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INSTITUTO  
DE CIENCIAS  
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CIMT  
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INFORMATICA MEDICA  
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LA SERENA SCHOOL  
FOR DATA SCIENCE  
Applied Tools for Data-driven Sciences

• AURA Campus  
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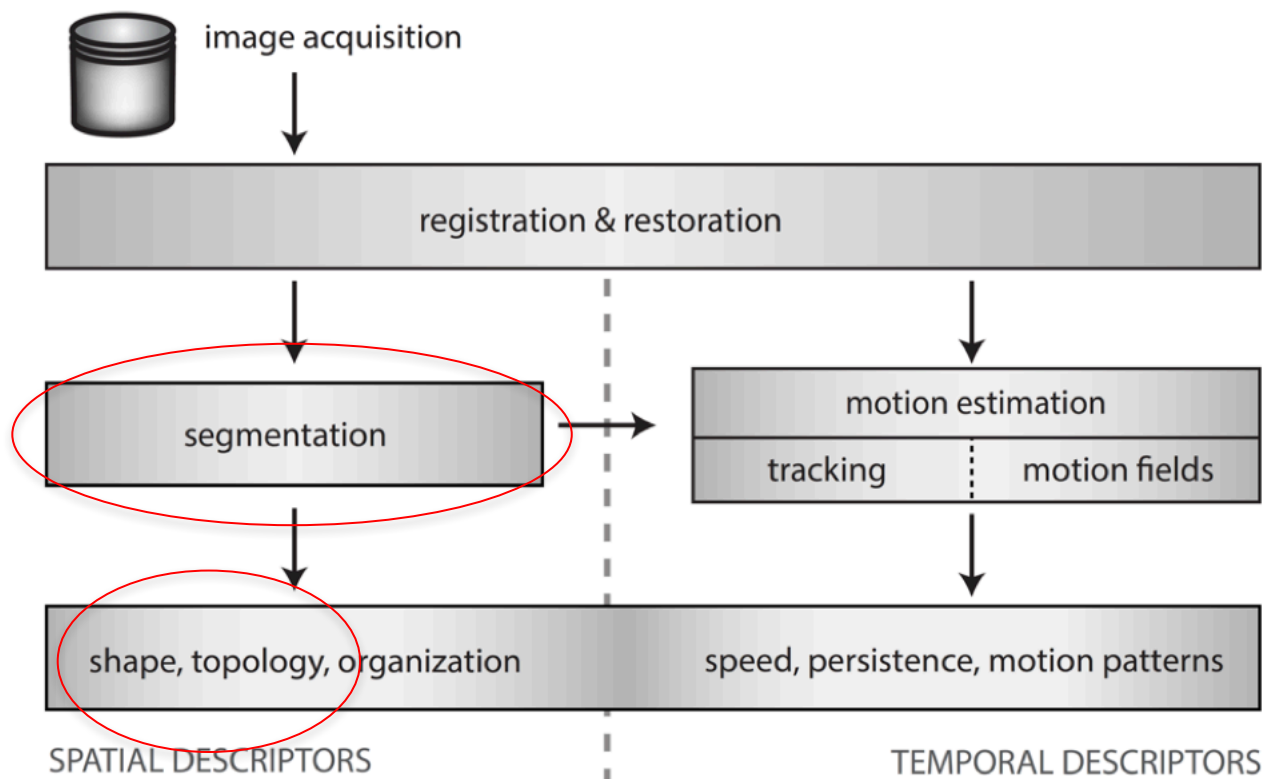
# Image Processing

image formation, segmentation

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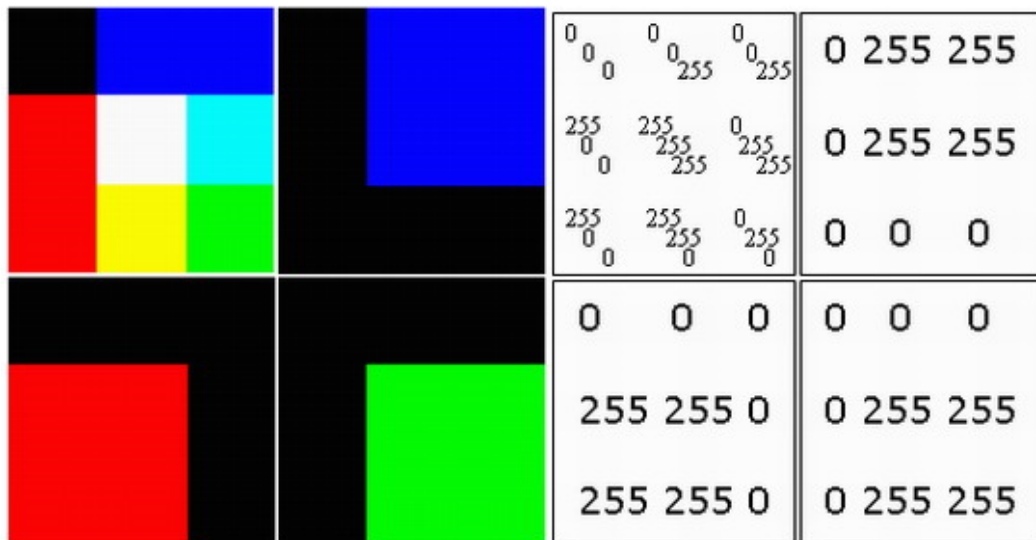
## 1. Introduction

- Images
- Segmentation\*

## 2. Descriptors

- Shape & Topology\*
- *Tracking*

- A **digital image** can be defined as a function over a discrete space
- A typical 2D image model is the **raster image**: array (matrix) of **pixels** in cartesian coordinates  $(x, y)$
- A numeric value for **brightness (intensity)** or **color** is associated to each pixel



$$I = f(x, y)$$

$$(x, y) \in [0, \dim_x - 1] \times [0, \dim_y - 1]$$

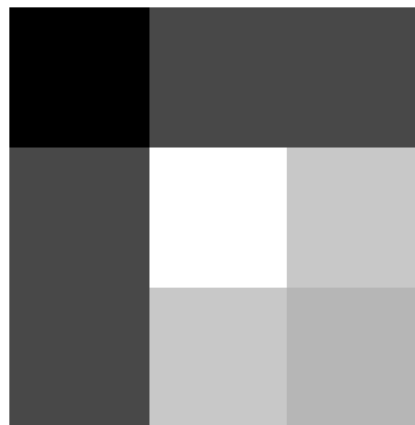
$$I[x_i, y_j] = f[x_i, y_j]$$

- ...so, a digital image can be treated as a **function** (in the mathematical sense)...
  - on a discrete domain
  - with numeric values associated to each elements, representing a property (such as color, brightness, depth, etc.)

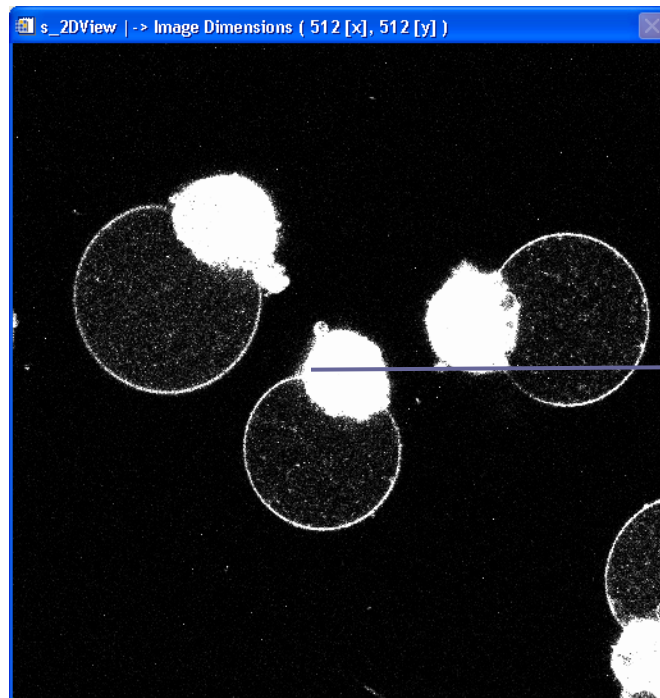
- Greyscale image
  - A brightness (intensity) level is defined for each pixel

|    |     |     |
|----|-----|-----|
| 0  | 85  | 85  |
| 85 | 255 | 170 |
| 85 | 170 | 85  |

$I[x,y]$



| Binary value | Decimal value |
|--------------|---------------|
| 0000 0000    | 0 (black)     |
| 0000 0001    | 1             |
| 0000 0010    | 2             |
| 0000 0011    | 3             |
| 0000 0100    | 4             |
| 0000 0101    | 5             |
| 0000 0110    | 6             |
| 0000 0111    | 7             |
| 0000 1000    | 8             |
| ...          | ...           |
| 1111 1011    | 251           |
| 1111 1100    | 252           |
| 1111 1101    | 253           |
| 1111 1110    | 254           |
| 1111 1111    | 255 (blanco)  |

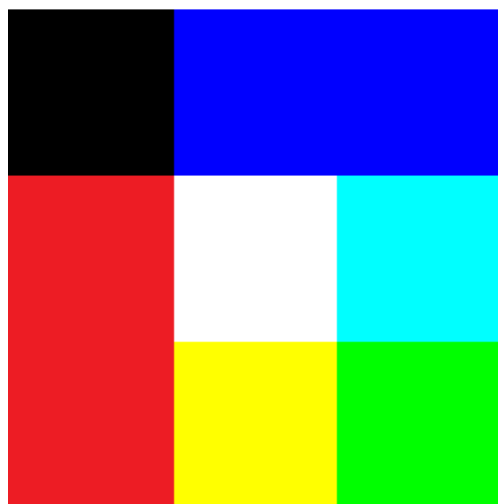


$I(290,267) = 220$

8 bit greyscale image

A  $n$  bit greyscale image encodes up to  $2^n$  intensity values

- RGB image
  - Three channels for respective primary colors: Red, Green, Blue



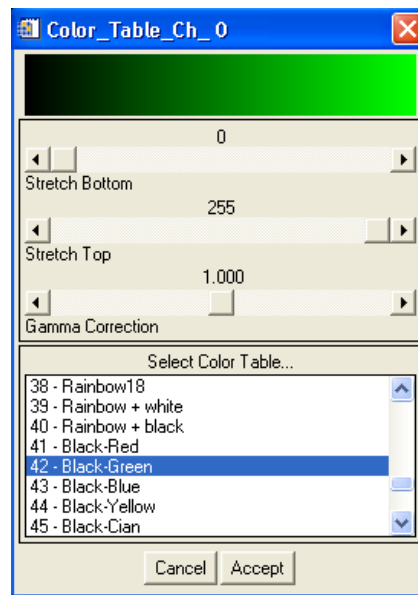
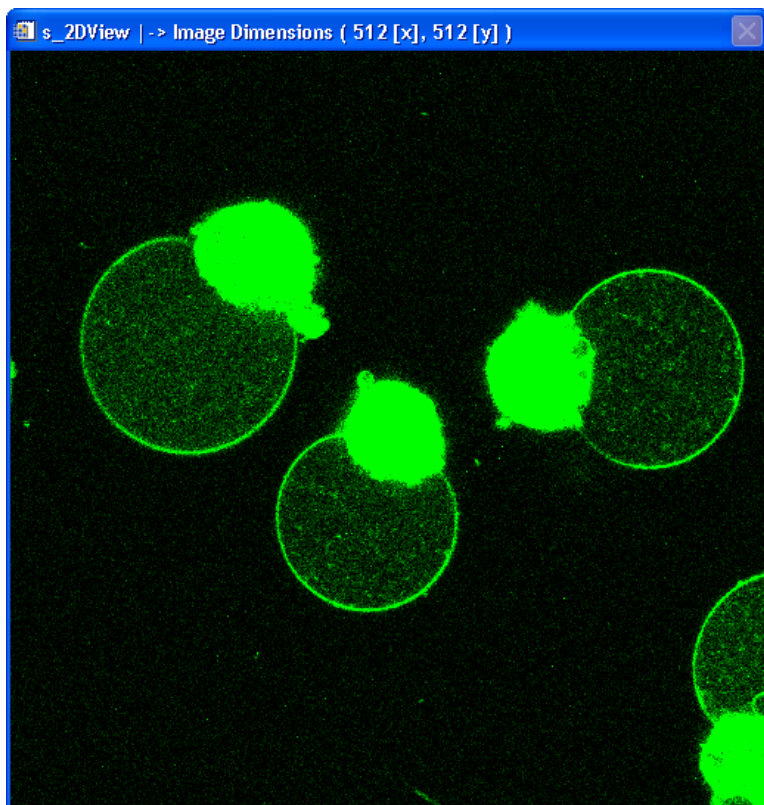
|     |     |     |
|-----|-----|-----|
| 0   | 0   | 0   |
| 0   | 0   | 0   |
| 0   | 255 | 255 |
| 255 | 255 | 0   |
| 0   | 255 | 255 |
| 0   | 255 | 255 |
| 255 | 255 | 0   |
| 0   | 255 | 255 |
| 0   | 0   | 0   |

$r[x, y] \ g[x, y] \ b[x, y]$

- Other color spaces are HSV, LAB



- It is possible to define color tables (or lookup tables, LUTs) for visualization purposes. A grayscale image can be displayed using a green scale.

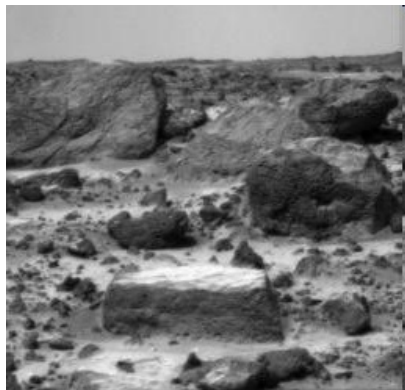


| r g b |     |   |
|-------|-----|---|
| 0     | 0   | 0 |
| 0     | 1   | 0 |
| 0     | 2   | 0 |
| :     | :   | : |
| :     | :   | : |
| :     | :   | : |
| :     | :   | : |
| 0     | 200 | 0 |
| :     | :   | : |
| :     | :   | : |
| 0     | 255 | 0 |

- Segmentation

- The partitioning of a given image into regions of interest (ROIs) according to given criteria (e.g. color).
- After segmentation, further characterizations can be performed upon the resulting ROIs.

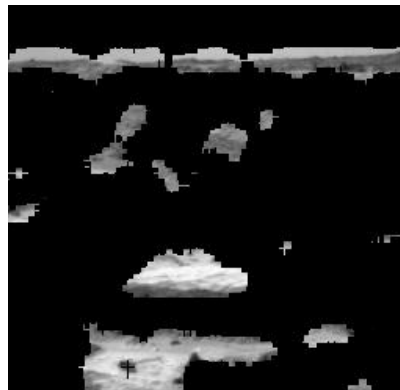
Shapiro LG and Stockman GC (2001):  
“Computer Vision”, pp 279-325  
New Jersey, Prentice-Hall



Sol 3, Mars  
Pathfinder Mission

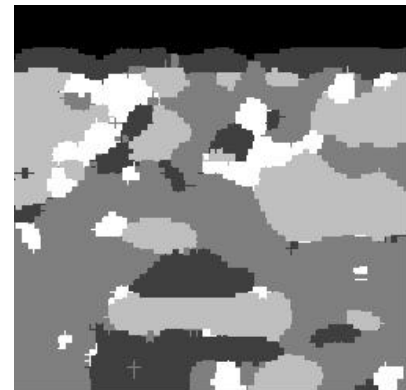


Sky / Flat



Dust / Horizon

...etc...

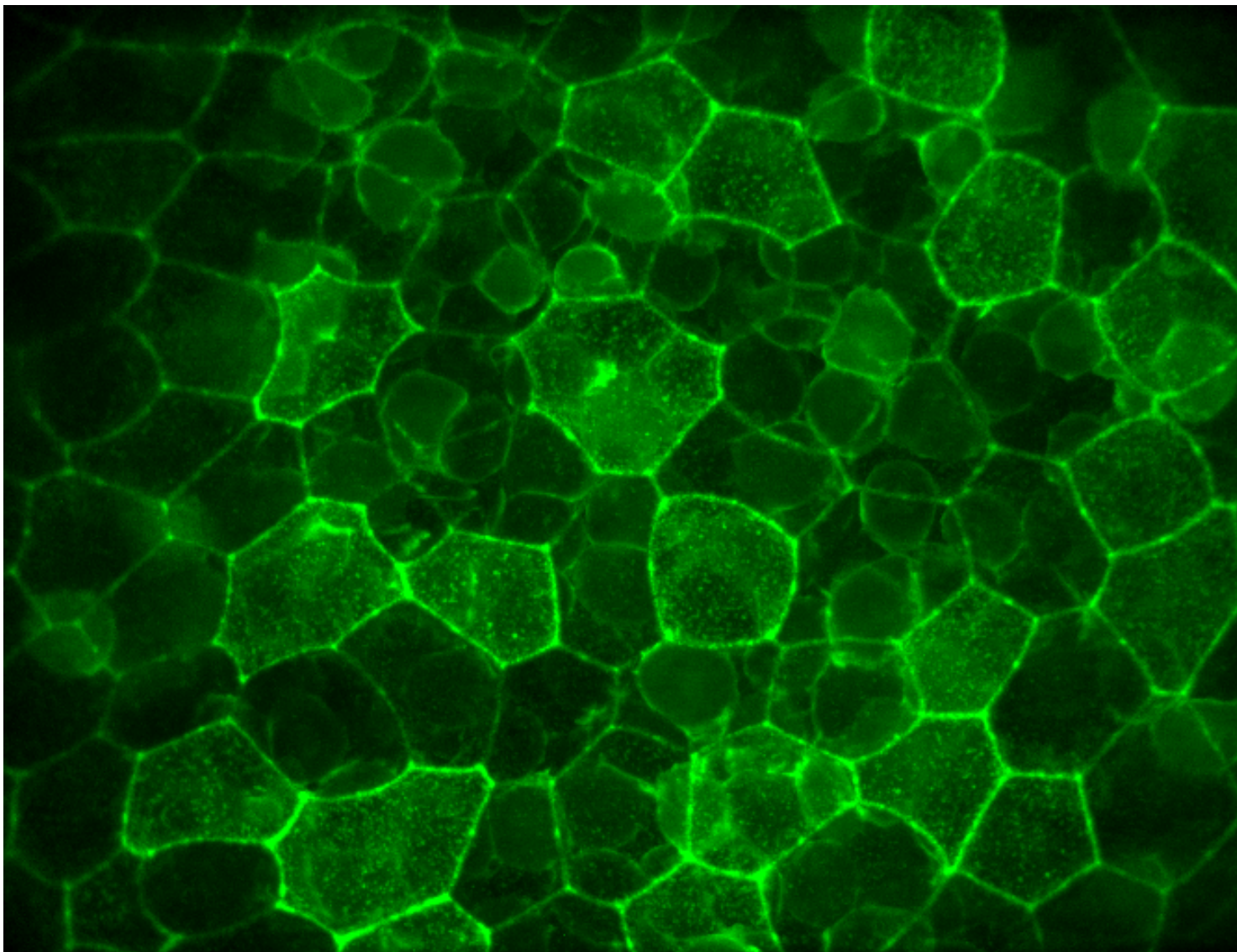


Final segmentation

- Not only objects as ROIs, but features...



Scale Invariant Feature Transformation (SIFT), D Lowe (2004). Image from J Clemons (2009)



Max Z-Project from confocal microscopy of Fundulus N. [data by German Reig 2015]

## Problems

- Lack of absolute criteria or standards (Ground Truth, Gold Standard [1,2])
- Missing or erroneous information (e.g. non-specific markers in samples)
- What to do? A “good” (i.e. carefully performed and controlled) acquisition ease this process

[1] Jason D. Hipp et al. Tryggo: Old Norse for truth: The real truth about ground truth. New insights into the challenges of generating ground truth maps for WSI CAD algorithm evaluation. Pathol. Inform 2012, 3:8

[2] Luc Bidaut, Pierre Jannin. Biomedical multimodality imaging for clinical and research applications: principles, techniques and validation. In Molecular Imaging: Computer Reconstruction and Practice (NATO Science for Peace and Security Series B: Physics and Biophysics), Springer, 2008, ISBN-13: 978-1402087516.

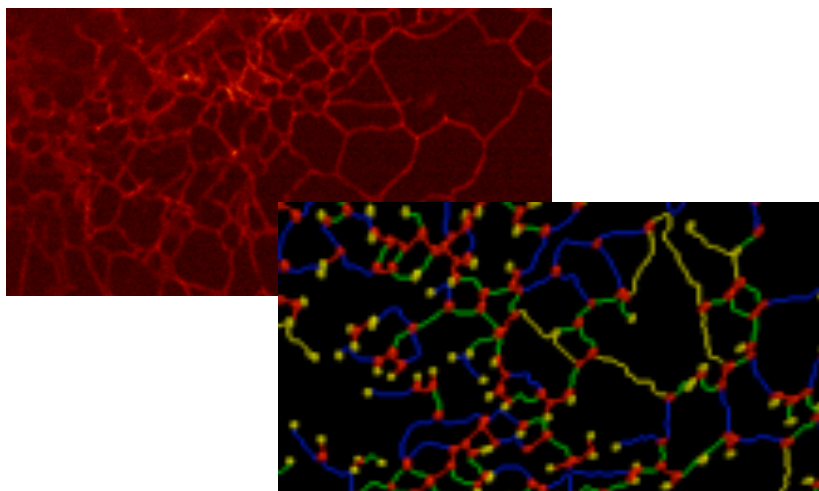


- Segmentation is the first step toward further quantifications

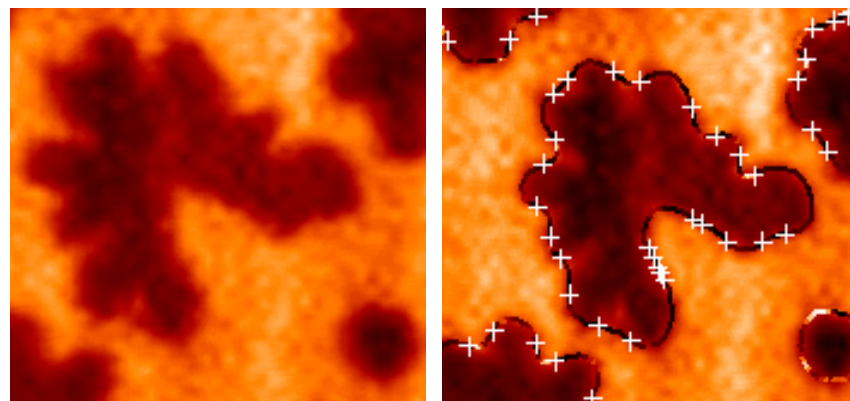
- In addition to images, ROI models and data structures can suit for different types of descriptions

Parameter estimation...

- Size: area, perimeter
- Boundary: inflections, shape
- Topology: connectivity, endpoints



Endoplasmic reticulum in a COS-7 cell  
O Ramírez, L Alcayaga (2012)

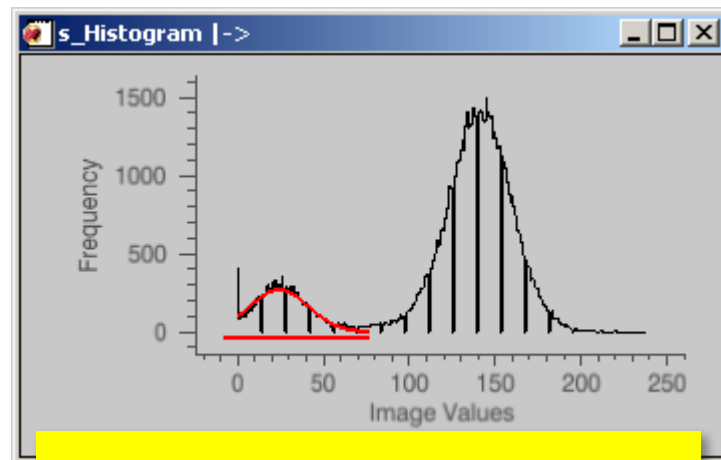
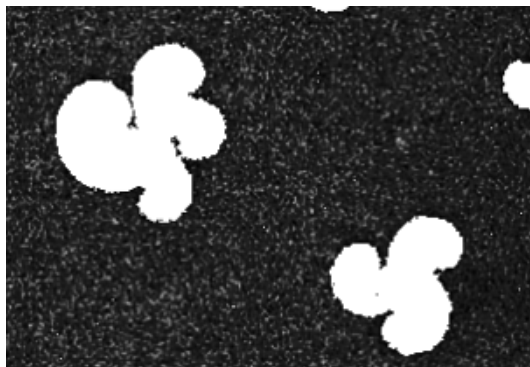
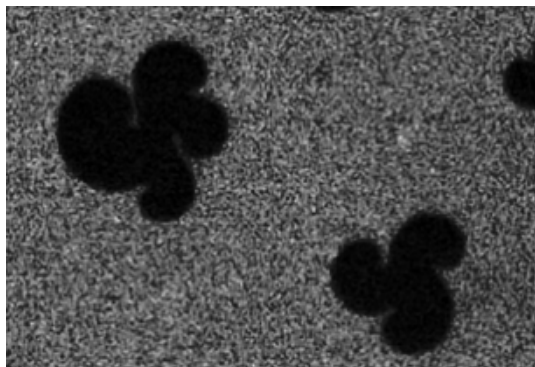


Lipid monolayers  
J Jara (2006), Fanani et al (2010)

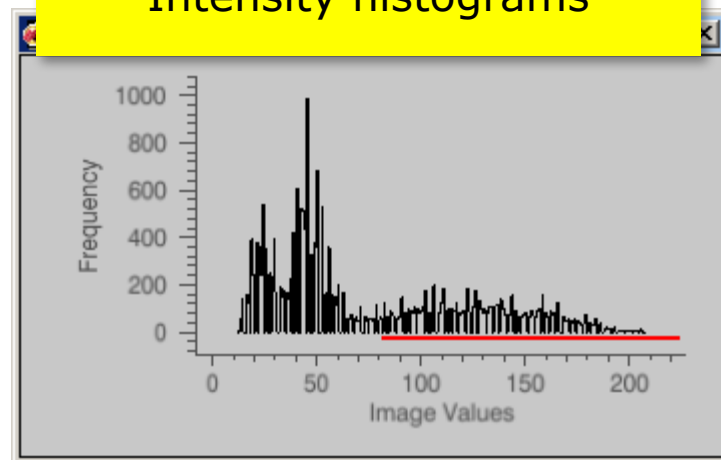
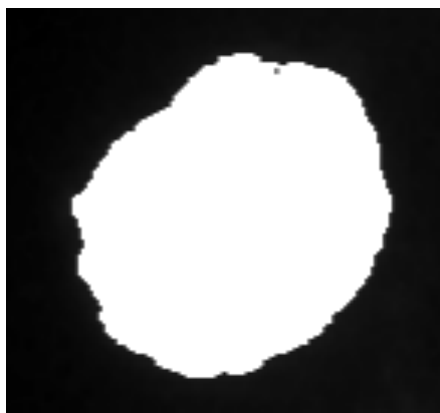
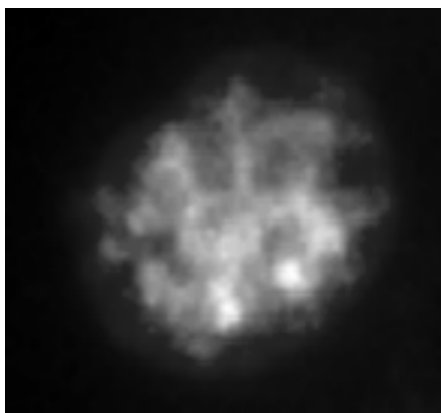
1. Classic approaches (filters)
  - Thresholding
  - Matrix convolution filters + threshold
  - Mathematical morphology
2. Advanced approaches
  - Shape priors (*pattern matching*)
  - Deformable models (active contours)
    - parametric
    - implicit



- Threshold filter  
segmentation: ROIs (white) / background (black)



Intensity histograms



- Otsu threshold
  - Idea: to separate the image pixel in two classes (sets), minimizing the sum of variances from both classes



$$\min \sigma_w^2(t) = \omega_1(t)\sigma_1^2(t) + \omega_2(t)\sigma_2^2(t)$$

$t$  : threshold,  $\omega_i$  : probability of class  $i$

$$\omega_1 = \sum_0^t p(i) \quad \omega_2 = \sum_{t+1}^{top} p(i)$$

## Algorithm

1. Compute histogram and probabilities of each intensity level
2. Set up initial  $\omega_i(0)$  and  $\mu_i(0)$
3. Step through all possible thresholds  $t = 1 \dots \text{maximum intensity}$ 
  1. Update  $\omega_i$  and  $\mu_i$
  2. Compute  $\sigma_b^2(t)$
4. Desired threshold corresponds to the maximum  $\sigma_b^2(t)$
5. You can compute two maximums (and two corresponding thresholds).  $\sigma_{b1}^2(t)$  is the greater max and  $\sigma_{b2}^2(t)$  is the greater or equal maximum
6. Desired threshold = 
$$\frac{\text{threshold}_1 + \text{threshold}_2}{2}$$

N. Otsu (1979).

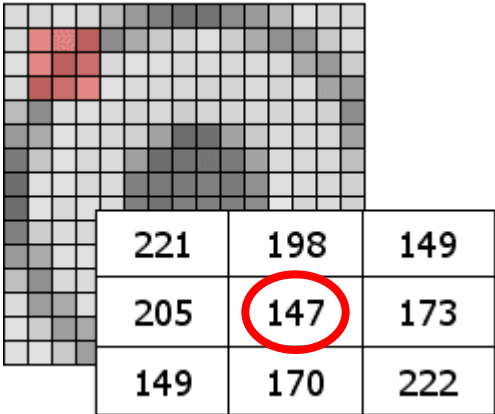
- $$(K \otimes I)_{i,j} = \begin{aligned} & (-1 \cdot 221) \\ & + (0 \cdot 198) \\ & + (1 \cdot 149) \\ & + (0 \cdot 205) \\ & + (0 \cdot \mathbf{147}) \\ & + (0 \cdot 173) \\ & + (-1 \cdot 149) \\ & + (0 \cdot 170) \\ & + (1 \cdot 222) = -63 \end{aligned}$$

<http://www.generation5.org/content/2002/convolution.asp>

$K$

|    |   |   |
|----|---|---|
| -1 | 0 | 1 |
| -2 | 0 | 2 |
| -1 | 0 | 1 |

$I$



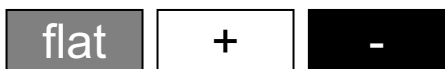
|     |            |     |
|-----|------------|-----|
| 221 | 198        | 149 |
| 205 | <b>147</b> | 173 |
| 149 | 170        | 222 |

$(K \otimes I)_{i,j} = (-1 * 222)$   
 $+ (0 * 170)$   
 $+ (1 * 149)$   
 $+ (-2 * 173)$   
 $+ (0 * \mathbf{147})$   
 $+ (2 * 205)$   
 $+ (-1 * 149)$   
 $+ (0 * 198)$   
 $+ (1 * 221) = +63$

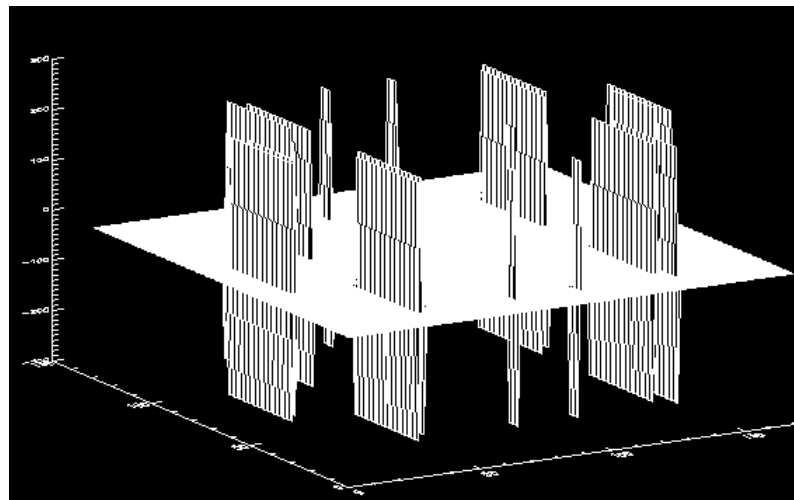
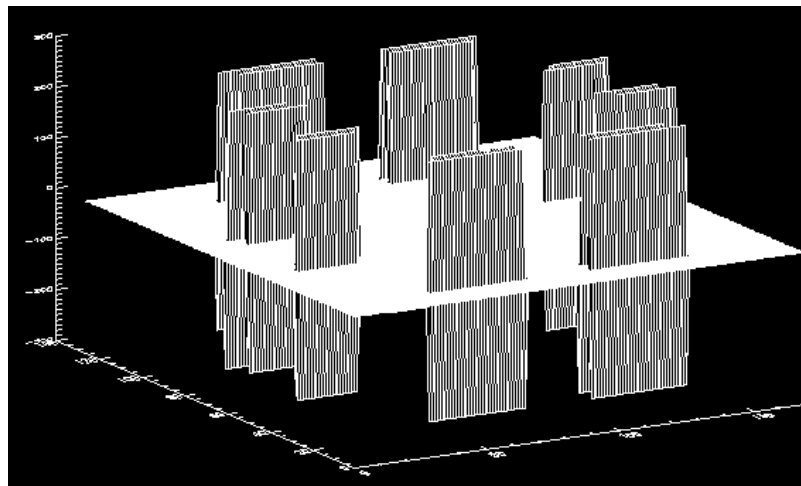
Matrix convolution can be implemented in different ways... beware of the algorithm!

- Intensity gradients (discrete approximation)

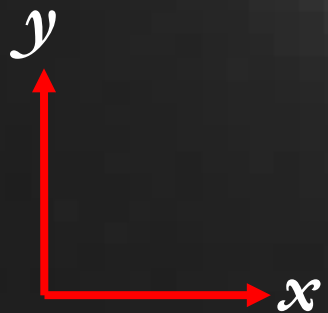
$$\frac{\partial I}{\partial x} \approx$$

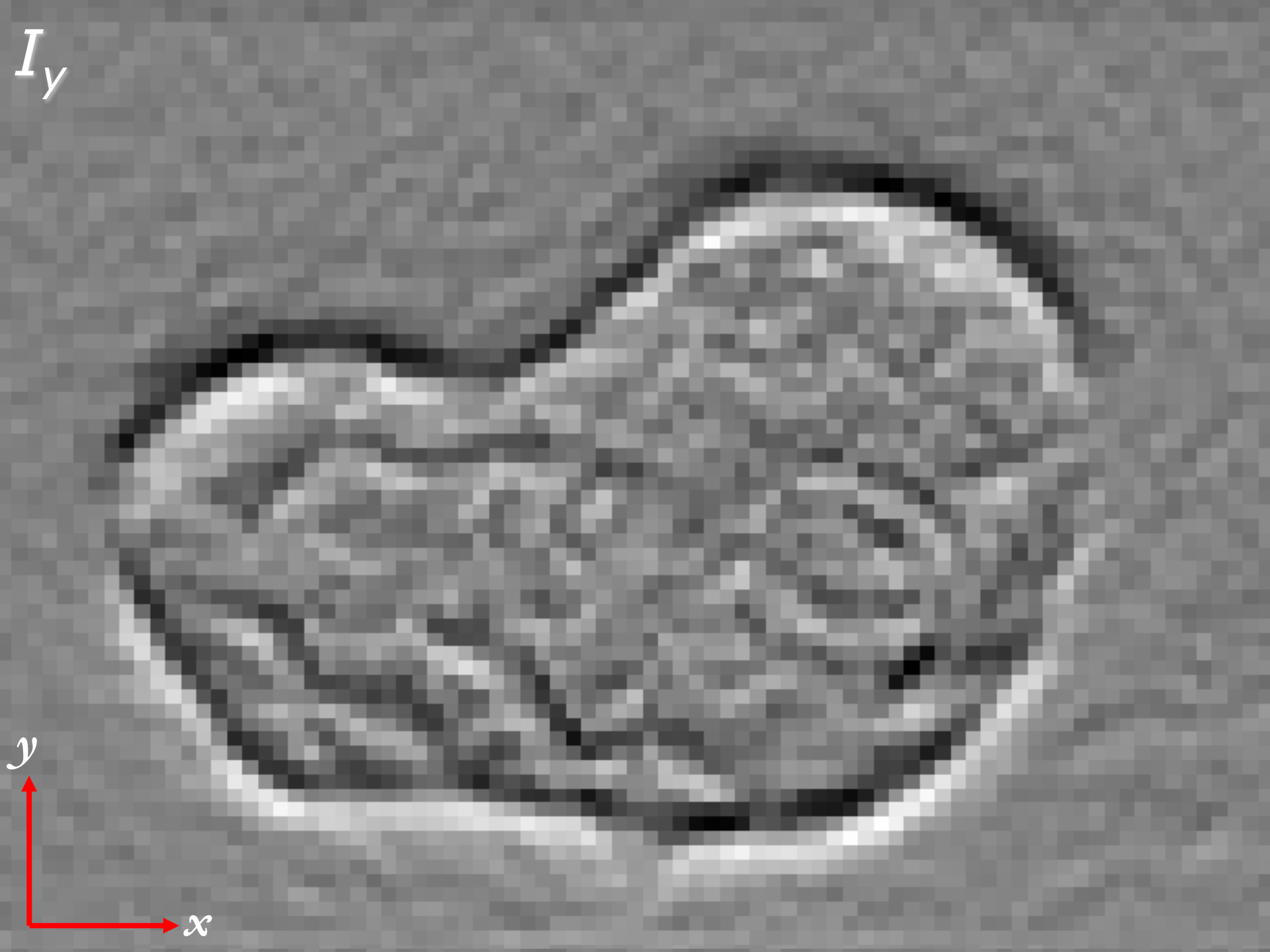


$$\frac{\partial I}{\partial y} \approx$$



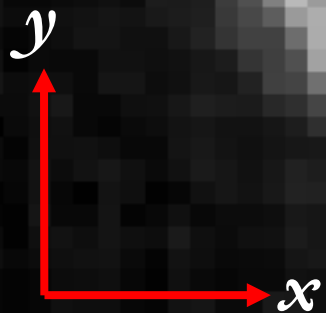
$$I = I(x, y)$$





“Edgemap”

$$|\nabla I| = |I_x| + |I_y|$$





- Intensity gradients (discrete approximation)

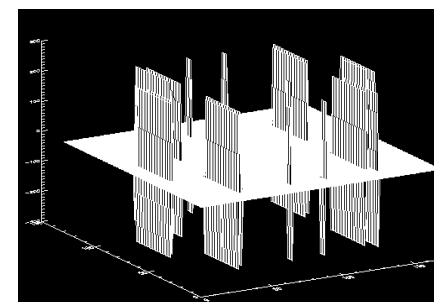


$$I = I(x, y)$$

$$\frac{\partial I}{\partial x} \approx \frac{I(x + \Delta x, y) - I(x, y)}{\Delta x} = Kx \otimes I$$

$\Delta x = 1 \text{ pixel}$

$$Kx = \begin{Bmatrix} 0 & 0 & 0 \\ 0 & -1 & 1 \\ 0 & 0 & 0 \end{Bmatrix}$$

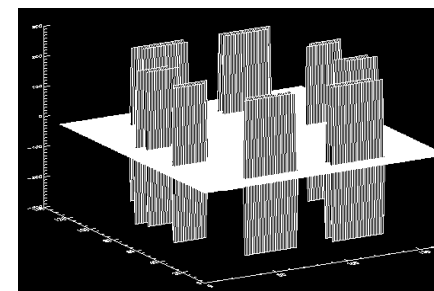


$$\frac{\partial I}{\partial y} \approx \frac{I(x, y + \Delta y) - I(x, y)}{\Delta y} = Ky \otimes I$$

$\Delta y = 1 \text{ pixel}$

$$Ky = \begin{Bmatrix} 0 & 1 & 0 \\ 0 & -1 & 0 \\ 0 & 0 & 0 \end{Bmatrix}$$

flat + -



- Kernels...

Laplace

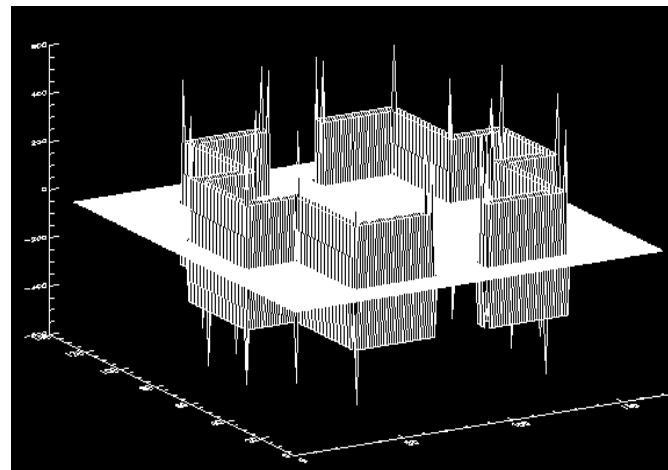
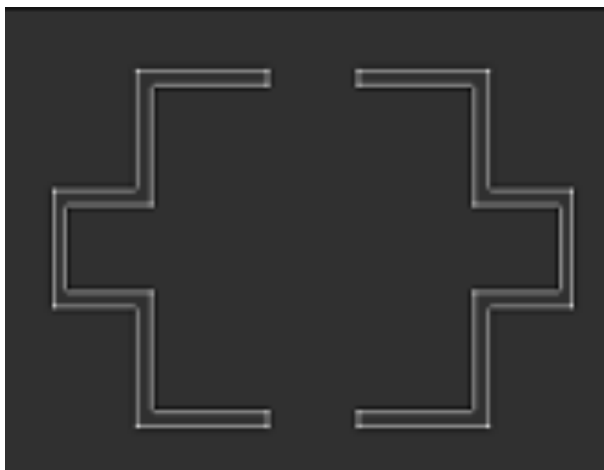
$$\nabla^2 I = \frac{\partial^2 I}{\partial x^2} + \frac{\partial^2 I}{\partial y^2}$$

$$\nabla^2 I \approx \frac{f(x + \Delta x, y) - 2f(x, y) + f(x - \Delta x, y)}{(\Delta x)^2} + \frac{f(x, y + \Delta y) - 2f(x, y) + f(x, y - \Delta y)}{(\Delta y)^2}$$

$$\nabla^2 I \approx \frac{f(x + \Delta x, y) + f(x, y + \Delta y) - 4f(x, y) + f(x - \Delta x, y) + f(x, y - \Delta y)}{(\Delta x)^2} = K_L \otimes I$$

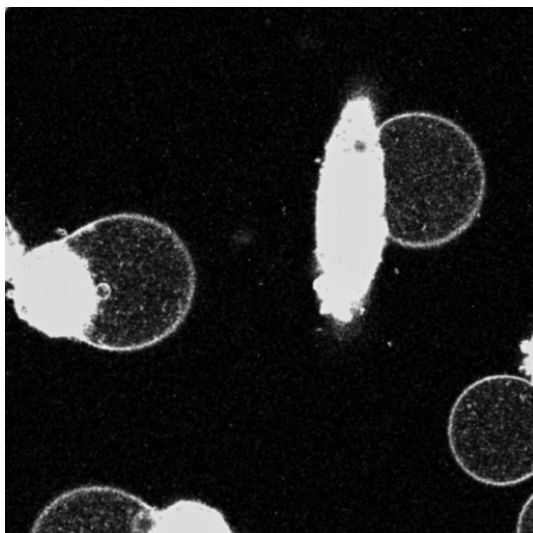
$\Delta x = \Delta y = 1$  pixel

$$K_L = \begin{Bmatrix} 0 & 1 & 0 \\ 1 & -4 & 1 \\ 0 & 1 & 0 \end{Bmatrix}$$

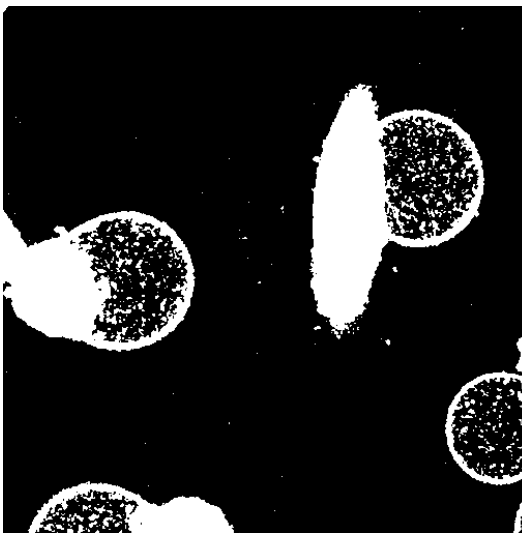
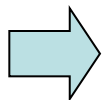


$$I = I(x, y)$$

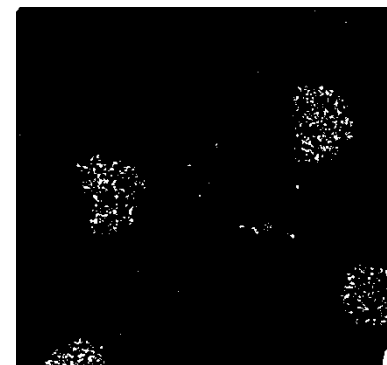
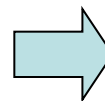
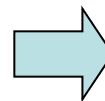
- Morphology based filters
  - Example: size selection



Input greyscale image



After thresholding...



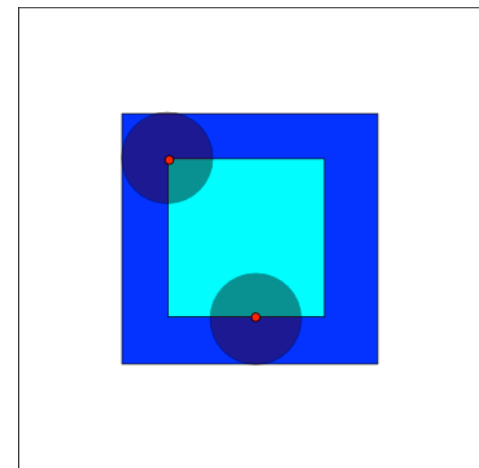
Size selection

How to define a size-select algorithm?

- Mathematical morphology

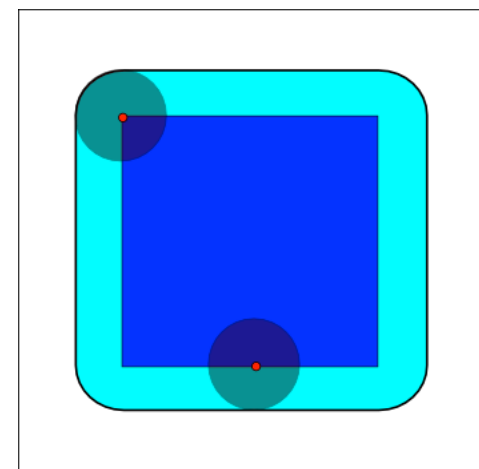
Erode

$$A \ominus B = \{z \in \mathbb{R}^2 | B_z \subseteq A\}$$



Dilate

$$A \oplus B = \bigcup_{b \in B} A_b$$



*What is B?*

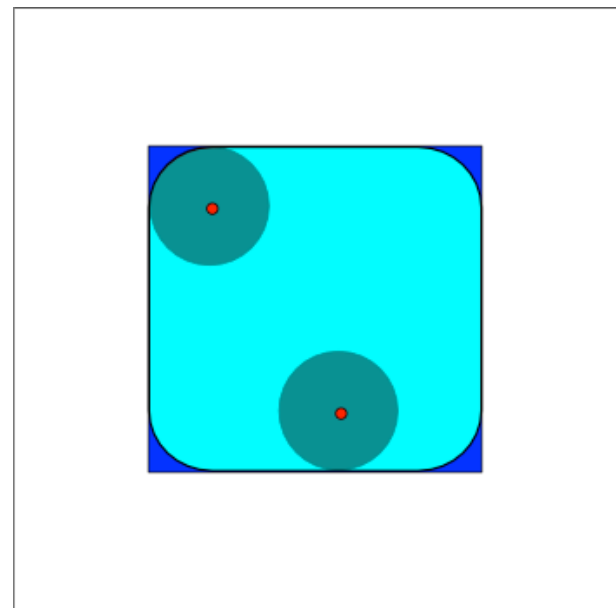
- Mathematical morphology

To open:

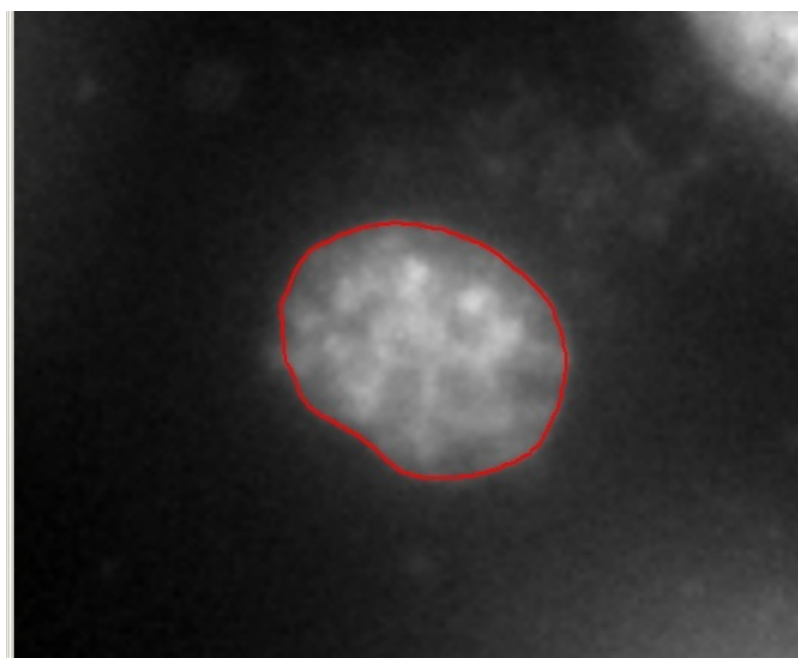
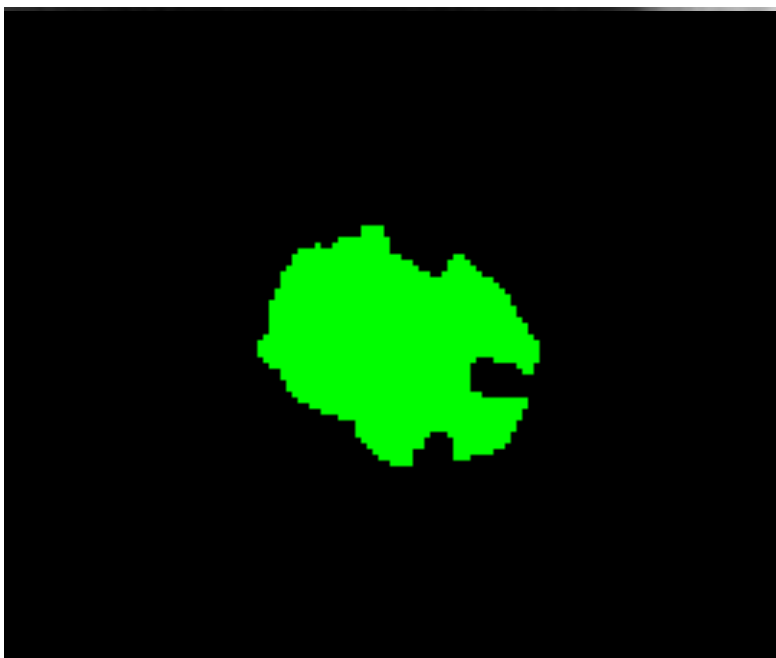
$$A \circ B = (A \ominus B) \oplus B$$

To close:

$$A \bullet B = (A \oplus B) \ominus B$$



- “Some” times more information is needed in order to achieve a good segmentation



- Template matching
  - “Classic model” Hough transform
  - Applies to circles, line segments and a variety of shapes

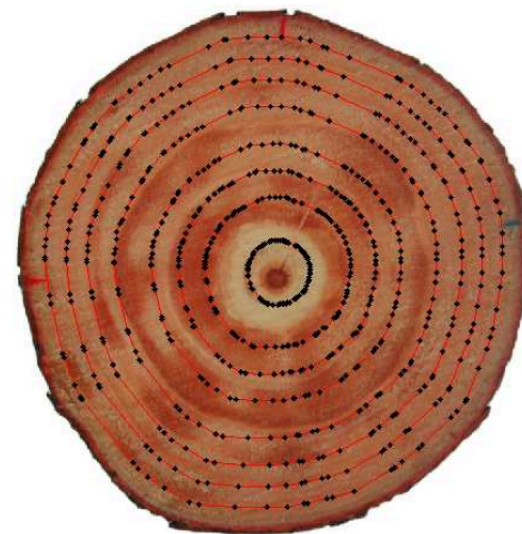
Hough P (1959)

If we can detect edges  
we can approximate a  
circle (or other shapes)

A test is performed to  
determine the circles  
with “best fit”



(c) Sample Tree 8.



(d) Fully automatic recognition with the input figure 5(c).

- Variational methods
  - Based on energy minimization, defining integral models
  - Idea: to include desirable features on segmented images (like homogeneous regions, short or smooth ROI boundaries)
  - Optimum solutions found by partial differential equations
  - Examples: Mumford-Shah, Ambrosio-Tortorelli, Chan-Vese (details in the book from Aubert & Kornprobst 2006)



image I



main discontinuities in I



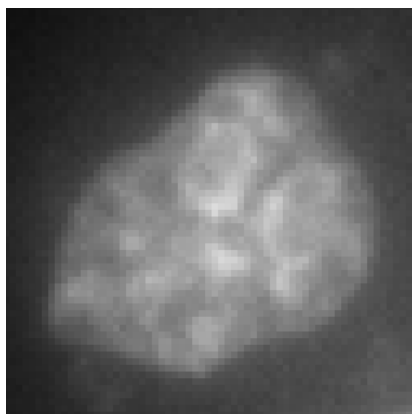
ROI boundaries B



piecewise smooth  
image J



- Active contour models
  - Optimization of different properties

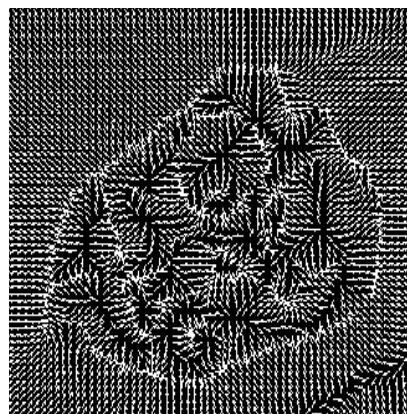


**input**  
image  
+ initial guess



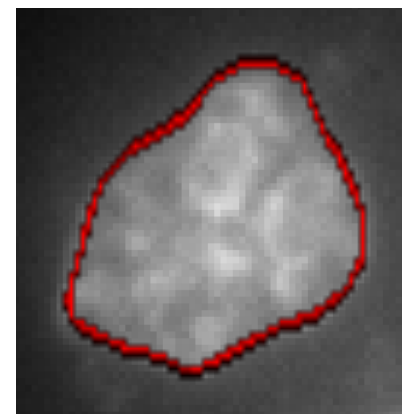
**contour  $C(s)$** 

- elasticity  
(contraction)
- rigidity  
(bending, cornering)



**force field**

- repulsion
- attraction



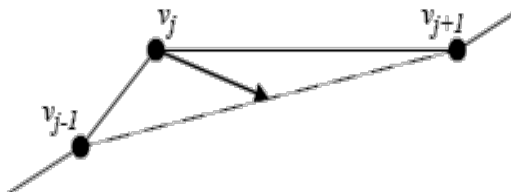
**output: force balance**  
minimal energy

First active contours approach:  
Kass, Witkin & Terzopoulos (1988)  
“Snakes”

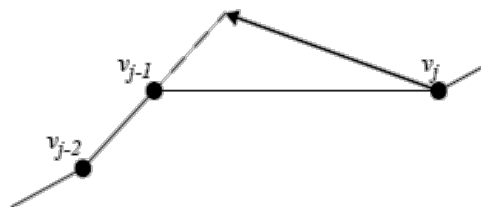
- Snakes: optimization derived from a **variational** approach
  - Minimization of an **integral functional**  
 “a snake minimizes its energy”

$$E = \int_0^1 \frac{1}{2} \left[ \alpha \left| \frac{\partial C(s)}{\partial s} \right|^2 + \beta \left| \frac{\partial^2 C(s)}{\partial s^2} \right|^2 \right] + E_{ext}[C(s)] ds$$

Elasticity term  
(coefficient  $\alpha$ )



Internal energy,  
contour dependant  
Rigidity term  
(coefficient  $\beta$ )



External energy,  
image dependant

Kass et al (1988) Snakes: active contour models  
Int. J. of Computer Vision 1(4): 321-331