Electron Microscopy

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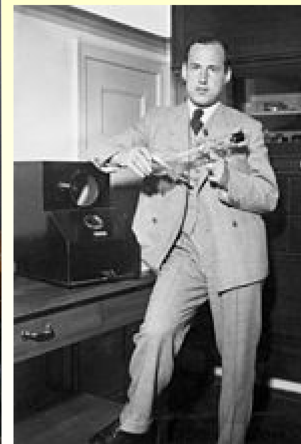
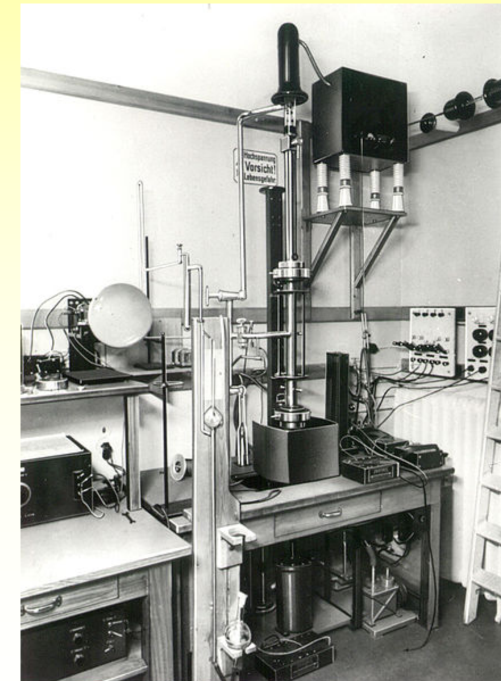
Introduction and History

- Electron microscopes are scientific instruments that use a beam of energetic electrons to examine objects on a very fine scale.
- Electron microscopes were developed due to the limitations of Light Microscopes which are limited by the physics of light.
- In the early 1930's this theoretical limit had been reached and there was a scientific desire to see the fine details of the interior structures of organic cells (nucleus, mitochondria...etc.).
- This required 10,000x plus magnification which was not possible using current optical microscopes.

- **The transmission electron microscope (TEM)** was the first type of Electron Microscope to be developed and is patterned exactly on the light transmission microscope except that a focused beam of electrons is used instead of light to "see through" the specimen. It was developed by Max Knoll and Ernst Ruska in Germany in 1931.

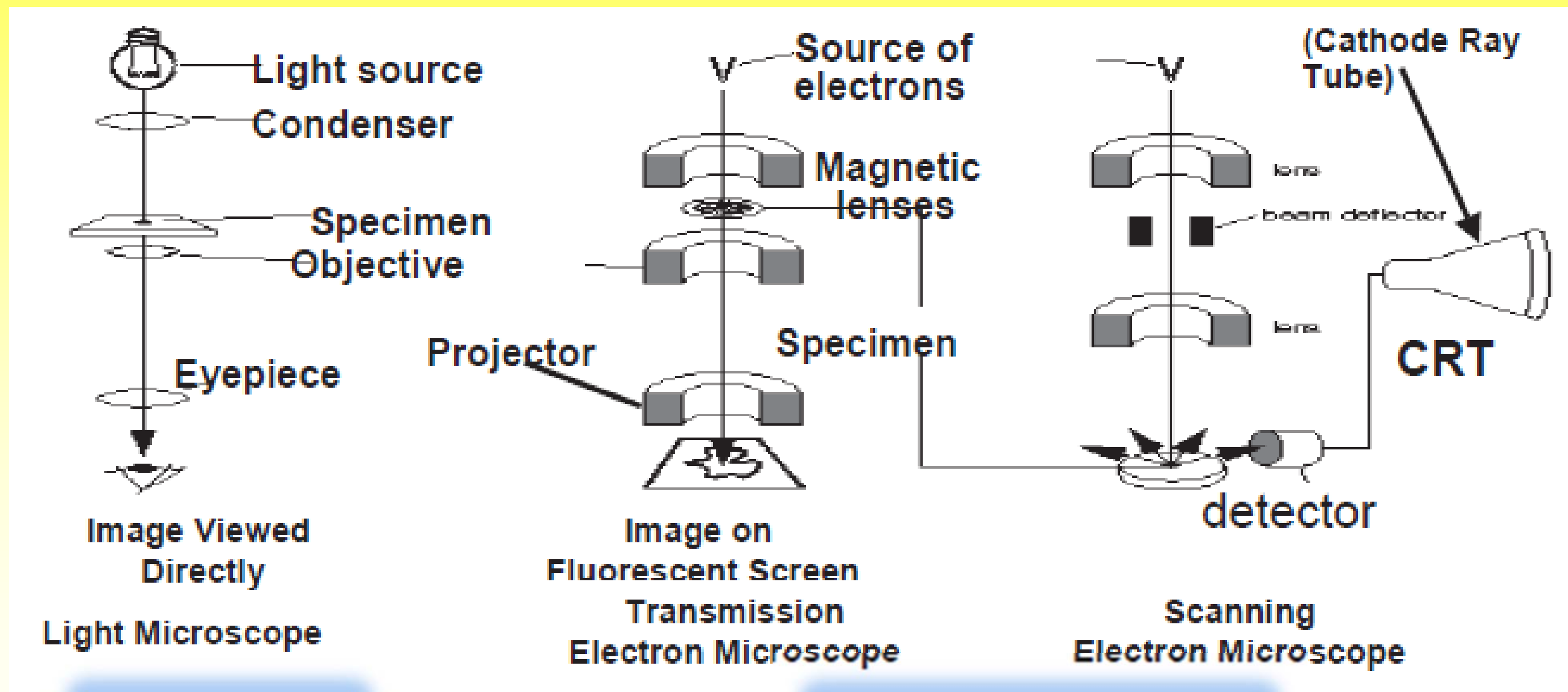
- The first **scanning electron microscope (SEM)** debuted in 1938 (Von Ardenne) with the first commercial instruments around 1965. Its late development was due to the electronics involved in "scanning" the beam of electrons across the sample.





Comparison of OM,TEM and SEM

Principal features of an optical microscope, a transmission electron microscope and a scanning electron microscope, drawn to emphasize the similarities of overall design.

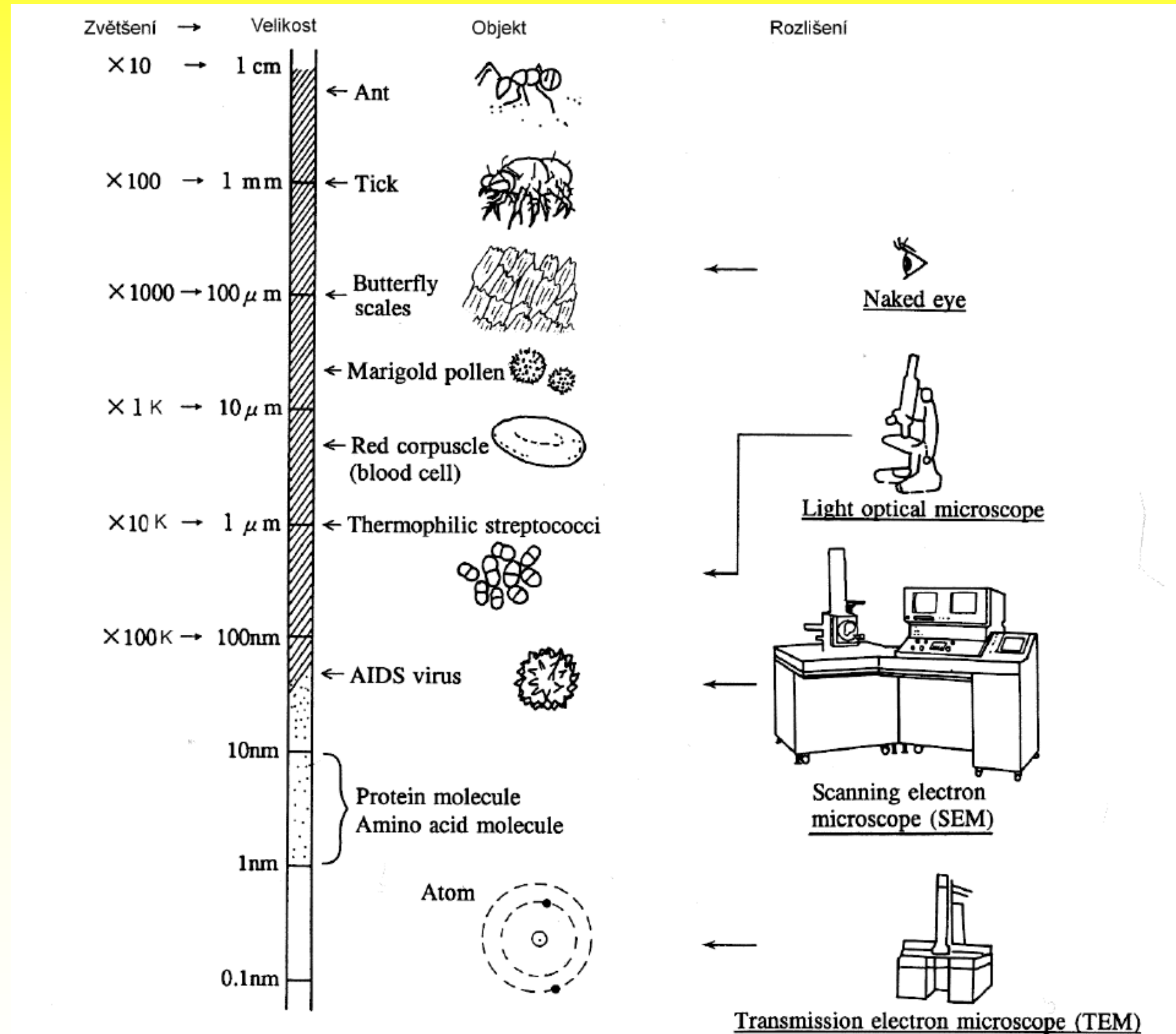


Light + glass lens

Electron + electomagnetic lens

The properties of the optical system and the radiation used depends what magnification and resolution will be achieved.

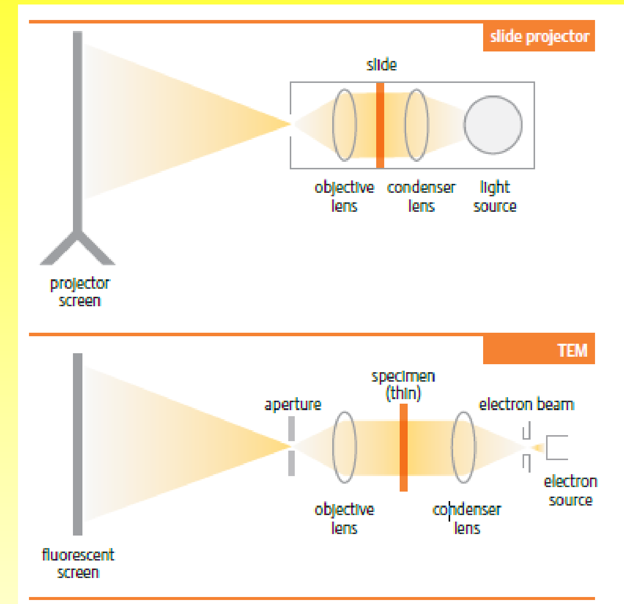
Scale and Microscopy Techniques



Transmission electron microscopy

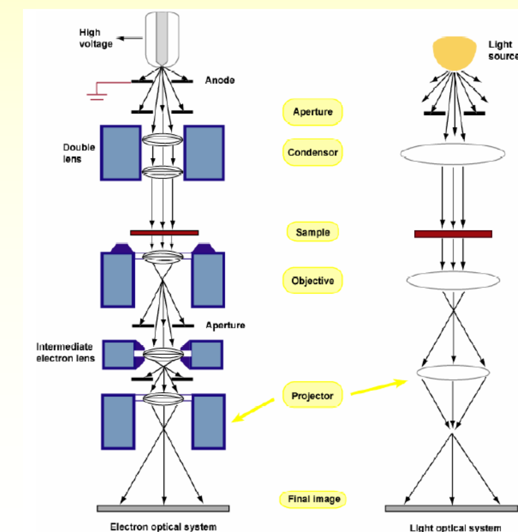
The transmission electron microscope can be compared with a slide projector.

- In a slide projector light from a light source is made into a parallel beam by the condenser lens; this passes through the slide (object) and is then focused as an enlarged image onto the screen by the objective lens.
- In the electron microscope, the light source is replaced by an electron source, the glass lenses are replaced by magnetic lenses, and the projection screen is replaced by a fluorescent screen, which emits light when struck by electrons, or, more frequently in modern instruments, an electronic imaging device such as a CCD (charge-coupled device) camera.

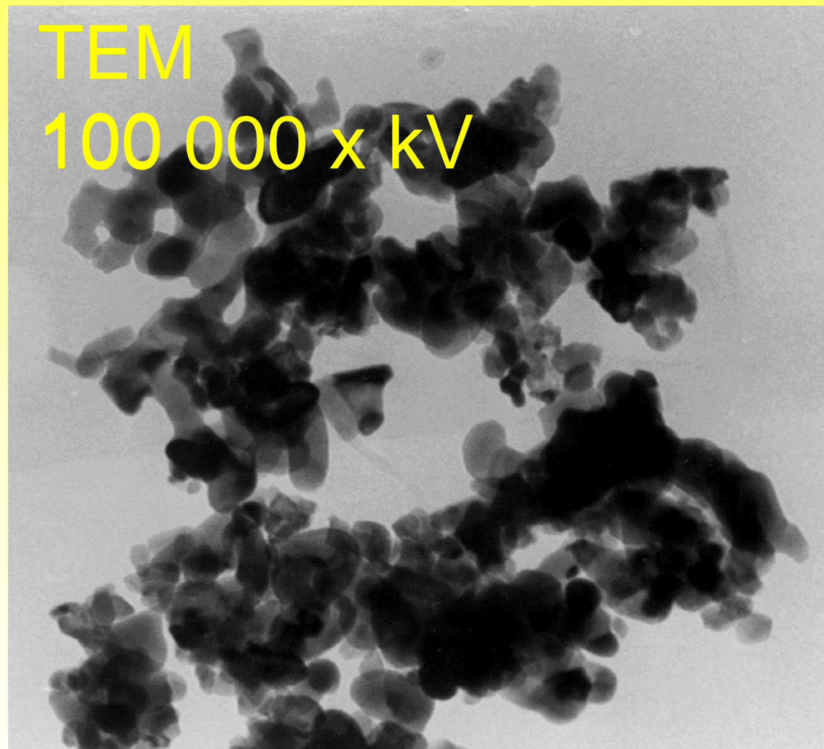


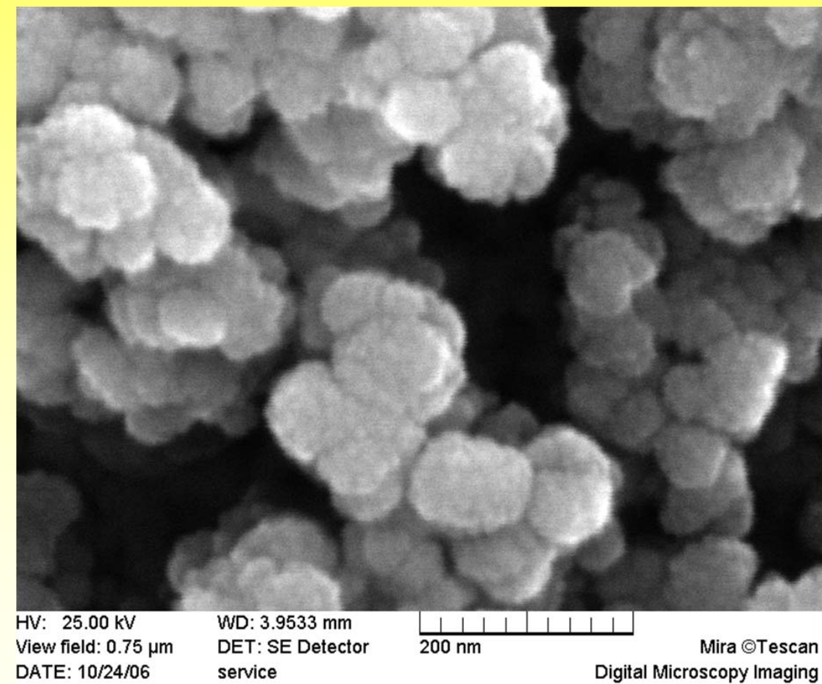
The whole trajectory from source to screen is under vacuum and the specimen (object) has to be very thin to allow the electrons to travel through it - light elements < 1 µm, heavy elements < 0.1 µm, lattice imaginig ~ 10 nm.
Not all specimens can be made thin enough for the TEM.
Alternatively, if we want to look at the surface of the specimen, rather than a projection through it, we use a scanning electron or ion microscope.

Comparison of OM and TEM



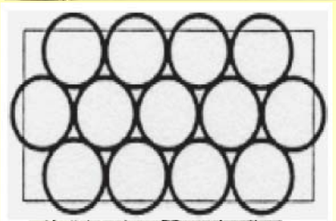
Comparison of TEM and SEM pictures

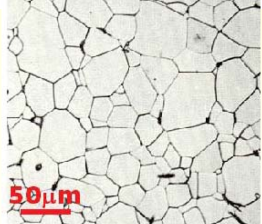


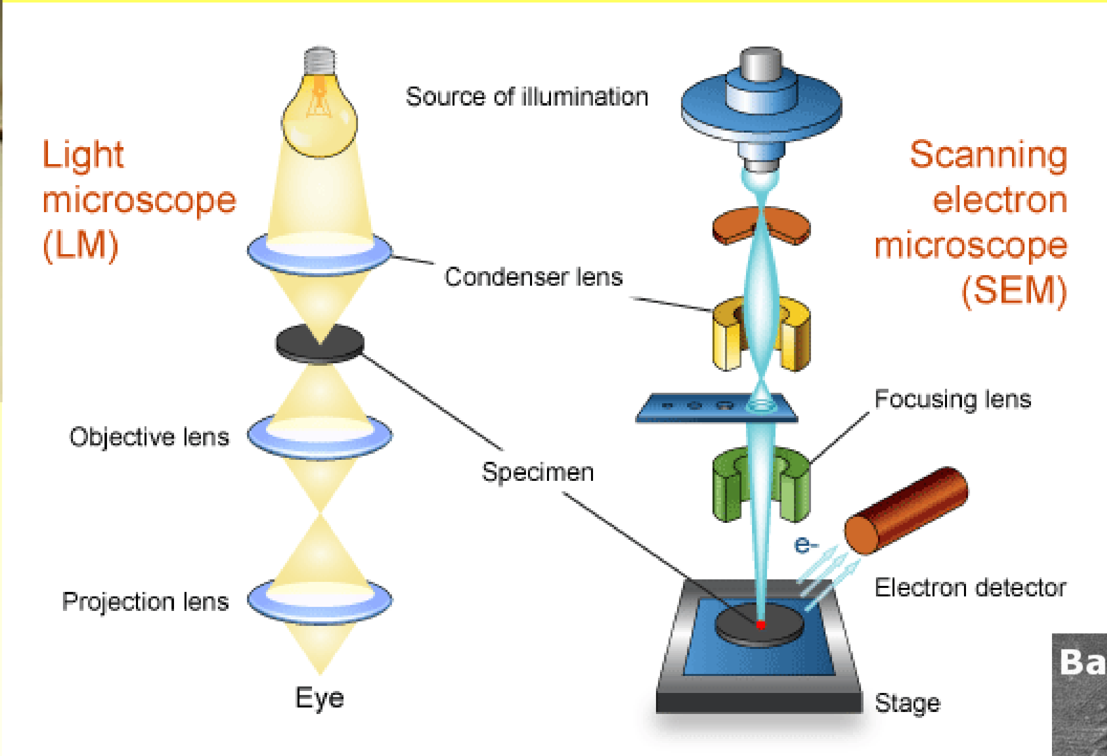


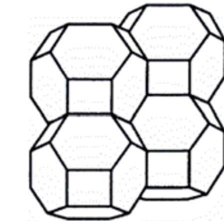
Scanning electron microscopy

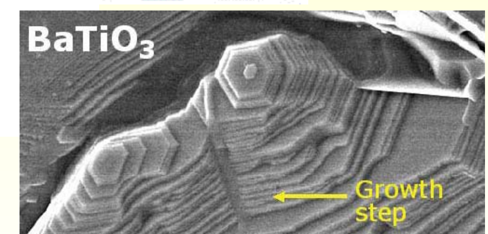
Comparison of OM and SEM











The scanning electron microscope (SEM) provides the competent user with **an advantage over the light microscope (LM)** in three key areas:

- ***Resolution at high magnification*.**

Resolution can be defined as the least distance between two closely opposed points, at which they may be recognized as two separate entities.
The best resolution possible in a **LM is about 200 nm** whereas a typical **SEM** has a resolution of **better than 10 nm** (typically 5 nm).

- ***Depth of field***

i.e. the height of a specimen that appears in focus in an image - **more than 300 times** the depth of field compared to the LM.
This means that great topographical detail can be obtained. For many users, the three dimensional (3D) appearance of the specimen image, is the most valuable feature of the SEM. This is because such images, even at low magnifications, can provide much more information about a specimen than is available using the LM.

- ***Microanalysis***

i.e. the **analysis of sample composition** including information about chemical composition, as well as crystallographic, magnetic and electrical characteristics.

	Mag	Depth of Field	Resolution
OM:	4x – 1400x	0.5mm	~ 0.2mm
SEM:	10x – 500Kx	30mm	1.5nm

One drawback to the use of the SEM

is that it operates **under vacuum** and in many SEMs the samples must be rendered conductive to be viewed. This is often achieved by **coating** with a very thin layer (~10 nm) of metal or carbon.
This coating is applied for two main reasons:

(1) Non conductive specimens are often coated to **reduce surface charging** that can **block the path of SE** and cause distortion of signal level and image form;

(2) **Low atomic number** (Z) specimens (e.g. biological samples) are coated to provide a surface layer that produces a **higher SE** yield than the specimen material.

It is important to leave the sample uncoated (in its natural state) if compositional information is required because the practice of coating samples with metals obscures this. If the sample is non-conductive then it can be coated with carbon (a low atomic number material) which will enhance conductivity without obscuring the compositional detail from below.

However, there are a number of different types of SEMs which all have specific purposes, often associated with additional pieces of equipment like specialised stages or collectors. Some of these do not require dry or conductive samples. They include the following:

- Low vacuum scanning electron microscopy (LVSEM)
- Using cryo on a scanning electron microscopy (Cryo-SEM)
- Environmental scanning electron microscope (ESEM)
- Focused ion beam (FIB) technology
- E-beam lithography (EBL)

History

In **1938**, von Ardenne added **scan coils** to a transmission electron microscope (TEM), producing a scanning transmission electron microscope (STEM).

In **1942**, Zworykin and his team developed the **first scanning electron microscope** to employ secondary electron detection.

He and his team recognized that secondary electron emission could be used to generate an image showing the topographic contrast of a specimen. The collector was biased to +50 volts to capture the secondary electrons, and the voltage drop across a connected resistor generated the image. Although the initial spatial resolutions were poor, approximately 200 nm, Zworykin and his colleagues reduced the beam spot size and obtained images with a resolution of 50 nm.

In the late **1940s and early 1950s**, researchers Oatley and McMullin introduced several notable **improvements** to the scanning electron microscope, including the electromagnetic lens, the stigmator, and signal amplification. By attaching the scintillator directly to the face of the photomultiplier, Everhart and Thornley greatly improved the signal-to-noise ratios.

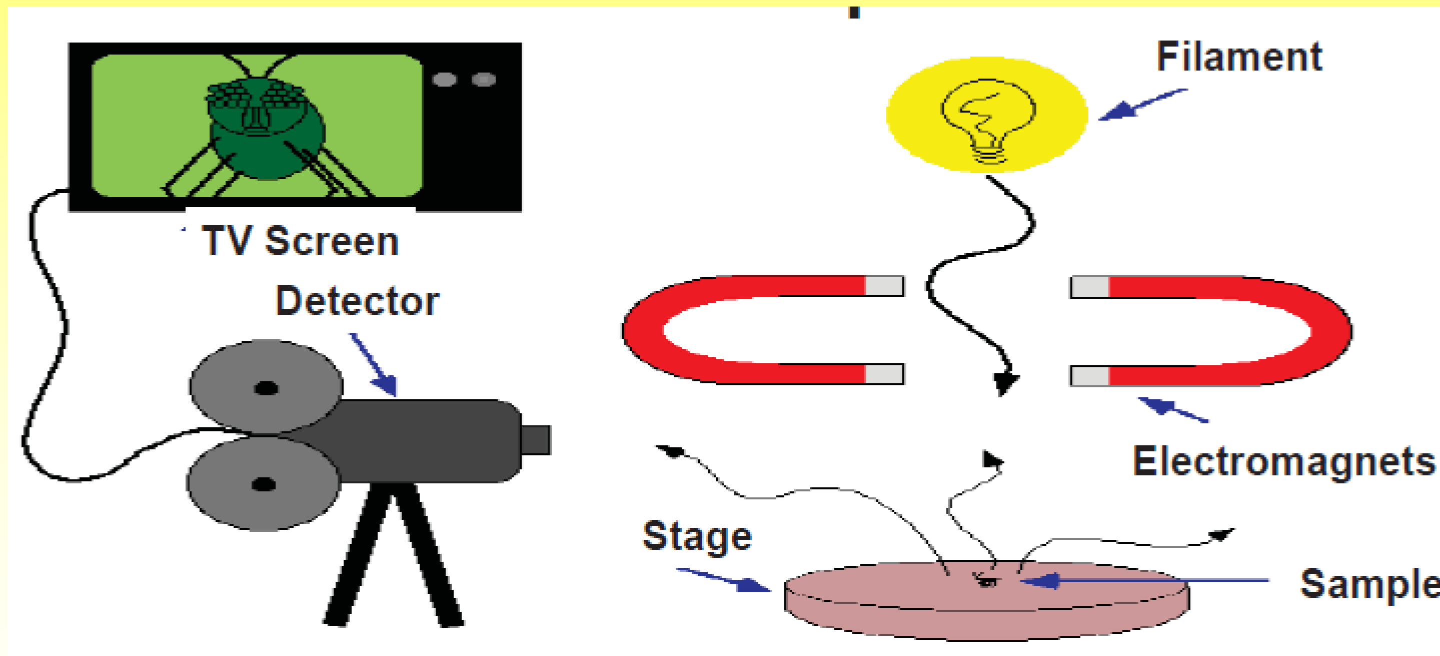
1960s were the development of the **LaB_6** (Lanthanum-hexaboride) electron cathode and the revival of the field emission tip electron source.

The LaB_6 improves resolution via a high-brightness electron gun. Yielding even higher resolution, the field emission tip electron source produces current densities that measure thousands of amps per cm^2. Consequently, today's commercial SEMs can obtain resolutions of about 10-20Å.

In **1968**, Fitzgerald demonstrated the addition of an **energy-dispersive x-ray detector** to an SEM, moving the SEM into the analytical probe arena.

SEM Layout

In simplest terms, an SEM is really nothing more than a television. We use a filament to get electrons, magnets to move them around, and a detector acts like a camera to produce an image.



SEM Layout

Princip:

The SEM uses a beam of high energy electrons generated by an electron gun, processed by magnetic lenses, focused at the specimen surface and systematically scanned (rastered) across the surface of a specimen.

Image formation:

Unlike the light in a light microscope (LM), the electrons in a scanning electron microscope (SEM) never form a real image of the sample. The ***SEM image*** is in the form of a serial data stream i.e. it is an ***electronic image***, which is a ***reflection of differences*** (e.g. topographical or compositional) in the sample.

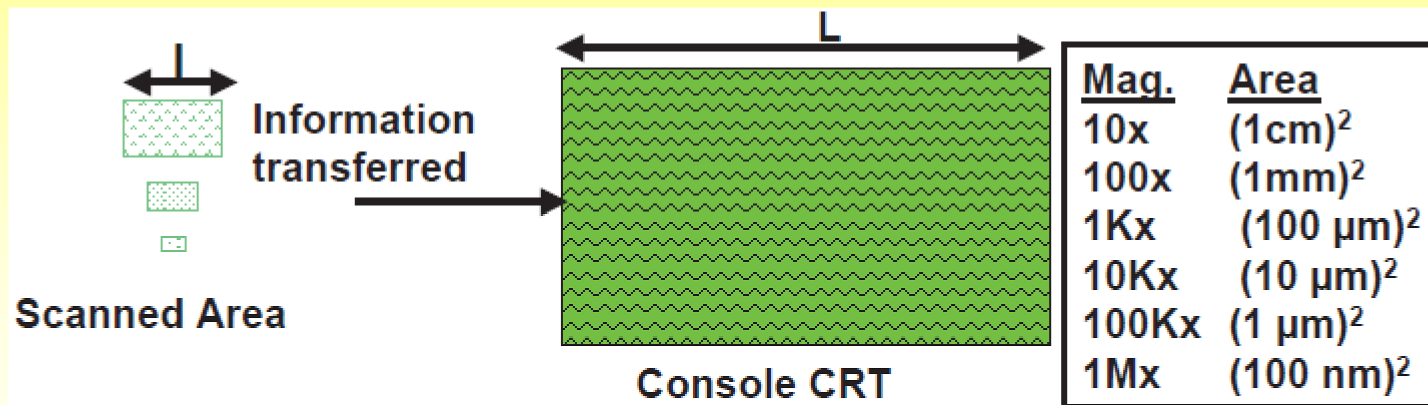
The formation of an image requires a scanning system (scan coils). The scanning system uses two pairs of electromagnetic deflection coils that scan the beam along a line then displace the line position to the next scan so that a rectangular raster is generated both on the specimen and on the viewing screen. The first pair of scan coils bends the beam off the optical axis of the microscope and the second pair bends the beam back onto the axis at the pivot point of the scan.

Signals generated from the specimen are collected by an **electron detector**,
converted to photons via a **scintillator**,
amplified in a **photomultiplier**,
and converted to electrical signals and used to modulate the intensity of the image on the viewing screen.

Magnification

- An image is obtained by taking the signal from the sample and transferring it to a CRT screen.
- **Increased magnification** is produced by **decreasing the size of the area scanned.**
- Magnification is determined by taking the ratio of the lengths of the scans:

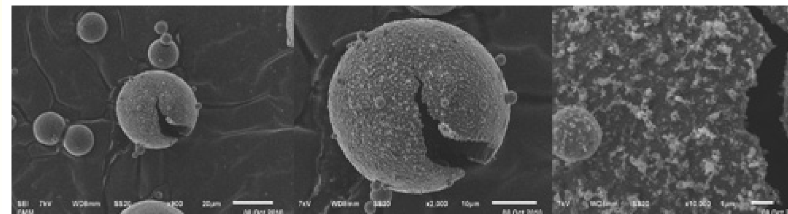
Mag. = L/l



Mag.	Area
10x	$(1cm)^2$
100x	$(1mm)^2$
1Kx	$(100\ \mu m)^2$
10Kx	$(10\ \mu m)^2$
100Kx	$(1\ \mu m)^2$
1Mx	$(100\ nm)^2$

Low magnification

High magnification



Resolution

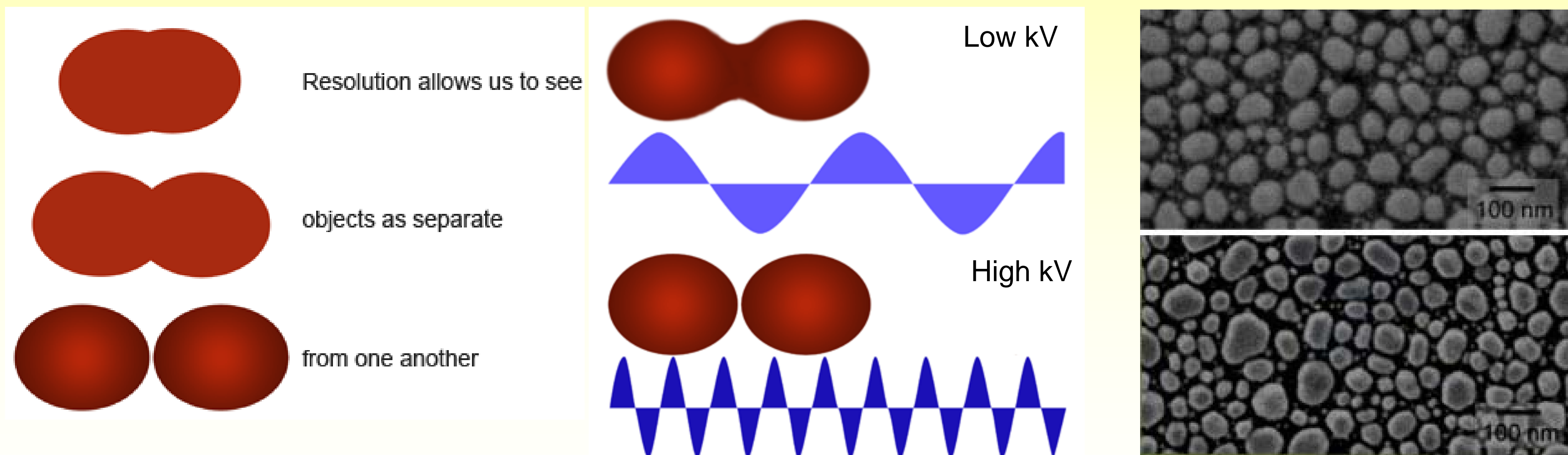
- Resolution is the ability to resolve two closely spaced points.

While you may have to be at a high magnification to see small features, resolution is NOT the same as magnification.

- Resolution is **dependent on wavelength** of the beam we use to see the material.
- Wavelengths of electron beams generated at **different accelerating voltages**
- One way to improve resolution is by reducing the size of the electron beam that strikes the sample.

We can also improve the resolution by:

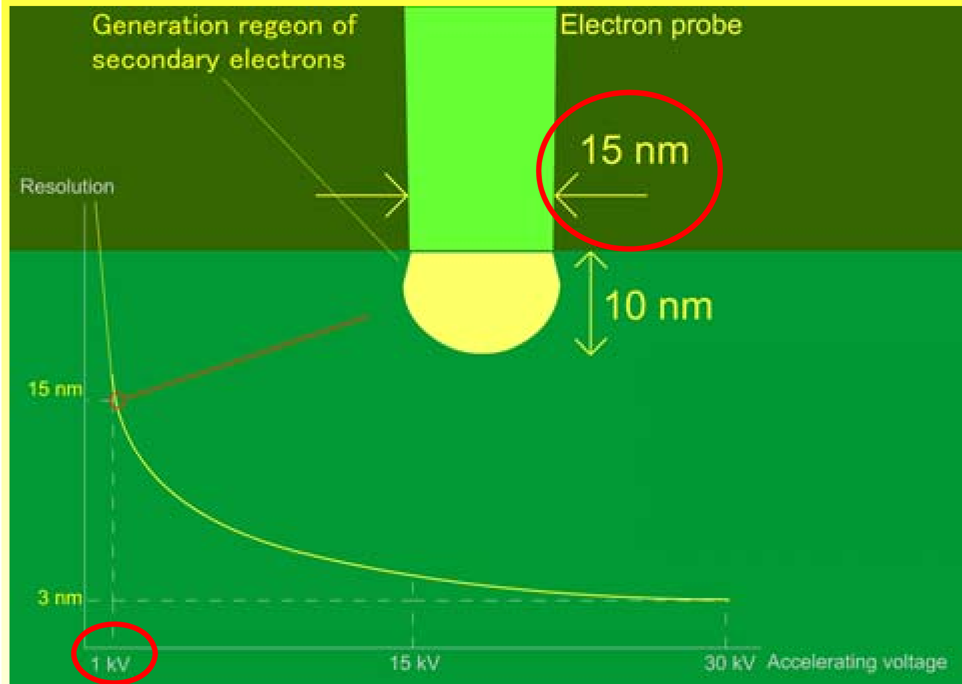
- Increasing the strength of the condenser lens
- Decreasing the size of the objective aperture
- Decreasing the working distance (WD = the distance the sample is from the objective lens)



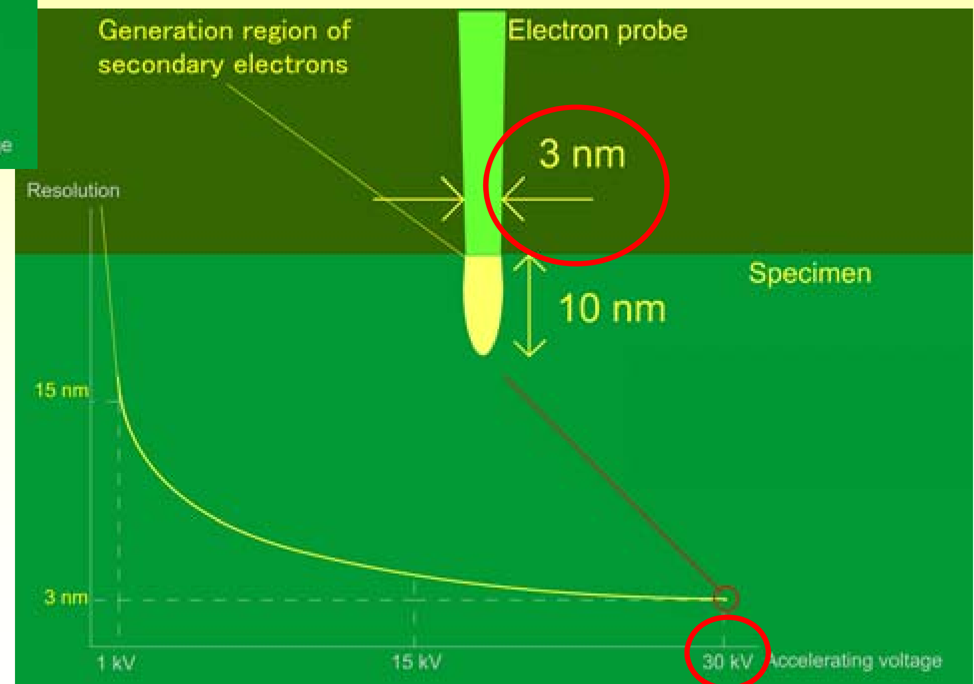
The smallest distance we can see between points in a light microscope (LM) is about 200 nm whereas a typical scanning electron microscope (SEM) can distinguish gaps smaller than 10 nm.

Resolution improvements achieved with aberration correction.

Resolution x Voltage



Probe diameter becomes larger as the accelerating voltage is lowered.



Resolution x Voltage

0.3 kV 5,000x

Resolution

0.5 kV 5,000x

15 nm

Extremely low voltages in tungsten SEM may not be suitable to observe fine surface structures.
FE SEM has better resolution in very low energy condition.

1 kV 5,000x

3 nm

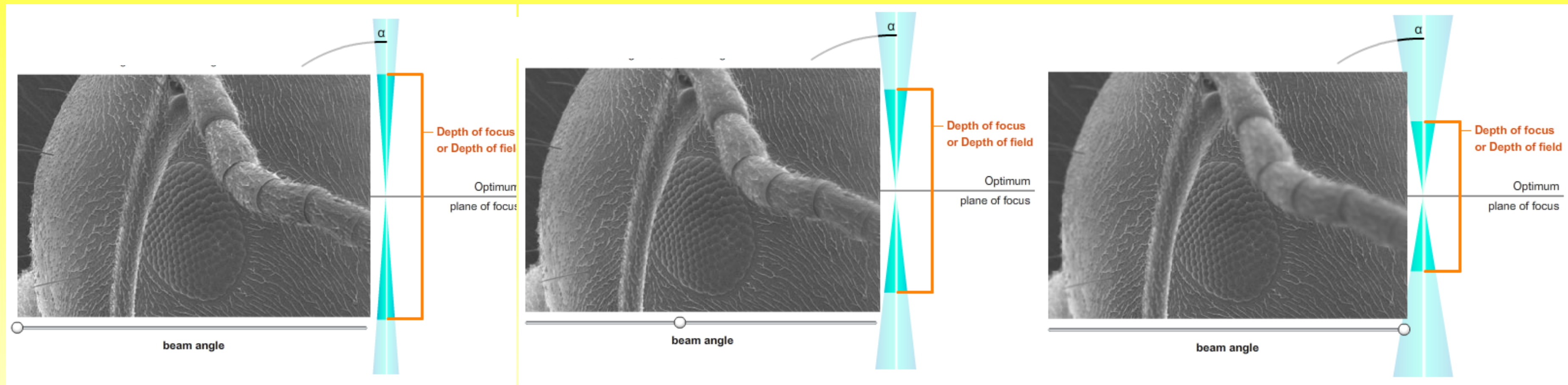
1 kV

15 kV

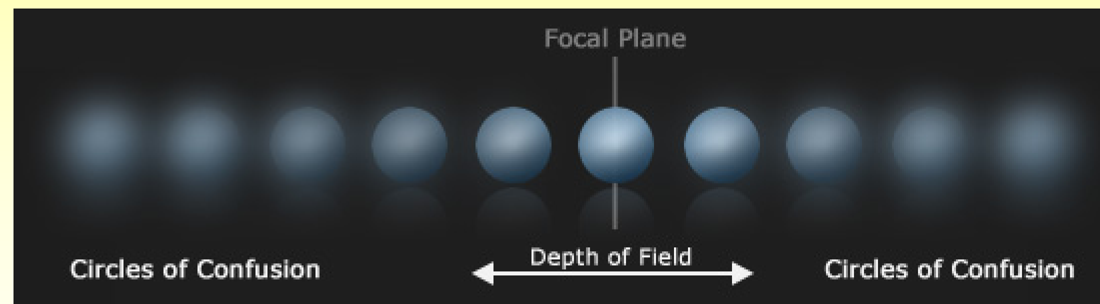
30 kV Accelerating voltage

Depth of Field

The height over which a sample can be clearly focused is called the Depth of Field. The SEM has a large depth of field which produces the images that appear 3-dimensional in nature.

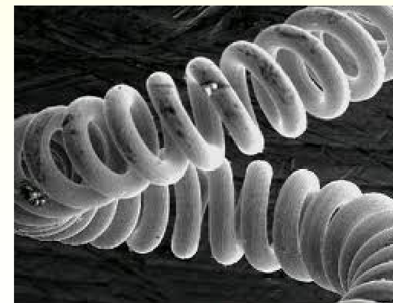


Depth of field is improved by:
- Longer working distance
- Smaller objective apertures
- Lower magnifications



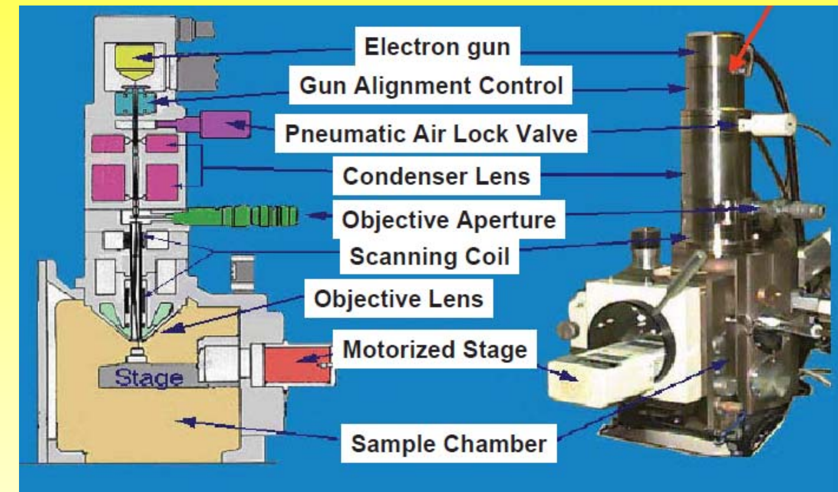
Depth of Field vs. Resolution

Depth of field and resolution have a reciprocal relationship:
- Improving resolution in conventional SEM's leads to a smaller depth of field
- While increasing depth of field decreases resolution.



Major Components of the Scanning Electron Microscope

Despite the differences between the light and electron microscopes, the components of the SEM have an analogous function to the parts of a light microscope.



Major Components of the Scanning Electron Microscope

All scanning electron microscopes consist of:

An **electron gun (1)** which generates a beam of electrons.

A **column** (2) which focuses and illuminates the specimen

A **specimen chamber (3)** where the electron beam interacts with the sample.

Detectors (4) to monitor the different signals that result from the electron beam/sample interaction.

A **viewing system** (5) that builds an image from the detector signal.

A **water chilling system**, which maintains a constant temperature of 20°C for the operation of the magnetic lenses in the microscope. If the chiller fails and the magnetic lenses heat up, the SEM will automatically shutdown.

A **vacuum system**, which consists of an **oil rotary (backing) pump** (for *rough evacuation*) and an **oil diffusion pump** (*higher vacuums)*.

1. Electron gun – generates the electron beam

Electron guns provide electrons for an electron beam by allowing them to escape from a cathode material.

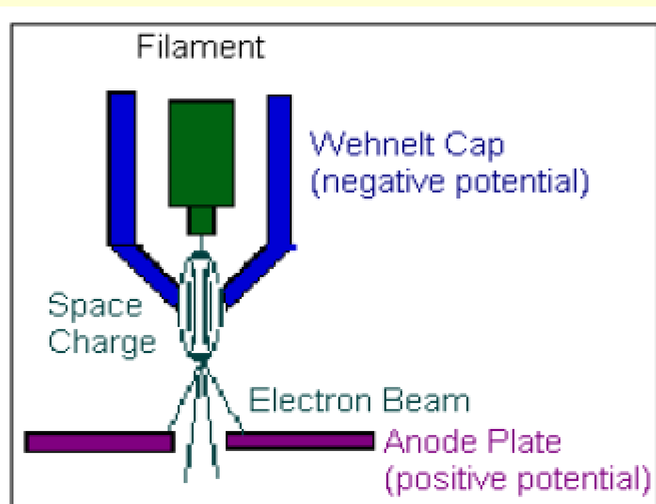
- An electron must be supplied sufficient energy to kick it into a high energy state within the material and additional energy for it to escape the surface.
- The electrons are emitted from a small area of the filament (point source).

A point source is important because it emits monochromatic electrons (with similar energy).

There are two main types of electron sources used in SEMs and microprobes:

Thermionic sources

Field emission source (FEG)



Emission	Thermionic		Field Emission
	W	LaB6	FE
Size (nm)	1 x 10^5	2 x 10^4	0.2
Brightness (A/cm2.steradian)	10^4 - 10^5	105 - 10^6	10^7 - 10^9
Energy Spread (eV)	1 - 5	0.5 – 3.0	0.2 – 0.3
Operating Lifetime (hrs)	>20	>100	>300
Vacuum (torr)	10-4 – 10^{-5}	10-6 – 10^{-7}	10-9 – 10

1. Electron gun - Thermionic sources

- electrons are **produced by heating a conductive material** to the point where the outer orbital electrons gain sufficient energy to escape.
- There are two main types of thermionic sources: tungsten metal filaments and LaB_6 crystals.

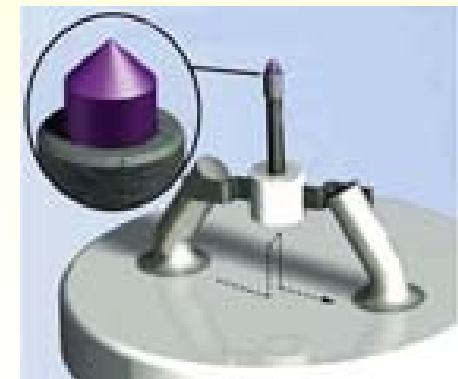
These two types of sources require vacuums of ~10^{-5} and ~10^{-7} torr, respectively.

The tungsten cathode

- is a fine wire approximately 100mm in diameter that has been bent into the shape of a hairpin with a V-shaped tip.
- The tip is heated by passing current through it; normally, the tip is heated to around 2400°C.
- The tungsten filament lasts approximately 50 hours

Lanthanum hexaboride (LaB_6) cathode

The most straightforward method to achieve higher resolution is to find a material which prodeces **more electrons at a given temperature**, hence a **brighter filament and higher resolution**. LaB_6, has been the best material developed to date for this application.



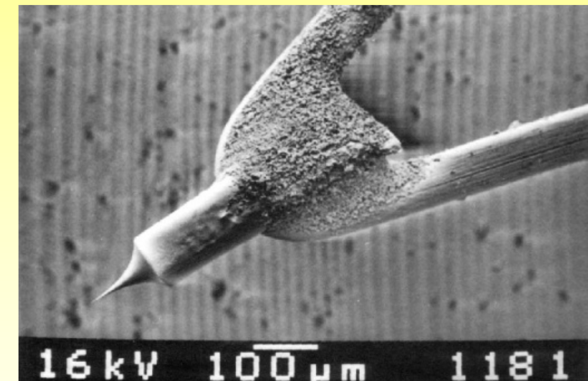
- průměr hrotu 20 μm
- The LaB_6 filament operates at approximately 2125°C and is **five times brighter than a tungsten filament** under the same conditions. However, LaB_6 filaments tend to be more expensive than tungsten filaments.
- životnost nad 500 hodin

1. Electron gun - Field emission source (FEG)

- electrons are **produced by a large electrical field**, 10^5 to 10^8 V/cm, which is placed between cathode and anode.
- the cathode forms a very sharp tip (typically 100 nm or less)
- Although the total current is lower than either the tungsten or the LaB6 emitters, the current density is between 103 and 106 A/cm. Thus, the field emission gun is hundreds of times brighter than a thermionic emission source.

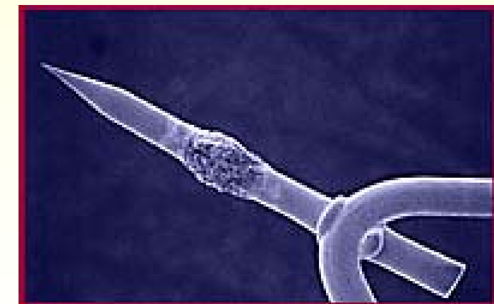
Cold source

works even at room temperature and depends only very slightly on temperature, indicating that it is not a temperature activated process. Instead, it is a purely quantum mechanical effect called "tunneling".
Field emission sources require vacuums of ~10^{-9} torr.

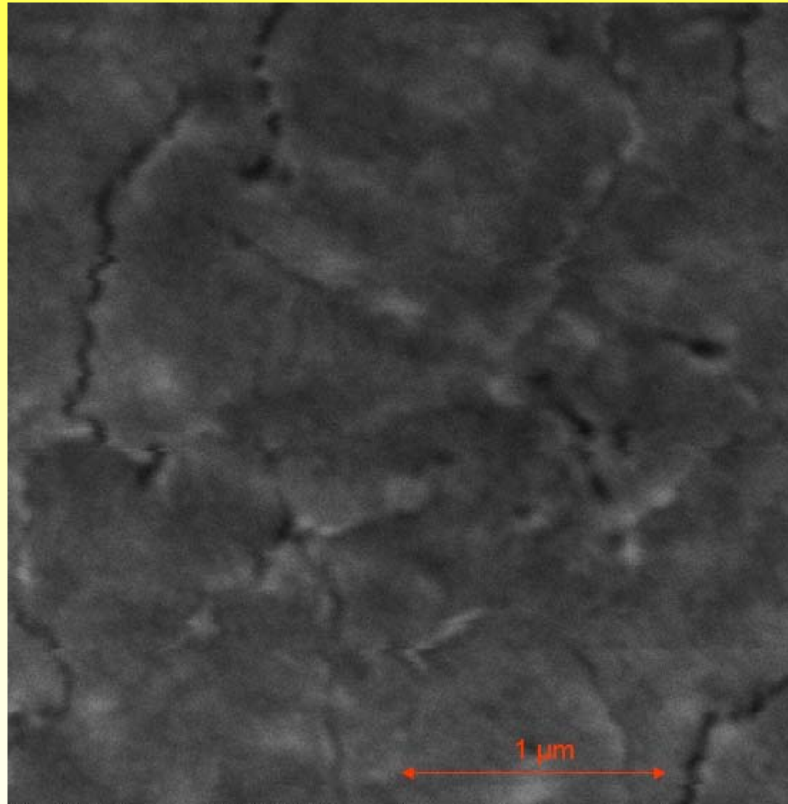


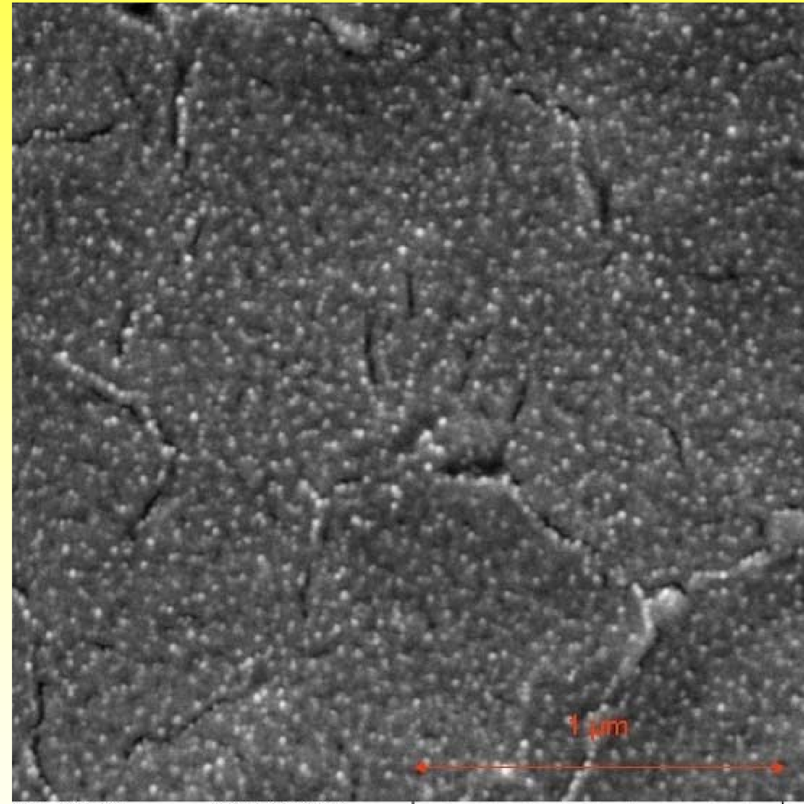
Thermal-field (TF) source

in which the tungsten point in a field emission source is heated, incorporating both thermionic and field emissions;
this is also referred to as a "'Schottky cathode".
A TF source requires a vacuum of ~10^{-8} torr.



Comparison of tungsten cathode and FEG





2. Electron column - The electron beam is focused using electromagnetic lenses.

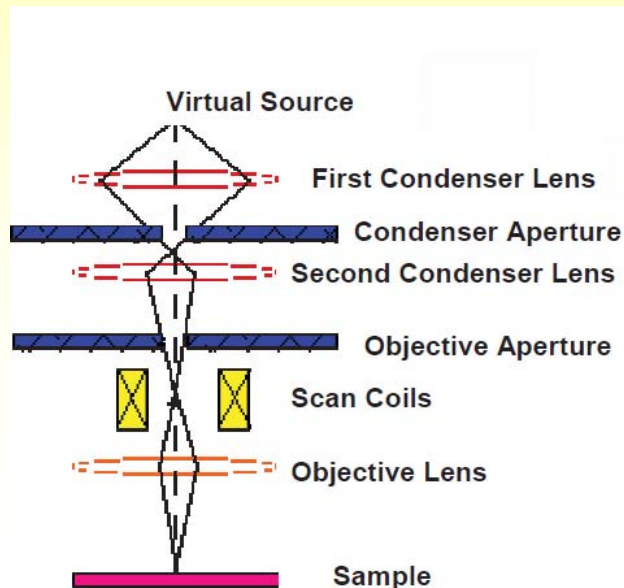
Magnetic lens system

The magnetic lens system consists of a:

- ***Condenser lens*** which is composed of one or more lenses, controls the intensity of the electron beam reaching the specimen
- ***The probe-forming lens***, often called the ***objective lens*** brings the electron beam into focus (de-magnifies) on the specimen.
- ***Scanning coils*** deflect the electron beam horizontally and vertically over the specimen surface. This is also called rastering.

Objective lens (OL) aperture

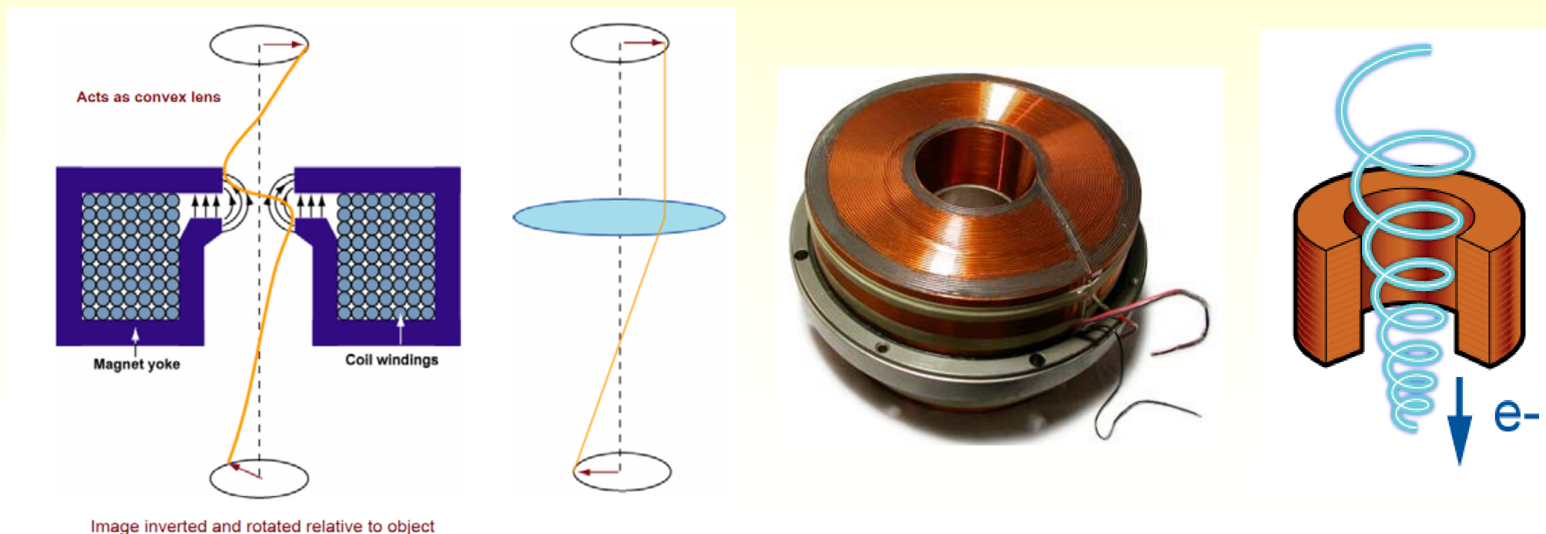
This aperture is used to reduce or exclude extraneous (scattered) electrons. An optimal aperture diameter should be selected for obtaining high resolution secondary electron images.



Lenses

The purpose of a lens is to change the path of the rays in a desired direction.
Glass or transparent plastic may **bend light** and so are used in optical lenses. However, glass or plastic lens will **stop electrons**. Therefore, it is not appropriate to use glass or plastic as lenses in an electron microscope.
Since **electrons are charged particles** and they can be bent in a **magnetic field**. Lenses for electrons are constructed with ferromagnetic materials and windings of copper wire. These produce **a focal length which can be changed by varying the current through the coil**. They are called **electromagnetic lenses**. The magnetic field bends electron paths in a similar way that solid glass lenses bend light rays. Under the influence of a magnetic field, electrons assume a helical path, spiralling down the column.

An **electromagnetic lens** is a coil of wire through which current flows. Because the current flow produces a magnetic field at right angles, the field pushes inwards into the hole in the centre. This acts to shape a beam of electrons travelling in their natural spiral path down the central hole.

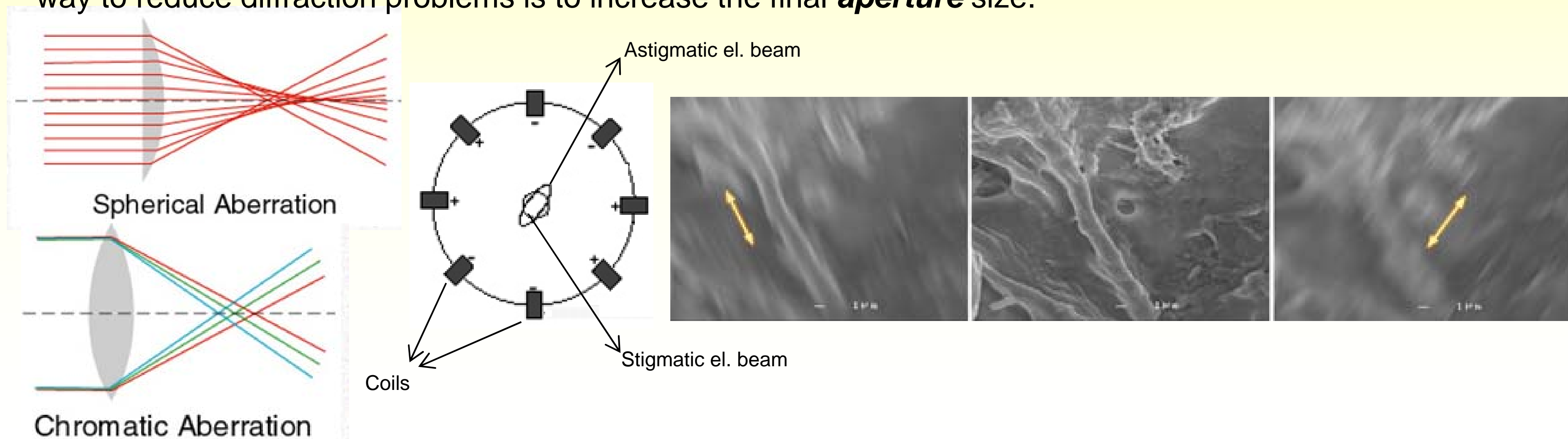


Abberations

- ***Spherical aberration*** results from **nonuniformity** of the lenses. Electrons which pass through the lens further off the optical axis are pulled more strongly than those that pass through near the center of the lens. The outer zones of a lens focus more strongly than inner zones. To reduce this effect, the final ***aperture*** can be reduced, but this reduction results in a lower beam current.
- ***Chromatic aberration*** results from **differences in electron velocity** through the lenses. The magnetic lenses will bend electrons with higher velocity or energy more strongly, resulting in a dull or blurred image. Electrons of slightly different energies are focused differently. ***No method*** exists to correct this problem other than using a more expensive LaB6 or field emission instrument. Tungsten filament systems typically have a 2 eV spread when leaving the cathode; LaB6 systems have a 1 eV spread, and field emission systems have a 0.2 to 0.5 eV spread.

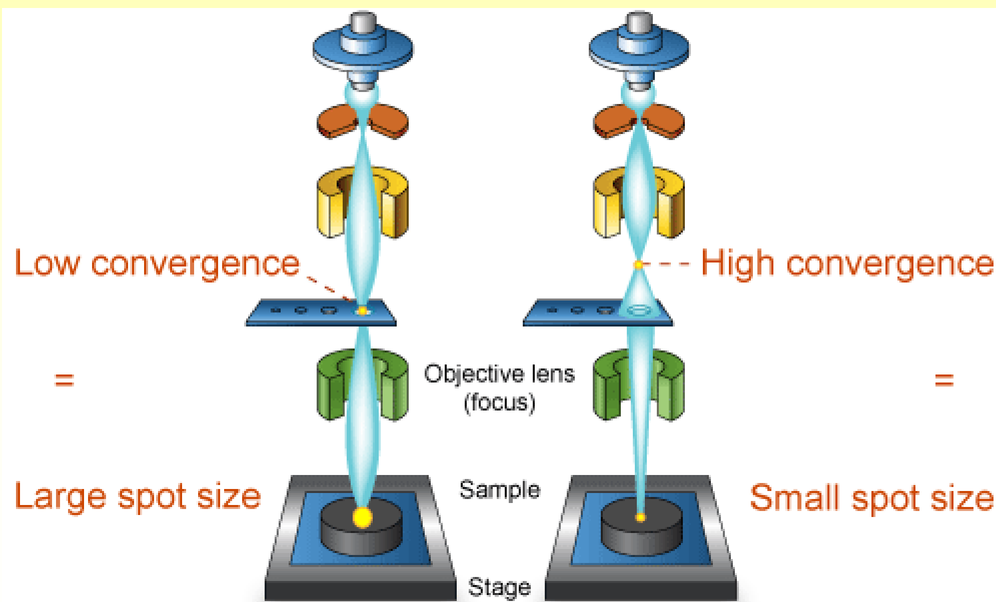
Astigmatism results from the fact that magnetic lenses **do not have perfect symmetry- the electron beam may be elliptical.** The ***stigmator*** can correct astigmatism by a correcting magnetic field to produce symmetrical electron beam at the sample. The operator can change both the strength and orientation (angle) of the magnetic field produced by stigmators to control the final beam shape.

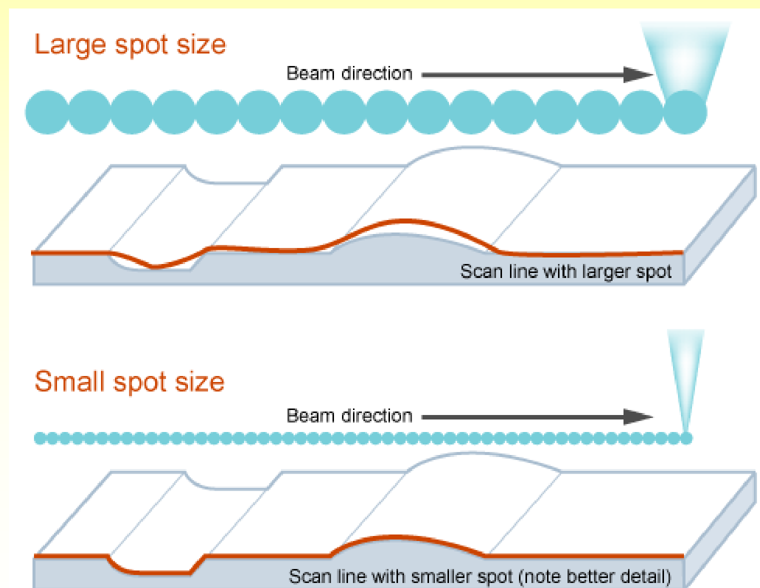
Diffraction occurs because of the **wave nature** of electrons and the aperture size of the final lens. The only way to reduce diffraction problems is to increase the final ***aperture*** size.



Condenser lens

The condenser lens convergences the cone of the electron beam to a spot below it, before the cone flares out again and is converged back again by the objective lens and down onto the sample.
This initial convergence can be at different heights, that is, close to the lens, or further away. **The closer** it is to the lens, **the smaller the spot** diameter at the point of convergence. The further away, the larger the diameter of this point.
So the condenser lens current controls this initial spot size and is referred to as the **spot size control**. The diameter of this initial convergence (also called a cross-over point) affects the final diameter of the spot the beam makes on the sample.

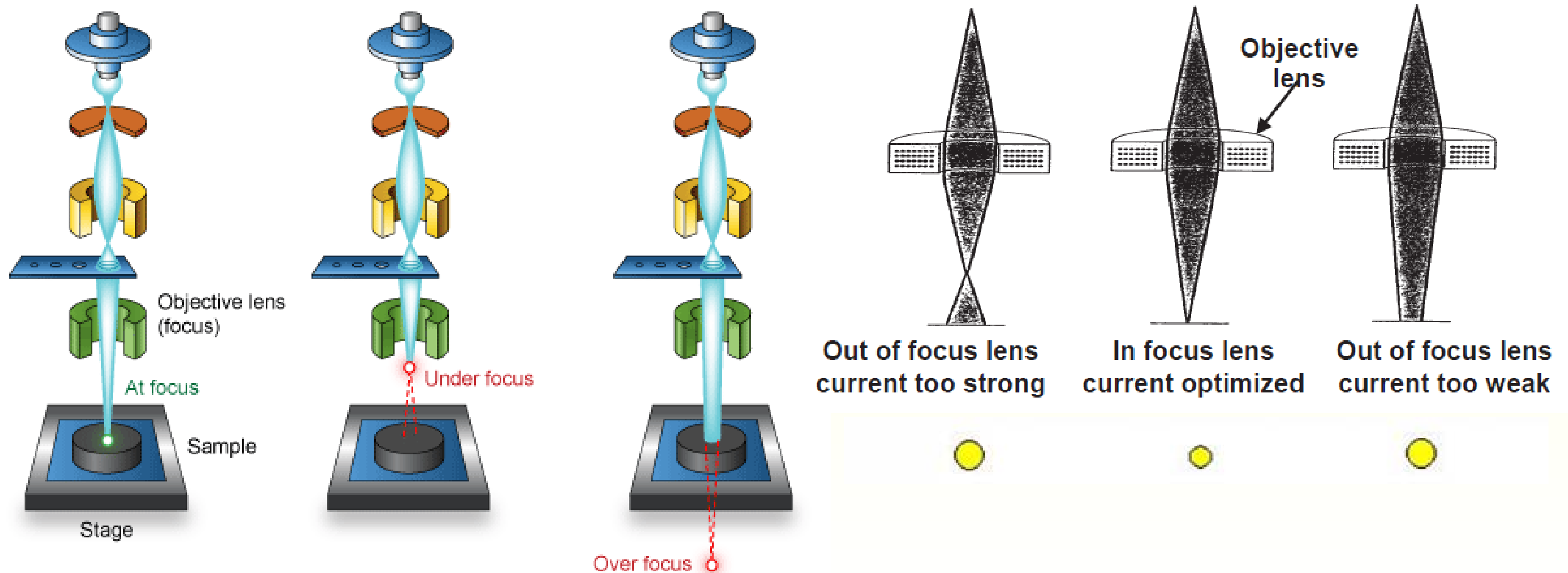




Objective lens

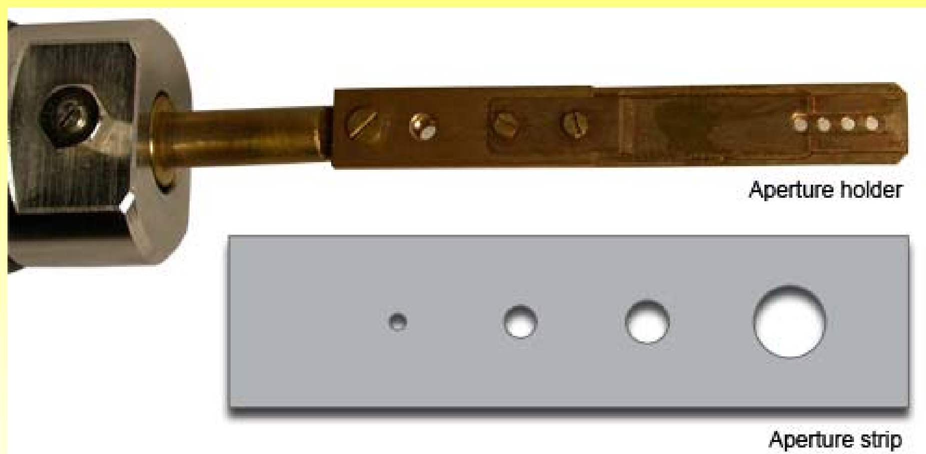
- By changing the current in the objective lens, the magnetic field strength changes and therefore the focal length of the objective lens is changed.
- **Its main role is in focusing the beam onto the sample.**
- The objective lens also has some influence over the diameter of the spot size of the electron beam on the specimen surface. A focused beam produces a smaller spot on the surface than an under or over-focused beam.

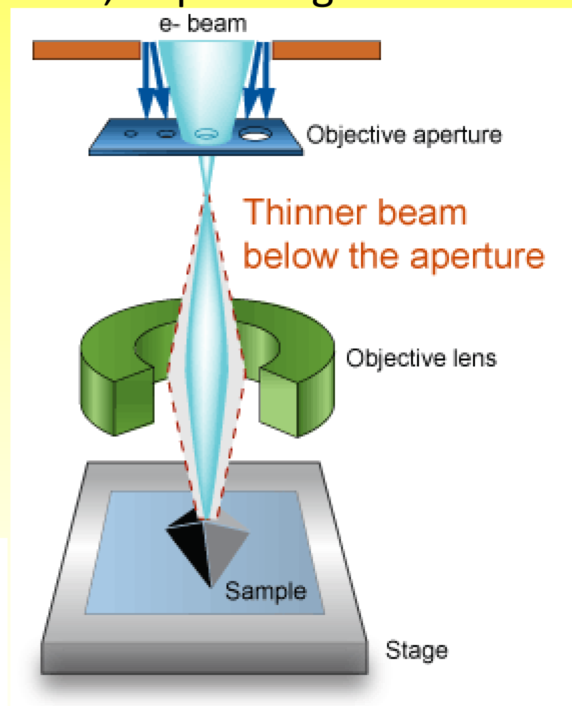
SEM works on a voltage between 2 to 50kV and its beam diameter that scans the specimen is 5nm-2μm.

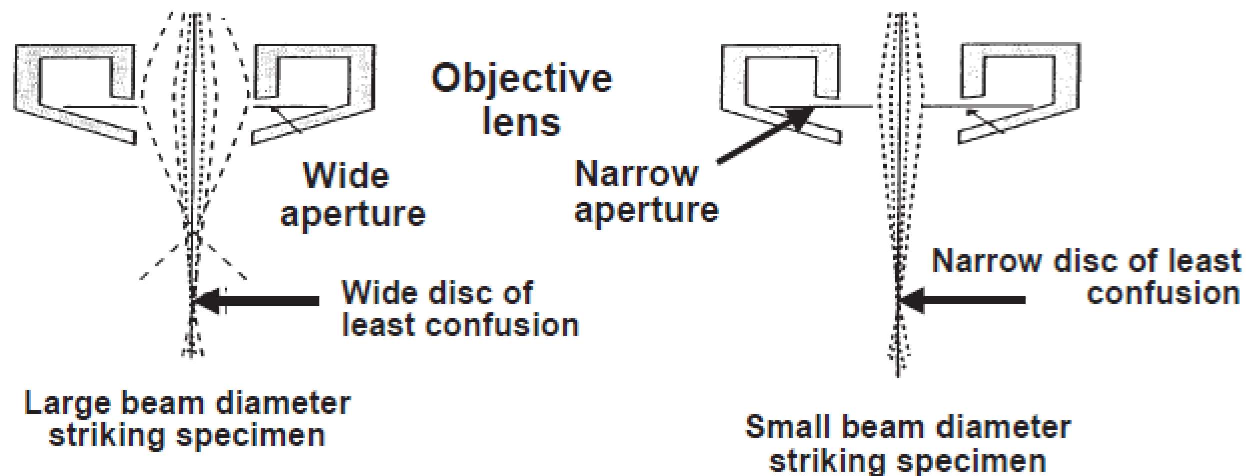


Aperture

- The **objective aperture** fits above the objective lens in the SEM.
- It is a thin metal strip with different sized holes that line up with the larger holes.
- The aperture stops electrons that are off-axis or off-energy from progressing down the column. It can also narrow the beam below the aperture, depending on the size of the hole selected.







Aperture

A large aperture is chosen for **low magnification** imaging to **increase signal** and for **BSE** and **microanalysis** work.

A smaller aperture is chosen for **high resolution** work and **better depth of focus** but has the disadvantage of fewer electrons and therefore a less bright image.

Some examples of aperture size and purposes

Scale	Aperture diameter (microns)	Probe current	Purpose
4	30	Smallest	Ultrahigh resolution; Low probe current; Large depth of field
3	50		Usual observation
2	70		High resolution at high probe current; Reduced depth of field
1	110	Largest	Observation at high probe currents; Shallow depth of field
0	1,000	----------------	Axis alignment

4. Specimen chamber

The specimen chamber is maintained **at high vacuum that minimises scattering of the electron beam before reaching the specimen**. This is important as scattering or attenuation of the electron beam will increase the **probe size** and **reduce the resolution**, especially in the SE mode. A high vacuum condition also optimises collection efficiency, especially of the secondary electrons.

Specimen stage

The specimen holder is fixed to the specimen stage. The stage can be moved along the X, Y (in the specimen plane), and Z directions (at right angles to the specimen plane). The Z adjustment is also known as the specimen height. The specimen stage can also rotate continuously or to be tilt.

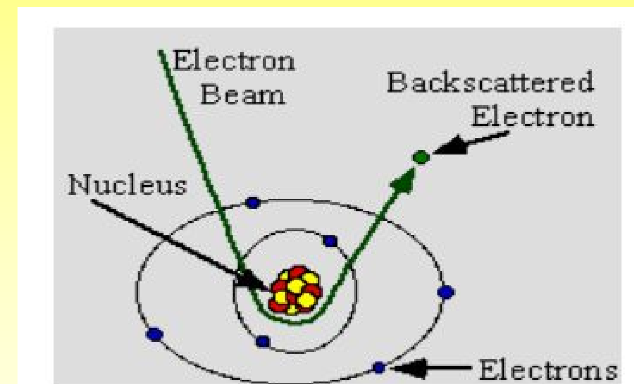
Electron-matter interactions

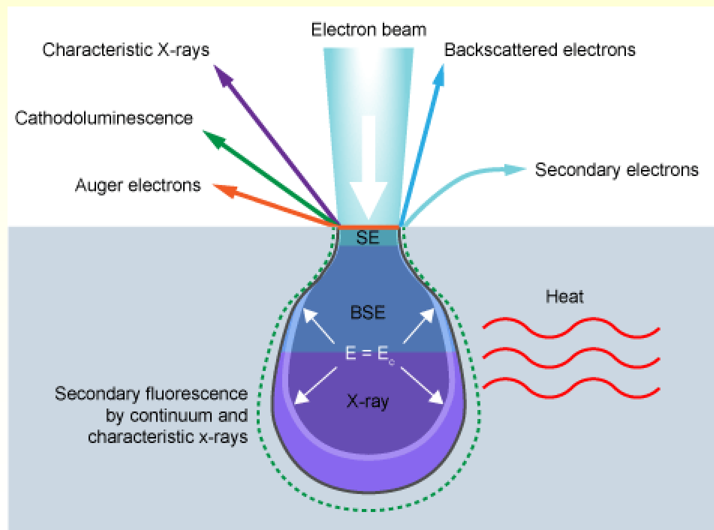
When the electron beam hits a sample it interacts with the atoms in that sample.

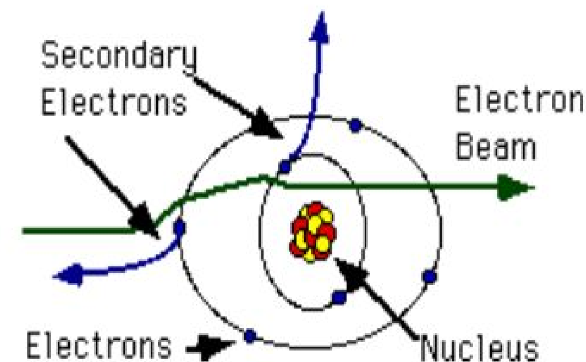
Electron-matter interactions can be divided into two classes:

1. **Elastic scattering** – the electron **trajectory within the specimen changes**, but its **kinetic energy and velocity remains essentially constant**. The result is generation of backscattered electrons (BSE).
2. **Inelastic scattering**– the incident electron **trajectory is only slightly perturbed, but energy is lost through the transfer of energy to the specimen**.

Inelastic scattering, places the atom in an excited (unstable) state. The atom "wants" to return to a ground or unexcited state. Therefore, at a later time the atoms will relax giving off the excess energy. XRays, cathodoluminescence and Auger electrons are three ways of relaxation. **The relaxation energy is the fingerprint of each element.**



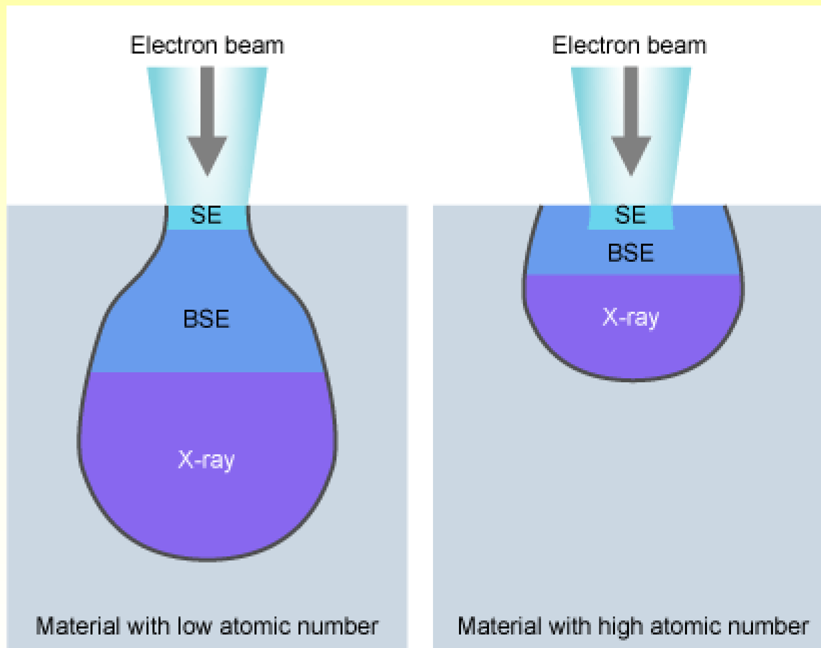




Electron-matter interactions

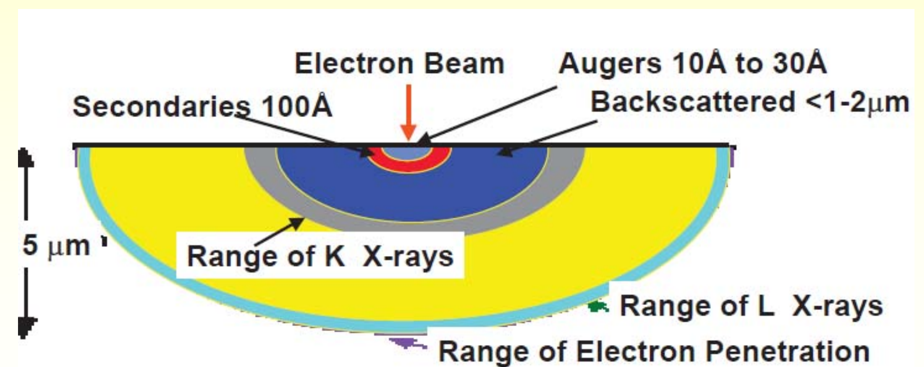
The volumes involved in the production of secondary electron (SE), backscattered electron (BSE) and X-rays, form into a shape that ranges from a tear-drop to a semi circle within the specimen. This shape is called an ***interaction volume*** and **its depth and diameter depends on the kV as well as the density of the specimen**.

Approximately the top 15nm of the volume comprises the zone from which SE can be collected, the top 40% is the region from which BSE can be collected and X rays can be collected from the entire region.



An example of a typical Interaction volume for:

- Specimen with atomic number 28, 20 kV
- 0° degrees tilt, incident beam is normal to specimen surface



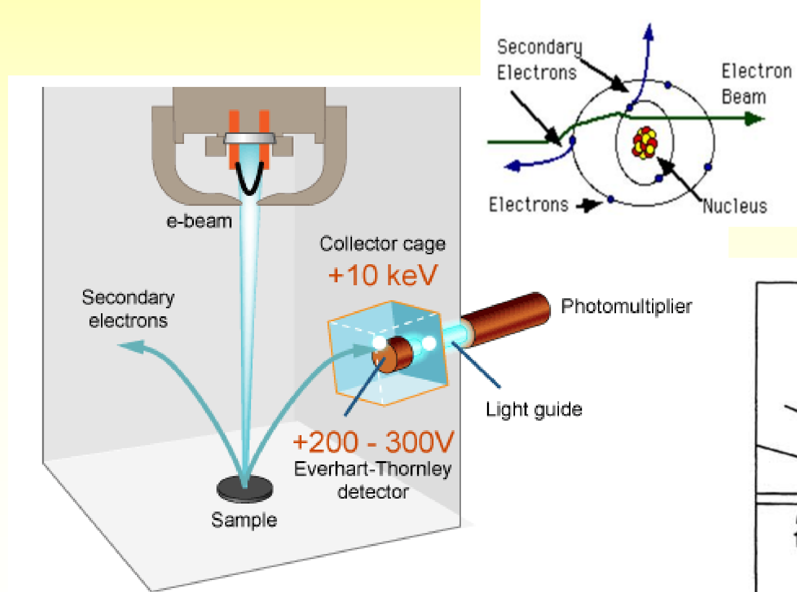
Secondary electrons

- Secondary electrons are **low energy electrons (E < 50 eV)** formed **by inelastic scattering.**
- The low energy of these electrons allows them to be collected easily. This is achieved by placing a positively biased grill on the front of the SE detector, which is positioned off to one side of the specimen. The positive grill attracts the negative electrons and they go through it into the detector. This is the case for the **Everhart-Thornley detector** which is most commonly used but there is another kind of **In-lens SE** detector in some machines.

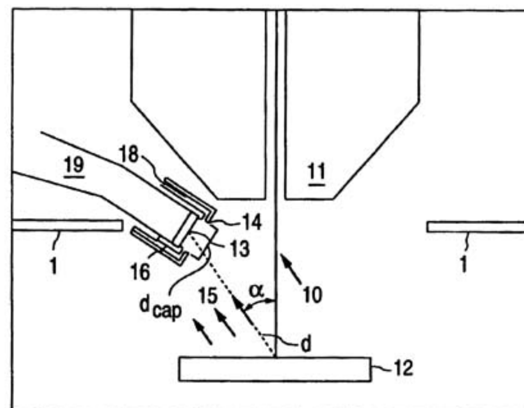
Since this lets us collect a large number of the secondaries (50 - 100%), we produce a “**3D” type of image of the sample with a large depth of field**.

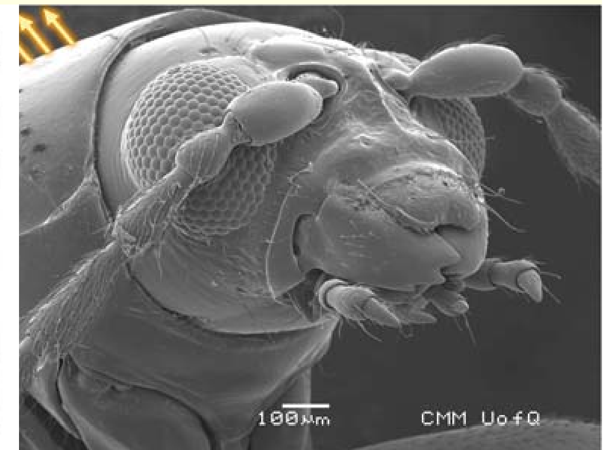
Electrons emitted from a surface that faces away from the detector or which is **blocked by the topography of the specimen**, will **appear darker** than surfaces that face towards the detector.

- The major influence on SE signal-generation is the **shape (topography) of the specimen surface**.



In the image of the beetle, the electron detector is in the top left corner, hence that region looks brightest. It is, however, not the only factor that contributes to the contrast and brightness in an SEM.





Secondary electrons

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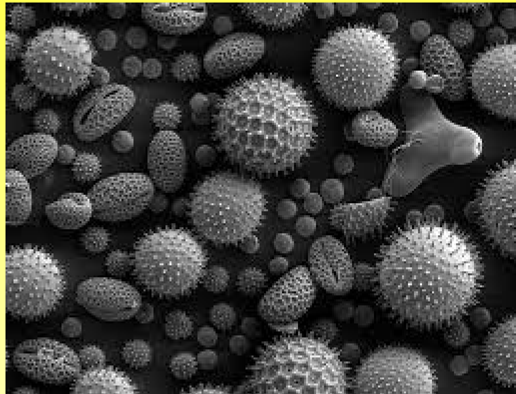
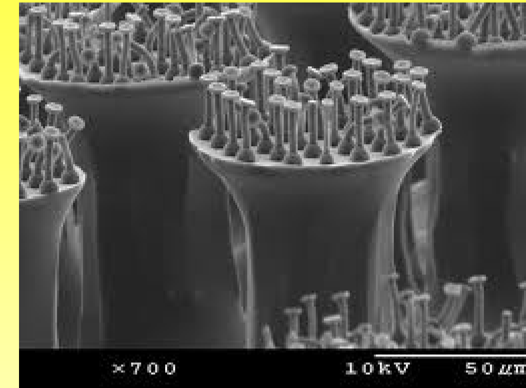


Pollen, http://www.mansic.eu/documents/PAM1/Giannakopoulos1.pdf

Vespula vulgaris - antenna



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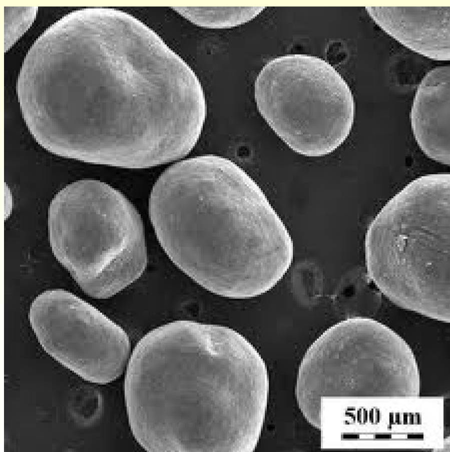
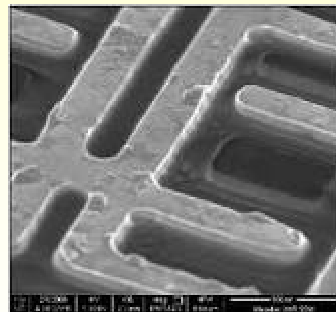
- Secondary electrons provide particularly good edge detail. Edges (and often pointy parts) look brighter than the rest of the image because they produce more electrons.



http://crysa.fzu.cz/MMaterialu2012/lectures.htm



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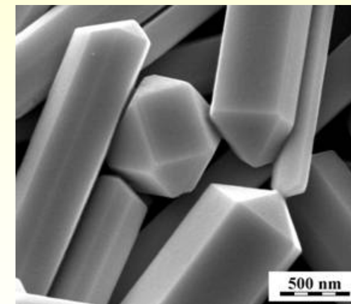

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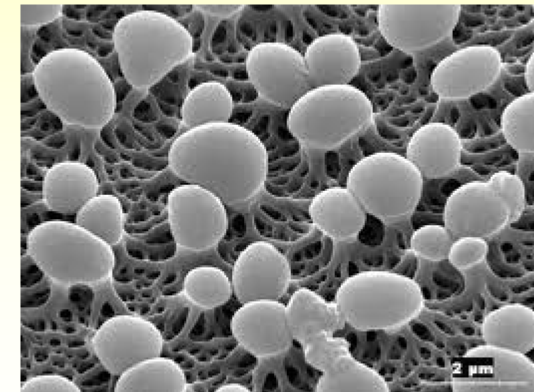
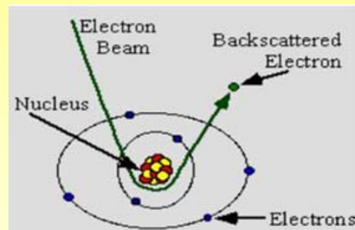
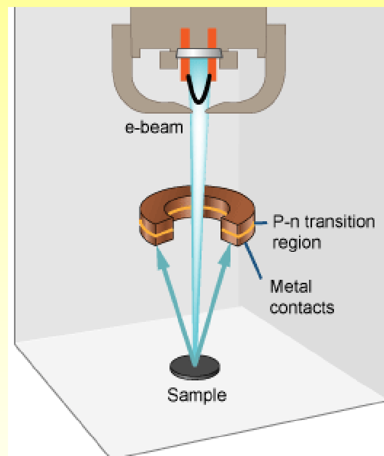


Rhyzopertha dominica egg
http://old.padil.gov.au/pbt/index.php?q=node/15&pbtID=187

Backscattered electrons

- **Backscattered (BS) electrons are high-energy electrons (>50 eV)**, which **arise due to elastic collisions** between the incoming electron and the nucleus of the target atom (i.e. Rutherford scattering).
- These BSE are used to produce a different kind of image. Such **an image uses contrast to tell us about the average atomic number of the sample.** The higher the average atomic number, the more primary electrons are scattered (bounced) back out of the sample. This leads to a brighter image for such materials.
- The angle of scattering can range from 0 to 180°.





Backscattered electon image (BSE)

Secondary electron image (SE)

- Since BSE have high energies, they can't be pulled in like secondaries. If you placed a potential on a grid to attract them, you would also attract the ncident beam!!
- The most common detector used is called a **surface barrier detector**. It sits **above the sample, below the objective lens**. BSE which strike it are detected.

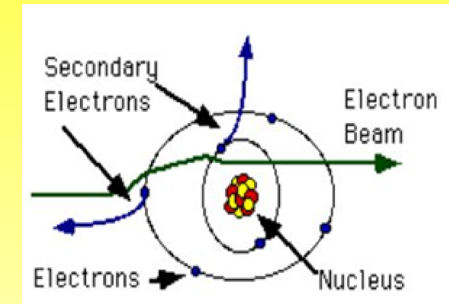
Microanalysis - The relaxation energy is the fingerprint of each element.

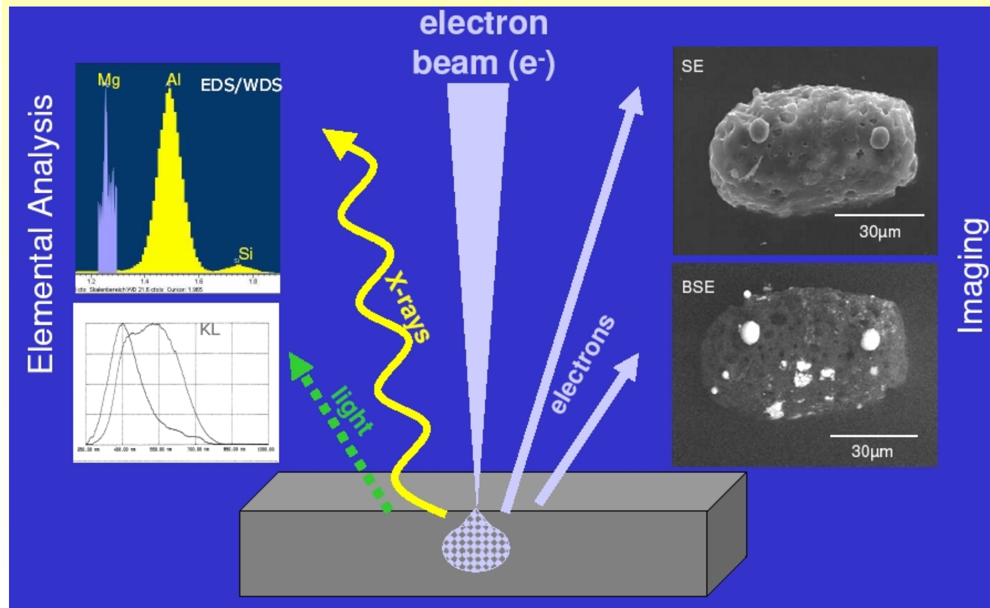
A technique to analyse composition in regions of a sample in the micron and nano ranges.

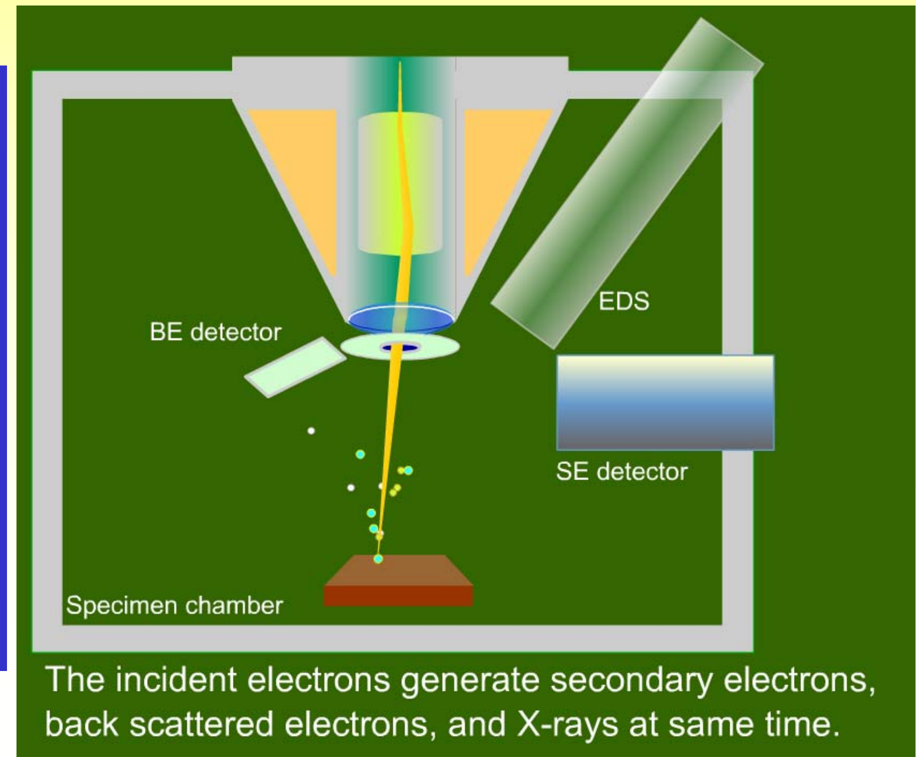
Techniques include spectroscopy

- ❖ Energy Dispersive or Wavelength dispersive X-ray
- ❖ Cathodoluminescence
- • Electron Backscatter Diffraction

Inelastic scattering





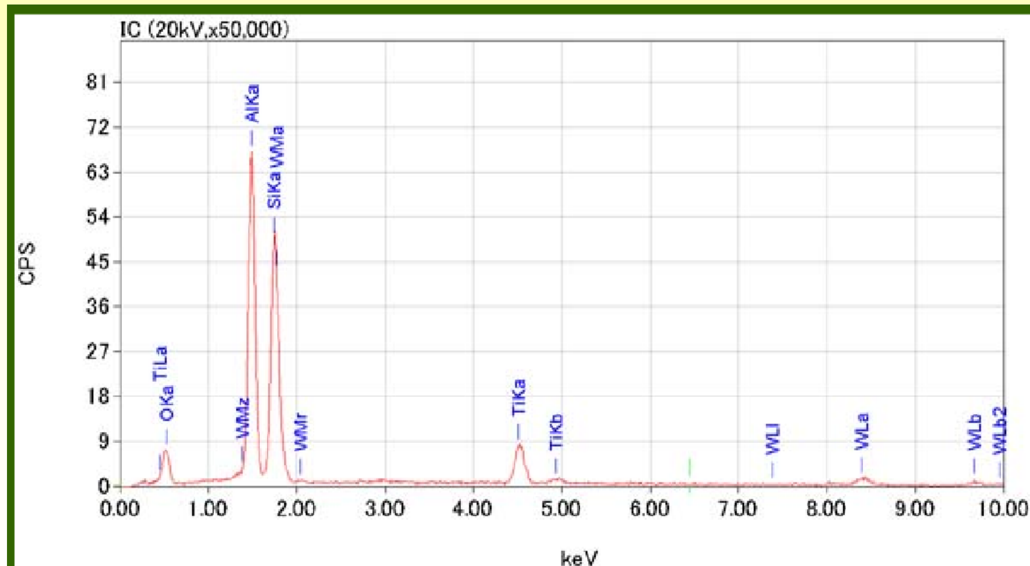


The incident electrons generate secondary electrons, back scattered electrons, and X-rays at same time.

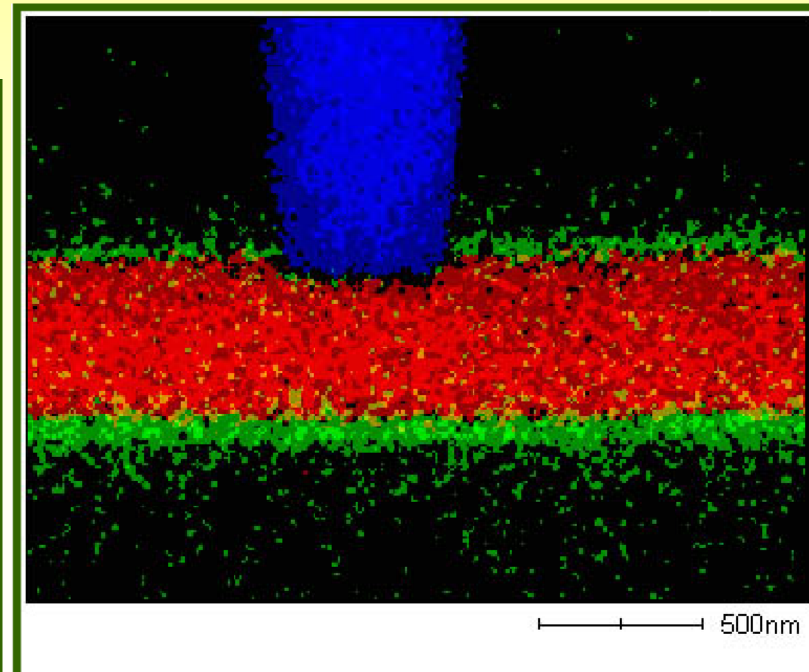
Energy Dispersive or Wavelength dispersive microanalysis

When the sample is bombarded by the electron beam of the SEM, electrons are ejected from the atoms on the specimens surface. A resulting electron vacancy is filled by an electron from a higher shell, and an X-ray is emitted to balance the energy difference between the two electrons. The EDS X-ray detector (also called EDS or EDX) measures the number of emitted x-rays versus their energy. **The energy of the x-ray is characteristic of the element from which the x-ray was emitted**.

- the elemental analysis or chemical characterization of a sample
- each element has a unique atomic structure allowing unique set of peaks on its X-ray spectrum



EDS spectrum of IC device

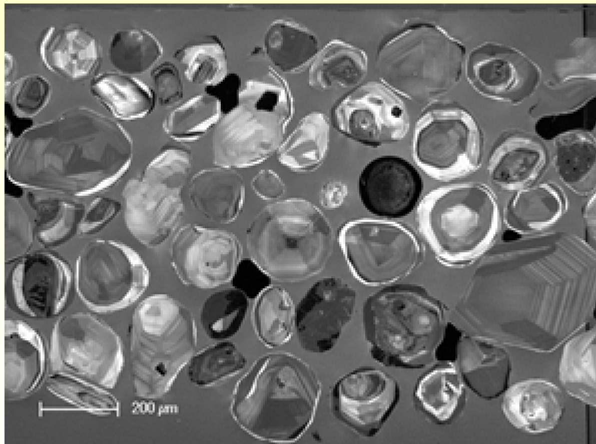


EDS X-ray mapping of IC device showing distribution of three elements

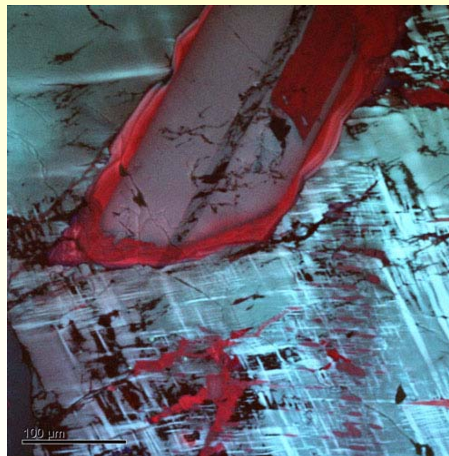
Cathodoluminescence (CL)

- Cathodoluminescence (CL) is the **emission of photons of characteristic wavelengths from a material that is under high-energy electron bombardment**.
- The CL response of the sample is recorded with digital images from the CL detector.
- The CL images can be obtained over a range of magnifications (10-10,000x), but the lowest magnification is constrained by the specific configuration of the CL detector system.

The **image** produced by cathodoluminescence **showing otherwise invisible microstructural defects and impurities**. It is used to examine internal structures of semiconductors, rocks, ceramics, glass, etc. in order to get information on the composition, growth and quality of the material.
The distribution of the CL in a material gives fundamental insights into such processes as crystal growth, replacement, deformation and provenance.



Zircon, showing crystal growth patterns. Microanalysis can provide details on composition, defects, and impurities.



http://serc.carleton.edu/research_education/geochemsheets/semcl.html

This photo is a CL image from granite and shows two minerals intergrown with each other. The bluish cross-hatched area is occupied by a grain of potassium feldspar (microcline), and the purple red-rimmed mineral in the upper half of the image is a grain of sodium feldspar (albite)

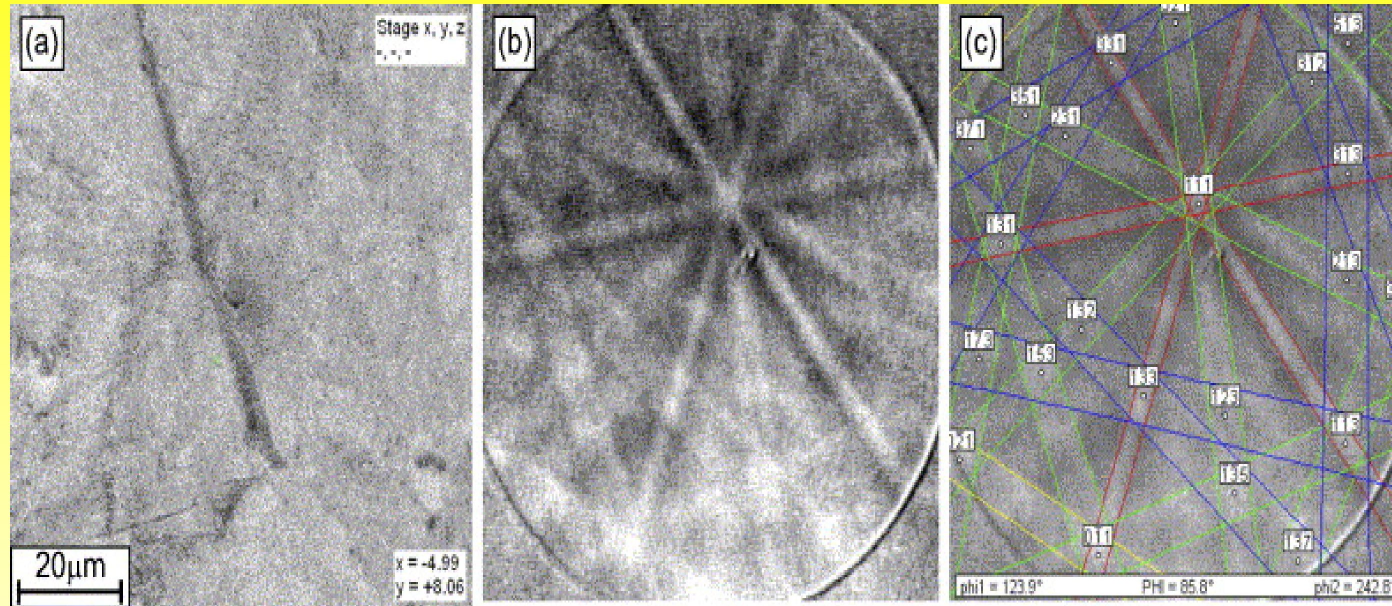
Electron backscatter diffraction (EBSD)

- EBSD is used **to examine the crystallographic orientation** of many materials, which can be used to elucidate texture or preferred orientation of any crystalline or polycrystalline material.
- EBSD can be used to index and identify the seven crystal systems, and as such it is applied to:
  - crystal orientation mapping,
  - defect studies,
  - phase identification,
  - grain boundary and morphology studies,
  - egional heterogeneity investigations,

  ...
- For an EBSD measurement a flat/polished crystalline specimen is placed in the SEM chamber at a highly tilted angle (~70° from horizontal) towards the diffraction camera, to increase the contrast in the resultant electron backscatter diffraction pattern.

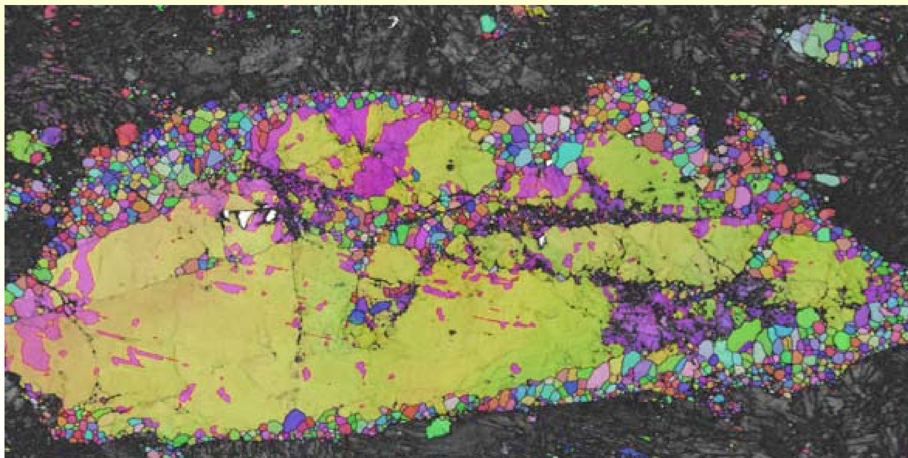
- An electron backscatter diffraction pattern is formed when many different planes diffract different electrons to form **Kikuchi bands** which correspond to each of the lattice diffracting planes.
- **Each band can be indexed individually by the Miller indices** of the diffracting plane which formed it.
- In most materials, **only three bands/planes which intercept are required to describe a unique solution to the crystal orientation** (based upon their interplanar angles) and most commercial systems use look up tables with international crystal data bases to perform indexing.

Electron backscatter diffraction (EBSD)



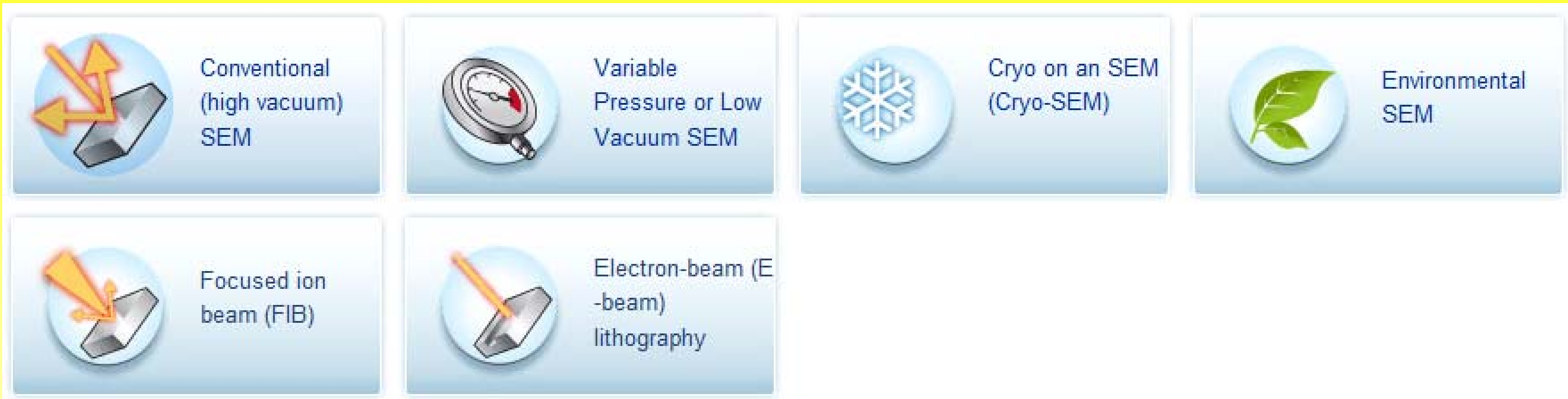
EBSD characterization of grain boundary network: (a) SE image from grain boundary, (b) EBSD pattern of grain boundary and (c) overlaying of ferrite pattern on the measured EBSD pattern.

http://www.sciencedirect.com/science/article/pii/S0921509306025172



An orientation map of a quartz clast in a pseudotachylite (earthquake rock) showing recrystallisation around the outer edge. Microanalysis can provide details on crystallographic information.

Types of SEM



Conventional (high vacuum) scanning electron microscopy (SEM)

- This is the most common type of machine.
- It requires a dry, conductive sample (often achieved by applying a thin layer of metal to the surface with a technique called sputtering).
- The sample must be able to withstand a high vacuum.
- This type of machine is used for routine imaging, using either secondary electrons (SE) or backscattered electrons (BSE).

Variable Pressure or Low Vacuum scanning electron microscopy (LVSEM)

- This type of machine is basically like a conventional SEM but has the advantage in low vacuum (LV) mode that the pressure can be adjusted in the sample chamber until the artefact of "electron charging" is removed from images.

- This means LVSEM can be used **to image the surface of non-conductive samples** (no metal needs to be added to the surface of such samples). It is particularly useful for viewing polymers, biological samples, and museum samples that cannot be changed in any way, particulate samples, and geological materials. **Imaging uses backscattered electrons (BSE)**.

- Backscattered electron imaging (BSE) of nonconductive, uncoated samples can provide **information about composition** via the contrast of the image: whiter regions have a higher average atomic number than darker regions.

- The LV mode can also be used to freeze-dry samples for viewing.

Using cryo on a scanning electron microscope (Cryo-SEM)

Cryo stands for frozen.

- A cryo-scanning electron microscope is a conventional SEM that has been fitted with specific equipment that allows samples to be viewed in the frozen state. This is particularly **useful for directly viewing hydrated (wet) samples, delicate biological samples, hydrogels, food, biofilms, foams, fats, and waxes, suspensions, pharmaceuticals and nanoparticles**.
- The sample can be snap frozen outside the machine and then inserted in its frozen state, or placed into the machine in an unfrozen state and frozen more slowly in the machine.
- Frozen samples can also be fractured or cut during preparation to reveal internal structures.
- It is **imaged using either secondary electrons (SE) or backscattered electrons (BSE).**

Environmental scanning electron microscope (ESEM)

- This machine is designed **to view a sample in its natural state**, without the need for desiccation.

- **Sample temperature and specimen chamber vapor pressure can both be controlled, allowing samples to be heated, cooled, wetted or dried.**

Relative humidity (RH) can be controlled within the chamber by adjusting the temperature of the conventional stage (±20° C) along with the pressure.

- Dynamic experiments can also be carried out on wet samples in real time, involving heating on a specialized hot-stage, anywhere up to 1500° C, cooling, wetting and drying. The samples can be imaged while these dynamic processes are occurring.

Focused ion beam (FIB) technology

- This technology involves using an ion beam (typically gallium ions) directed onto a hard sample. The beam is focused to an extremely fine probe size (<10 nm) onto the surface of a specimen. **The sample can be sectioned or shaped with the ion beam while it is being monitored by scanning electron microscopy (SEM)**.

FIB can cut 10-nm-thick sections from very hard materials. These sections can be taken off as sequential sections, each viewed in turn with the SEM mode, and this imaging information used to construct a 3D image.
FIB can also be used to shape needles that can then be viewed by other techniques such as transmission electron microscopy or atom probe tomography.

- **It can also be used for deposition of materials in a small area (approx 100nm) from chemical vapor from specific gasses.**

Electron-beam lithography (E-beam lithography or EBL)

- EBL is a maskless lithography technique used for patterning of computer generated layout structures on photoresists on Si wafers.
- Upon irradiation of focused electron beam, electron-sensitive resists undergo chain-scission or crosslinking, resulting in solubility switch of materials during the subsequent development process (remove/retain exposed material in development depending on the tone of the resist).
- To date, EBL remains the highest resolution patterning tool in lithography, it is widely used in photomask fabrication and low volume production of semiconductor components.

Summary - Applications and practical uses - what the SEM can do

Scanning electron microscopy is a remarkably versatile technique. There are many different types of SEMs available, tailored to specific needs. **With SEM one can**:

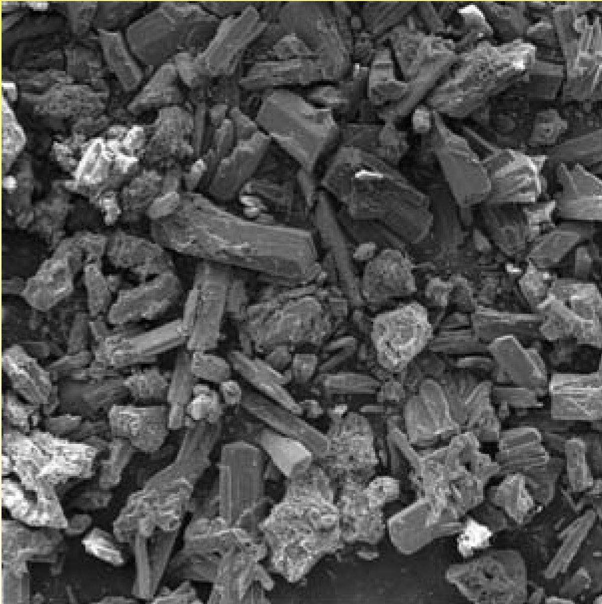
- ***Image morphology of samples*** (e.g. view bulk material, coatings, sectioned material, foils, even grids prepared for transmission electron microscopy).
- ***Image compositional and some bonding differences*** (through contrast and using backscattered electrons).
- ***Undertake micro and nano lithography***: remove material from samples; cut pieces out or remove progressive slices from samples (e.g. using a focussed ion beam).
- ***Heat or cool samples while viewing them*** (while possible in many instruments it is generally done only in ESEM or during Cryo-scanning electron microscopy).
- ***Wet and dry samples while viewing them*** (only in an ESEM)
- ***View frozen material*** (in an SEM with a cryostage)
- ***Generate X-rays from samples for microanalysis*** (EDS; WDS)
- ***Study optoelectronic behaviour of semiconductors*** using cathodoluminescence
- ***View/map grain orientation/crystallographic orientation*** and study related information like heterogeneity and microstrain in flat samples (Electron backscattered diffraction).
- ***Electron diffraction*** using electron backscattered diffraction. The geometry may be different to a transmission electron microscope but the physics of Bragg Diffraction is the same

Summary - What the SEM can't do

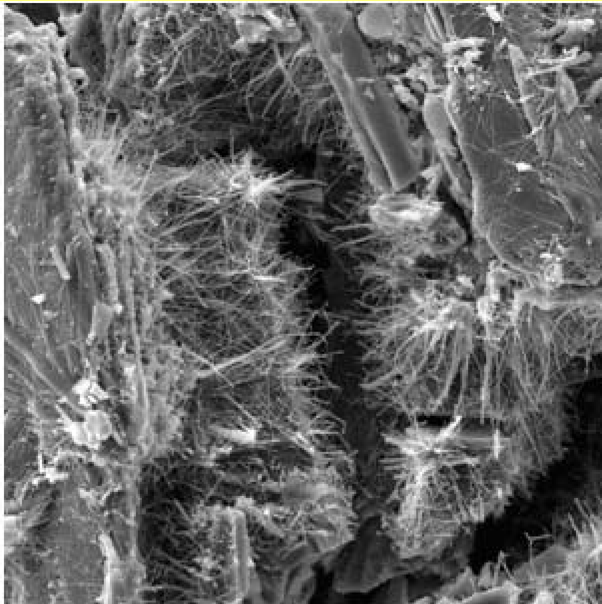
There are some things SEM can't do:

- ***SEM cannot take colour images***. The colour is often added artificially in coloured SEM images. However, some SEMs can collect true colour images via a wavelength selective cathodolumenence (CL) detector.
- ***SEM cannot image through water***. An ESEM using a wet Scanning Transmission Electron Microscope (STEM) detector can be used to image through thin water films.
- ***Generally, SEMs are not used for experiments involving liquids, chemical reactions, and air-gas systems*** although some specialised machines and sample chambers do allow for these experiments.
- The resolution of the SEM is not high enough ***to image individual atoms*** (use a transmission electron microscope).
- ***The SEM cannot reliably image charged molecules that are mobile in a matrix***. For example, some species (e.g. Na+) are volatile under the electron beam because the negative electron beam exerts a force on charged material.
- ***SEM is not good for quantifying surface roughness at small scale***. Atomic Force Microscope (Scanning Probe Microscopy) is more useful for this task.
- ***Elemental analysis below micron scale*****.** Analysis in the < 7kV range can provide elemental information on the sub-micron scale but is often problematical.

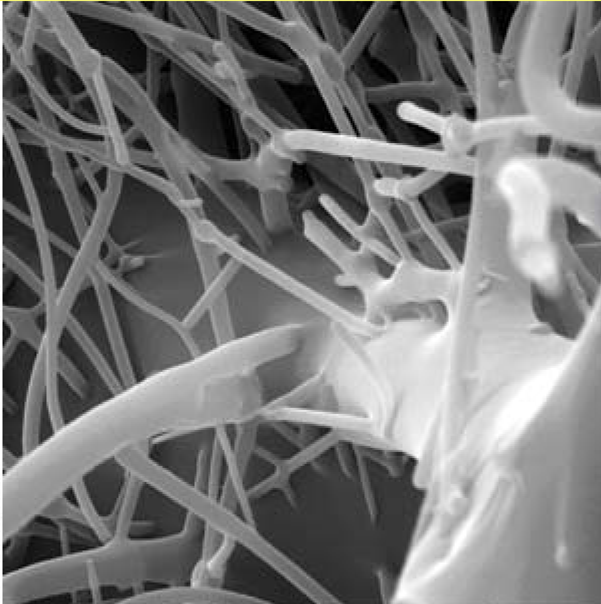
Recrystalization of API



HV: 20.00 kV | WD: 11.9400 mm | 1 mm
View field: 2.26 mm | Det: SE Detector | MIRA\ TESCAN
SEM MAG: 133 x | service | Digital Microscopy Imaging

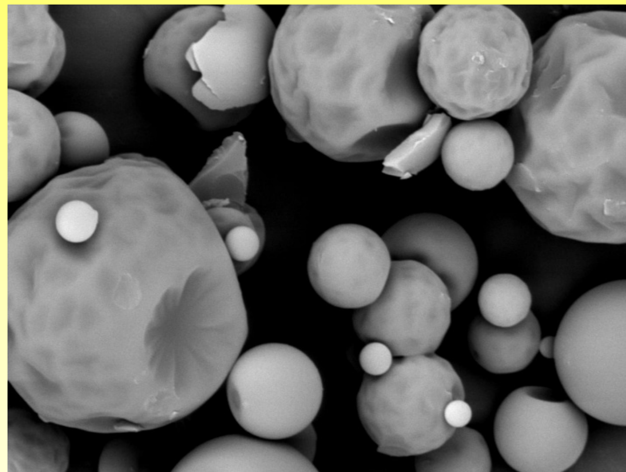


HV: 20.00 kV | WD: 11.9170 mm | 100 µm
View field: 255.74 µm | Det: SE Detector | MIRA\ TESCAN
SEM MAG: 1.18 kx | service | Digital Microscopy Imaging

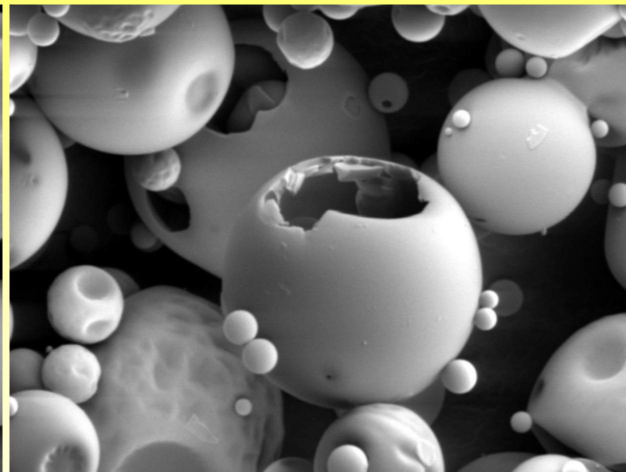


HV: 20.00 kV | WD: 11.8940 mm | 2 µm
View field: 6.74 µm | Det: SE Detector | MIRA\ TESCAN
SEM MAG: 44.74 kx | service | Digital Microscopy Imaging

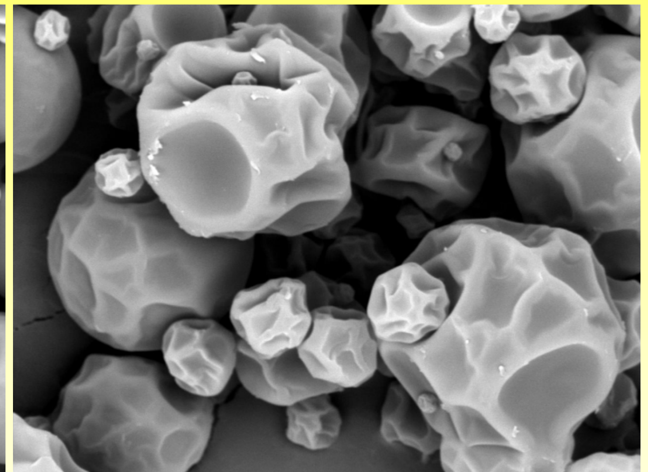
Drying under different conditions



SEM MAG: 5.00 kx DET: BSED
HV: 20.0 kV DATE: 11/09/07 20 um Vega ©Tescan
VAC: HiVac Name: 4000_bse Digital Microscopy Imaging



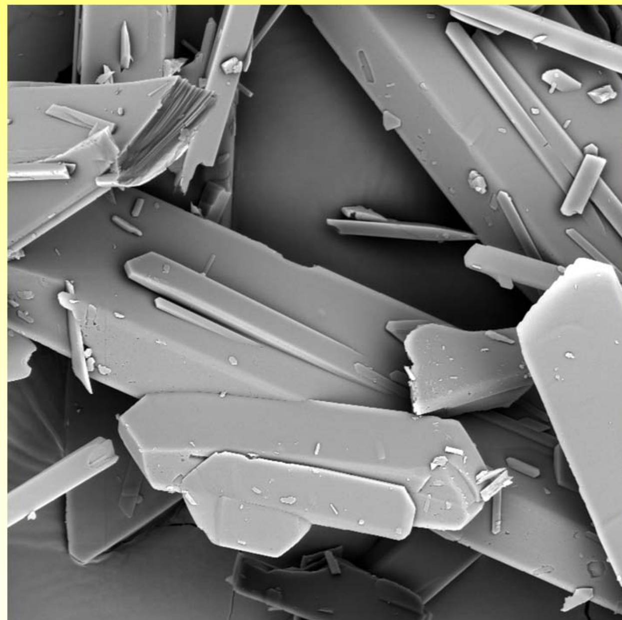
SEM MAG: 5.00 kx DET: BSED
HV: 20.0 kV DATE: 11/09/07 20 um Vega ©Tescan
VAC: HiVac Name: 4000_bse Digital Microscopy Imaging



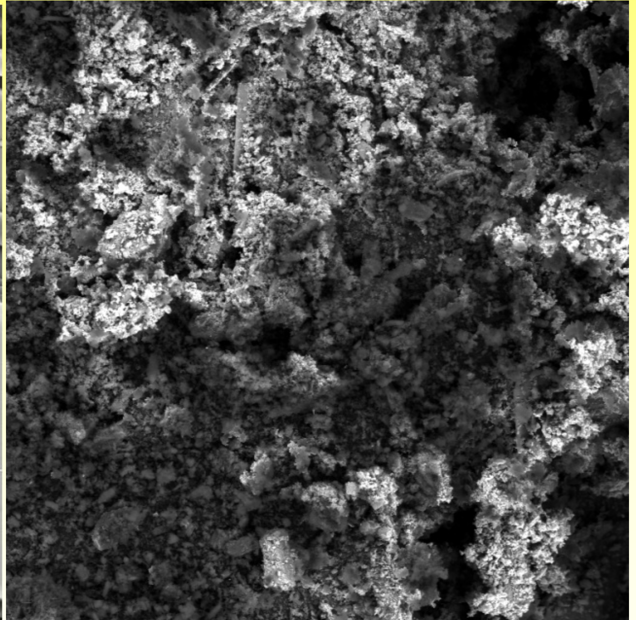
SEM MAG: 5.05 kx DET: BSED
HV: 20.0 kV DATE: 11/09/07 20 um Vega ©Tescan
VAC: HiVac Name: 4000_bse Digital Microscopy Imaging

Crystalization under different conditions

SEM HV: 15.00 kV | SEM MAG: 1.00 kx | MIRA\\ TESCAN
Det: BSE | SM: DEPTH | 50 µm
Date(m/d/y): 09/02/09 | Name: ABAA0449_1_1000x | Performance in nanospace

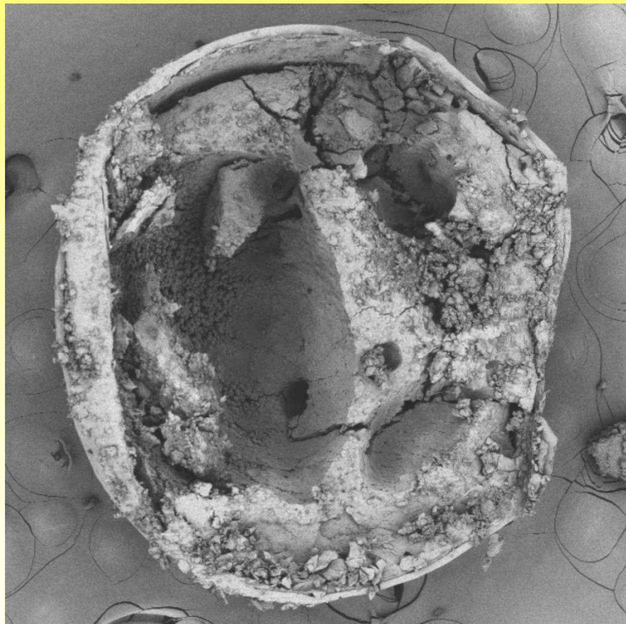


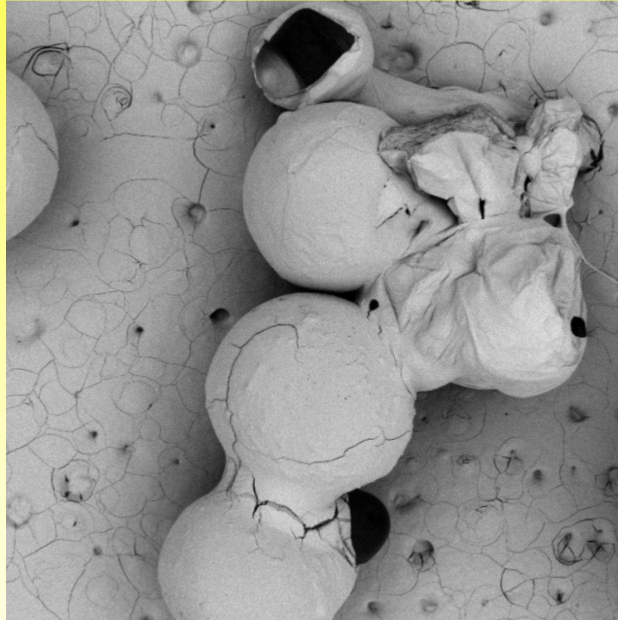
SEM HV: 15.00 kV | SEM MAG: 1.00 kx | MIRA\\ TESCAN
Det: BSE | SM: DEPTH | 50 µm
Date(m/d/y): 11/19/09 | Name: ABAA0449_24_1000x | Performance in nanospace

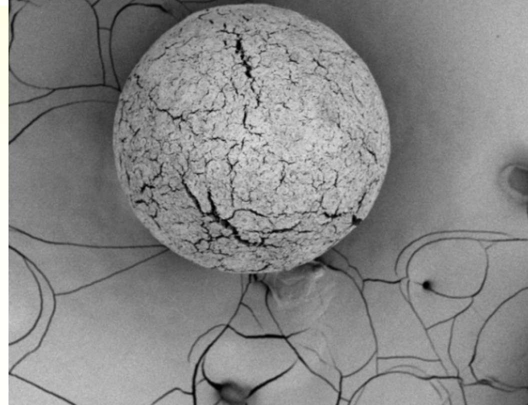


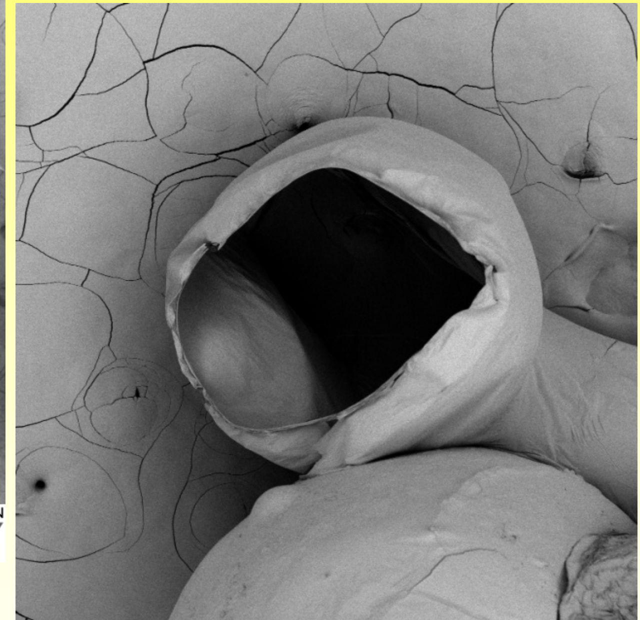
SEM HV: 15.00 kV | SEM MAG: 1.00 kx | MIRA\\ TESCAN
Det: BSE | SM: DEPTH | 50 µm
Date(m/d/y): 10/09/09 | Name: ABAA0449_19_1000x | Performance in nanospace

Pelets after dissolution tests





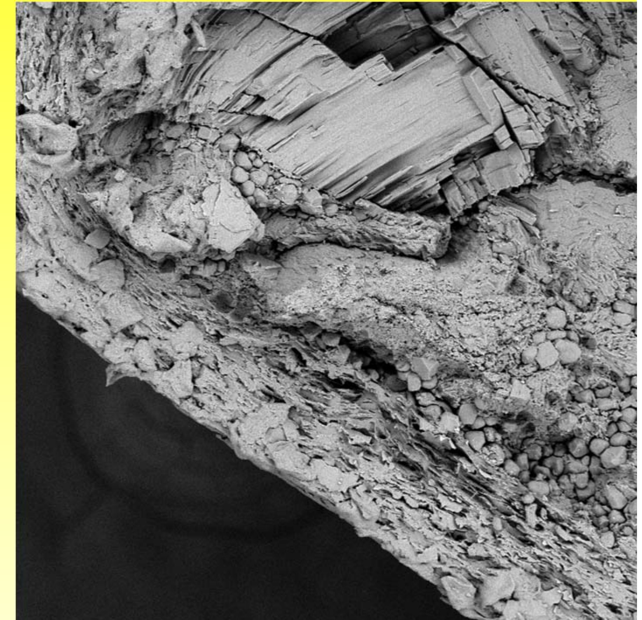




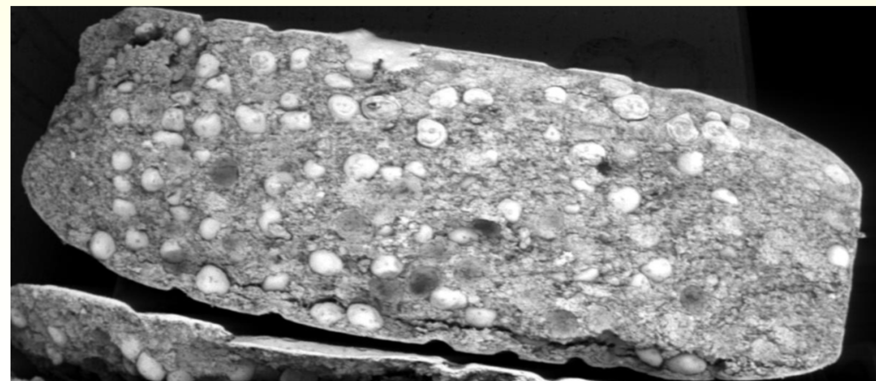
Tablets

SEM HV: 10.00 kV SEM MAG: 500 x MIRA\\ TESCAN
Det: BSE SM: DEPTH 100 μm
Date(m/d/y): 11/24/10 Name: H150_povrch500x Performance in nanospace

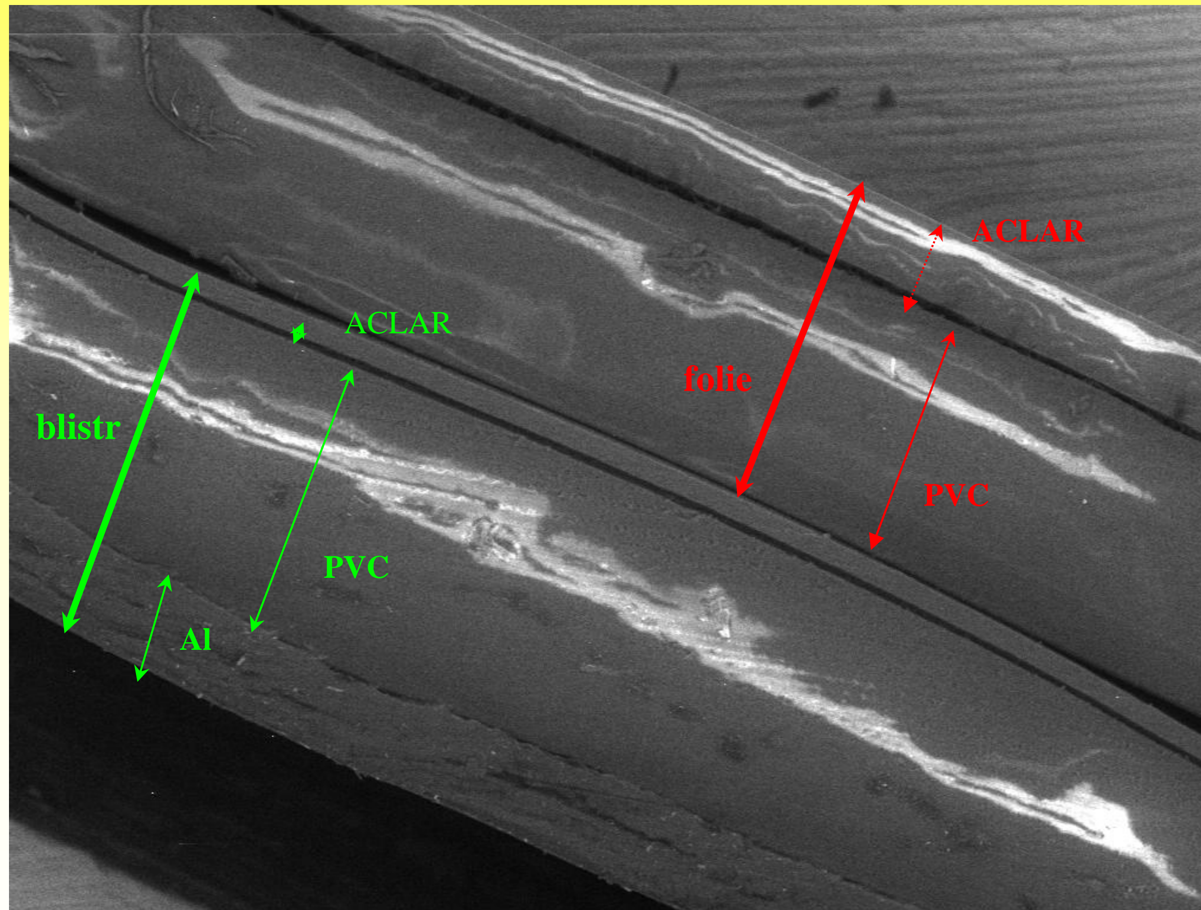
SEM HV: 10.00 kV SEM MAG: 500 x MIRA\\ TESCAN
Det: BSE SM: DEPTH 100 μm
Date(m/d/y): 11/24/10 Name: H150_řez500x Performance in nanospace



SEM HV: 10.00 kV SEM MAG: 500 x MIRA\\ TESCAN
Det: BSE SM: DEPTH 100 μm
Date(m/d/y): 11/24/10 Name: H150_okraj500x_2 Performance in nanospace



Microanalysis – identification of package

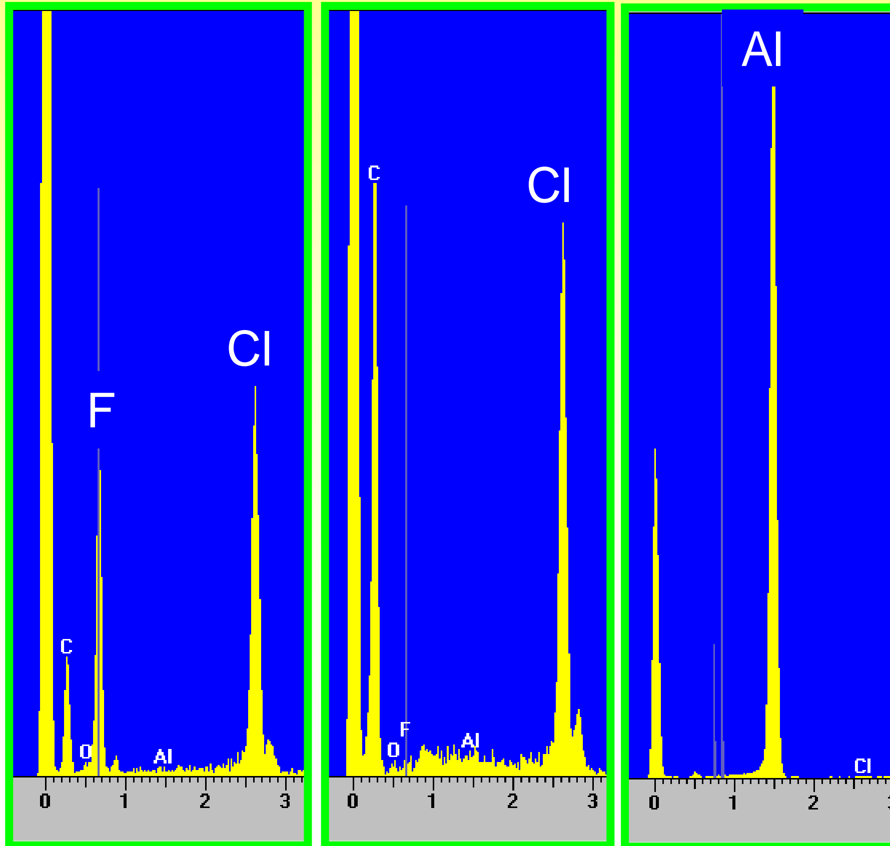


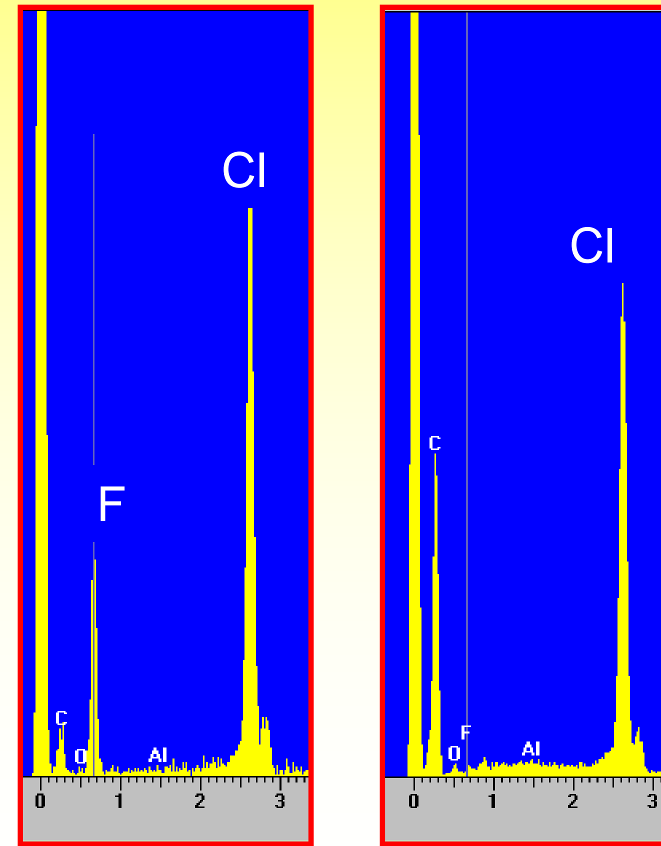
SEM MAG: 287 x | DET: BSED | 200 um
HV: 20.0 kV | DATE: 07/17/07 | Vega ©Tescan
VAC: HiVac | Name: 4 | Digital Microscopy Imaging

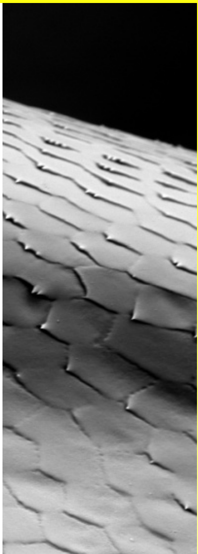
Microanalysis – identification of package

Blistr: ACLAR, PVC a Al.

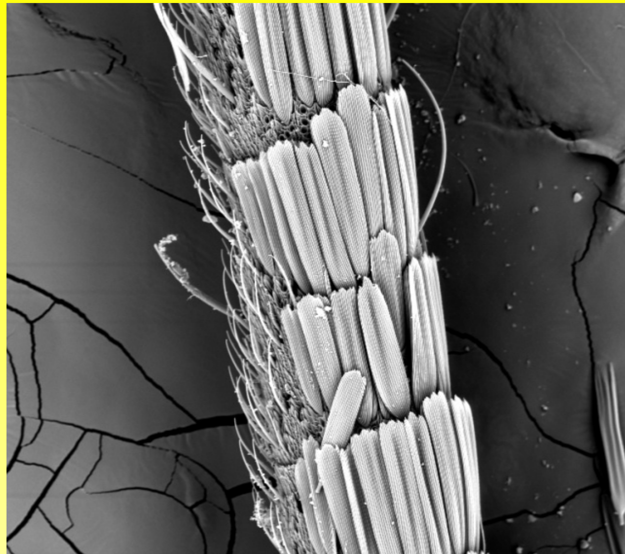
Folie: ACLAR a PVC

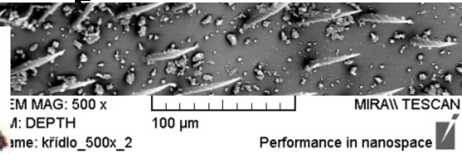




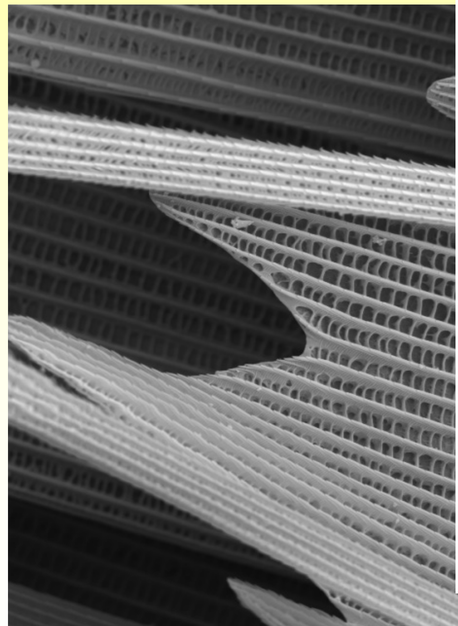


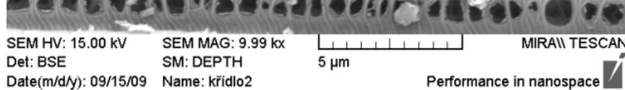
SEM HV: 15.00 kV SEM MAG: 2.01 kx 20 µm Perf
Det: BSE SM: DEPTH
Date(m/d/y): 09/21/09 Name: k_2000x

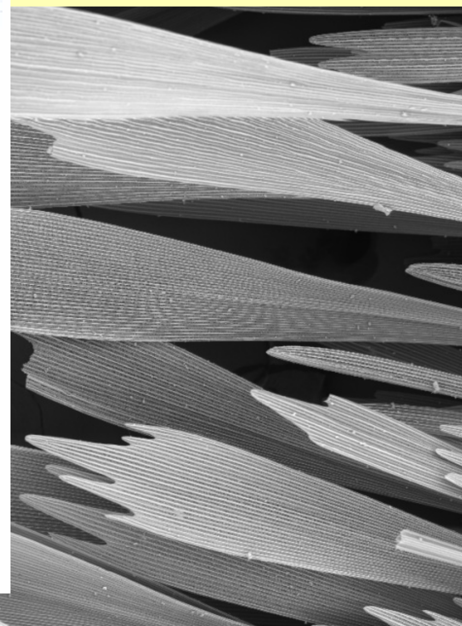




EM MAG: 500 x MIRA\\ TESCAN
/: DEPTH 100 µm
ame: křídlo_500x_2 Performance in nanospace







SEM HV: 15.00 kV SEM MAG: 5.00 kx MIRA\\ TESCAN
Det: BSE SM: DEPTH 10 µm
Date(m/d/y): 09/15/09 Name: křídlo7 Performance in nanospace

SEM HV: 15.00 kV SEM MAG: 9.99 kx MIRA\\ TESCAN
Det: BSE SM: DEPTH 5 µm
Date(m/d/y): 09/15/09 Name: křídlo2 Performance in nanospace

SEM HV: 15.00 kV SEM MAG: 500 x MIRA\\ TESCAN
Det: BSE SM: DEPTH 100 µm
Date(m/d/y): 09/15/09 Name: křídlo4 Performance in nanospace