Bioimage Informatics

Lecture 18, Spring 2012

Bioimage Data Analysis (V)

Single Particle Tracking (part 3)
Outline

• Overview of single particle tracking techniques

• Review: linear assignment based single particle tracking

• Multiple hypothesis tracking

• Application I: fluorescence speckle microscopy
• Overview of single particle tracking techniques
  • Review: linear assignment based single particle tracking
  • Multiple hypothesis tracking

• Application I: fluorescence speckle microscopy
Overview of Single Particle Tracking Techniques (I)

- **Method 1: simple nearest neighbor tracking**
  - Each particle is assigned to its nearest neighbor
  - Often a search radius is adopted
  - Rarely used; applicable only when particles are well separated

- **Method 2: global nearest neighbor tracking**
  - Association cost between a pair of particles is their distance

\[
\begin{align*}
\min & \sum_{i \in G_k} \sum_{j \in G_{k-1}} a^k(i,j)w^k(i,j) \\
\text{st.} & \quad \sum_i a(i,j) = 1 \quad \sum_j a(i,j) = 1 \quad a(i,j) \in \{0,1\} \\
& \quad w^k(i,j) = \|x_{j}^{k+1} - x_{i}^{k}\|
\end{align*}
\]
Overview of Single Particle Tracking Techniques (II)

• Method 3: global smooth motion tracking

- Association cost between a pair of particles is their motion smoothness

\[
\min \sum_{i \in G_k} \sum_{j \in G_{k-1}} a^k(i, j) w^k(i, j) \\
\text{st.} \quad \sum_{i} a(i, j) = 1 \quad \sum_{j} a(i, j) = 1 \quad a(i, j) \in \{0, 1\}
\]

\[
c^k(i, j) = w_1 \left[ 1 - \frac{(x^k_i - x^{k-1}_i)(x^{k+1}_j - x^k_i)}{\|x^k_i - x^{k-1}_i\| \|x^{k+1}_j - x^k_i\|} \right] + w_2 \left[ 1 - 2 \frac{\sqrt{\|x^k_i - x^{k-1}_i\| \|x^{k+1}_j - x^k_i\|}}{\|x^k_i - x^{k-1}_i\| + \|x^{k+1}_j - x^k_i\|} \right]
\]
Overview of Single Particle Tracking Techniques (III)

- There are other ways to define the association cost.
  - Example 1: to incorporate particle intensity

- There are a variety of other tracking techniques.
  - Example: joint-probabilistic data-association filter (JPDAF)
  - Many of such techniques come from military applications
  - Many of these techniques are based on assumptions that may not necessarily hold in biological applications
Overview of Single Particle Tracking Techniques (IV)

- **Distance** → Global nearest neighbor
  \[
  w^k(i, j) = \|x_{j}^{k+1} - x_{i}^{k}\|
  \]

- **Smooth motion** → Global smooth motion
  \[
  w^k(i, j) = w_1 \left[ 1 - \frac{(x_{i}^{k} - x_{j}^{k-1})(x_{j}^{k+1} - x_{i}^{k})}{\|x_{i}^{k} - x_{j}^{k-1}\|\|x_{j}^{k+1} - x_{i}^{k}\|} \right] + w_2 \left[ 1 - 2 \frac{\sqrt{\|x_{i}^{k} - x_{j}^{k-1}\|\|x_{j}^{k+1} - x_{i}^{k}\|}}{\|x_{i}^{k} - x_{j}^{k-1}\| + \|x_{j}^{k+1} - x_{i}^{k}\|} \right]
  \]

- **Mahalanobis distance**, where the prediction comes from typically a Kalman filter
  \[
  w^k(i, j) = (x_{i}^{k} - \hat{x}_{j}^{k})^T S(x_{i}^{k})^{-1} (x_{i}^{k} - \hat{x}_{j}^{k})
  \]
  \[
  S(x_{i}^{k}) = \text{cov}(x_{i}^{k} - \hat{x}_{j}^{k})
  \]
• Overview of single particle tracking techniques

• **Review: linear assignment based single particle tracking**

• Multiple hypothesis tracking

• Application I: fluorescence speckle microscopy
Definition of Particle Tracking (II)

• Different cases
  - Constant number of features
  - Feature appearance
  - Feature disappearance

• Cases of feature appearance & disappearance
  - Moving in or out of field of view
  - Moving in or out of the focal plane
  - Assembly/disassembly
  - Feature merging/splitting
Example: Particle Tracking (I)

- Frame $i-1$
- Frame $i$
- Frame $i+1$
Example: Particle Tracking (II)

- Frame $i-1$
- Frame $i$
- Frame $i+1$
Example of Particle Tracking (III)

Accumulated evidence from multiple frames makes tracking more reliable.

- Frame $i-1$
- Frame $i$
- Frame $i+1$
Particle Tracking Based on Global Linear Assignment (I)

- An optimization strategy is required to resolve conflicts between competing assignments.
- Selection of assignment weight will critically influence outcomes.

Particle Tracking Based on Global Linear Assignment (II)

• Formulation of the tracking problem as a bipartite graph assignment

\[
\min \sum_{i \in G_k} \sum_{j \in G_{k+1}} a^k(i, j) w^k(i, j)
\]

\[
st. \sum_i a(i, j) = 1 \quad \sum_j a(i, j) = 1 \quad a(i, j) \in \{0, 1\}
\]

• There are efficient numerical algorithms to solve large scale assignment problems.

• Why not use a tripartite graph?
  - Optimal assignment of tripartite graph is NP-complete.
  - Difficult to resolve conflicts between two tripartite assignments.
Handling Particle Appearance & Disappearance

• Track appearance and disappearance are handled by introducing virtual points.

G. Yang, A. Matov, G. Danuser, Reliable tracking of large scale dense antiparallel particle motion for fluorescence live cell imaging, *IEEE CVPR, 2005*
References on Linear Assignment


(Downloadable from class web page)
Assessment of Linear Assignment Based Particle Tracking

• Disadvantage: The algorithm fundamentally only looks for the best solution over two consecutive frames (greedy).

• Disadvantage: It can not handle feature merging and splitting.

• Advantage: It has low computational complexity and can be used to tracking very large number of particles.
• Overview of single particle tracking techniques

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MHT-Based Particle Tracking (I)

• “MHT is a deferred decision logic in which alternative data association hypotheses are formed whenever observation-to-track conflict situations occur.”

• Multiple competing hypotheses are represented in a tree structure.

Fig. 4. Family (node) structure with $N$-scan pruning.
MHT-Based Particle Tracking (II)

- The tree structure provides a flexible way to handle feature appearance/disappearance and merging/splitting.

Fig. 5. Formation of hypotheses from tracks in families.
MHT-Based Particle Tracking (III)

• **Advantages**
  - An effective framework to incorporate multi-frame information.
  - A natural way to handle feature appearance and disappearance.
  - A natural way to handle feature merging/splitting.

• **Disadvantages**
  - Combinatorial explosion if the tree is not pruned.
  - Many variations in implementation.
  - High computation and memory cost.

• Overall a very important approach, especially when the number of features to be tracked is small.
Methods to Handling Merging & Splitting

• Tracklet-based approaches
  – http://celltracking.intel-research.net/

• Graph-based approaches
  – http://www.farsight-toolkit.org/wiki/Main_Page
Tracking Migrating Cells

Courtesy of Lee Weiss & Takeo Kanade
• Overview of single particle tracking techniques
• Review: linear assignment based single particle tracking
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• Application I: fluorescence speckle microscopy
Overview of Cell Cycle

Interphase

- Prophase
- Prometaphase
- Metaphase
- Anaphase
- Telophase

Mitosis

Metaphase-to-anaphase transition

Cytokinesis

DNA replication

M Phase

Interphase
Dynamic Microtubules in the Mitotic Spindle

Alberts et al., MBoC 5

Green: microtubule
Red: kinetochore
5 \mu m
Confirmation of Poleward Flow of Spindle Microtubules

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?
Movement of a Free Molecule (I)

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

$$\left\langle \frac{1}{2}mv_x^2 \right\rangle = \frac{kT}{2}$$

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

- Principle of equipartition of energy

$$\left\langle \frac{1}{2}mv^2 \right\rangle = \frac{3 \cdot kT}{2}$$

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

1 Joule $= 1$ N·m

$t_K = t_C + 273.15$

Howard Berg, Random walks in biology, Princeton University Press, 1993
Movement of a Free Molecule (II)

- 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·cm²/sec².

$$\sqrt{\langle v^2_x \rangle} = \sqrt{\frac{kT}{m}} = 961.51 \text{ cm/sec}$$

Howard Berg, Random walks in biology, Princeton University Press, 1993
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 an average step size of $\delta = v_x \cdot \tau$

$$x_i(n) = x_i(n-1) \pm \delta$$

| -3$\delta$ | -2$\delta$ | -$\delta$ | 0 | +$\delta$ | +2$\delta$ | +3$\delta$

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

$$= \frac{1}{N} \sum_{i=1}^{N} x_i(n-1) = \langle x(n-1) \rangle$$

- 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.

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

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

\[
\langle r^2(n) \rangle = \langle x^2(n) + y^2(n) \rangle = 4Dt
\]

\[
\langle r^2(n) \rangle = \langle x^2(n) + y^2(n) + z^2(n) \rangle = 6Dt
\]

## 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>K⁺</td>
<td>0.25ms</td>
</tr>
<tr>
<td>Protein</td>
<td>5ms</td>
</tr>
<tr>
<td>Organelle</td>
<td>1s</td>
</tr>
</tbody>
</table>

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

Application of the Microscopic Theory (II)


![Graph showing mean square displacement vs. time for pure diffusion, diffusion with external flow, and diffusion in a cage.](image-url)
Application of the Microscopic Theory (III)

- Calculation of diffusion coefficient (Einstein-Stokes equation)
  - diffusion of spherical particles through liquid in which viscous force dominates

\[ D = \frac{kT}{6\pi\eta r} \]

- Boltzmann constant: \( k = 1.381 \times 10^{-23}\text{J/k} = 1.381 \times 10^{-17}\text{N\cdot\mu m/k} \)
- Absolute temperature: \( T = 273.15 + 25 \)
- Viscosity: \( \eta = 0.8904\text{mPa\cdot s} = 0.8904 \times 10^{-3} \times 10^{-12}\text{N\cdot\mu m^{-2}\cdot s} \)
- Sphere radius: \( r = 500\text{nm} = 0.5\mu m \)
- Calculated diffusion coefficient: \( D = 0.5\mu m^2/s \)

An Overview of Axonal Transport

- Axonal transport is critical to survival and function of neurons.
- Axonal transport is a powerful model of intracellular transport.
- Axonal transport may be a good model to study spatiotemporal cell signaling.
- Many mitotic motors also drive axonal transport.

Hirokawa N., JCB, 94:129, 1982
Bars: 0.1μm
Molecular Motor Machinery of Axonal Transport

Potential Mechanisms of Axonal Transport Defects

![Diagram showing axonal transport mechanisms]

- **Dynein, DLC, DLIC, DHC**
- **Kinesin-1, KHC**
- **KLC**
- **Cargo**
- **Microtubule**
- **Docking protein**
- **Dynactin complex**

**Abbreviations**
- LC = light chain
- HC = heavy chain
- LIC = light intermediate chain