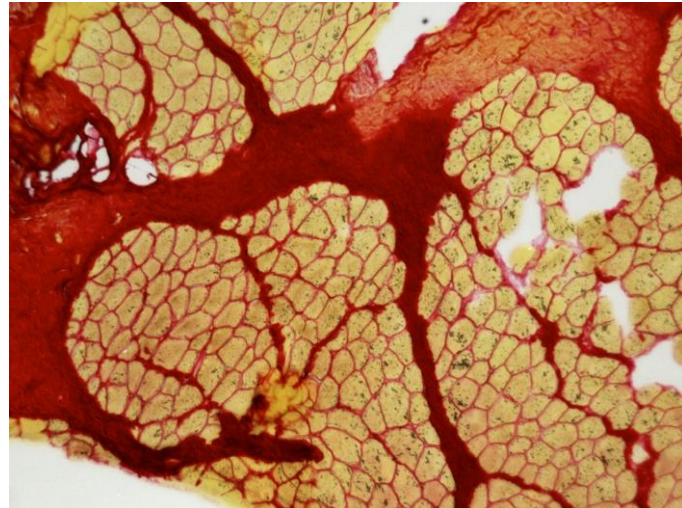
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Thickening of the perimysium is common



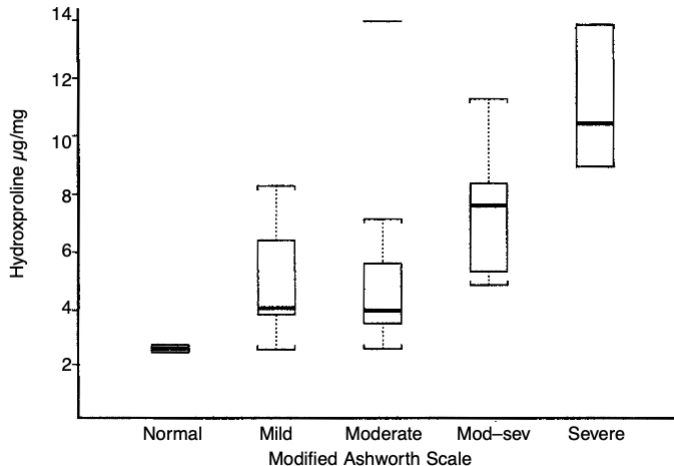
Typically developed



Cerebral palsy

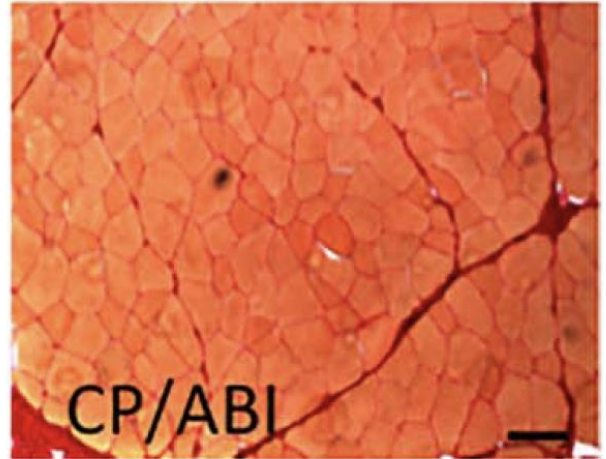
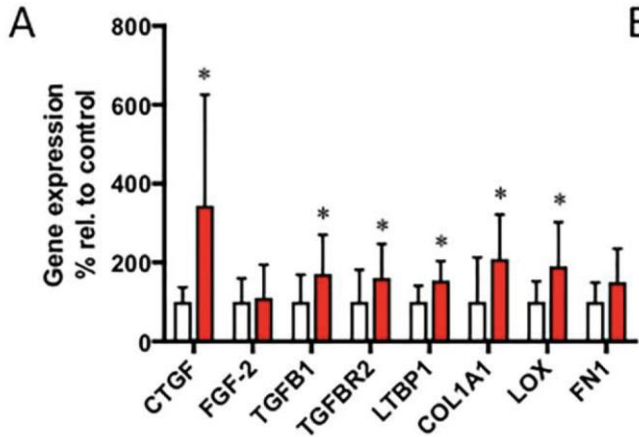
Von Walden et al 2018. Muscle and Nerve

Higher collagen content in skeletal muscle with increased severity of Cerebral Palsy



Booth et al 2001. Dev Med Child Neur.

Fibrogenic gene expression and increased collagen content in CP muscle

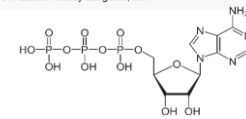
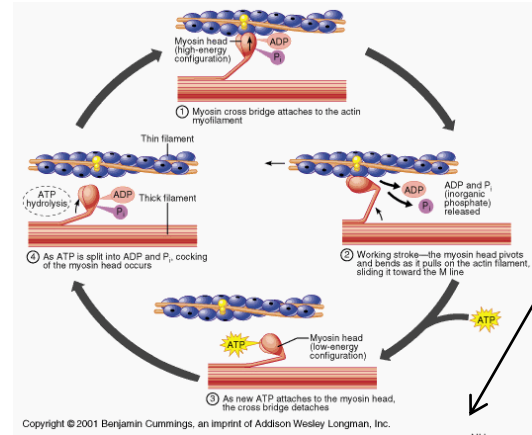
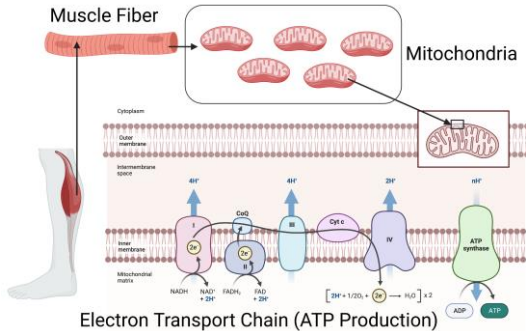
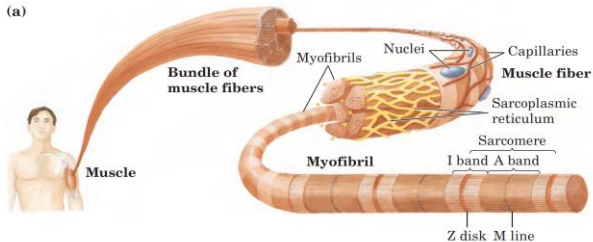


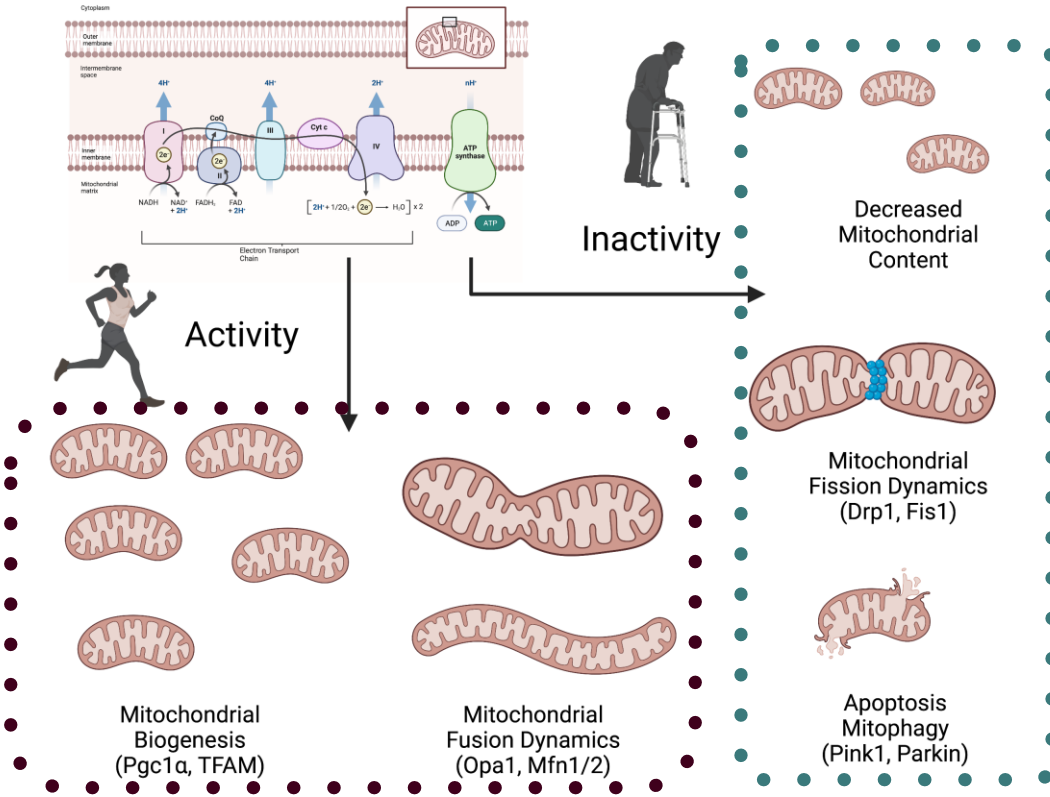
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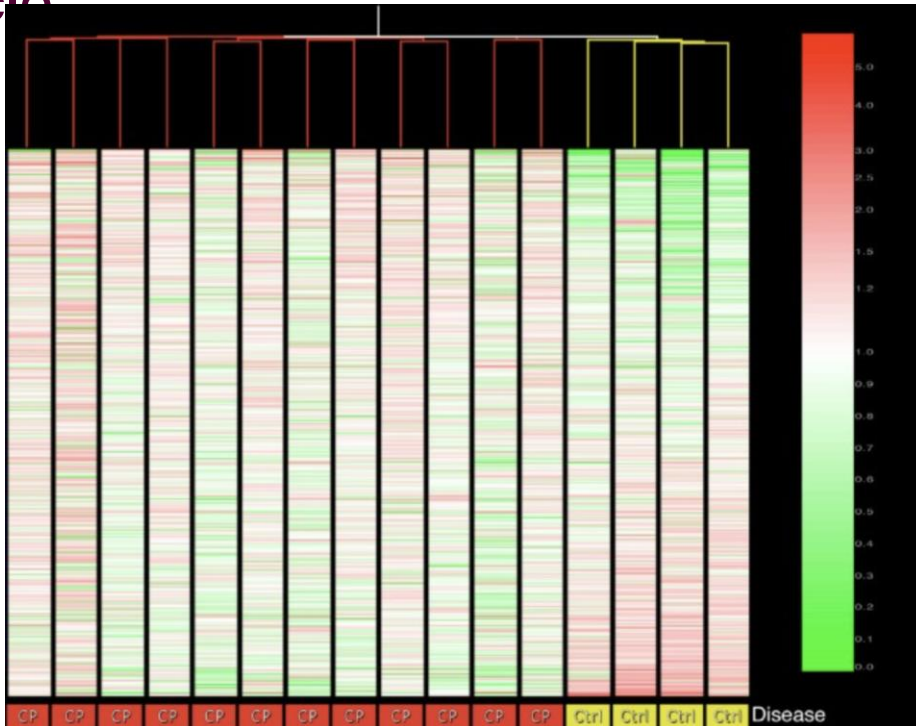


Peripheral component – the muscle





Global gene expression is altered in CP skeletal muscle






Smith, Pontén et al 2009. *BMC Genomics*

Mitochondria and Skeletal Muscles in Children with CP

DEVELOPMENTAL MEDICINE & CHILD NEUROLOGY

ORIGINAL ARTICLE

Skeletal muscle maximal mitochondrial activity in ambulatory children with cerebral palsy

SUDARSHAN DAYANIDHI^{1,2,3}  | ELISA H BUCKNER^{1,4} | ROBIN S REDMOND³ | HENRY G CHAMBERS^{1,5}  | SIMON SCHENK^{1,4} | RICHARD L LIEBER^{1,2,3,6} 

DEVELOPMENTAL MEDICINE & CHILD NEUROLOGY

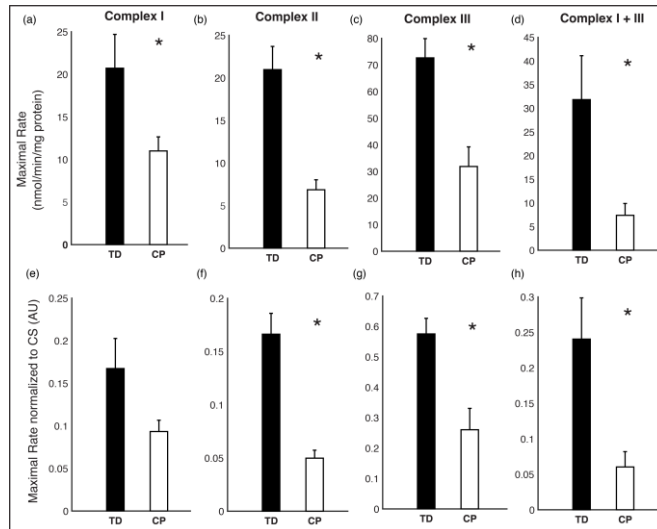
ORIGINAL ARTICLE

Reduced mitochondrial DNA and OXPHOS protein content in skeletal muscle of children with cerebral palsy

FERDINAND VON WALDEN^{1,2,3,*}  | IVAN J VECHETTI JR^{2,3,4,*}  | DAVIS ENGLUND^{3,5}  | VANDRÉ C FIGUEIREDO^{2,3}  | RODRIGO FERNANDEZ-GONZALO^{6,7}  | KEVIN MURACH^{3,5}  | JESSICA PINGEL⁸  | JOHN J MCCARTHY^{2,3} | PER STÅL⁹  | EVA PONTÉN¹ 

EC Editor's
Choice

Mitochondrial Activity Assays in Leg Muscles



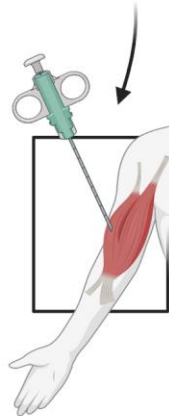
Mitochondrial activity in ambulatory children with CP are lower

Reduced expression of key factors in mitochondrial

k Typically Developing Child's Muscle



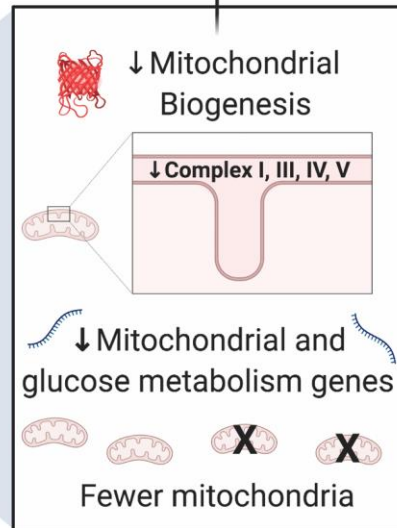
Cerebral Palsy Child's Muscle



In biopsy samples

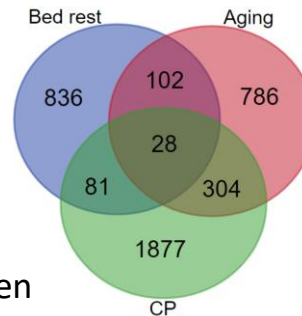
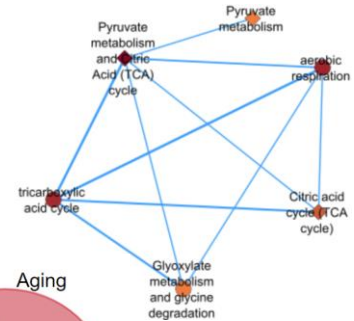
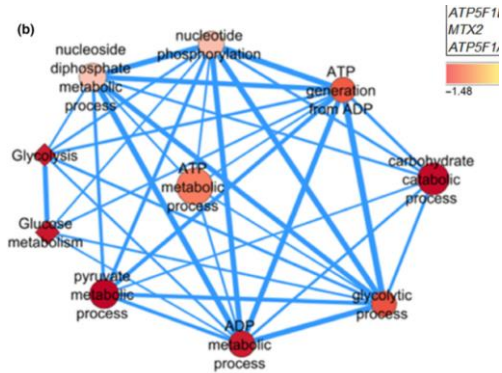


Comparative analysis:
Similar to aging and prolonged bed rest



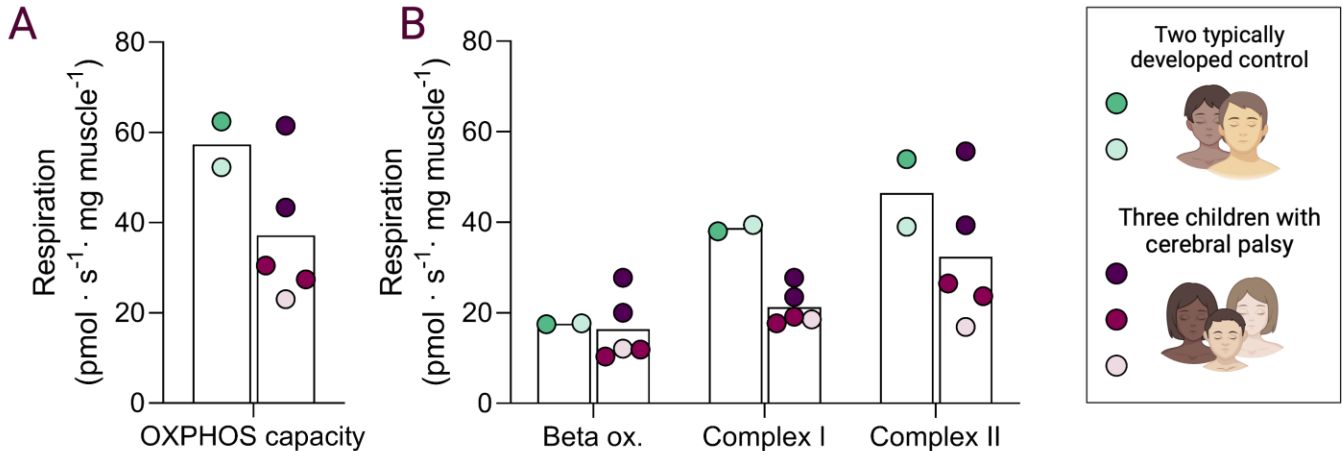
Versus Typically Developing

Metabolic Gene Networks in Leg Muscles



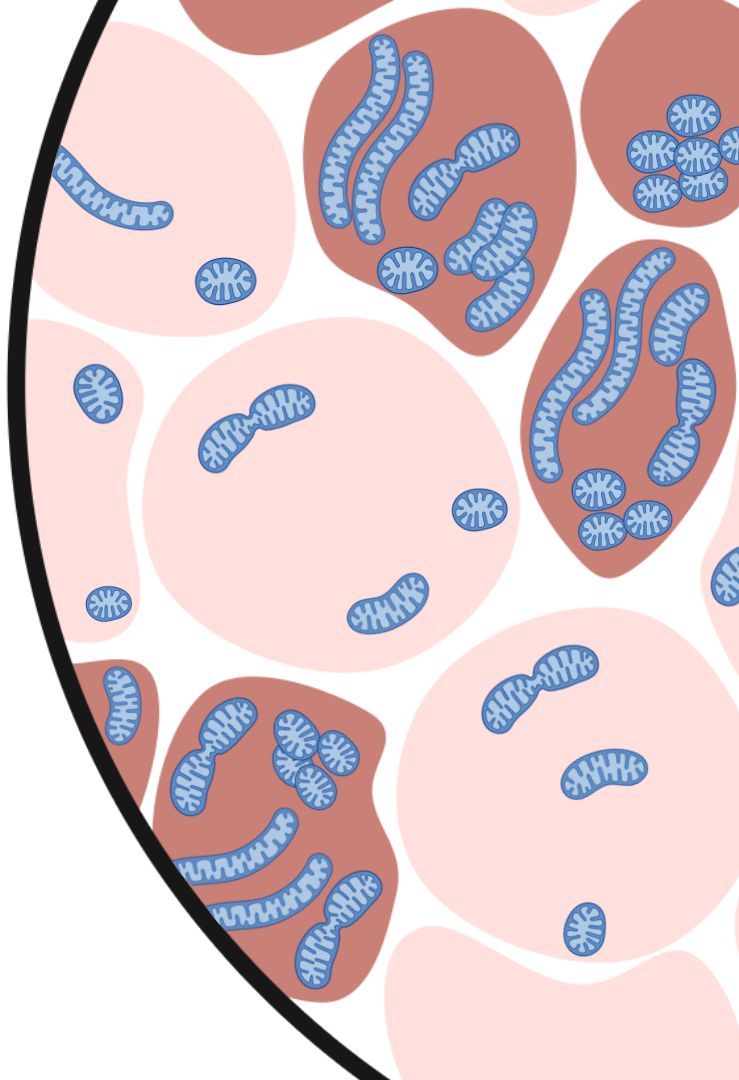
- Metabolic gene networks are downregulated
- Shared commonalities between Aging, Bed Rest and CP

Mitochondrial respiration in skeletal muscle

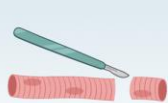


 **SLOW-TWITCH**

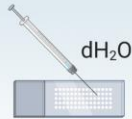
 **FAST-TWITCH**



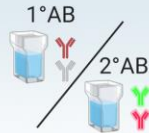
THRIFTY - high THROUGHput Immunofluorescence Fibre TYping



① Cut off 0.5 mm segment



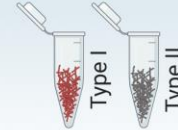
② Mount fibre segment on slide



③ Fluorescent antibody staining



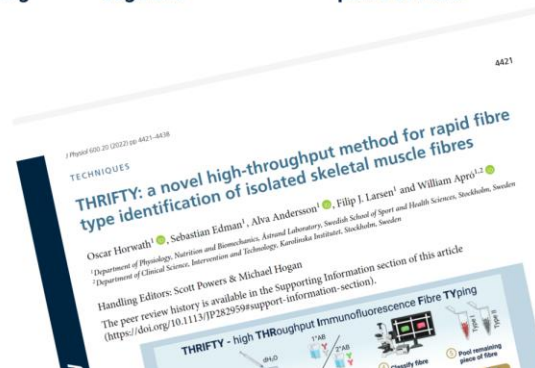
④ Classify fibre segment



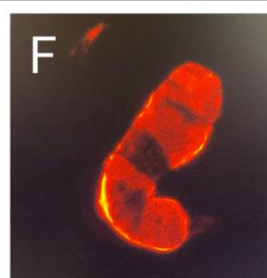
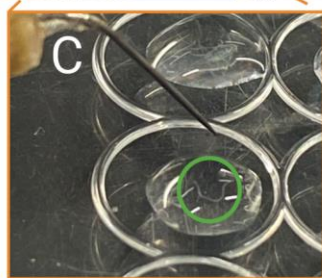
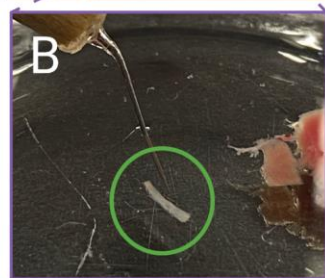
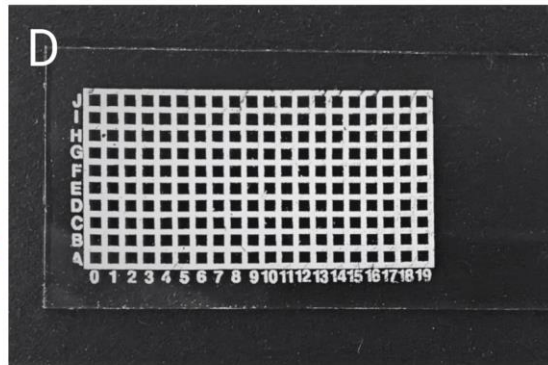
⑤ Pool remaining piece of fibre

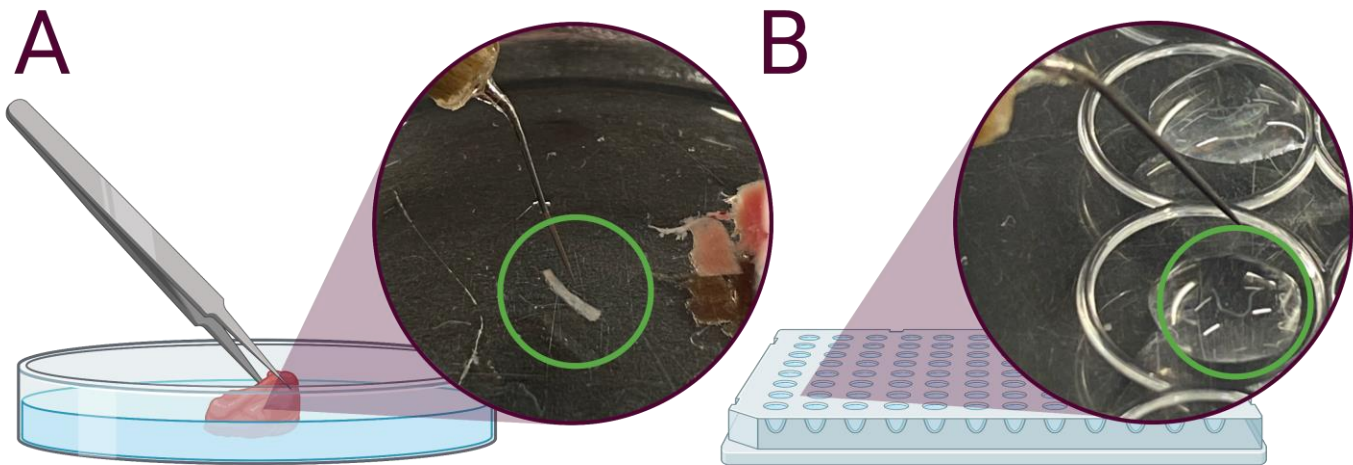


Sebastian Edman, new postdoc

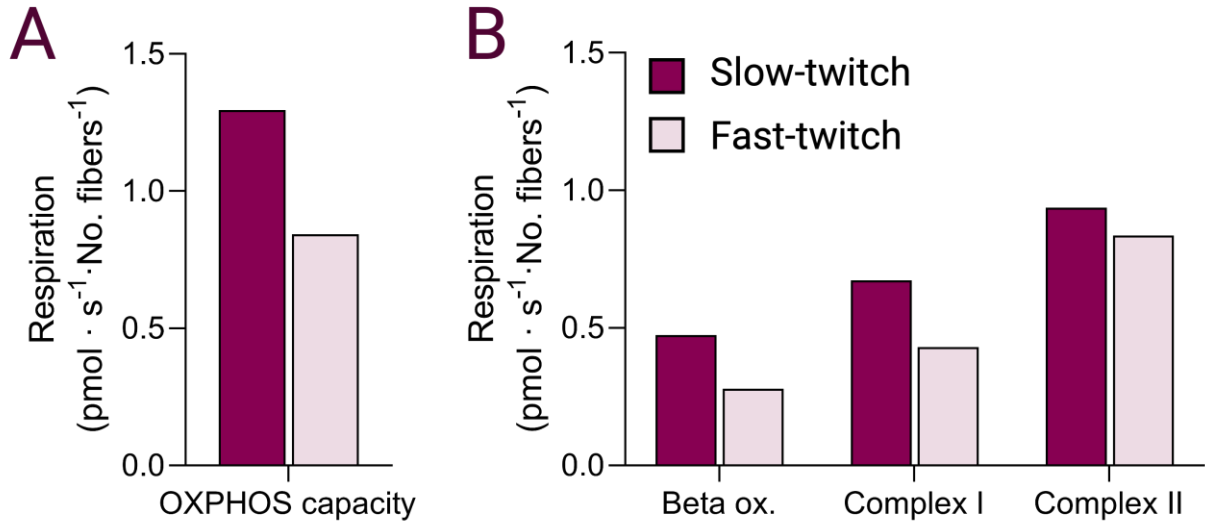


23 oktober 2023





Proof of concept data



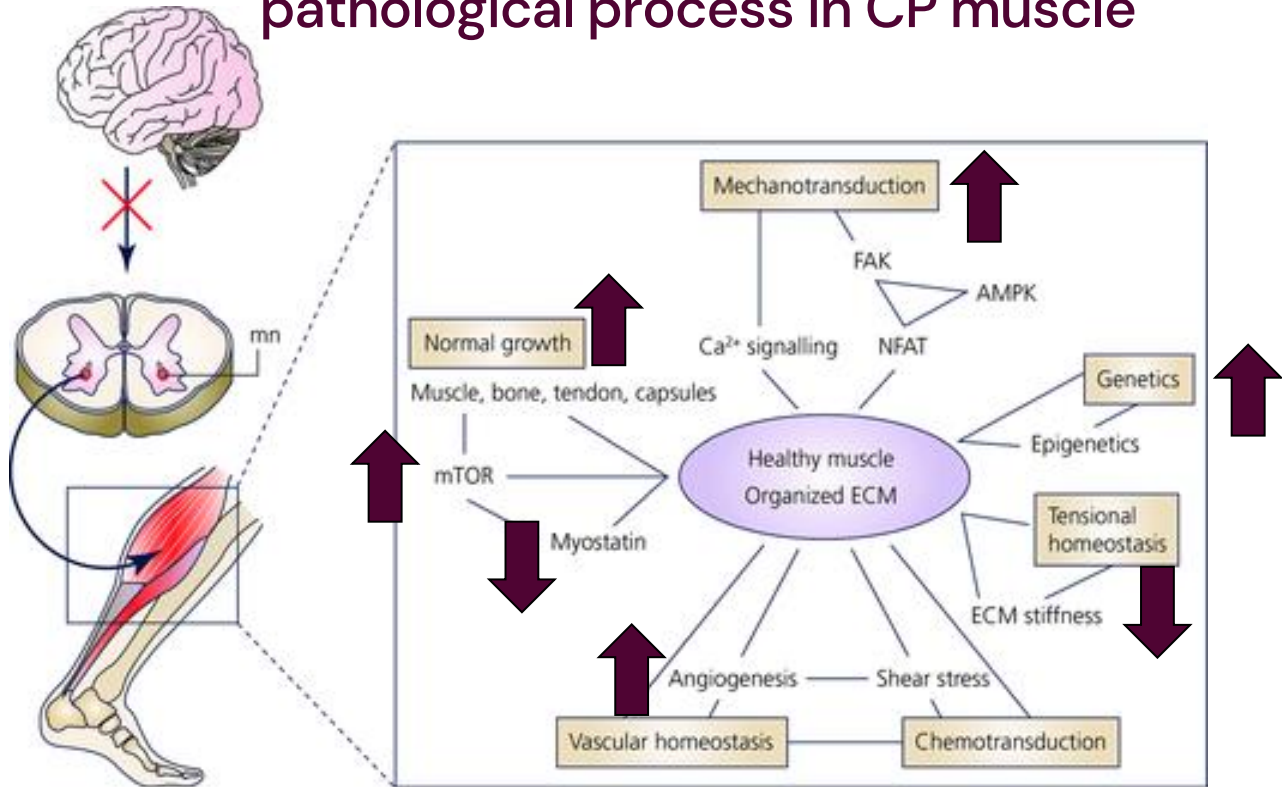
Repetition

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Physical activity likely counteracts pathological process in CP muscle



Acknowledgement

Team von Walden

Linnea Corell, PhD-student
Minying Cui, PhD-student
Sebastian Edman, PhD, Postdoc
Baptiste Jude, PhD, Postdoc
Paulo Jannig, PhD, Postdoc
Ola Kvist, MD, PhD, Postdoc

Alumni team members

Emma Hjalmarsson, PT, PhD

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Professor
Johanna Lanner, PhD, Associate
Professor
Rodrigo Fernandez-Gonzalo, PhD,
Associate Professor
Jessica Norrbom, PhD, Senior
researcher

VUMC, Amsterdam, the Netherlands

AnneMieke Buizer, MD, PhD, Professor
Petra van Schie, PhD
Arnoud Edelman Bos, PhD, student



Norrbacka-Eugeniastiftelsen

Stiftelsen Sunnerdahls
Handikappfond



CARLSSONS STIFTELSE



Vetenskapsrådet

