CURSO DE POSTGRADO

Procesamiento de Imágenes y Bioseñales I & II

Image Segmentation I

Basics

Jorge Jara W.

Outline

1. Segmentation I

   - Digital image processing
   - Segmentation basics

2. Segmentation II
   - Advanced techniques

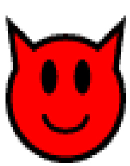
Introduction

Experience

Training

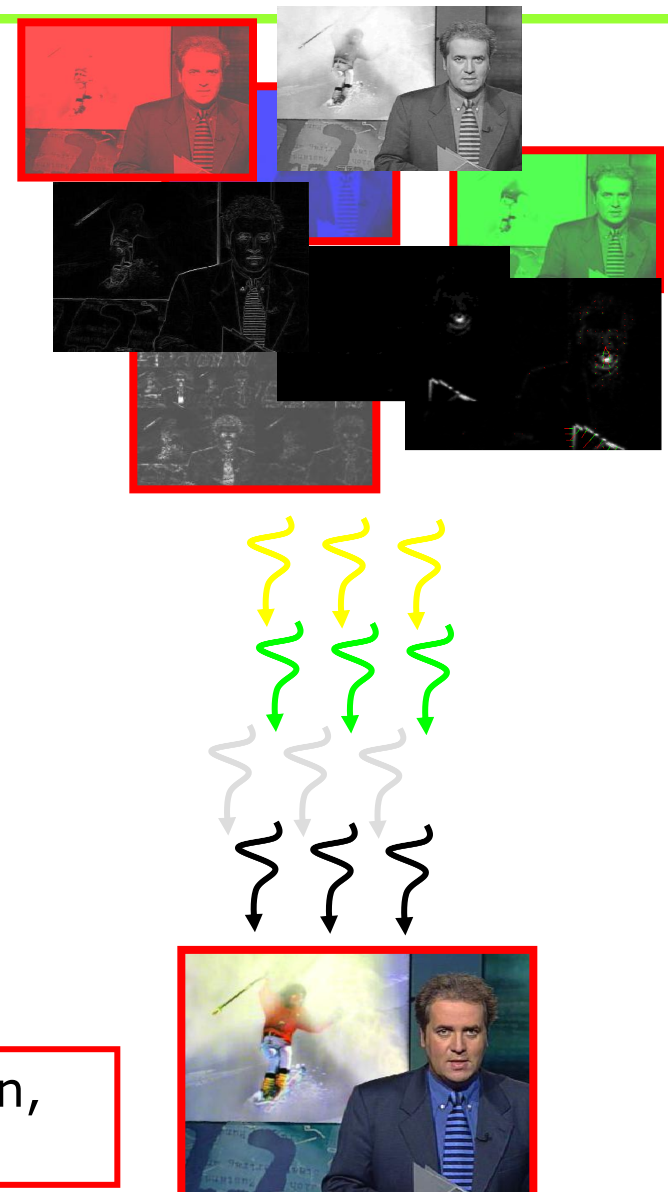
Expectation

Anticipation



Cognitive complement, ...

Symbolic representation, model



Introduction

- Common criteria

Color similarity (regions)

Color transitions or gradients (boundaries)

Introduction

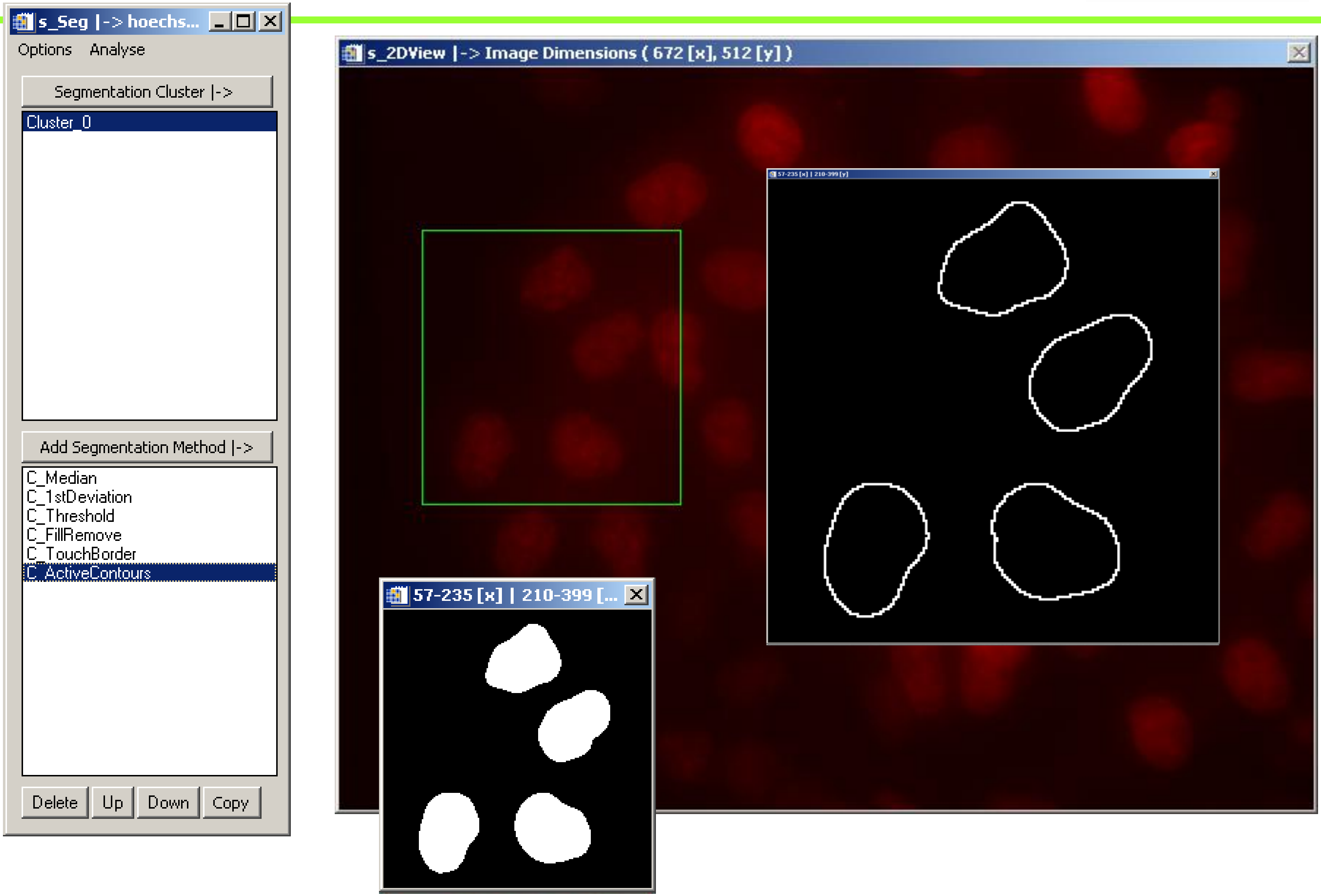
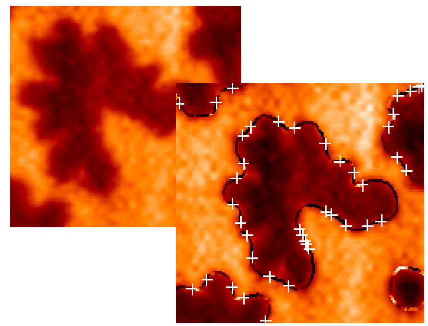
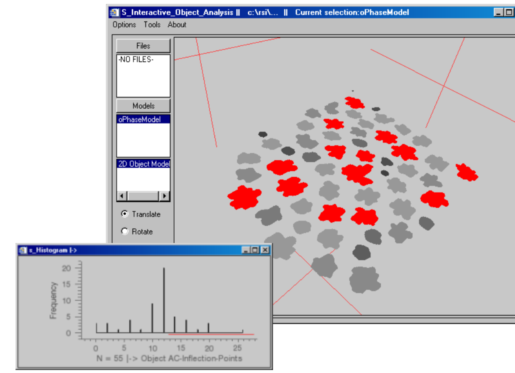


Image Processing



Acquisition
- Microscopes
- Cameras
- …

(pre)Treatment
- Restoring (e.g. PSF deconvolution)
- Contrast enhancement
- Artifact removal

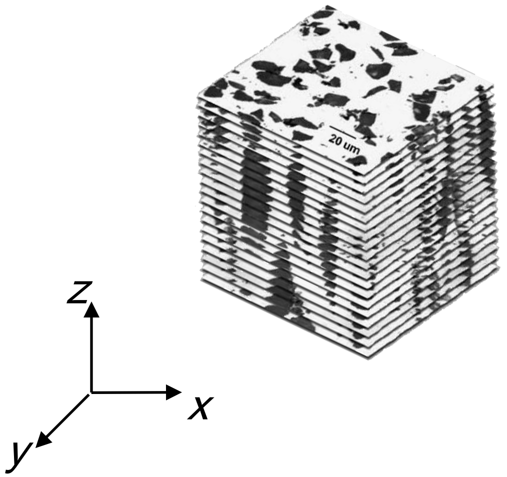
Analysis
- ROIs / models
- Measurements

Understanding
- Higher level task
- Hypotheses support

images **images** **images + descriptions**

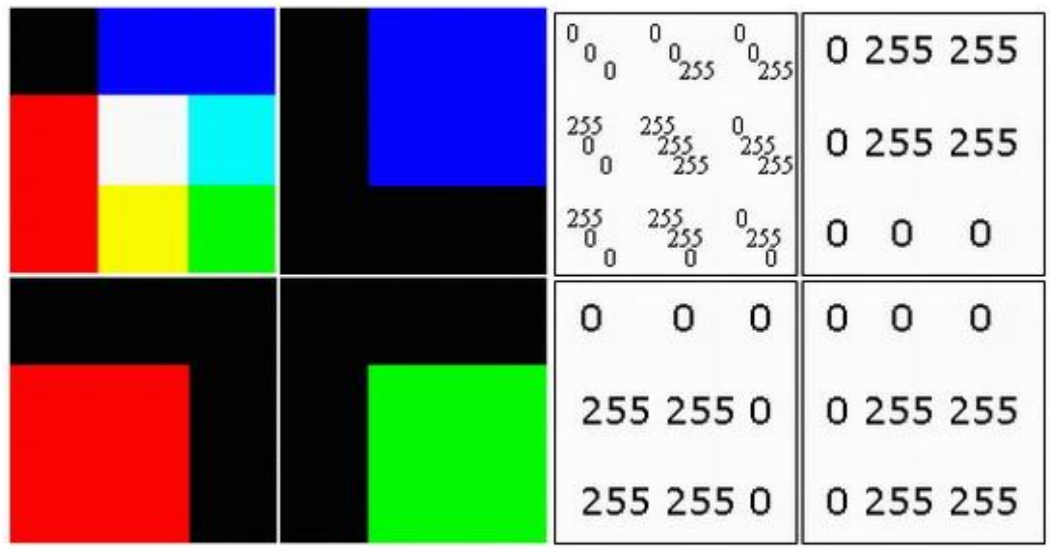
Digital image processing

i.e. with math & computers (among others...)



- Digital (computational) image processing
  - Digital… discrete, finite
    - A discrete set is composed by elements that are “isolated” one from another
      Examples:
      - Natural numbers {1, 2, 3, …} (infinite set)
      - Natural numbers from 1 to 10 (finite set)
    - If not discrete? Continuum
      Example: real numbers in the [0,1] interval (infinite set)

- 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 pixeles in cartesian coordinates (*x*, *y*)
  - A numeric value for brightness (intensity) or color is associated to each pixel



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

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

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

Digital Images

- Other than raster images…
  - A vector image is defined by using a set of base elements (like shapes or curves), instead of explicitly give the color/intensity for each pixel
  - Example: SVG images; base functions (wavelet, splines, Fourier)
- In order to show a vector image in a common digital screen (pixel matrix) a rasterization algorithm is applied

http://en.wikipedia.org/wiki/SVG

Digital Images

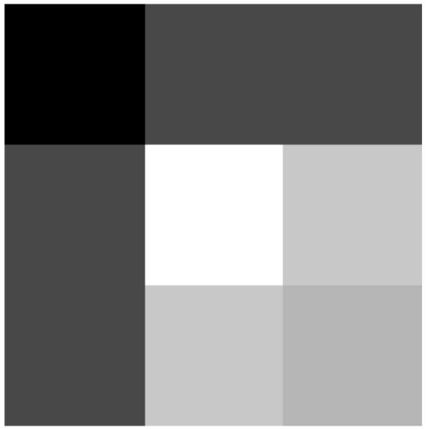
- ...so, a digital image can be treated as a function (in the mathematical sense)...
  - on a discrete domain
  - with numeric values associated to each elements, representing a property (such as color, brightness, depth, etc.)

Aubert & Kornprobst (2006)
Mathematical Problems In Image Processing

Digital Images

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

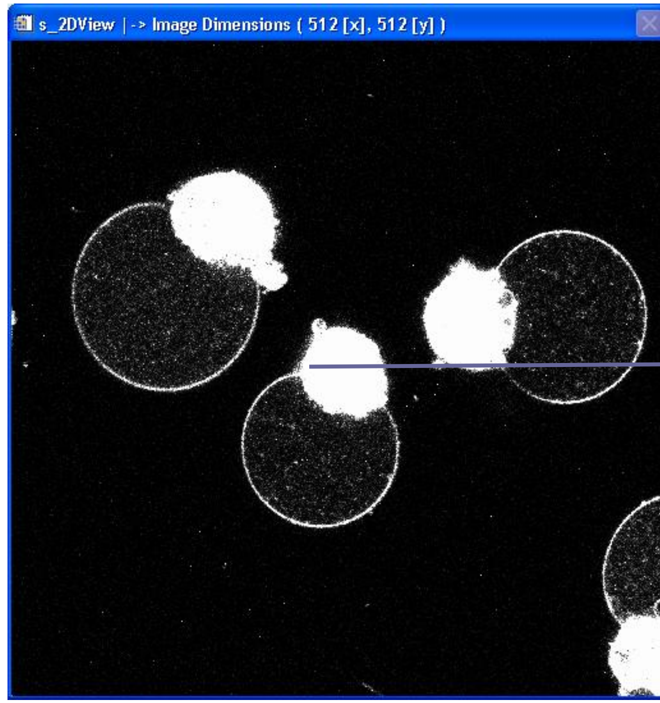
$$\begin{matrix} 0 & 85 & 85 \\ 85 & 255 & 170 \\ 85 & 170 & 85 \end{matrix}$$



$$I[x,y]$$

Digital Images

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)



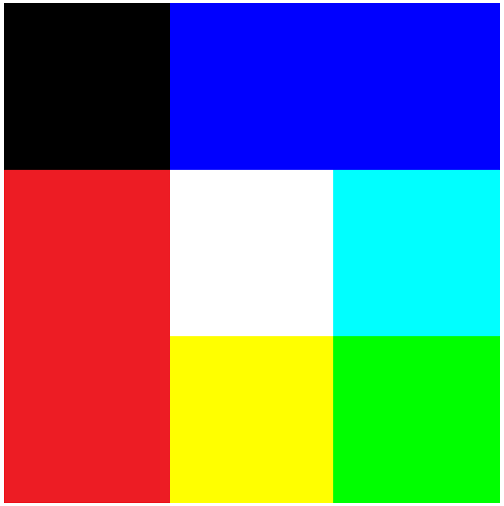
I(290,267) = 220

8 bit greyscale image

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

Digital Images

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



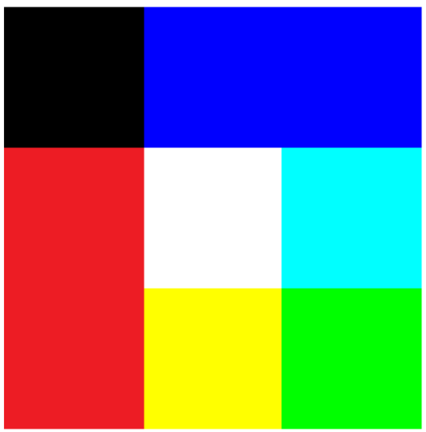
0	0	0
0	0	0
0	255	255
255	255	0
0	255	255
0	255	255
255	255	0
0	255	255
0	0	0

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

  - Analogous case: CMYK (Cyan, Magenta, Yellow, Black)

Digital Images

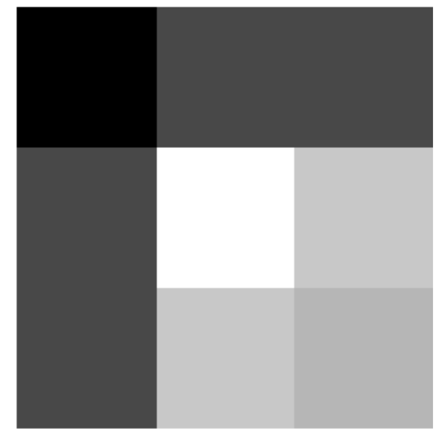
- From RGB to greyscale
  - The conversion might not be an average…



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

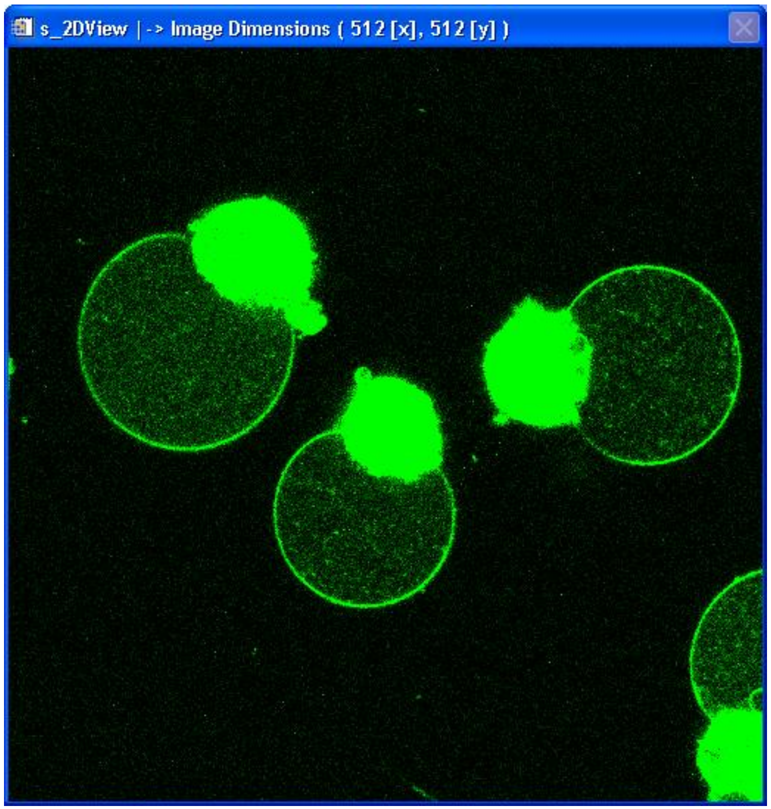
$I[x,y]$:

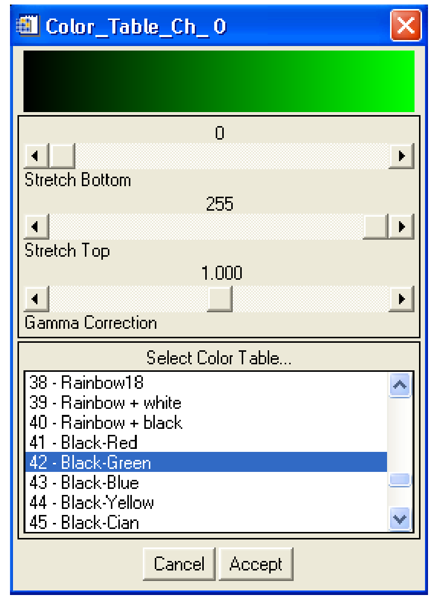
0	85	85
85	255	170
85	170	85



How good is the human eye resolving colors in R, G or B tones?

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

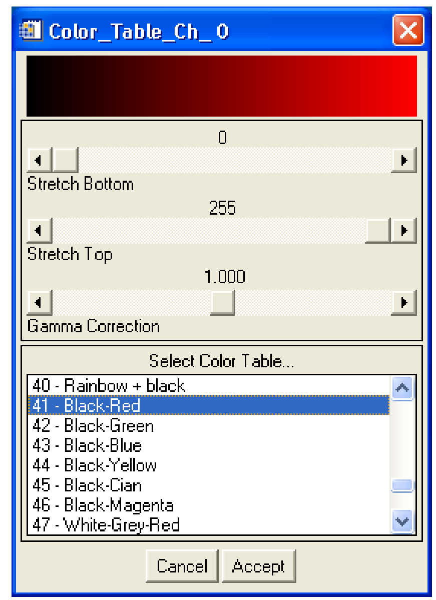




r	g	b
0	0	0
0	1	0
0	2	0
:	:	:
:	:	:
:	:	:
:	:	:
0	200	0
:	:	:
:	:	:
0	255	0

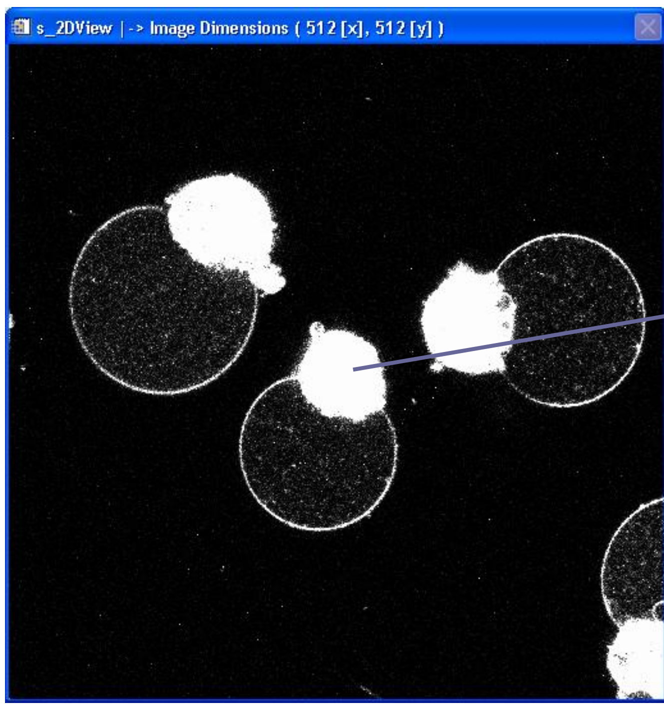
Digital Images

- Red scale...



r	g	b
0	0	0
1	0	0
2	0	0
:	:	:
:	:	:
:	:	:
:	:	:
220	0	0
:	:	:
:	:	:
255	0	0

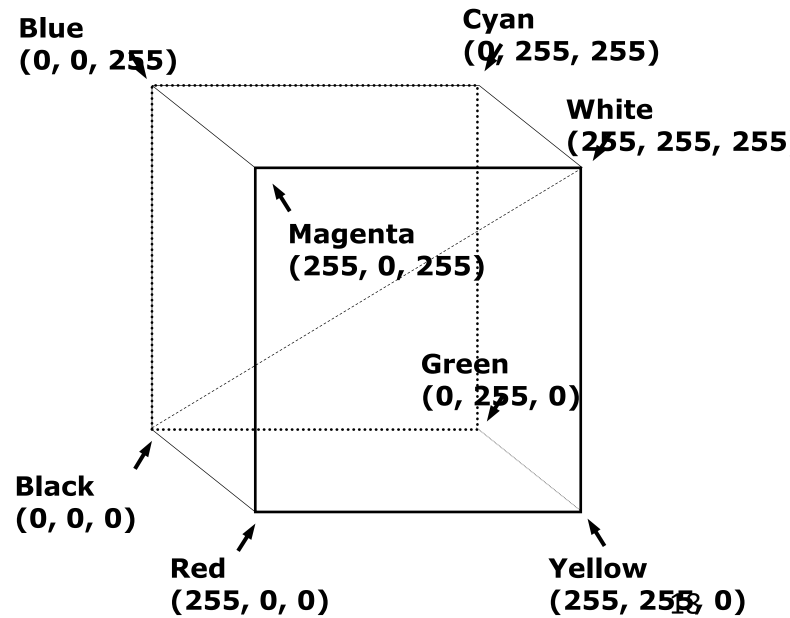
- ...or any custom color table



$I(290,267) = 220$

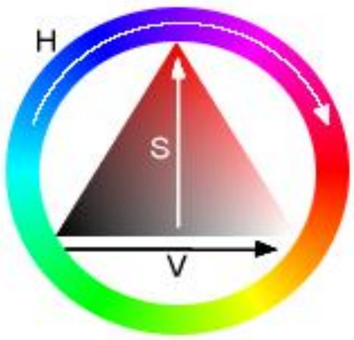
r	g	b
0	0	0
:	:	:
220	220	220
:	:	:
255	255	255

$$\text{Red} = [r_0, r_1, \ldots, r_{255}]$$
$$\text{Green} = [g_0, g_1, \ldots, g_{255}]$$
$$\text{Blue} = [b_0, b_1, \ldots, b_{255}]$$



HSV (hue, saturation, value) model

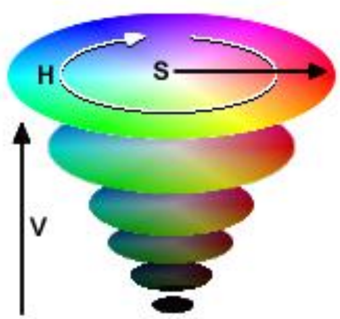
http://en.wikipedia.org/wiki/HSV_color_space



- **Hue**
  color „type", range 0-360° (0° red, 120° green, 240° blue)

- **Saturation**
  color „intensity", range 0-100%.

- **Value**
  brightness, range 0-100%.



HSV is a **non linear** transformation from the RGB color space.

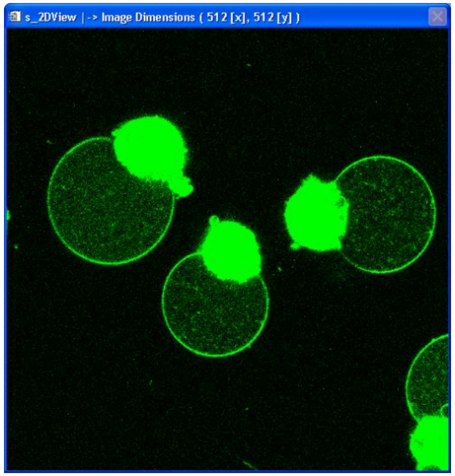
$$H = \begin{cases} \Theta & G \geq B \\ 2\pi - \Theta & G \leq B \end{cases}$$

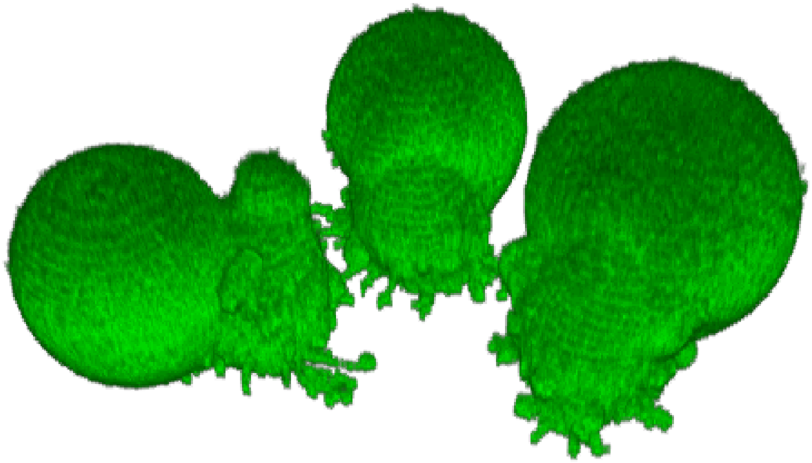
$$S = 1 - 3\frac{\min(R,G,B)}{R+G+B}$$

$$I = \frac{R+G+B}{3}$$

$$\Theta = \arccos\left[\frac{1}{2}\frac{(R-G)(R-B)}{\sqrt{(R-G)^2+(R-B)(G-B)}}\right]$$

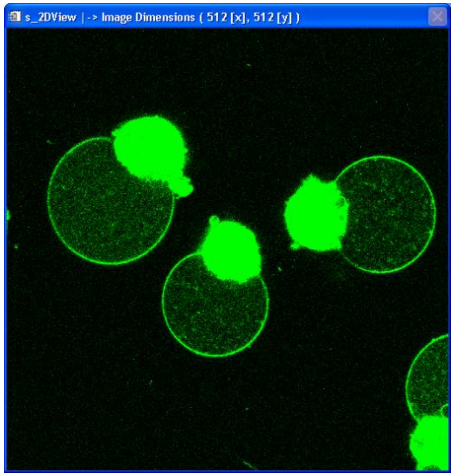
Digital Images

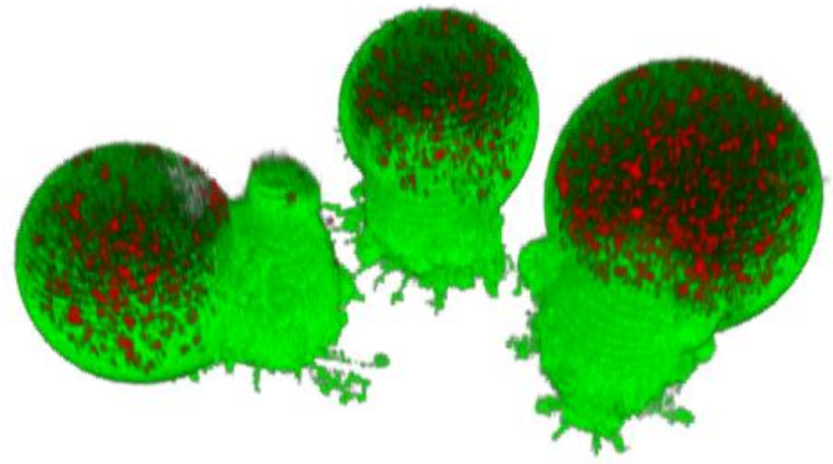




Occlusions may occur in 3D visualization

Digital Images





[R, G, B, α]

Opacity values can be associated to pixels (or voxels in 3D) for visualization purposes

Digital Images

- Function representation
  - Raster, SVG, base functions
- Color mode
  - grayscale
  - color (RGB, CMYK, HSV, Lab, etc.)
- Color depth (bit depth)
  - How many bits for how many values (e.g. 8 bits, 32 bits)
  - Number format
    - Integer (typically unsigned, e.g. TIFF)
    - Decimal (can be signed, e.g. ICS)
- Also important: storage mode
  - “Raw”: each pixel value is stored (lots of space)
  - Compressed, with or without information loss (e.g. JPEG *lossy* format, TIFF can be compressed or uncompressed)

- Image Analysis
  - The extraction of meaningful descriptions of features of interest from images

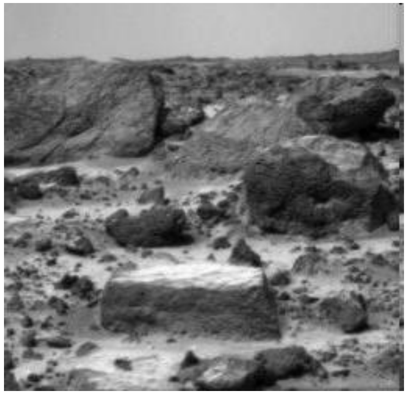
Adapted from
Young I, Gerbrands J, van Vliet L (1995)
Fundamentals of Image Processing. Delft: PH

- Some examples of analysis tasks
  - Objects/regions identification (segmentation)
    - Cells and/or their organelles
  - Registration: image, region and/or feature "matching"
    - Drift correction of the sample (from acquisition)
    - Relative speeds/displacements inside a given cell or reference system
    - Correspondence finding between images, objects or sections of these
  - Motion estimation, object tracking
    - Individual & collective migration
  - Morphology, topology, texture characterization
  - Classification
    - Detection of different populations, anomalies

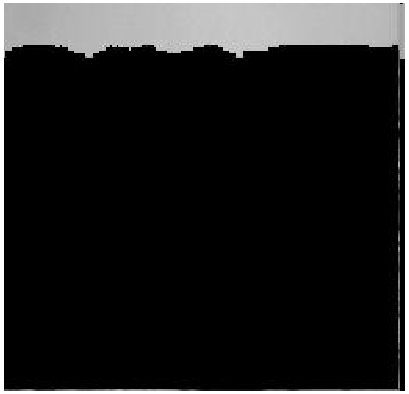
- Segmentation
  - The partitioning of a given image into regions of interest (ROIs) according to given criteria (e.g. color).
  - After segmentation, further characterizations can be performed upon the resulting ROIs.

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

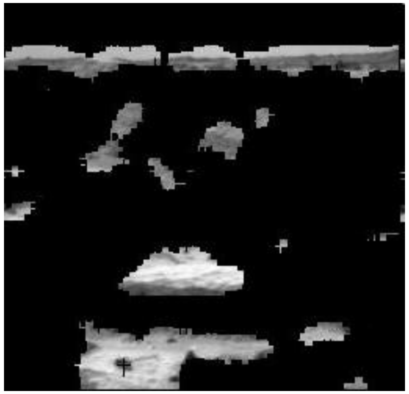
Segmentation



Sol 3, Mars
Pathfinder Mission

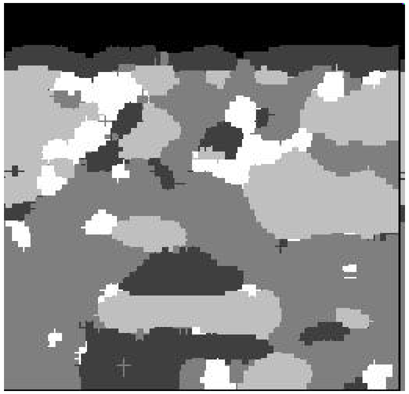


Sky / Flat



Dust / Horizon

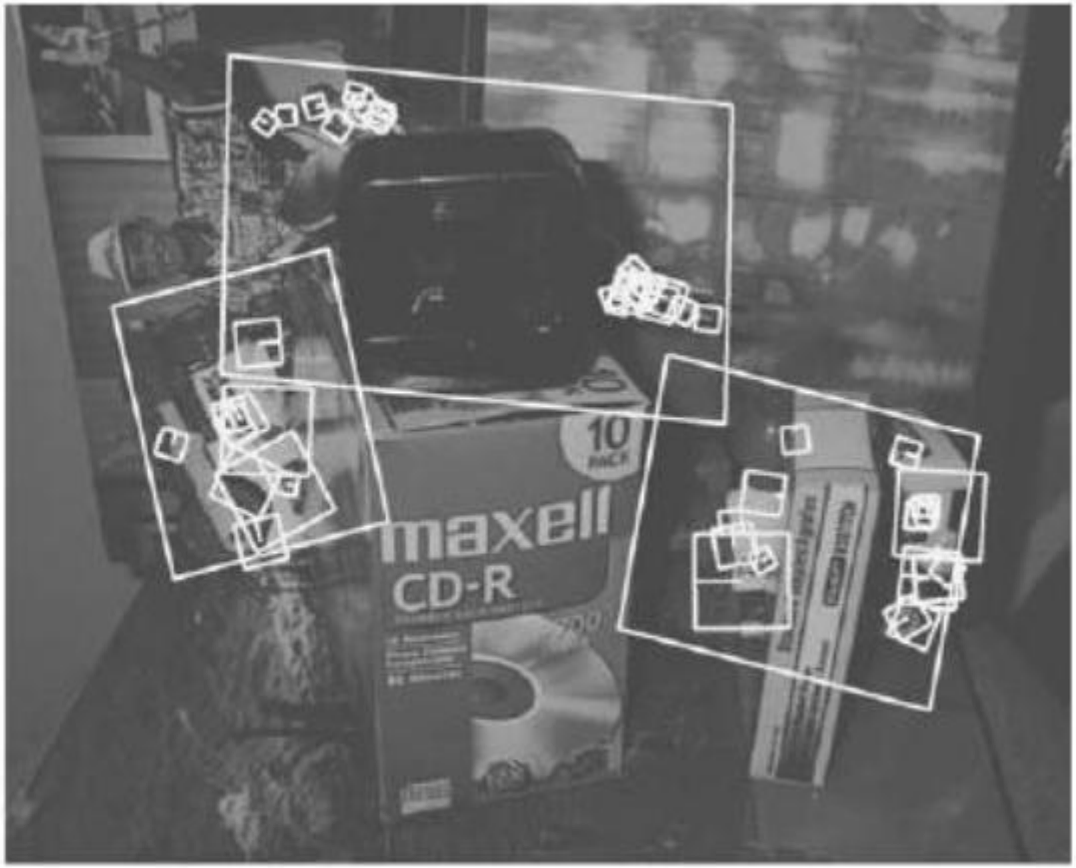
...etc...



Final segmentation

Images from NASA

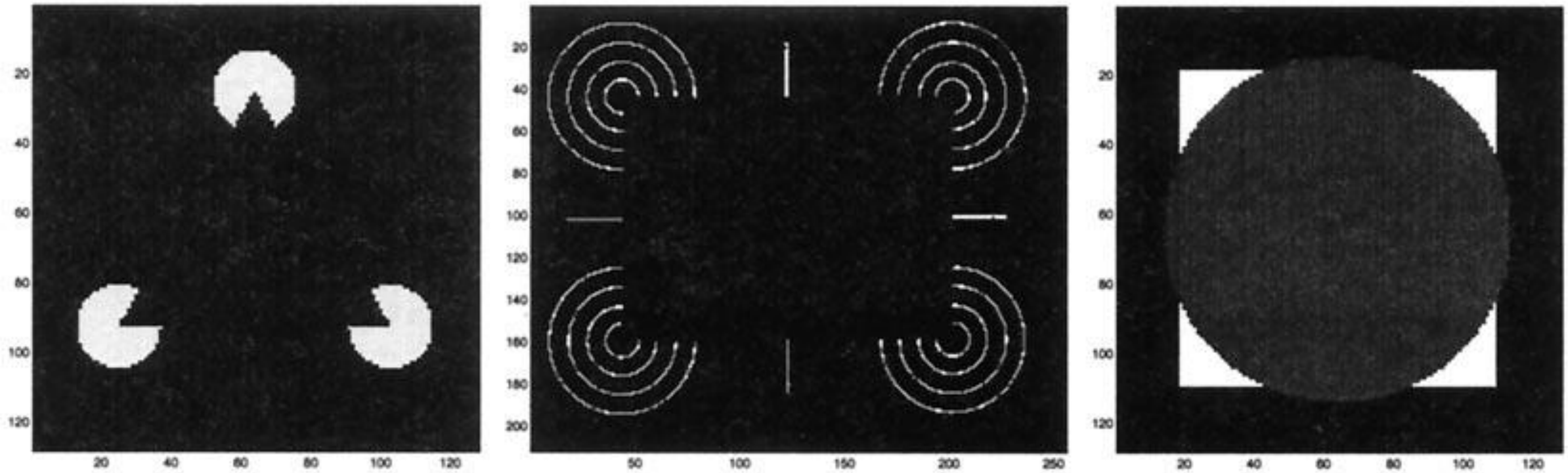
- Not only objects as ROIs, but features…



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

Segmentation

- Segmenting… ¿Which features? ¿Which criteria?

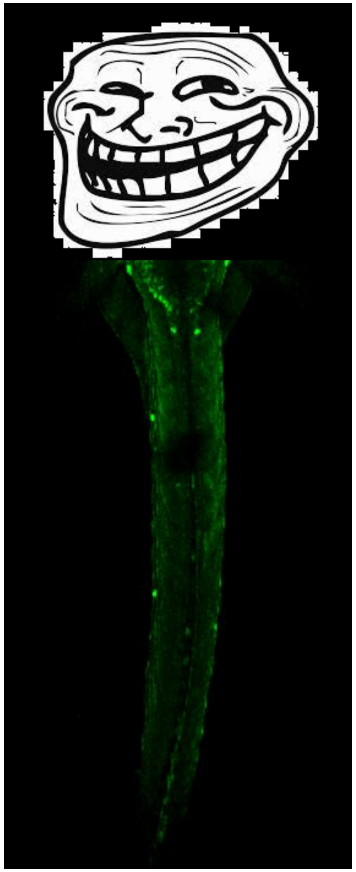


  - …not always (almost never) enough information for a 100% accurate segmentation.

Sarti & Sethian (2000) Subjective surfaces. PNAS

Segmentation

Problem?



CG Lemus (LEO, SCIAN-Lab, BNI)

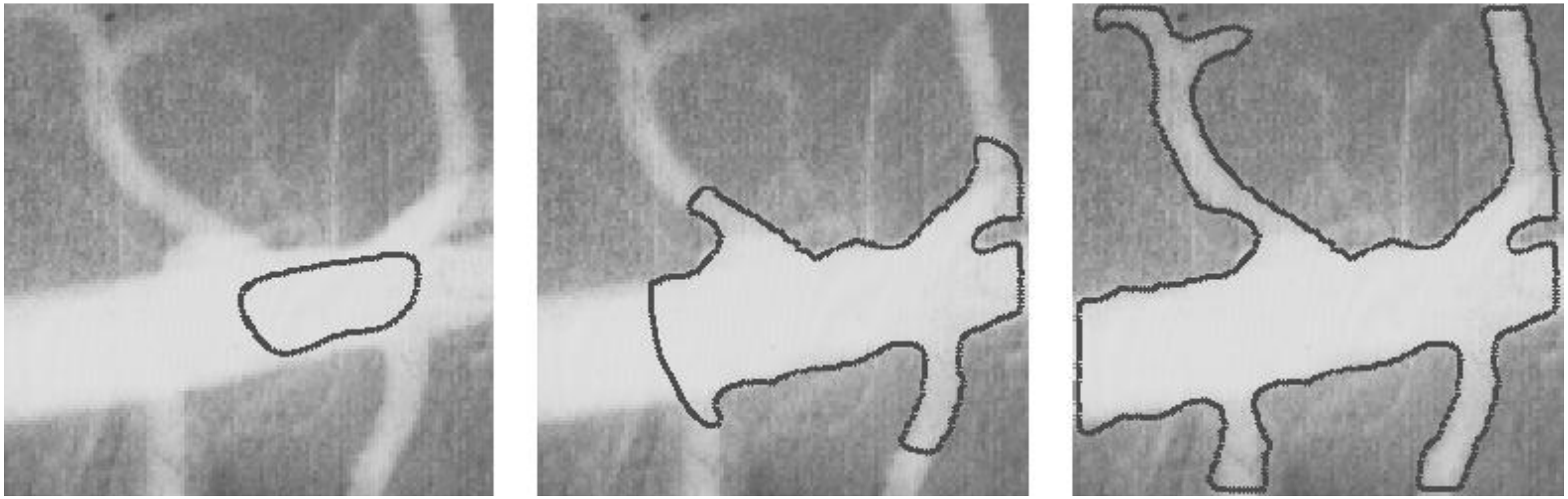
Problems

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

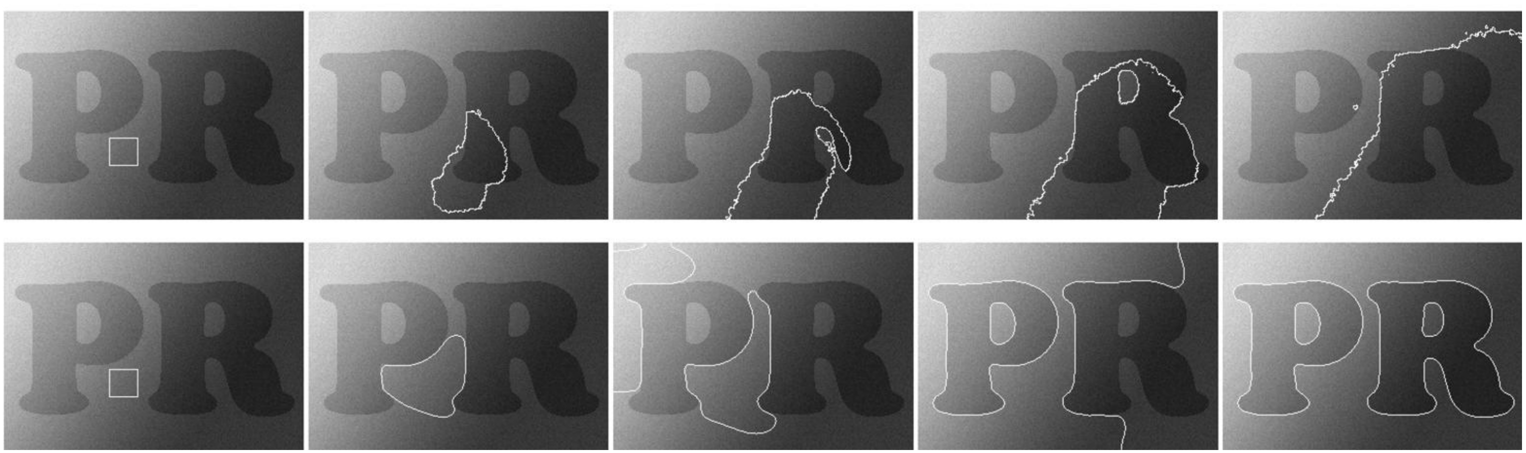
[1] Jason D. Hipp et all. Tryggo: Old norse for truth: The real truth about ground truth. New insights into the challenges of generating ground truth maps for WSI CAD algorithm evaluation. Pathol. Inform 2012, 3:8

[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 models regarded as “good” for a given scenario can be “bad” for other



J A Sethian – Fast marching and level set methods
http://math.berkeley.edu/~sethian/2006/Applications/Medical_Imaging/artery.html



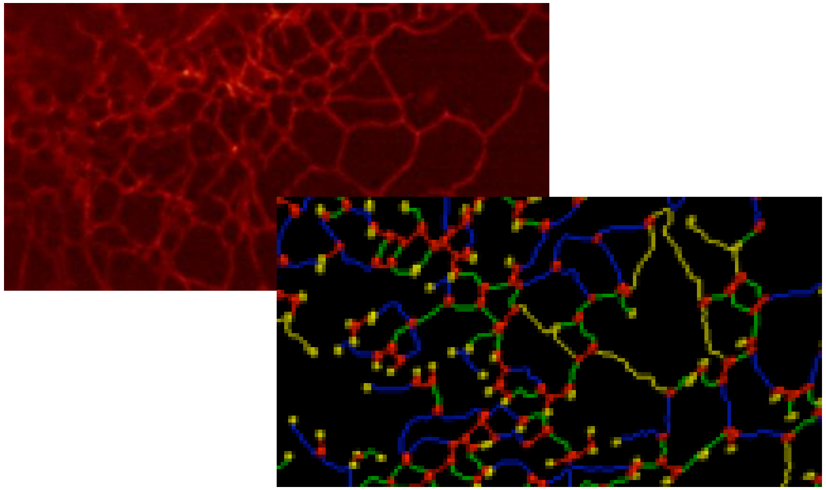
X Xie (2010) Magnetostatic Active Contours

Segmentation

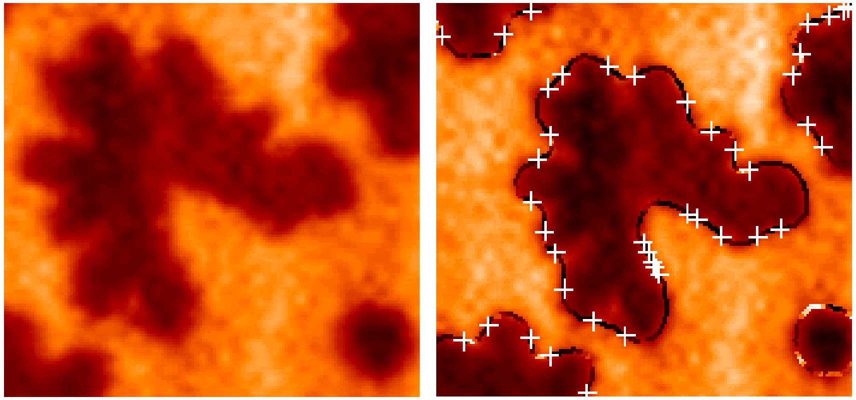
- Segmentation is the first step toward further quantifications
  - In addition to images, ROI models and data structures can suit for different types of descriptions

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)

Segmentation

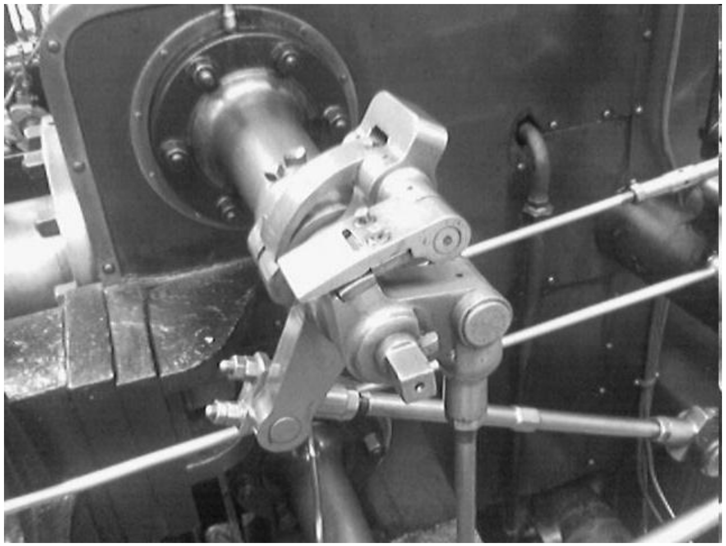
1. Classic approaches (filters)
   - Thresholding
   - Matrix convolution filters
   - Mathematical morphology
   - Fourier

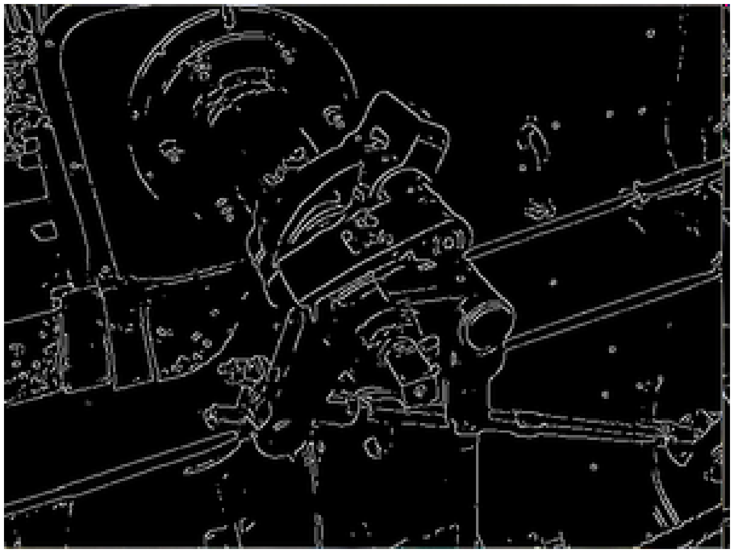
2. Advanced approaches
   - Shape priors (*pattern matching*)
   - Clustering methods (graph cuts, entropy)
   - Deformable models (active contours)
     - parametric
     - implicit

Segmentation – basics

- Filter

image → filter → filtered image

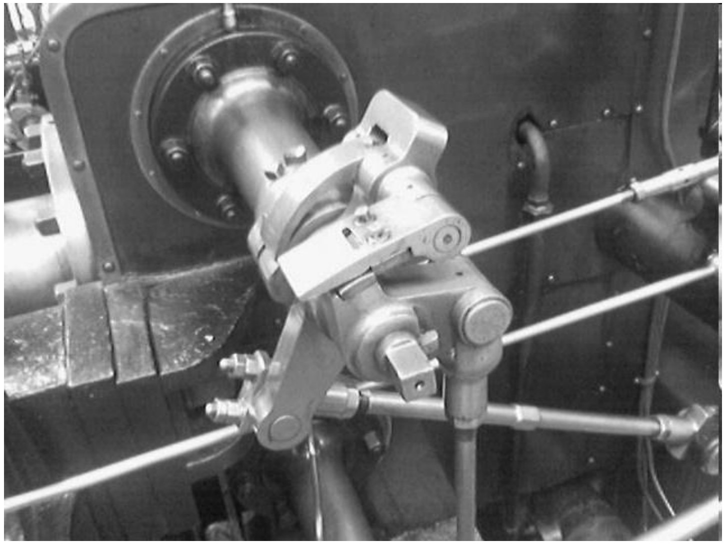


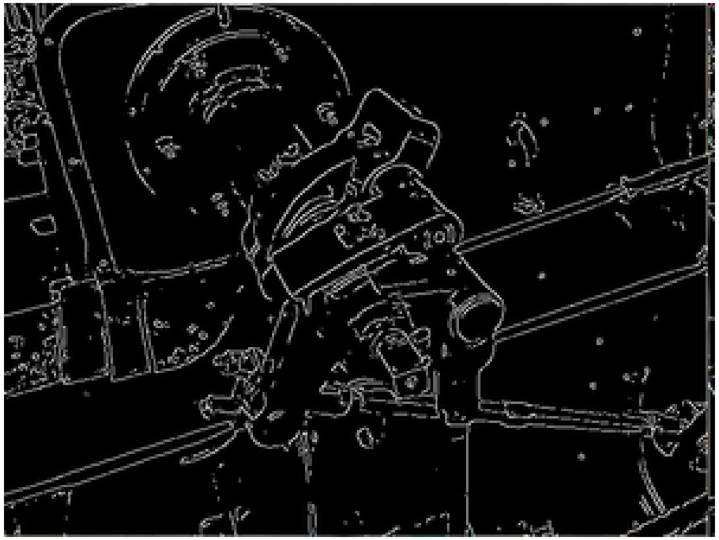


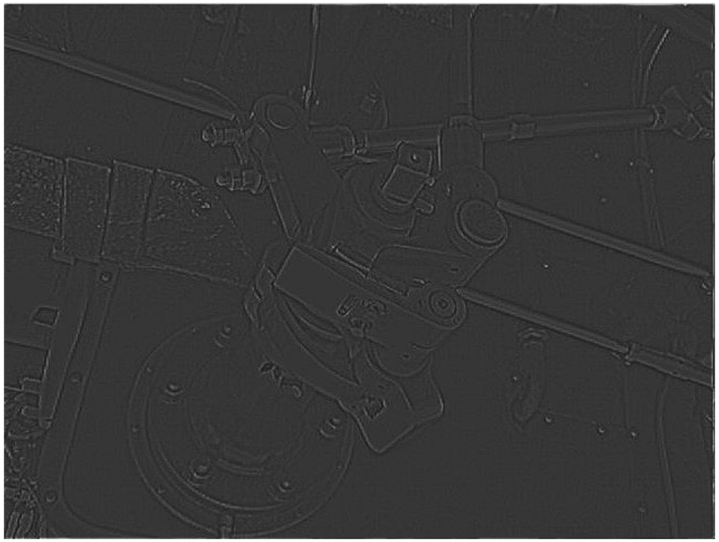
Imágenes from Wikipedia

Segmentation – basics

- A sample filter…



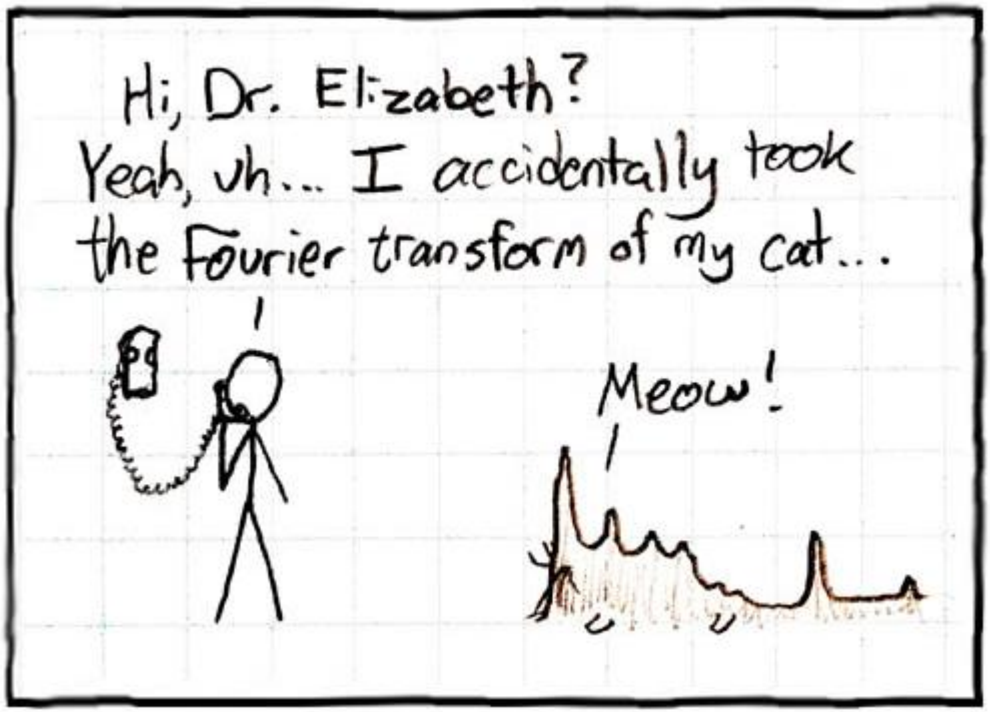




Domain transformation… frequency (Fourier), Wavelets, …

Jean-Baptiste Joseph Fourier

Jean-Baptiste-Joseph Fourier (21 de marzo 1768 en Auxerre - 16 de mayo 1830 en París), matemático y físico francés conocido por sus trabajos sobre la descomposición de funciones periódicas en series trigonométricas convergentes llamadas Series de Fourier, método con el cual consiguió resolver la ecuación del calor. La transformada de Fourier recibe su nombre en su honor. Fue el primero en dar una explicación científica al efecto invernadero en un tratado. Se le dedicó un asteroide que lleva su nombre y que fue descubierto en 1992.



Segmentation – basics

Jean-Baptiste Joseph Fourier

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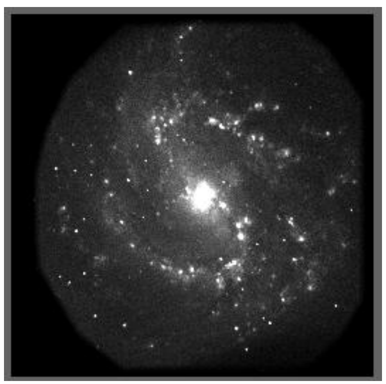
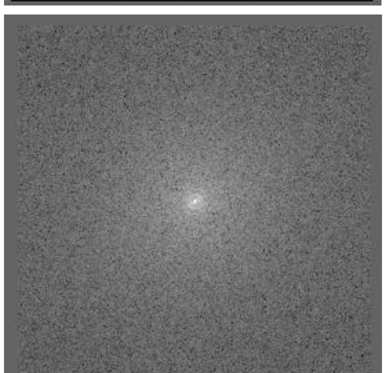


$$F(k,l) = \frac{1}{N^2} \sum_{a=0}^{N-1} \sum_{b=0}^{N-1} f(a,b)\, e^{-\iota 2\pi \left(\frac{ka}{N} + \frac{lb}{N}\right)}$$



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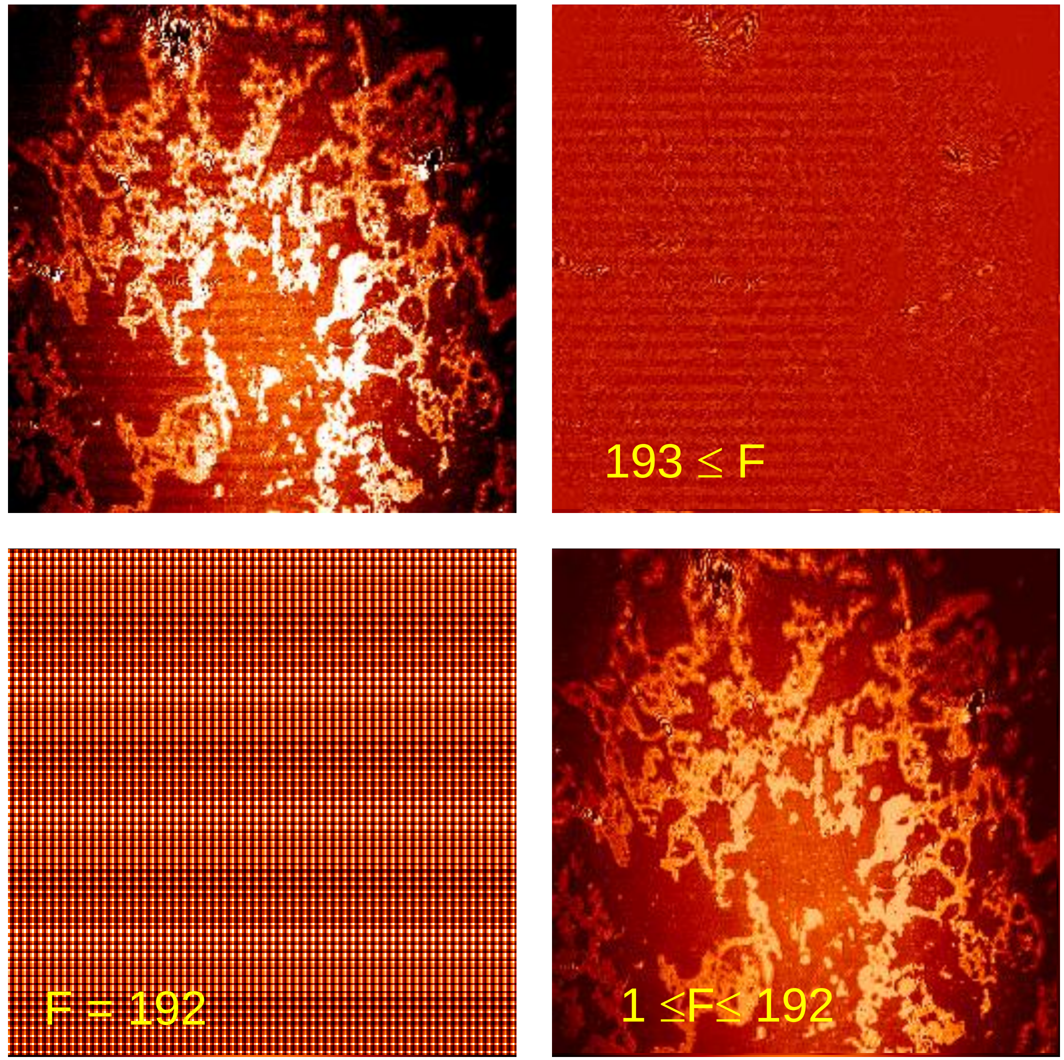
Segmentation – basics

Local filters:
- Linear
- Non linear

Global filters:
-Fourier
-Wavelets
- ...

Others:
- Adaptive analysis
- ...



Segmentation – basics

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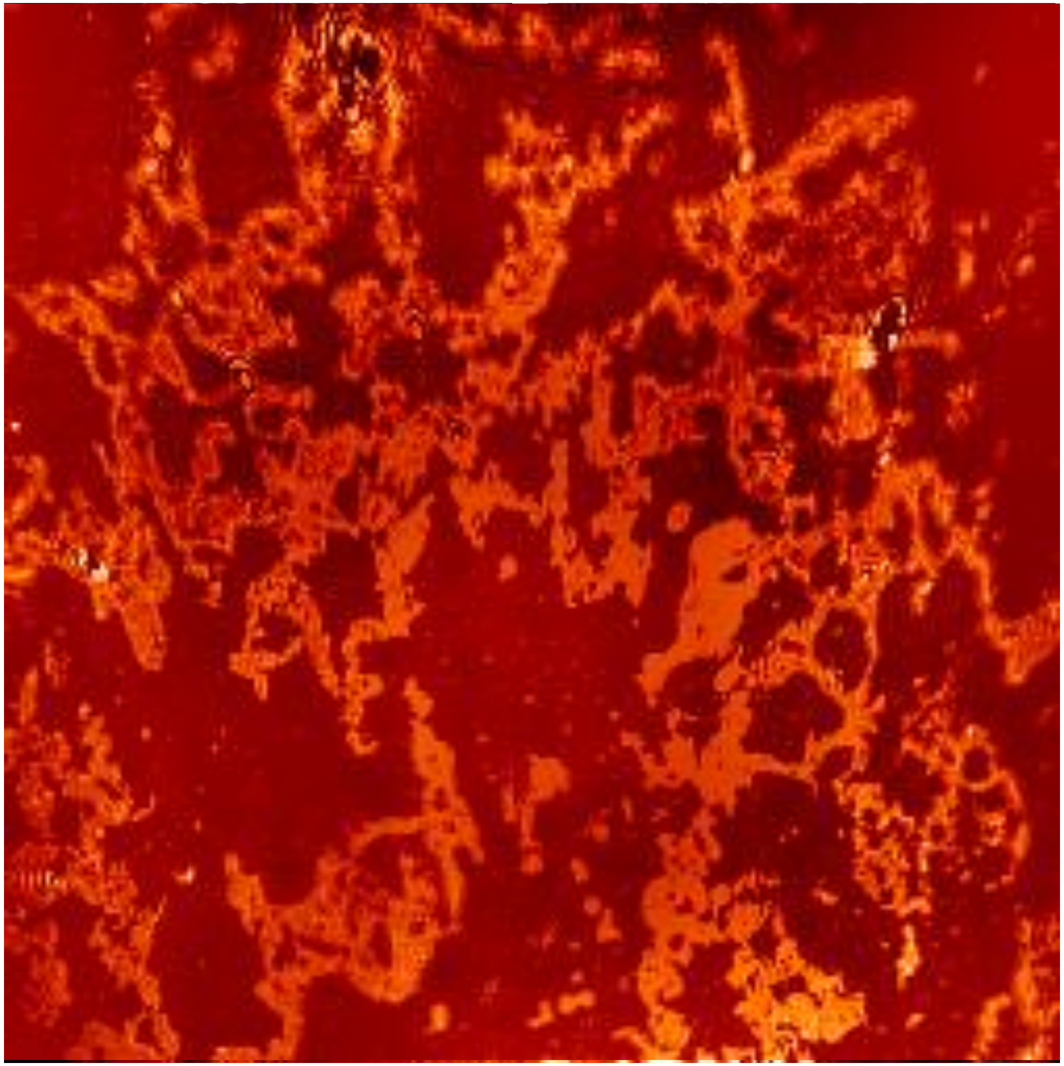


Segmentation – basics

Thresholding
- example: Otsu

Convolution based
- Convolution operation
- Examples: gradient, Laplace, Sobel, Gaussian

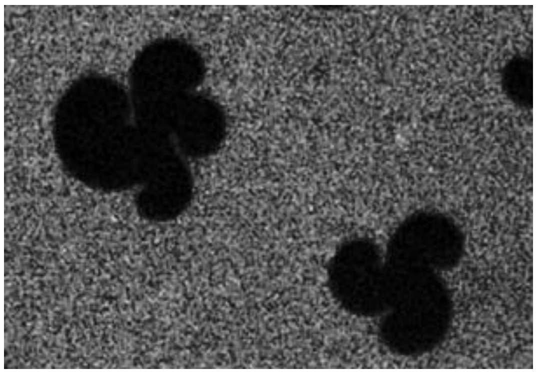
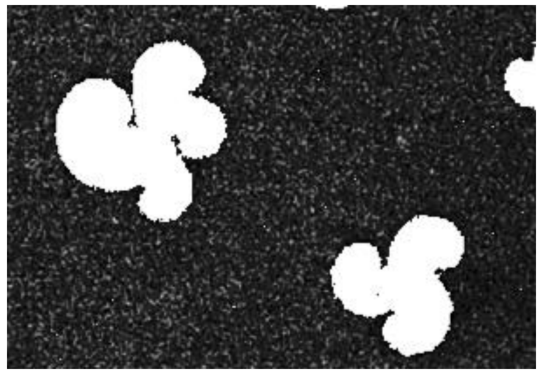
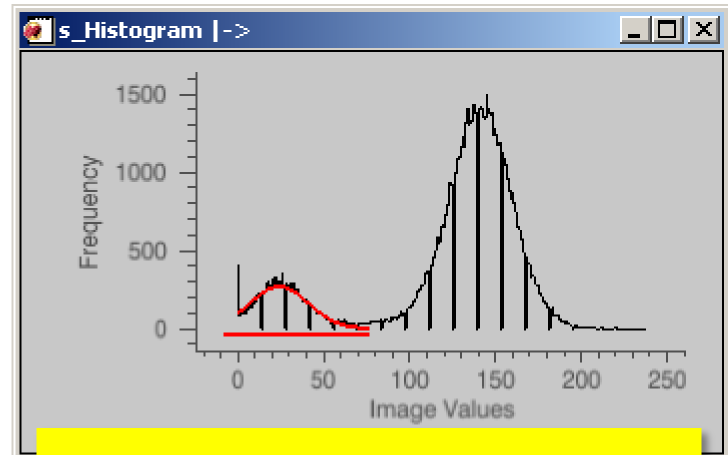
Morphological
- Morphological operators
- Size
- Thinning / skeletonization

Arithmetic-logic
- AND, OR, XOR

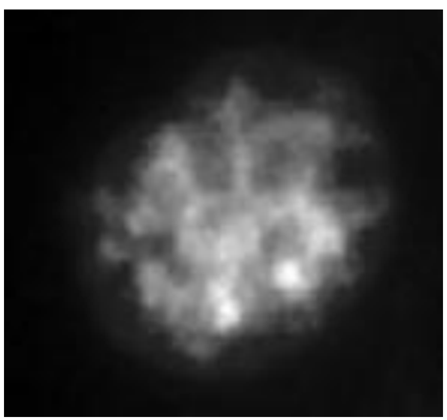
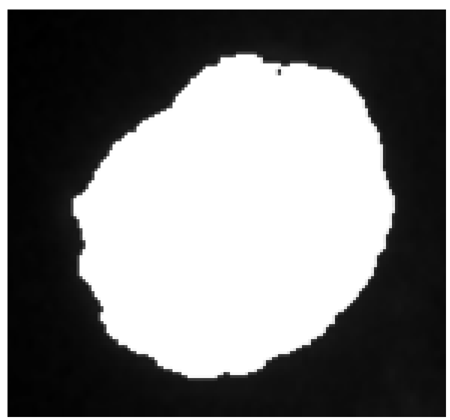
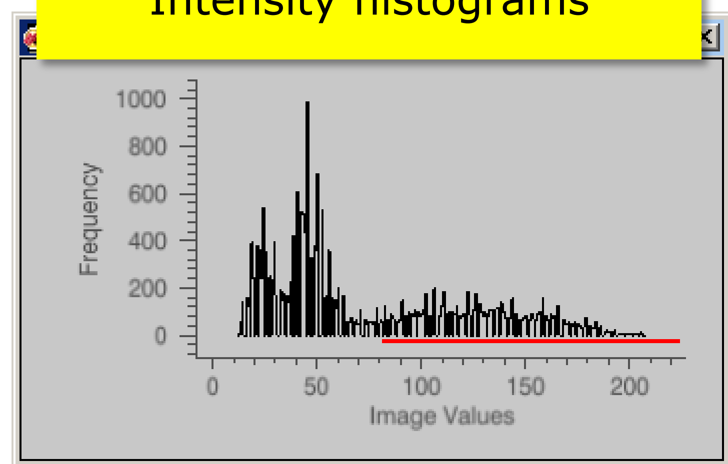
And a long etc.

Segmentation – basics

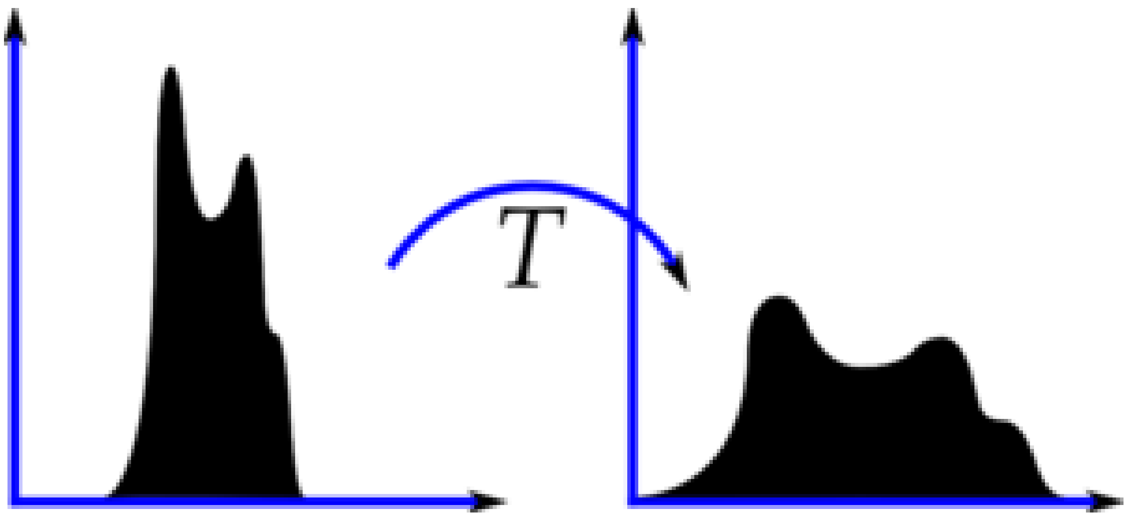
- Threshold fillter
  segmentation: ROIs (white) / background (black)

Intensity histograms

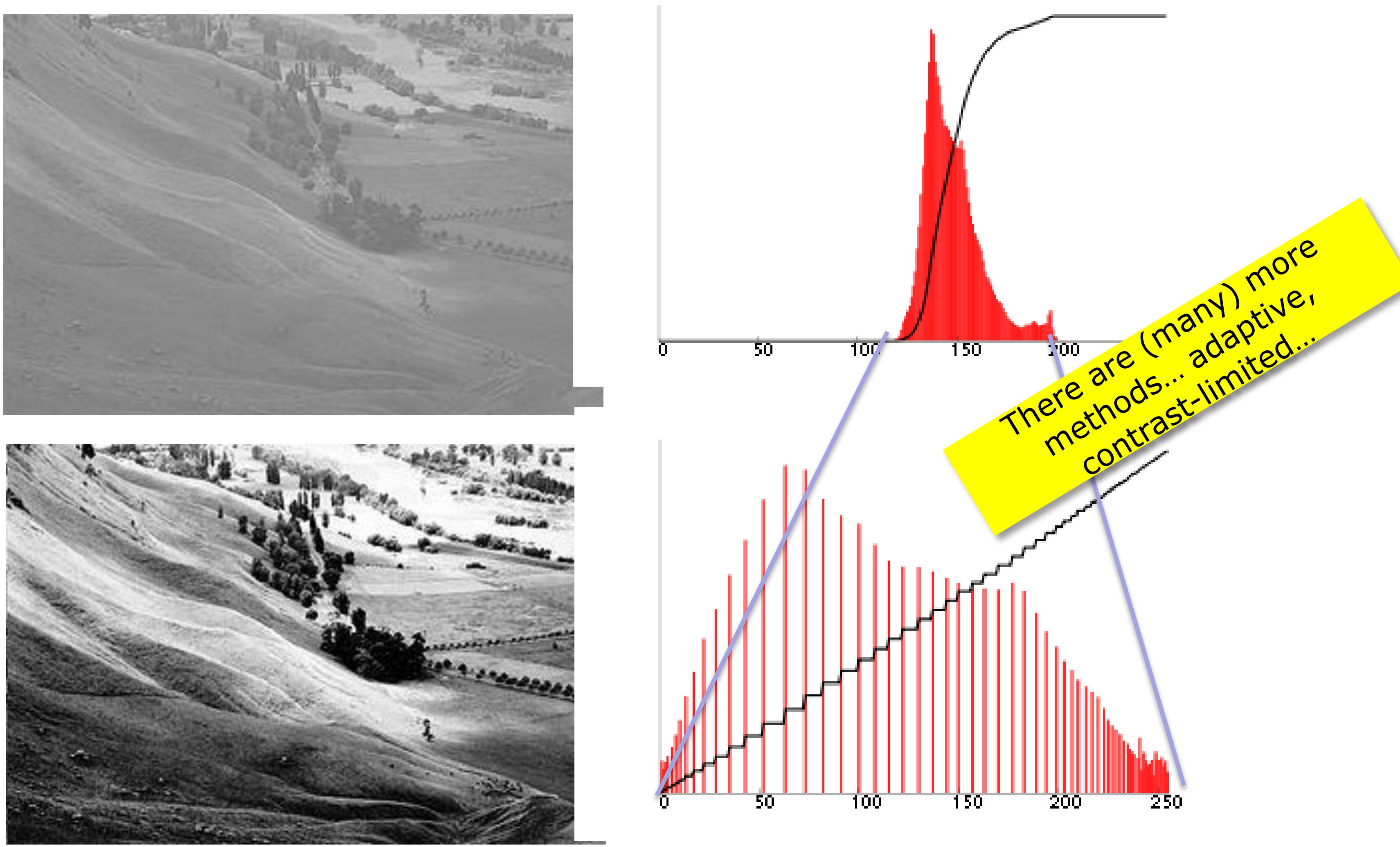
- Histogram equalization



http://en.wikipedia.org/wiki/Histogram_equalization

Segmentation – basics

- Histogram equalization



http://en.wikipedia.org/wiki/Histogram_equalization

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

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

$$t : \text{threshold}, \quad \omega_i : \text{probability of class } i$$

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

Algorithm

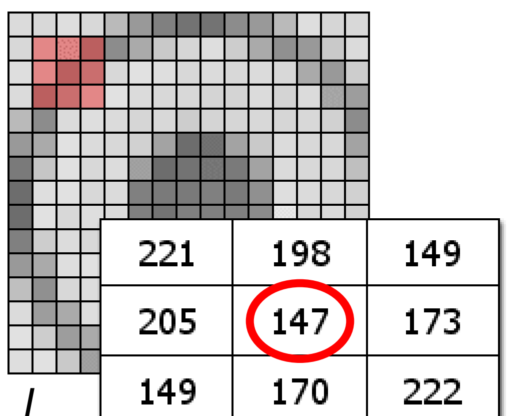
1. Compute histogram and probabilities of each intensity level
2. Set up initial $\omega_i(0)$ and $\mu_i(0)$
3. Step through all possible thresholds $t = 1 \ldots$ maximum intensity
   1. Update ω_i and μ_i
   2. Compute $\sigma_b^2(t)$
4. Desired threshold corresponds to the maximum $\sigma_b^2(t)$
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 = $\dfrac{threshold_1 + threshold_2}{2}$

N. Otsu (1979).

- Convolution
  - Lots of filters based on this principle
    http://en.wikipedia.org/wiki/Convolution
- Matrix convolution, in our case, is an operation between two matrices, namely…
  - the input image, *I*
  - a *kernel*, *K*

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

K



I

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

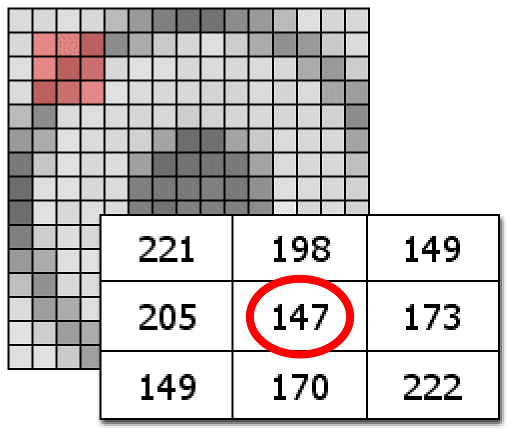
Adapted from James Matthews, 2002
http://www.generation5.org/content/2002/convolution.asp

Segmentation – basics

K

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

I



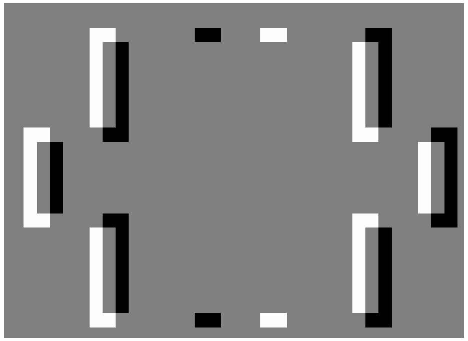
$$(K \otimes I)_{i,j} = (-1 * 222) + (0 * 170) + (1 * 149) + (-2 * 173) + (0 * \mathbf{147}) + (2 * 205) + (-1 * 149) + (0 * 198) + (1 * 221) = +63$$

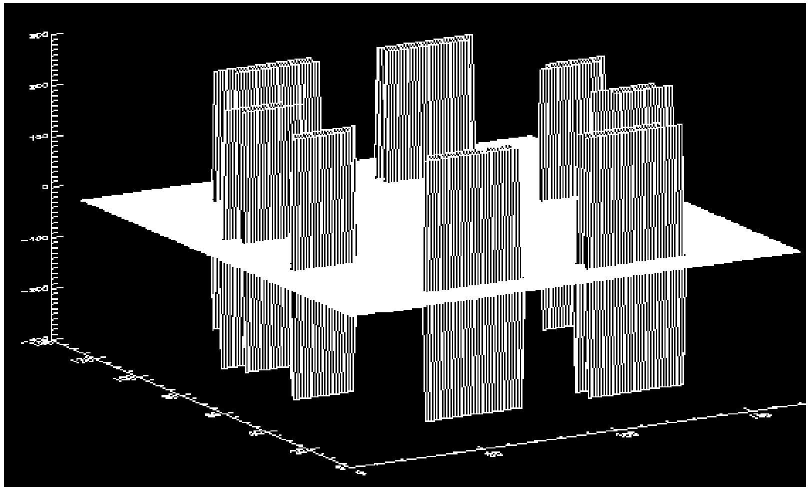
Matrix convolution can be implemented in different ways… beware of the algorithm!

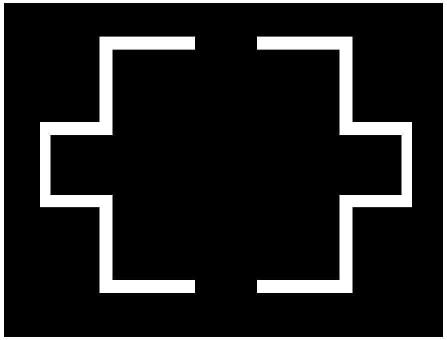
James Matthews, 2002
http://www.generation5.org/content/2002/convolution.asp

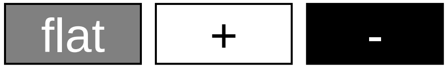
- Intensity gradients (discrete approximation)

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

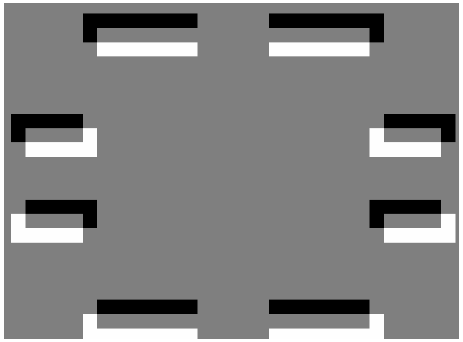


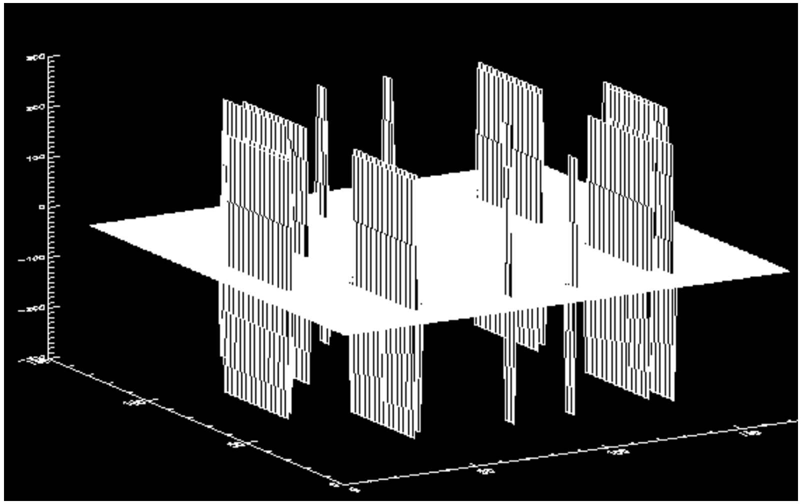


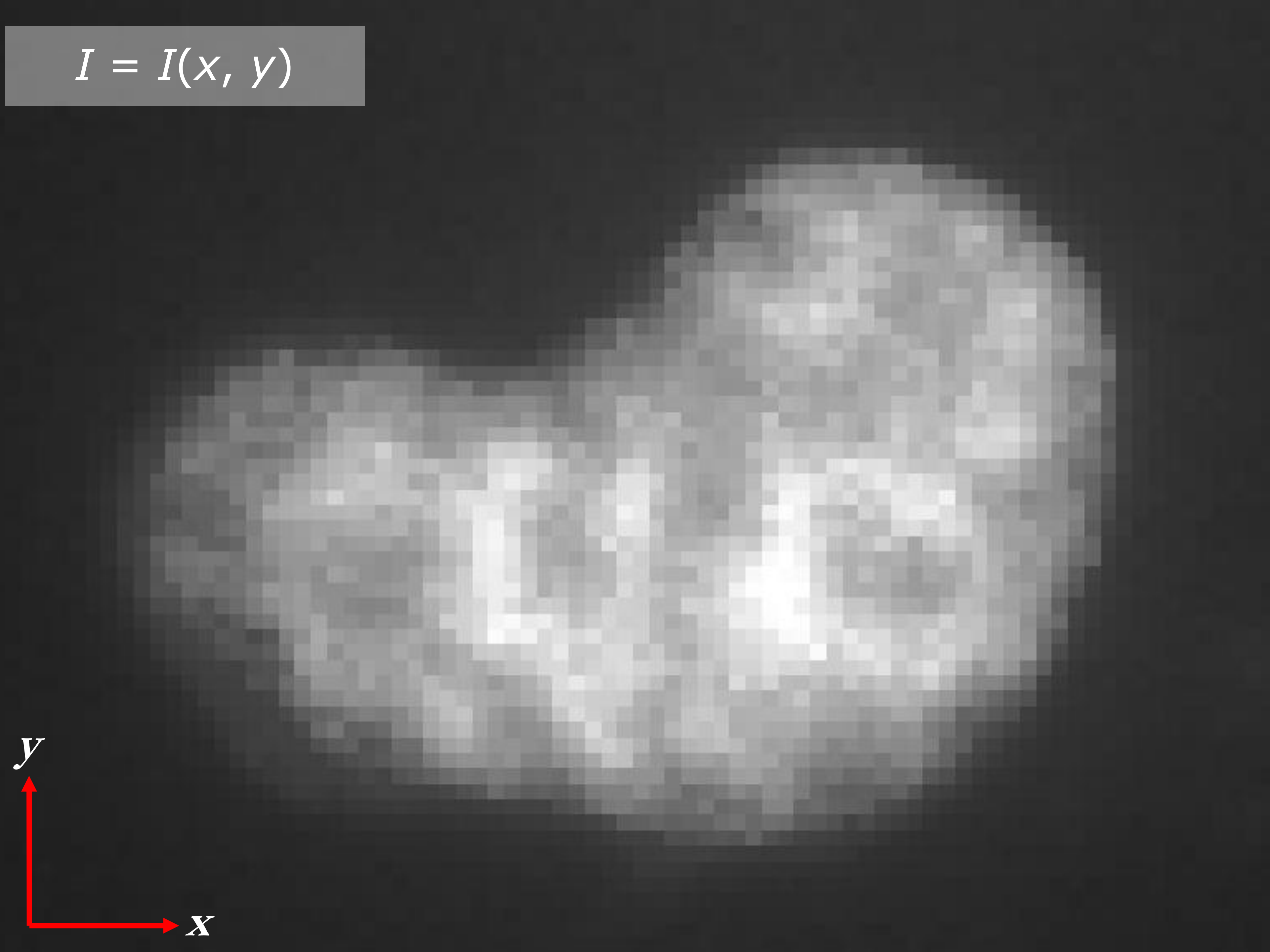




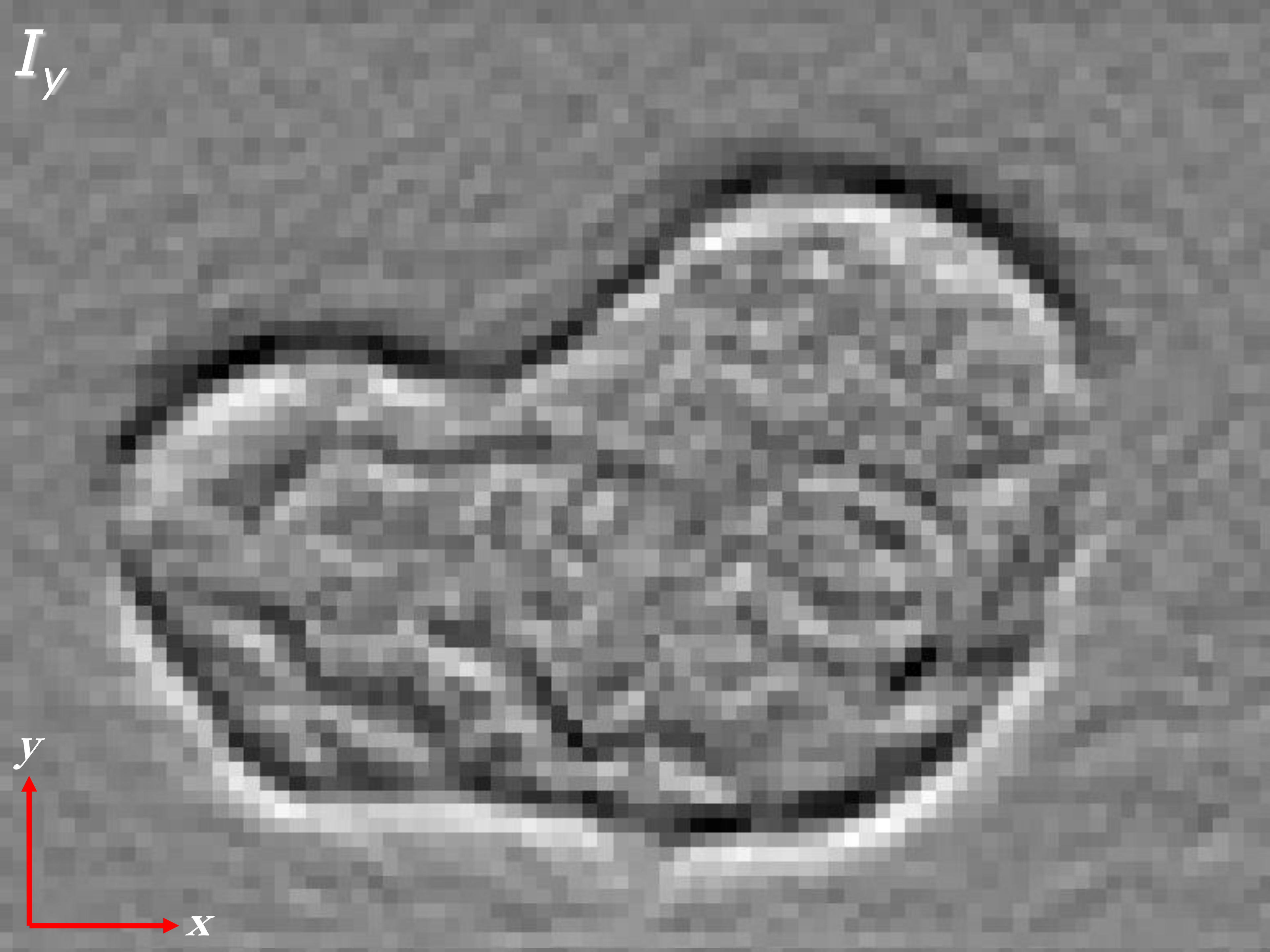
$$\frac{\partial I}{\partial y} \approx$$





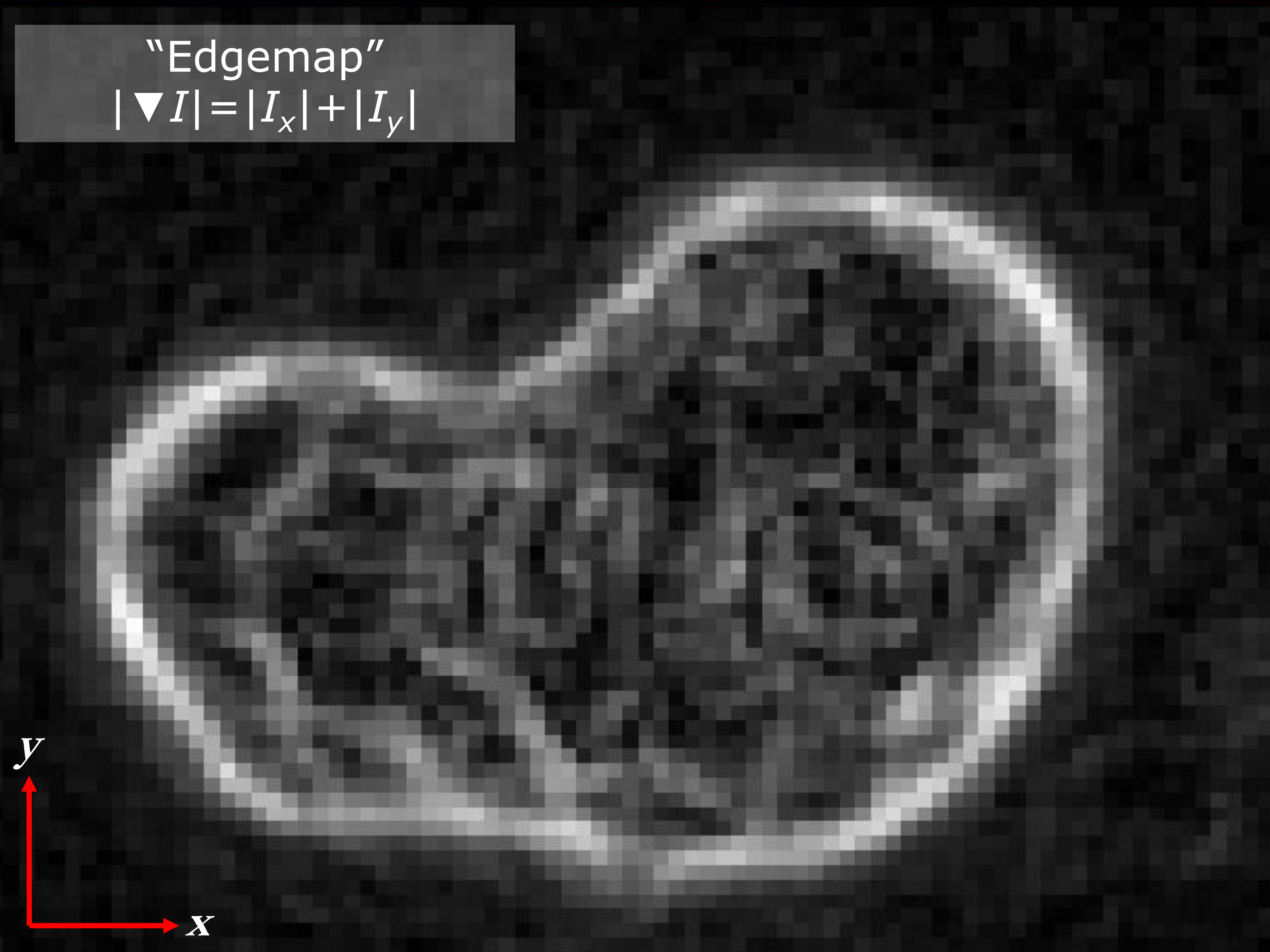


I = I(x, y)
y
x


I_y
y
x

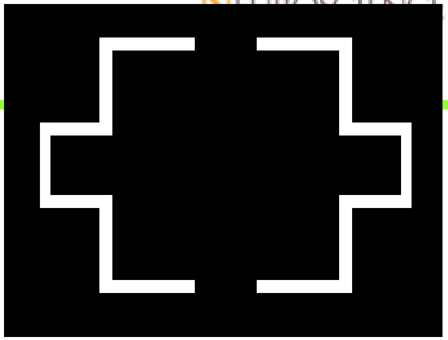
“Edgemap”

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



Segmentation – basics

- Intensity gradients (discrete approximation)

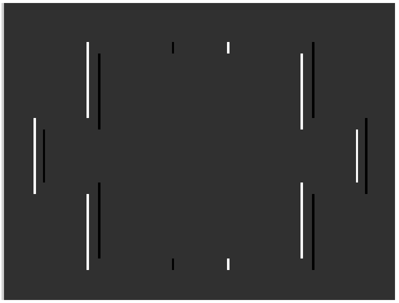


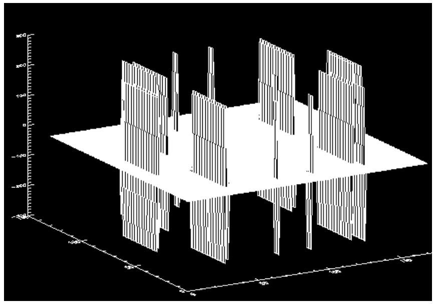
$$I = I(x, y)$$

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

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

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



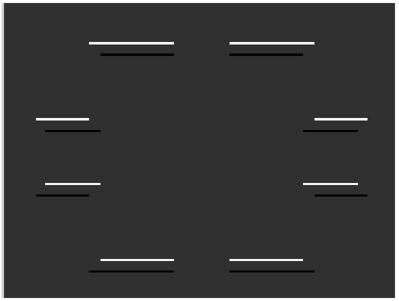


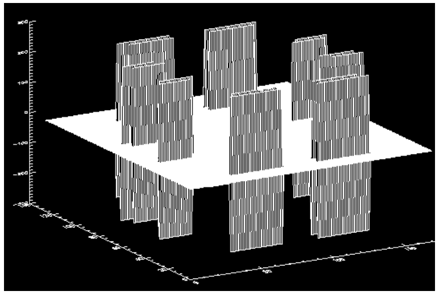
flat | + | -

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

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

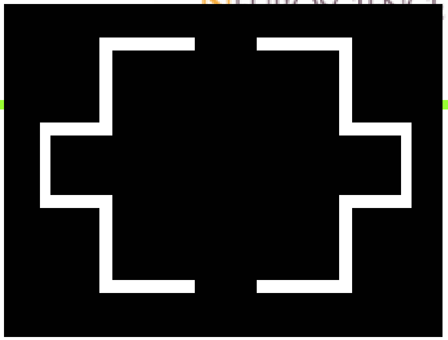
$$Ky = \begin{Bmatrix} 0 & 1 & 0 \\ 0 & -1 & 0 \\ 0 & 0 & 0 \end{Bmatrix}$$





Segmentation – basics

- Kernels…



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

Laplace

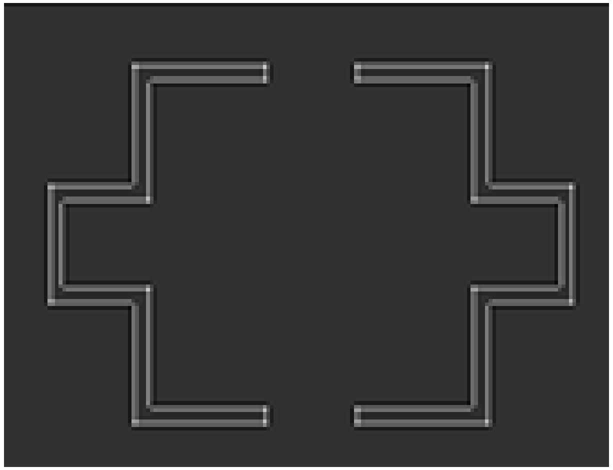
$$\nabla^2 I = \frac{\partial^2 I}{\partial x^2} + \frac{\partial^2 I}{\partial y^2}$$

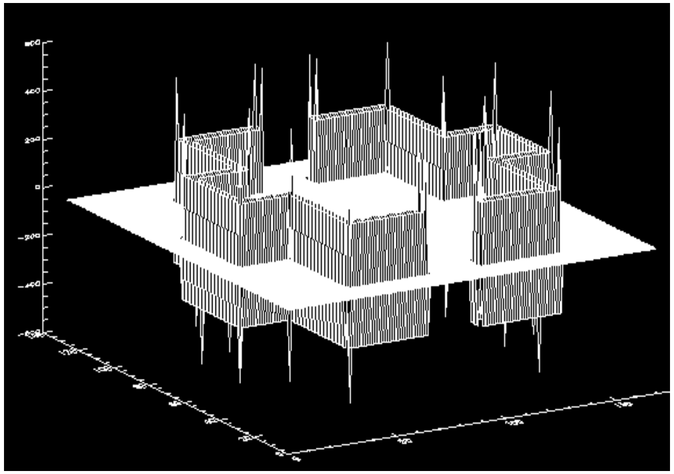
$$\nabla^2 I \approx \frac{f(x+\Delta x, y) - 2f(x,y) + f(x-\Delta x, y)}{(\Delta x)^2} + \frac{f(x, y+\Delta y) - 2f(x,y) + f(x, y-\Delta y)}{(\Delta y)^2}$$

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

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

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





Segmentation – basics

- Edgemaps

An ***edgemap*** filter takes intensity changes as ROI boundaries or “edges”

$$f = \sqrt{(Kx \otimes I)^2 + (Ky \otimes I)^2}$$

Example:Sobel filter
(notice the thick ROI edges)

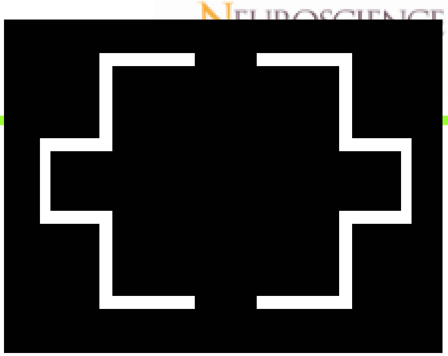
$$\begin{Bmatrix} -1 & 0 & 1 \\ -2 & 0 & 2 \\ -1 & 0 & 1 \end{Bmatrix} \qquad \begin{Bmatrix} 1 & 2 & 1 \\ 0 & 0 & 0 \\ -1 & -2 & -1 \end{Bmatrix}$$

$$Sx \qquad\qquad Sy$$

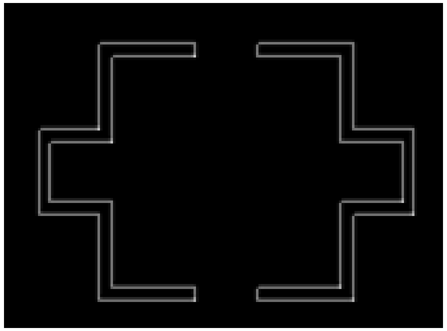
$Sx \otimes I$?

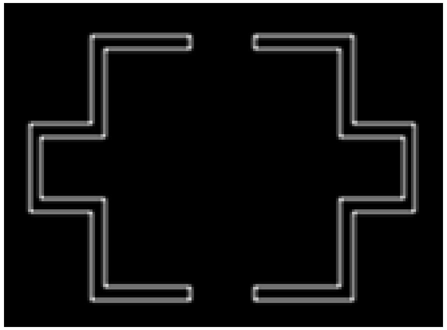
$Sy \otimes I$?

$$f_{Sobel} = \sqrt{(Sx \otimes I)^2 + (Sy \otimes I)^2}$$



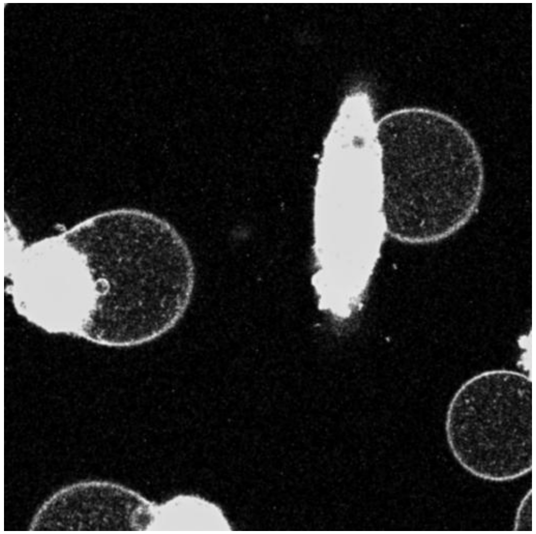
$I = I(x, y)$



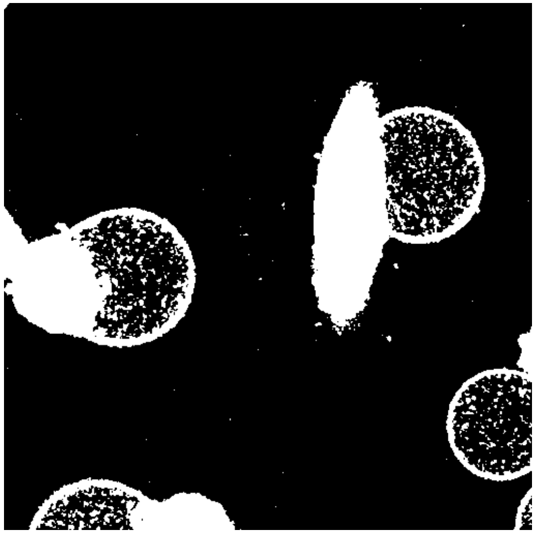


Segmentation – basics

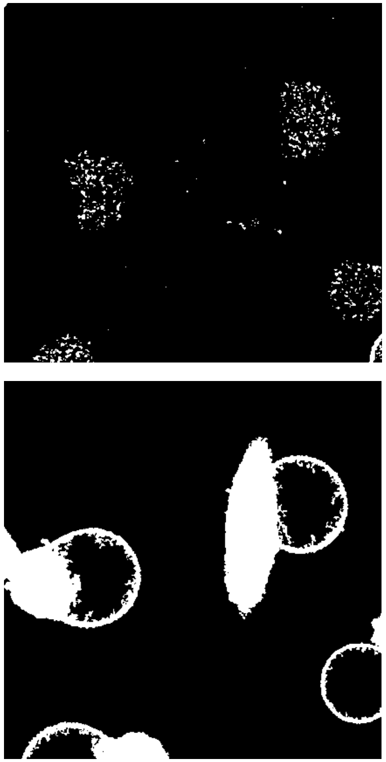
- Morphology based filters
  - Example: size selection



Input greyscale image



After thresholding…



Size selection

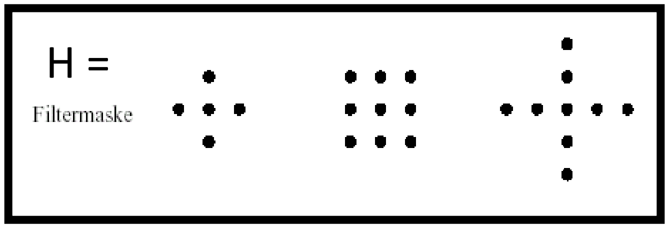
How to define a size-select algorithm?

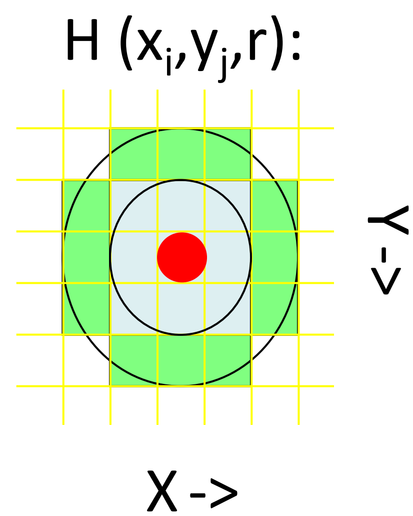
Segmentation – basics

Filtering with morphological operators:

- Structuring element, template or mask H ...

- Additional rules...





Polynomial filters $y(m,n) = \bar{h}_1[x(m,n)] + \bar{h}_2[x(m,n)],$

$$\bar{h}_1[x(m,n)] = \sum_{\substack{p=0 \\ (p,q)\neq(0,0)}}^{P-1} \sum_{q=0}^{Q-1} a(p,q) \cdot x(m-p, n-q)$$

$$\bar{h}_2[x(m,n)] = \sum_{\substack{p=0 \\ (p,q)\neq(0,0)}}^{P-1} \sum_{q=0}^{Q-1} \sum_{\substack{k=0 \\ (k,l)\neq(0,0)}}^{P-1} \sum_{l=0}^{Q-1} b(p,q,k,l) \cdot x(m-p, n-q) \cdot x(m-k, n-l)$$

- Mathematical morphology

Minkowski Operations

Addition... dilation: $A \oplus S = \{(m,n) | [S + (m,n)] \cap A \neq 0\}.$

Substraction ... erosion: $A \ominus S = \{(m,n) | [S + (m,n)] \subseteq A \neq 0\}$

...opening : $A \circ S = (A \ominus S) \oplus S,$

...closing : $A \bullet S = (A \oplus S) \ominus S,$

- A: image
- S: structuring element

Segmentation – basics

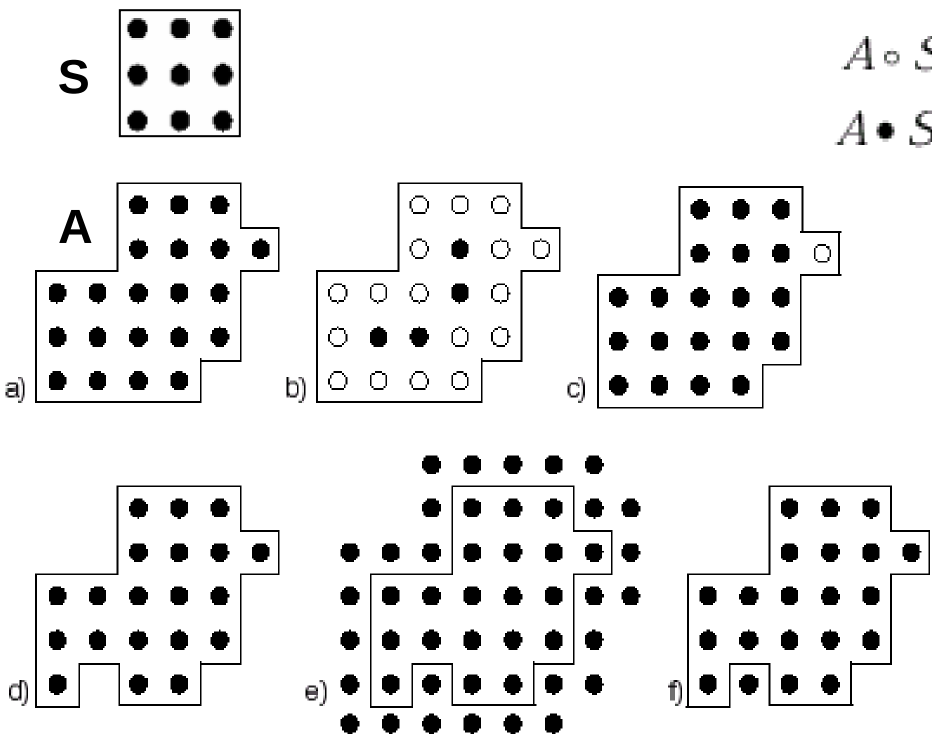
- Mathematical morphology

$$A \oplus S = \{(m,n) \| [S+(m,n)] \cap A \neq 0\}.$$

$$A \ominus S = \{(m,n) \| [S+(m,n)] \subseteq A \neq 0\}$$

$$A \circ S = (A \ominus S) \oplus S,$$

$$A \bullet S = (A \oplus S) \ominus S,$$



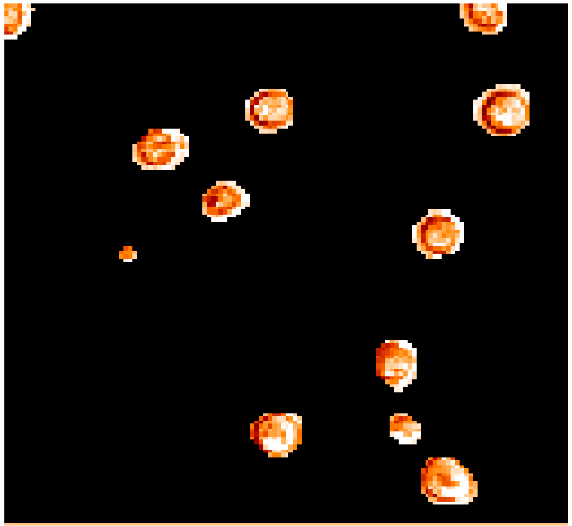
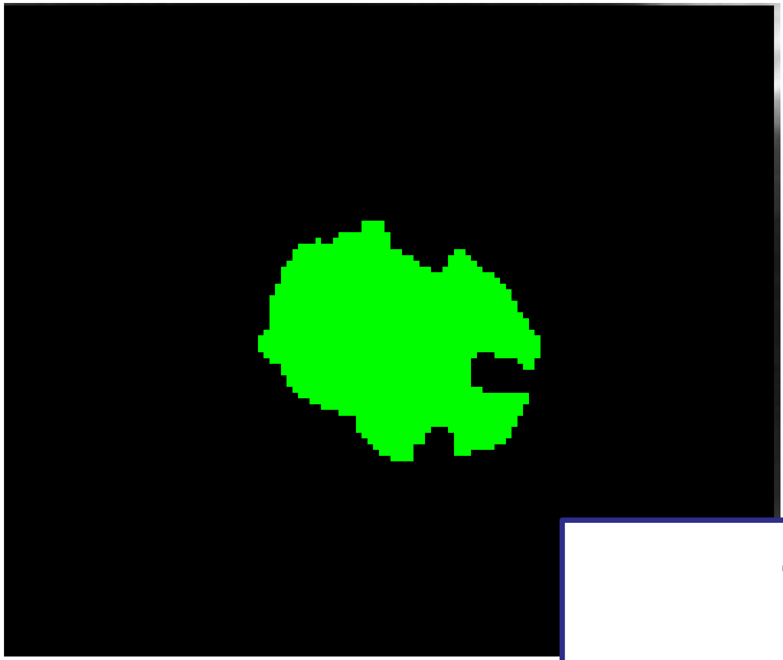
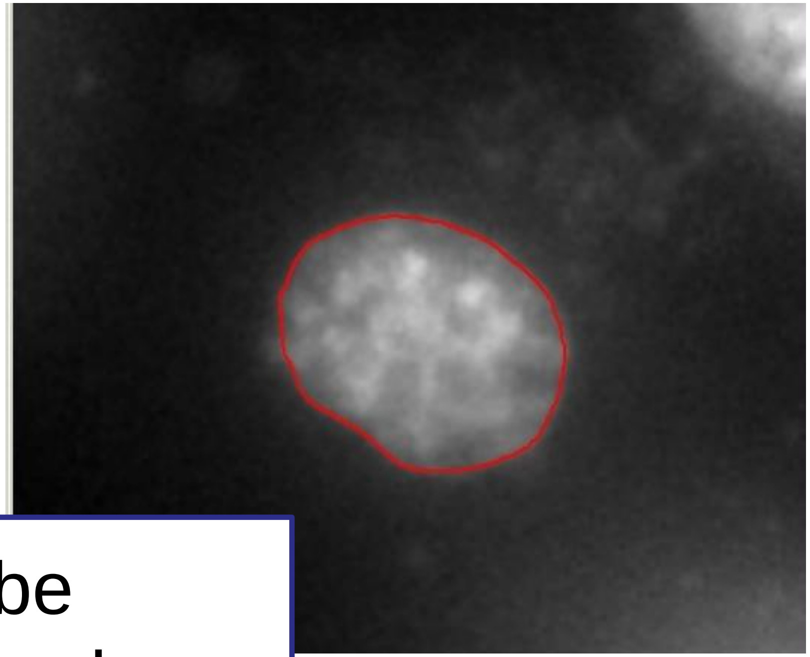


Abb. 2.5. **a** Originalform **b** erodiert **c** opening (Dilatation von b)
d Originalform **e** dilatiert **f** closing (Erosion von e)

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





To be continued…

References

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  MIT press, 1982

- John Russ. The image processing handbook, 4^{th} ed.
  CRC Press, 2002

- Nixon, Aguado. Feature extraction & image processing, 1^{st}|2^{nd}|3^{rd} ed.
  Academic Press, 2002|2008|2012

- Aubert & Kornprobst. Mathematical Problems In Image Processing
  Springer, 2006

- From SCIAN-Lab (see publications section on the website)
  - Jara, 2006. 2D/3D active contours
  - Olmos, 2009. 2D topology adaptive active contours (t-snakes)

References

Some free /open source software tools

- Java based (Java runtime requried)
  - ImageJ (http://rsbweb.nih.gov/ij/, public domain)
  - Fiji (http://fiji.sc; GPL license)
  - Icy (http://icy.bioimageanalysis.org; GPLv3 license)

- Others
  - CellProfiler (http://cellprofiler.org; GPL, BSD licenses)
  - Slicer (www.slicer.org; BSD license)
  - ilastik (https://ilastik.org; GPL license)
  - IPOL (Image Processing Online): open access electronic journal with peer reviewed articles + code (in C language) + examples (www.ipol.im; BSD / GPL / LGPL licenses or similar)