Outline

• Review: computational analysis of axonal transport
• Basic diffusion theory
• Computational analysis of Spindle Microtubule Flux
• Review: computational analysis of axonal transport

• Basic diffusion theory

• Computational analysis of spindle microtubule flux
Axonal Cargo Transport (I)

- Axonal transport is critical to survival and function of neurons.
- Axonal transport provides a powerful model of intracellular transport.
A Drosophila Model of Alzheimer’s Disease

- Two pathological hallmarks of AD: Aβ plaques & tau tangles
- Control:
  - SG26.1 GAL4/+; UAS-APPYFP/+ ↔ transport is driven by kinesin-1
  - SG26.1 GAL4/+; UAS-SynGFP ↔ transport is driven by kinesin-3
- Mutants:
  - SG26.1 GAL4/+; UAS-APPYFP/+; UAS-wt hTau/+  
  - SG26.1 GAL4/+; UAS-APPYFP/+; UAS-R406W hTau/+  

The questions:

1) What are the differences between normal and degenerative neurons in their axonal transport behaviors?

2) What causes transport defects in degenerative neurons?
Tracking Vesicle Movement Using Computer Vision Techniques (I)
Some General Comments (I)

- To select or build effective visualization tools is very important to the development of biological image analysis algorithms.

- It is critical to recognize and prevent potential information loss in the analysis workflow.

- Because of the small number of features, it is feasible to use algorithms with high computational complexity.
Some General Comments (II)

• A more comprehensive description of the work flow of particle tracking.

• What problems do you see in this picture?

Tracking Vesicle Movement Using Computer Vision Techniques (II)
APP Vesicle Transport and its Impairment is Region-Specific

How can we make sure that information loss is minimized?
Questions for identifying potential information loss

1) Does each vesicle change its velocity over time? If so, how?

2) How are the vesicles spatially distributed?
Tau Overexpression Differentially Affects Axonal Transport
Axon Swelling and Vesicles Accumulation
What can we learn from this?

1) How the images should be analyzed is strongly dependent on the biological questions to be addressed.

2) It is important to identify research questions from applications.
Challenge: To Infer Mechanisms from Behaviors
• Review: computational analysis of axonal transport

• **Basic diffusion theory**

• Computational analysis of spindle microtubule flux
Thermal Movement of a Free Molecule

• The average kinetic energy of a particle of mass $m$ and velocity $v_x$ is

$$\frac{1}{2} m v_x^2 = \frac{kT}{2}$$

Boltzmann constant = $1.381 \times 10^{-23}$ J/K

1 Joule = 1 N·m
t$_K$ = t$_C$ + 273.15

where $k$ is Boltzmann's constant and $T$ is absolute temperature (Einstein 1905).

• Molecular mass of GFP is 27 kDa. One atomic mass unit (Da) is $1.6606 \times 10^{-24}$g. So the mass of one GFP molecule is $4.4836 \times 10^{-20}$g.

At 27 degree C, kT is $4.1451 \times 10^{-14}$g·cm$^2$/sec$^2$.

Howard Berg, Random walks in biology, Princeton University Press, 1993

$$\sqrt{\langle v_x^2 \rangle} = \sqrt{\frac{kT}{m}} = 961.51 \text{ cm/sec}$$
1D Random Walk in Solution (I)

- **Assumptions**: *consider an ensemble of N particles*,

  1. A particle $i$ has equal probabilities to walk to the left and to the right.
  2. Particle movement at consecutive time points are independent.
  3. Movement of different particles are independent.
  4. Each particle moves at a average step size of $\delta = v_x \cdot \tau$

\[
x_i(n) = x_i(n-1) \pm \delta
\]

\[
\begin{align*}
\langle x(n) \rangle &= \frac{1}{N} \sum_{i=1}^{N} x_i(n) \\
&= \frac{1}{N} \sum_{i=1}^{N} [x_i(n-1) \pm \delta] \\
&= \frac{1}{N} \sum_{i=1}^{N} x_i(n-1) = \langle x(n-1) \rangle
\end{align*}
\]

- **Property 1**: The mean position of an ensemble of particles undergoing random walk remains unchanged.

Howard Berg, Random walks in biology, Princeton University Press, 1993
1D Random Walk in Solution (II)

- Property 2: The mean square displacement of a particle undergoing random walk increases linearly w.r.t. time.

\[
\langle x^2(n) \rangle = \frac{1}{N} \sum_{i=1}^{N} x_i^2(n) = \frac{1}{N} \sum_{i=1}^{N} x_i^2(n-1) \pm 2\delta x_i(n-1) + \delta^2 \\
= \langle x^2(n-1) \rangle + \delta^2
\]

\[
\langle x^2(n) \rangle = n\delta^2 = \frac{t}{\tau} \delta^2 = 2Dt
\]

Application of the Microscopic Theory (I)

<table>
<thead>
<tr>
<th>Object</th>
<th>Distance diffused</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1 μm</td>
</tr>
<tr>
<td></td>
<td>100 μm</td>
</tr>
<tr>
<td></td>
<td>1 cm</td>
</tr>
<tr>
<td></td>
<td>1 m</td>
</tr>
<tr>
<td>K⁺</td>
<td>0.25 ms</td>
</tr>
<tr>
<td></td>
<td>2.5 s</td>
</tr>
<tr>
<td></td>
<td>2.5 × 10⁴ s (7 hrs)</td>
</tr>
<tr>
<td></td>
<td>2.5 × 10⁸ s (8 yrs)</td>
</tr>
<tr>
<td>Protein</td>
<td>5 ms</td>
</tr>
<tr>
<td></td>
<td>50 s</td>
</tr>
<tr>
<td></td>
<td>5.0 × 10⁵ s (6 days)</td>
</tr>
<tr>
<td></td>
<td>5.0 × 10⁹ s (150 yrs)</td>
</tr>
<tr>
<td>Organelle</td>
<td>1 s</td>
</tr>
<tr>
<td></td>
<td>10⁴ s (3 hrs)</td>
</tr>
<tr>
<td></td>
<td>10⁸ s (3 yrs)</td>
</tr>
<tr>
<td></td>
<td>10¹² s (31710 yrs)</td>
</tr>
</tbody>
</table>

K⁺: Radius = 0.1 nm, viscosity = 1 mPa·s⁻¹; T = 25°C; D=2000 μm²/sec
Protein: Radius = 3 nm, viscosity = 0.6915 mPa·s⁻¹; T = 37; D = 100 μm²/sec
Organelle: Radius = 500 nm, viscosity = 0.8904 mPa·s⁻¹; T = 25°C; D = 0.5 μm²/sec

Application of the Microscopic Theory (II)


\[ \langle X^2(t) \rangle \]

- **Pure diffusion**
- **Diffusion with external flow**
- **Diffusion in a cage**
• Review: computational analysis of axonal transport

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• Computational analysis of spindle microtubule flux
Overview of Cell Cycle

Diagram showing the stages of the cell cycle:
- Interphase
  - Prophase
  - Prometaphase
  - Metaphase
  - Anaphase
  - Telophase
  - DNA replication
  - Metaphase-to-anaphase transition
  - Mitosis
  - Cytokinesis
Dynamic Microtubules in the Mitotic Spindle

Alberts et al., MBoc5
Confirmation of Poleward Flow of Spindle Microtubules

Cameron et al, JCB, 173:173-179,2006
Fluorescent Speckle Microscopy (FSM)
FSM of Dynamic Spindle Architecture

Fluorescent speckle microscopy
Quantitative Mapping of Spatial-Temporal Spindle Dynamics

Regional Variations of Microtubule Flux

Questions?