



Neuronal Mitochondrion Trafficking

BCH441 Project: Defining a System

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December 26, 2015

The source code, notebook, and data pipeline can be found at github.com/thejmazz/biologicalsystem.
Cover image (mitochondrion in Purkinje neuron) by *Atlas of Ultrastructural Neurocytology*¹

¹synapses.clm.utexas.edu/atlas/1_1_2_8.stm

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1 Introduction

The “powerhouse of the cell” as it is so commonly called, the mitochondria is one of the most vital organelles in eukaryotes. This structure is thought to have developed through a symbiotic relationship among engulfed prokaryotic cells and their hosts. As such, it is rooted quite deeply evolutionarily, and one might expect its proper functioning to be absolutely vital, that is, knock-out mutants will not survive. This is true - but as we will see, it is not just the performance of this organelle which is centrally important, but where it is localized within the cell as well.

Images of isolated mitochondria were first observed in 1979 by Johnson et al.:

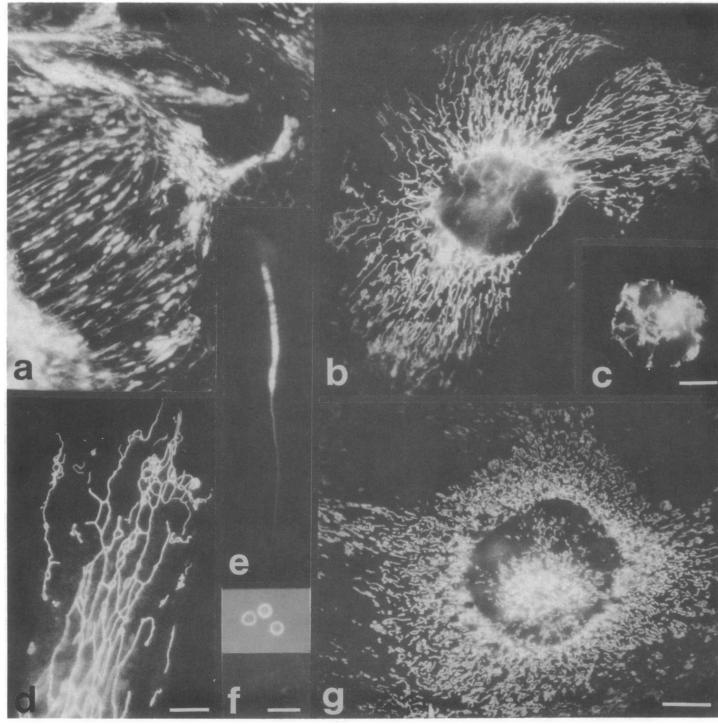


FIG. 5. Living cells stained with rhodamine 123: (a) rat cardiac muscle; (b) Pt KI marsupial kidney; (c) mouse B lymphocyte; (d) mouse 3T3; (e) mouse sperm; (f) human erythrocytes (phase-contrast above and rhodamine 123-treated but unstained below); (g) rat embryo fibroblast. Bar represents: 15 μm in a, b, e, and g; 10 μm in c; 8 μm in d; 10 μm in f.

The variety of mitochondrion shape and size is clear, ranging from globular to filamentous to networked structures. As well, the authors observed movement during 15-30 sec intervals, between fluorescent and phase-contrast photographs.

The primary role of a mitochondrion is to supply energy to the cell in the form of ATP units, through the electron transport chain among the cristae. Where is that energy needed? Consider highly polar and elongated cells such as neurons. The cell body of a neuron is distant from its synaptic endings, where as it happens, large amounts of energy are required for neurotransmitter release and absorption. Following, we will investigate the **system** whose **functional role** is the **localization of mitochondrion within neurons**.

2 The System

<i>Name</i>	Localization/Trafficking of mitochondrion within neurons
<i>Description</i>	The collective of functional units represented by genes which process signals, transduce these events, initiate, and maintain the actions necessary to transport mitochondrion to distal points along the axon of a neuron.
<i>Associated GO Terms</i>	GO:0051646 (mitochondrion localization) <ul style="list-style-type: none"> • GO:0051659 (maintenance of mitochondrion localization) <ul style="list-style-type: none"> – GO:1990456 (mitochondrion-ER tethering) • GO:0034643 (establishment of mitochondrial localization, microtubule mediated) <ul style="list-style-type: none"> – GO:0034642 (mitochondrial migration along actin filament) – GO:0034643 (establishment of mitochondrial localization, microtubule mediated) <ul style="list-style-type: none"> * GO:0034640 (establishment of mitochondrion localization by microtubule attachment) * GO:0047497 (mitochondrion transport along microtubule) – GO:0090146 (establishment of mitochondrial localization involved in mitochondrial fission)

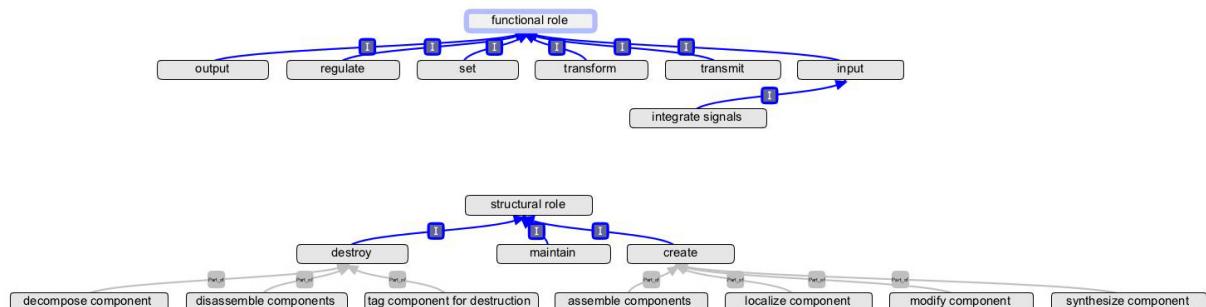
- * [GO:0090147](#) (regulation of establishment of mitochondrion localization involved in mitochondrial fission)
- [GO:0048311](#) (mitochondrion distribution)
 - [GO:0048312](#) (intracellular distribution of mitochondria)
 - [GO:0000001](#) (mitochondrion inheritance)

Why this system? Originally I was looking into “mitochondrial localization.” Amongst the genes returned by the ontology, there appeared those related to mitochondrial localization during cellular reproduction, transport, microtubules, tethers, mRNA-binding, and various “popular” genes such as ubiquitins, serum albumin, leucine-rich repeat serine/threonine-protein kinase, basic helix-loop-helix protein. There was a fair amount of variety. In order to gather together a structured list of genes I would need to filter these out, and to filter these out I would need a functional goal. I decided to choose the neuronal process because it is one of the most extreme cases of mitochondrial movement in all cell types, there was a decent amount of related literature available, some elements of its processes had been recently elucidated, and it has important neurophysiological consequences. A review by Reis et al. (2009) explored the atypical Miro GTPases and their role in transporting mitochondria in neurons. The authors note that abnormal mitochondrial dynamics can contribute to Amyotrophic Lateral Sclerosis (ALS), Huntington’s, Parkinson’s, and Alzheimer’s diseases. A more recent experiment by Loss and Stephenson (2015) examines the role of TRAK1 and TRAK2 kinesin adaptor proteins which link mitochondria to kinesin motor proteins. Furthermore, Miro proteins are expressed in a large variety of cell types, potentially extending this current analysis to new domains (Reis et al., 2009).

2.1 Systems Role Ontology

To define this system in a structured manner, I considered its functionality in the context of the Systems Roles Ontology, which can be found at github.com/hyginn/SyRO:

SyRO – Systems Role Ontology (2015-10-27)



Name	ID	Context Within Neuronal Mitochondrion Localization
input	16	compounds or signals prompting the directed movement of mitochondrion
integrate signals	22	machinery which directs input signals/compounds to the system
output	21	mitochondrion transport towards synaptic endings
regulate	20	components which ensure regular mitochondrial motility
set	19	preparing the functional units, “setting the stage” as it were
transform	18	altering the system status so that it may be reversed, stopped, reinstated
transmit	17	the physical machinery to move mitochondria

These functions collaborate to produce the **functional role** of localizing mitochondria **for the proper functioning of neural communication**. I will define the bounds of this system as any genes which can be annotated with the functional role annotations above. Structural role annotations will be considered second to function, and will be used largely to describe how that component physically determines its function. With this goal in mind, I fetched and filtered data from the Gene Ontology Consortium, QuickGO, UNIPROT, IntAct, and STRING. Those genes which did not make the cut can be seen through the *Summary of first pool* in my [notebook](#).

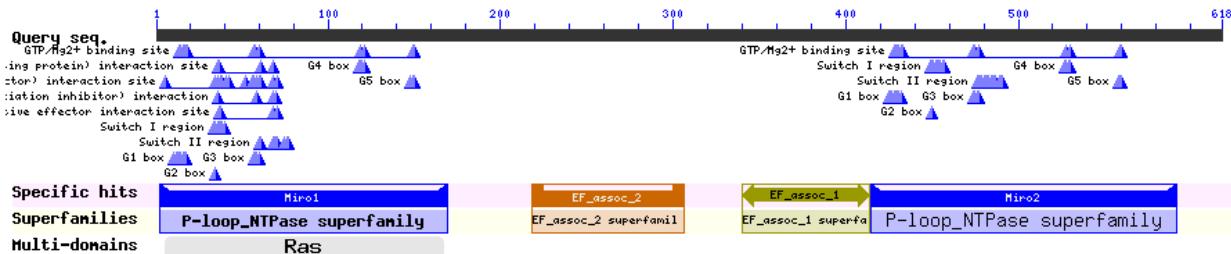
3 Gene Collection

Gene	Accession	Name	SyRO
ATCAY	Q86WG3	Caytaxin	regulate
BHLHA15	Q7RTS1	Class A basic helix-loop-helix protein 15	transmit
CLUH	I3L2B0	Clustered mitochondria protein homolog	set
GABA	P80404	Gamma-amino-N-butyrate transaminase	transmit
GAN	Q9H2C0	Gigaxonin	transform
KIF1B	O60333	Kinesin-like protein KIF1B	output
KIF5B	P33176	Kinesin-1 heavy chain	output
KLCA1	Q07866	Kinesin light chain 1	output
MAPT	P10636	Microtubule-associated protein tau	set
MAP1B	P46821	Microtubule-associated protein 1B	set
MGARP	Q8TDB4	Mitochondria-localized glutamic acid-rich protein	regulate
MTM1	Q13496	Myotubularin	set
MSTO1	Q9BUK6	Protein misato homolog 1	regulate
RHOT1	Q8IXI2	Mitochondrial Rho GTPase 1	integrate signals
RHOT2	Q8IXI1	Mitochondrial Rho GTPase 2	integrate signals
SNPH	O15079	Syntaphilin	regulate
SYBU	Q9NX95	Syntabulin	set
TIAM2	Q8IVF5	T-lymphoma invasion and metastasis-inducing protein 2	integrate signals
TRAK1	Q9UPV9	Trafficking kinesin-binding protein 1	transmit
TRAK2	Q8IU62	Trafficking kinesin-binding protein 2	transmit
TTL	Q8NG68	Tubulin–tyrosine ligase	transform

3.1 Signal Integration

3.1.1 RHOT1, RHOT2 Mitochondrial Rho GTPase 1, 2

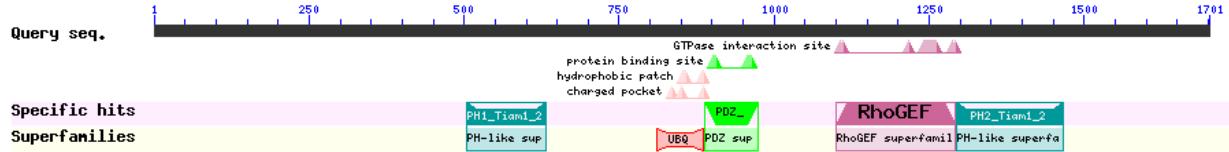
RHOT1 and RHOT2, which are also known as MIRO1 and MIRO2, were first reported as a new family of Rho GTPases with in 2003 by Fransson et al.. An NCBI CDD search presents us with a two GTPase domains each terminus, with two EF hands in between. The C terminal TM domain targets this protein to the mitochondria.



NTPases are a large superfamily and operate in a large variety of systems. The on/off state of these proteins confers their ability to integrate signals into a pathway. In this way we can observe that whichever is activating Rhot, Rhot is integrating that signal downwards to its effector. Fransson et al. also observed an interesting property: overexpression of Miro1/Val-13 led to an aggregation of the mitochondrial network. The same authors separated this response into two distinct phenotypes later in 2006. They observed that Miro-1 induced aggregation and thread-like mitochondria, whereas Miro-2 only induced aggregation. Fransson et al. also demonstrated interactions of Miro with GRIF-1 (TRAK2) and OIP106 (TRAK1), trafficking kinesin binding proteins.

3.1.2 TIAM2 T-lymphoma invasion and metastasis-inducing protein 2

TIAM2 regulates the activity of RHO-like proteins (UNIPROT). This is confirmed with the existence of a GEF domain from the CDD.



In this way TIAM2 has the ability to activate Rhot. TIAM2 has also been shown to promote the migration of neurons in the cerebral cortex (UNIPROT).

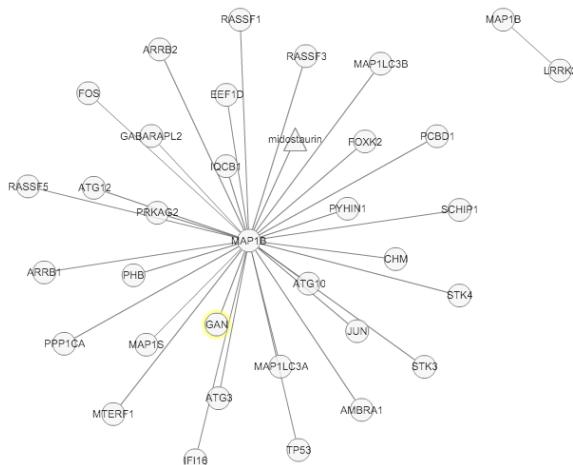
3.2 Set

3.2.1 CLUH *Clustered mitochondria protein homolog*

A CDD search with CLU1 from yeast presents the CLU domain (CLustered mitochondria). This domain is required for mitochondrial positioning and transport; improper function can lead to mitochondrion clustering at the microtubule plus ends (CDD).

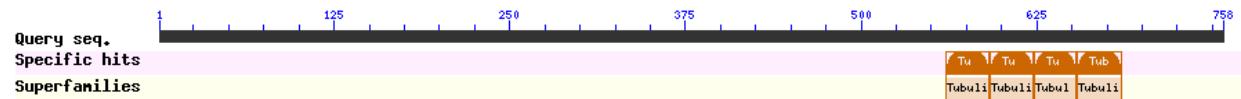
3.2.2 MAP1B *Microtubule-associated protein 1B*

By similarity, MAP1B facilitates tyrosination of α -tubulin in neuronal microtubules. Interacts with TIAM2 and TTL (UNIPROT). IntAct suggests an interaction with GAN.



3.2.3 MAPT *Microtubule-associated protein tau*

CDD for MAPT presents tubulin binding repeat domains. MAPT is expected to stabilize microtubules and potentially establish and maintain neuronal polarity (UNIPROT).

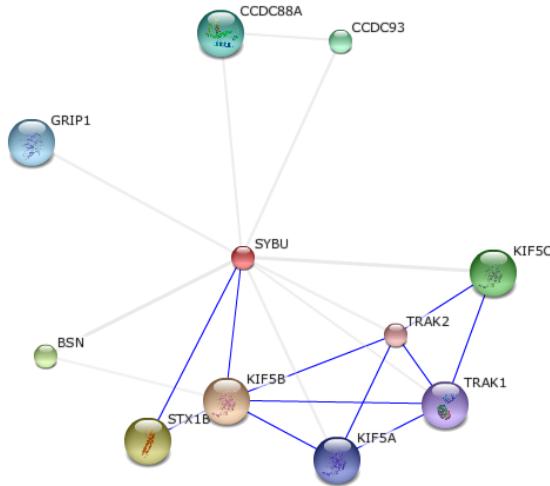


3.2.4 MTM1 *Myotubularin*

Hnia et al. (2011) observed that decreased MTM1 expression and mutations induced abnormal mitochondrial positioning, shape, dynamics, and function.

3.2.5 SYBU *Syntabulin*

SYBU belongs to a kinesin motor-adapter complex. It is critical for forward axonal transport (UNIPROT). STRING demonstrates binding interactions with KIF5B, which in turn binds with KIF5A and TRAK2, of which both bind TRAK1.



3.3 Transmit

3.3.1 BHLHA15 *Class A basic helix-loop-helix protein 15*

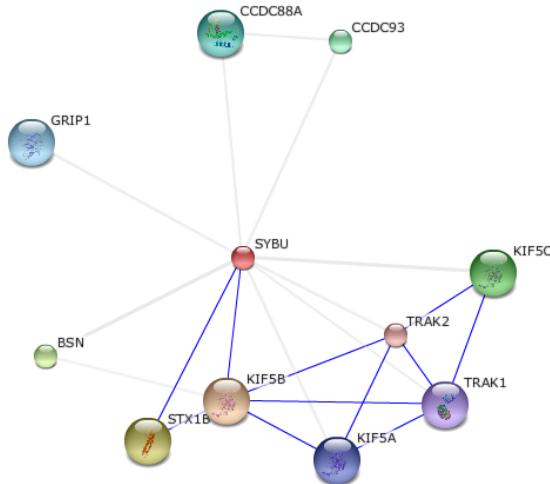
Required for mitochondrial calcium ion transport (UNIPROT).

3.3.2 GABA *Gamma-amino-N-butyrate transaminase*

GABA is located within the mitochondrial matrix (UNIPROT).

3.3.3 TRAK1 *Trafficking kinesin-binding protein 1*

Trafficking of GABA-A receptors (UNIPROT). Contains coiled-coiled domain. STRING corroborates with interactions for SYBU above.



3.3.4 TRAK2 *Trafficking kinesin-binding protein 2*

Milton domain. Interact with GABA-A receptors.

3.4 Transform

3.4.1 GAN *Gigaxonin*

Controls degredation of MAP1B and MAP1S; critical for neuronal maintenance and survival (UNIPROT).

3.4.2 TTL *Tubulin-tyrosine ligase*

ATP + detyrosinated alpha-tubulin + L-tyrosine = alpha-tubulin + ADP + phosphate

3.5 Regulate

3.5.1 ATCAY *Caytaxin*

May regulate the localization of mitochondria within axons and dendrites (UNIPROT).

3.5.2 MGARP *Mitochondria-localized glutamic acid-rich protein*

Regulates kinesin-mediated axonal transport of mitochondria to nerve terminals. Translocation of TRAK2 from cytoplasm to mitochondrion.

3.5.3 MSTO1 *Protein misato homolog 1*

Regulation of mitochondrial dist. and morphology.

3.5.4 SNPH *Syntaphilin*

Inhibits SNARE complex formation by absorbing free syntaxin-1.

3.6 Output

3.6.1 KIF1B *Kinesin-like protein KIF1B*

MT + end directed motility.

3.6.2 KIF5B *Kinesin-1 heavy chain*

Kinesin heavy chain.

3.6.3 KLC1 *Kinesin light chain 1*

Kinesin light chain.

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