What is ImageJ?

❖ One of the most important image processing software that employed in scientific researchs.

❖ Developed by Wayne Rasband for National Institute of Health (Washington, USA).

❖ It is completely free, open source Java program.

❖ It can be installed on Windows, Mac OS X and Linux in both 32-bit and 64-bit modes.

What can ImageJ do?

❖ Open and save most Image formats like GIF, JPEG, BMP, PNG, PGM, FITS , ASCII, DICOM, and many other formats using plugins.

❖ Considered as fastest image processing Java program in the world which can filter 2048 x 2048 image (40 million pixels) in 0.1 second.

❖ Provide zooming ranges (1:32 to 32:1) with scrolling and guaranteed functioning pf all analysis processes at any magnification factor.

❖ Analyze images of 8-bit, 16-bit, 32-bit and RGB color.

❖ Split a 32-bit color image into RGB or HSV components. Merge 8-bit components into a color image. Convert an RGB image to 8-bit indexed color. Apply pseudo-color palettes to grayscale images.

What can ImageJ do?

❖ Measure area, mean, standard deviation, min and max of selection or entire image. Measure lengths and angles. Use real world measurement units such as millimeters. Calibrate using density standards. Generate histograms and profile plots.

❖ Supports smoothing, sharpening, edge detection, median filtering and thresholding on both 8-bit grayscale and RGB color images. Interactively adjust brightness and contrast of 8, 16 and 32-bit images.

❖ Create rectangular, elliptical or irregular area selections. Create line and point selections. Edit selections and automatically create them using the wand tool. Draw, fill, clear, filter or measure selections. Save selections and transfer them to other images.

What can ImageJ do?

❖ Cut, copy or paste images or selections. Paste using AND, OR, XOR or "Blend" modes. Add text, arrows, rectangles, ellipses or polygons to images.

❖ Display a "stack" of related images in a single window. Process an entire stack using a single command. Open a folder of images as a stack. Save stacks as multi-image TIFF files.

Other features?

❖ Extend ImageJ by developing plugins using ImageJ's built in text editor and Java compiler. More than 500 plugins are available.

❖ ImageJ and its Java source code are freely available and in the public domain. No license is required.

Basic concepts – Analog images and Digital images

❖Analog images are the type of images that we, as humans, look at. What we see in an analog image is various levels of brightness (or film density) and colors. It is generally continuous and not broken into many small individual pieces. (Required for human viewing)

❖Digital images are recorded as many numbers. The image is divided into a matrix or array of small picture elements, or pixels. Each pixel is represented by a numerical value. The advantage of digital images is that they can be processed, in many ways, by computer systems. (Required for computer viewing)

Basic concepts – Analog images and Digital images

Basic concepts - Digital images

❖DIGITAL IMAGES are electronic snapshots taken of a scene or scanned

from documents, such as photographs, manuscripts, printed texts, and

artwork. The digital image is sampled and mapped as a grid of dots or

picture elements (pixels).

❖Each pixel is assigned a tonal value (black, white, shades of gray or color),

which is represented in binary code (zeros and ones). The binary digits

("bits") for each pixel are stored in a sequence by a computer and often

reduced to a mathematical representation (compressed). The bits are

then interpreted and read by the computer to produce an analog version

for display or printing (http://preservationtutorial.library.cornell.edu/intro/intro-01.html).

Pixel Values: As shown in this bi-tonal image, each pixel is assigned a tonal value, in this example 0 for black and 1 for white.

Basic concepts - Digital images
What is Pixel ?

❖ Pixel is the smallest visual element (component) of image .

❖ Each pixel is a sample of an image where more samples

provide more accurate representation of the original image.

One Pixel

One pixel

1 inch

inch

Low Density   Medium Density   High Density
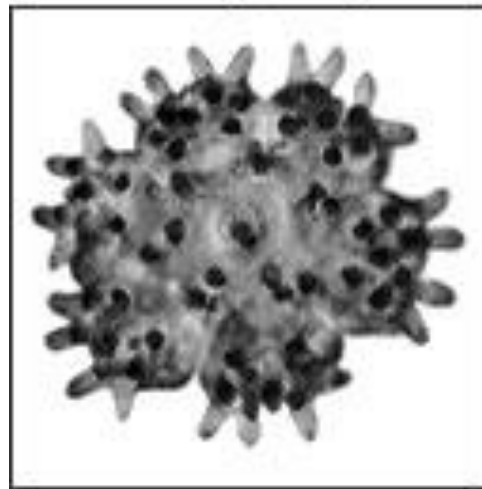
■ 1 pixel   ■ 1 pixel   ▪ 1 pixel

Basic concepts - Digital images - Raster Image

❖Raster Image or bitmap image is a rectangular grid of pixels *i.e.* dot matrix structure.

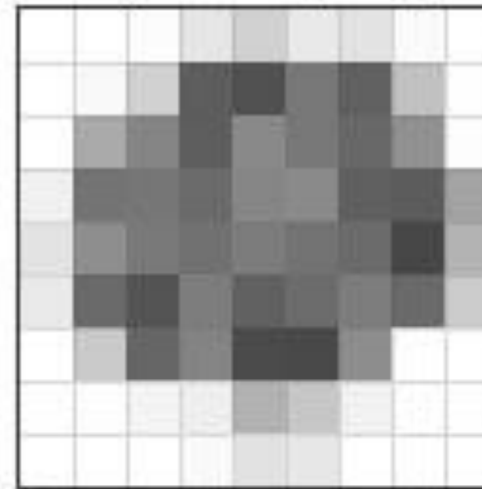❖Image formats like BMP, GIF, JPEG and PNG are an examples of raster images
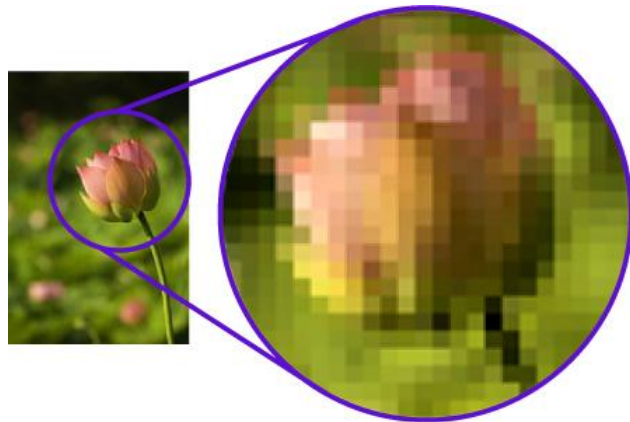
Creation of a Digital Image

Analog Image	Digital Sampling	Pixel Quantization

249	244	240	230	209	233	227	251	255
248	245	210	93	81	120	97	193	254
250	170	133	94	137	120	104	145	253
241	116	118	107	134	138	96	92	163
277	142	121	113	124	115	107	71	179
234	106	84	125	97	108	125	106	204
241	202	102	132	75	73	141	248	252
253	252	244	239	178	199	242	250	245
255	249	244	250	226	231	240	251	253

(a) (b) (c)

Figure 1

Pixel values: As shown in this bilonal image, each pixel is assigned a tonal value, in this example 0 for black and 1 for white

Computer Applications

ImageJ- Lecture 8

Basic concepts - Resolution

❖ RESOLUTION is the ability to distinguish fine spatial detail. The spatial frequency at which a digital image is sampled (the sampling frequency) is often a good indicator of resolution. This is why dots-per-inch (dpi) or pixels-per-inch (ppi) are common and synonymous terms used to express resolution for digital images. Generally, but within limits, increasing the sampling frequency also helps to increase resolution.(http://preservationtutorial.library.cornell.edu/intro/intro-02.html).
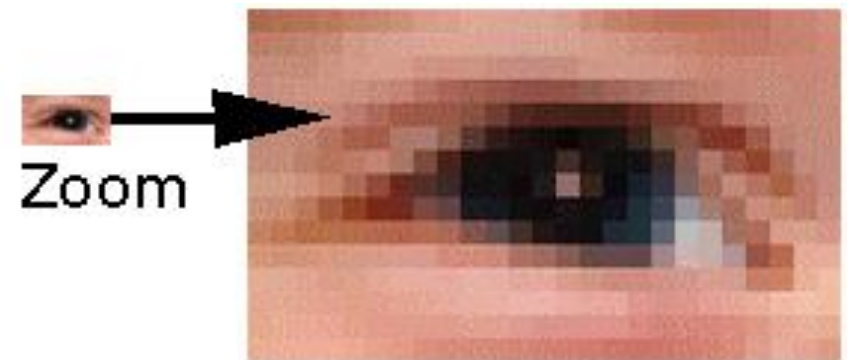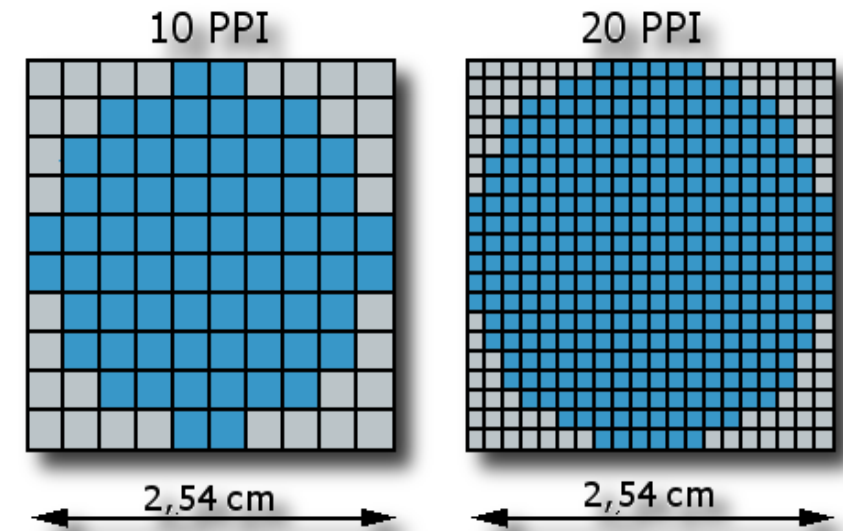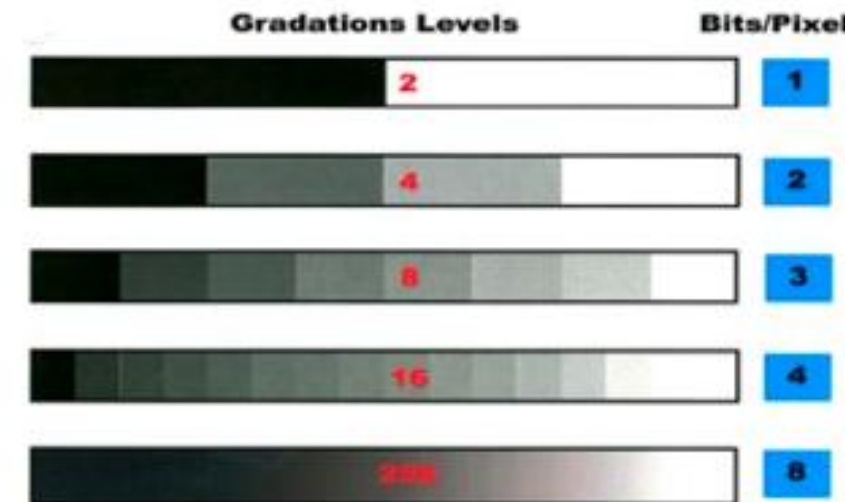
Pixels: Individual pixels can be seen by zooming in an image.

Basic concepts - Bit - depth

❖ BIT DEPTH is determined by the number of bits used to define each pixel. The greater the bit depth, the greater the number of tones (grayscale or color) that can be represented resolution (http://preservationtutorial.library.cornell.edu/intro/intro-04.html).

❖ In a 2-bit image, there are four possible combinations: 00, 01, 10, and 11. If "00" represents black, and "11" represents white, then "01" equals dark gray and "10" equals light gray. The bit depth is two, but the number of tones that can be represented is 2^2 or 4. At 8 bits, 256 (2^8) different tones can be assigned to each pixel.
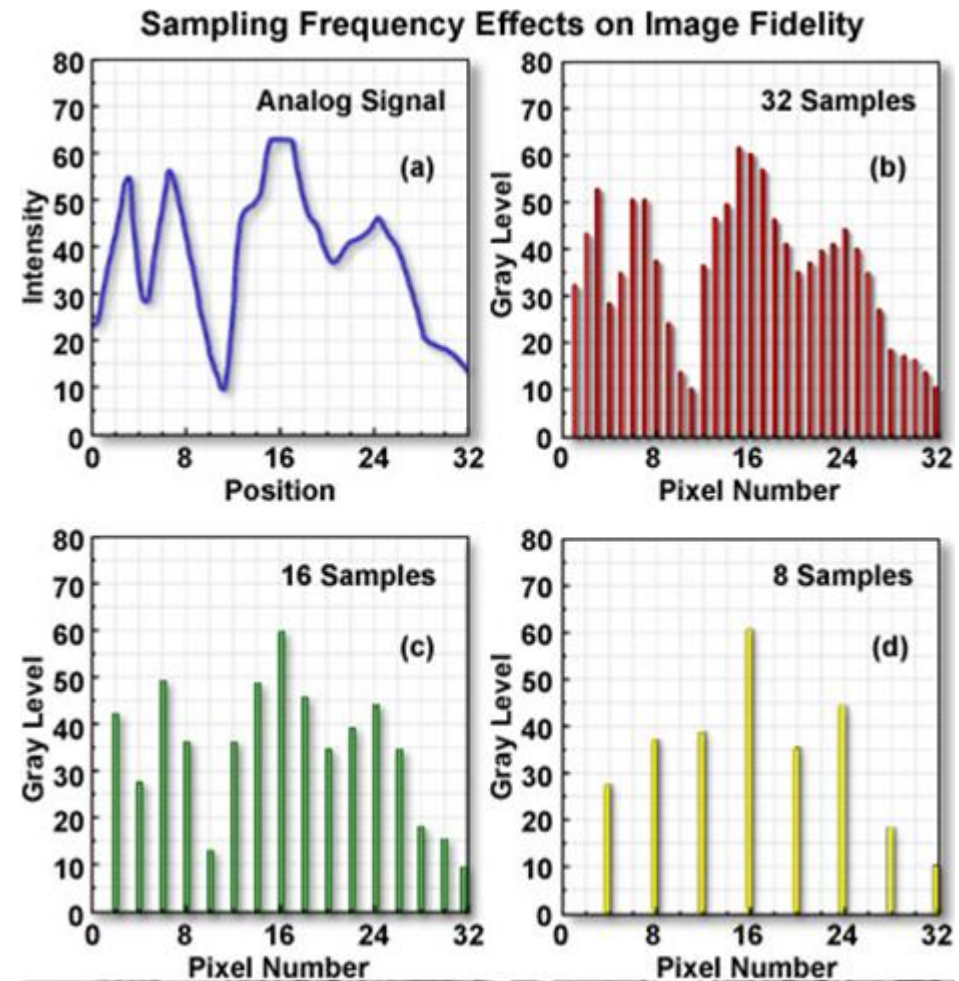
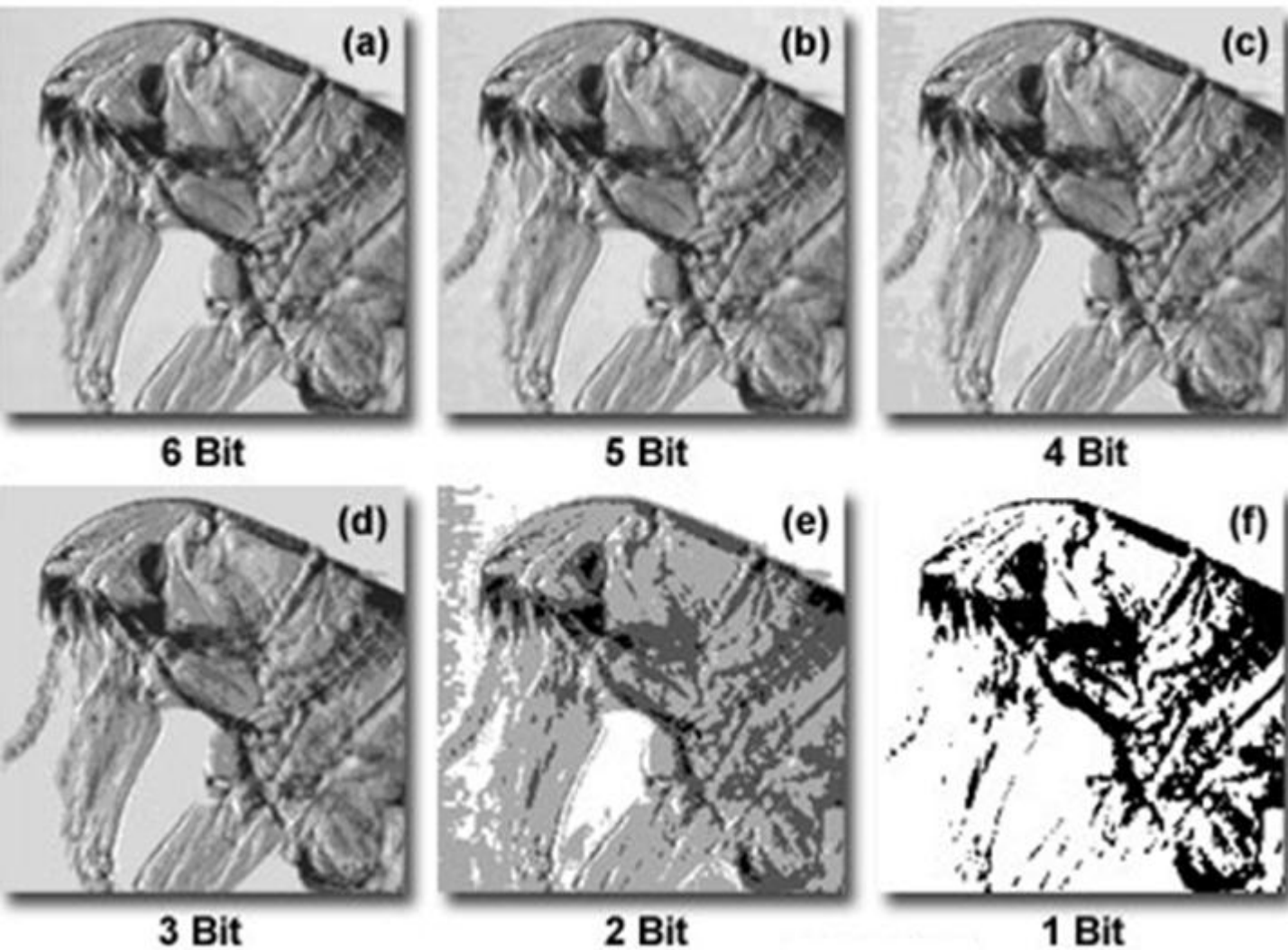1 bit (2^1) = 2 tones
2 bits (2^2) = 4 tones
3 bits (2^3) = 8 tones
4 bits (2^4) = 16 tones
8 bits (2^8) = 256 tones
16 bits (2^{16}) = 65,536 tones
24 bits (2^{24}) = 16.7 million tones

http://copierwise.com/17rules/bitdepth.html

Basic concepts - Bit - depth

Grayscale Resolution and Digital Image Appearance

(a) 6 Bit
(b) 5 Bit
(c) 4 Bit
(d) 3 Bit
(e) 2 Bit
(f) 1 Bit

Sampling Frequency Effects on Image Fidelity

(a) Analog Signal
(b) 32 Samples
(c) 16 Samples
(d) 8 Samples

https://www.olympus-lifescience.com/en/microscope-resource/primer/digitalimaging/digitalimagebasics/

Pixel values: As shown in this bitonal image, each pixel is assigned a tonal value, in this example 0 for black and 1 for white

Computer Applications

ImageJ- Lecture 8

Basic concepts – Bit - depth

❖A color image is typically represented by a bit depth ranging from 8 to 24 or higher. With a 24-bit image, the bits are often divided into three groupings: 8 for red, 8 for green, and 8 for blue. Combinations of those bits are used to represent other colors. A 24-bit image offers 16.7 million (2 ^{24}) color values (http://preservationtutorial.library.cornell.edu/intro/intro-04.html).

Green layer (8 bits)
Red layer (8 bits)
Blue layer (8 bits)
Composite RGB image
8+8+8 = 24 bits
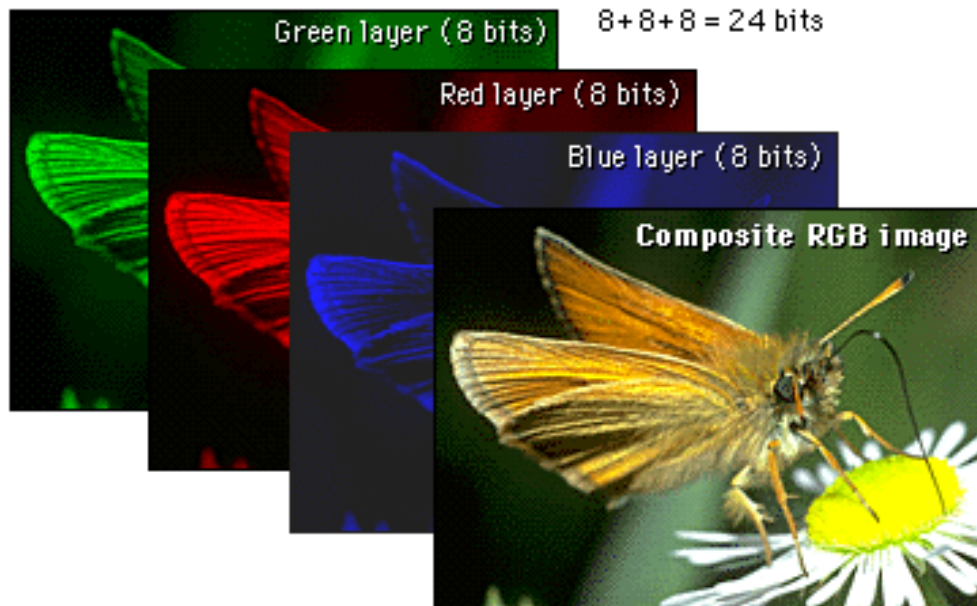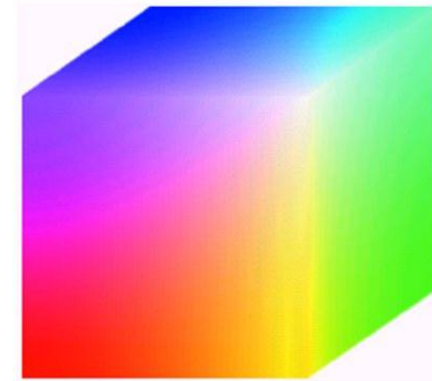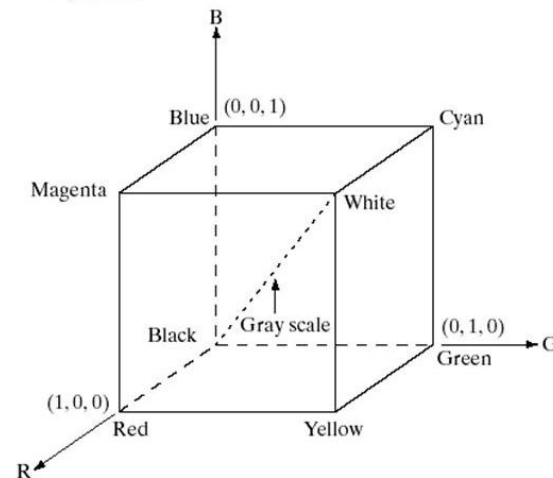
RGB color cube

RGB 24-bit color cube

Conclusion

To analyze image with any image processing program we have to:

❖Work on digital image.

❖Specify the suitable Bit-depth which plays a significant role in image processing.

❖Specify the suitable resolution as it affects the analysis accuracy.

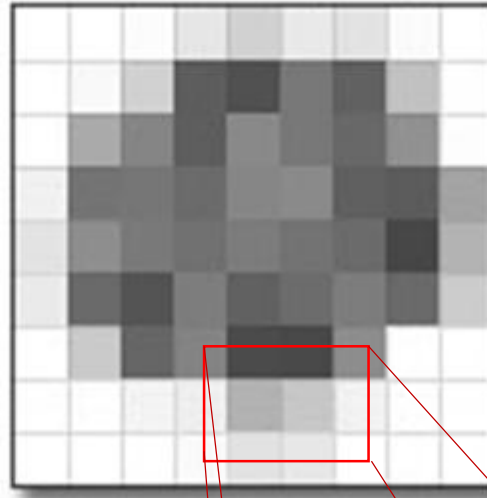❖Using greyscale mode making processing easiest to perform.

Conclusion

Analysis

Analog Image

Digital Sampling

Pixel Quantization

249	244	240	230	209	233	227	251	255
248	245	210	93	81	120	97	193	254
250	170	133	94	137	120	104	145	253
241	116	118	107	134	138	96	92	163
277	142	121	113	124	115	107	71	179
234	106	84	125	97	108	125	106	204
241	202	102	182	75	73	141	246	252
253	252	244	239	178	199	242	250	245
255	249	244	250	228	231	230	251	253

Bit Depth and Gray Levels in Digital Images

2 Bit	4 Bit	6 Bit	7 Bit	8 Bit	10 Bit
4	16	64	128	256	1,024

Gray Levels
(Bit Depth)
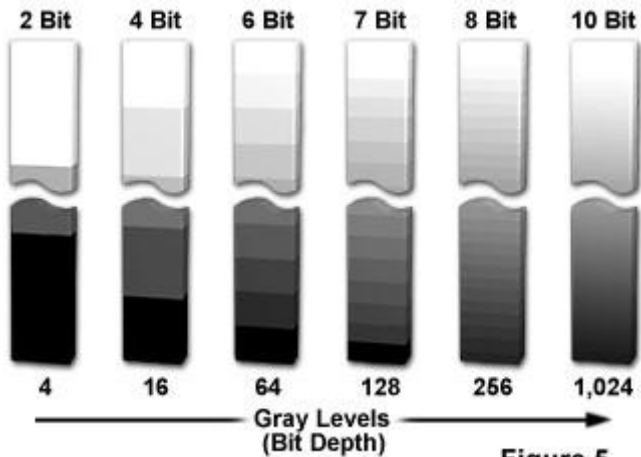
Figure 5

Pixel

Pixel value

178   199