

Particle Analyzer: an hybrid system for **microscopic granulometry**

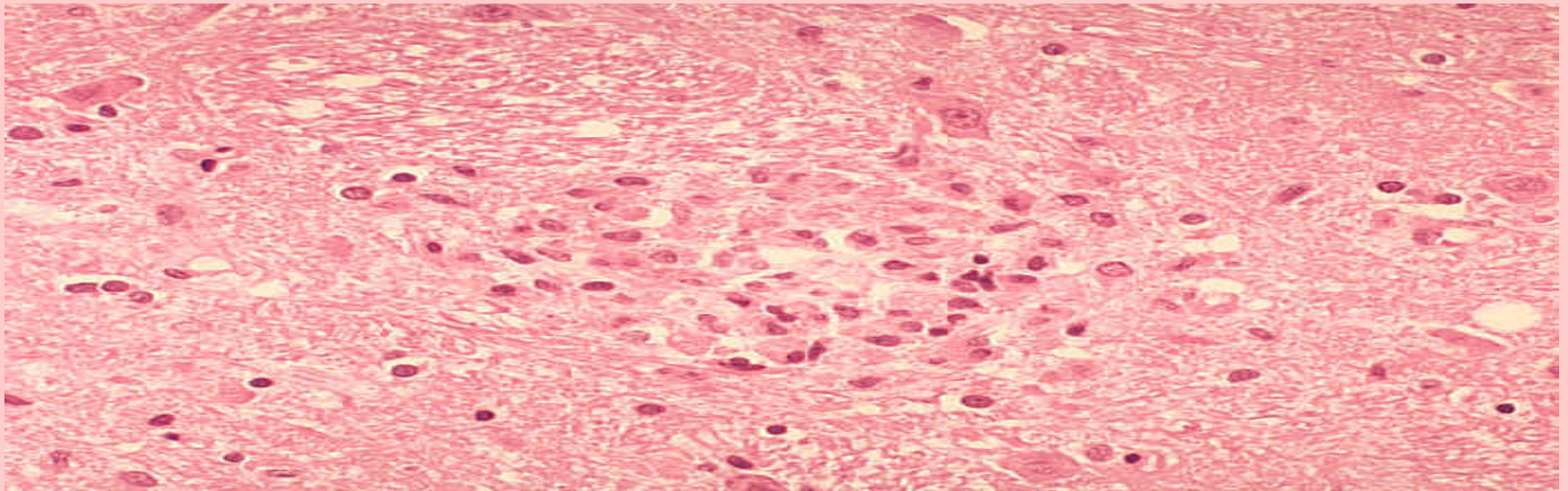
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Melo

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Importance:

- **HIV** - causative agent of **AIDS**;
- Slower progression of immune deficiency with use of **Highly Active Anti-Retroviral Therapy (HAART)**;
- To **monitoring - count** and a **viral load** laboratory test are **periodically recommended**;
- Generic approaches to **identify and count** cells of interest



Synopsis:

- **particles size distribution and identification** = granulometrie
- morphological granularity

adapted to:

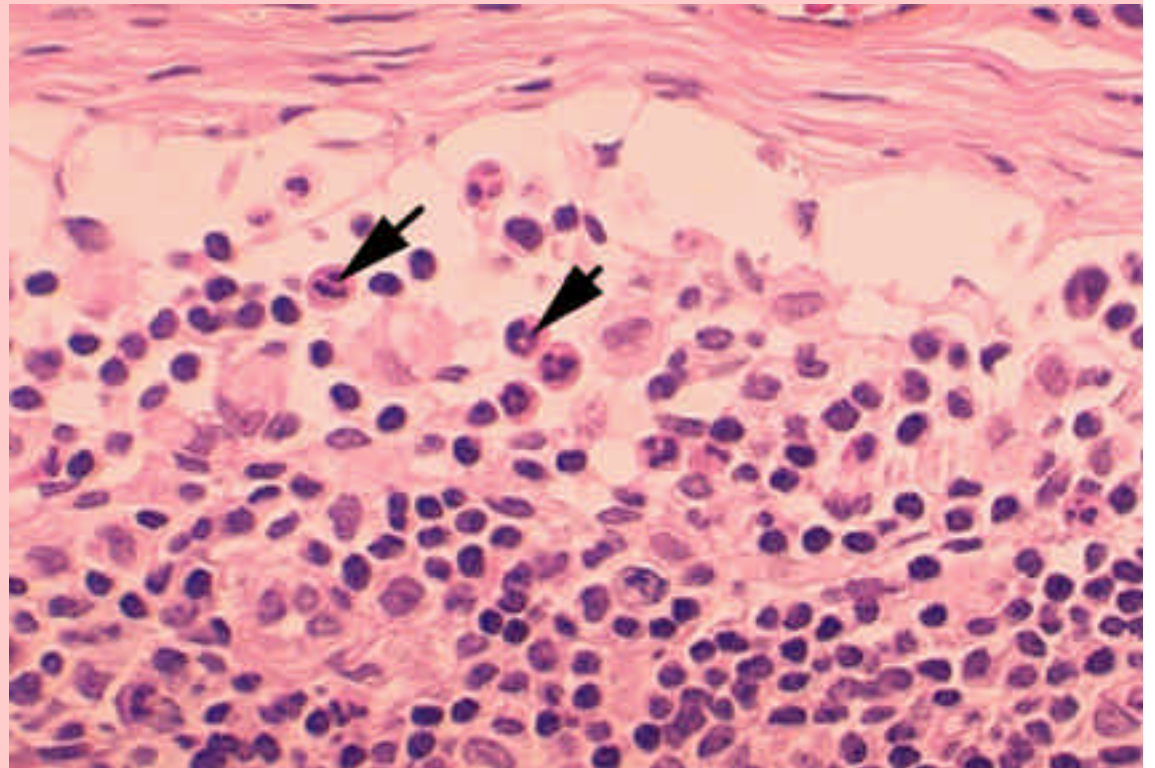
extract

blood and

HIV cells and

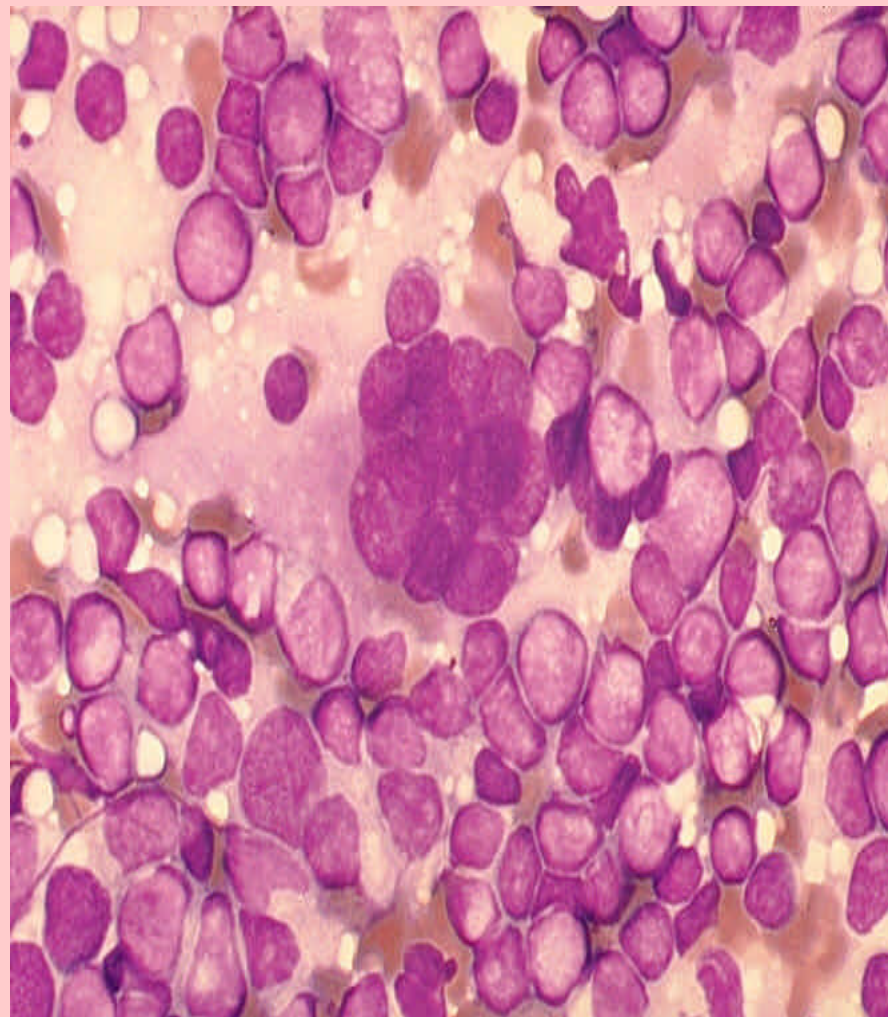
from microscopic

images



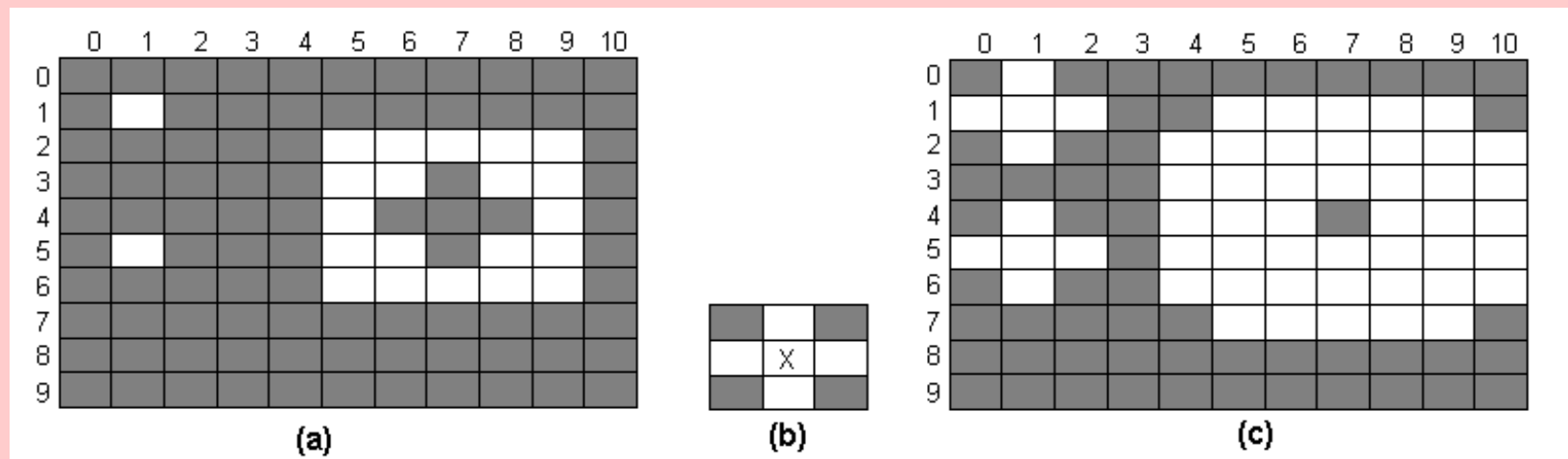
Counting Cells by Mathematical Morphology

- **Main idea:** image simplification by removing not relevant details **preserving** essential characteristics of **form**
- Basic operations : **dilation, erosion, opening and closing**



The **dilation** on binary sets
are defined by:

$$A \oplus B = \{ c \mid c = a + b, \text{ for some } a \in A, b \in B \}$$

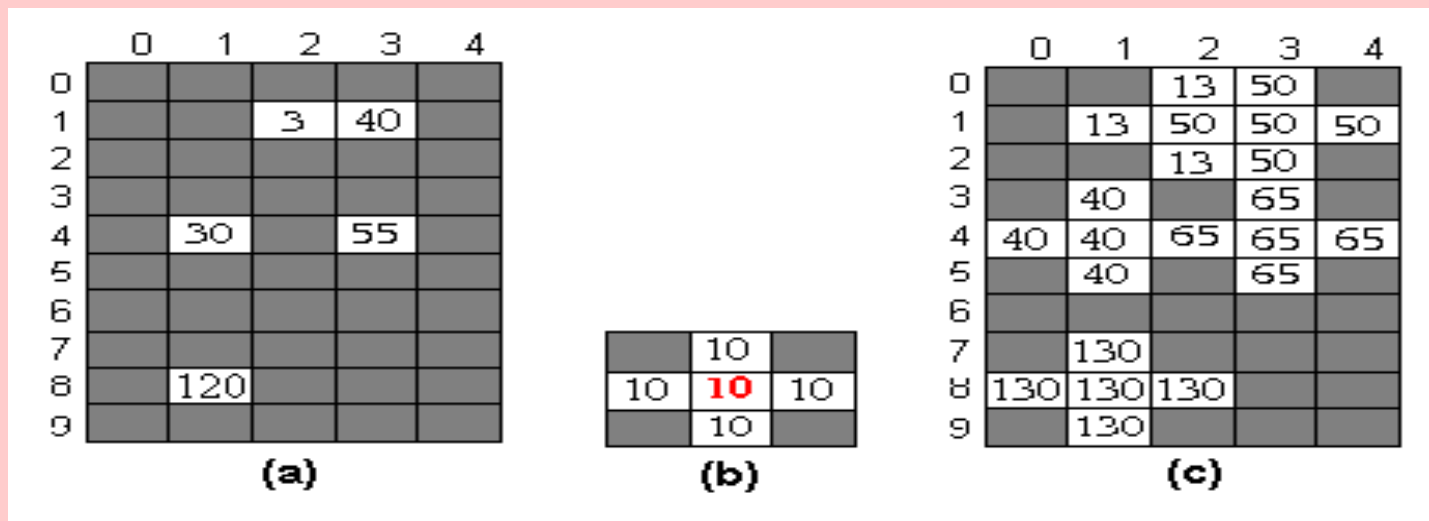


- it gives an "expansion" or "grow" on the image

Dilation applied to gray value image

- Let F, K be the domains of functions: $f(x, y)$ and $k(x, y)$ where $x, y \in \mathbb{Z}$, then **Dilation** can be defined as:

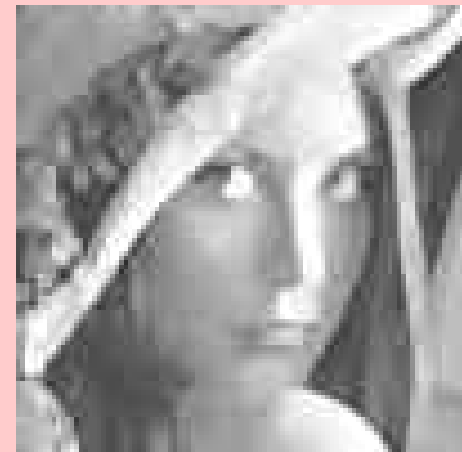
$$f \oplus k = (f \oplus k)(x) = \max \{ z \in K, x-z \in F \mid f(x-z) + k(z) \}$$



- f is normally the image, while k is named **structuring element**.
- on color image, each **RGB**, **HSV** or **YIQ** channel must be considered

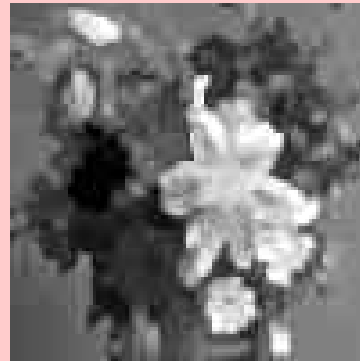
Dilation results: bright

- On the left is the original image on the center and right the result after Dilation with a discrete $3 \times 3\{1\}$ and $3 \times 3\{50\}$ element .

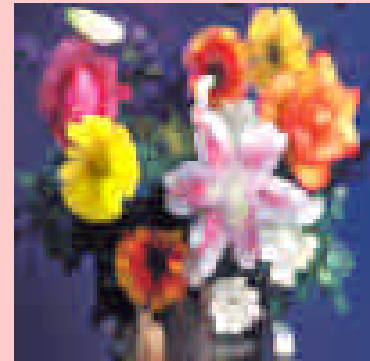


Dilation to color images

original



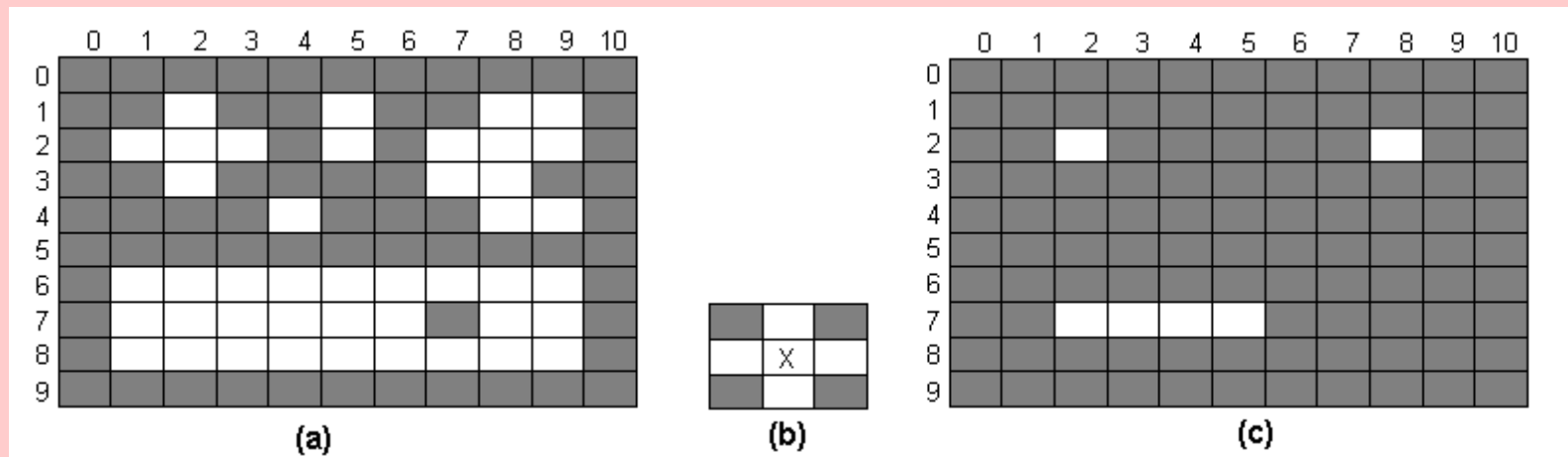
$3 \times 3 \{1\}$ for each channel: *red, green e blue*



result

Erosion on binary sets are defined by:

- several equivalent definitions which can be given for erosion on binary sets but all resulting:



in "shrinking" of the original image

Erosion on gray value image

- Let F , K be the domains of functions: $f(x, y)$ and $k(x, y)$

where $x, y \in \mathbb{Z}$, then Erosion can be defined as:

$$f \ominus k = \min \{ z \in K, x+z \in F \mid f(x+z) - k(z) \}$$

	0	1	2	3	4
0				66	
1			3	40	7
2				150	
3					
4		30		55	
5					
6					
7		70			
8	95	120	12		
9		40			

(a)

	10	
10	10	10
	10	

(b)

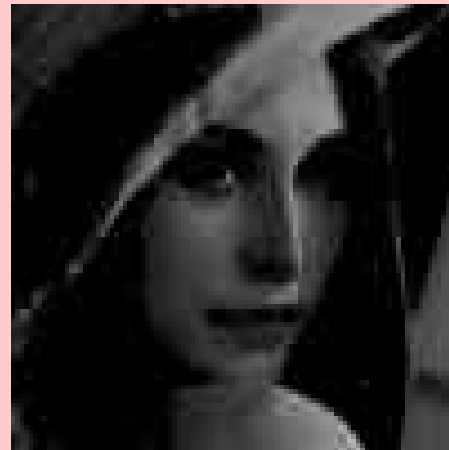
	0	1	2	3	4
0					
1					
2					
3					
4					
5					
6					
7					
8		2			
9					

(c)

- f is normally the image, while k is often the **structuring element**.
- on color image, each **RGB**, **HSV** or **YIQ** channel must be considered

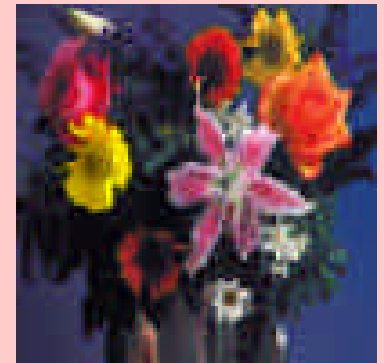
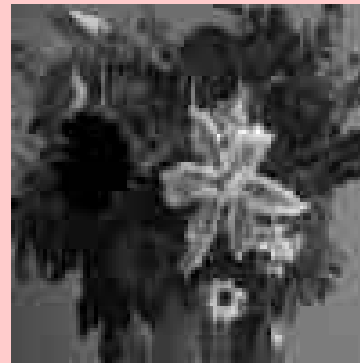
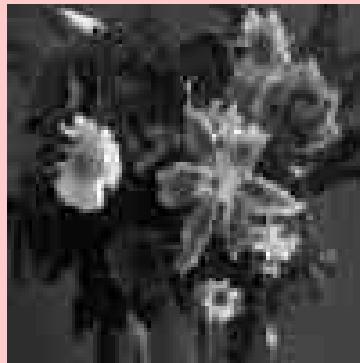
Erosion results: a dark image

- on the left result after erosion with a discrete $3 \times 3\{1\}$ element .
- on the right the result after erosion with a discrete $3 \times 3\{50\}$ element .

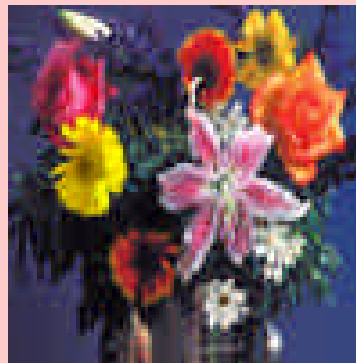


Erosion results to color images

- $3 \times 3 \{1\}$ for each channel: *red, green e blue*



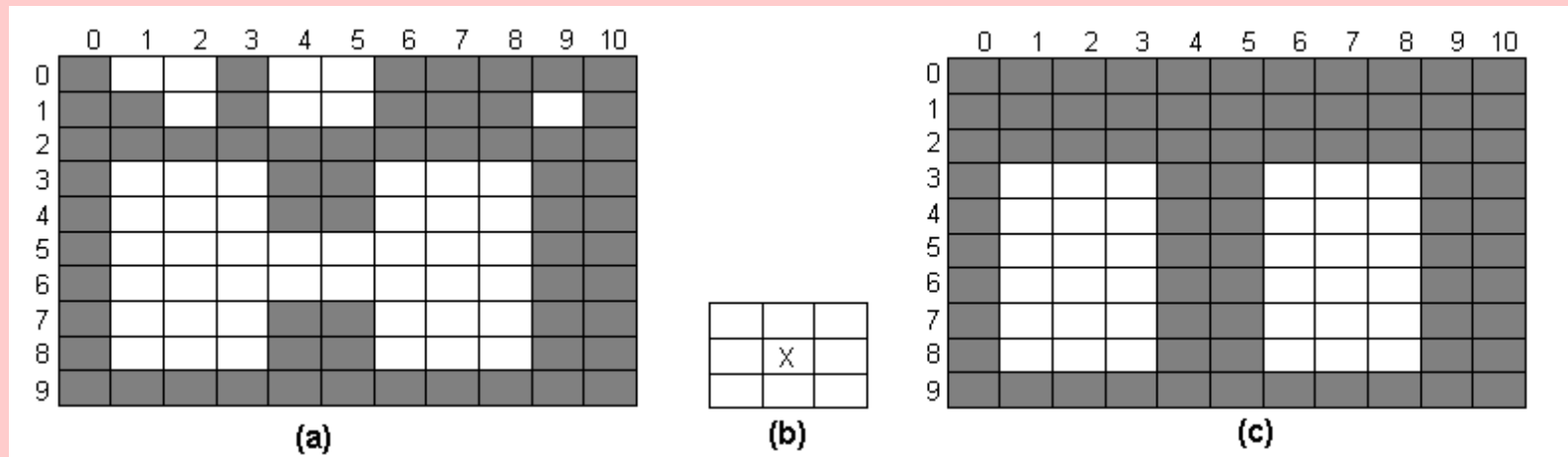
result



original

Opening

- This operation is a combination of an **erosion** and then an **dilation** :

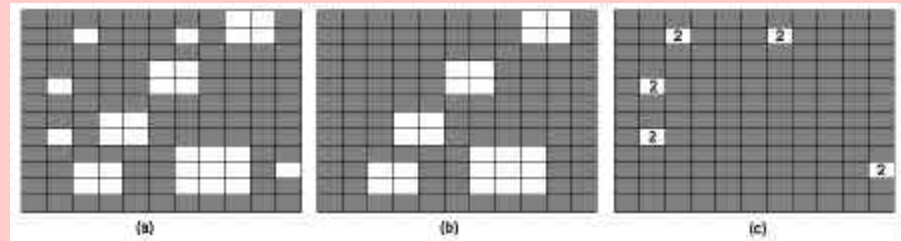


- Thus opening :
 - breaks thin connections within an object
 - eliminates small islands and sharp protrusions

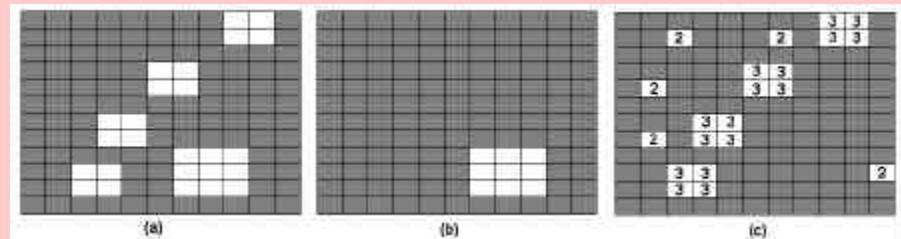
Morphological Granulometrie:

- by a set of Opening using $I B$

(series of virtual **sieve** with meshed defined by the **structuring element** size)



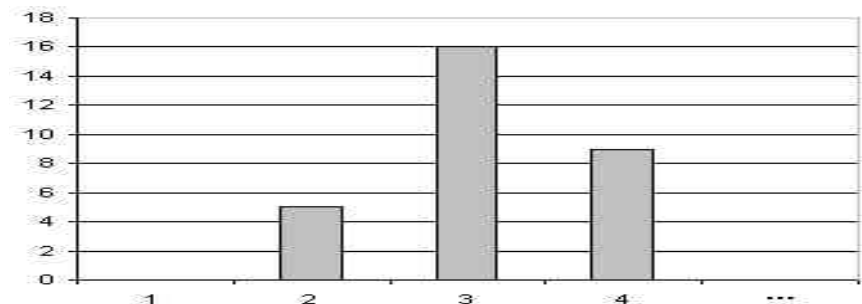
1st step: (a) original image, (b) open by a square **2x2**, (c) result



2nd step: (a) Image of first step (b) open by a square **3x3**, (c) result

etc. to size distribution:

Number
of
pixels
erased
in
the
image



structuring element size

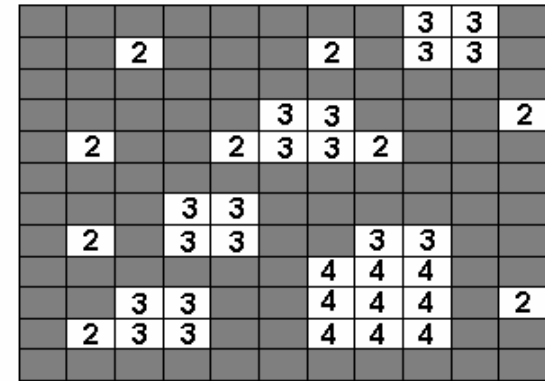
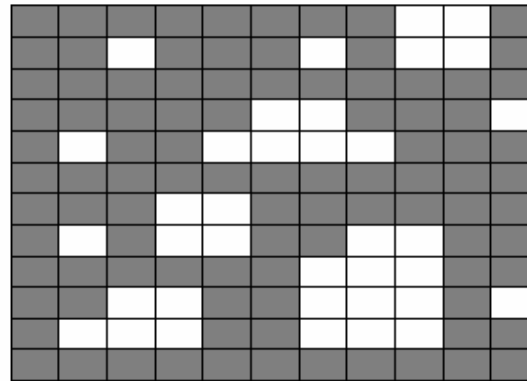
Morphological Reconstruction

- inserts the **concept of connectivity** in images;
- reconstruction can be built by an **infinite sequence of dilation and intersection**, until the result reaches a stability
- **with** Reconstruction :
 - only **complete cells eliminated** by opening using $I \ B$
 - black **nucleus inside** cells can be identify
 - only **complete cells** in the frame are counted

Example: **pixels** sieving

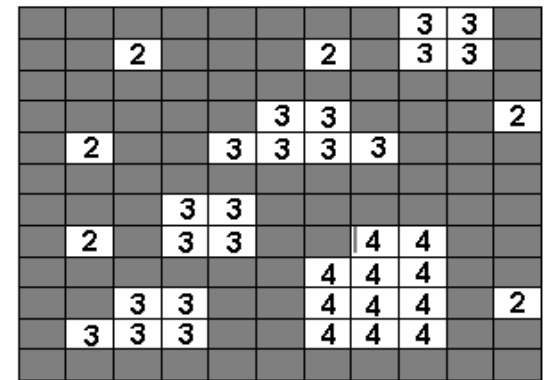
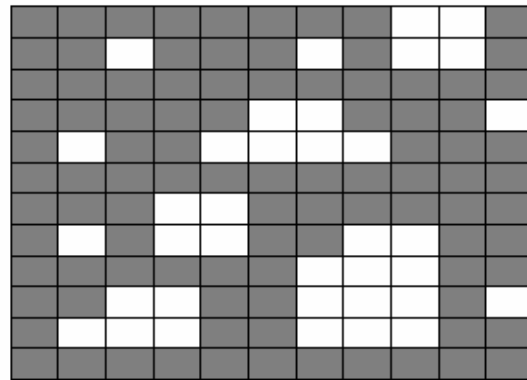
without
reconstruction:

partial cells
eliminated by a
square element B



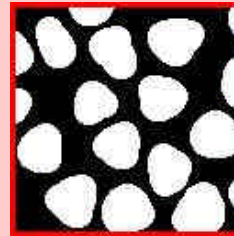
with reconstruction:

only **complete** cells
eliminated by
opening using $I \ B$



Detection **cells** on the **image border** that is not complete because they touch the acquired limits

- the image frame, shown in red.

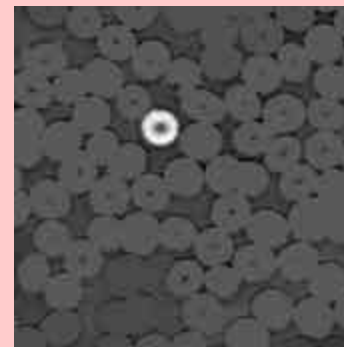
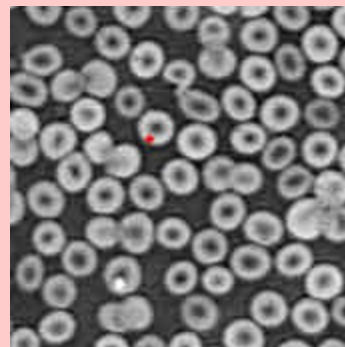


- **Reconstruction detects all the pixels that are connected to the borders.** The reconstruction of the image detects **all the blobs that touch the image frame.** It is possible then, to **subtract** this from the original to get only complete blobs.



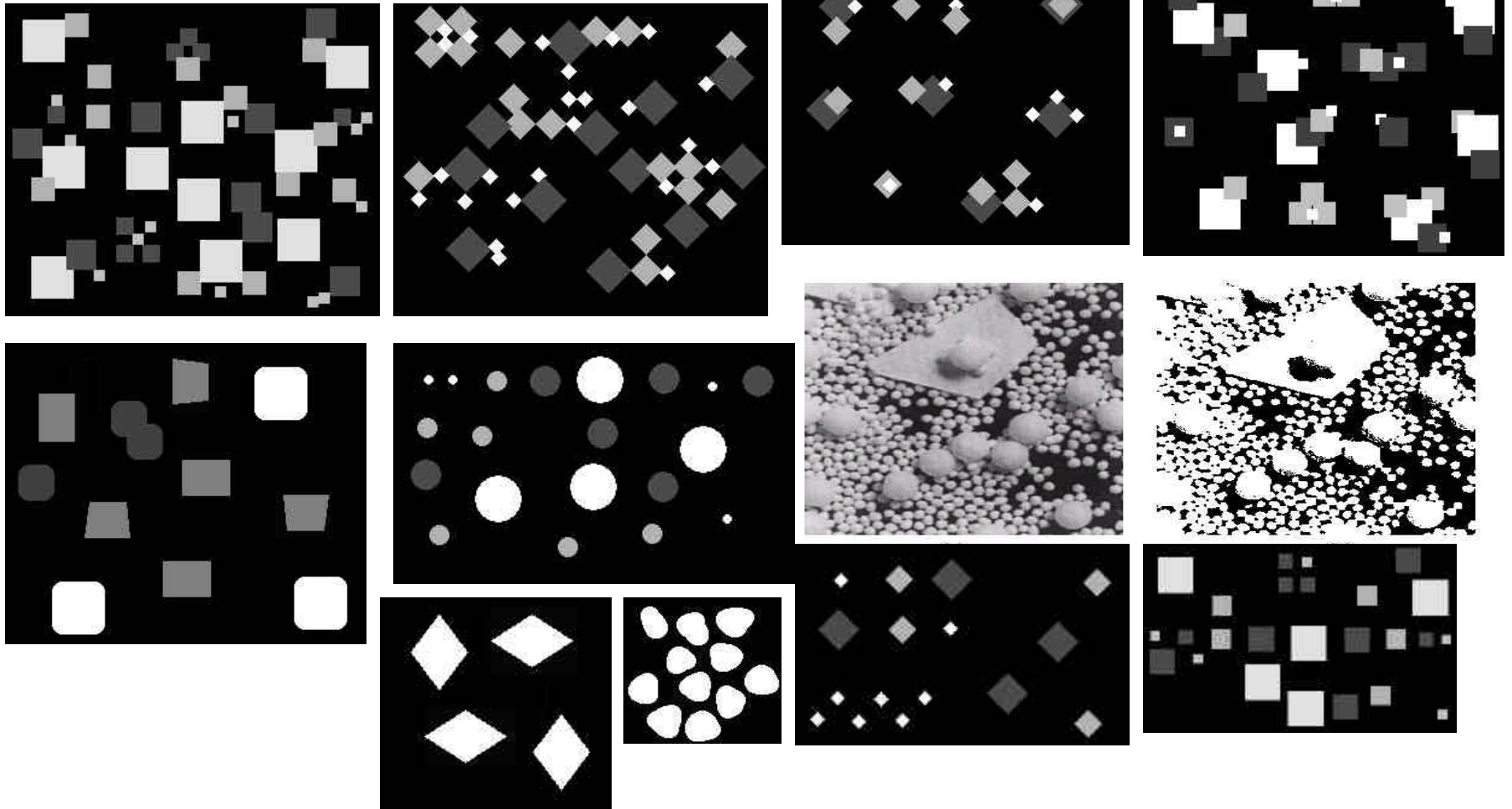
Detecting a single Black Nucleus cell

- A marker is placed over **one nucleus** in the negated cell image. The result of the **reconstruction** enhances **this black blood** cell because it is surrounded by dark pixels.



System result of granulometrie using morphologic techniques

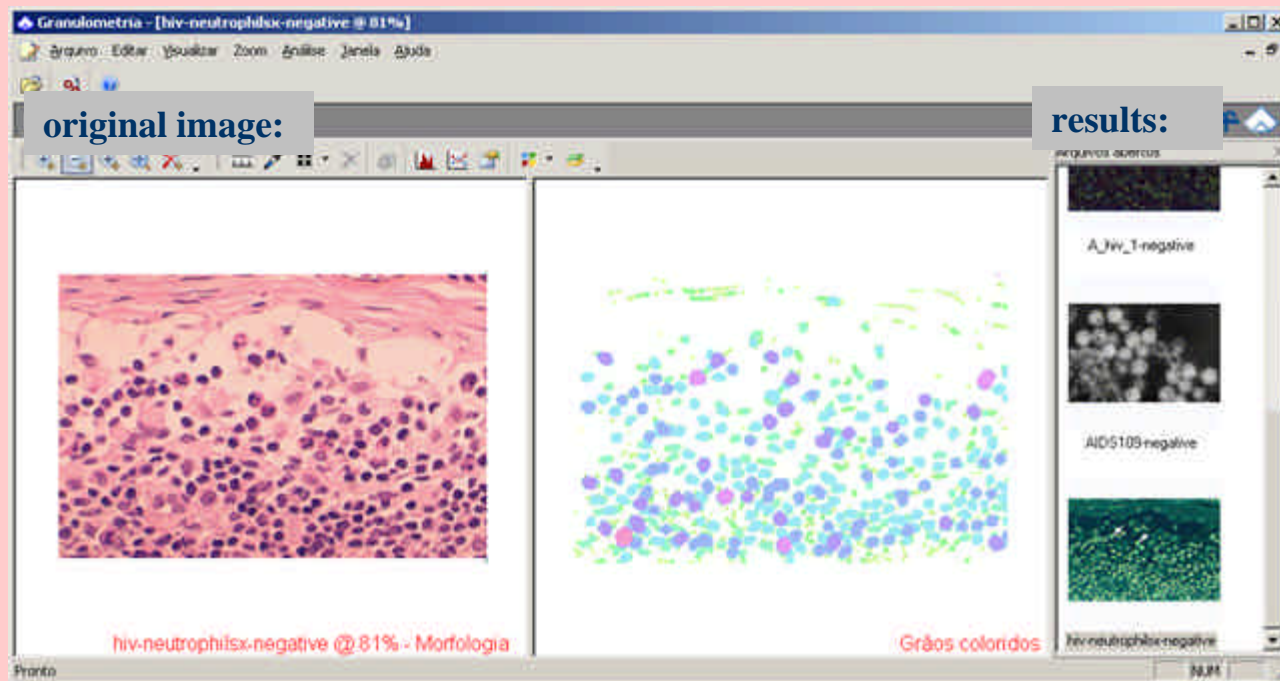
Some synthetic images or known results



Real result

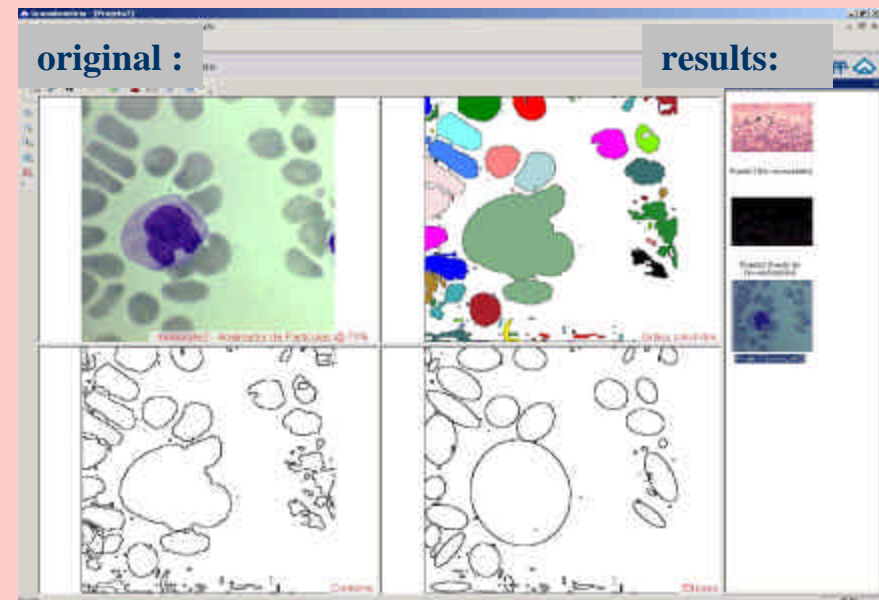
- identify count the HIV and blood cells

(different color => different number of nuclei)

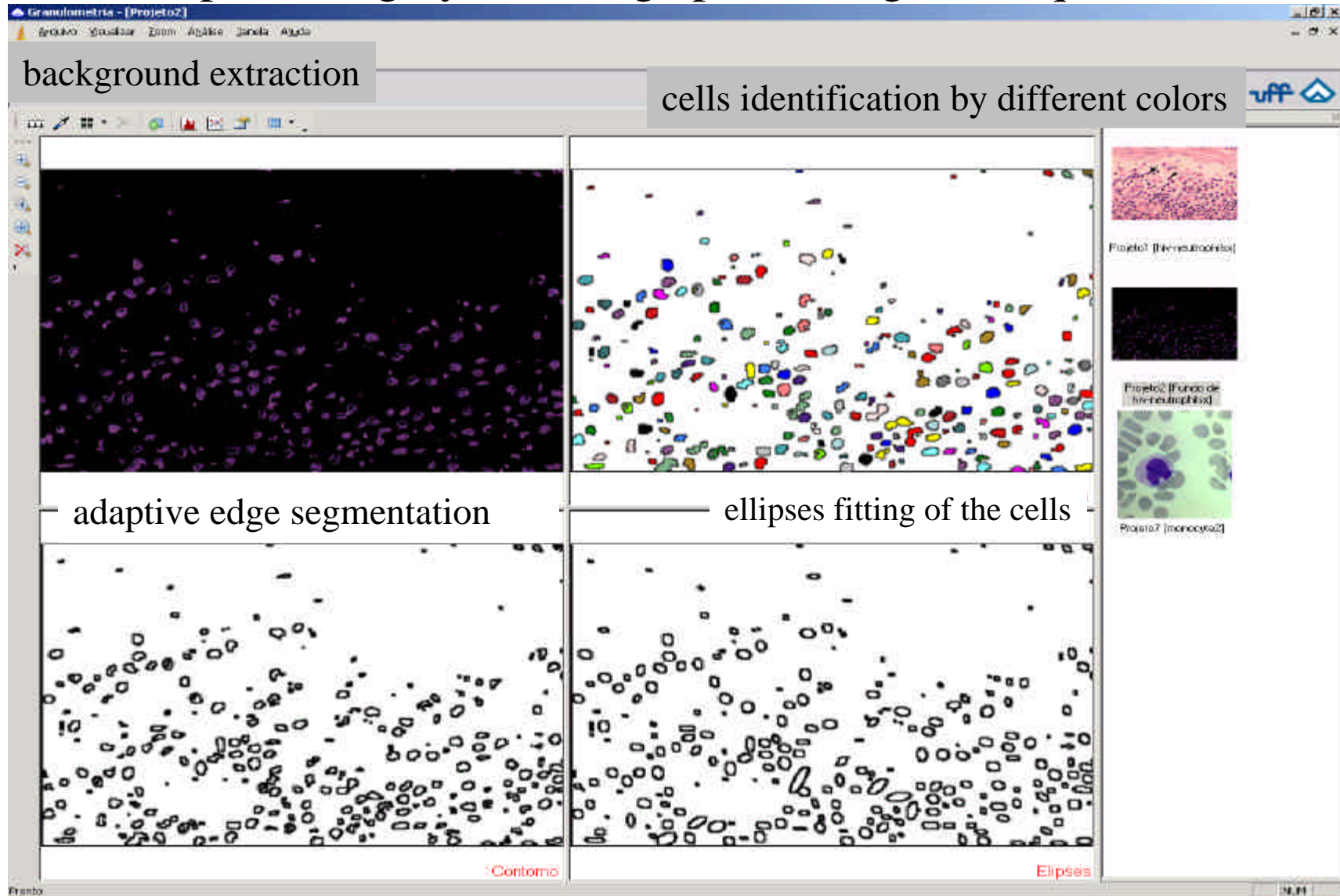


Hybrid image processing techniques:

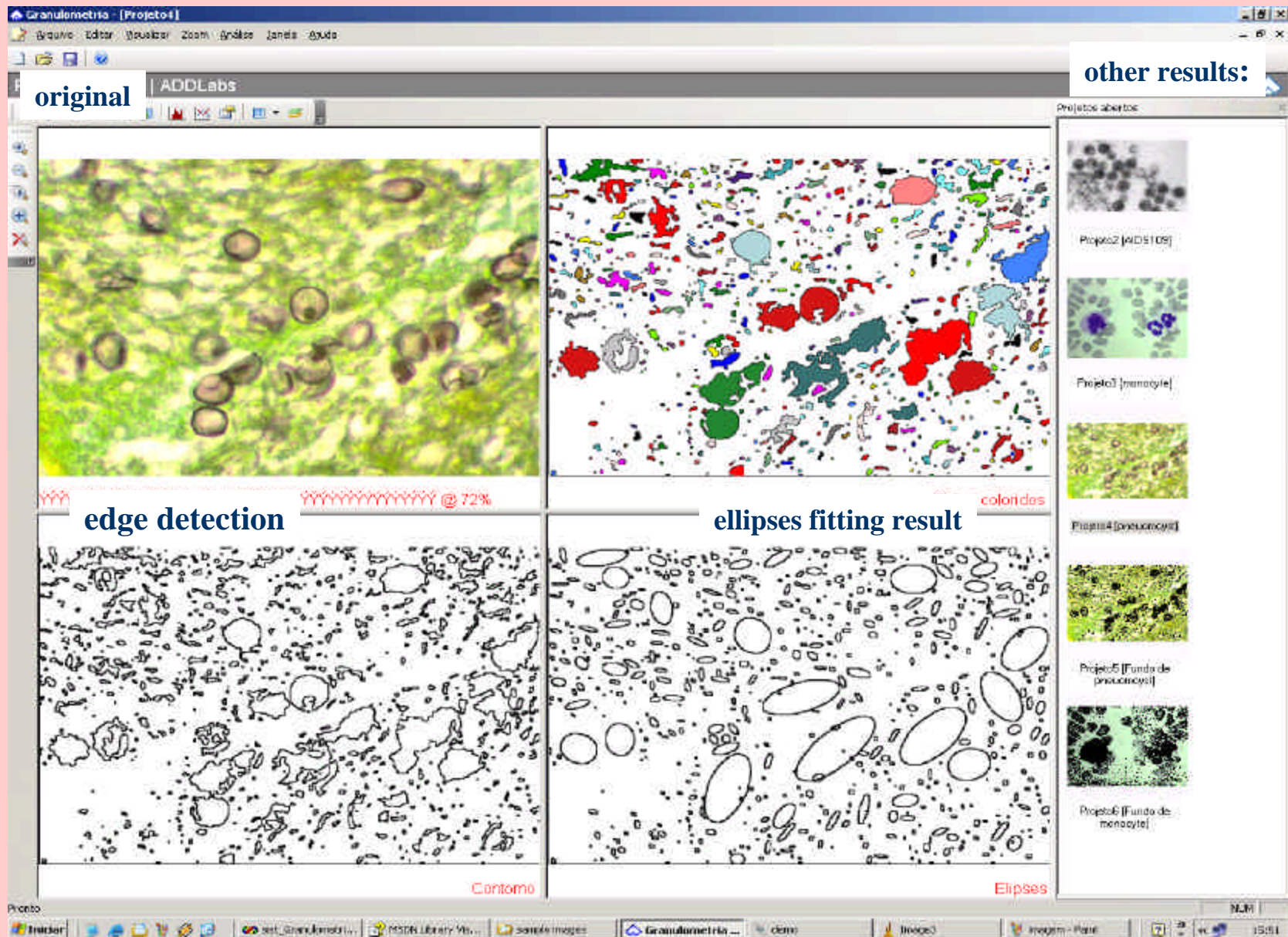
- background extraction
- HSV color space
- windowed threshold
- adaptive edge segmentation
- ellipses fitting of the cells



Examples using hybrid image processing techniques:



Hybrid image processing techniques:



Conclusions:

- It was possible to extract and count virusor blood cells by the proposed approach;
- Techniques here presented for cell-size statistics can be used in others types of cells analysis whether **differentiation** is relevant as
 - ceramic micrography
 - petrology and petrography
 - mineralogy
 - metallurgy

