Introduction to Image Processing & Analysis

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The Nature of Images

- What is an image?
  - A two-dimensional structure
  - A “matrix” composed of *n* rows and *m* columns
  - The value at each of the coordinates is a discrete quantity.
  - Each element of the matrix array is called an *image element*, *picture element*, *pixel*, or *pel*.

The Nature of Images

- Each pixel location is denoted by the index (i, j) where $0 \le i < n$ and $0 \le j < m$

$$\begin{array}{|c|c|c|c|}
\hline
0,0 & 0,1 & \cdots & 0,m \\
\hline
1,0 & 1,1 & \cdots & 1,m \\
\hline
\vdots & \vdots & \ddots & \vdots \\
\hline
n,0 & n,1 & \cdots & n,m \\
\hline
\end{array}$$

Quantization

- A set of *n* *quantization* levels comprises the integers $0, 1, 2, \ldots, n-1$
- 0 and *n*-1 are usually black and white respectively, with intermediate levels rendered in various shades of gray.
- Quantization levels are commonly referred to as *gray levels* or *grayscale values*

Quantization

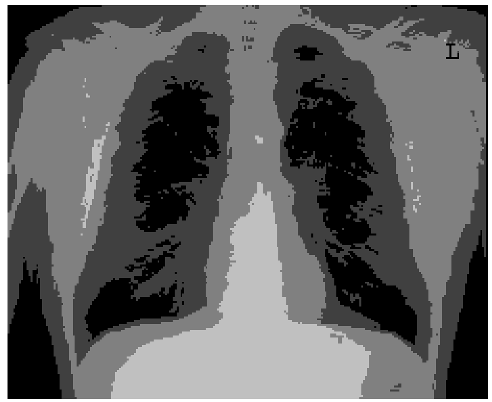
- n is usually an integral power of two

$$n = 2^b$$

- b is the number of bits used for quantization
- Typically b=8 → 256 possible gray levels.
- If b=1, then there are only two values: 0 and 1
  - This image is known as a BINARY image
- Sometimes the range of values spanned by the gray levels is called the *dynamic range* of an image.

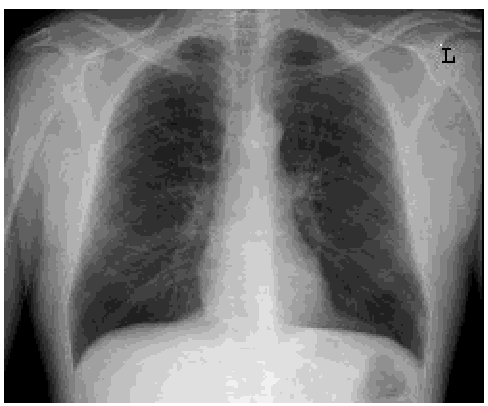
Quantization

n=2

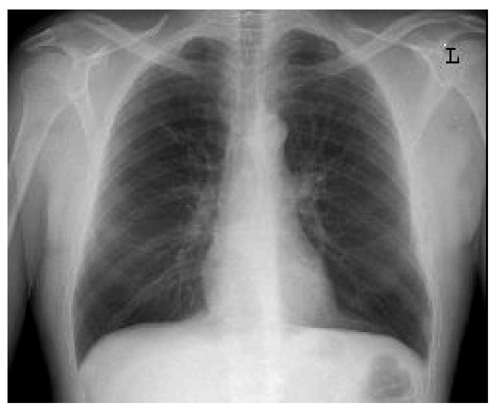


n=4

n=8



n=16



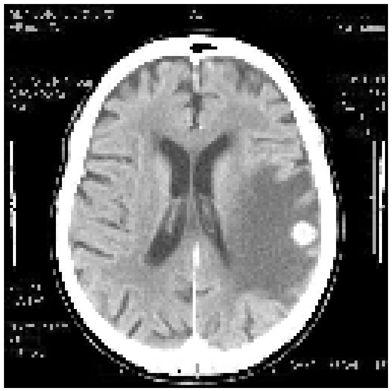
n=64

n=256

Spatial Resolution

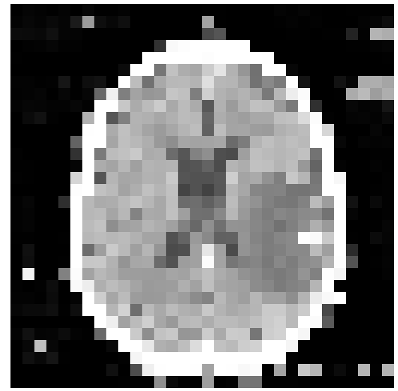
- The spatial resolution of an image is the physical size of a pixel in an image
  - Basically it is the smallest discernable detail in an image.

Spatial Resolution

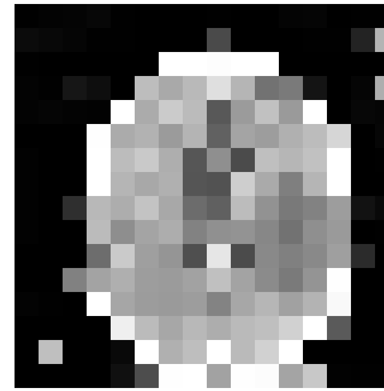


128x128

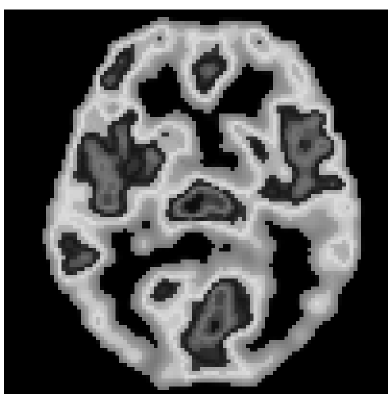
64x64



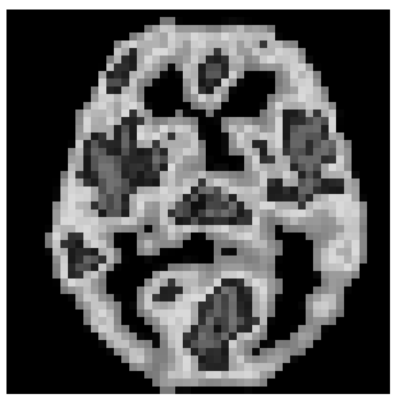
32x32



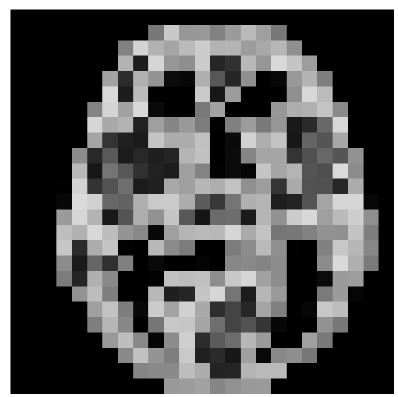
16x16



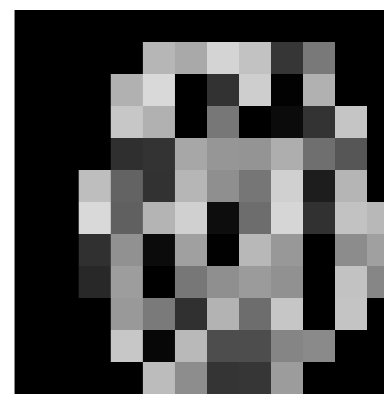
100x100



50x50



24x24



12x12

Image Representation

- Usually a single quantized value is associated with each pixel of an image
  - A two-dimensional array of bytes
  - Denote a pixel location by (i, j) where i is the row index and j is the column index
- JAVA

```
byte [][] image = new byte[height][width]
```

- MATLAB

```
image = zeros(width,height)
```

Looking Inside the Human Body

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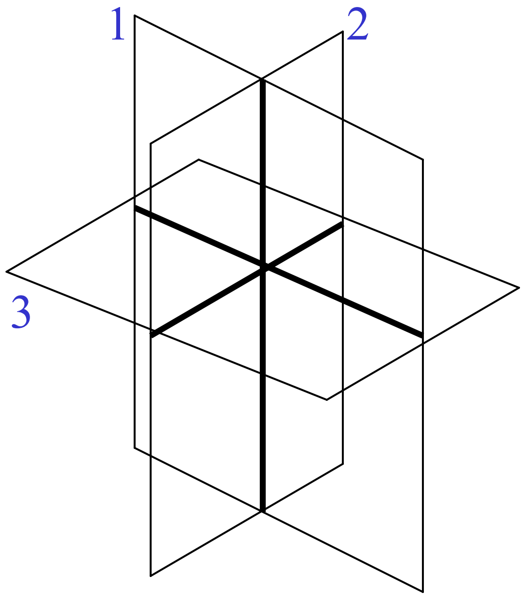
Medical Imaging

- Medical imaging is the visualization and measurement of characteristics associated with internal anatomical structures
  - helps physicians detect, interpret, predict and plan
  - 2D (serial and projection) and 3D images
  - temporal and dynamic images

Multiplanar Imaging

1. Coronal: divides a region into anterior (front) and posterior (back) sections
2. Sagittal: divides a region into left and right sections
3. Axial: divides a region into superior (upper) and inferior (lower) sections



Medical Imaging Modalities

- What is a modality?
  - A *modality* is the manner in which an image is obtained from a specific kind of imaging device
- Medical imaging modalities are based on various physical phenomena
  - x-ray attenuation, the relaxation of magnetized hydrogen nuclei, sound reflections, or radioactive decay of radiopharmaceuticals

Structural vs. Functional Information

- Imaging modalities can be divided into two broad categories:
  - Structural (anatomical, morphological)
  - Functional (physiological and biochemical)
- Morphological
  - characteristics such as size, shape, texture, position, colour and composition of anatomical structures
- Physiological
  - characteristics such as flow, uptake, perfusion, metabolism, and chemistry

Dividing the Modalities

Morphological

Ultrasonography (US)

X-ray

Computed Tomography (CT)

Magnetic Resonance (MR)

Portal and video images

Physiological

Positron Emission Tomography (PET)

Single Photon Emission Computed Tomography (SPECT)

functional MR (fMR)

MR Spectroscopy (MRS)

perfusion MR (pMR)

perfusion CT (pCT)

X-ray

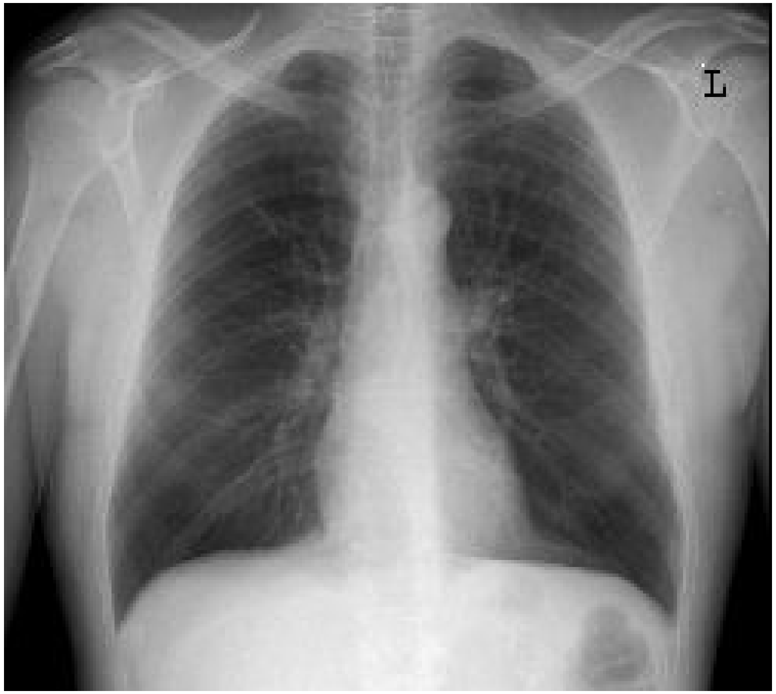
- X-rays were discovered by W. C. Röntgen in 1895.
  - The first form of medical imaging.
  - Radiography, computed radiography.
  - They penetrate most biological tissues with little attenuation, and thus provide a comparatively simple means to produce shadow, or projection, images of the human body.

X-ray

- An x-ray is a 2D projection image which measures the amount of ionizing radiation which is absorbed by tissues
  - As x-rays pass through an anatomical region they are attenuated (weakened) by the different density tissues they encounter
  - Bone is very dense and attenuates a great deal of x-rays, whereas soft-tissue is much less dense and attenuates far less x-ray energy

Examples of X-rays

Chest X-ray

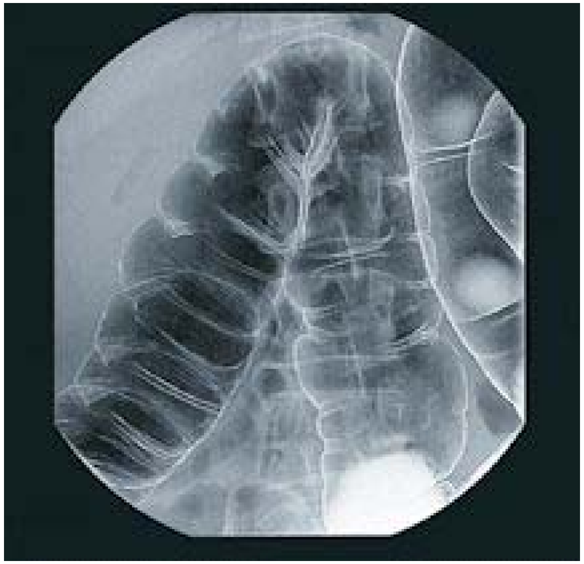


Hand X-ray

Fluoroscopy

- Fluorography
  - A form of dynamic x-ray imaging
- Uses contrast agents

  e.g. barium is used to highlight the gastrointestinal tract

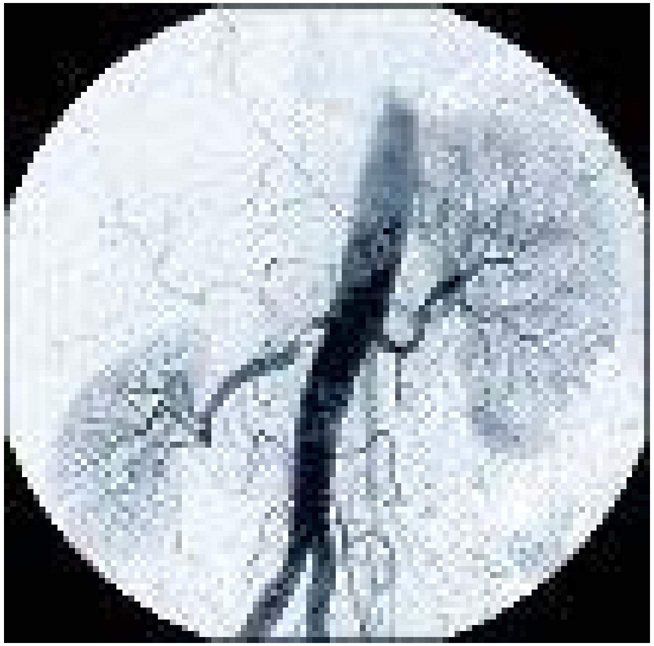


Angiography

- Angiography
  - A contrast-enhanced x-ray which uses a contrast agent to enhance vasculature
  - Specifically image and diagnose diseases of the blood vessels of the body
  - Involves the administration of intravenous contrast agent (i.e. iodine)
  - Digital Subtraction Angiography (DSA)

Examples of Angiograms

- Angiogram of the descending aorta, kidneys and renal arteries
- Angiogram of the carotid artery and arteries of the brain

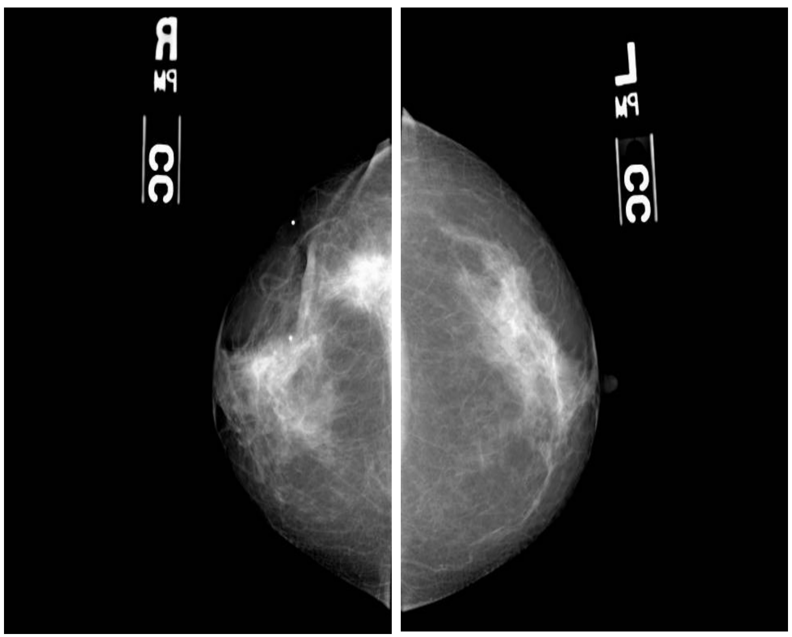


Mammography

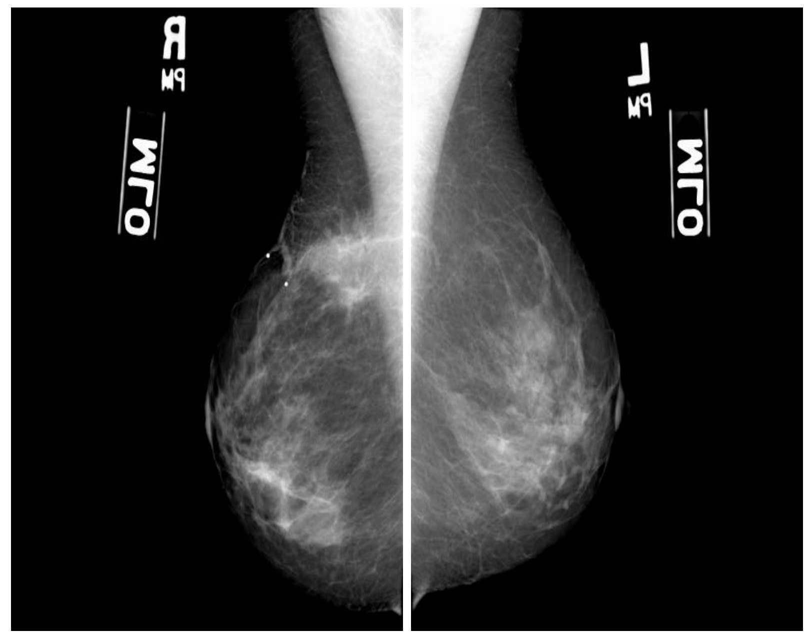
- Mammogram
  - A soft-tissue x-ray projection image of the three-dimensional structures of the breast
  - Requires compression of the breast

Examples of Mammograms

Left and right Cranio-Caudal Mammograms



Left and right Medio-Lateral Oblique Mammograms



Magnetic Resonance

- Magnetic resonance imaging is based on the measurement of magnetic radiofrequency (RF) signals from hydrogen molecules.
  - The contrast differences in MR signals are based primarily on the amount of hydrogen present in different tissues

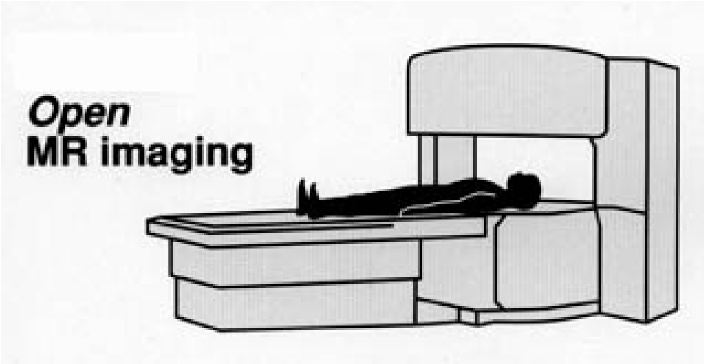
  e.g. bone and air, which contain a minimum amount of hydrogen, produce a low signal intensity, whilst fat and water produce high signal intensities

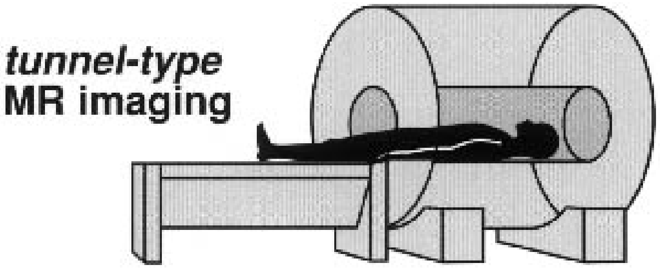
Magnetic Resonance

- Magnetic resonance imaging can be used to map:
  - perfusion
  - temperature
  - electrical conductivity
  - viscoelasticity
  - pressure
  - micromorphology

Magnetic Resonance

- Two types of MR scanner:
  - “Tunnel” using cylindrical magnets
  - “Open” using C-shaped magnets



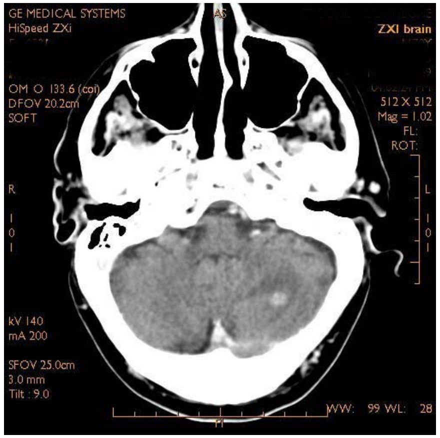


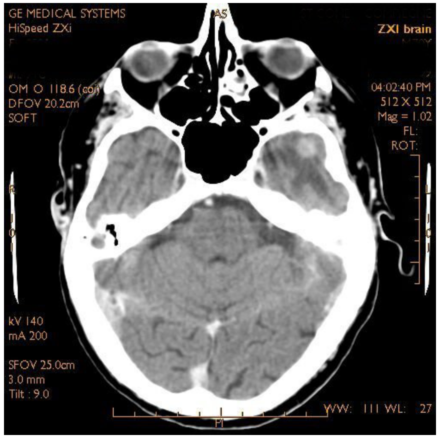
Types of MR

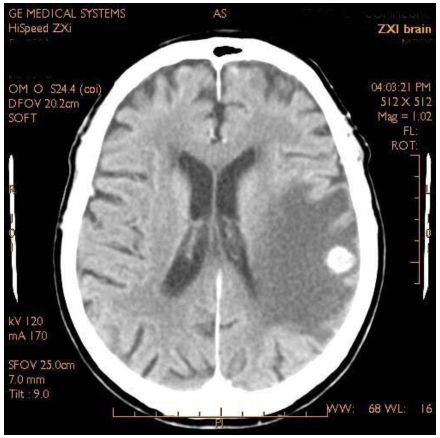
- MR Angiography (MRA):
  - uses paramagnetic contrast agents to enhance vasculature.
- functional MR (fMR):
  - used to measure blood flow
- MR Spectroscopy (MRS):
  - measures in-vivo chemicals within the body.
- Perfusion MR (pMR):
  - measurement of regional blood-flow
- MR Elastography (MRE):
  - measures the rheological (elastic) characteristics of tissue
- Diffusion MR (dMR)
  - reveal diffusion characteristics of water in tissues
- MR Fluoroscopy (MRF):
  - dynamic MR using a contrast agent

Examples of MR Images

- MR images of the brain

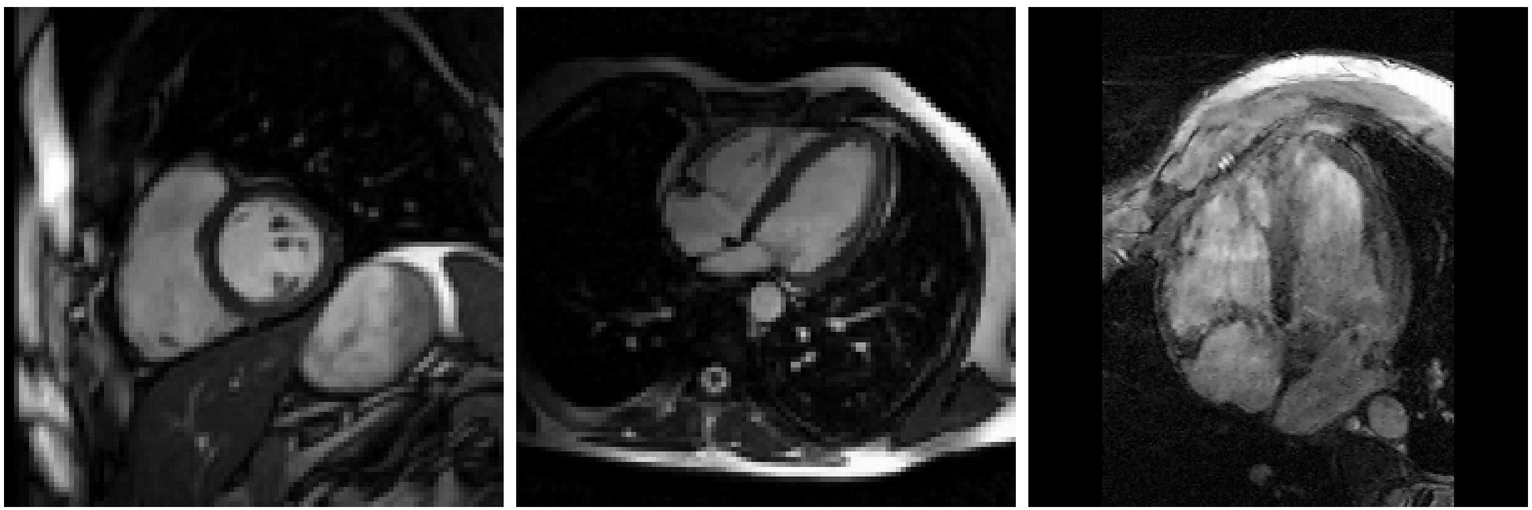






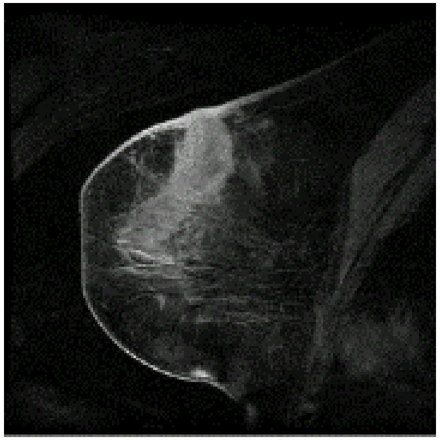
Examples of MR Images

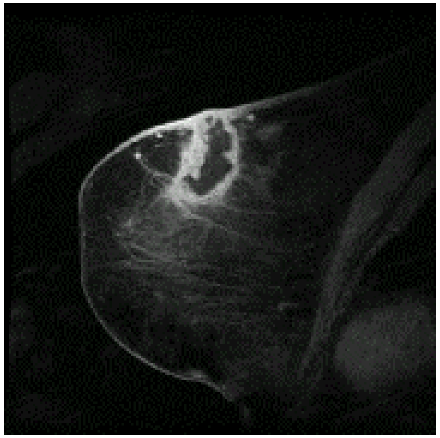
- Dynamic MR cardiac images showing the structure and motion of the heart



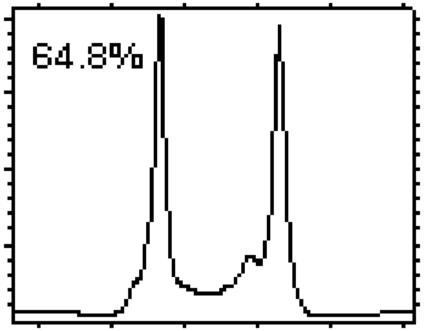
Examples of MR Images

- Contrast-enhanced MR images
  - Pre and post contrast enhanced MR images of the breast





- MR Spectroscopy



Examples of MR Images

- MR Fluoroscopy
- MR Angiography

Computed Tomography

- Computed Tomography (CT) images measure ionising radiation absorbed by tissue
  - Based on the principal of x-rays
  - Forms a 2D image by gathering many projection images at different angles
  - Transmission CT
  - A 3D CT image is composed of a series of 2D CT images

Types of Computed Tomography

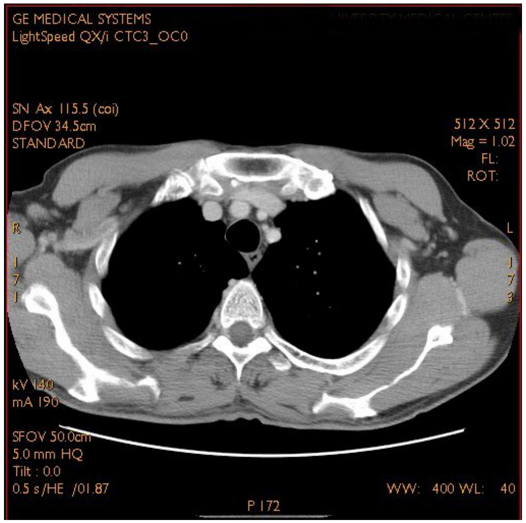
- CT Angiography (CTA)
  - uses contrast agents to enhance vasculature
- functional CT (fCT)
  - used to measure blood flow
- Perfusion CT (pCT)
  - measurement of regional blood-flow
- Electron-Beam CT (EBCT)
  - uses an electron beam which is electromagnetically guided to form a rotating fan-beam of x-ray.

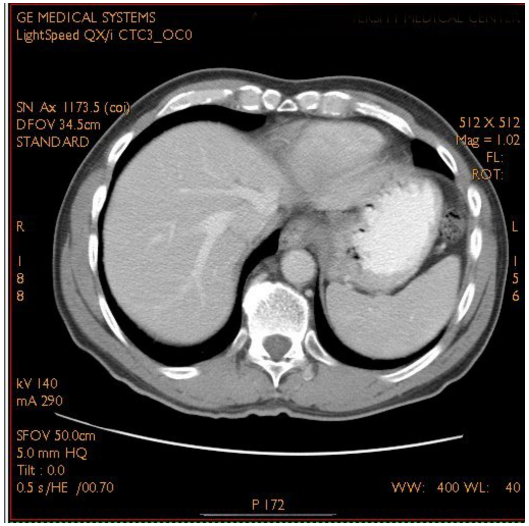
Spiral CT

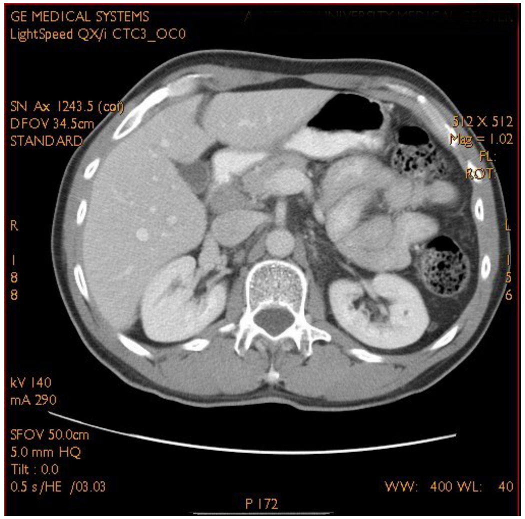
- Spiral (or helical) CT is the continuous acquisition of a 3D CT image
  - A recent development in CT scanning involves moving the patient at a continuous, regular speed of about 1 cms^{-1} through a rotating fan-shaped X-ray beam.
  - The resulting ‘corkscrew’ block of data is analysed by computer, giving improved 3D imaging and shorter examination times (approximately 30–40s).

Examples of CT Images

- CT Images of the chest and abdomen

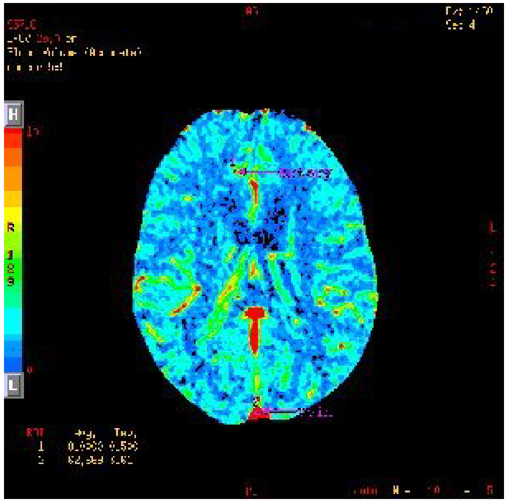


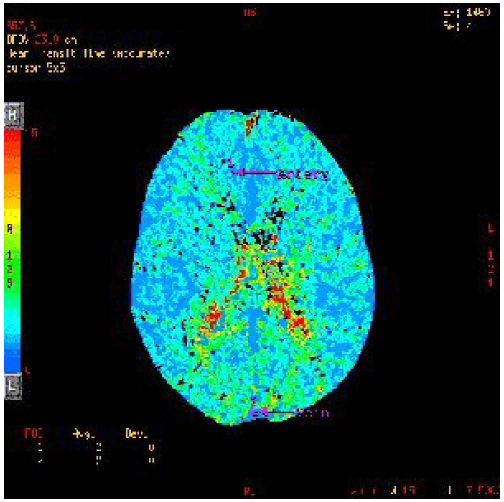




CT: Perfusion CT

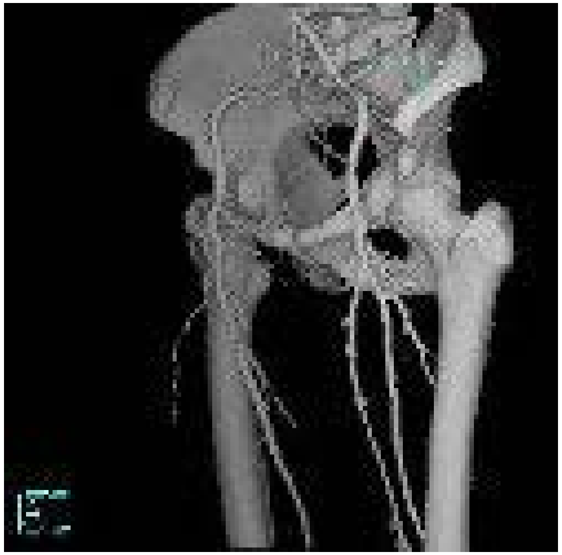
- Affords a quantitative measurement of regional blood flows
  - Involves sequential acquisition of images during administration of an iodinated contrast agent





CT: CT Angiography

- Derivative of CT used to enhance vascular structures using contrast agents



Electrical Impedance Tomography

- Electrical Impedance Tomography (EIT)
  - Measures low level bioelectric currents to produce real-time images of the electrical impedance properties of tissue.
  - Various tissues conduct electricity differently.
  - Can determine if the region-of-interest is normal tissue or cancerous tissue

Ultrasonography

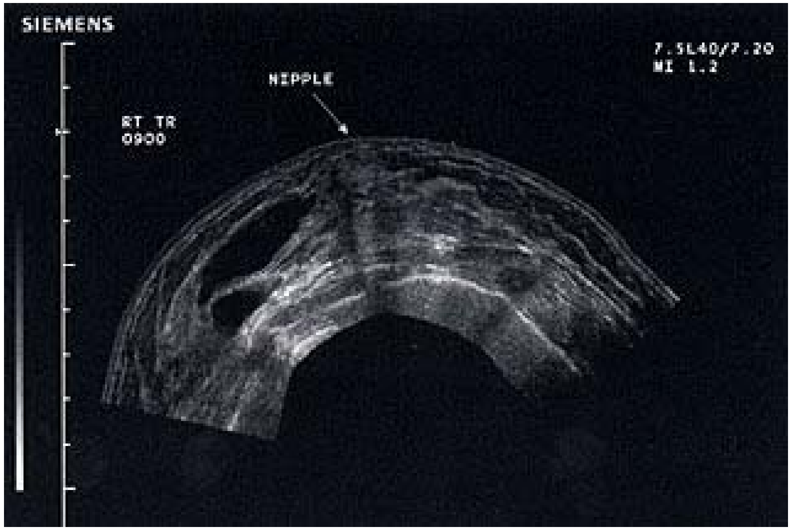
- Ultrasonography (US) generates dynamic images using high-frequency sound waves
  - Ultrasound imaging uses the reflective and refractive interactions of sound with tissue to image different tissues.
  - Continuous ultrasound beams are emitted from a transducer and reflected by discontinuities in the acoustic impedance of tissues.

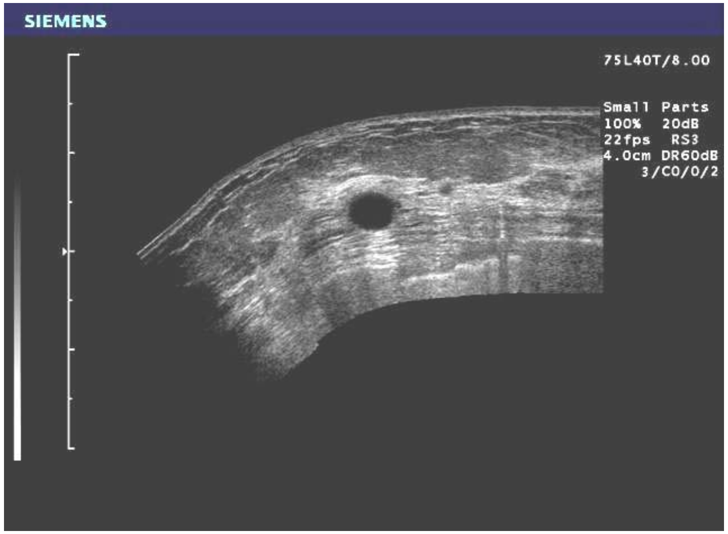
Ultrasonography

- Various forms of ultrasound:
  - Colour Doppler US: provides information on blood flow velocities
  - 3D US: provides a dynamic 3D ultrasound image

Examples of Ultrasound

- Ultrasound images of the breast





Nuclear Medicine

- Emission computed tomography
- Nuclear medicine measures in-vivo substances labelled with positron-emitting isotopes or radiopharmaceuticals
  - Planar Scintigraphy
  - Single Photon Emission Computed Tomography (SPECT)
  - Positron Emission Tomography (PET)
- Imaging signals are collected from the external detection of gamma emissions from the radionuclides within body tissues and used to generate images reflecting tissue biochemistry and physiology

Radiopharmaceuticals

- Specifically formulated to be collected temporarily in a specific part of the body
- Oral or intravenous introduction of very low-level radioactive chemicals into the body
  - radiopharmaceuticals, radionuclides, radioisotopes, radiotracers

    e.g. lung cancer imaging → inhalation of the radionuclide
- Radionuclide substances:
  - Synthesized radioactive substances

    e.g. Tc-99m (Technetium)
  - Radioactive forms of elements found naturally in the body

    e.g. Iodine

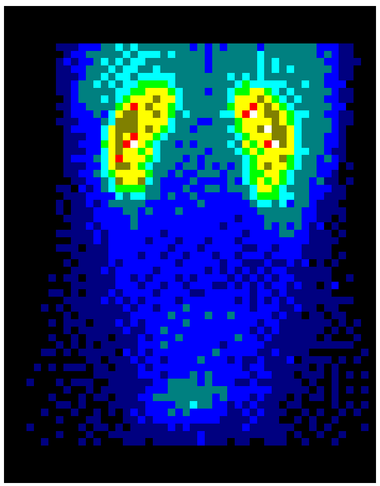
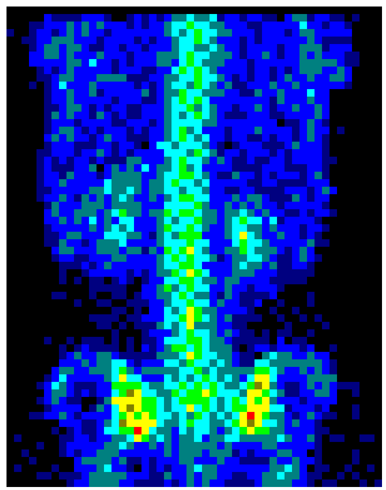
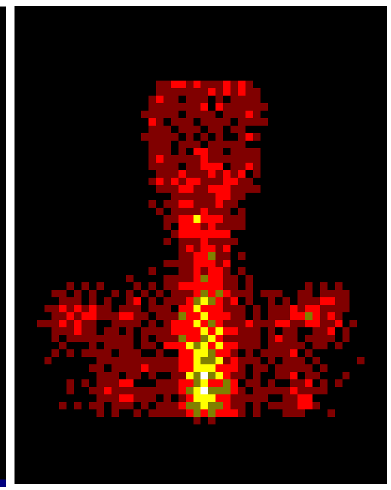
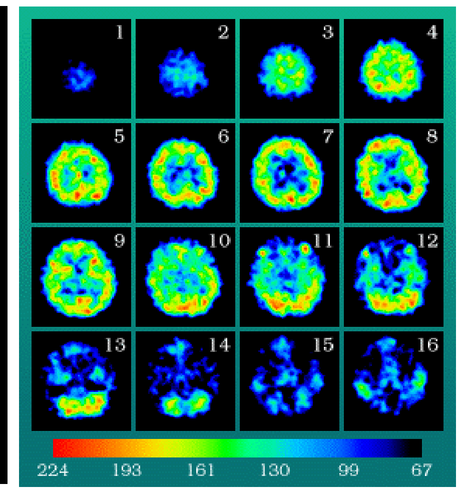
Radiopharmaceuticals

- The amount of uptake also has clinical importance.
  - Metabolically active regions, such as tumours or inflamed tissue, may show increased uptake, whereas tissues with reduced blood flow, such as heart tissue after a heart attack, may show decreased uptake.
- Radiopharmaceutical vs Region of application
  - thallium-201 to image heart muscle that may be inadequately perfused with blood; Gallium-67, technetium-99, or thallium-201 to image tumors; technetium-99 to image bone; xenon-133 to image lung perfusion and fluorine-18 to detect regions of increased metabolism.

Single Photon Emission Computed Tomography

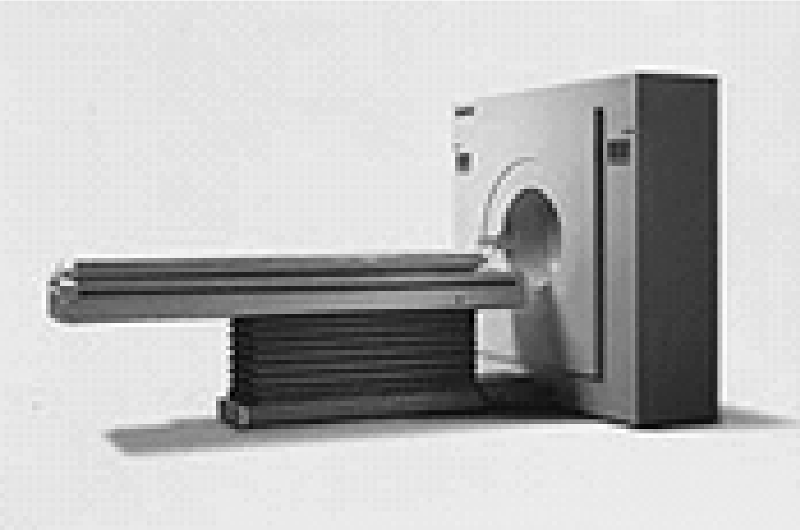
- Single Photon Emission Computed Tomography
  - This involves the detection of γ-rays (gamma-rays) emitted singly (hence 'single photon') from radionuclides such as 99m Tc and 201 Tl (thallium).
  - A gamma came … 60° around the patient, taking a … fixed locations.

Examples of SPECT

Kidneys	Bones	Head	Brain
			

Positron Emission Tomography

- Positron Emission Tomography uses isotopes in which two photons are produced
  - Radionuclides decay via emission of positrons
  - When a positron (β^+), ‘meets’ an electron (β^-), they ‘annihilate’ each other.

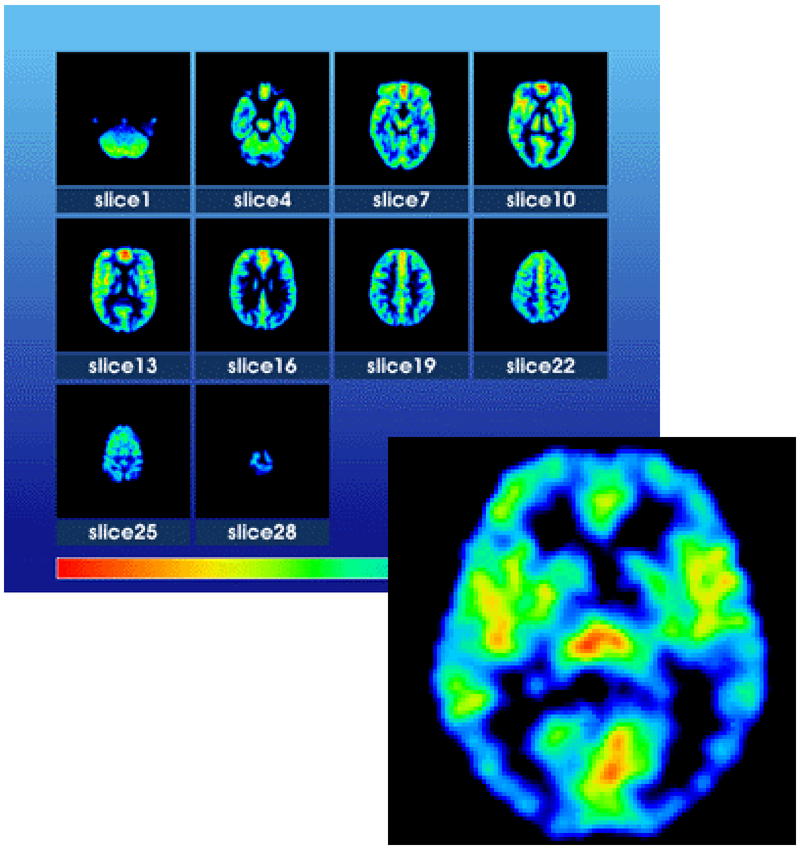


Positron Emission Tomography

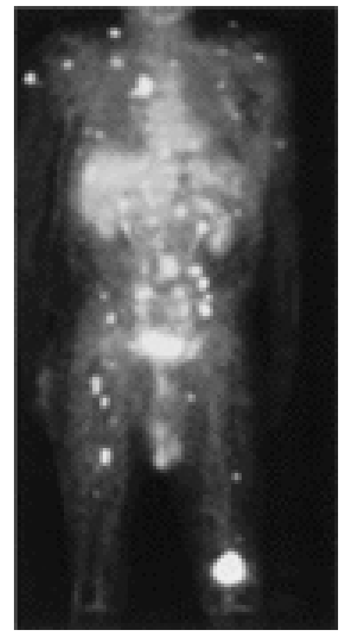
- Specific applications
  - Brain imaging: study of epilepsy, evaluation of stroke, evaluation of brain tumours
  - Oncologic imaging: differentiation between recurrent active tumour growth and necrotic (dead) soft-tissue masses in cancer treatment

Examples of PET

Brain PET

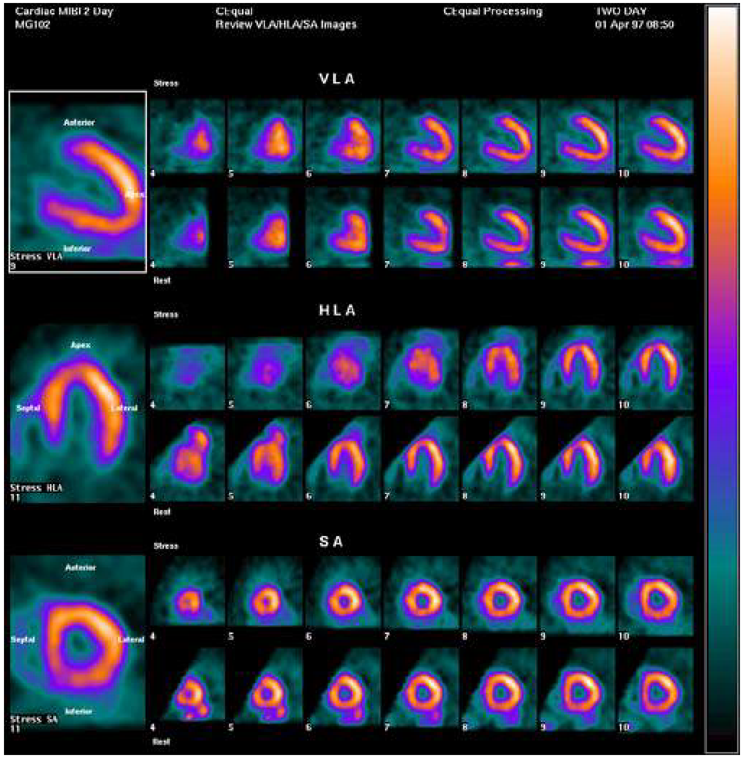


Whole-body PET



Examples of PET

Cardiac PET



Planar Scintigraphy

- Often used for bone scanning

Spatial Resolutions

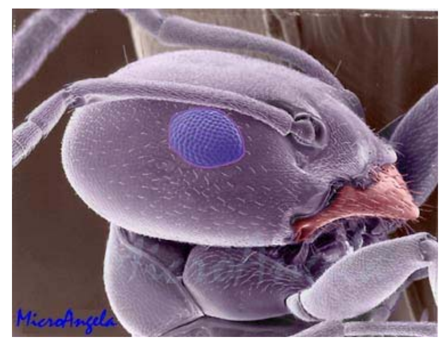
Modality	Image Size	Resolution
Computed Tomography	512x512	1-2mm
Magnetic Resonance	256x256	2mm
Mammograms	4096x2048	
Single Photon Emission Computed Tomography	128x128	10-20mm
Positron Emission Tomography	128x128	4-6mm

Spatial vs. Temporal Resolution

- High spatial = lower temporal resolution
- High temporal = low spatial resolution
- For example:

	Spatial Resolution	*Temporal Resolution*	*Image Size*
Dynamic MR	2-3mm	10-30 seconds	128x128
MR	<1.5mm	5 minutes	256x256

Biological Imaging



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Biological Imaging vs The Human Eye

The Human Eye	Biological Imaging
spatial resolution: ~ 0.1mm	spatial resolution: ~ 1nm
temporal resolution: ~ 100ms	temporal resolution: ~ 20ms
sensitivity: ~ 100 photons (510nm)	sensitivity: ~ 1 photon
wavelength range: 400-700nm	wavelength range: 10^{-13} – 1m

- Biological imaging can:
  - watch processes too rapid to perceive
  - see objects too small for the eye to see
  - see wavelengths too faint for the eye, or that the eye is not sensitive to
  - see inside living objects

Relative Sizes in Microns

- 1 metre = 1,000 millimeters (mm) = 1,000,000 microns (Greek letter mu, μ) = 1,000,000,000 nanometers (nm) = 10,000,000,000 angstroms (Å)
  - a nanometre is one-millionth of a millimetre
  - red blood cell = 7.0 microns in diameter
  - antibody protein = 0.003 microns long

Biological Imaging Modalities

- Electron Microscopy
  - Transmission Electron Microscopy (TEM)
  - Scanning Electron Microscopy (SEM)
- Light Microscopy
  - Fluorescence Microscopy (FM)
  - Confocal Microscopy (CM)
  - Multi-Photon Microscopy
- Near-Field Microscopy
  - Atomic Force Microscopy (AFM)
- Autoradiography

Electron Microscopy

- Why use electrons instead of photons for microscopy?
  - The lower wavelength of electrons allows a much higher limit of resolution.
  - Uses a beam of electrons instead of light.
  - Views objects smaller than 2μm
  - 10,000 to 100,000 times magnification
  - The resolution of a compound light microscope is 0.2 micrometers; maximum magnification is 2000x.

Transmission Electron Microscopy

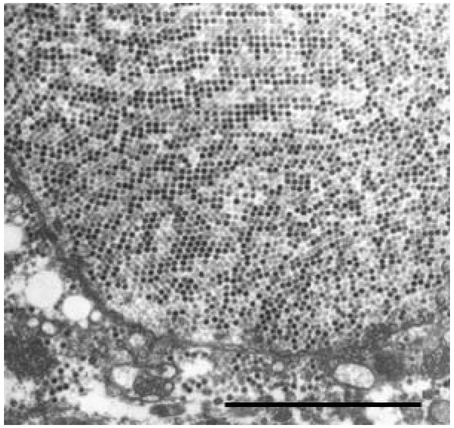
- In *Transmission Electron Microscopy* (TEM) the electrons traverse the sample producing images of the specimen that are a projection of the whole sample volume, not only its surface
  - A complete 3D reconstruction is possible
  - Structures which are less than one nanometre apart can be resolved

Transmission Electron Microscopy

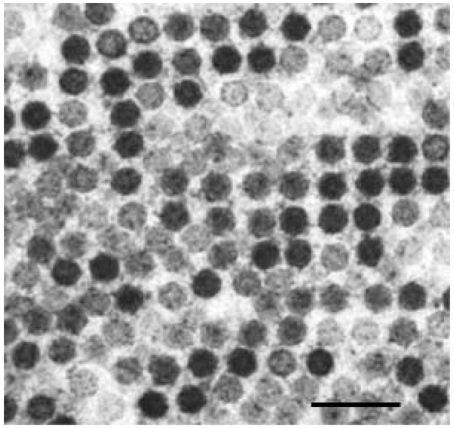
- This type of microscopy uses an electron beam instead of light to visualize biological tissues.
  - The beam is focused with electromagnetic lenses and goes through a thin section of the specimen.
  - It is then expanded thousands of times by another series of lenses until it reaches a photographic plate or a light-emitting screen to form an image.
- The primary contrast mechanism used in electron microscopy arises from scattering of electrons.
  - Scattered electrons fail to reach the image plane and reveal the position of electron dense material.
  - There is no absorption of electrons.

TEM Image Examples

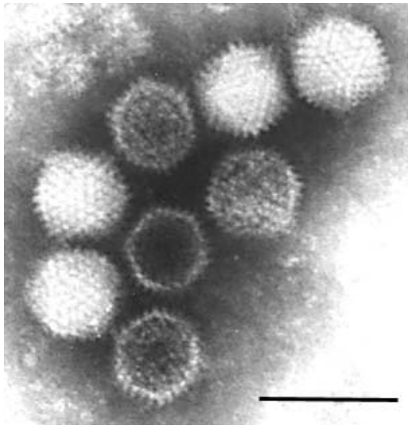
- Adenoviridae virus



Bar = 2μm



Bar = 250nm



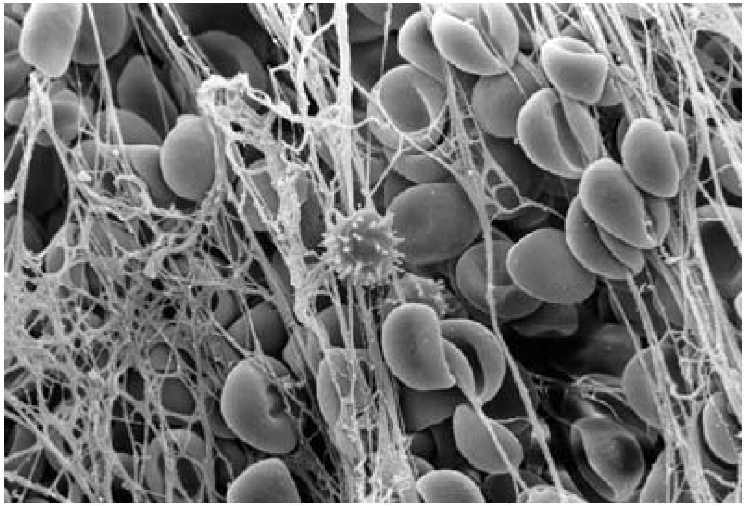
Bar = 100nm

Scanning Electron Microscopy

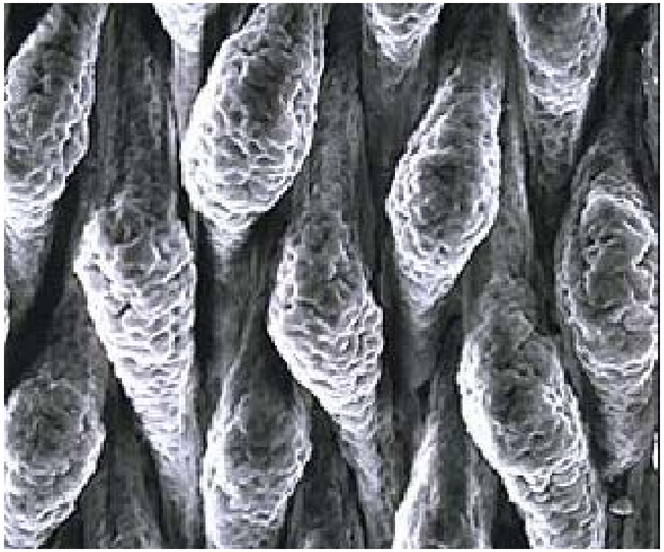
- *Scanning Electron Microscopy* (SEM) uses an electron microscope to visualize surface features of a biological tissue.
  - The incident electron beam causes electrons to be emitted from the surface of the subject and it is the pattern of this electron emission that forms the image.
  - An electron beam is focused to a tiny spot on the surface of the specimen. When the electron beam hits the sample, the amount of reflected electrons or the amount of secondarily emitted electrons from the specimen is measured.

SEM Image Examples

A blood clot viewed with a scanning electron microscope. Strands of the protein fibrin are deposited to form a mesh across the wound, trapping red blood corpuscles and a single white blood cell (centre).



The inner surface of the small intestine is covered with millions of tiny folds known as the intestinal villi.



Scanning Probe Microscopy

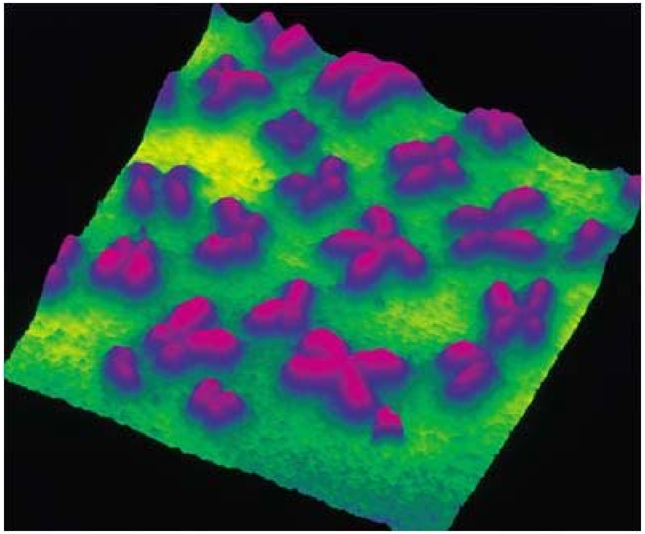
- Scanning Probe Microscopy (SPM) is the generic name for a family of local probe techniques that operates at the nanoscale
  - A scanning probe microscope scans the surface of a structure.
  - Atomic Force Microscopy (ATM)
  - Scanning Tunnelling Microscopy (STM)
  - Scanning Near-Field Optical Microscopy (SNOM)
  - Shear-force Microscopy (ShFM)

Atomic Force Microscopy

- An Atomic Force Microscope provides true 3D images of a surface at nanometre resolution.
  - It does this by measuring the nanonewton-sized forces (a newton is a unit of force) between the surface and the ultra-sharp tip of a force-sensing cantilever (this may be thought of as a soft spring).
  - Measured at each point of a two-dimensional array, this information builds up to yield a three-dimensional image.
  - The image is colour-coded according to the height of the various regions

SPM Image Examples

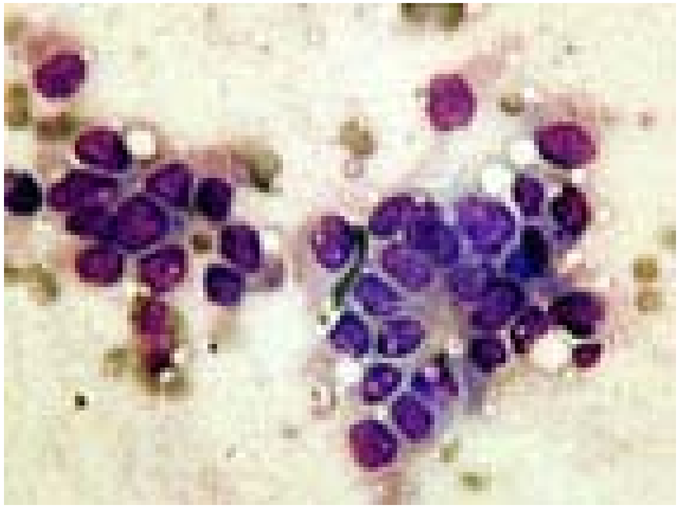
Human chromosomes seen here during nuclear division.

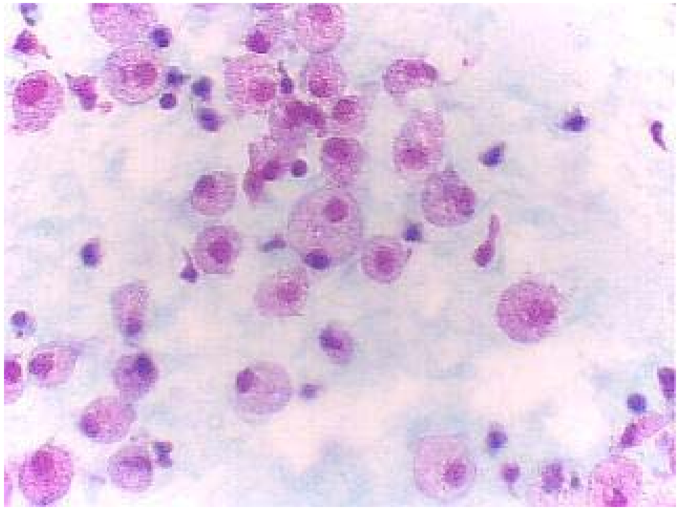


A human chromosome (chromosome no. 1) as seen at the metaphase stage of division. The chromosome has already replicated into two parts (chromatids), which are joined at a point near the centre of the chromosome (the centromere).

Light Microscopy

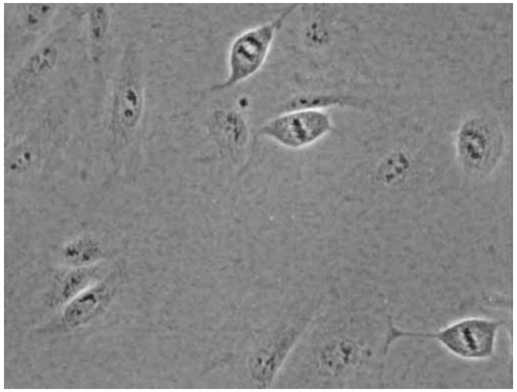
- Histological (tissue) images
  - Certain components of the tissue, such as proteins, can be viewed by staining them with specific dyes.





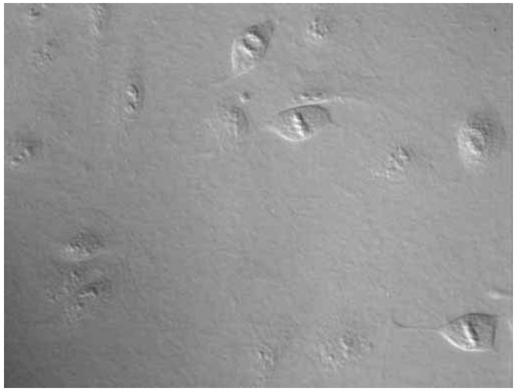
Phase-Contrast Microscopy

- Because biological samples are a complicated mixture of different materials, light passes through different parts of the sample at different speeds.
  - During *phase contrast microscopy* the phase differences are converted to intensity differences (contrast-enhanced).



Differential Interference Contrast Microscopy

- Differential Interference Contrast (DIC) (or Normarski) Microscopy recombines incident and refracted light waves from a single source at the plane of the image.
  - The interference effects between the incident and refracted light enhance small differences in contrast.
  - Produces images that have a shadowed relief.

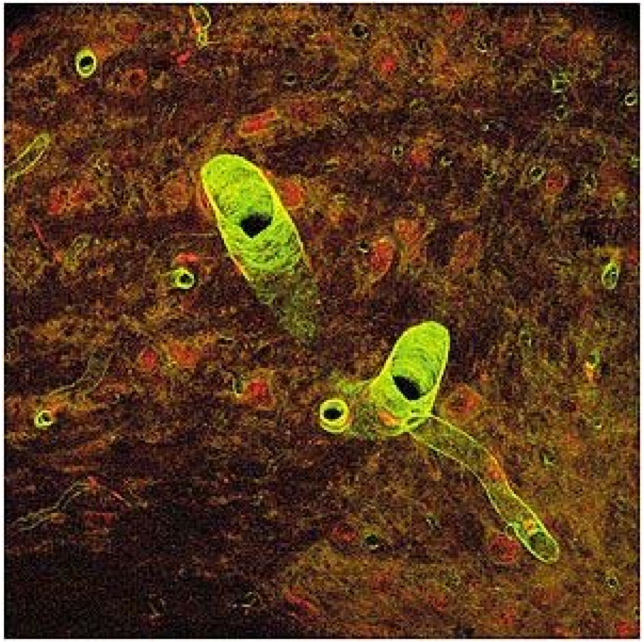


Confocal Microscopy

- A scanning Confocal Microscope makes optical sections through a tissue specimen.
  - Uses a highly coherent light source such as a laser
  - The laser beam is used to scan a pin point of light at a fixed depth within the specimen and rejects the out-of-focus information from other planes, thus providing a crisp, high-resolution image at that depth.
  - Images light from a thin confocal slice rather than from the entire specimen.
  - A 3D reconstruction is produced when all these layers are put together to provide a sharp two-dimensional representation of the three-dimensional information

CM Image Examples

- Brainstem blood vessels



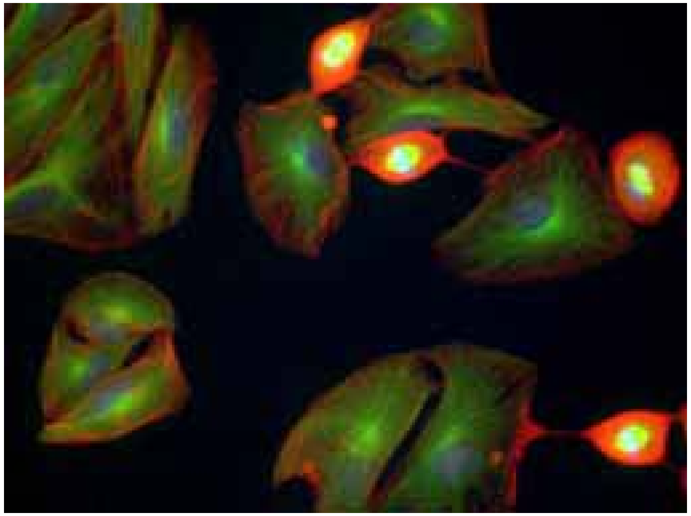
- The green tube-like structures are small arteries called arterioles running through the brainstem.
- Branching of the blood vessels can be glimpsed to the lower right where two smaller arterioles join the larger vessel. The green stain highlights the actin protein in the muscle cells of the arteriole walls. Actin is one of the proteins that causes the muscle to contract.
- Cells called pericytes, which act as support cells for the blood vessel walls, are stained red.

Fluorescence Microscopy

- In fluorescence microscopy specimens are stained with fluorochromes/fluorochrome complexes.
  - Light of high energy or short wavelengths is then used to excite molecules within the specimen or dye molecules attached to it.
  - These excited molecules emit light of different wavelengths, often of brilliant colours.
  - This emission is known as *fluorescence* if the emission takes place very shortly after the absorption of light

Fluorescence Microscopy

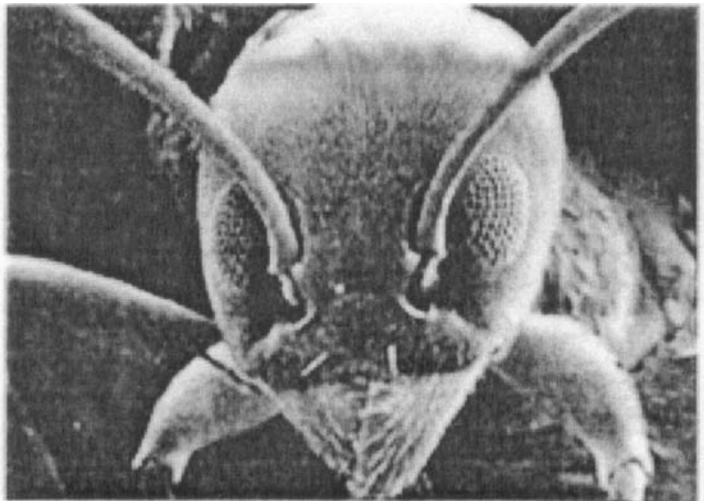
– Dyes such as fluoescein, rhodamine
– Viewing specimens that absorb visible light, such as histological stains.



Light Microscope vs. SEM

- Light Microscope
  – Resolution and depth of focus are greatly limited

- SEM
  – Resolution of stuctural detail and depth of field are improved



A Comparison of Microscopes

Microscope	*Resolution*	*Magnification*
Light	1.0μm	10x-10^3x
SEM	2nm	10x-10^6x
TEM	0.1nm	10^8x
SPM	01-3.0nm	10^8x

Autoradiography

- A 2D image of the patterns of distribution of radiation emitted by a specimen

  e.g. DNA sequencing gel autoradiograph