U.S. DEPARTMENT OF ENERGY | Office of Science

SAND2013-3494C

Active Assembly of Dynamic and Adaptable Materials

April 23-24, 2013

George D. Bachand

Requested Funding				
	FY13	**FY14**	**FY15**	**Total**
Operating	$1,290K	$1,290K	$1,290K	$3,870K
Capital	$ 0K	$ 0K	$ 0K	$ 0K
Total	**$1,290K**	**$1,290K**	**$1,290K**	**$3,870K**

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Sandia National Laboratories

Outline and Agenda

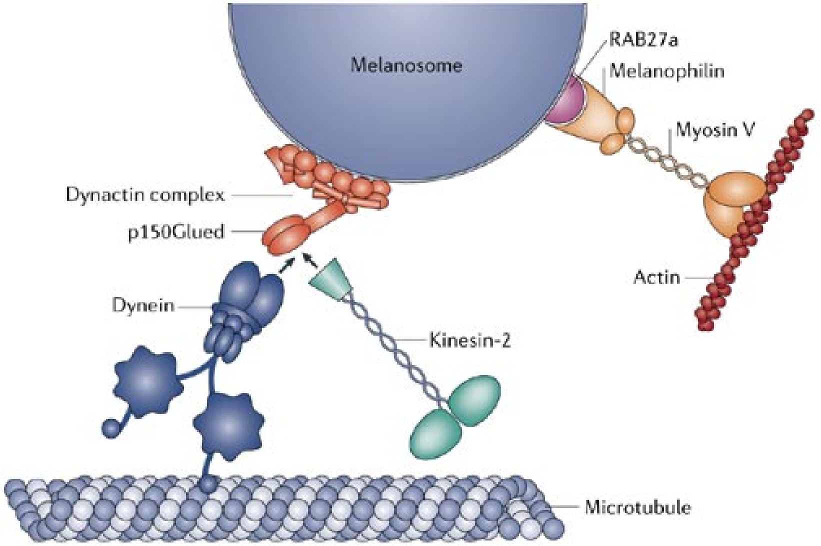
- **Project Description**
  - Project Goal and Objectives
  - Background and Significance
  - Project Structure
- **Project Team & Expertise**
  - Investigator / Institution / Role
- **Project Highlights**
  - Task 1: Active Protein Assemblies
  - Task 2: Artificial Microtubules
- **Future Work**
  - Task 1: Active Protein Assemblies
  - Task 2: Artificial Microtubules
- **Acknowledgments**

Project Description

- **Project Goal:**
  - Explore the extent to which energy-consuming proteins can be used in artificial systems for the active transport, assembly, and reconfiguration of nanomaterials

Mesoscale Behaviors & Active Proteins

Video Credit: Caldwell Lab, UC-Berkeley

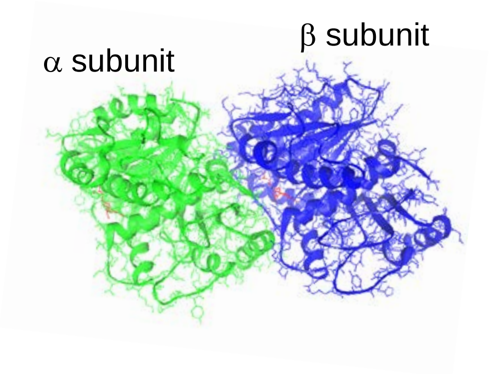


Soldati & Schliwa, 2006, Nat. Rev. Mol. Cell Biol., 7, 897

Background & Significance

Kinesin motors and microtubule (MT) filaments – active transport of cellular nanomaterials; enable emergent behaviors

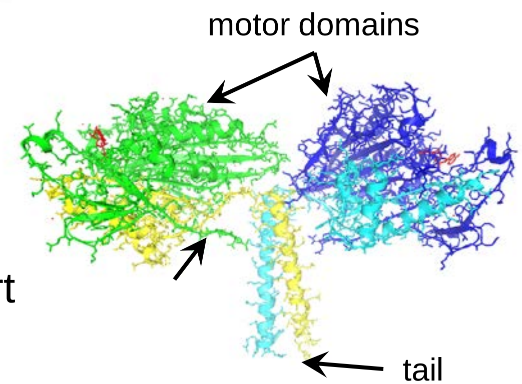
Can these principles, components, and/or synthetic analogs be used to assemble dynamic and adaptive materials?



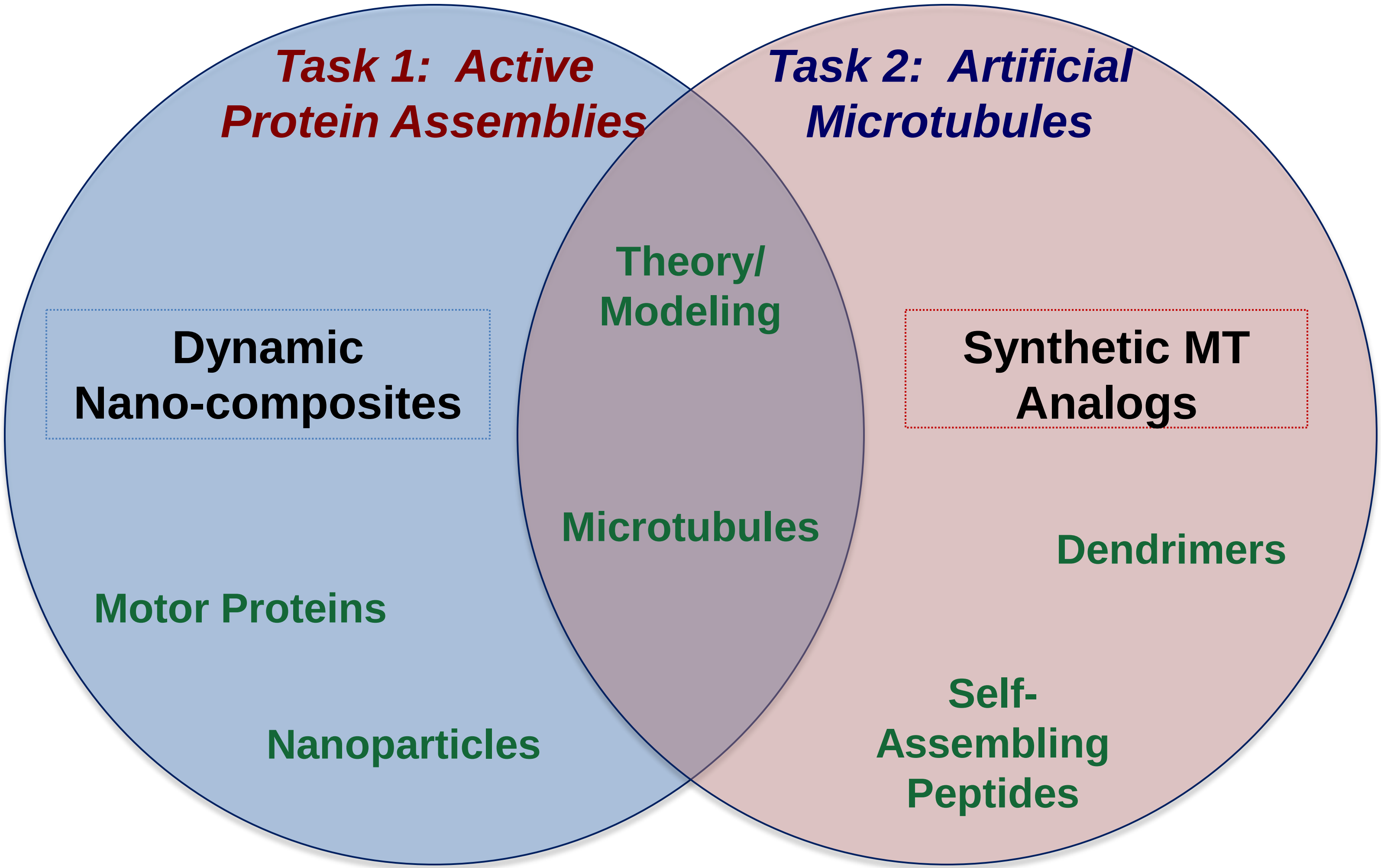
Tubulin
- Dimer
- Fuel = GTP
- Polymerization forms MT filaments

Video Credits: “Inner Life of Cell”
Conceptualized by *Dr. Alain Viel Ph.D.*, and *Dr. Robert Lue Ph.D.*, Molecular and Cellular Biology, Harvard University
Animated by *John Lieber* of XVIVO, Inc.
Funded by the *Howard Hughes Medical Institute*

Kinesin
- Dimer
- Fuel = ATP
- Use MTs as “tracks” for materials transport



Project Structure



Key Expertise

Investigator	Role
George Bachand (SNL)	PI & Task Lead (Active Proteins) + biomaterials
Erik Spoerke (SNL)	Task Lead (Artificial Microtubules) + bio-inspired materials
Bruce Bunker (SNL)	Former PI, chemistry of programmable materials <u>(retired SNL)</u>
Darryl Sasaki (SNL)	Lipid synthesis, supported bilayers, vesicles
James McElhanon (SNL)	Dendrimers, polymer synthesis
Mark Stevens (SNL)	Theory & modeling of natural & artificial microtubules
Dominic McGrath (Univ. Arizona)	Dendrimer synthesis & self-assembly





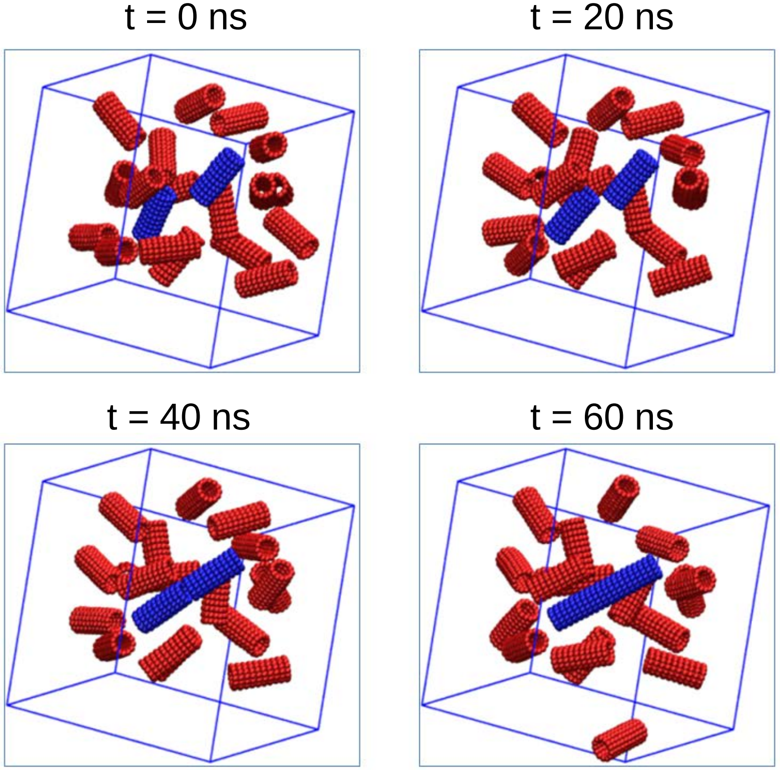


Self-Assembling MT Nanowires

Do stabilized MT grow, and if so, how?

- MD simulations – large oligomers and even assembled tubes can “fuse” during assembly
- Do natural MTs grow through a similar mechanism?

Time-lapse from MD simulation

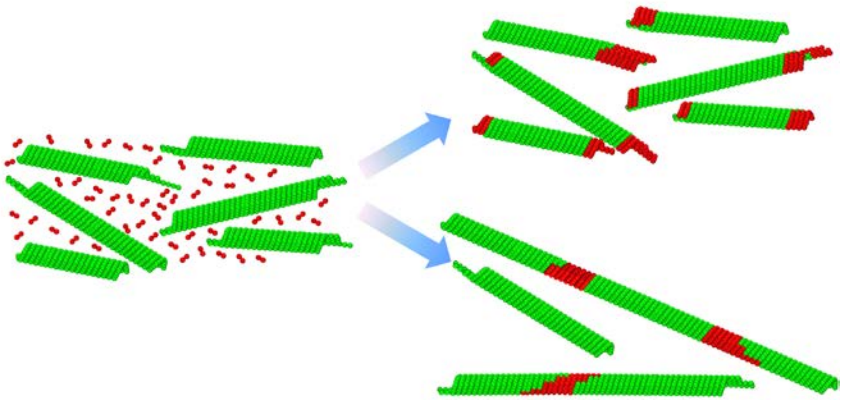


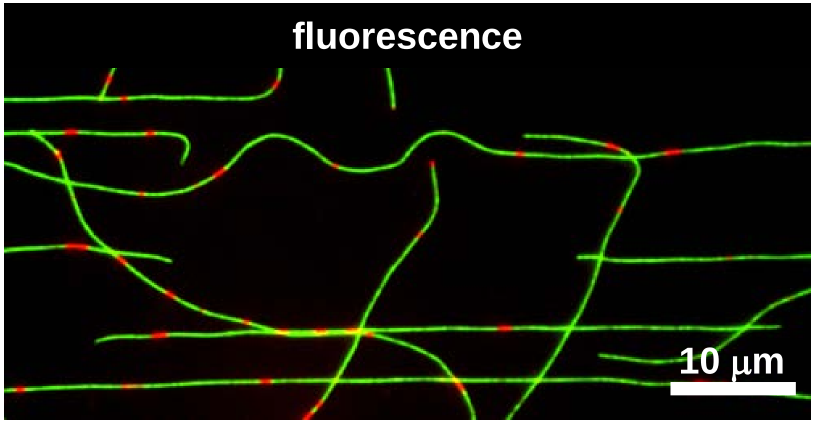
Impact: Established a mechanistic understanding of unique mode of MT growth *via* self-organization of stabilized filaments

Self-Assembling MT Nanowires

Do stabilized MT grow, and if so, how?

- MD simulations – large oligomers and even assembled tubes can "fuse" during assembly
- Do natural MTs grow through a similar mechanism?
- Unique MT growth mode:
  - [tubulin] >> [MTs] = spontaneous polymerization
  - [tubulin] > [MTs] = nucleated polymerization
  - **[tubulin] << [MTs] = MT fusion**



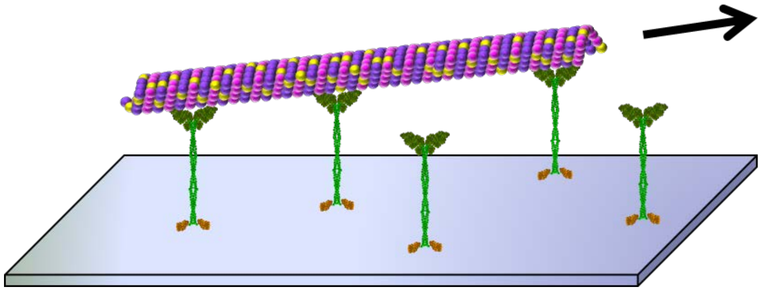


Impact: Established a mechanistic understanding of unique mode of MT growth *via* self-organization of stabilized filaments

Confining Motor-Driven Assembly

How does microscale confinement affect motor-driven self-assembly?

- Inverted motility of microtubule filaments
  - Kinesin monolayer on substrate with motor domains extending outward from surface
  - Microtubules "glide" across the kinesin monolayer when ATP is present
  - Gliding direction is stochastic and limited only by the presence of fuel



Impact: Understanding how energy dissipative assembly and microscale confine collaborate in non-equilibrium materials self-assembly

Liu & Bachand (2011), *Soft Matt*. **7**, 3087; Liu & Bachand (2013), *Cell. Mol. Bioeng.* **6**, 98

Confining Motor-Driven Assembly

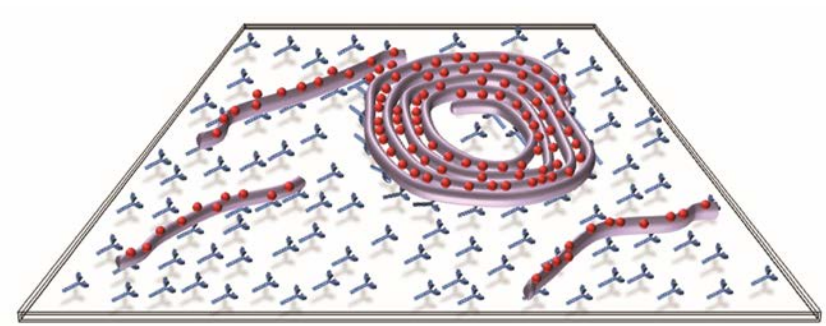
How does microscale confinement affect motor-driven self-assembly?

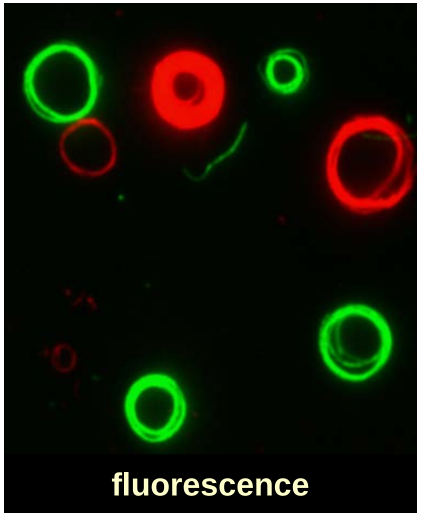
- Addition of nanoparticles (e.g., Qdots) to gliding microtubules leads to self-assembly of ring nanocomposites

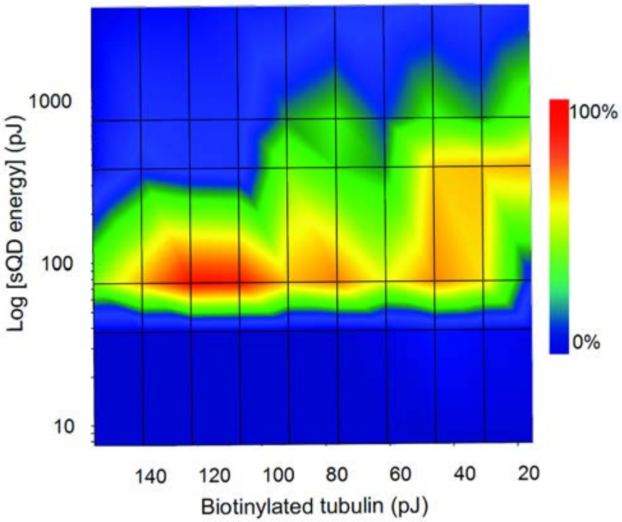
See:

Bachand et al., (2005), *J. Nanosci. Nanotechnol.*, **5**, 718;

Liu et al., (2008), *Adv. Mater.*, **20**, 4476







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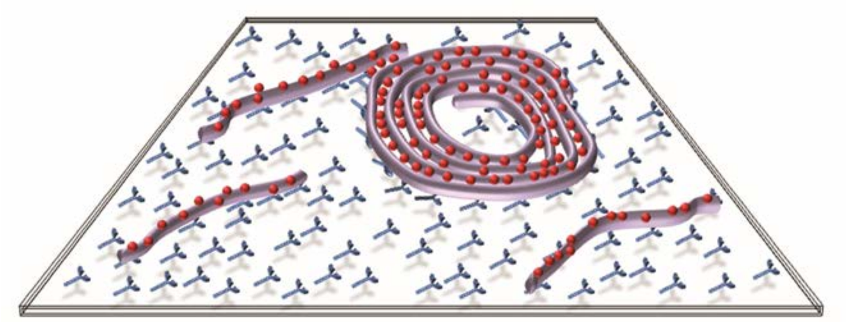
Confining Motor-Driven Assembly

How does microscale confinement affect motor-driven self-assembly?

- Physical patterns = channels and posts
- Selective kinesin adhesion using SAMs
- Kinesin adhesion and microtubule transport defined by chemical and physical confinement



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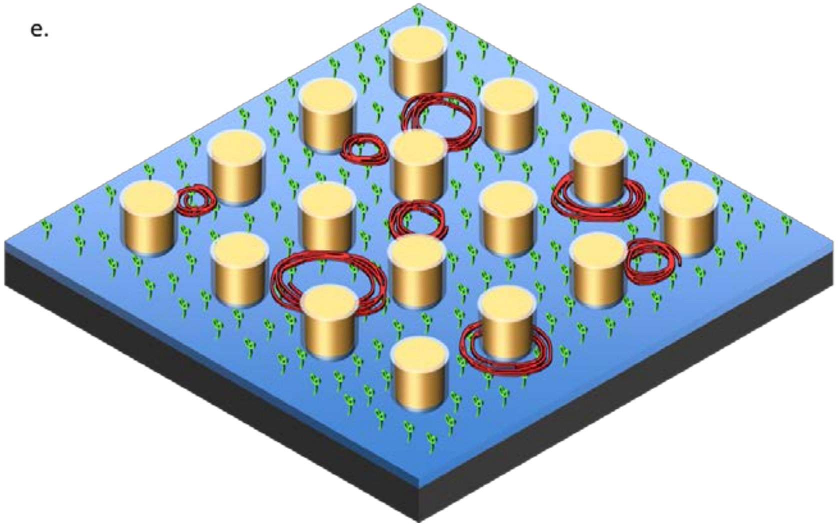

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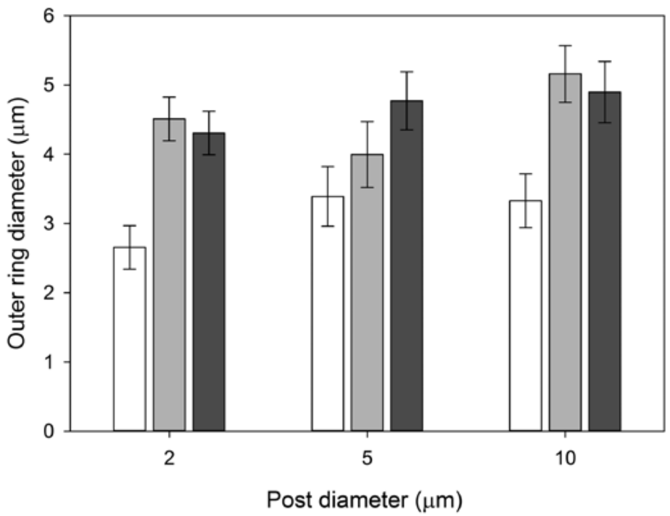
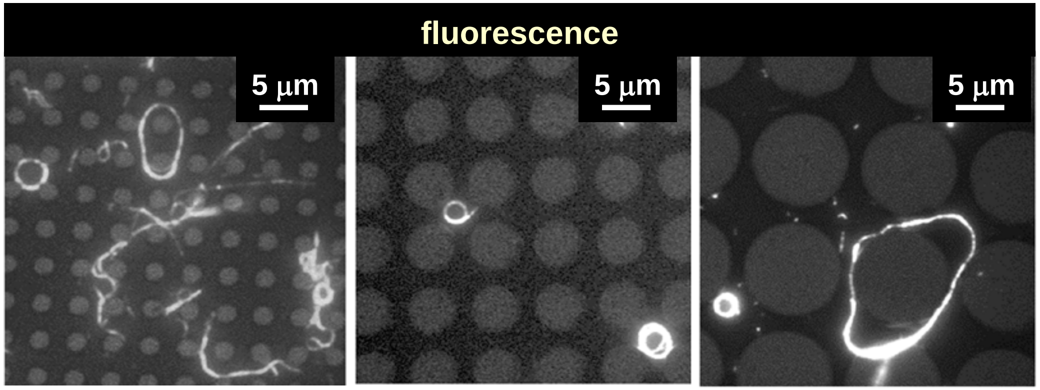
Confining Motor-Driven Assembly

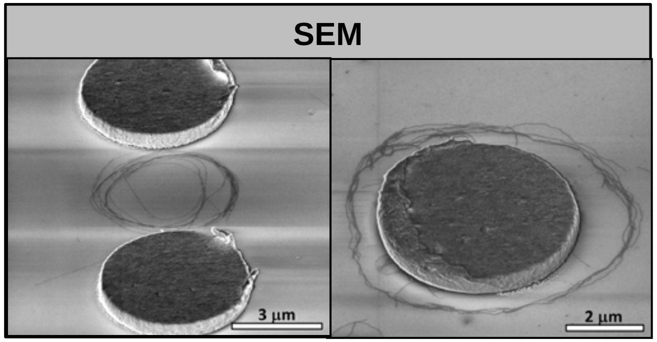
How does microscale confinement affect motor-driven self-assembly?

- Post patterns – lattice spacing and feature size define:

Size	Density
Shape	Location







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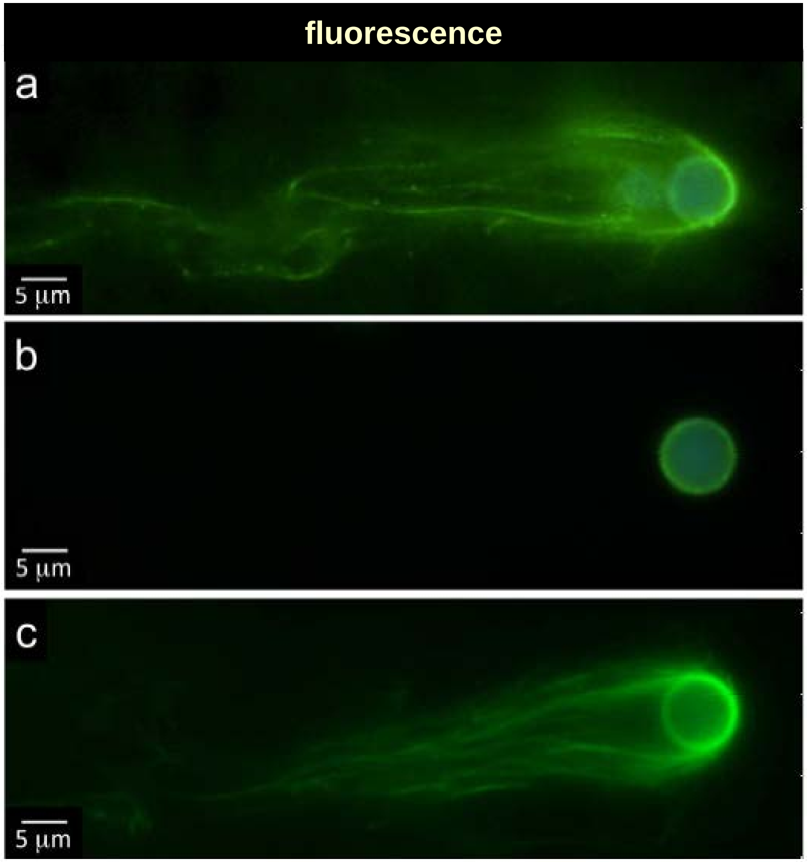
3D Nanomaterials Assembly

How are cellular nanomaterials organized into functional 3D assemblies?

- MTs naturally assemble into complex 3D structures based on interactions with MT-associated proteins (MAPs)
- Biomolecular templating of QDs on MT constructs crates unique 3D composite architectures
- Dynamic assembly – Qdot-MT aster composites reversibly assemble & disassemble



Impact: Learned how cooperative biomolecular interactions may be used for hierarchical 3D assembly and organization of nanomaterials

Spoerke et al. (2013), *ACS Nano* **7**(3), 2012

3D Nanomaterials Assembly

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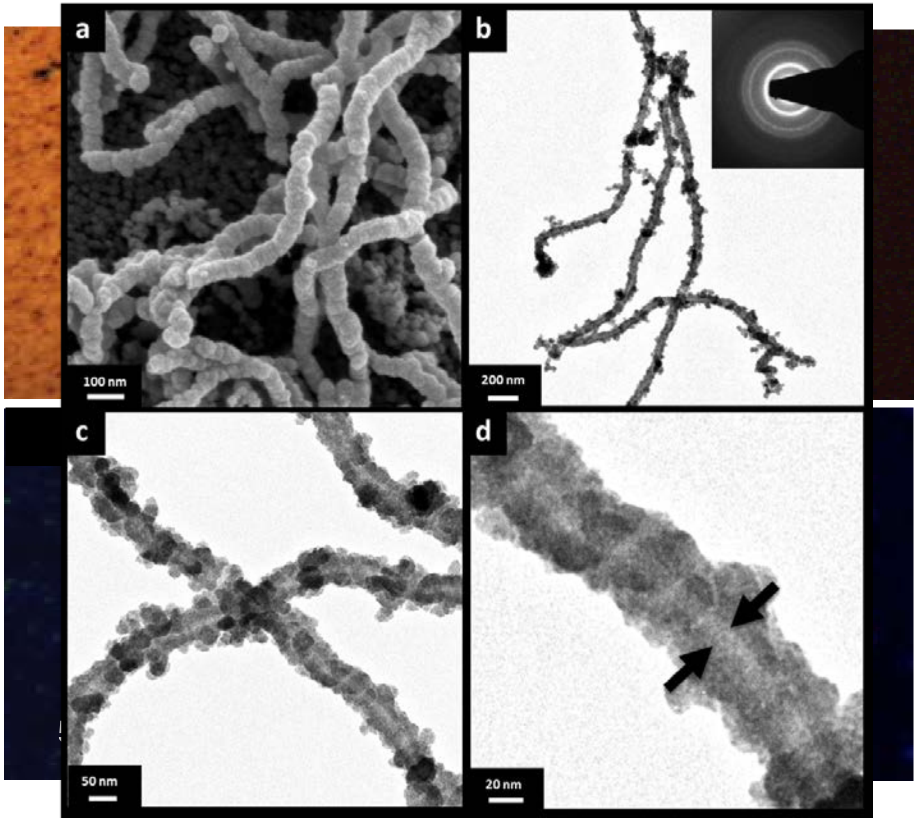
- MTs naturally assemble into complex 3D structures based on interactions with MT-associated proteins (MAPs)
- Dynamic assembly – Qdot-MT aster composites reversibly assemble & disassemble
- Mineralization – functional semiconductor asters and rings formed using 3D MT templates



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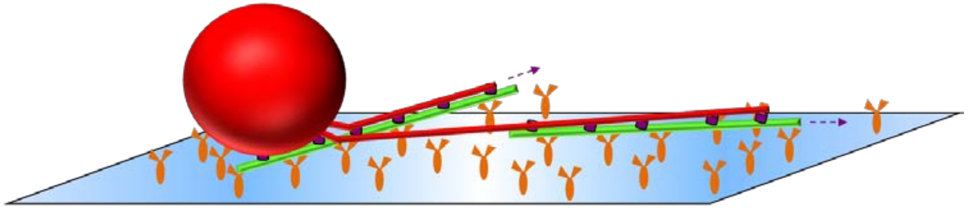
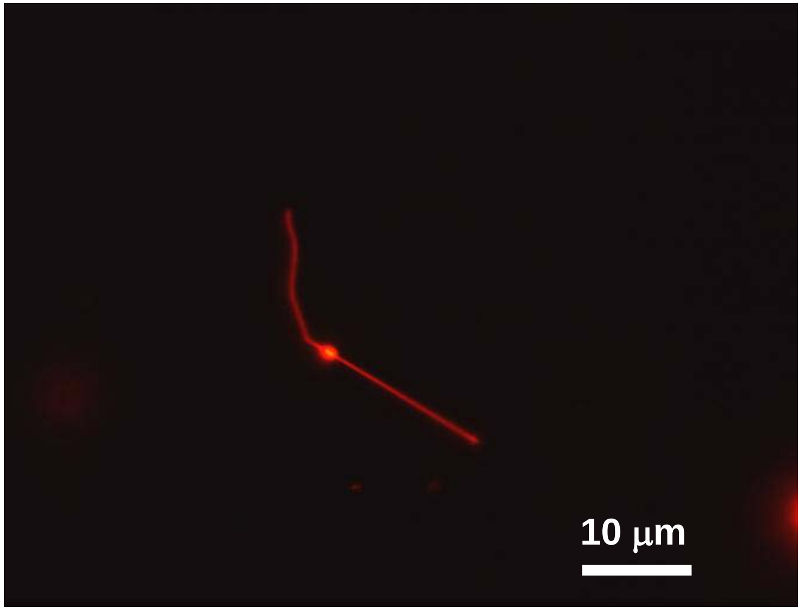
"Living" Nanotube Networks

Can active transport impart self-healing behaviors?

- Branches grow, fuse, & collapse, while network maintains overall morphology & function = self-healing
- Nanotubular networks of >10 mm in total length may be dynamically assembled by motors & MTs within 5-15 min





Impact: Understanding of how energy-dissipation (i.e., MT gliding) can self-assemble lipid networks capable of self-healing

Bouxsein et al. (2013), *Langmuir* **29**, 2992

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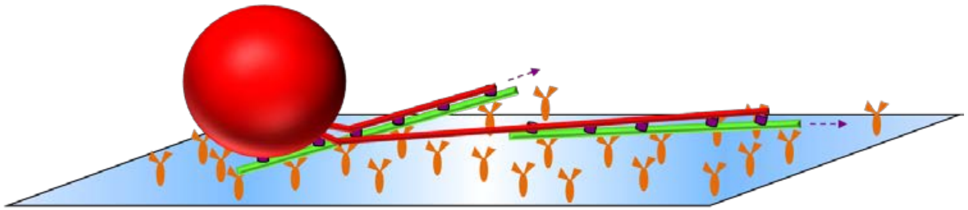

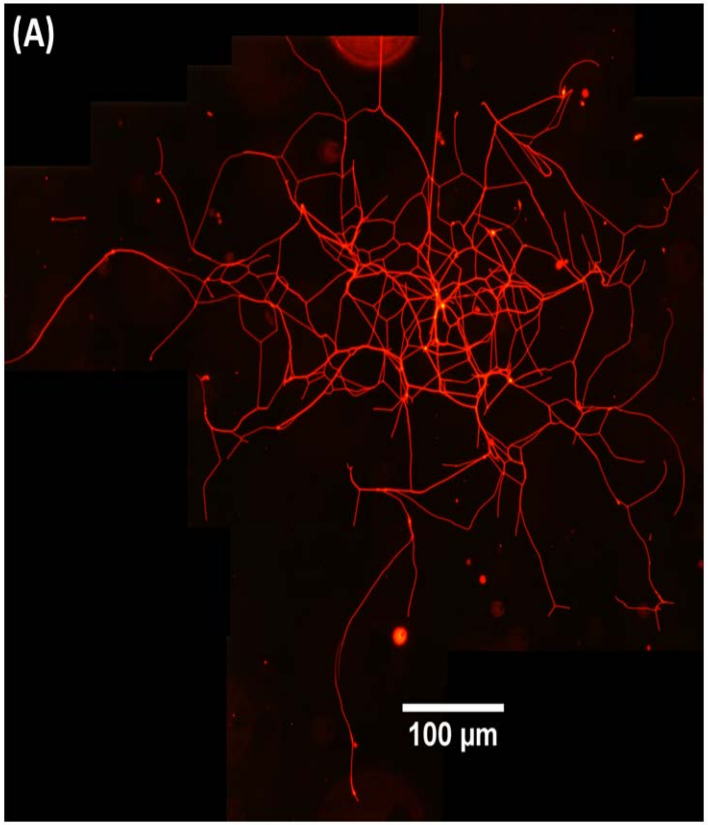

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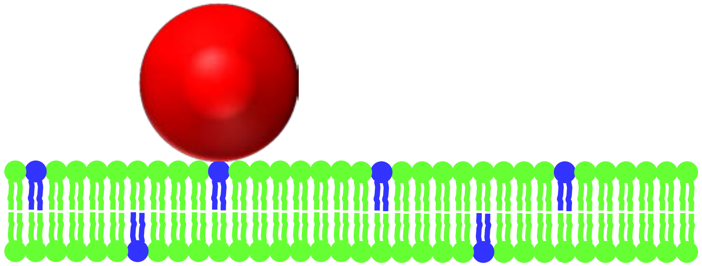
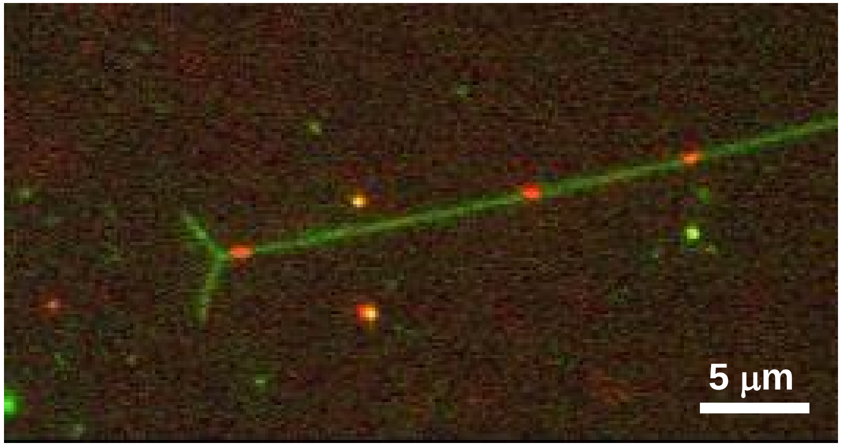
Nanotube Transport Highways

Can lipid networks support nanomaterials transport?

- Branches grow, fuse, & collapse, while network maintains overall morphology & function = self-healing
- Nanotubular networks of >10 mm in total length may be dynamically assembled by motors & MTs within 5-15 min
- "Surfing" – Qdots bound to lipid on outer leaflet move through networks via 1D thermal motion (D = 2.3 ± 0.4 μm s^{-1})





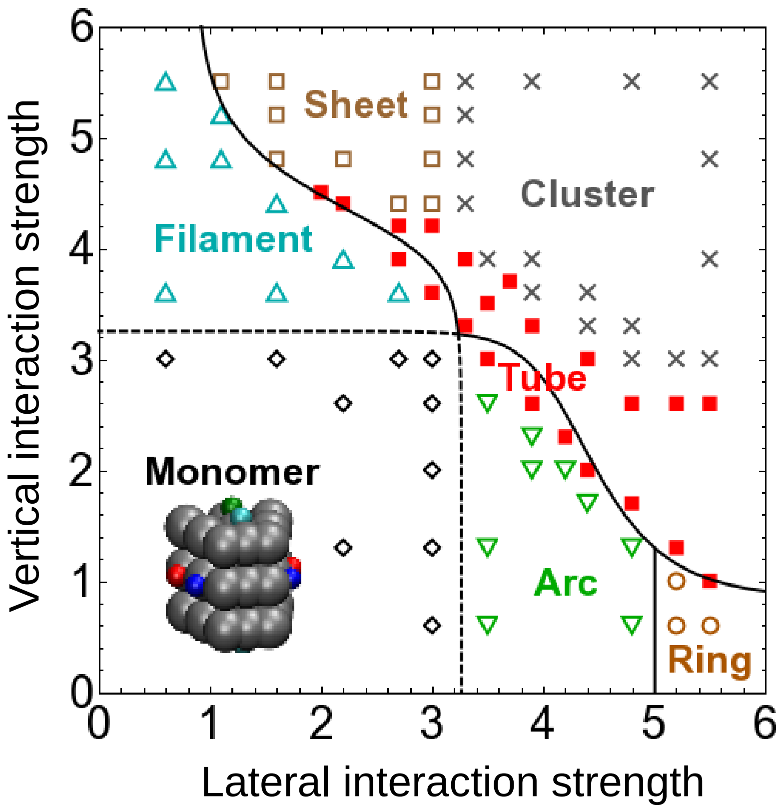
Impact: Measurement of 1D diffusive transport of nanomaterials on highly interconnected tubular networks

Bouxsein et al. (2013), *Langmuir* **29**, 2992

Simulating Artificial MT Assembly

How do molecular interactions govern molecular self-assembly of artificial MTs?

- Initial simulations – asymmetric shape and attractive sites crucial to tubule formation
- Optimal tubule formation – both the attractive lateral and vertical binding interaction energies ~10 $k_B T$
- Fewer defects when lateral interaction strength slightly exceed vertical interaction strength

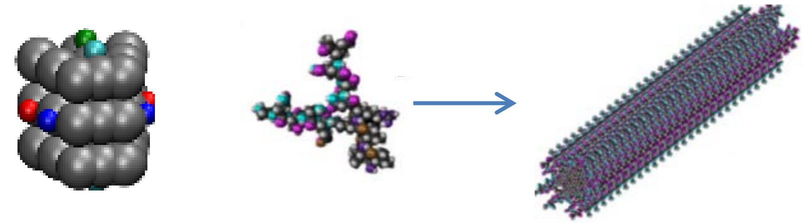


Impact: Established design rules for developing synthetic tubulin analog capable of assembling into tubule structures

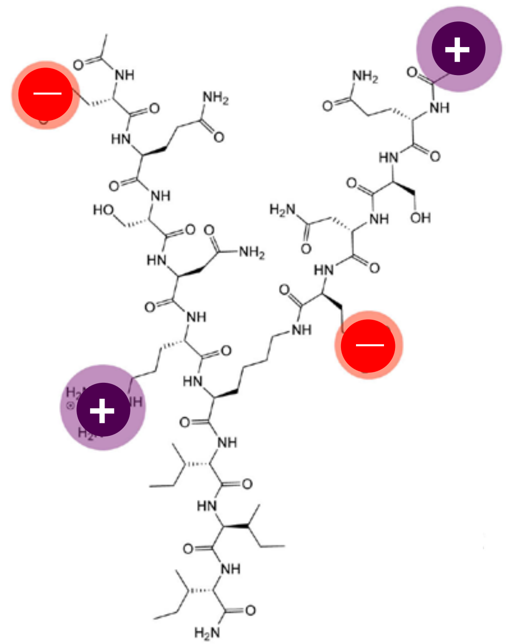
Cheng et al. (2012), *Soft Matter* **8, 5666**

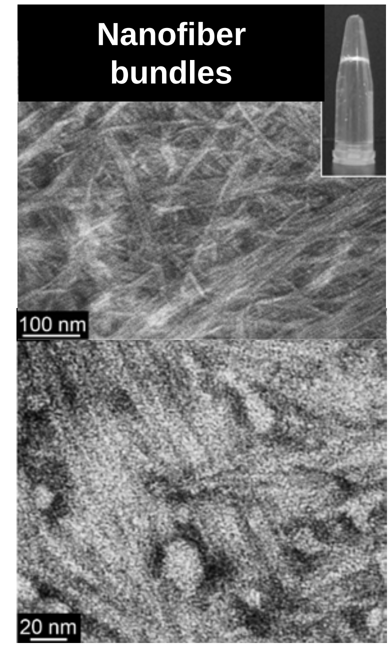
Peptide Nanofiber Assembly

Do MD simulation design rules translate into synthetic MT mimics?



- Asymm-Wedge: hydrophilic, charged branches and hydrophobic tail
- Amphiphilicity (hydrophilic asymmetry) – critical driving force for assembly
- β-sheet formation from isoleucines creates vertical interactions, drives assembly of extended fibers
- Temperature-dependent disassembly by disrupting hydrogen bonds





Impact: Peptide chemistry and versatility provide a platform for studying artificial MT assembly and tailoring functional interactions

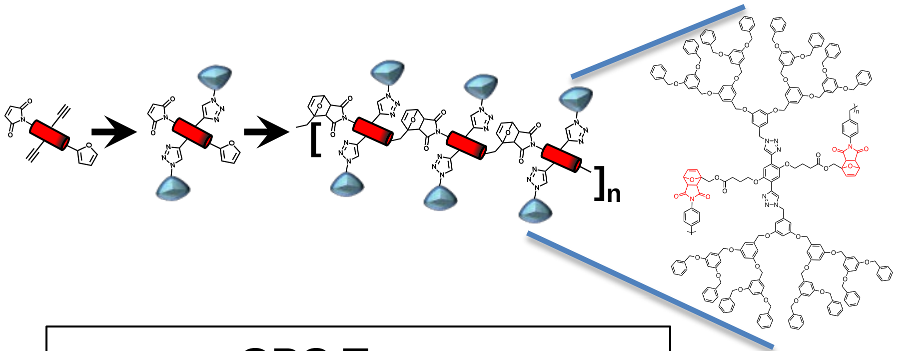
Gough et al. (2013), *Soft Matter* (in review)

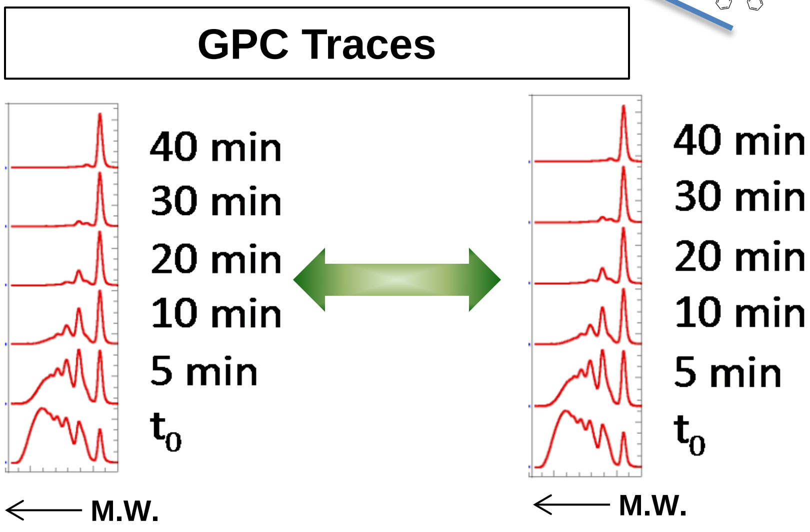
Project Highlight

Dendronized Polymer Assembly

Can thermally reversible assembly of polymers mimic MT dynamics?

- Furan-maleimide Diels-Alder chemistry
- AA-BB system: A = furan; B = maleimide
  AB system: maleimide and furan @ opposing ends
- Polymerization ~50°C
  Depolymerization ~110°C
- Rate of polymerization << rate of depolymerization





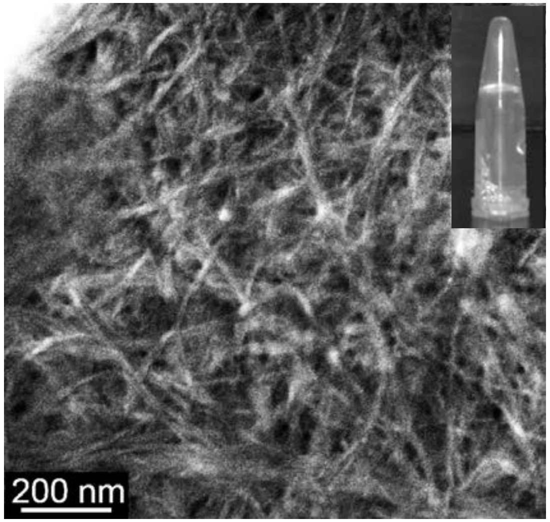
Impact: Assembly/disassembly of synthetic "tubulin" dendimers informs a path toward more complex MT analogs

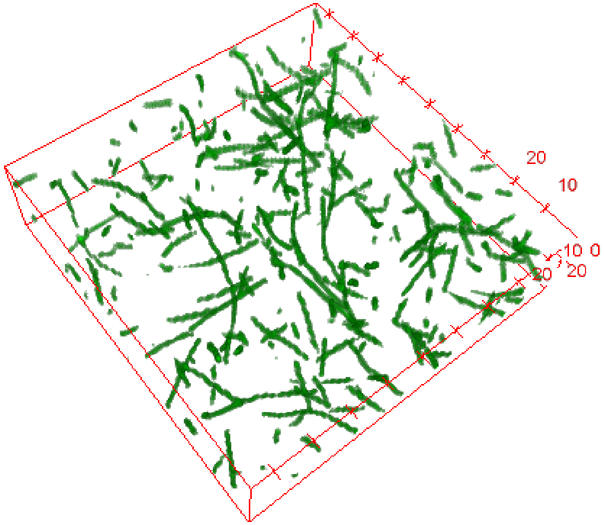
Polaske et al. (2011), *Macromolecules,* **44,** 3203; Polaske et al., (2010), *Macromolecules* **43,** 1270

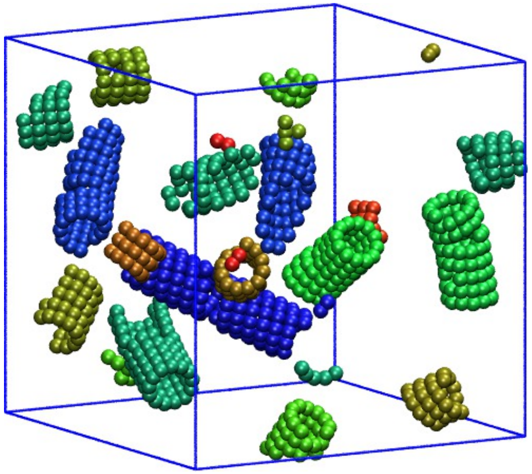
Proposed Work

Major themes underlying proposed work:

- Understanding of non-equilibrium materials assembly, both for native proteins and artificial materials
- Exploring the use of non-equilibrium assembly in novel nanocomposite materials and to elicit emergent phenomena
- Extending materials assemblies into 3D architectures





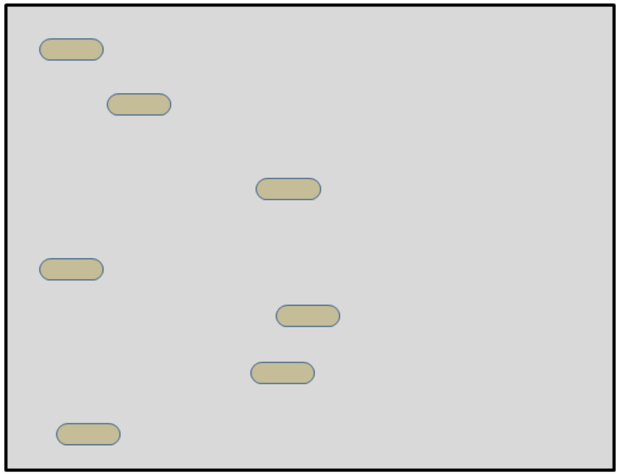


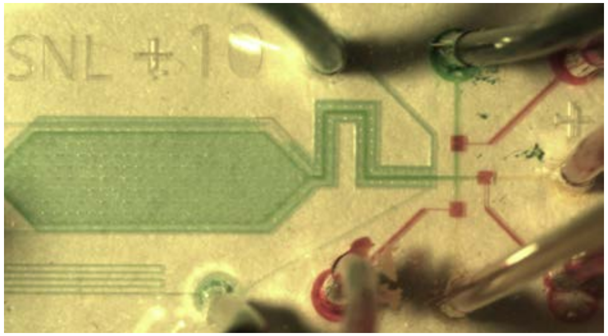
MT Dynamics & Nanowire Growth

Exploring nanowire growth based on MT polymerization & self-organization

- Understanding/manipulate transition between nucleated polymerization and MT fusion to generate MTs with chemically unique regions
- Nucleated polymerization (and potentially fusion) on planar surfaces for step-growth of heterostructured nanowires
- PDMS microfluidic devices for rapid exchange of molecular building blocks to regulate dynamic growth

Polar aligned, minus end-capped MTs



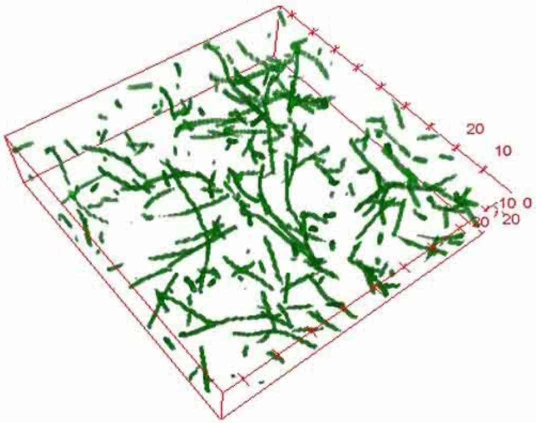


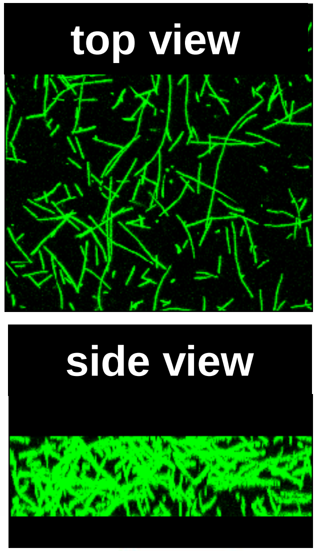
Expected Impact: Platform to study MT dynamic assembly and self-organization and understand how such processes can be used for the directed and logical growth of nanowires and heterostructures

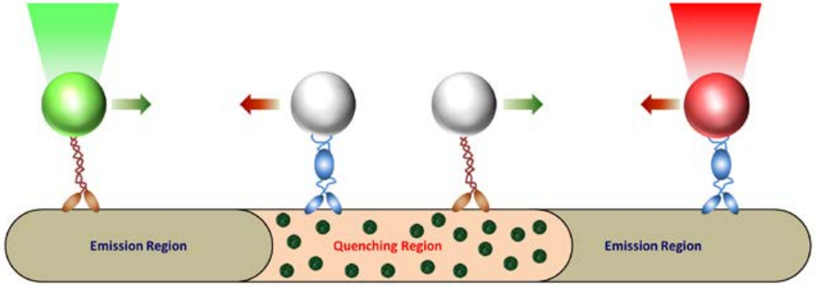
Assembly & Transport in 3D

How does molecular confinement define and enhance functionality?

- Biological phenomena – coordinated interactions of processes across crowded 3D space
- MT assembly in biocompatible gels (sol-gel, peptide gels, etc.)
- Hypothesis: molecular confinement alter system dynamics such as MT polymerization and kinesin run length
- 3D scaffold for active color change based on nanoparticle transport across segmented MTs





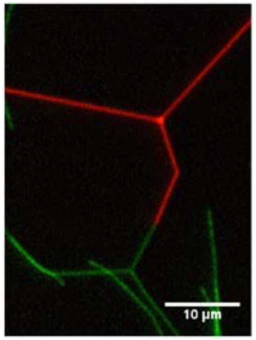


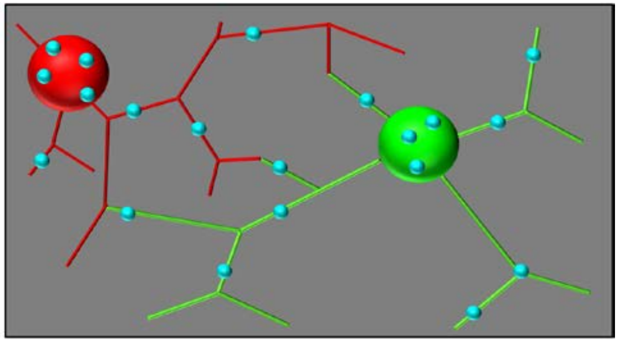
Expected Impact: A model system to study how active transport and MT dynamics in 3D architectures enhance emergent properties such as color changing observed in organisms

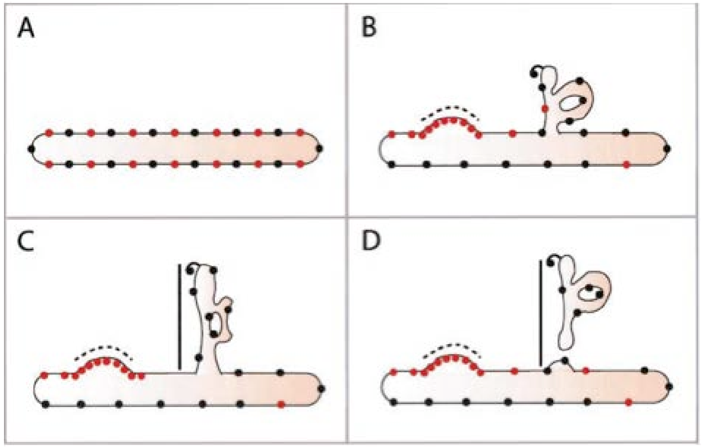
LNT Communication Networks

Learning to manipulate of lipid nanotubes and vesicles

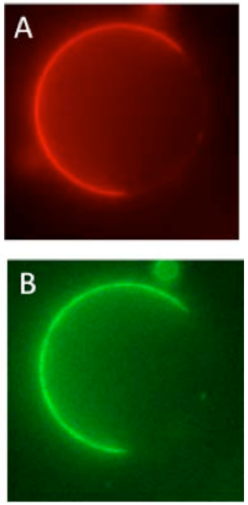
- Understand nanotube junctions among different networks; continuous or discontinuous
- Co-transport of particles surfing outside as well as within the confined, interstitial space
- Lipid nanotube formation w/ biphasic liposomes
- Vesicle budding – can vesicles be extruded based on motor protein transport?







Polishchuk et al. (2013). *Mol. Biol. Cell* **14**, 4470.

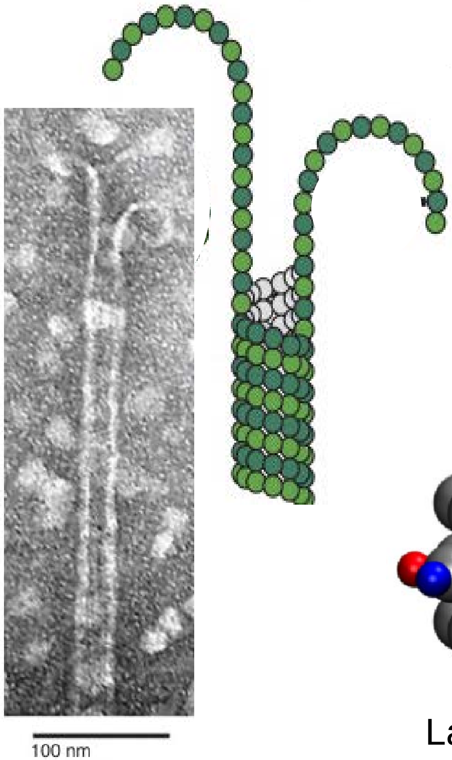


Expected Impact: LNT networks and vesicle dynamics can be used as primitive “communication networks” to study 1D diffusion in larger, interconnected networks (surfing and interstitial transport)

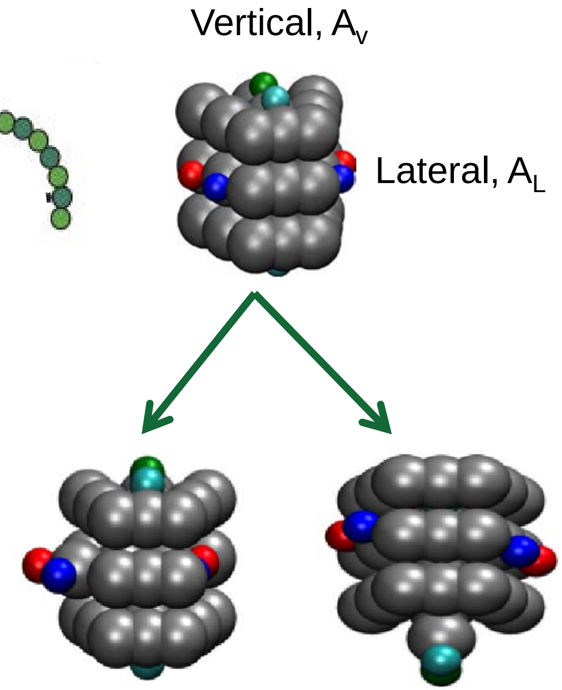
Energy & Artificial MTs

Advanced MD simulations of artificial MT assembly

- GTP/GDP bound states of tubulin essential to the catastrophic depolymerization
- Explore how shape changes in GDP- and GTP-bound tubulin affect polymer assembly mechanical properties, stability
- Monomer with two (or three) states; transitions implemented with Monte Carlo type step within the MD simulations



Hawkins et al., 2010, J. Biomechanics, 43, 23

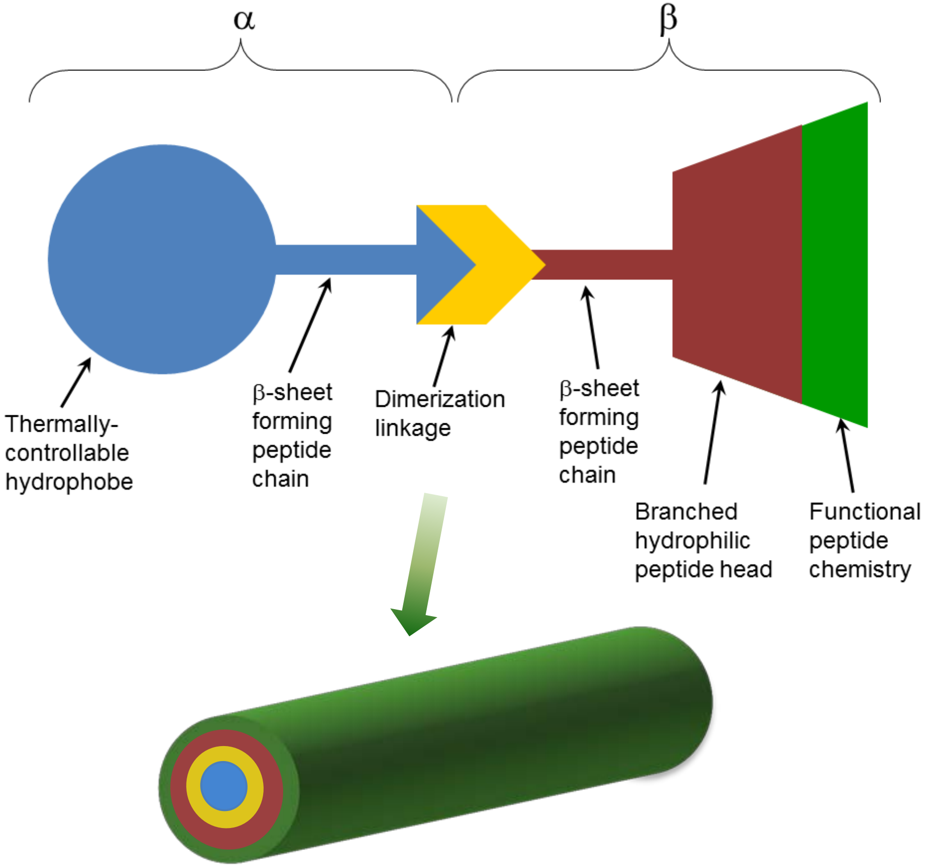


Expected Impact: Simulations will lay foundation for understanding the role of energy-dissipative, conformationally dependent elements in the dynamic assembly/ disassembly of artificial MTs

Platform for Artificial MT Assembly

Versatile molecular toolbox for artificial MT exploration

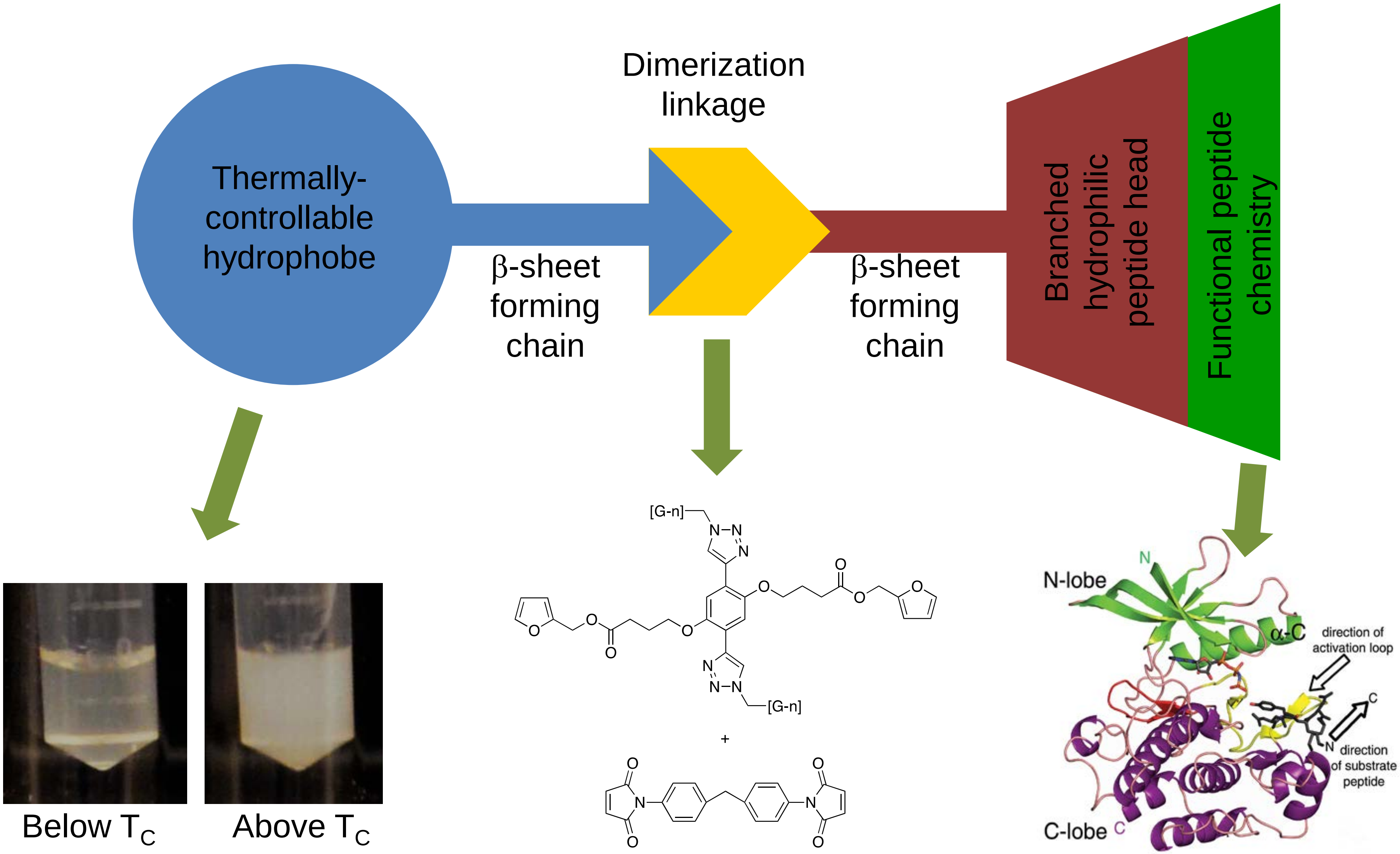
- Drive nanofiber assembly and disassembly through controllable amphiphilicity
- Enable β-sheet formation (vertical interactions)
- Establish mechanism for interactions with other materials (e.g., motors)
- Dimerizing linkage: reduce synthetic challenges + allow element swapping



Expected Impact: Modular nature of platform facilitates the exploration of an extremely diverse set of chemistries and approaches for biomimetic assembly of artificial MTs

Proposed Work

Platform for Artificial MT Assembly



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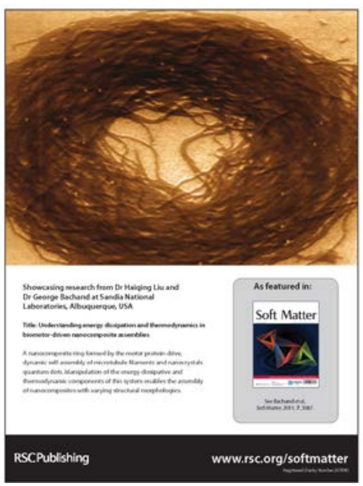

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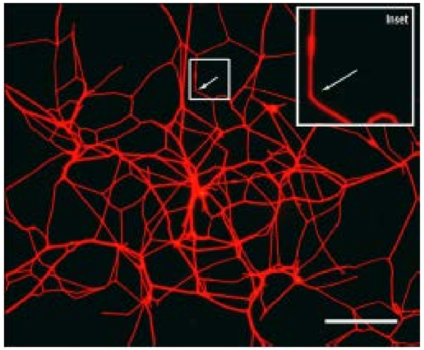
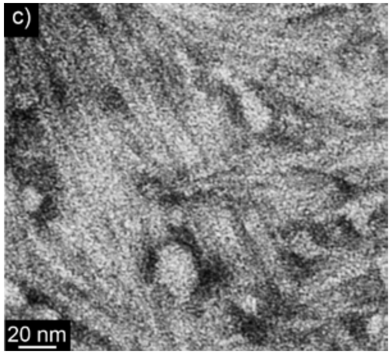
Poster Session

Lipid Membrane Transport Systems via Microtubule Processing



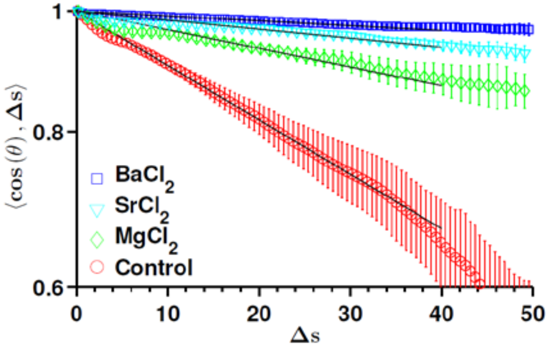
Darryl Sasaki

Molecular Tools for Artificial Microtubule Development



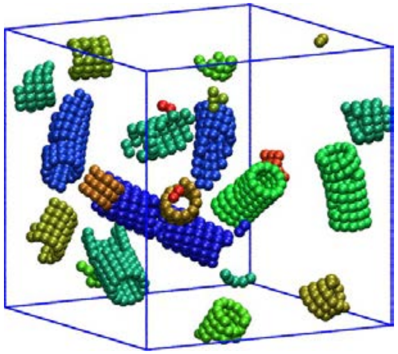
Erik Spoerke

Passive Steering and Structural Stability of Bio-Motor Driven Filaments



Nathan Bouxsein

Simulations of Artificial Microtubule Assembly



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