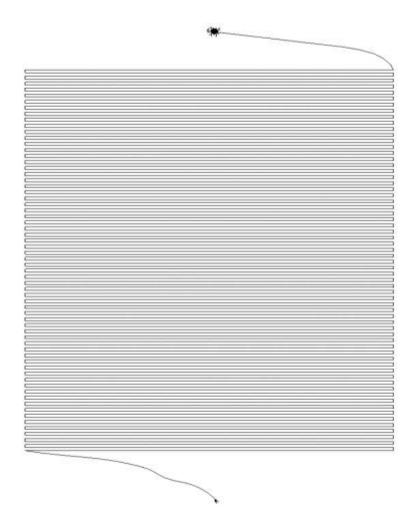




Historical Perspectives and Current Questions

Peter J. Hollenbeck Dept of Biological Sciences Purdue University West Lafayette, IN

School lecture 13 ICTS-TIFR Advanced School on Axonal Transport & Neurodegenerative Disorders 18 January 2013



The scientific method is our greatest "Baloney Detector"

Carl Edward Sagan (1934-1996) Professor of Astronomy, Cornell University



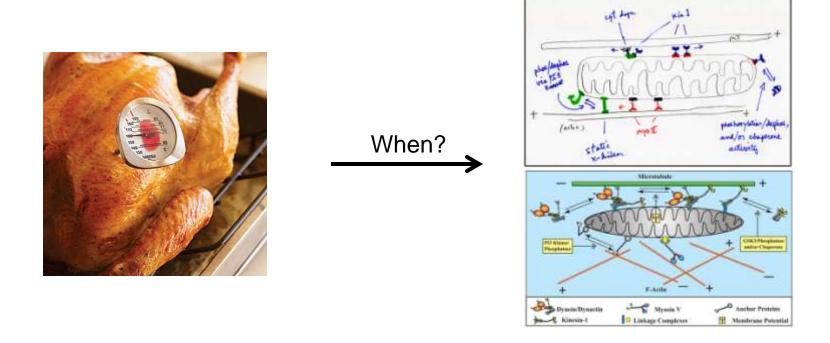
"Somewhere, something incredible is waiting to be known."

"For me, it is far better to grasp the Universe as it really is than to persist in delusion, however satisfying and reassuring."

On-line version of Sagan's Baloney Detector: http://users.tpg.com.au/users/tps-seti/baloney.html A more recent issue: lots of papers with lots of data, from lots of labs...

When is a body of work and its interpretation "fully cooked"?

When do we view something as settled science and draw the cartoon?

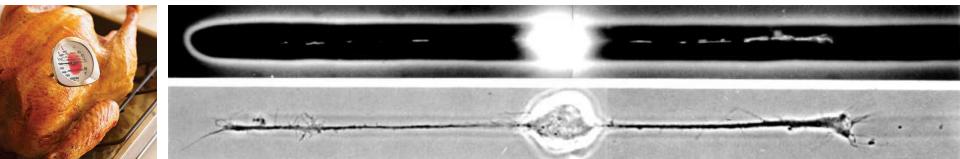


Are there facets of axonal organelle transport that are already in the "cartoon" stage but are perhaps still under-cooked?

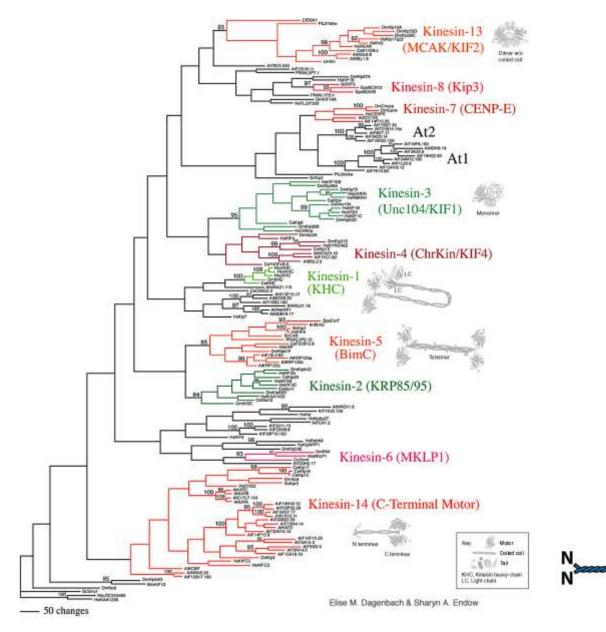
(1) Motor diversity and cargo diversity

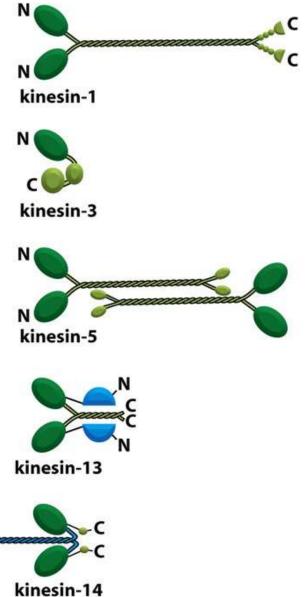
(2) Motor-organelle adaptor/regulator proteins

(3) Neuronal polarity and organelle traffic

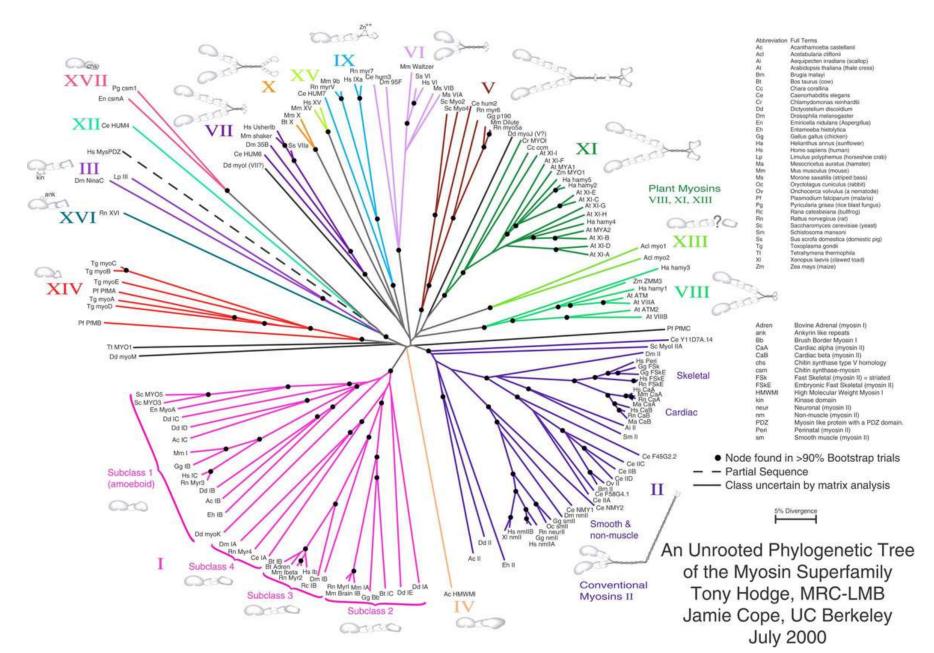


Kinesin: 14 families, plus orphans

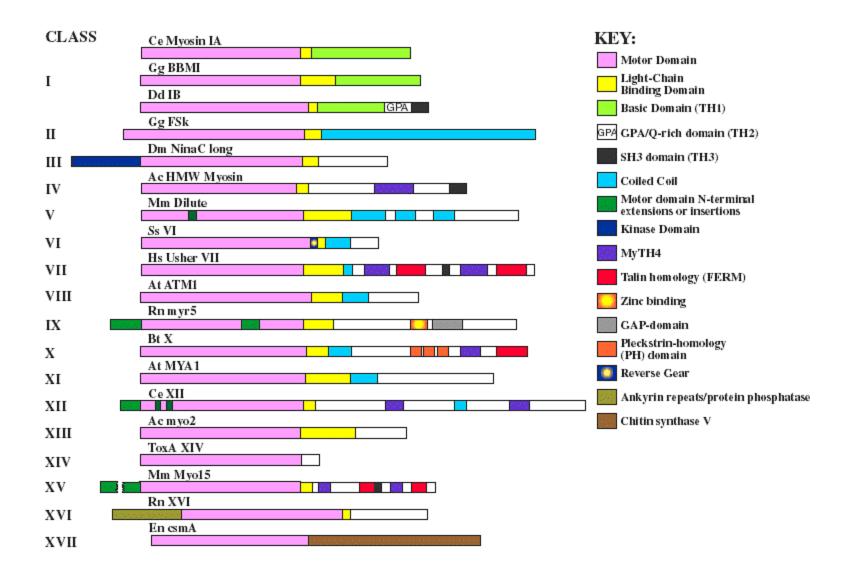




Myosin: 17-19 families



Myosin: domain structures



Mark Mooseker Ross Granville Harrison Professor of MCD Biology Yale University School of Medicine

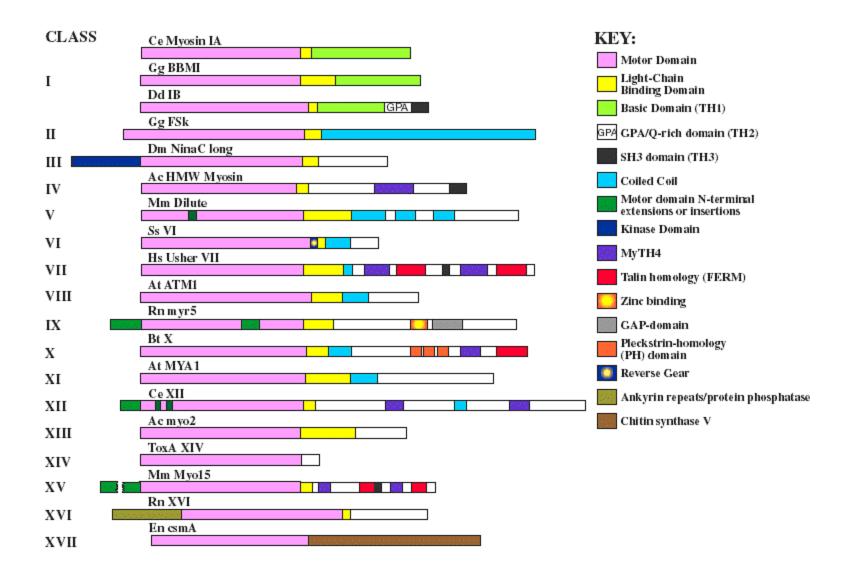


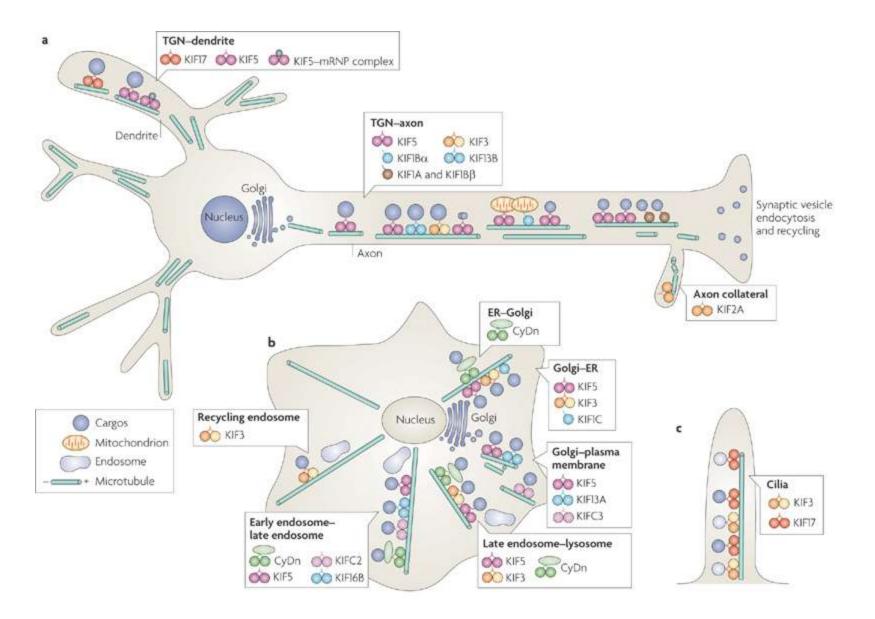
1990s: PCR approach revealed multitude of myosins

"Are you kidding? I'm miserable. The interesting part is over. With this many myosins, there'll be a generation of nothing but spade-work in cell biology."

--After a seminar at HMS, around 1996

Myosin: domain structures





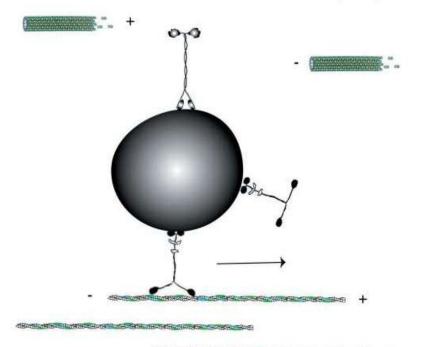
Nature Reviews | Molecular Cell Biology

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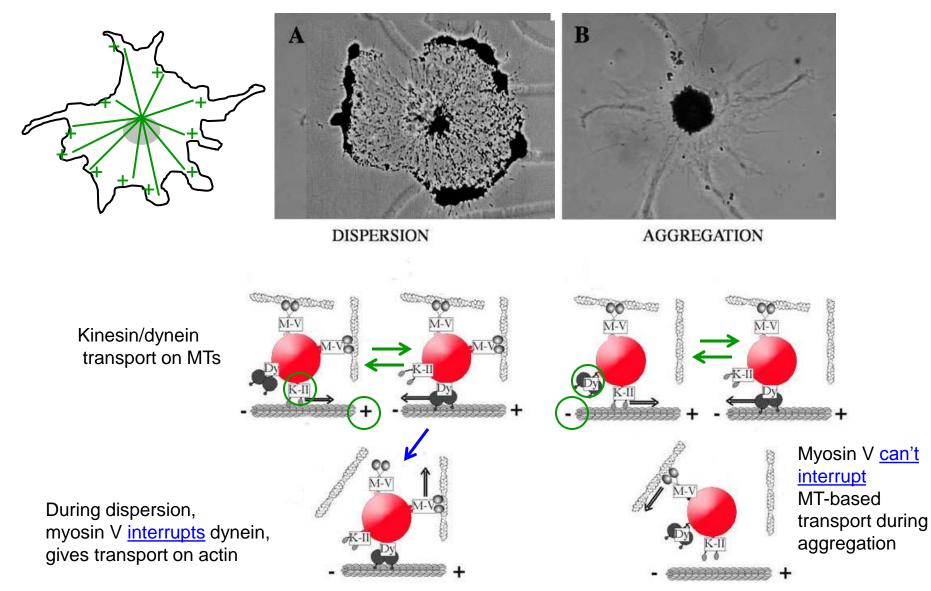
Sorry, you're stuck reading these!

Turn-of-the-millenium model: how do actin- and MT-based transport cooperate to drive FAT?



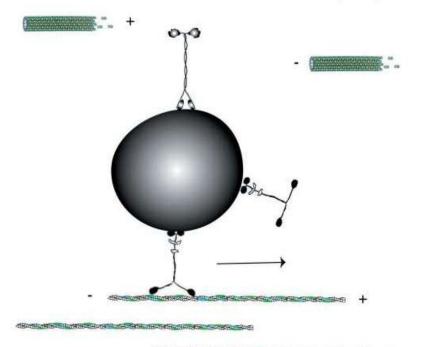
Myosin-Dependent Transport

Pigment cells: tug of war between actomyosinand MT-based movements



Gross et al, 2002

Turn-of-the-millenium model: how do actin- and MT-based transport cooperate to drive FAT?



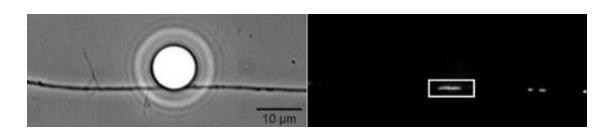
Myosin-Dependent Transport

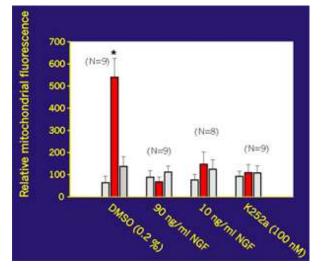
Mitochondrial movement and docking

Chick DRG neurons in culture:

When axonal actin is extensively depolymerized, mitochondria move faster and more persistently. (Morris & Hollenbeck, 1995)

Mitochondria can be halted along the axon in response to local NGF/TrkA signaling, with a role for PI3K signaling downstream. However, in the absence of F-actin, mitochondria do not dock (Chada & Hollenbeck, 2003, 2004).

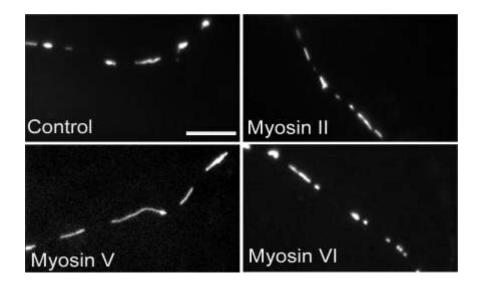


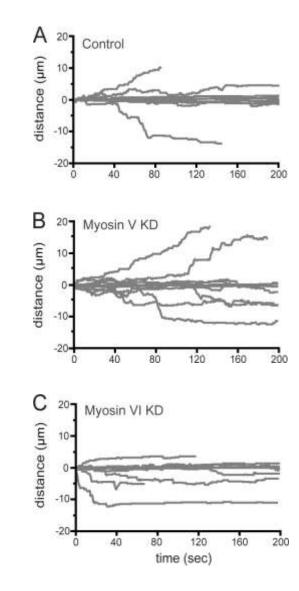


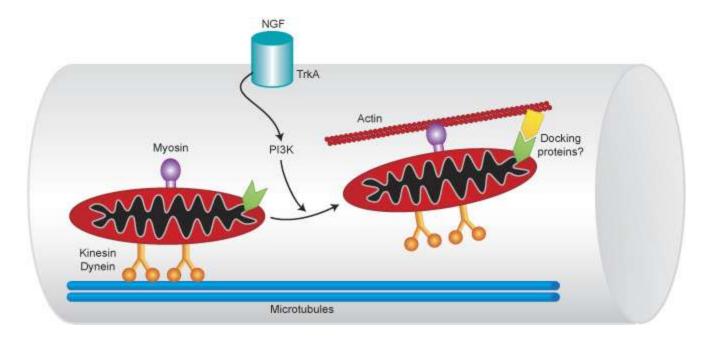
Mitochondrial movement and docking

Drosophila neurons in culture:

RNAi knock down of myosin V results in increased flux of axonal mitochondria, which move faster, more of the time, and more persistently than when normal myosin levels are present (Pathak et al, 2010).







Reynolds & Rintoul, Science STKE, 2004

Before we become satisfied that the science is settled, and rely on a cartoon mechanism, we need to have first-rate studies from :

more than one system, cell type or species.

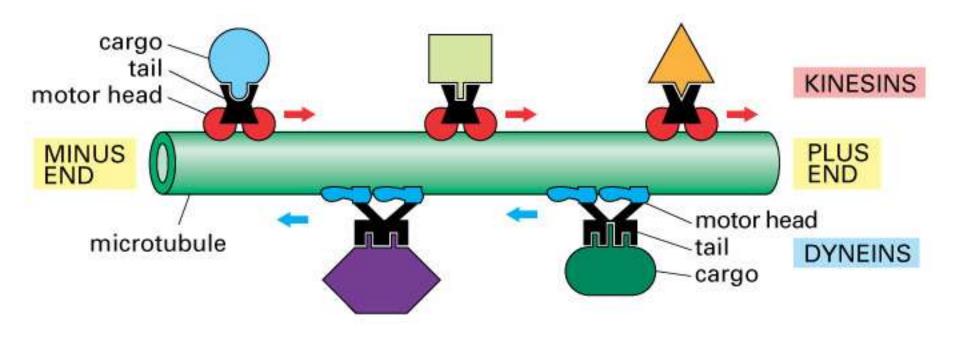
- vertebrates, invertebrates with different features
- CNS, PNS, special senses, etc

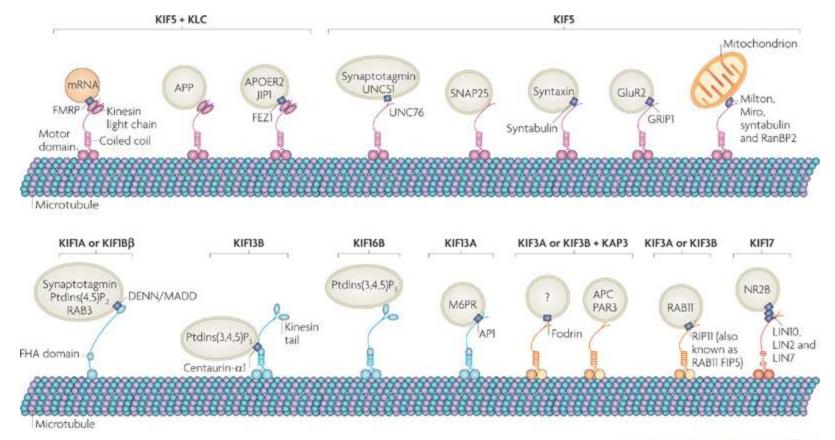
more than one technical approach.

- anatomy, EM, LM
- quantitive cellular phenotype
- in vitro systems
- biochemistry & physiology
- genetics

more than one laboratory.

Motor-organelle adaptor proteins: getting the right motor on the right cargo

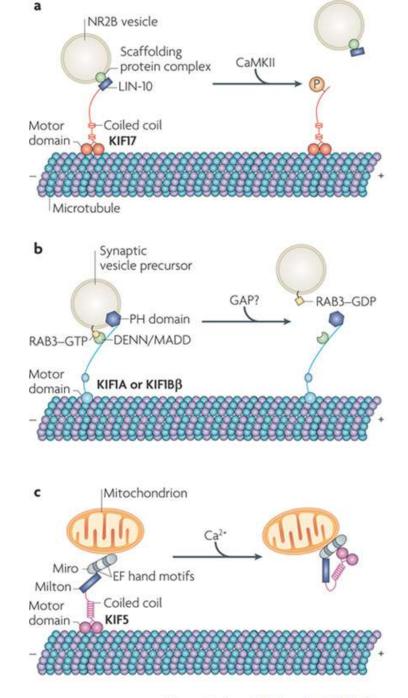




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¹ Attempting protein constructions are provided by construction protein strugglers 1, 99°, additional strugglers and provide and protein strugglers 1, 99°, additional strugglers strugglers 1, 99°, additio



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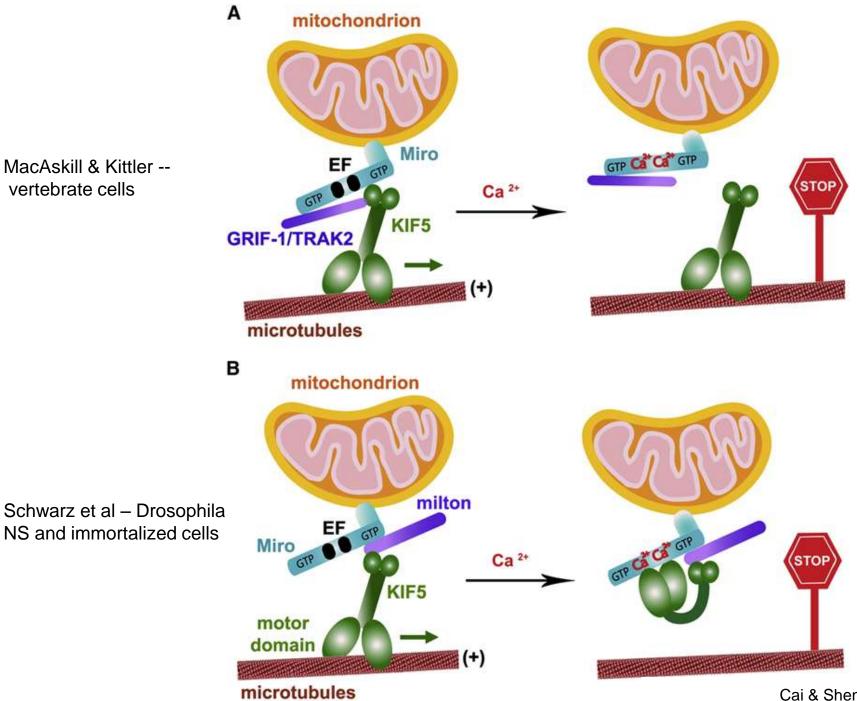
Example of a recent, exciting result: a complex that serves as a motor-mitochondrial adaptor and transport regulator: Miro/Milton and Ca regulation of FAT

Milton mutant fly: fails to deliver mitochondria to distal axon (Schwarz lab)

Miro mutant fly: fails to deliver mitochondria to synapses (Zinsmaier lab)

Studies of GRIF1/Milton and Miro-1 in vertebrate neurons (Kittler lab)

Studies of Milton and Miro in fly NS and various cell lines and neurons (Schwarz lab)



Cai & Sheng, 2009

Other organelle-kinesin adaptors and regulators "in play"

•JIP scaffolding proteins (kinesin-vesicle)

•RanBP2 (kinesin-vesicle etc)

•APP (kinesin-vesicle, contentious)

•Kinectin (kinesin-vesicles, gone but not forgotten :-)

•Syntabulin (kinesin-vesicle or mitochondria?)

•Syntaphilin (docking, mitochondria)

•HUMMR (anterograde regulator, mitochondria)

•GluR2-receptor interacting protein (GRIP1, kinesin-vesicle, dendrites)

•Dynactin-dynein (any cargo specificity?)

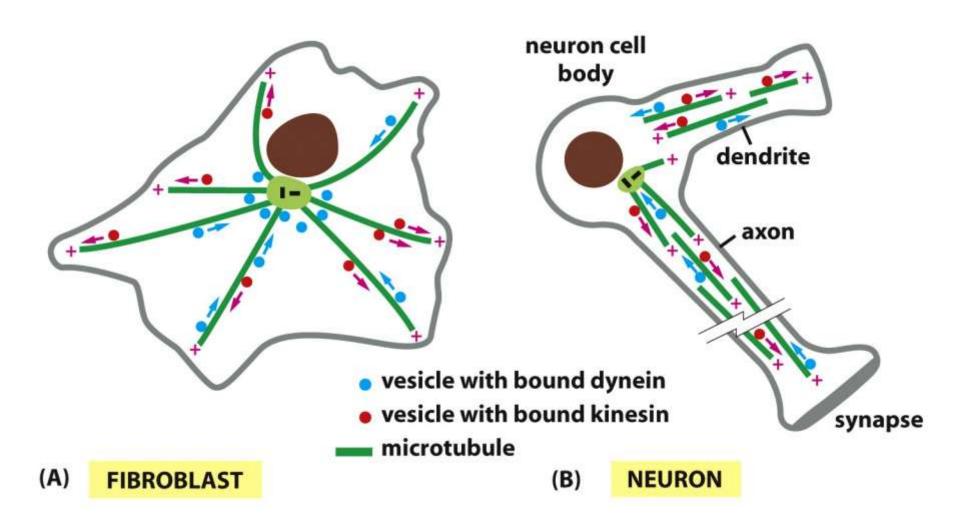
Neuronal polarity and organelle transport

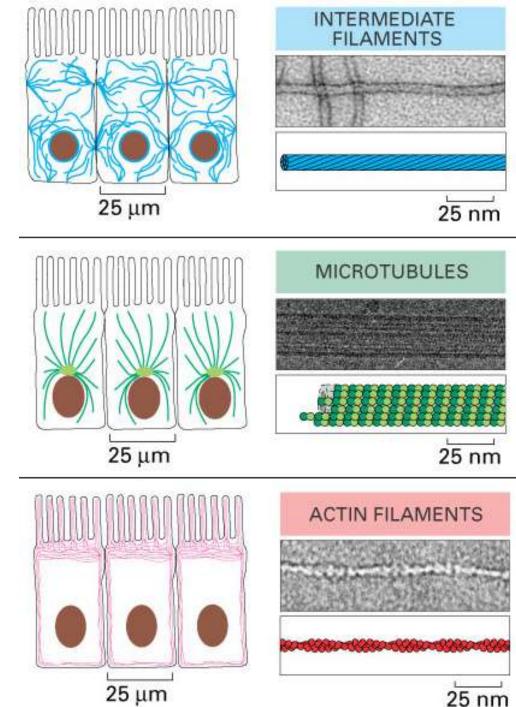
<u>Axons vs dendrites</u>: almost uniform (axon) vs mixed (dendrite) polarity of MTs is now bankable.

But what does it mean for organelle traffic? Can MT polarity and motors keep some organelles in or out of axons, or dendrites?

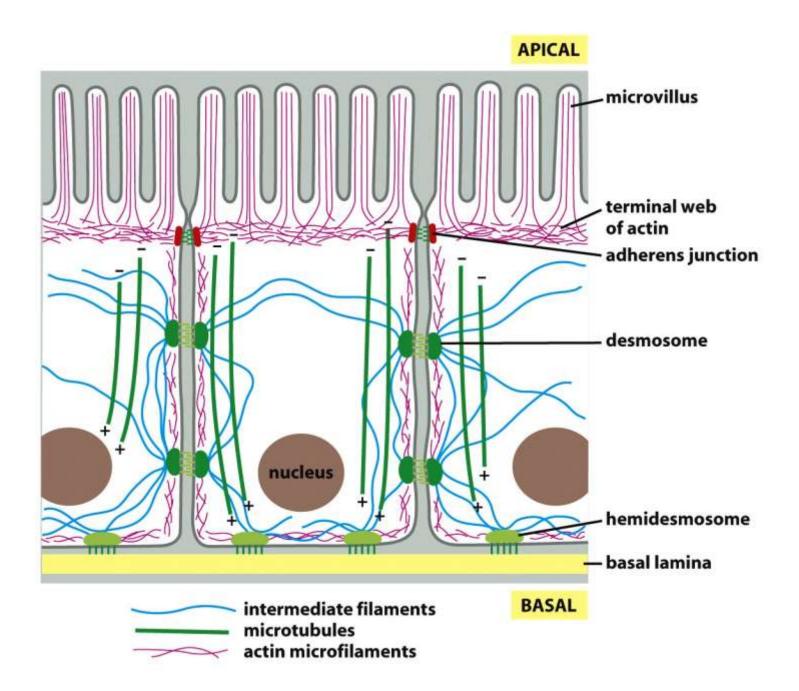
<u>Epithelial cells vs neurons</u>: existing models posit the cell polarity of these two v different cell types to be analogous – the distal axon being equivalent to the apical domain of epithelial cells.

There are similarities, but organelle traffic on MTs cannot be one of them – epithelia in real tissues have their (-) ends apical!



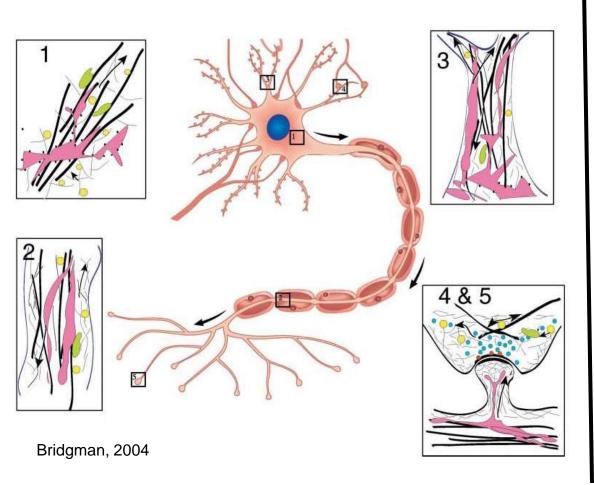


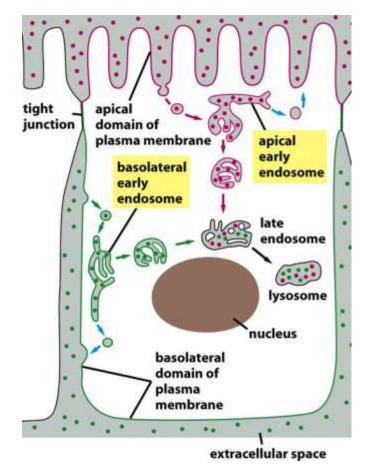
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Some features of organelle traffic, secretion, endocytosis, etc are similar between the apical domains of epithelia the growth cone of synapse of a neuron.

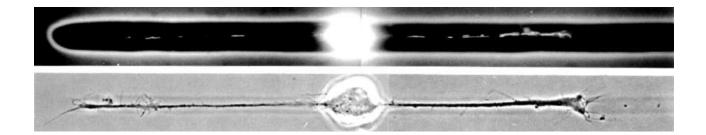
Others are not!





Other exciting and "in play" axonal transport areas:

- •Regulation of motor activity & function (refer to STB talk!)
- Motor cooperativity / tug of war
- •Slow axonal transport of "soluble" proteins (ask SR!)
- •(Any you'd like to add?)



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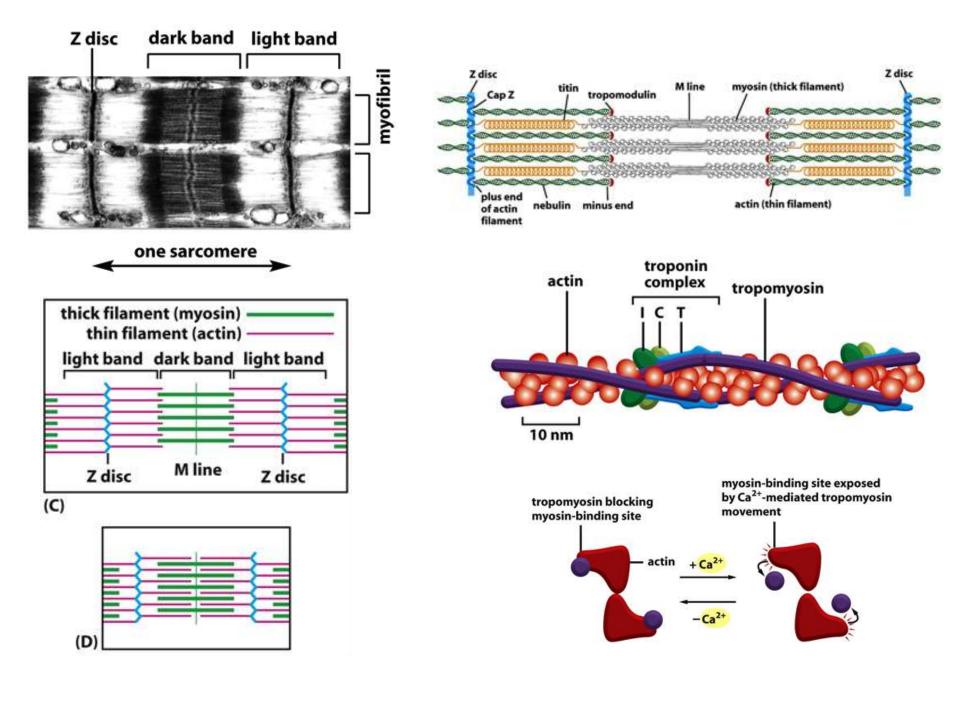
more than one system, cell type or species.

- vertebrates, invertebrates with different features
- CNS, PNS, special senses, etc

more than one technical approach.

- anatomy, EM, LM
- quantitive cellular phenotype
- in vitro systems
- biochemistry & physiology
- genetics

more than one laboratory (many more).







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