Using active contours for yeast cell segmentation

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Outline

1. Problem description
   - Understanding the yeast metabolism
   - Confocal fluorescence microscopy
   - Extracting quantifiable information

2. Yeast cell segmentation
   - Image characteristics
   - An active contour algorithm
   - Extra ingredients

3. Results

4. Summary and outlook
Using active contours for yeast cell segmentation

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Understanding the yeast metabolism

- Yeast is one of the simplest eukaryotes
- Serves as a model organism
- Grows fast → easy to produce statistically relevant data
- Genes can easily be manipulated
  → useful for functional analysis of proteins
Examining the yeast metabolism

- Green fluorescence protein (GFP)
- GFP is inserted into gene sequence $\rightarrow$ marker
- Gene expression $\rightarrow$ respective protein is fluorescent
- Track fluorescence via microscopy

New technique: Laser confocal microscopy $\rightarrow$ high quality 3D-images
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**Extracting quantifiable information**

**Given:**
- Fluorescence image
- Transmission image
  (non-invasive measurement)
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- Transmission image (non-invasive measurement)

**Sought:** Characteristics of fluorescent material
- Volume per cell
- Localization within cell
- Shape information
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- Fluorescence image
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**Needed:** Cell segmentation method
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Image characteristics

- Densely populated with cells, non-uniform background
- Cells separated by dark or bright ridges
- Complex structures inside the cells
- Different cell sizes and mother/daughter cells (buds)
- Different types of clutter
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**Tracing the contour of a cell**

**Subtask:** Segment a single cell  
**Idea:** Minimize energy for a parametrization  
\( \gamma : S^1 \rightarrow \mathbb{R}^2 \)  

Active contour model: ("Snakes")

\[
\min_{\gamma} \int_{S^1} G(\gamma(t)) \, dt + \alpha \int_{S^1} |\gamma'(t)|^2 \, dt + \beta \int_{S^1} |\gamma''(t)|^2 \, dt
\]

*Kass, Witkin, Terzopoulos ['87]*

\( G \) "rewards" cell boundaries in \( I \), e.g.

\[
G(x) = \lambda^2 + |\nabla I(x)|^2
\]
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- **External energy**
- **Internal energy**

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Limitations of the classical model

Algorithm:
Start with a contour and do functional descent
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Start with a contour and do functional descent

Problems:

1. Approach the boundary from outside
   - More than one cell could be caught
   - Hangs at interior structures

2. Approach the boundary from inside
   - Likely to segment a single cell
   - Internal energy forces contour to a point
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Remedy:
Add an “inflating force” to the model
Introducing a volume term

**Idea:** Reward greater volumes

Divergence theorem:

\[
\text{Vol}(\Omega') = \frac{1}{2} \int_{S^1} \gamma(t) \cdot \gamma'(t) \perp \, dt
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Introducing a volume term

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Inflating snakes:

\[
\min_{\mathbf{\gamma}} \int_{S^1} G(\mathbf{\gamma}(t)) + \alpha |\mathbf{\gamma}'(t)|^2 \, dt - \left( \frac{\beta}{2} \int_{S^1} \mathbf{\gamma}(t) \cdot \mathbf{\gamma}'(t)^\perp \, dt \right)^p
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\[ 0 < p < 1 \]
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$$0 < p < 1$$

- **$p$-th power:** Volume terms grows slower than $\|\gamma'\|_2^2 \sim$ still regularizing
- Self-intersecting parametrizations forbidden (no problem in practice)
- Connections to the “balloons” of Cohen ['91]
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Extra ingredients

**Preprocessing:**
- Normalize and smooth image \( \mapsto \) TV regularization or nonlinear degenerate lifting
- Find constant regions (background)
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**“Seeding”:**
- Find points where to inflate snakes ($\geq 1$ per cell)
- Heuristic algorithm based on local maxima of smoothed edge data
- Cluster local maxima $\rightarrow$ seeding points
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Postprocessing:
- Detect overlapping cells $\Rightarrow$ discard/join
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**Example: Evolution of a snake**

Gradient descent on cell with complicated structure:
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**Segmentation of a whole dataset**

![transmission image](image1.png) ![segmented image](image2.png)
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Segmentation of a whole dataset

transmission image

segmented image
Using active contours for yeast cell segmentation

**Detailed discussion**

**Features:**

- Many cells are segmented accurately
- Works for cells with complicated inner structure
- Catches also non-circular shapes
Detailed discussion

Features:
- Many cells are segmented accurately
- Works for cells with complicated inner structure
- Catches also non-circular shapes

Problems:
- Some cells are missed
- Cell-like interior structures are not captured right
- Sometimes background is falsely classified as cell
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Summary and outlook

- A modified snake model is suitable for the segmentation of a single yeast cell
- Can be integrated into a processing pipeline to segment whole images
- According to the microscopists, the results are good
- A single set of parameters work for many images
- Still: Some components can be improved, e.g. seeding or background detection
- In the moment, the algorithm is very slow (\( \sim 30 \) minutes)
- Eventually, the segmentation has to be combined with the processing of the fluorescence data