

Representación, filtrado y segmentación de imágenes I

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Session Overview

1. Digital Image
 - Basics
 - Grays & Colors
 - 3D (a *bit*)
 - Compression
 - Storage Formats
 - Brightness & Contrast / Intensity Histograms

2. Image Processing Overview

- Image Analysis...
 - Segmentation

3. Try It Yourself (free & open tools)

- **ImageJ**
- **FIJI**
- FIJI + Weka plug-in (machine learning)
- Icy
- Ilastik (trainable/machine learning)
- Cellpose
- 3D Slicer
- QuPath (digital pathology)

<https://imagej.net/nih-image/>

<https://fiji.sc>

<https://imagej.net/plugins/tws/>

<https://icy.bioimageanalysis.org/>

www.ilastik.org

www.cellpose.org

<https://slicer.org>

<https://qupath.github.io/>

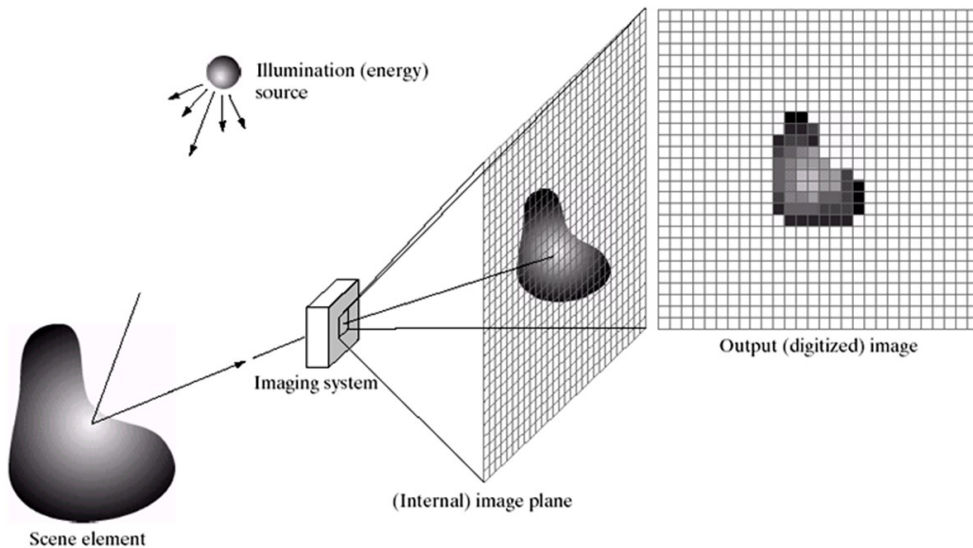
Digital Image



What is...

- ...a pixel?
- ...a voxel?
- ...a bit?
- ...a (raw) image?
- ...JPEG?

Digital Image



a b c d e

FIGURE 2.15 An example of the digital image acquisition process. (a) Energy (“illumination”) source. (b) An element of a scene. (c) Imaging system. (d) Projection of the scene onto the image plane. (e) Digitized image.

Analog Image*

A snapshot of objects by the a camera, telescope, microscope, or any other device (often optical) that registers tone and/or light-shade variations.



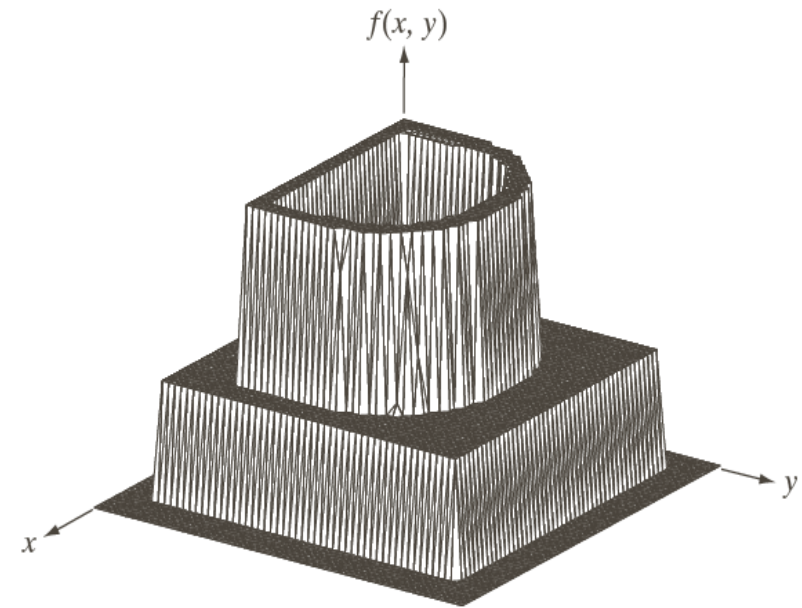
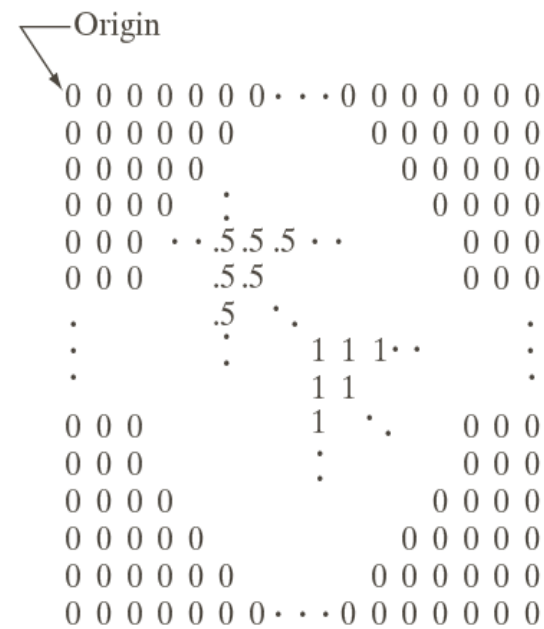
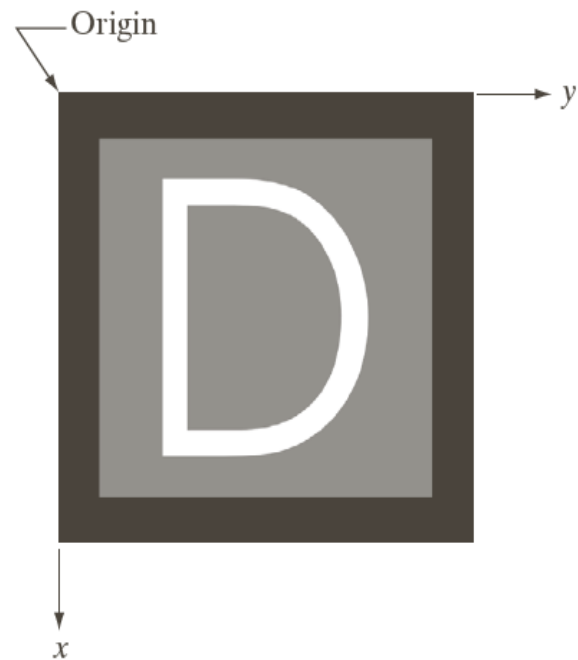
Digital Image

*Analog signal: a representation of a time-or-space varying quantity (*analogous* to it).

Digital Image – Basics

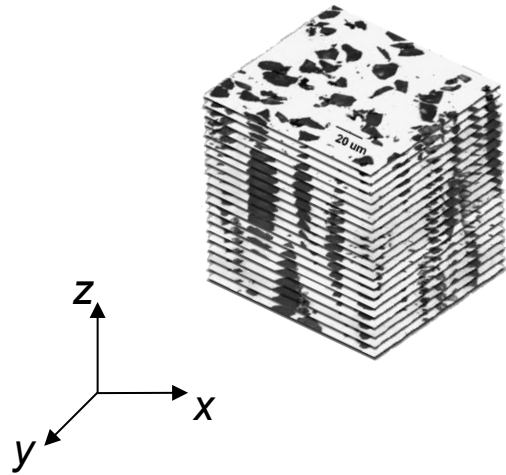
We can think of the color or intensity of an image as a function (f) of the x, y, z coordinates.

In 2D, the gray level intensity $I = I(x, y)$



Digital image means...

...discrete (and also finite!)



A 3D “image stack”
(three spatial dimensions)

- A discrete set is composed by elements which are “isolated” one from another
Examples:

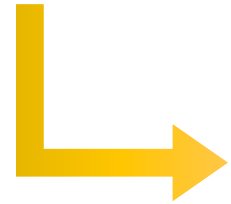
- Natural numbers $\{1, 2, 3, \dots\}$ (infinite set)
- Natural numbers from 1 to 10 (finite set)

- If not discrete? Continuum
Example: real numbers in the $[0,1]$ interval
(infinite set)

Digital Image – Basics (Acquisition)

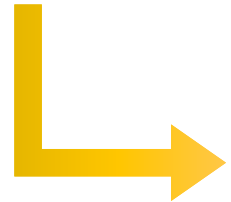
Conversion / Transduction

Light (photons) → Voltage (tension)



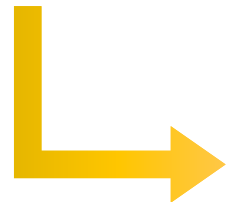
Sampling

Signal capture at defined intervals (time & space)



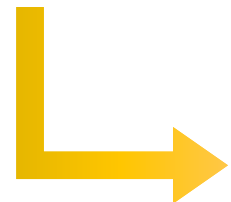
Quantization

Numerical value mapping



Coding

Numerical value data coding
(Raw, TIFF, PNG, JPEG, N5, Zarr)



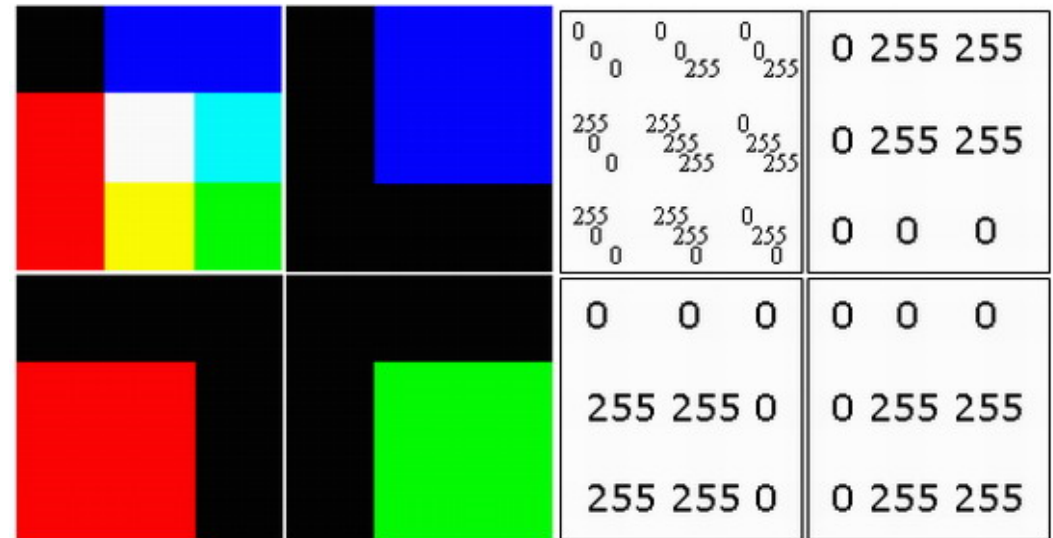
“Bonus”: *Metadata-ing*

Timestamp, Acquisition Device Data, File permissions, ...

Digital Image – Basics

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



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

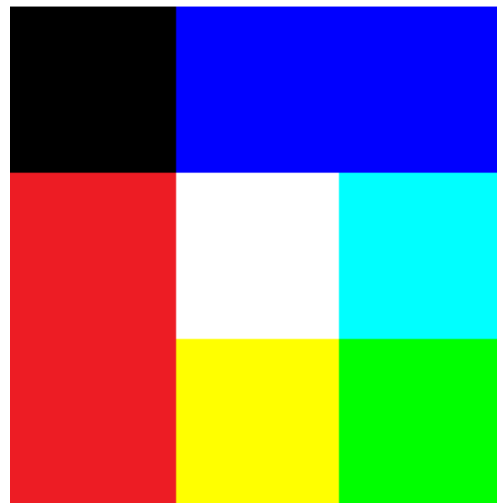
$$(x, y) \in [0, \dim_x - 1] \times [0, \dim_y - 1]$$

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

Digital Image – Basics

The ubiquitous RGB image

- Three *channels* coding primary colors: Red, Green, Blue. 8 bits determine [0,255] range for each channel



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 channels can be used, *e.g.* CMYK (Cyan, Magenta, Yellow, Black)

Digital Image – Basics

The gray scale

- One brightness (intensity) level per pixel

0	85	85
85	255	170
85	170	85

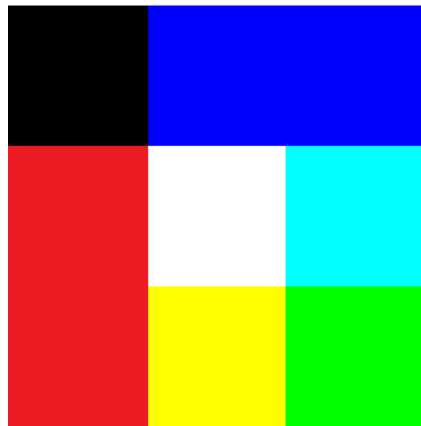
$I[x,y]$



How to go from an RGB to a greyscale image?

Digital Image – Basics

From RGB to grays



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

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

0	85	85
85	255	170
85	170	85

$I[x,y]$



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

Digital Image – Basics

- 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)
- To show a vector image in a common digital screen (matrix of pixels), a **rasterization** algorithm is applied

Scalable Vector Graphics

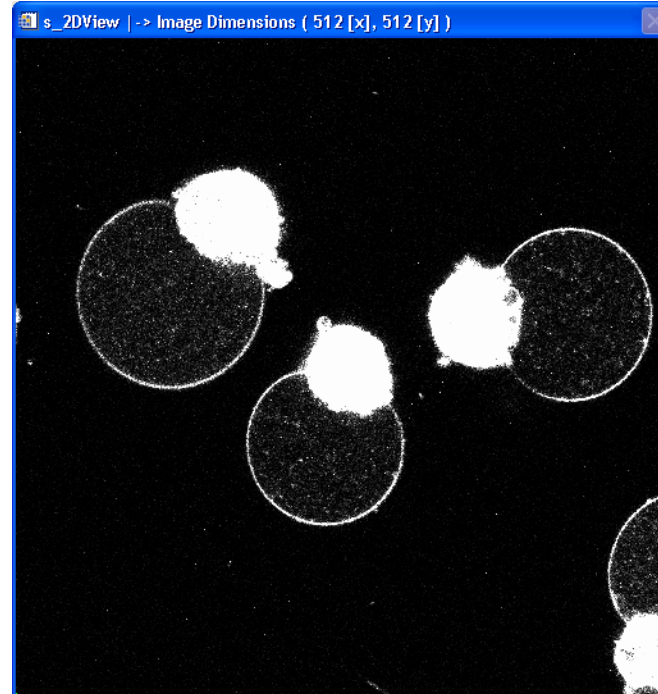


```
<?xml version="1.0" encoding="UTF-8" standalone="no" ?>
<svg version="1.0" xmlns="http://www.w3.org/2000/svg" >
  <defs>
    <linearGradient x1="99.7" x2="99.7" y1="0" y2="100" id="box_gr" />
  </defs>
  <use xlink:href="#box_gr" x="100" y="300" width="90" height="90" />
  <use xlink:href="#circle" x="100" y="300" width="90" height="90" />
  <!--add more content here-->
  <circle cx="100" cy="300" r="45" />
</svg>
```



Digital Image – Gray Levels

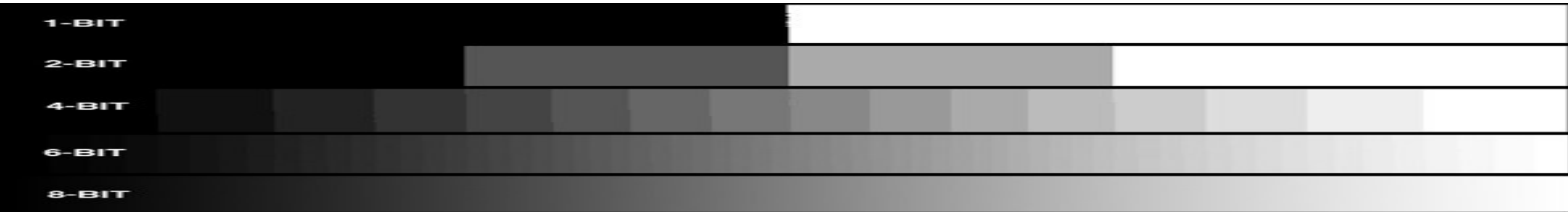
Binary value	Decimal value
0000 0000	0 (black)
0000 0001	1
0000 0010	2
0000 0011	3
0000 0100	4
0000 0101	5
0000 0110	6
0000 0111	7
0000 1000	8
...	...
1111 1011	251
1111 1100	252
1111 1101	253
1111 1110	254
1111 1111	255 (white)



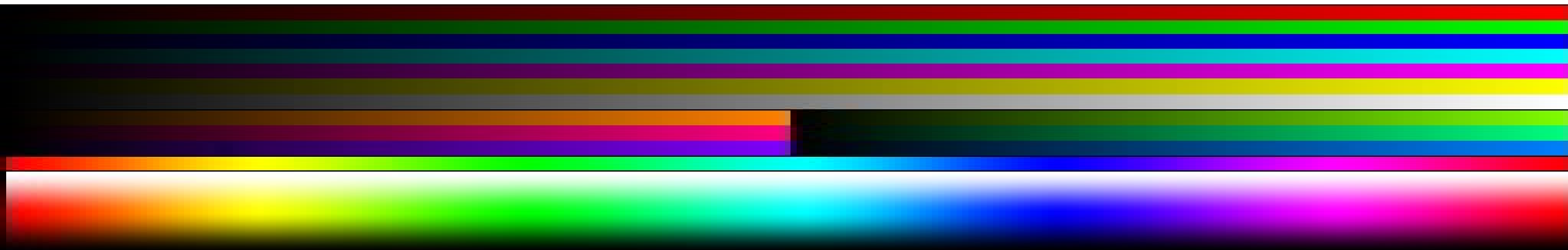
$$I(290,267) = 220$$

8 bit single-channel (grays) image
 n bits $\rightarrow 2^n$ intensity values

Digital Image – Gray & Color Levels



<https://www.projectorcentral.com/All-About-Bit-Depth.htm> (8-bit rendering in most web browsers and monitors)



https://en.wikipedia.org/wiki/File:RGB_24bits_palette_color_test_chart.png

How many levels can
YOU distinguish?

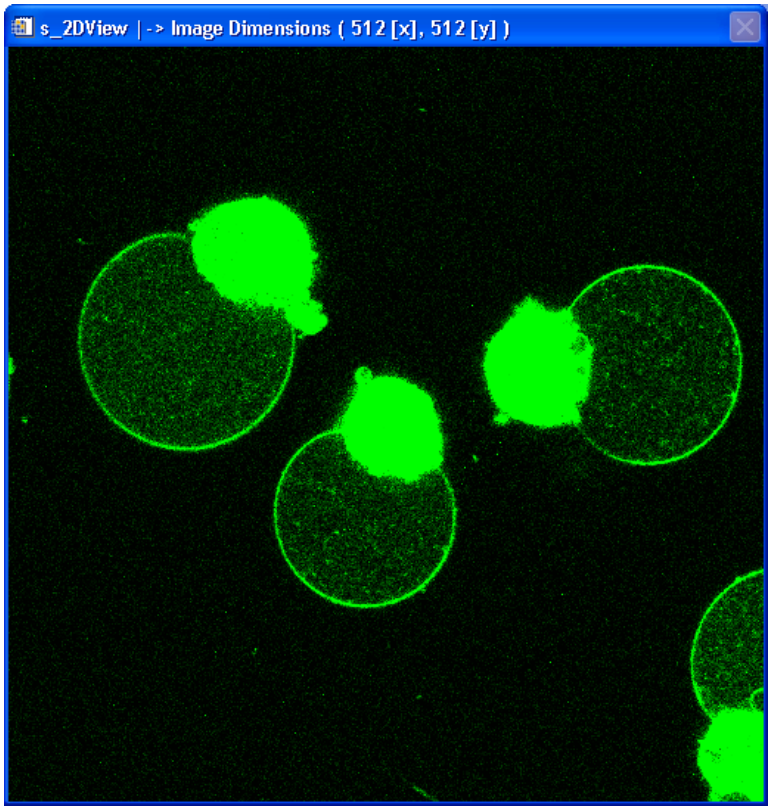
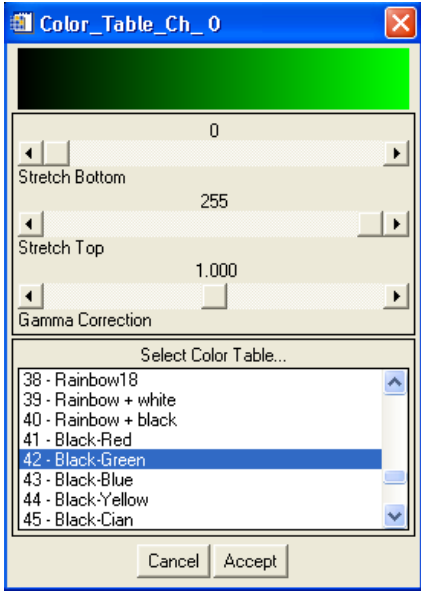
How many levels can
YOUR DISPLAY show?

https://en.wikipedia.org/wiki/Color_depth

Digital Image – Color Spaces

- *Lookup tables* (LUTs) are useful for visualization purposes. For instance, a greyscale image can be displayed using a black-to-green LUT...

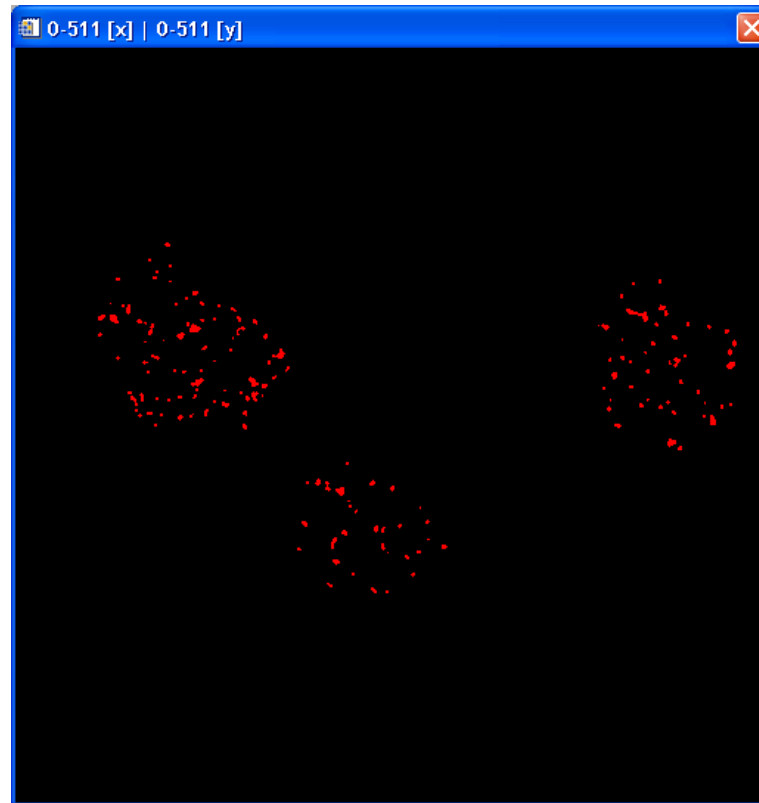
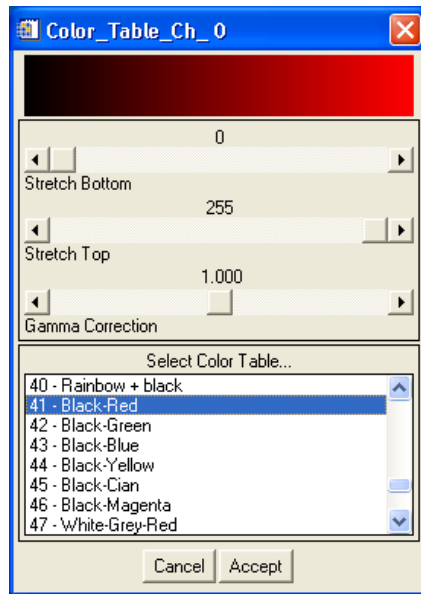
R	G	B
0	0	0
0	1	0
0	2	0
:	:	:
:	:	:
:	:	:
0	200	0
:	:	:
:	:	:
0	255	0



Digital Image – Color Spaces

- Now with a black-to-red LUT...

R	G	B
0	0	0
1	0	0
2	0	0
:	:	:
:	:	:
:	:	:
:	:	:
220	0	0
:	:	:
:	:	:
255	0	0

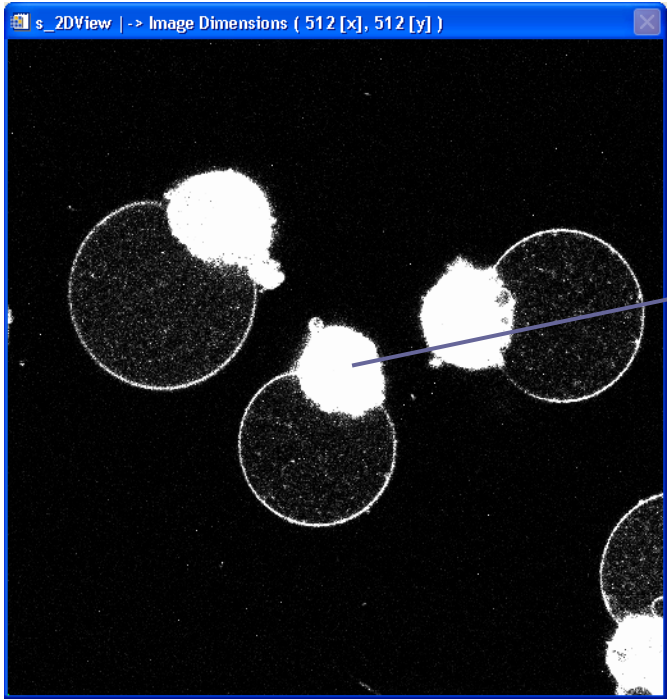


In ImageJ/FIJI:
Go to the
"Image" → "Lookup
Tables" menu

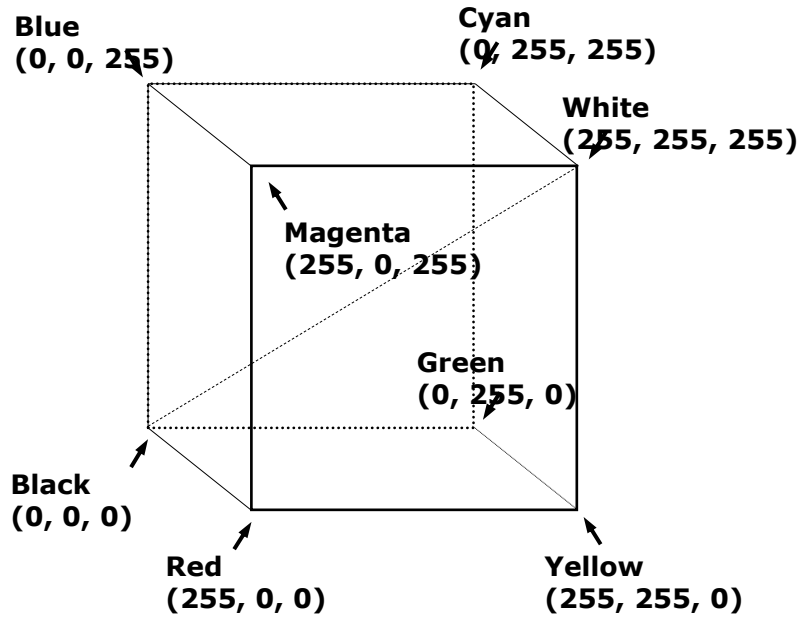
Digital Image – Color Spaces

- ...or any custom table

R	G	B
0	0	0
⋮	⋮	⋮
⋮	⋮	⋮
⋮	⋮	⋮
⋮	⋮	⋮
⋮	⋮	⋮
⋮	⋮	⋮
⋮	⋮	⋮
220	220	220
⋮	⋮	⋮
⋮	⋮	⋮
255	255	255



$I(290,267) = 220$



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

Digital Image – Color Spaces

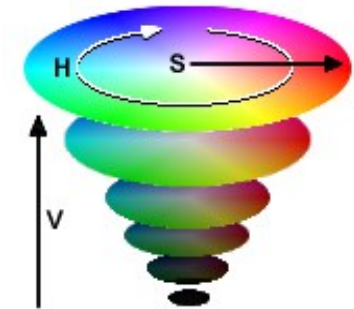
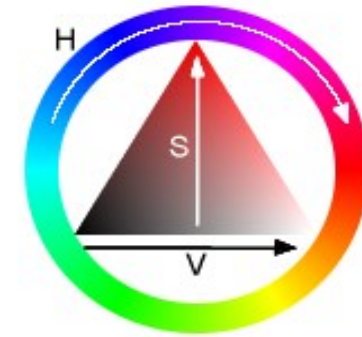
HSV (hue, saturation, value) model

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.

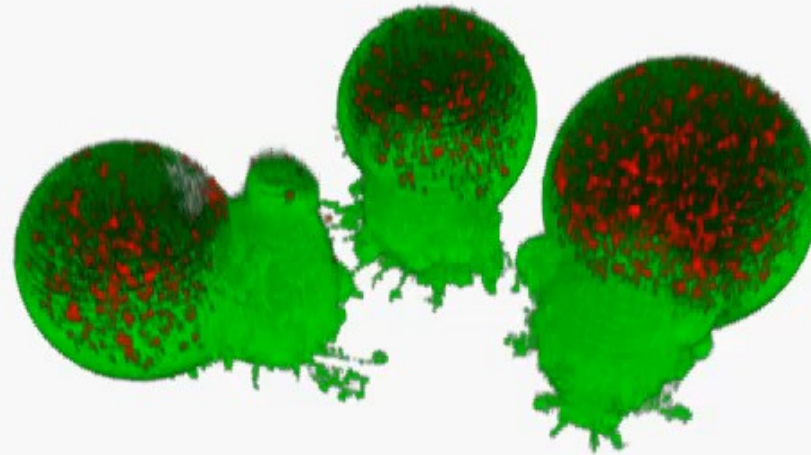
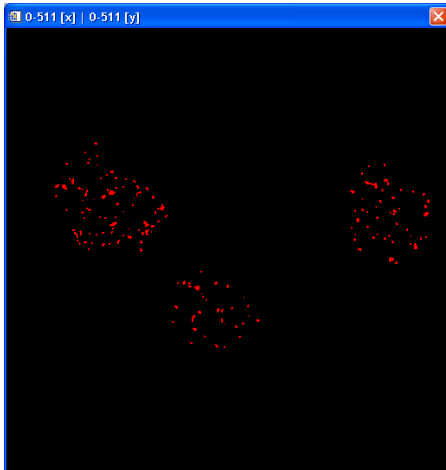
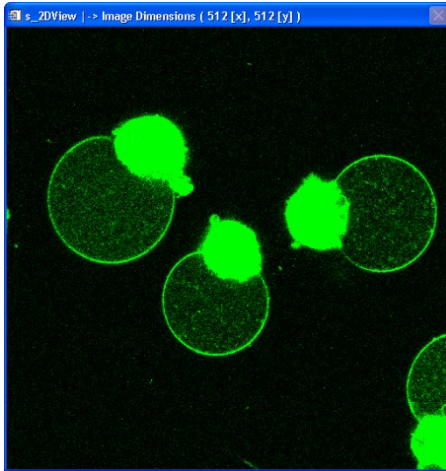
See also HSL (hue, saturation, lightness/intensity)



<http://color.lukas-stratmann.com/color-systems/hsv.html>

<http://color.lukas-stratmann.com/color-systems/hsl.html>

Digital Image – 3D



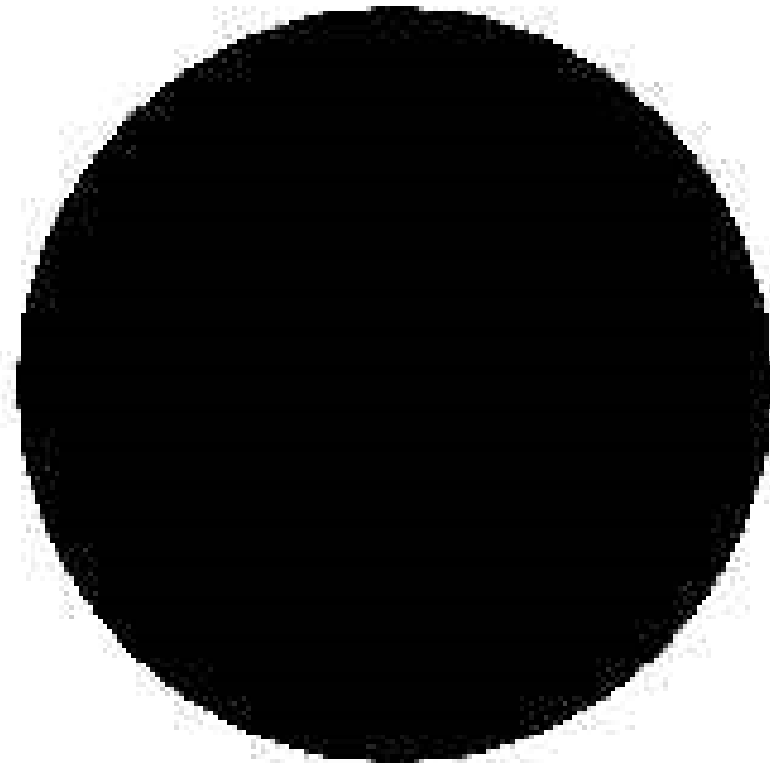
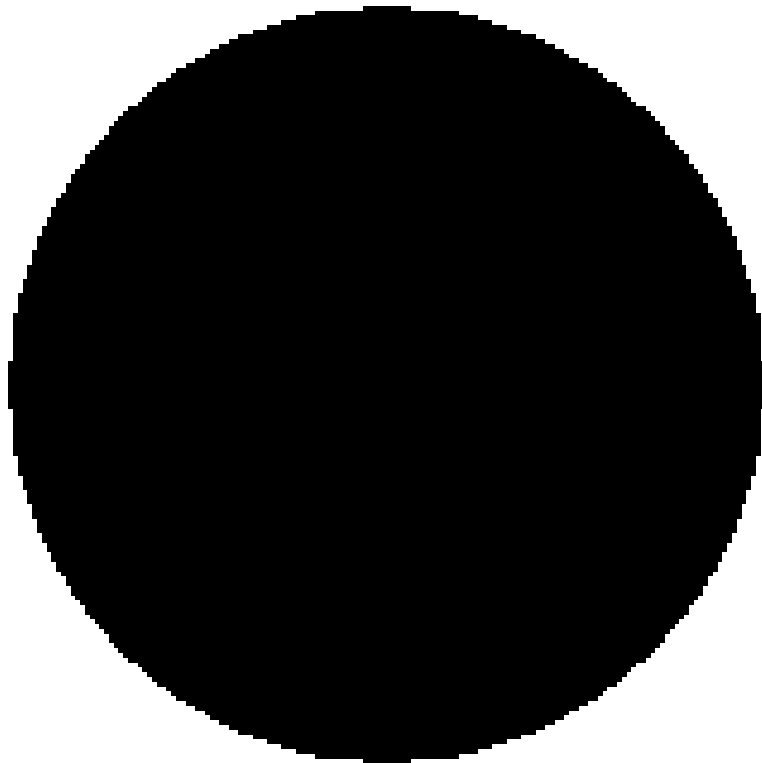
Occlusions may occur in 3D visualization

[R, G, B, α]

Opacity values associated to voxels

Digital Image – Compression

- Lossless vs. Lossy Compression

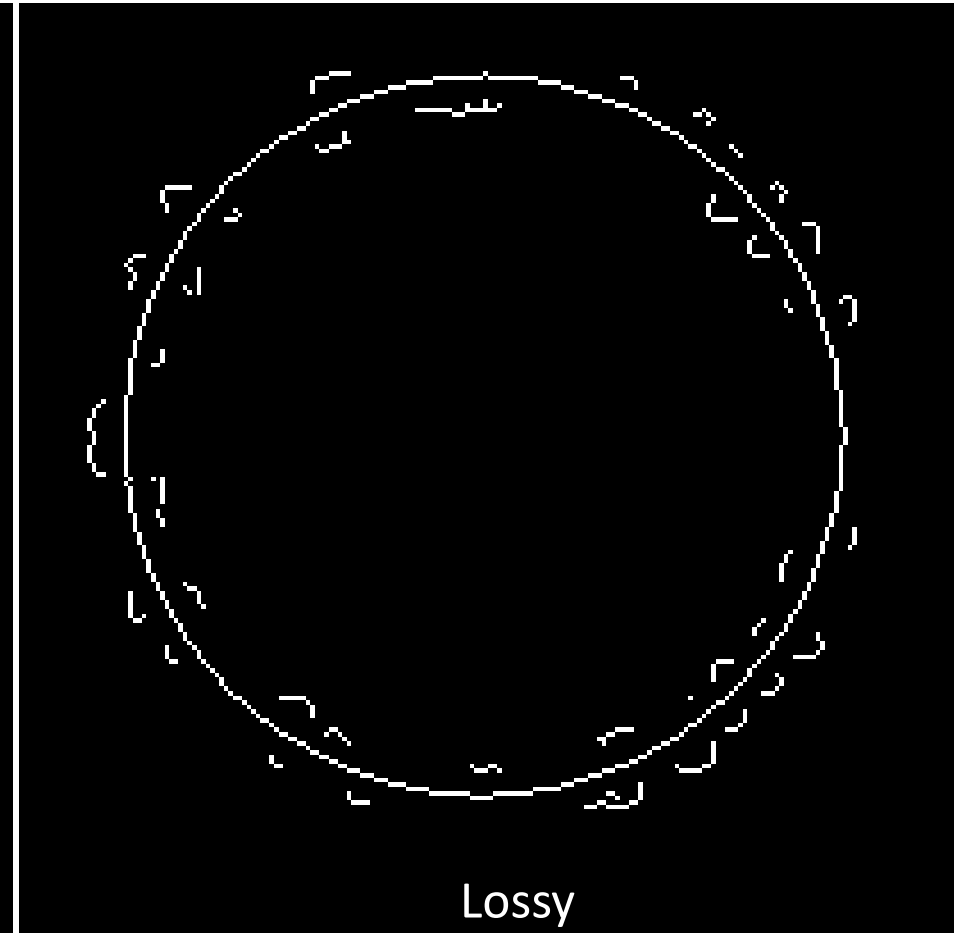
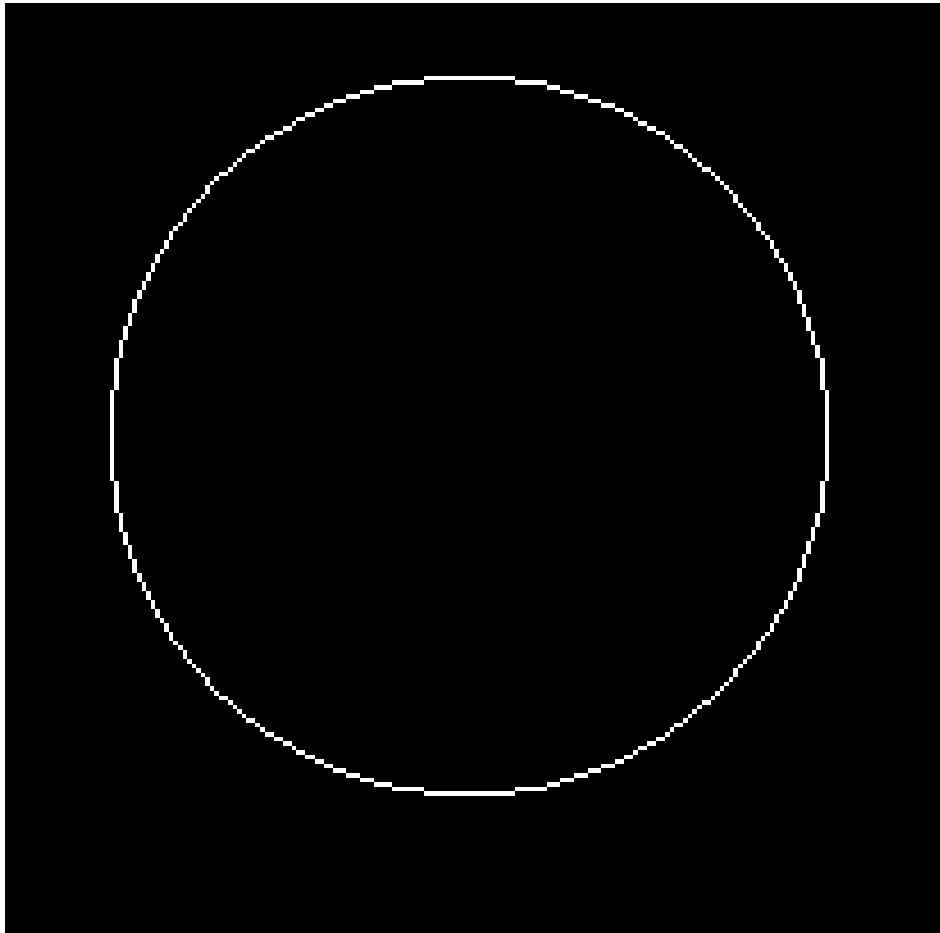


Lossy

Digital Image – Compression

- Lossless vs. Lossy Compression

Canny edge detector



Digital Image – Storage Formats

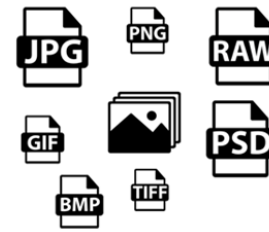
Main aspects to consider:

- Image representation
 - Raster, SVG, base functions
- Color mode
 - Grayscale or 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)
- Storage format
 - “Raw”: each pixel value is stored (lots of space)
 - Compressed, with or without information loss (e.g. JPEG *lossy*, TIFF compressed or uncompressed)

Digital Image – Storage Formats

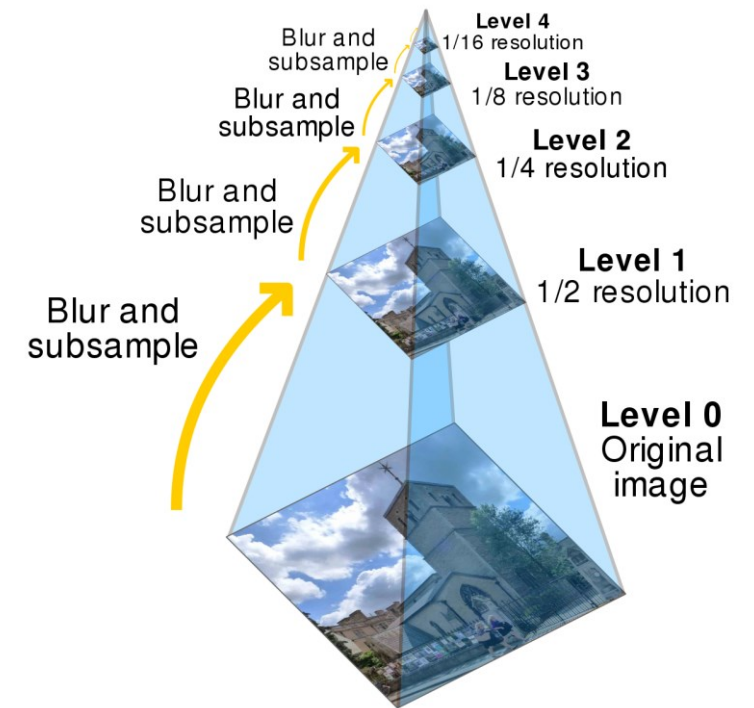
Standard formats

- Portable Network Graphics PNG
- Tagged Image File Format TIFF (2D/3D multi-channel, LUT and/or compression support)
- DICOM (container, 2D/3D)



Formats for large n-dimensional arrays

- N5 <https://github.com/saalfeldlab/n5>
- Zarr <https://imagej.net/N5>
https://www.youtube.com/watch?v=lnqZ_NsgB4g
<https://zarr.readthedocs.io/en/stable/>



A 2D image pyramid. Source: [https://en.wikipedia.org/wiki/Pyramid_\(image_processing\)](https://en.wikipedia.org/wiki/Pyramid_(image_processing))

Digital Image – Brightness & Contrast



Brightness? Contrast?



Digital Image – Brightness & Contrast

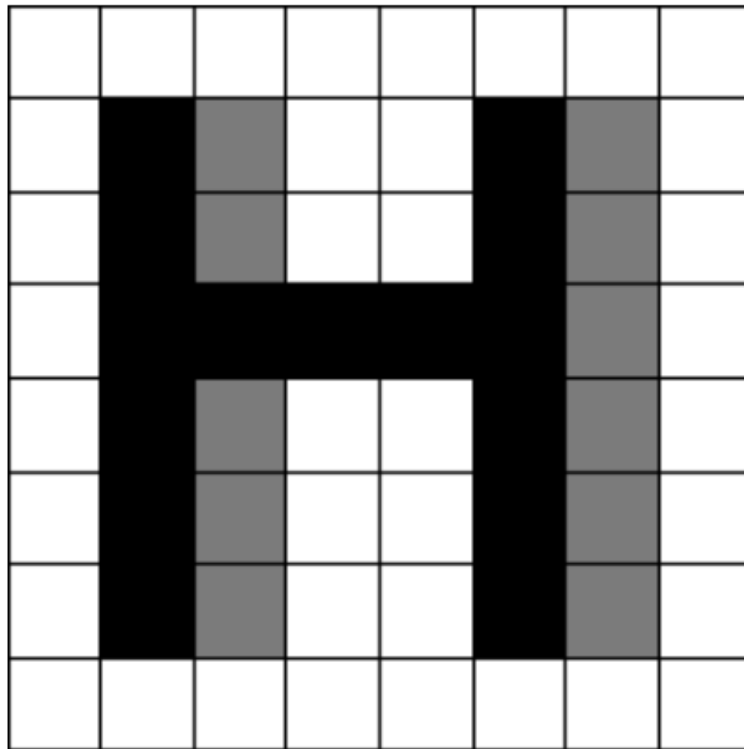


Figura 2. Imagen de 8x8 píxeles.

Nivel de gris	Brillo
0	Negro
1	Gris oscuro
2	Gris claro
3	Blanco

Digital Image – Brightness & Contrast

La gráfica que aparece en la Figura 3 es el histograma correspondiente a esta imagen.

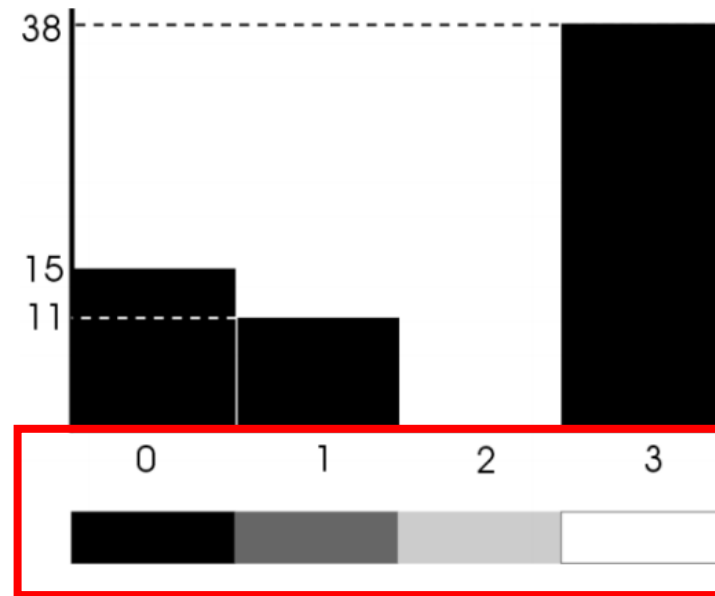


Figura 3. Histograma correspondiente a la Figura 2.

El eje horizontal representan los niveles de gris que pueden aparecer en la imagen: a la izquierda está el valor más oscuro (negro) y en el extremo derecho el más claro (blanco). El resto de niveles se distribuyen uniformemente. Se ha puesto una escala con los tonos de gris correspondientes para facilitar la comprensión. En un histograma real habitualmente no encontrará numerado el eje vertical, ni la escala de tonos para el eje horizontal.

Digital Image – Intensity Histograms

La gráfica que aparece en la Figura 3 es el histograma correspondiente a esta imagen.

La altura de cada barra representa el número de píxeles de la imagen que presentan ese nivel de gris concreto.

Se puede deducir entonces que la imagen tiene :

15 píxeles completamente negros (con nivel 0)

11 de tono gris oscuro (nivel 1)

38 píxeles completamente blancos (nivel 3).

No hay ningún píxel en la imagen con un nivel de gris 2.

¿Cuánto deberán sumar las alturas de todas las barras?

64, que es el número total de píxeles que tiene la imagen.

Por lo tanto, con sólo mirar el histograma podemos deducir algunas cosas interesantes sobre la imagen, lo que demuestra su utilidad

La mayor parte de los píxeles son blancos, así que probablemente se aprecie un fondo blanco uniforme. Hay un número significativo de píxeles totalmente blancos y totalmente negros, por lo que presentará un aspecto bien contrastado.

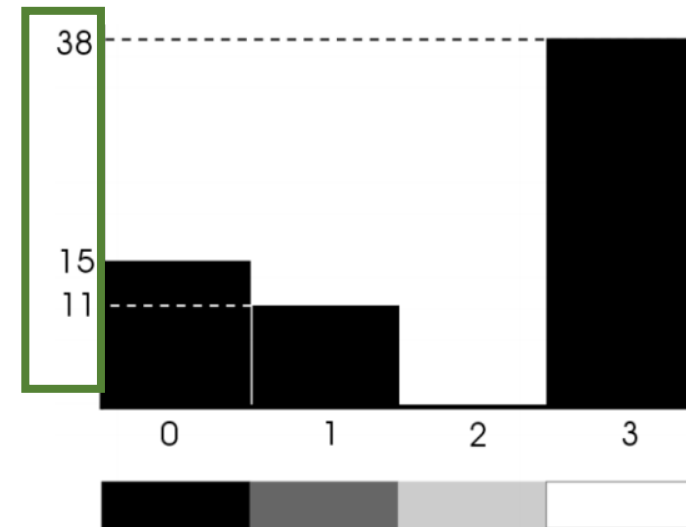
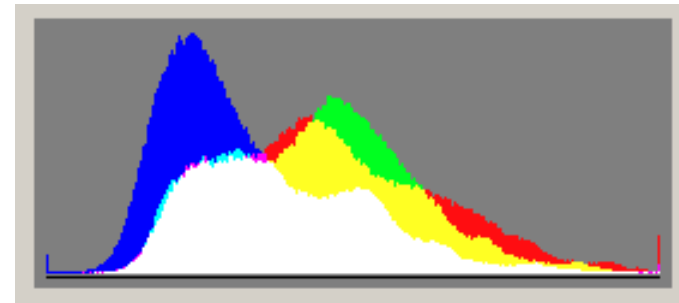
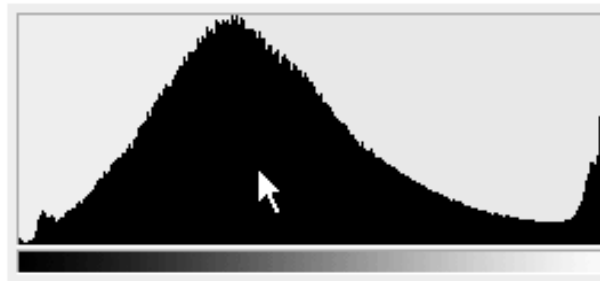
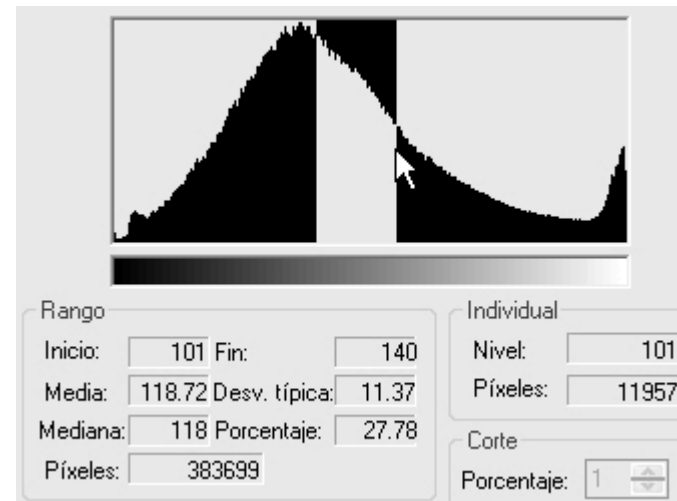


Figura 3. Histograma correspondiente a la Figura 2.

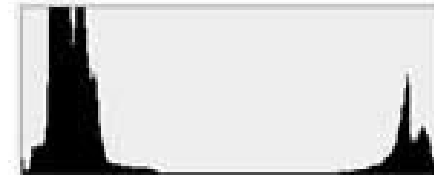
Digital Image – Intensity Histograms



Podemos obtener información de un área determinada del histograma.



Digital Image – Intensity Histograms



Digital Image – Intensity Histograms

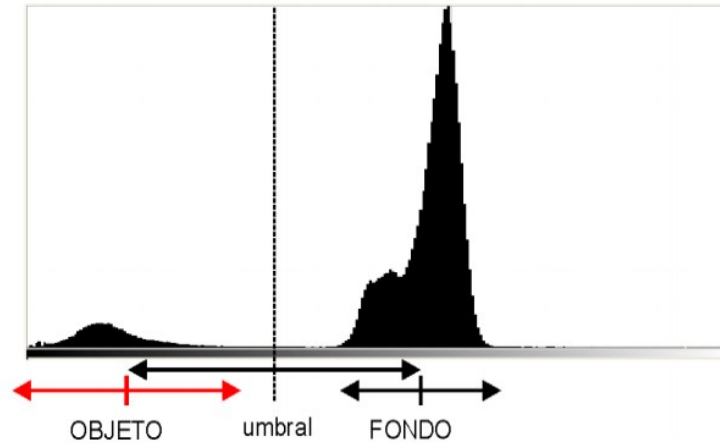


Si una fotografía está sobreexpuesta el histograma estará inclinado hacia la derecha (más píxeles iluminados)

Si una fotografía está subexpuesta el histograma estará inclinado hacia la izquierda (más píxeles oscuros)

Si una fotografía esta correctamente expuesta el histograma estará en el centro o extendido.

Digital Image – Intensity Histograms



Histograma
Bimodal

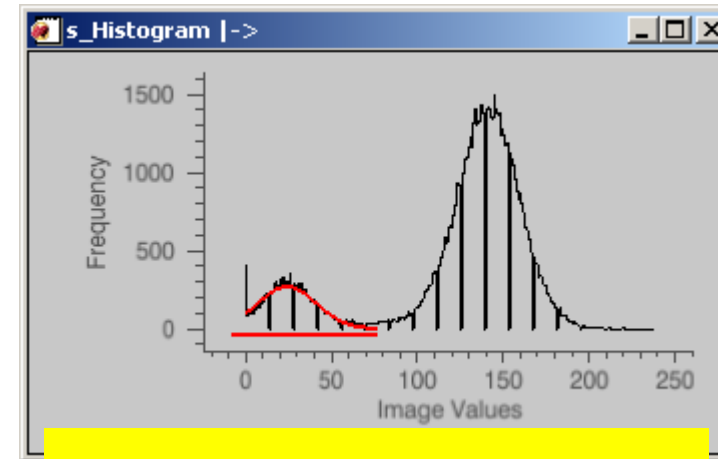
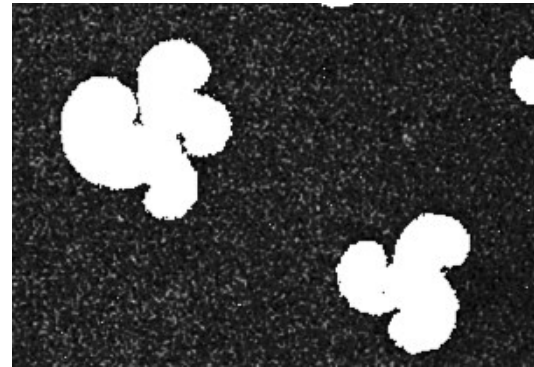
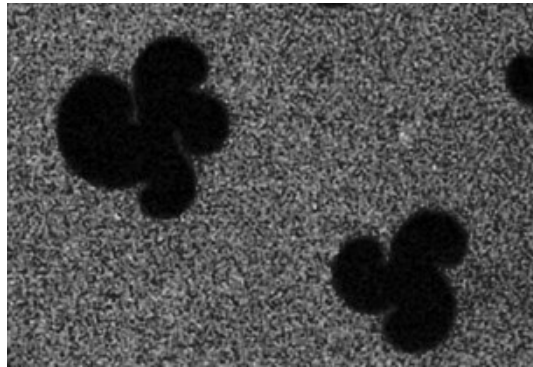
Figura 12. Ejemplo de umbralización de una imagen con ayuda de su histograma.



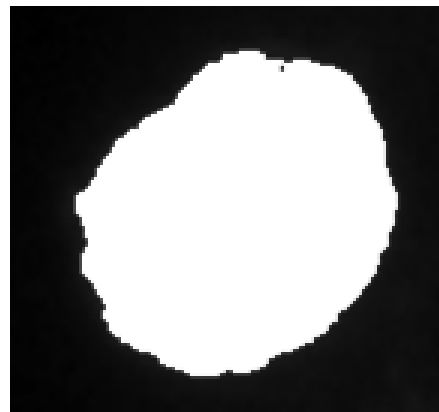
Figura 13. Resultado de la umbralización automática de la imagen anterior.

Digital Image – Intensity Histograms

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



Intensity histograms



Digital Image – Intensity Histograms

Observación	Diagnóstico
Histograma amplio, cubre todo o casi todo el rango tonal	Imagen óptima, buen contraste
Histograma estrecho	Falta de contraste
Histograma desplazado a la izquierda	Escena oscura Imagen poco expuesta
Histograma desplazado a la derecha	Escena con mucho brillo o tonos predominantemente claros. Imagen demasiado expuesta.

Tabla 2. Resumen de la interpretación de un histograma.