The role of endoplasmic reticulum-mitochondria crosstalk in axonal maintenance

Roman Chrast

The RE(ACT) Congress / Bologna

8th of March, 2018

Karolinska Institutet

ÉCOLE POLYTECHNIQUE FÉDÉRALE DE LAUSANNE

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Dr. Nathalie Bernard-Marissal
(U1251, Aix Marseille Université)

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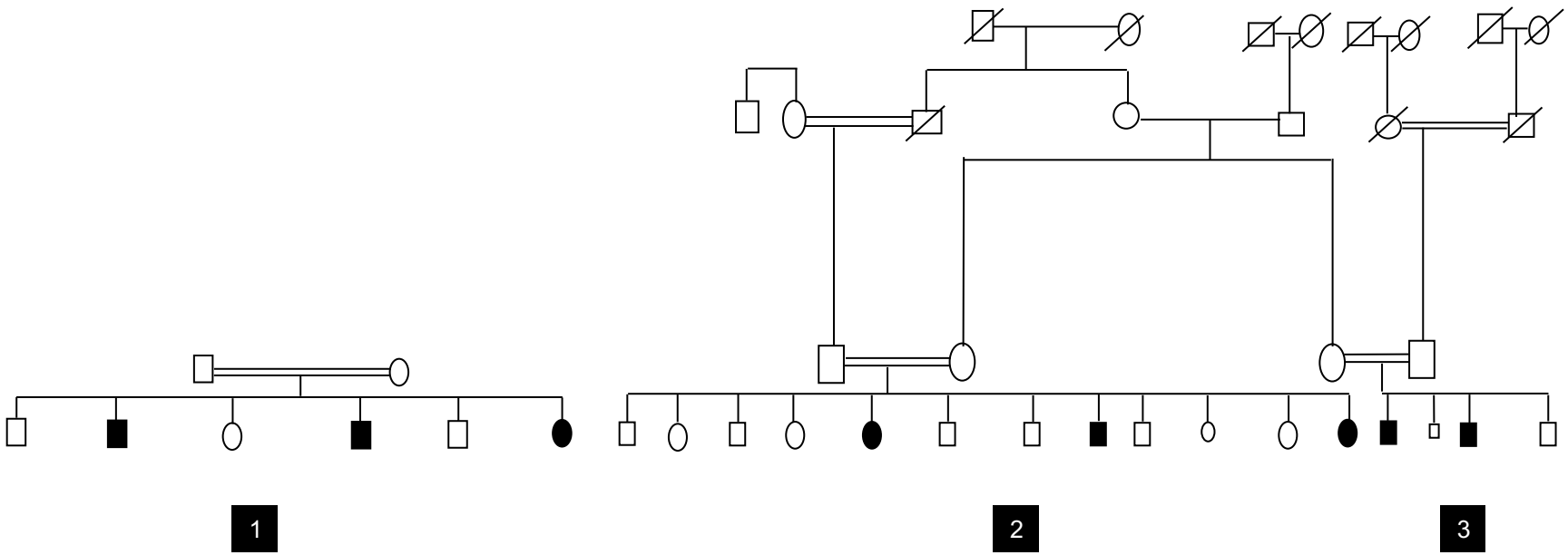
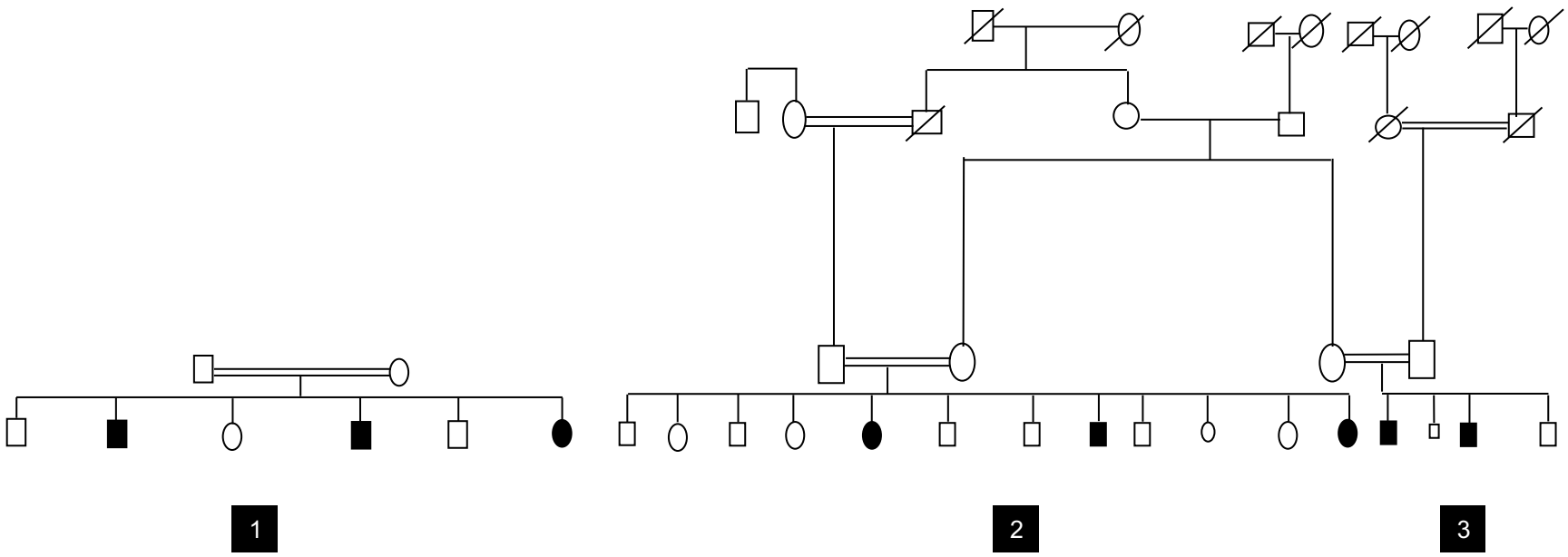
3 families affected by a hereditary motor neuropathy



Onset 2nd-3rd decade
Progressive muscle wasting and weakness in distal upper and lower limbs
Normal sensory examination

Dr. Hamid Azzedine

Loss of SIGMAR1 function in 3 families affected by a hereditary motor neuropathy



Homozygosity mapping & sequencing

SIGMAR1

Dr. Hamid Azzedine

Sigmar1 mutations and motor neuron disease

Sigma Nonopioid Intracellular Receptor 1 Mutations Cause Frontotemporal Lobar Degeneration–Motor Neuron Disease

Agnes A. Luty, BSc,[1–3]* John B.J. Kwok, PhD,[1–3]* Carol Dobson-Stone, PhD,[1–3]* Clement T. Loy, MD,[1–3] Kirsten G. Coupland, BSc,[1] Helena Karlström, PhD,[4] Tomasz Sobow, MD,[5] Joanna Tchorzewska, MD,[5] Aleksandra Maruszak, BSc,[6] Maria Barcikowska, MD,[6] Peter K. Panegyres, MD,[7,8] Cezary Zekanowski, PhD,[6] William S. Brooks, MD,[1,2] Kelly L. Williams, BSc,[9] Ian P. Blair, PhD,[9,10] Karen A. Mather, PhD,[11] Perminder S. Sachdev, MD,[11,12] Glenda M. Halliday, PhD,[1,2] and Peter R. Schofield, PhD, DSc[1–3]

ANN NEUROL 2010;68:639–649

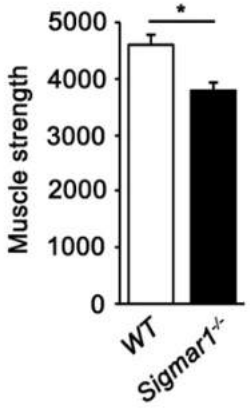
A Mutation in Sigma-1 Receptor Causes Juvenile Amyotrophic Lateral Sclerosis

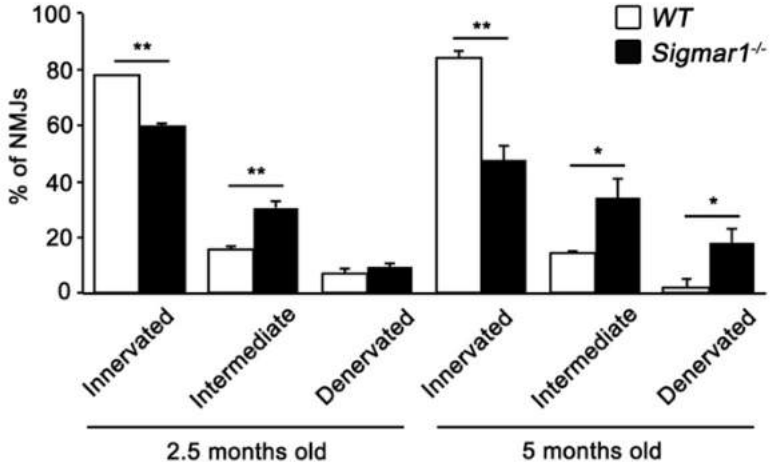
Amr Al-Saif, MD,[1] Futwan Al-Mohanna, PhD,[2] and Saeed Bohlega, MD[3]

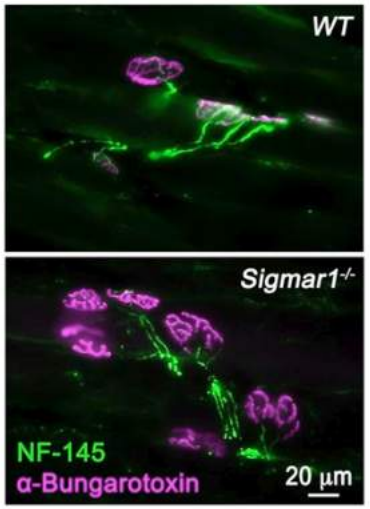
ANN NEUROL 2011;70:913–919

SIGMAR1 deficiency leads to motor neuron degeneration *in vivo*

Tibialis anterior - fast-twitch muscle
(No change in soleus - slow-twitch muscle)







Bernard-Marissal et al., Brain 2015

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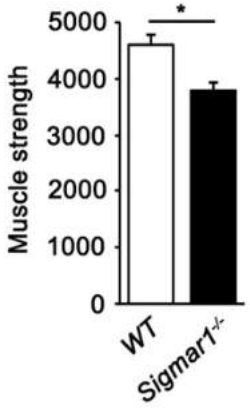
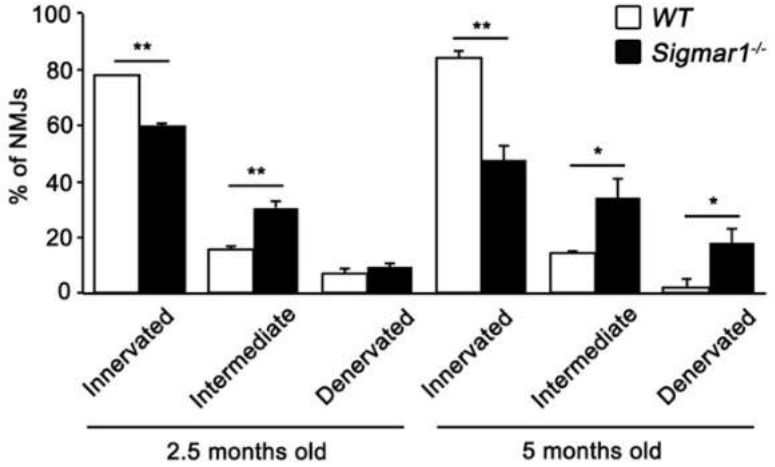
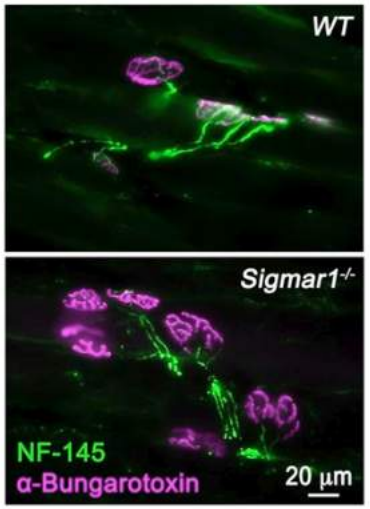
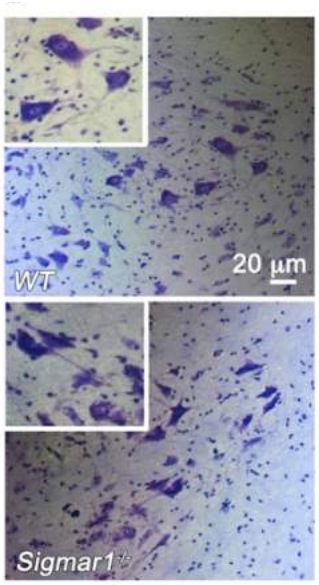
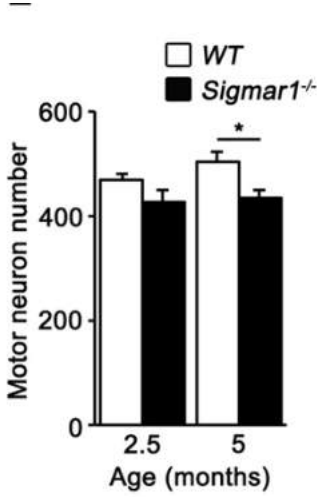
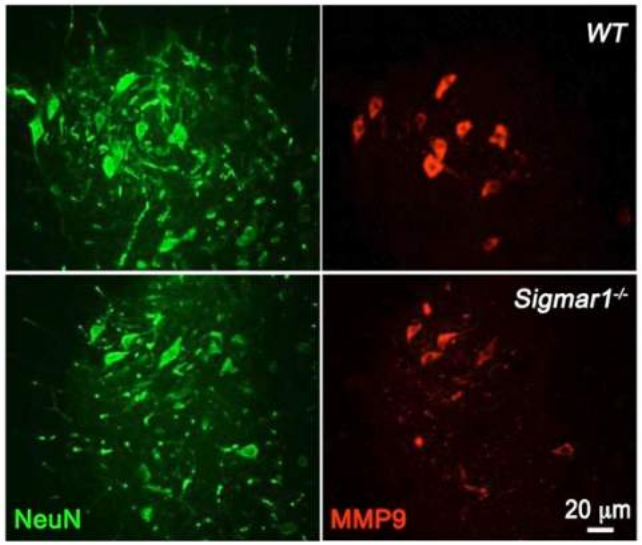




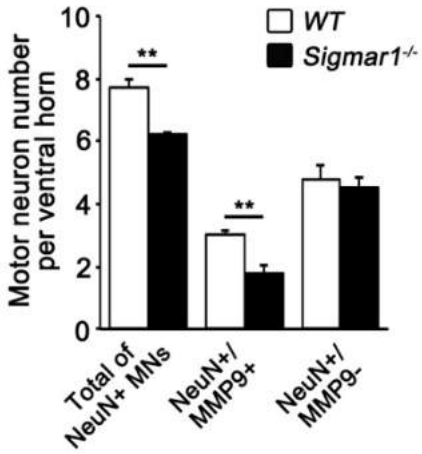




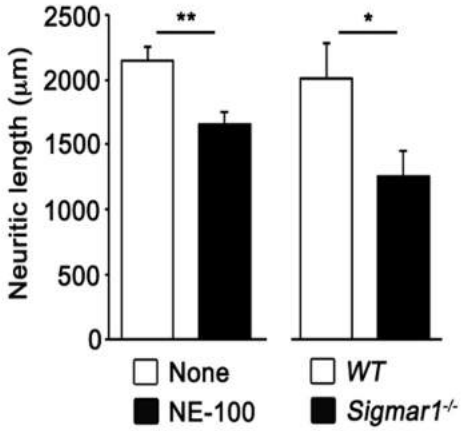


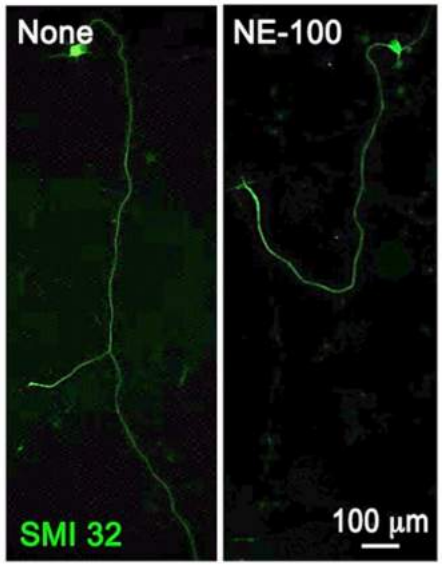


MMP9: fast vulnerable motor neurons



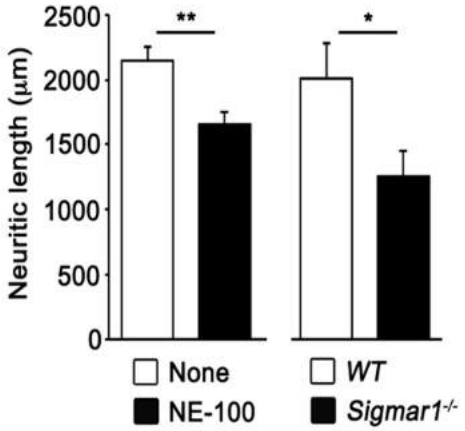
SIGMAR1 deficiency leads to motor neuron degeneration *in vitro*

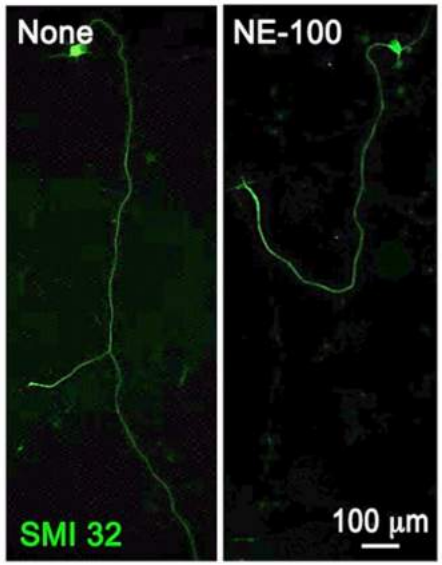


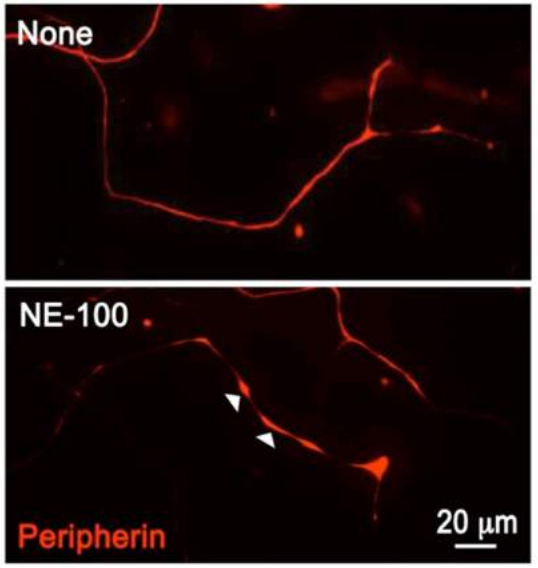


NE-100: SIGMAR1 antagonist

SIGMAR1 deficiency leads to motor neuron degeneration *in vitro*

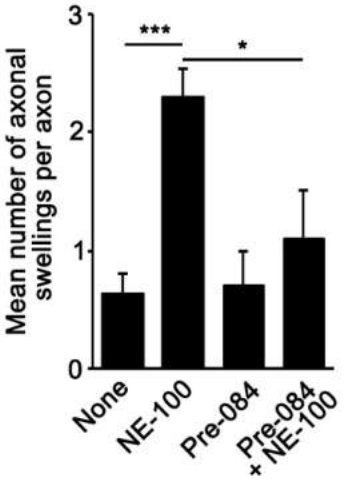


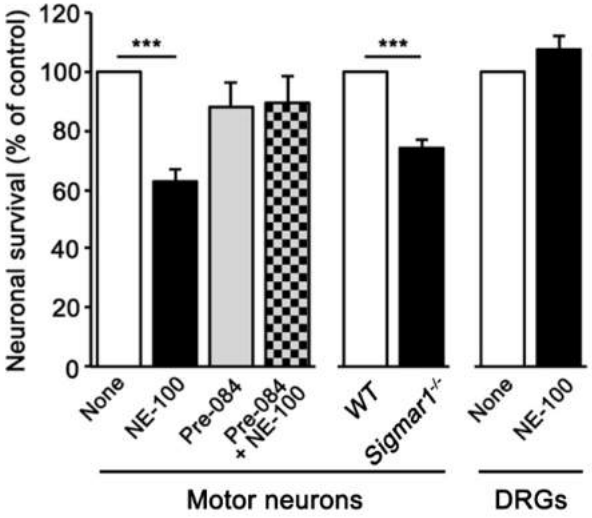




NE-100: SIGMAR1 antagonist
Pre-084: and SIGMAR1 agonist

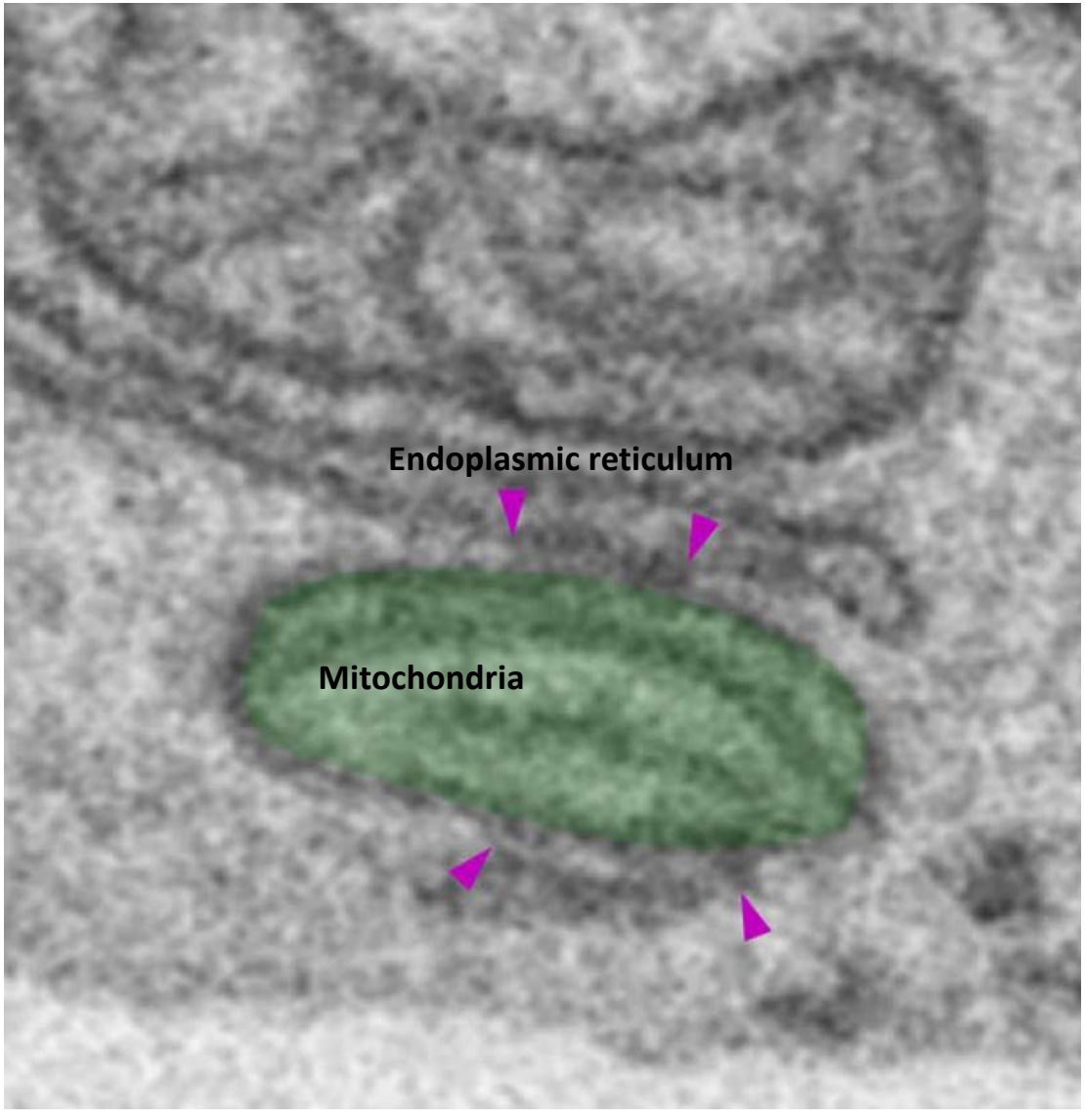
Peripherin: axonal swellings and spheroids





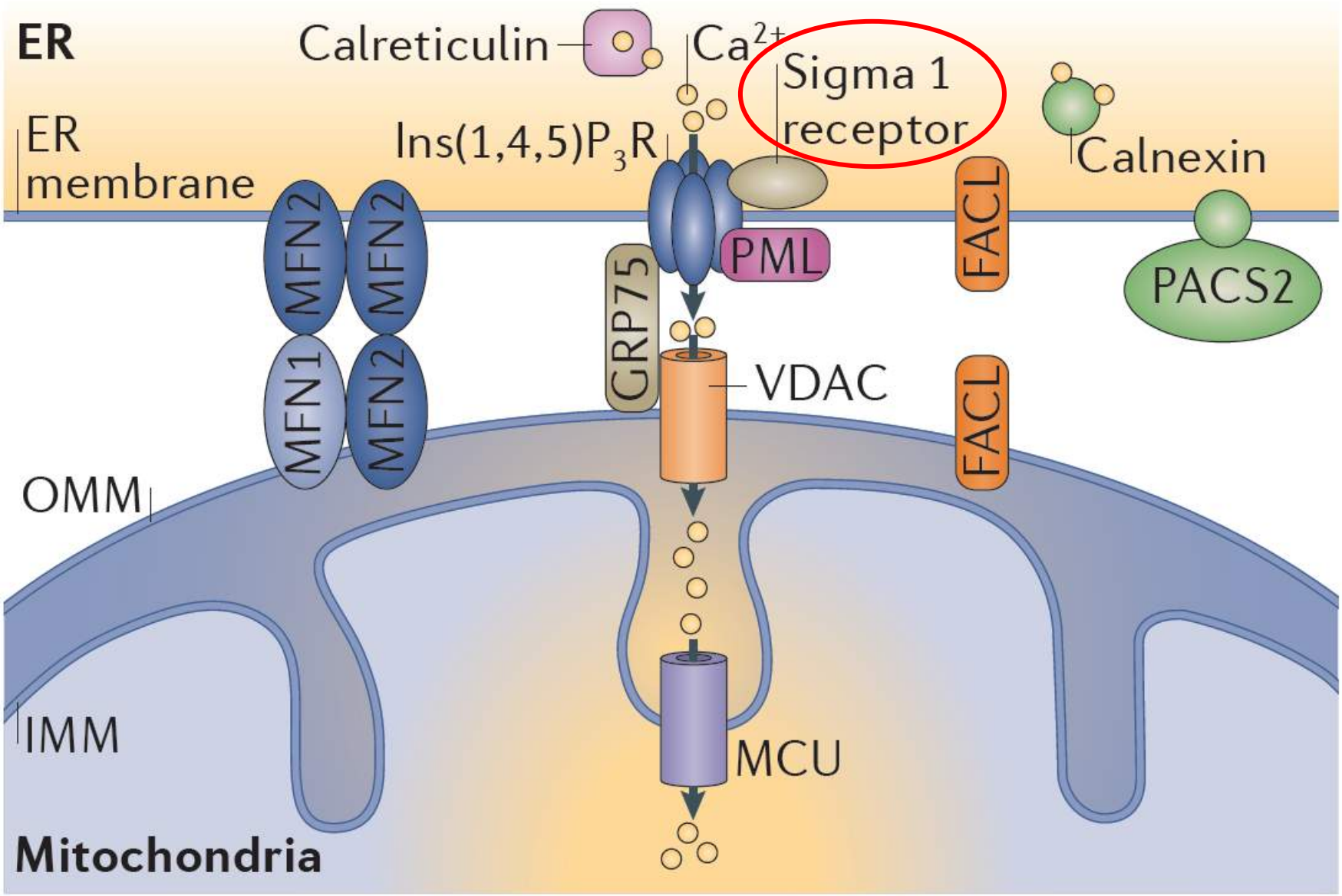
SIGMAR1 and mitochondria-associated membranes (MAMs)

In Chinese Hamster Ovary (CHO) cells SIGMAR1 is localized at the outer membrane of the ER at contact points between ER and mitochondria called mitochondria-associated membranes - MAMs *(Hayashi and Su, 2007)*



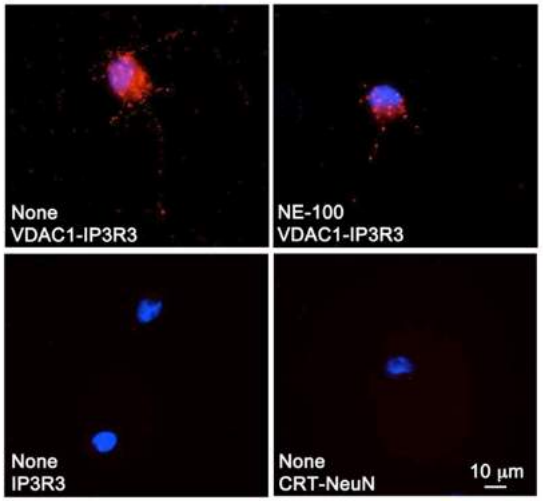
Krols et al., Acta Neuropathol. 2016

Mitochondria-associated membranes (MAMs)



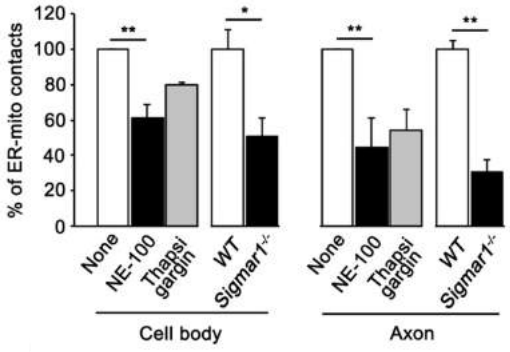
Rizzuto et al., Nat. Rev. Mol. Cell Biol. 2012

Inhibition and loss of SIGMAR1 function reduce contacts between mitochondria and ER in MNs



anti-ITPR3: ER side
anti-VDAC1: mitochondrial side

Thapsigargin: ER calcium depletor – induces ER stress



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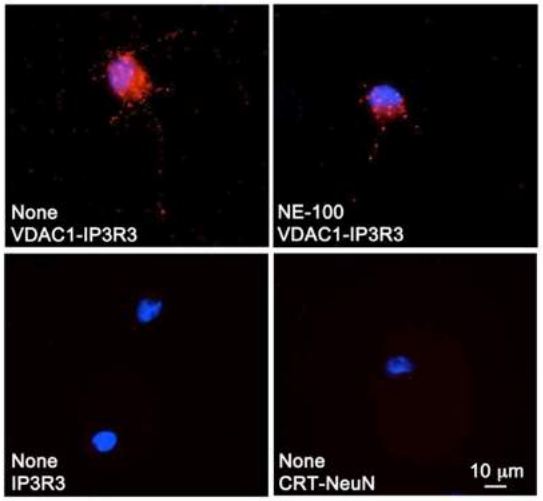

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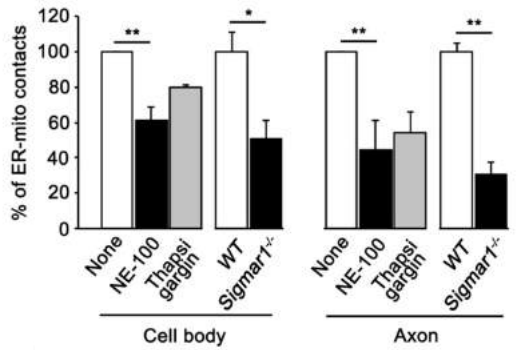
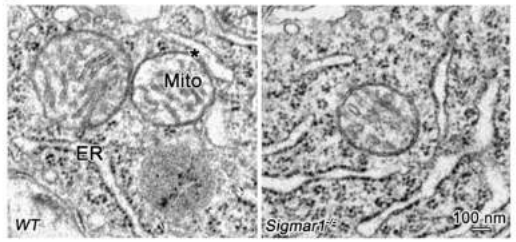
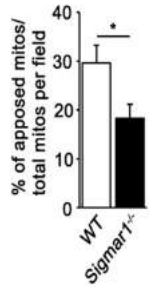
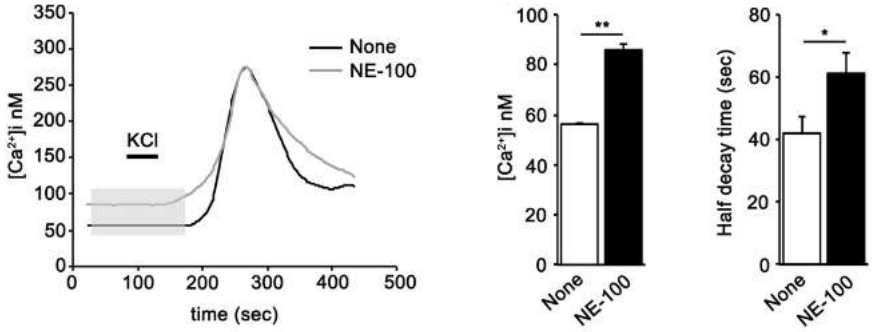






Calcium signaling and ER stress are involved in motor neuron degeneration induced by SIGMAR1 deficiency

At the level of MAMs, ER transfers calcium towards the mitochondria and this is necessary for its metabolic function (Rizzuto et al., 2012)



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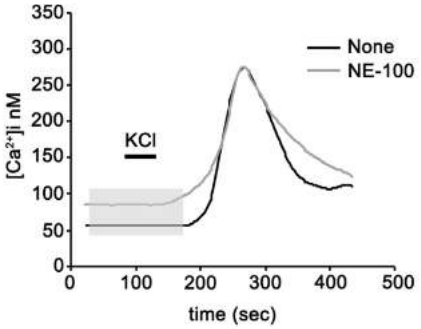
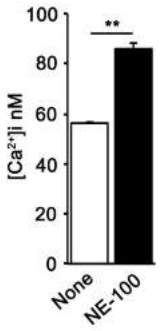
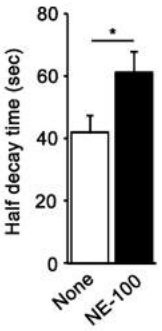
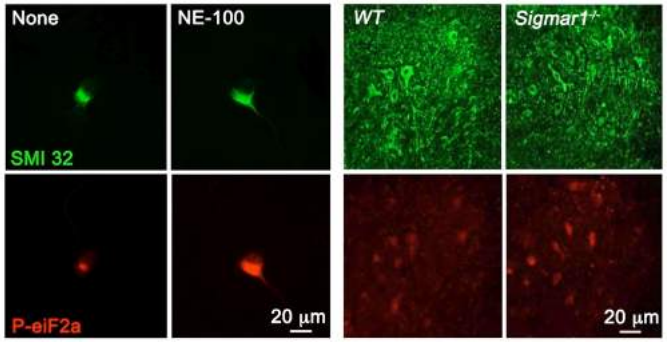
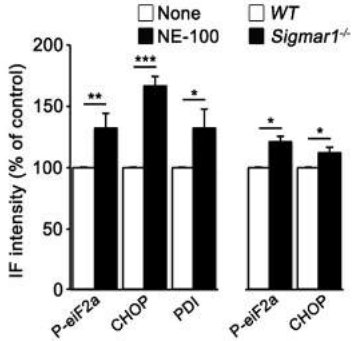


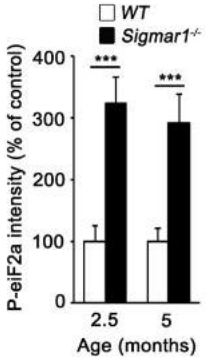




P-eiF2A, CHOP, PDI: ER stress indicators







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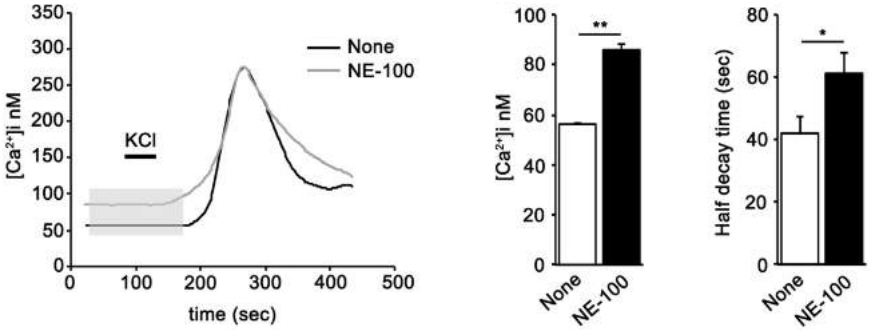

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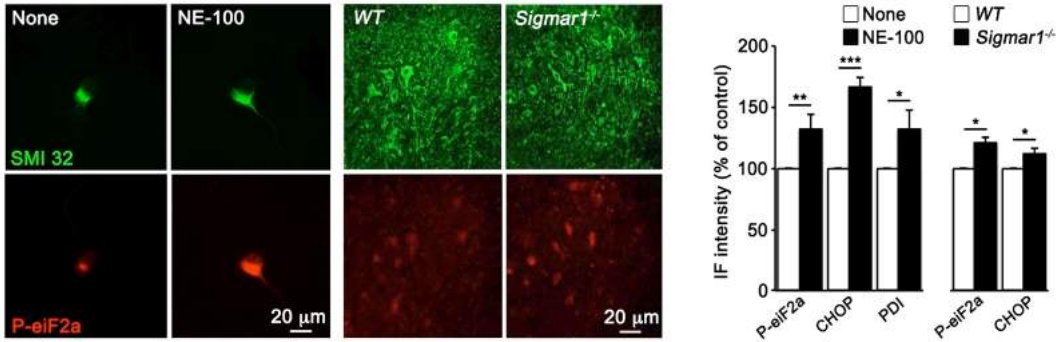
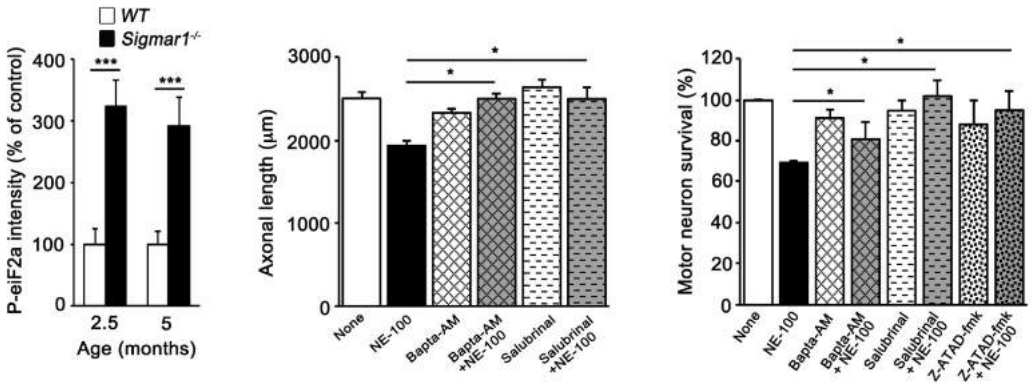


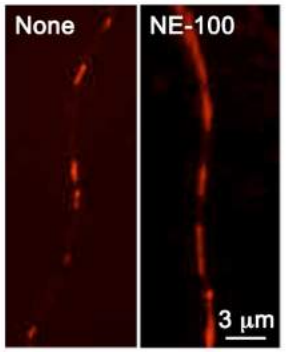
BAPTA-AM: cytoplasmic calcium scavenger
Salubrinal: ER stress inhibitor
Z-ATADfmk: inhibitor of the ER connected caspase-12

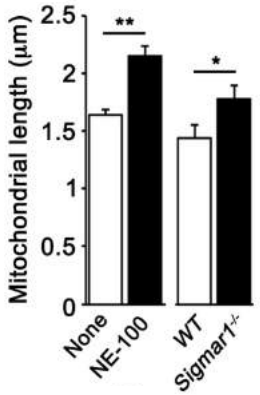


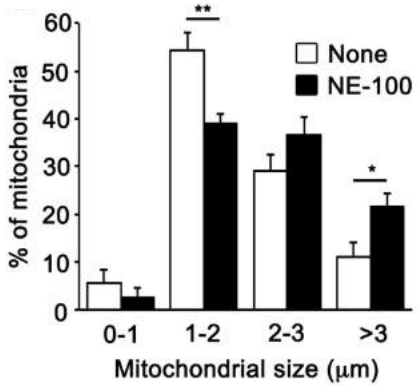
Deficit in mitochondrial dynamics induced by SIGMAR1 dysfunction triggers axonal degeneration and cell death

At MAMs, ER tubules have been shown to circumscribe the mitochondria thus promoting mitochondrial fission (Westermann, 2011)

MitoTracker Red







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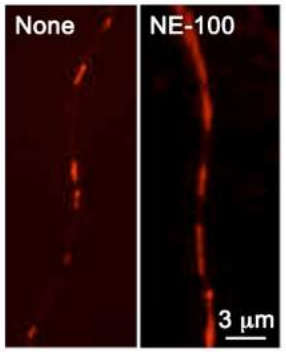
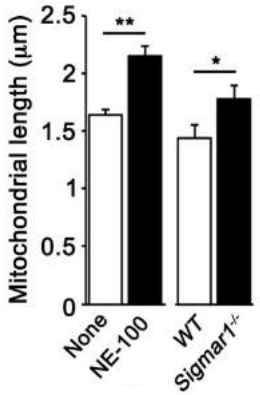
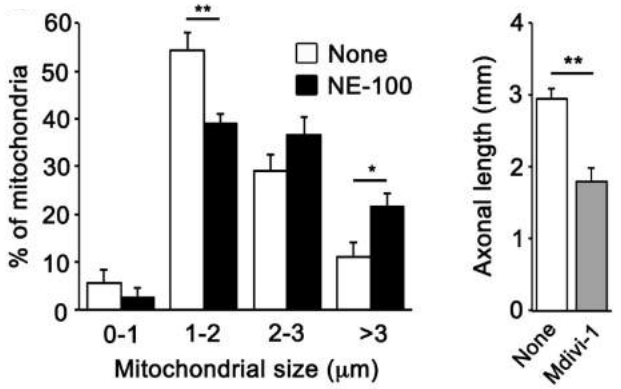
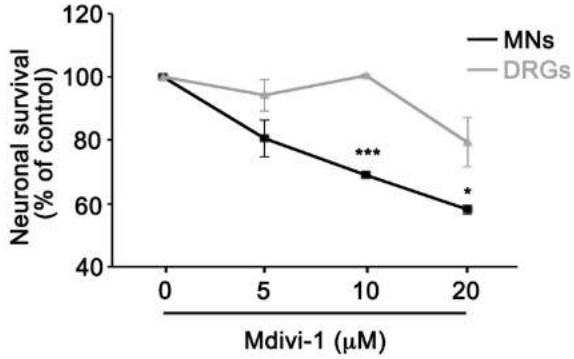


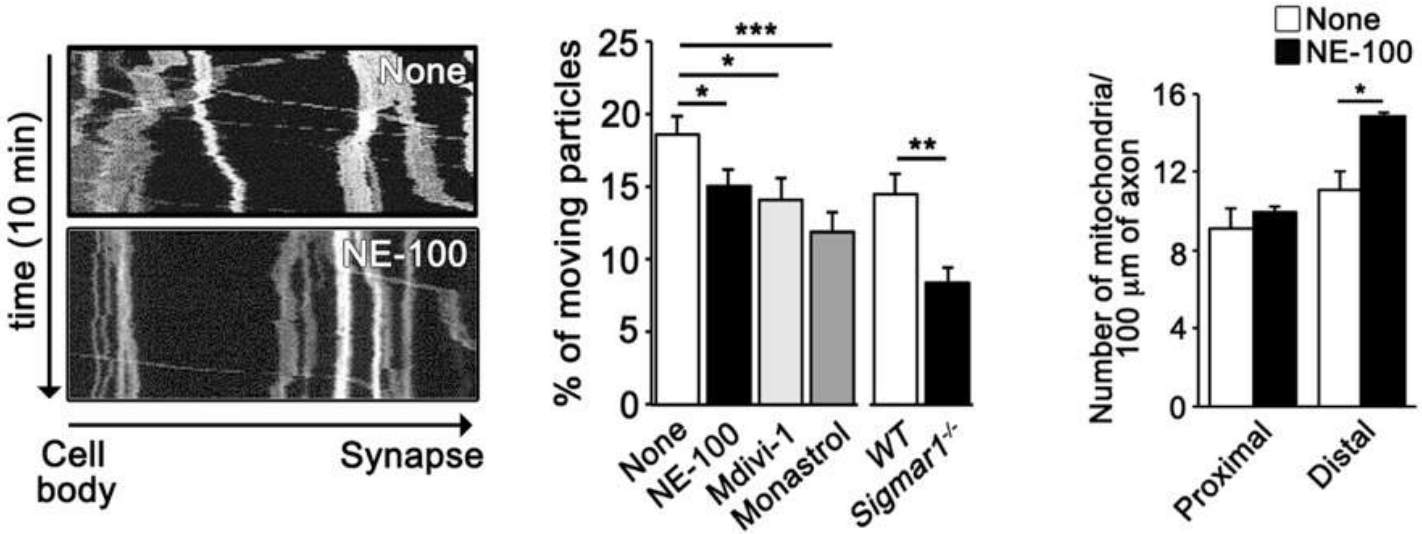


Mdivi-1: a selective inhibitor of DNM1L that blocks mitochondrial fission (Cassidy-Stone et al., 2008)



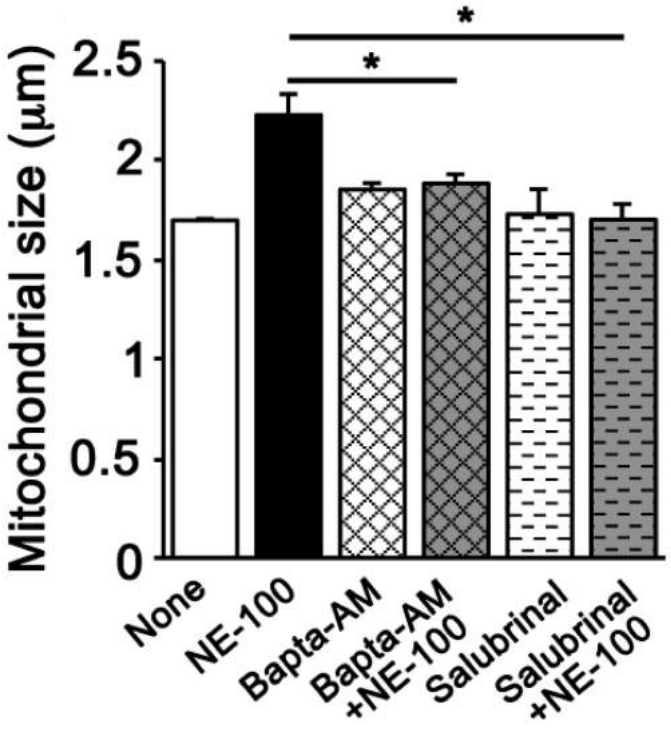


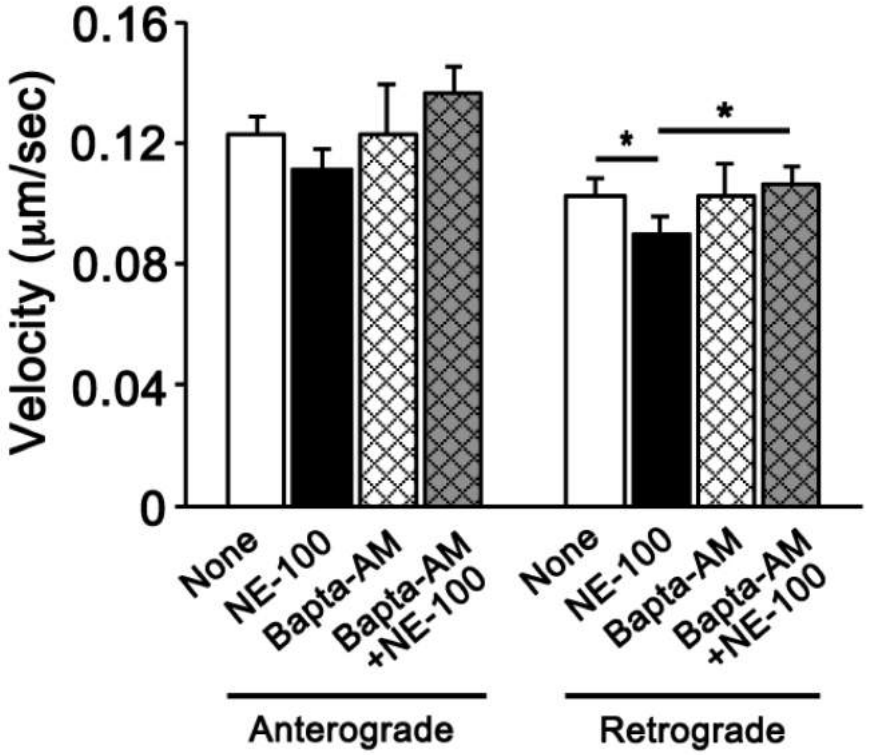
SIGMAR1 dysfunction disrupts mitochondrial axonal transport



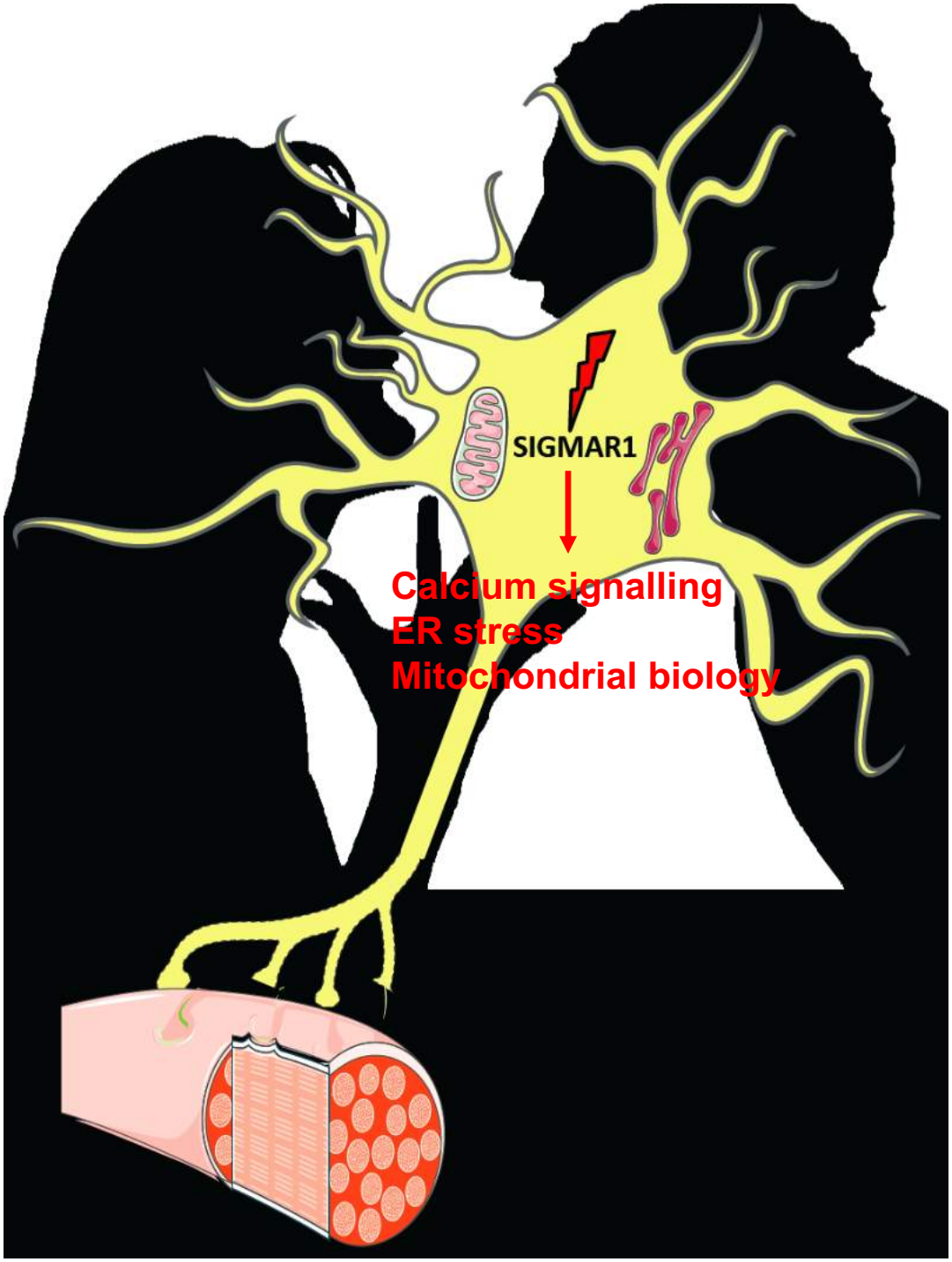
Monastrol: specifically affects anterograde transport

SIGMAR1 dysfunction disrupts mitochondrial axonal transport

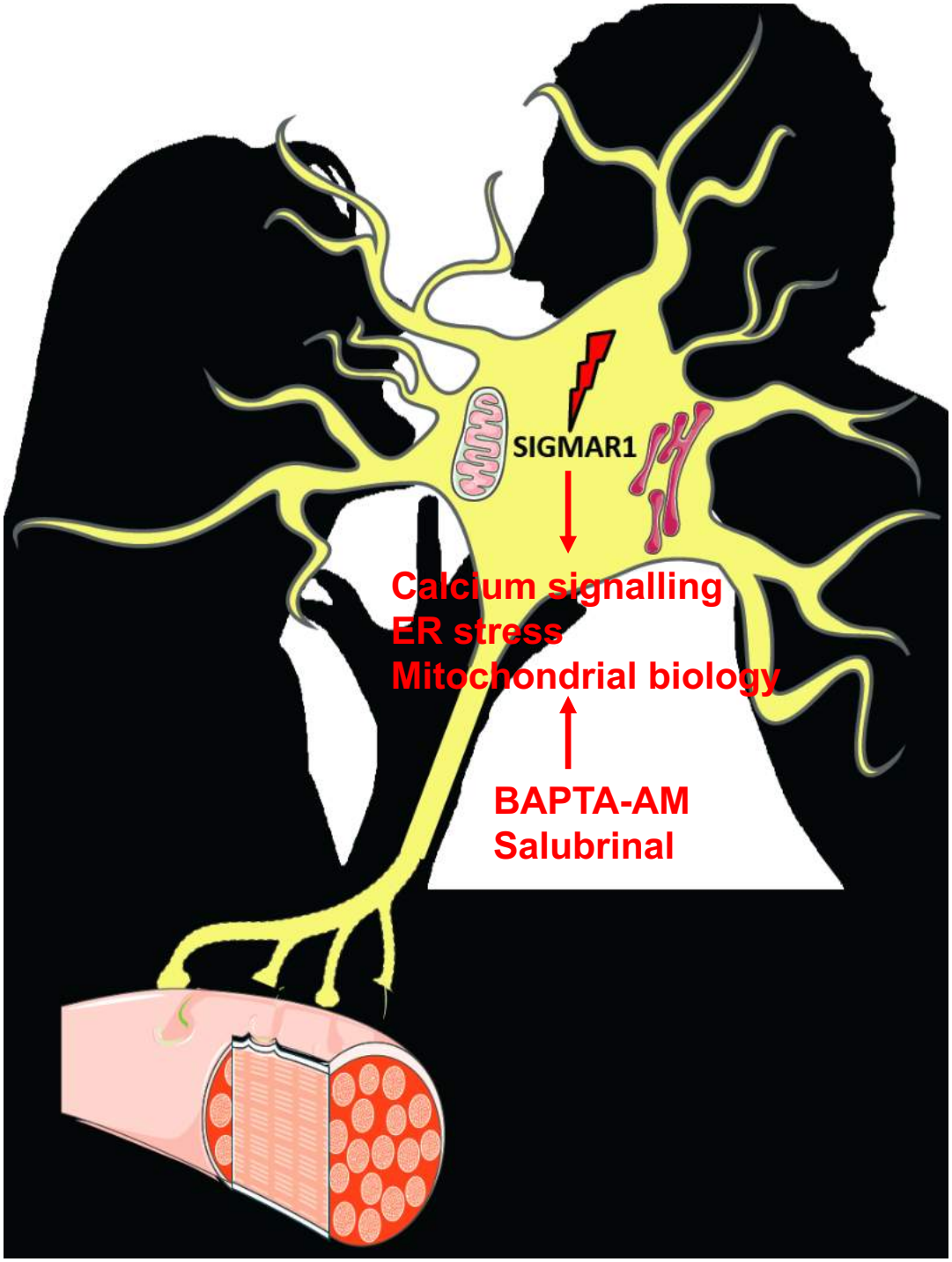




BAPTA-AM: cytoplasmic calcium scavenger
Salubrinal: ER stress inhibitor



Bernard-Marissal et al., Brain 2015



Bernard-Marissal et al., Brain 2015

Sigmar1 mutations and neuropathy

A *SIGMAR1* splice-site mutation causes distal hereditary motor neuropathy

Xiaobo Li, Zhengmao Hu, Lei Liu, Yongzhi Xie, Yajing Zhan, Xiaohong Zi, Junling Wang, Lixiang Wu, Kun Xia, Beisha Tang, and Ruxu Zhang

Neurology 84 June 16, 2015

SIGMAR1 mutation associated with autosomal recessive Silver-like syndrome

Horga A., Tomaselli P.J., Gonzalez M.A., Laurà M., Muntoni F., Manzur A.Y., Hanna M.G., Blake J.C., Houlden H., Züchner S., Reilly M.M.

Neurology 87 October 11, 2016

Further Validation of the *SIGMAR1* c.151+1G>T Mutation as Cause of Distal Hereditary Motor Neuropathy

Jessica J. Y. Lee, BSc[1,2,3], Clara D. M. van Karnebeek, MD, PhD[1,2,3,4], Britt Drögemoller, PhD[3,5], Casper Shyr, PhD[1,2,3], Maja Tarailo-Graovac, PhD[1,2,3,5], Patrice Eydoux, PhD[2,3,6], Colin J. Ross, PhD[1,2,3,5], Wyeth W. Wasserman, PhD[1,2,3,5], Bruce Björnson, MD[3,7], and John K. Wu, MD[3,8]

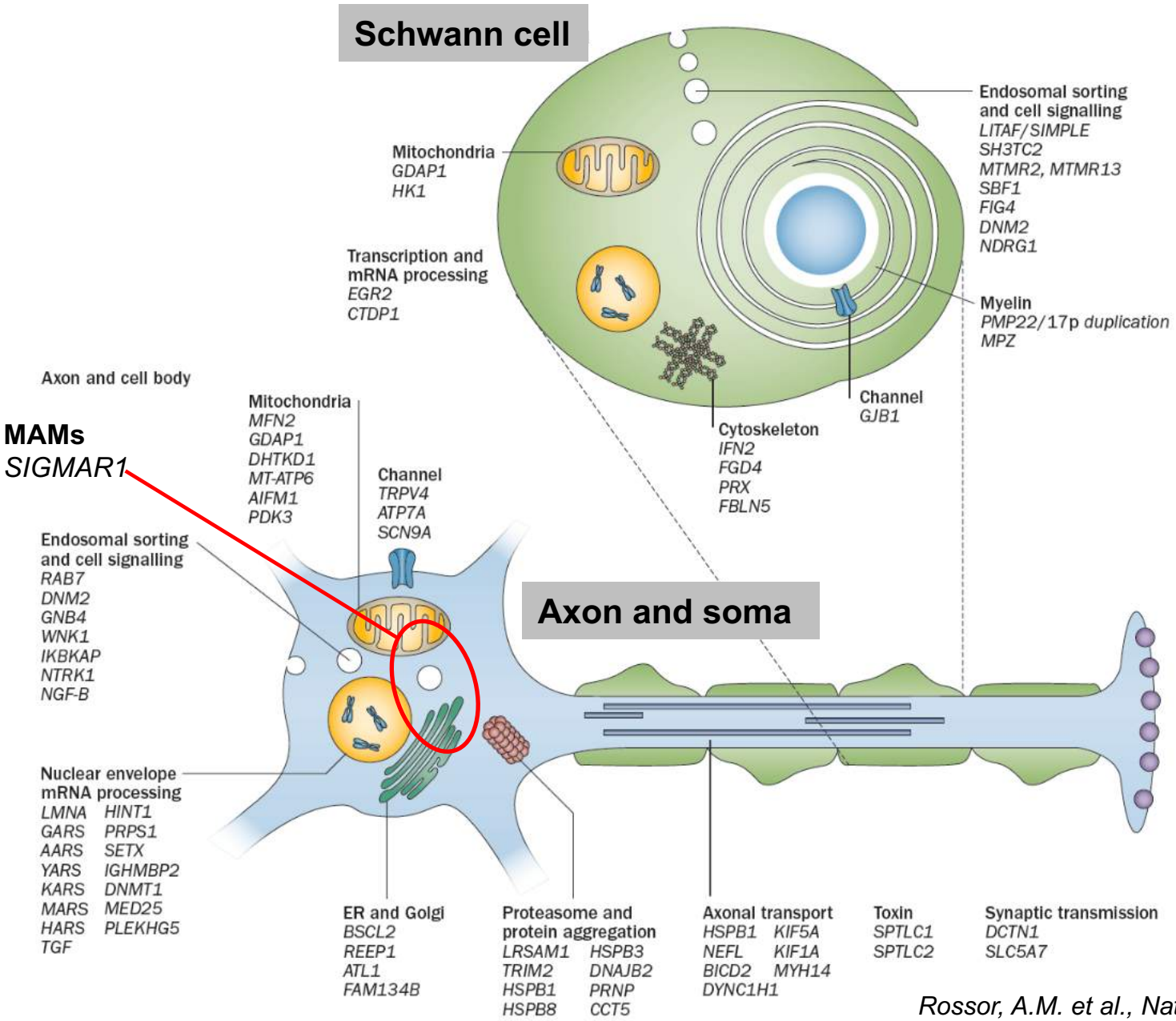
Child Neurology Open
Volume 3: 1-5

Recessive distal motor neuropathy with pyramidal signs in an Omani kindred: underlying novel mutation in the *SIGMAR1* gene

R. Nandhagopal[a], D. Meftah[b], S. Al-Kalbani[c] and P. Scott[c]

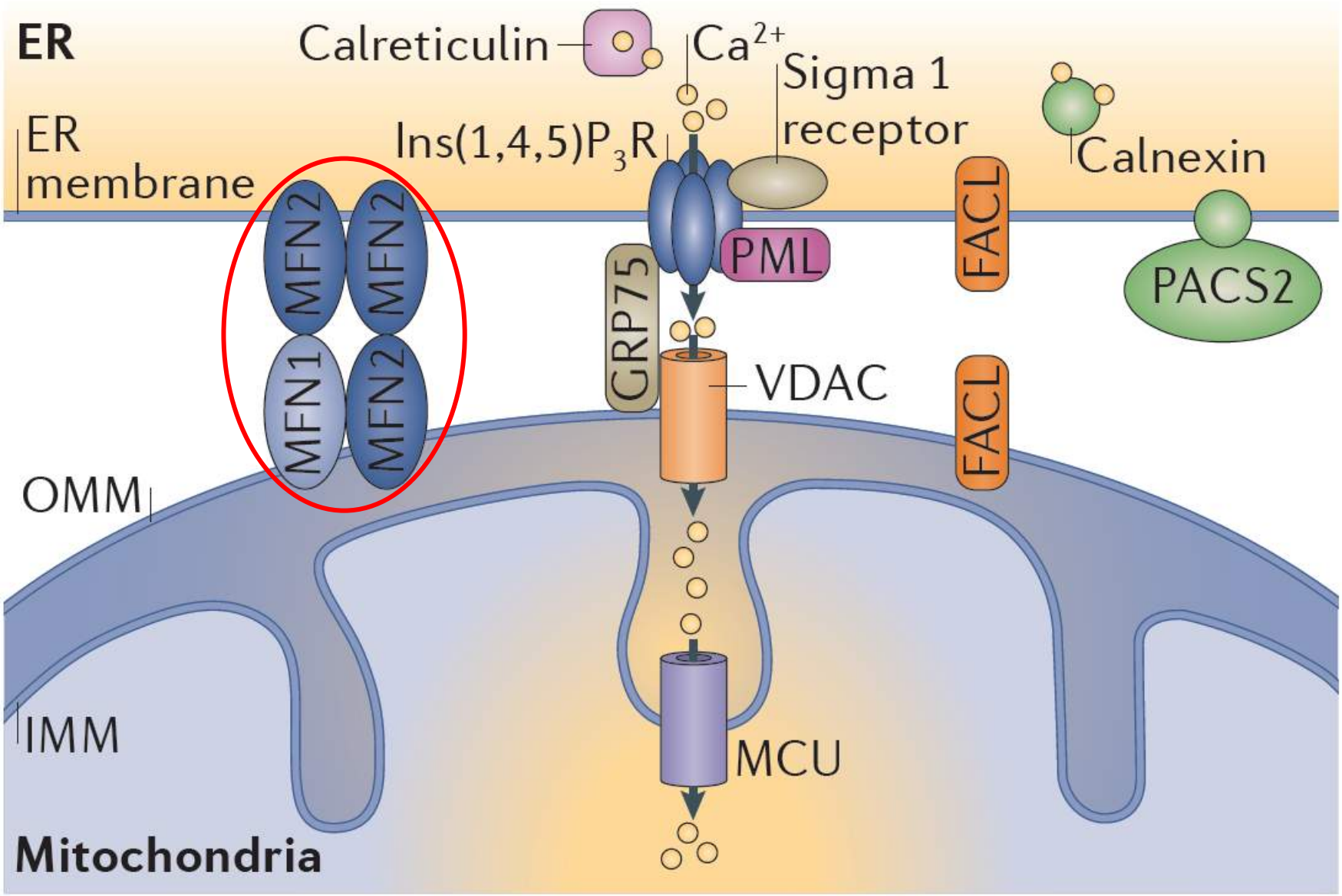
European Journal of Neurology 2018, **25:** 395–403

Inherited neuropathies



Rossor, A.M. et al., Nat. Rev. Neurol. 2013

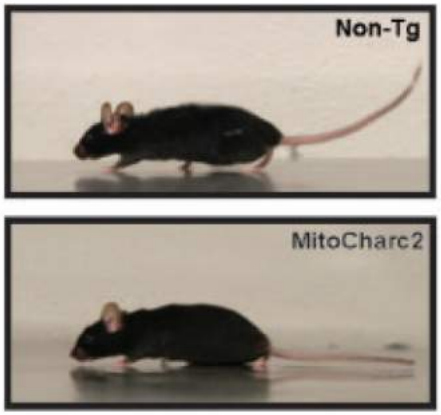
MFNs and MAMs

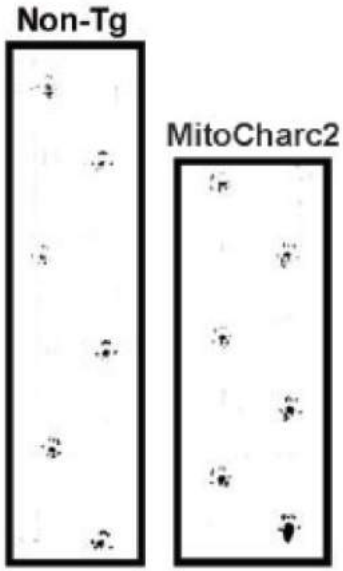


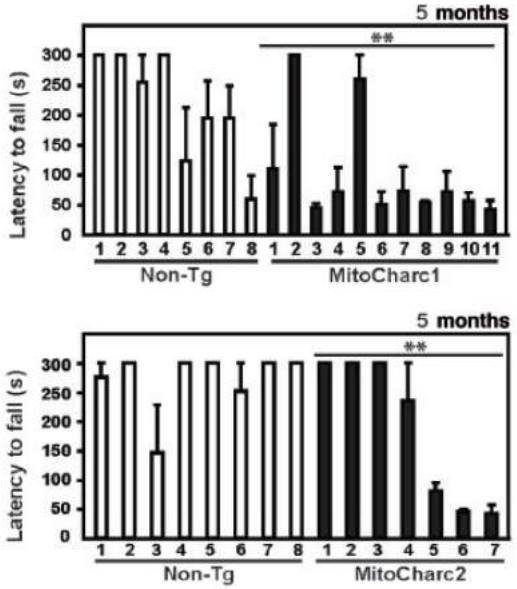
Rizzuto et al., Nat. Rev. Mol. Cell Biol. 2012

CMT2A-like phenotype in MFN2^{R94Q} Tg mice

CMT2A: dominant form of axonal peripheral sensorimotor neuropathy caused by mutations in MFN2

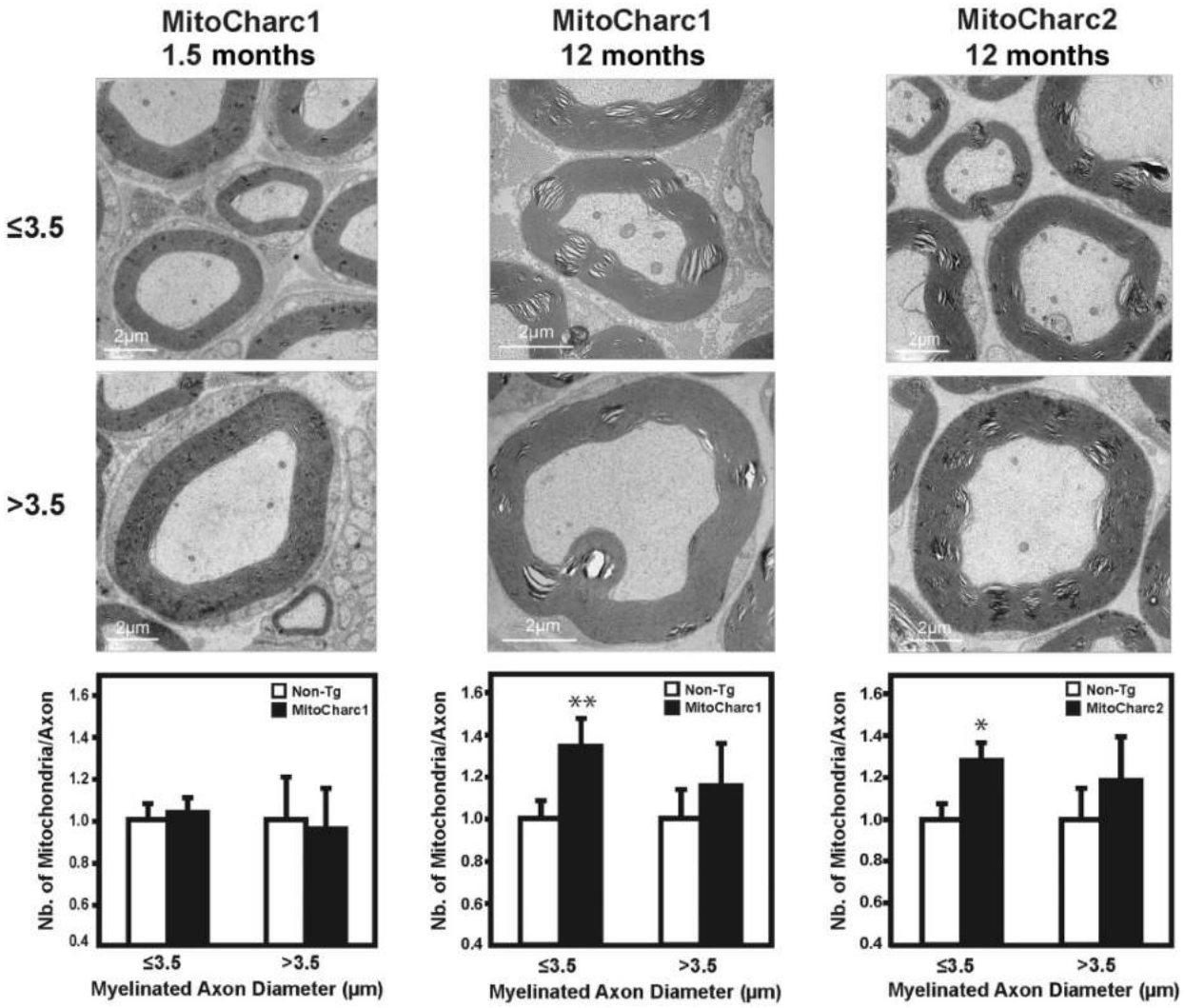






Cartoni et al., Brain 2010

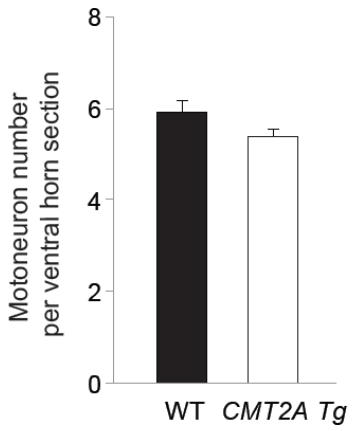
Increased number of mitochondria in the distal part of the sciatic nerve from MFN2^{R94Q} Tg1 and 2 mice



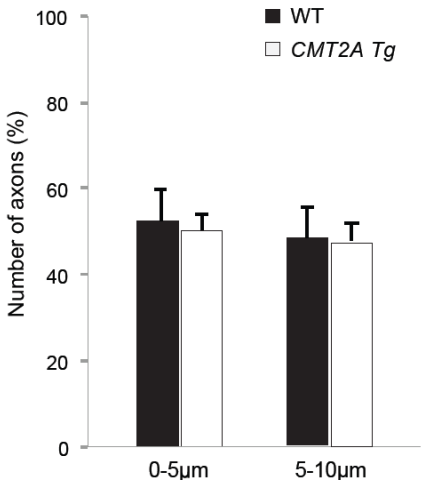
Cartoni et al., Brain 2010

Motoneurons and root fibers in CMT2A MFN2^{R94Q} Tg mice

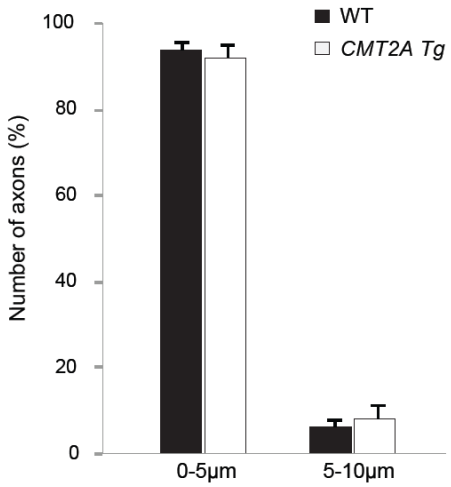
Lumbar spinal cord



Ventral roots



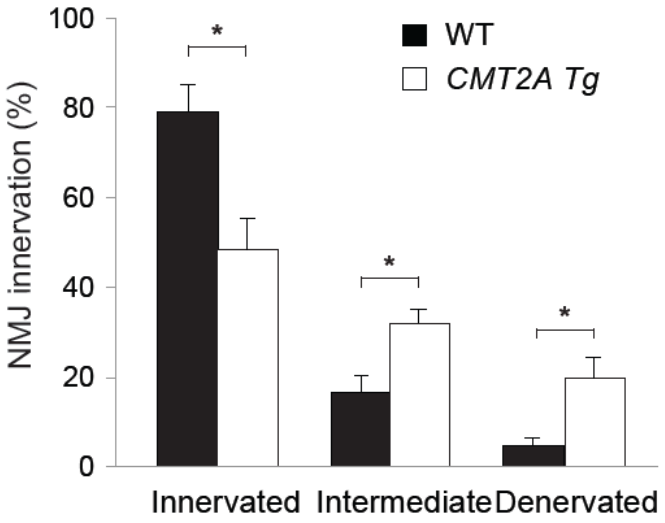
Dorsal roots

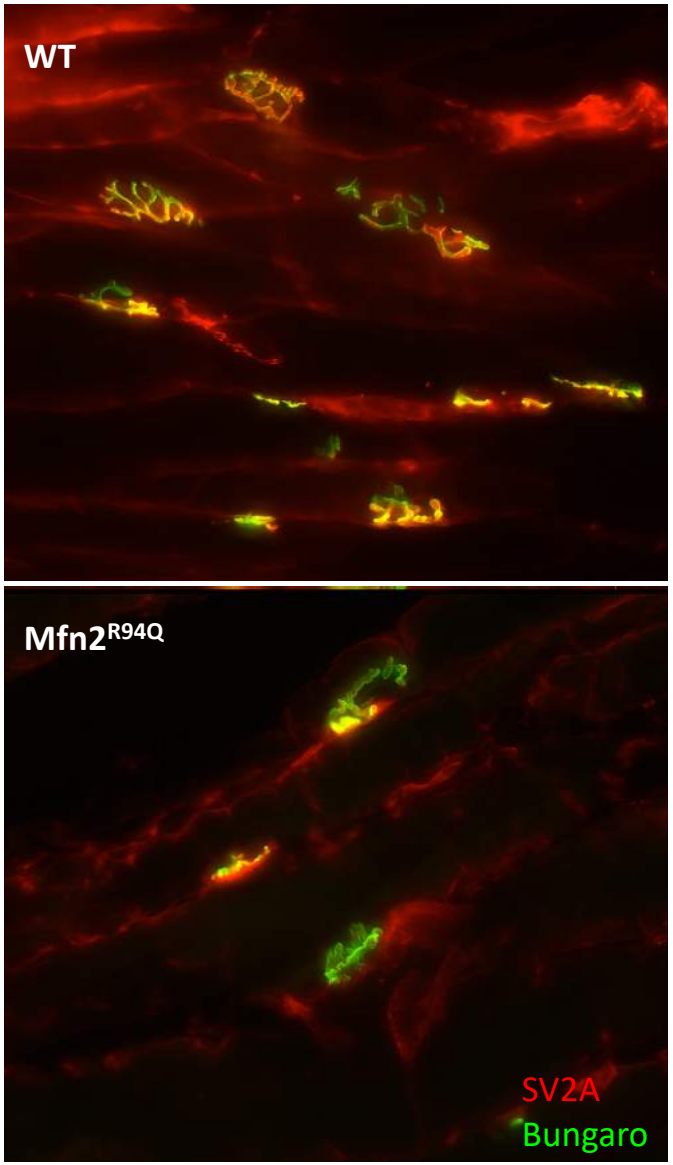


Age: 1 year

MFN2^{R94Q} Tg mice display muscle denervation

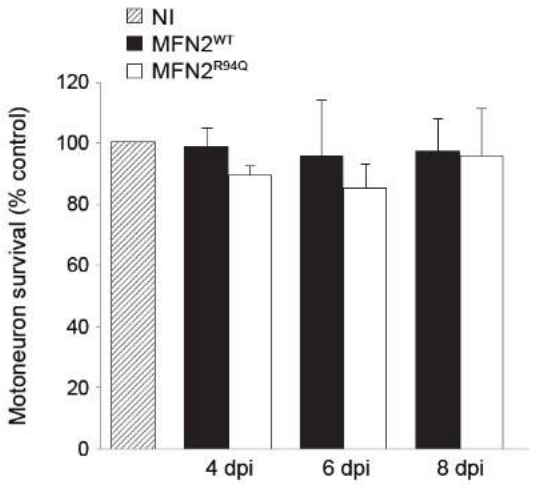
Innervation of soleus muscle

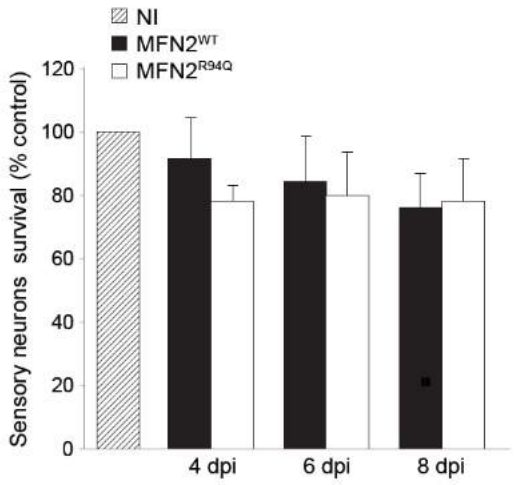




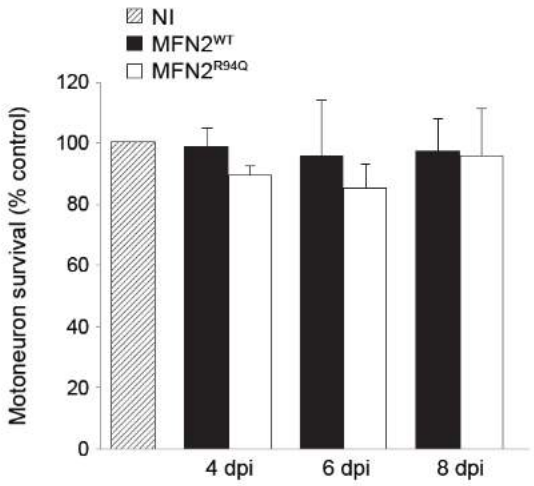
Age: 1 year

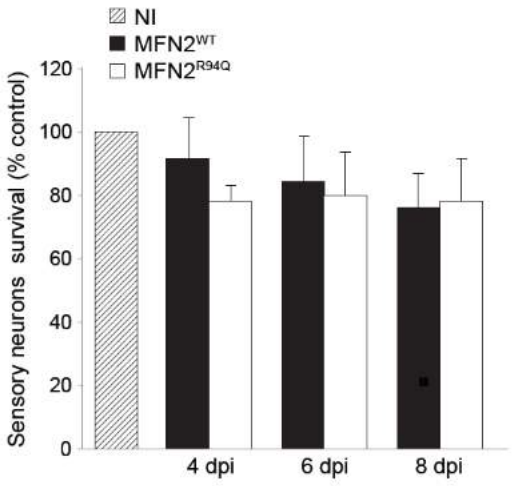
MFN2^{R94Q} induces axonal degeneration in the absence of neuronal death

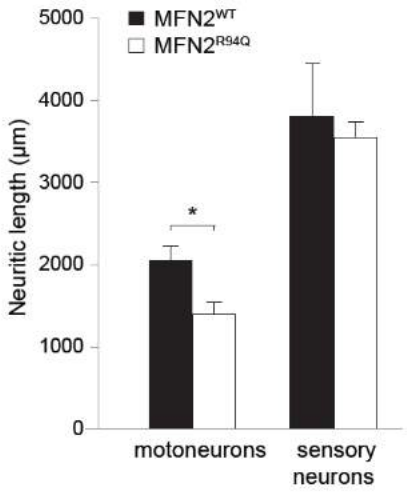


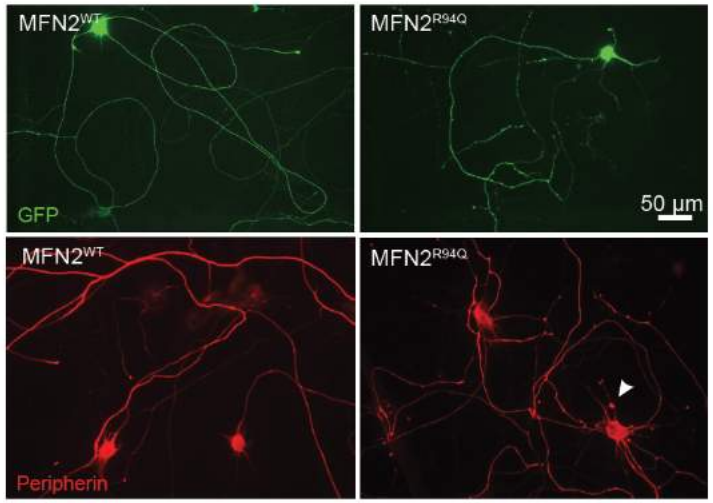


MFN2^{R94Q} induces axonal degeneration in the absence of neuronal death



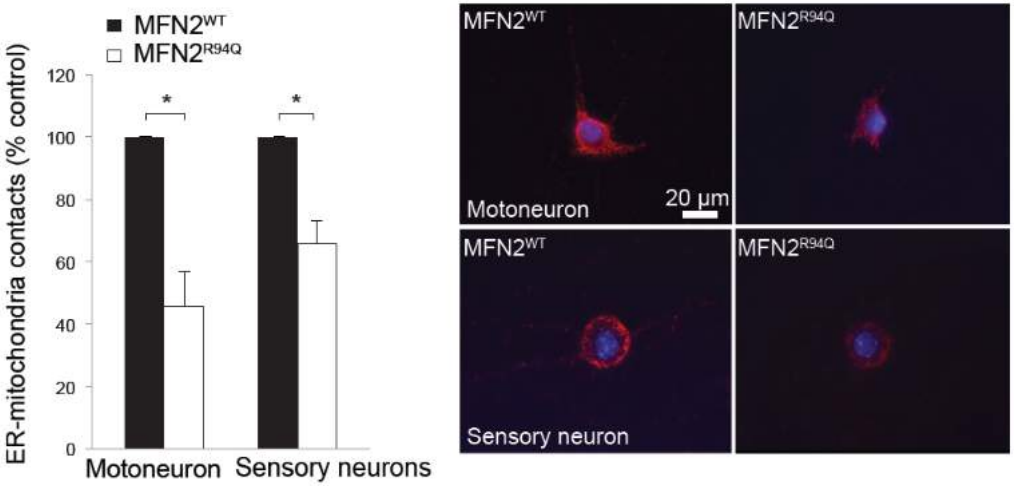




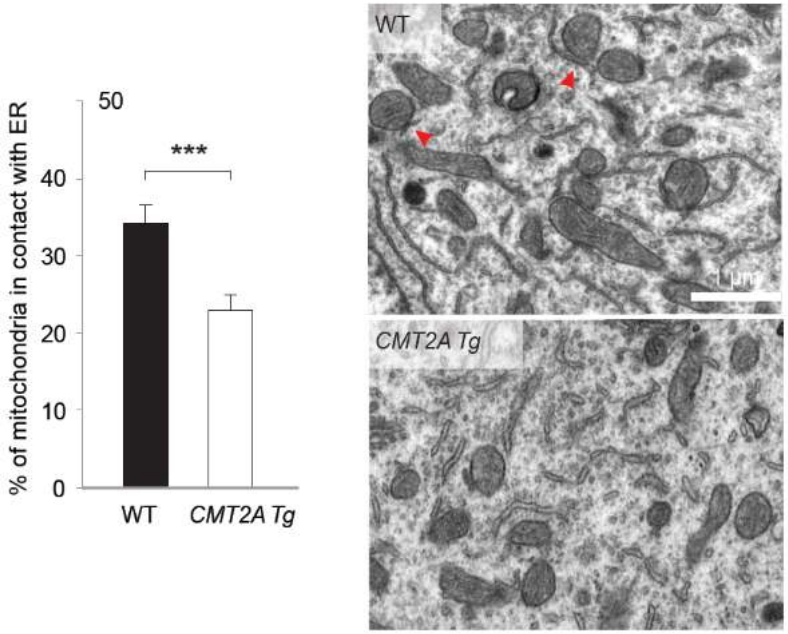


MFN2^{R94Q} affects neuronal ER-mitochondria contacts *in vitro* and *in vivo*

in vitro



in vivo



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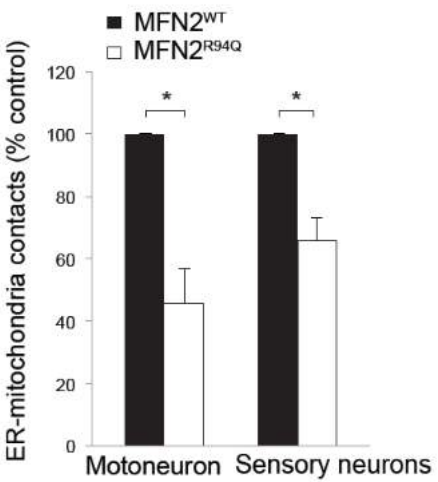
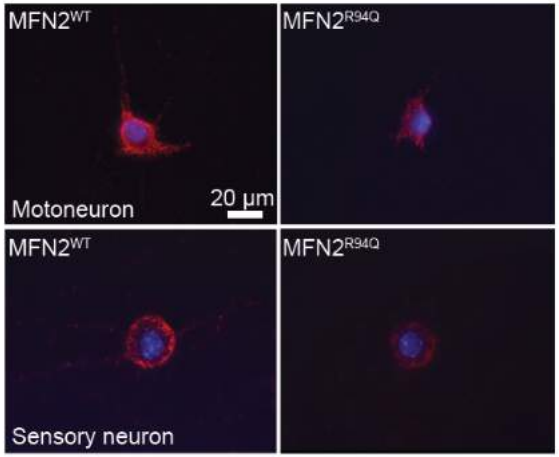
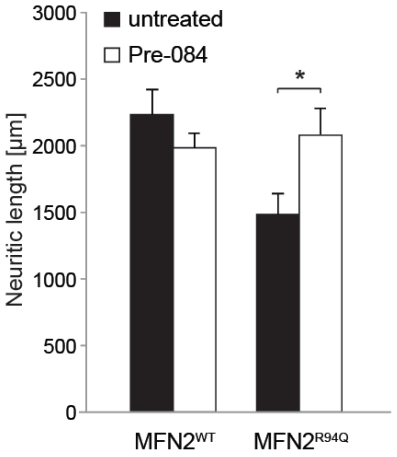
in vitro

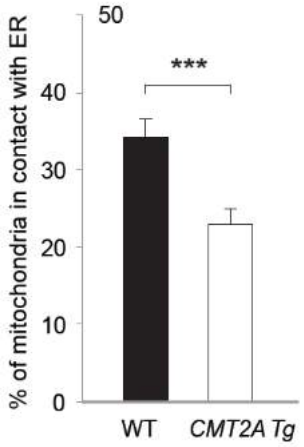


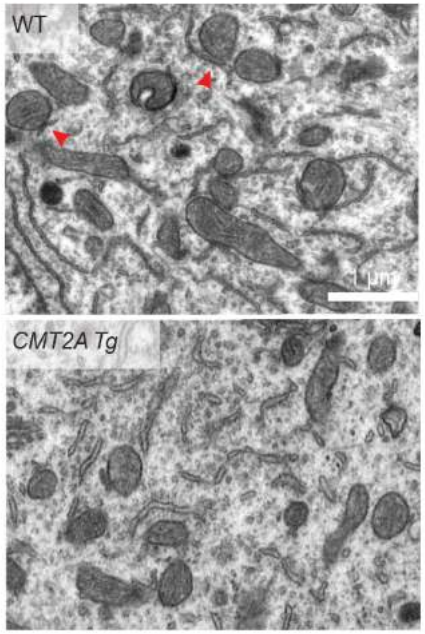




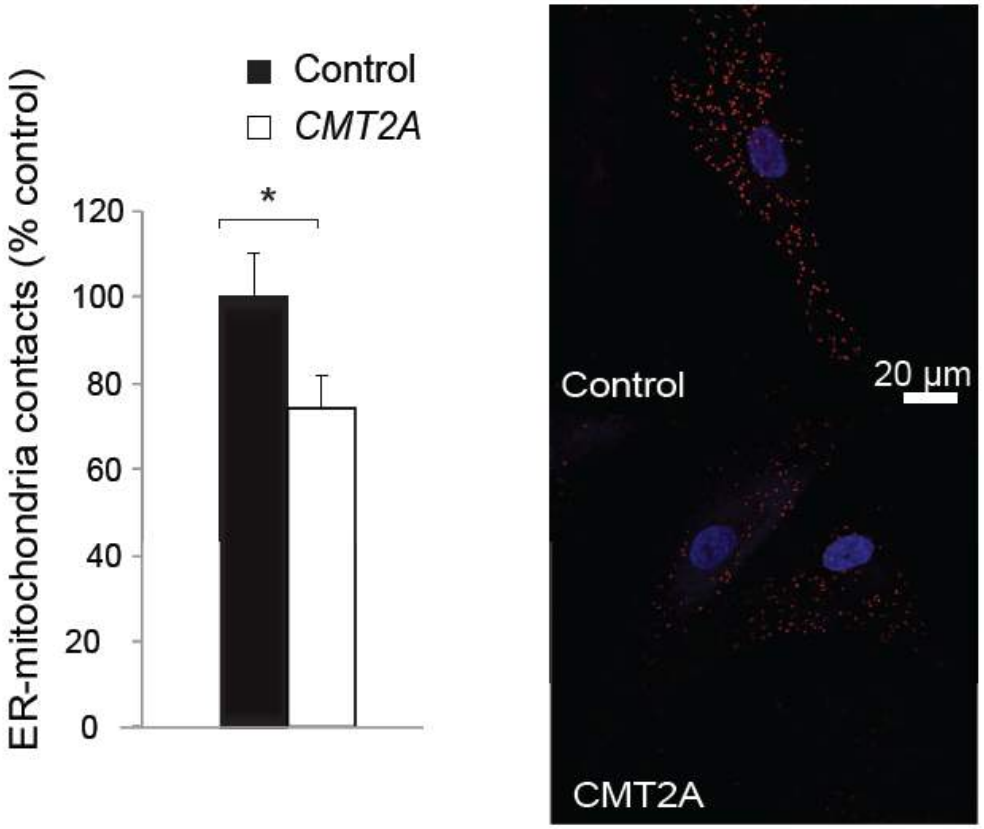
Pre-084: SIGMAR1 agonist

in vivo



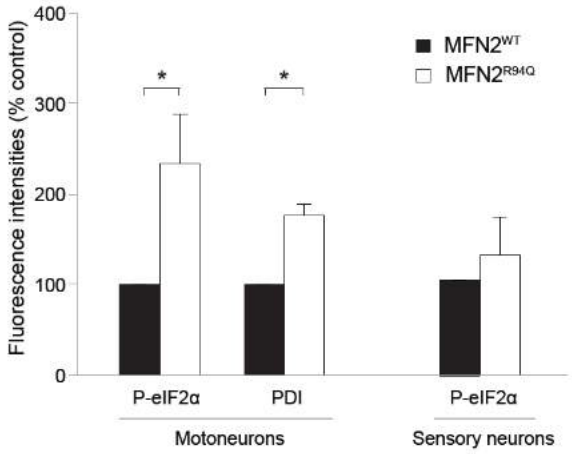


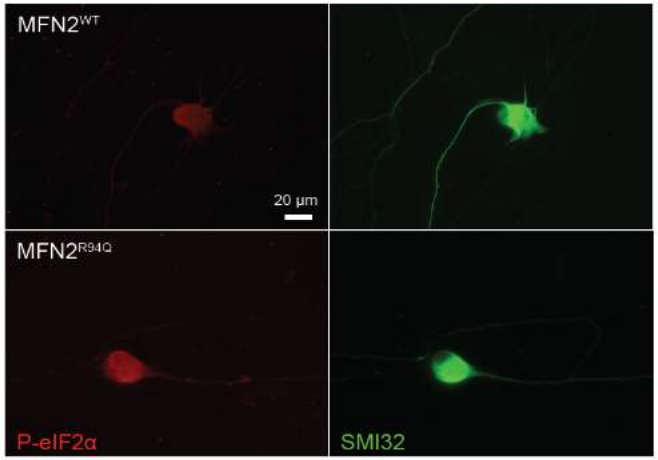
Proximity ligation assay in control and CMT2A fibroblasts



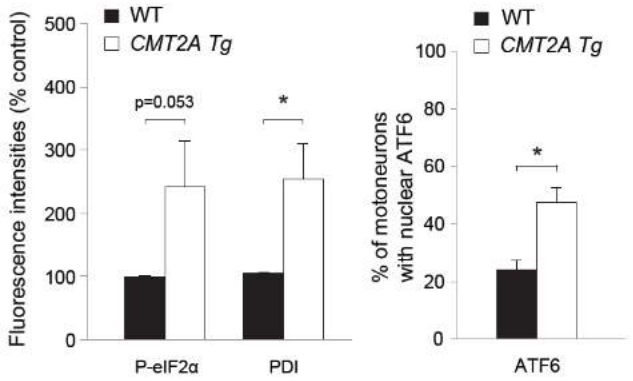
MFN2^{R94Q} induces ER stress both *in vitro* and *in vivo*

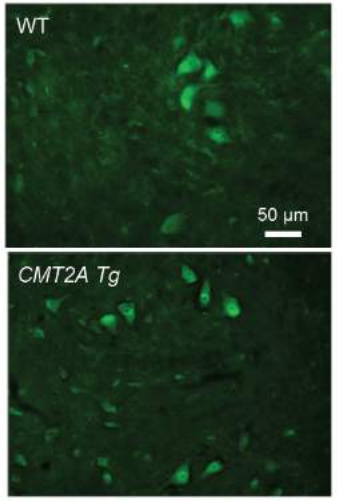
in vitro





in vivo





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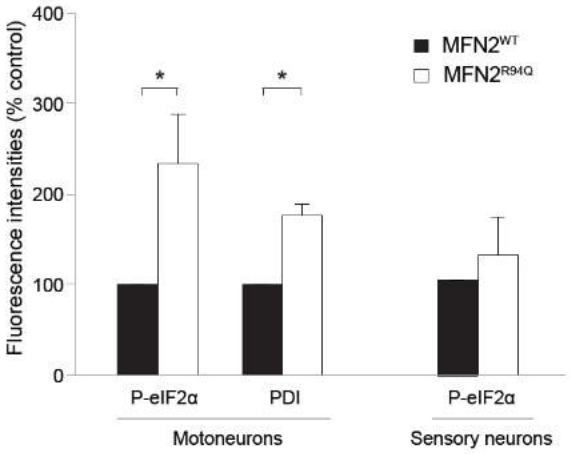
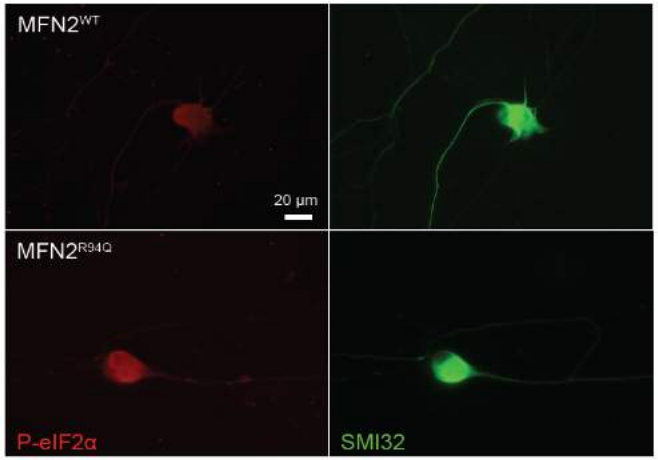
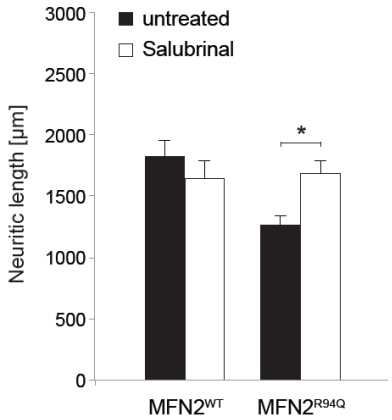
in vitro

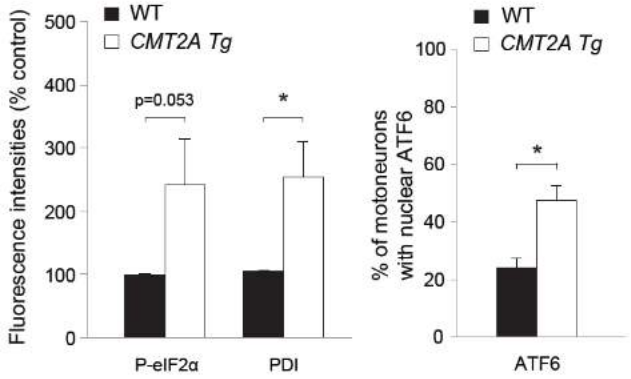


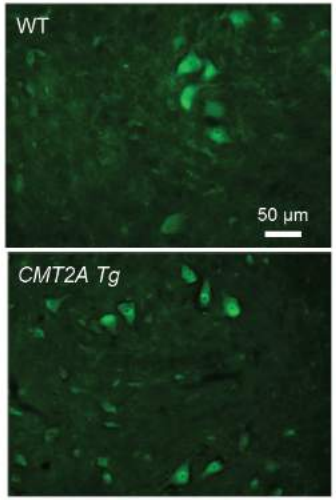




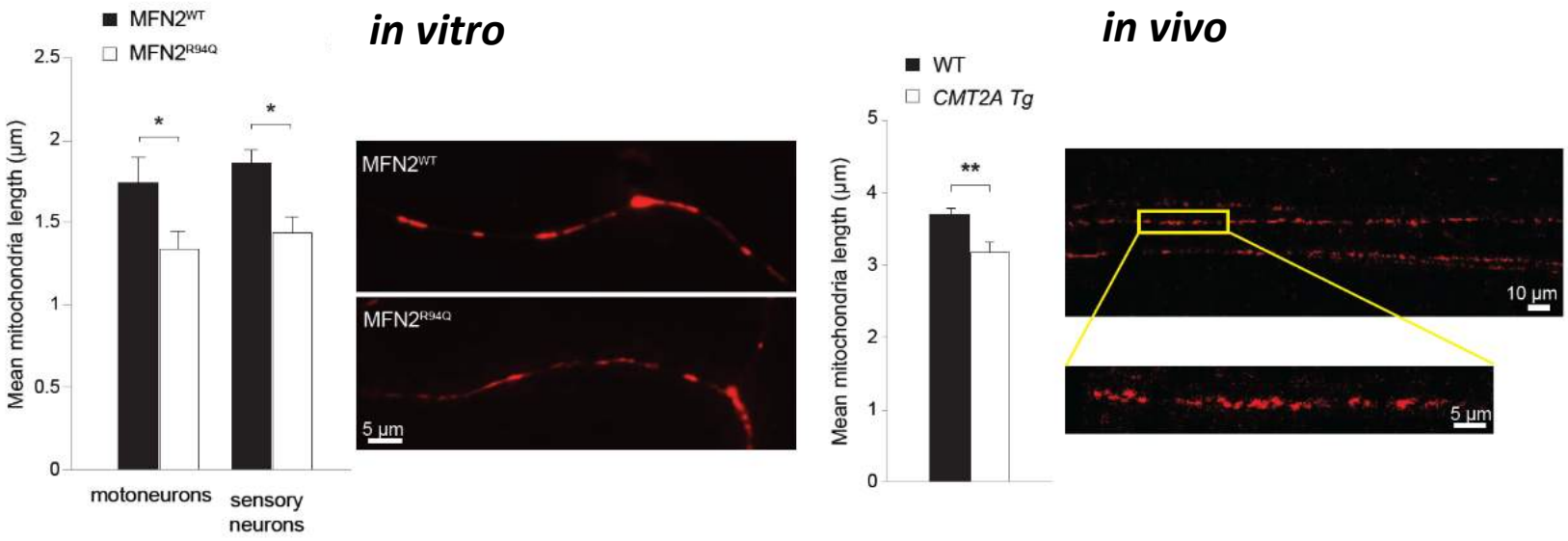
Salubrinal: ER stress inhibitor

in vivo





Mitochondrial morphology is affected by MFN2^{R94Q} both *in vitro* and *in vivo*



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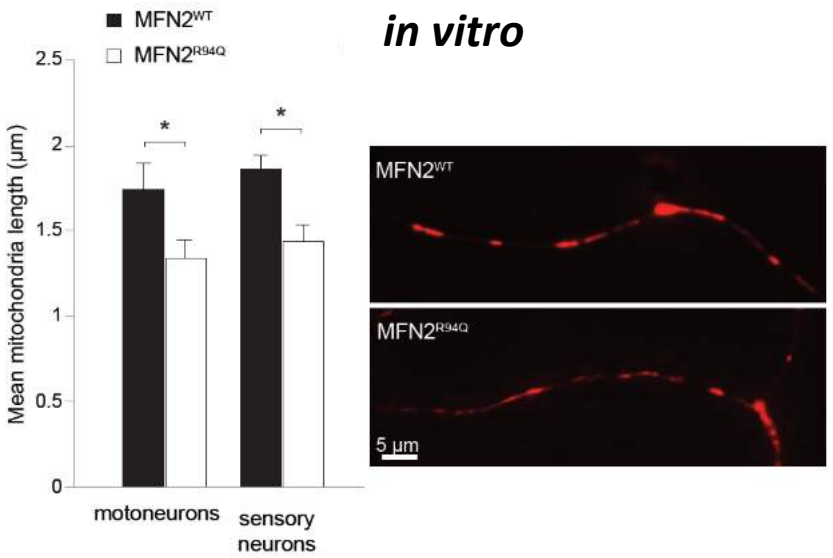
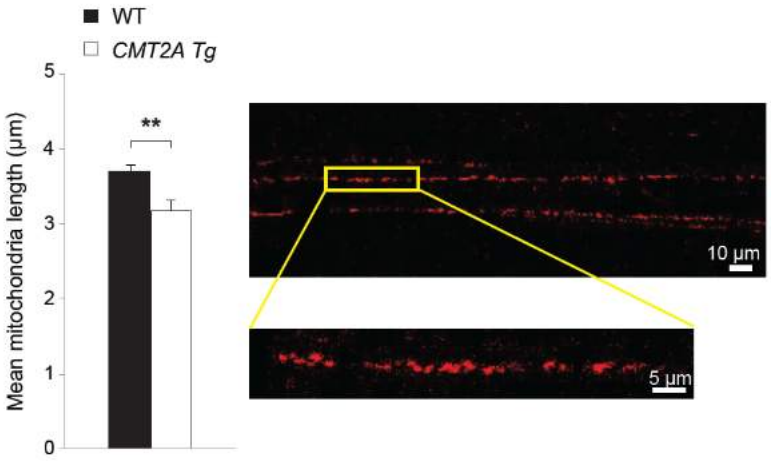
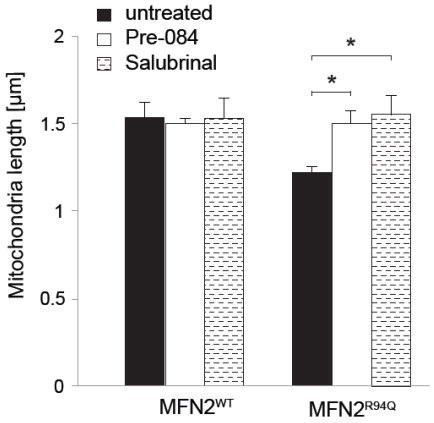
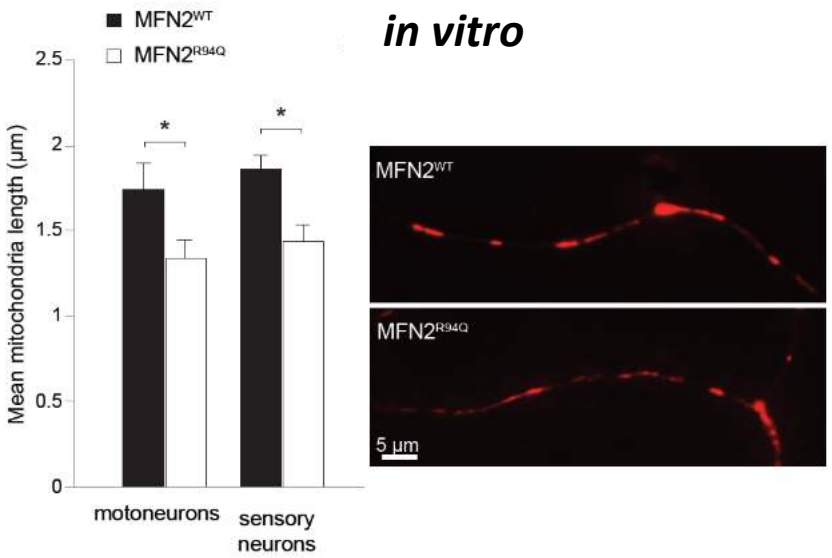


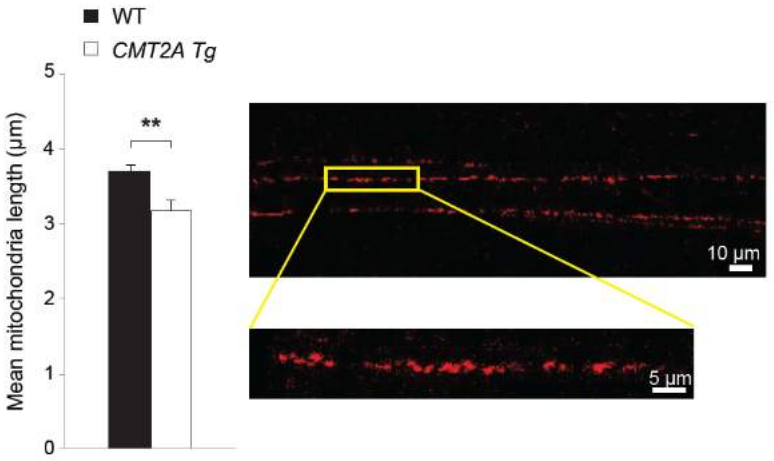


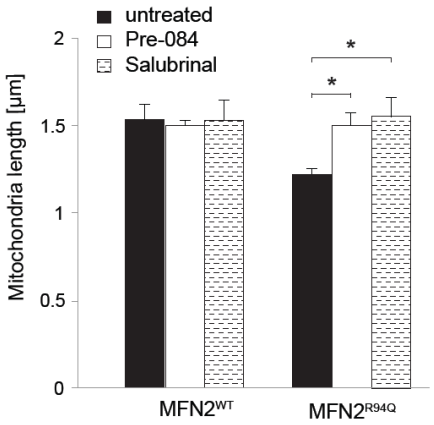


Pre-084: SIGMAR1 agonist
Salubrinal: ER stress inhibitor

Mitochondrial morphology is affected by MFN2^{R94Q} both *in vitro* and *in vivo*







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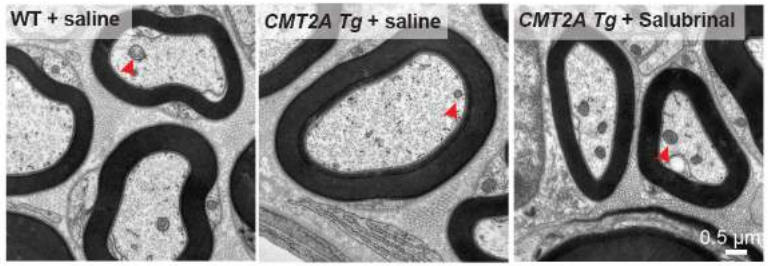
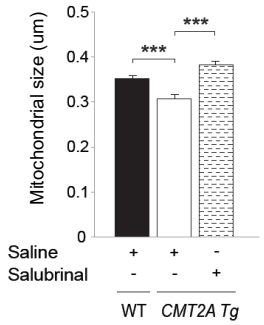
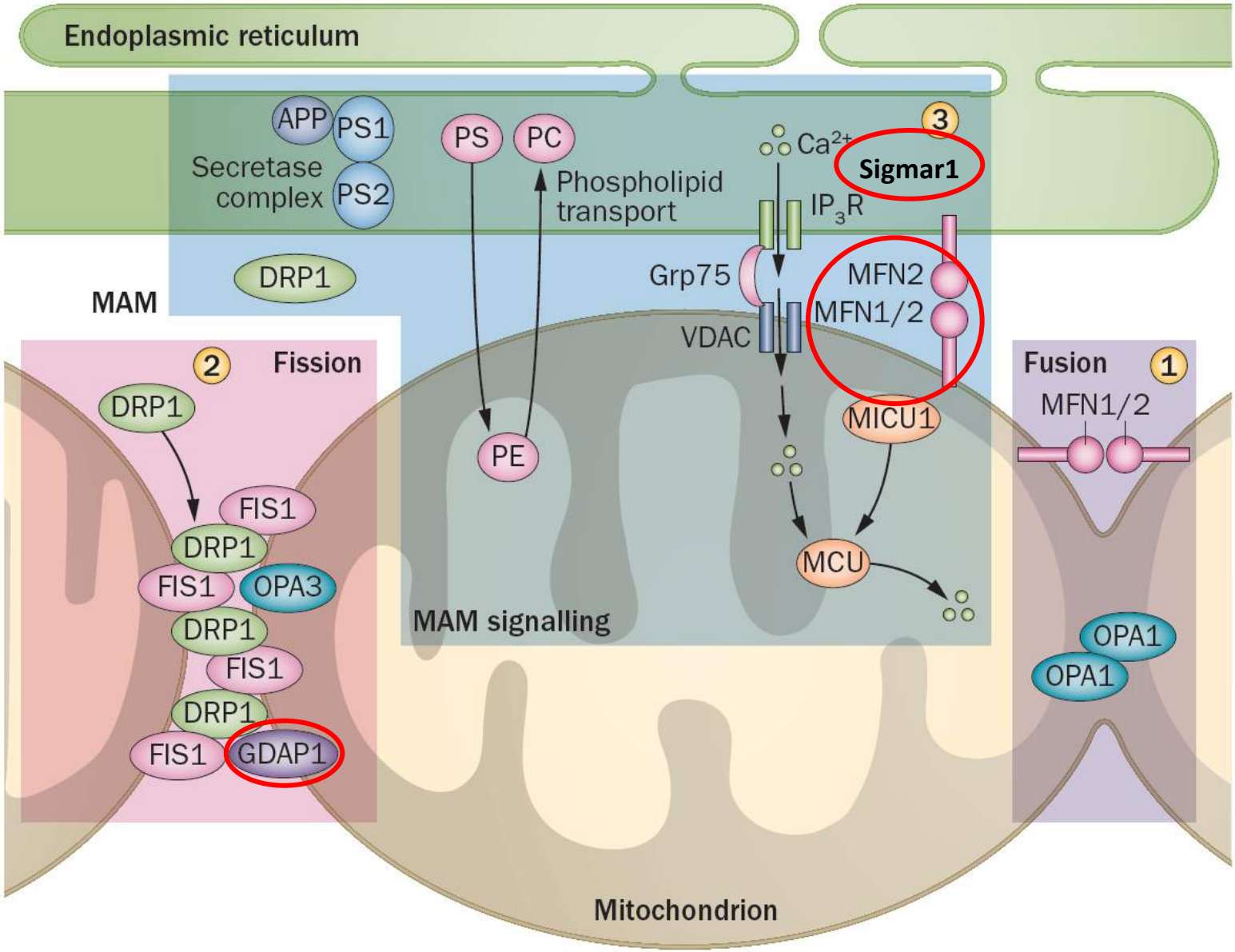
Salubrinal: ER stress inhibitor





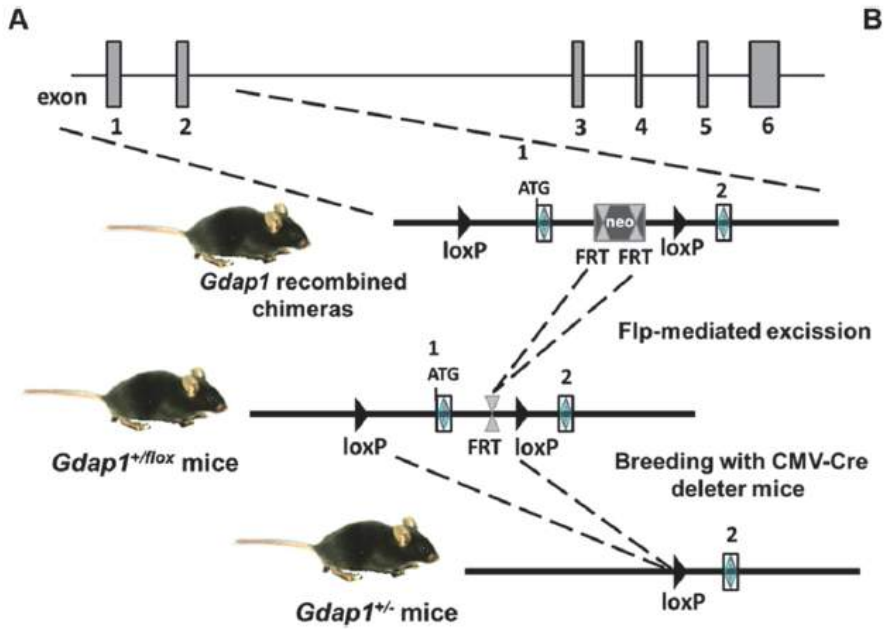
Mitochondrial dynamics and interactions

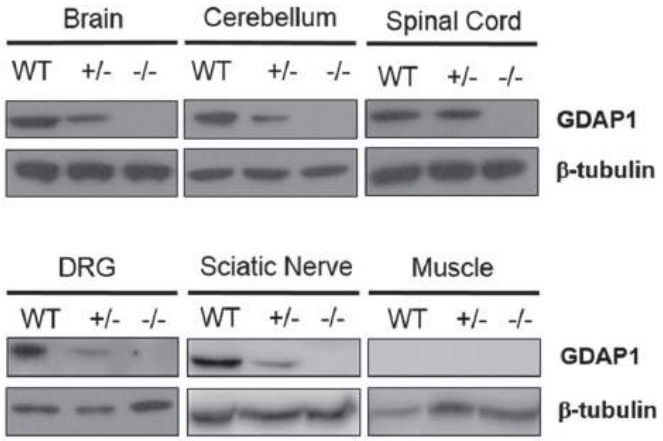


Burté et al., Nat. Rev. Neurol. 2015

GDAP1

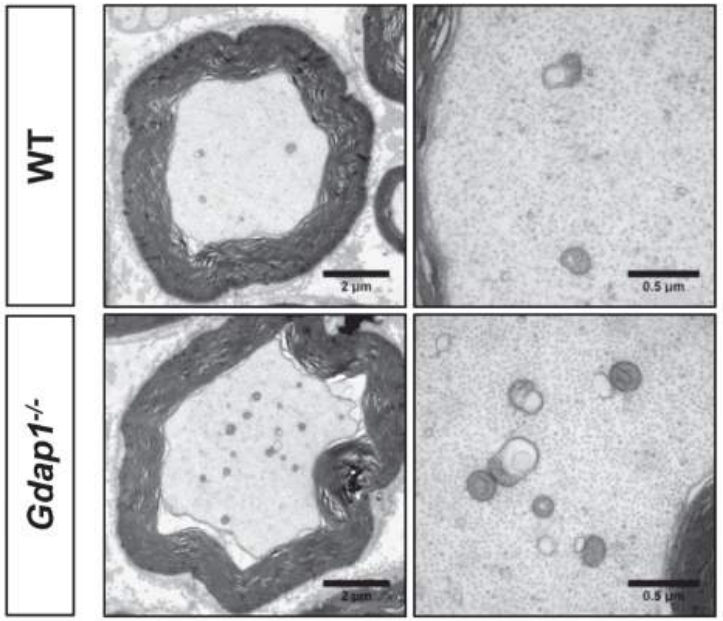
Mutations in GDAP1 cause axonal recessive (AR-CMT2), axonal dominant (CMT2K) and demyelinating recessive (CMT4A) forms of Charcot-Marie-Tooth (CMT) neuropathy

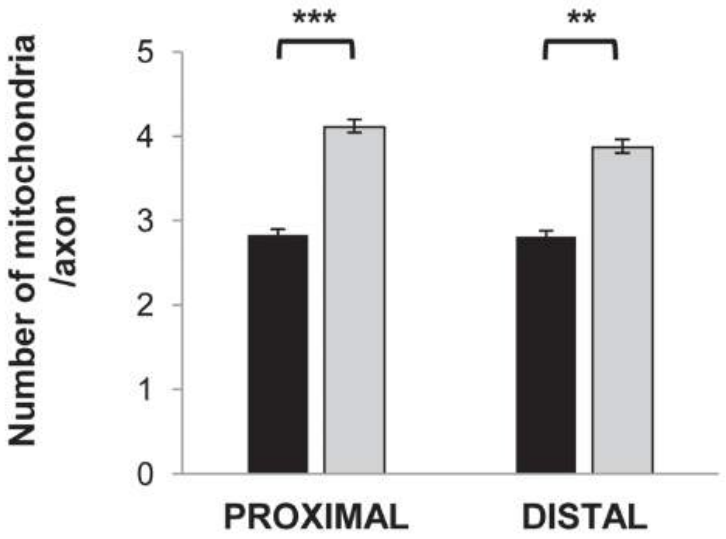




Barneo-Muñoz et al., PLOS Genetics 2015

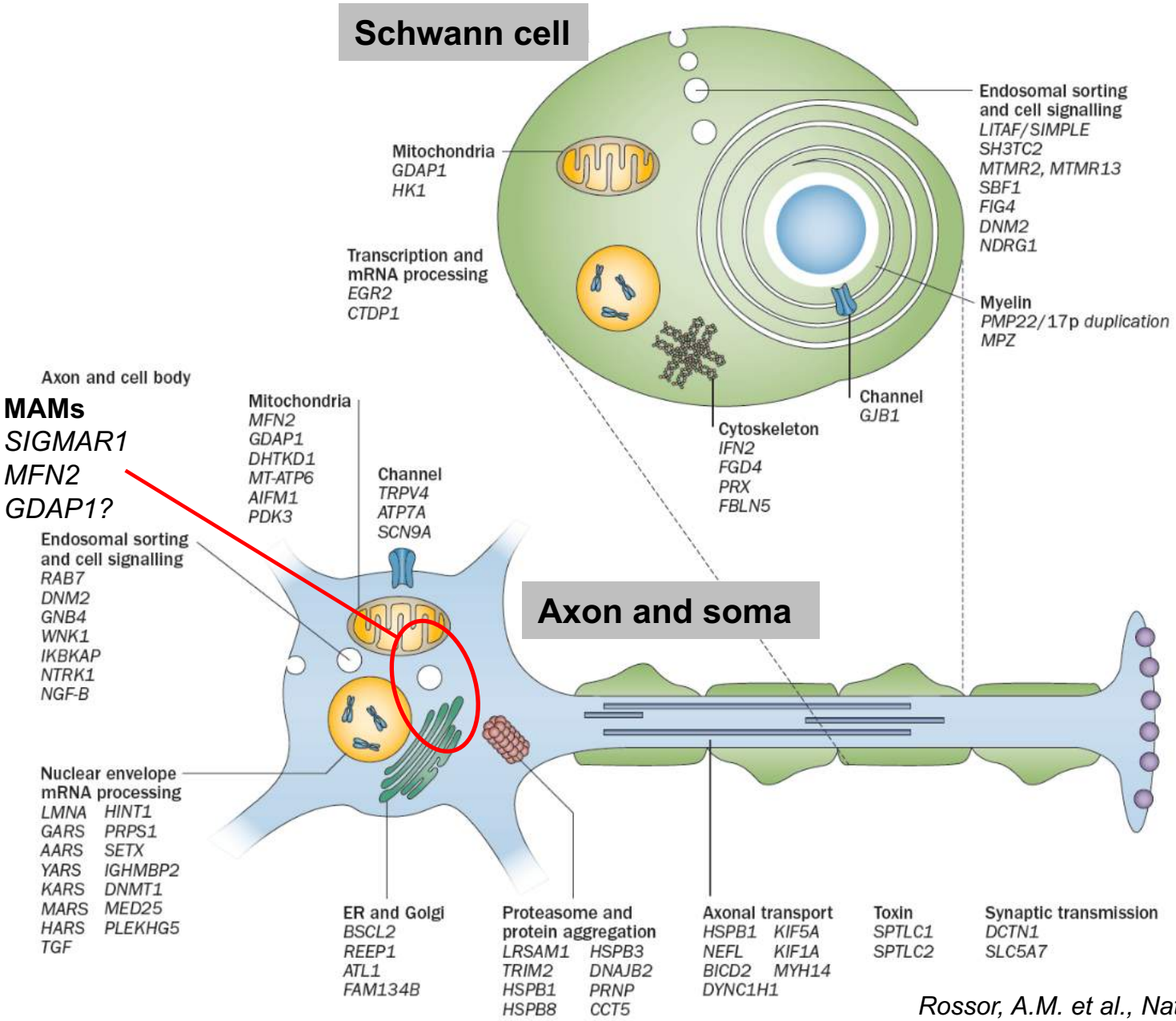
Mitochondrial quantitation in sciatic nerves from WT and Gdap1$^{-/-}$ mice





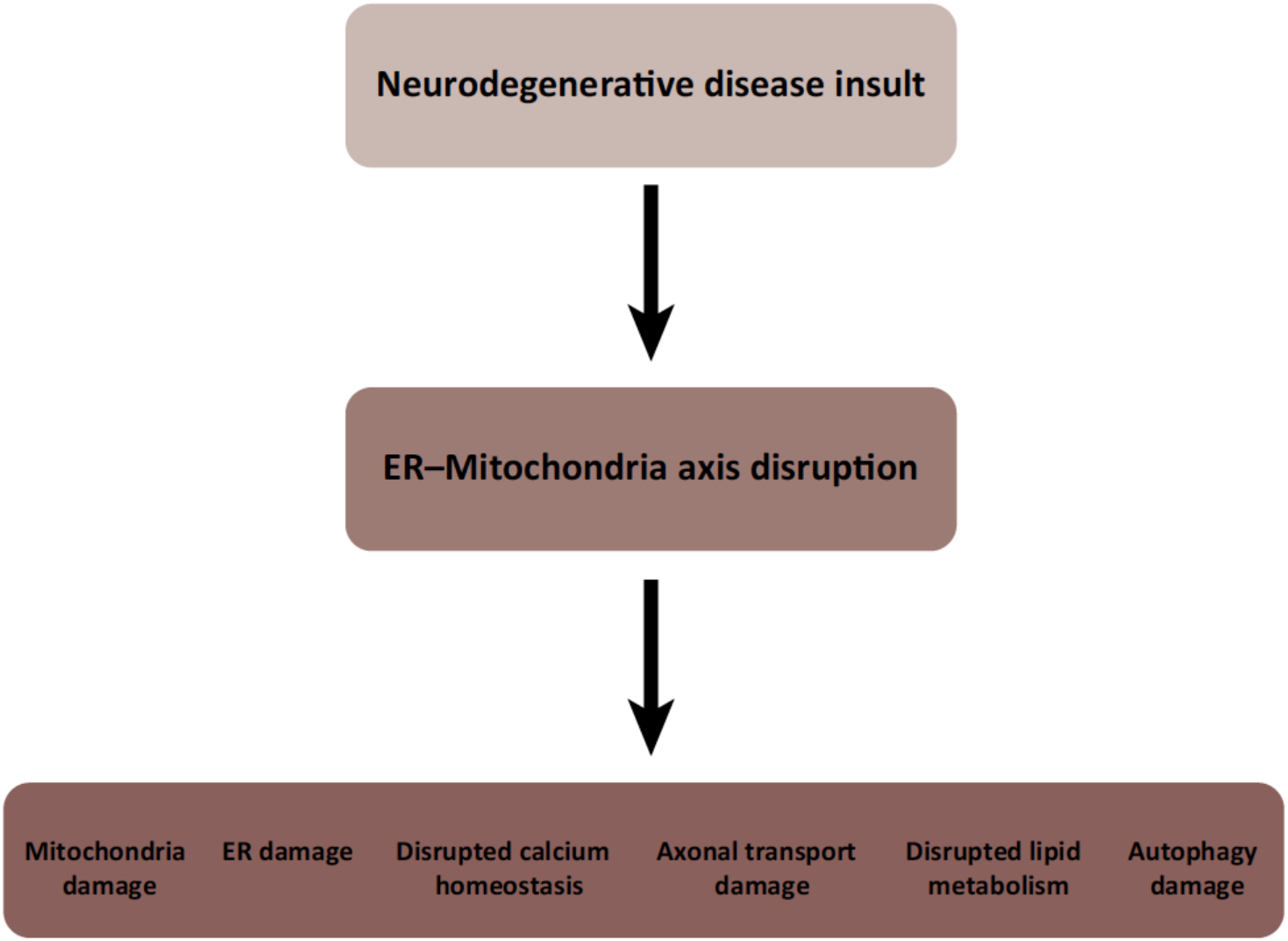
Barneo-Muñoz et al., PLOS Genetics 2015

Inherited neuropathies



Rossor, A.M. et al., Nat. Rev. Neurol. 2013

Disruption to ER–mitochondria associations provides a mechanism by which many of the disparate pathological features of neurodegenerative diseases might arise



Paillusson et al., Trends in Neurosciences 2016

Lausanne / Stockholm

H. Azzedine

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AFMTÉLÉTHON CURE THROUGH INNOVATION

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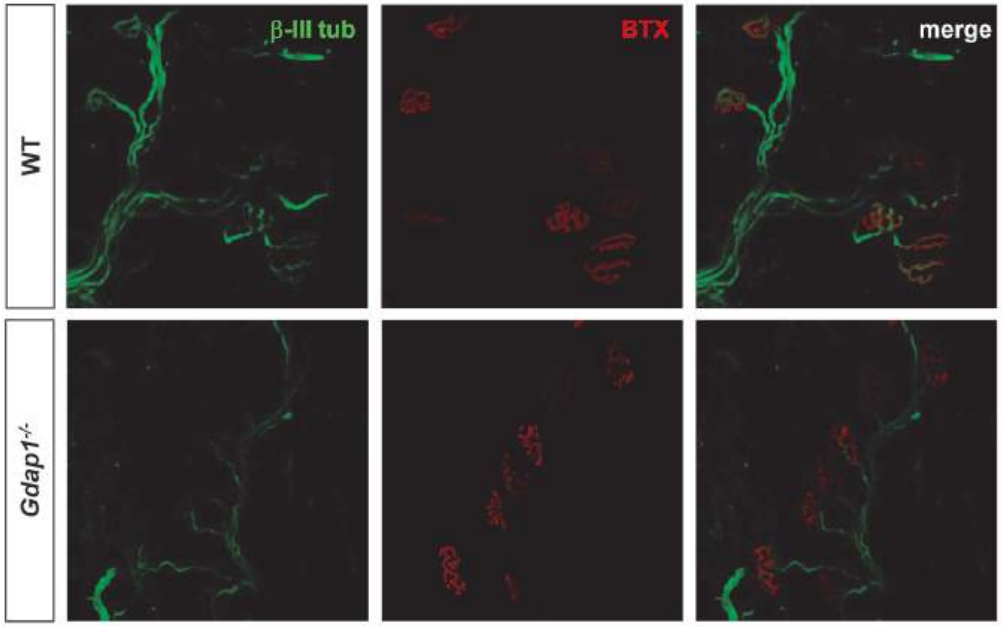
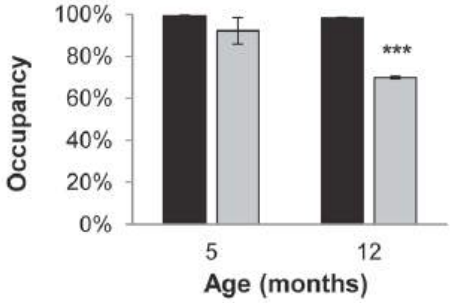
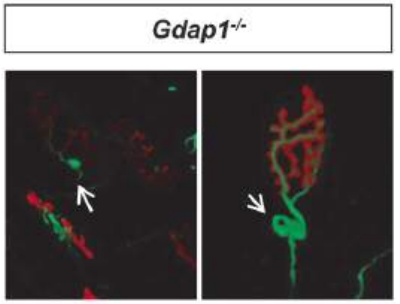
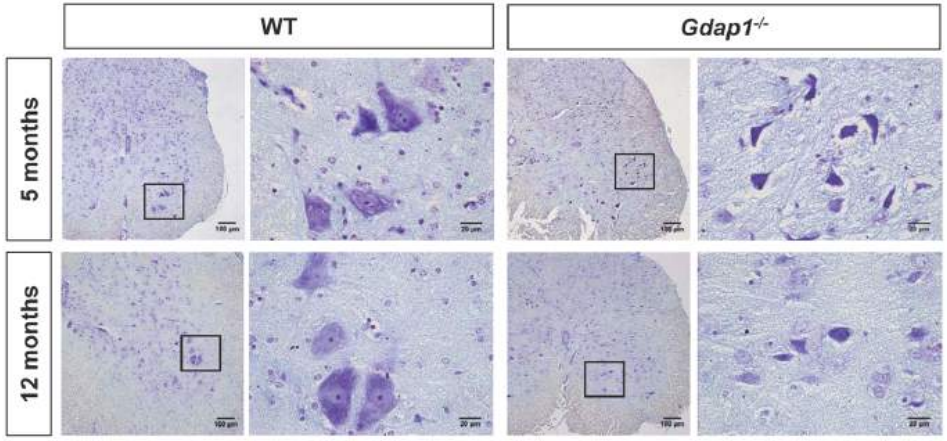
Karolinska Institutet

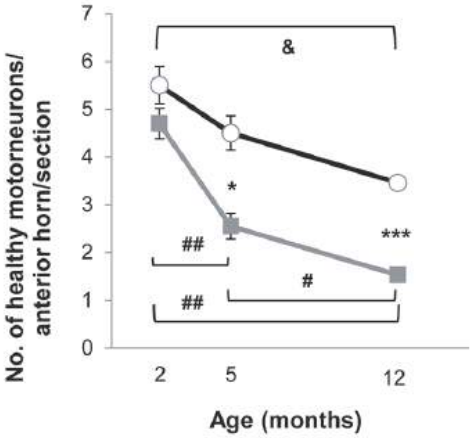
Lack of GDAP1 leads to loss of motor neurons and abnormal neuromuscular junctions





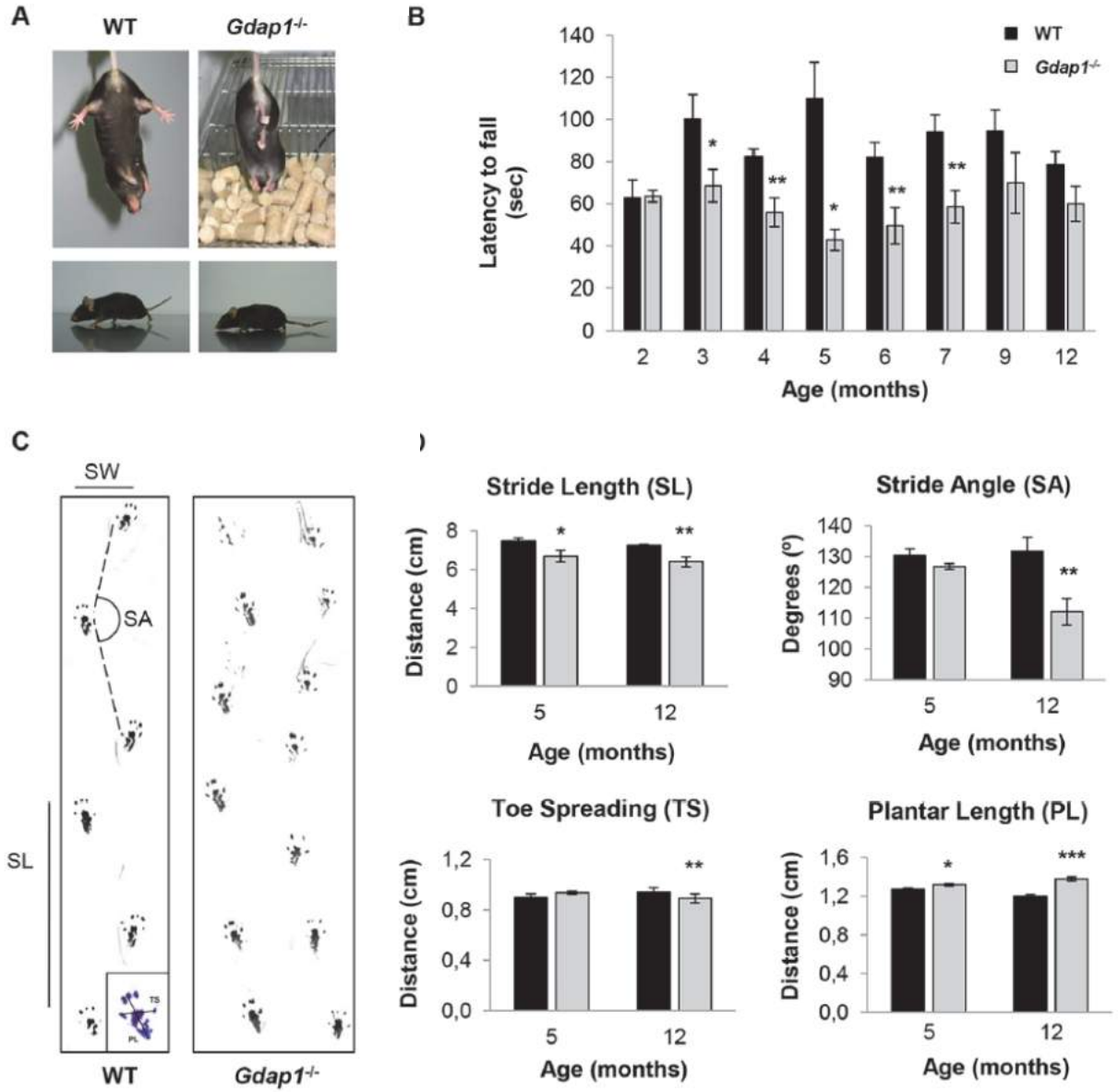






Barneo-Muñoz et al., PLOS Genetics 2015

Behavioral phenotypes in Gdap1$^{-/-}$ mice



Barneo-Muñoz et al., PLOS Genetics 2015