Chapter 7  GUIDELINE SAMPLE PREPARATION.

Samples come in all types, shapes and sizes, techniques therefore have to be found to cope with all variations and deviations from the perfect sample, a naturally conducting, regular solid providing high emission of electrons. Some of the problems the microscopist is frequently face with can be cataloged as follows:

Fractures

Fractures can be performed on most materials, the outcome is a result of the technique combined with the nature of the sample material. We can first split materials into two grouped metals and non-metals, this also splits the basic techniques.

Metals

Metals will usually be fractured deliberately by mechanical machinery such as an Instrom device, which gradually fractures test pieces of different metals for the study of mechanical properties. Fatigue fractures can be induced but generally are the product of wear or accidental flexing of the metal over long periods of time. The type of fracture reflects the nature in which it was created, therefore care should be taken to produce a representative fracture relative to the nature of study.

Within the laboratory small fractures can be made from steels by inducing a cut partially through the metal and breaking the rest with either shock (heavy weight) or levering after keeping it immersed in liquid nitrogen for some time. Alloys are more elastic and may need cooling in liquid nitrogen before any shock treatment.

Non-metals

This category contains plastics, ceramics, and organic materials. Techniques differ in complexity for these materials but one common condition is that fracturing is best made at low temperature.

Plastics, generally can be fracture by immersing in liquid nitrogen and breaking between two grips when at low temperature.

Some ceramics are not so willing to fracture, without low temperature. They have resilient elastic properties, therefore after removal from the liquid nitrogen a hard impact is usually needed. Others can shatter at the smallest impact and probably do not need low temperature, i.e. silicon wafers.

Organic materials need more care and attention to fine detail otherwise damage due to ice forming and collapse of structures can ruin a fine sample. Because of this problem there is a dedicated equipment known as a "Cryo-system" for
the SEM. This is a method of cooling the sample in an ice/LN2 slush bath, and then transferring the sample to a complete LN2 environment attached to a SEM port. Here it can be fractured, and, or even be coated by sputtering before finally entering the microscope for observation.

Typical samples are food stuffs, plant and animal tissue, semi-solids(fats/oils).

**Sections**

This category can be split into hard and soft materials to distinguish a difference between polishing or cutting methods to produce the open surface of the sample's inner structure.

**Hard samples:** Cross-sections

The hard materials are either composite, i.e. commercial products or dedicated materials such as metals, rocks, ceramics and glass. Some can be embedded in resin or Bakelite such as metals for easy handling when cutting and polishing. Impregnation with resin (Lakeside) is popular for rocks to free the mass of air, water or oil before cutting and polishing.

**Surface preparation**

Cutting methods can be by diamond wheel, or diamond saws, although metals are usually just ground down from a rough surface to a smooth one using different grades of carborundum paper ending with diamond paste down to 1/4 micron.

Rocks need a similar grinding process after cutting, but on glass with carborundum powder, ending again with diamond paste to 1 or 1/4 micron.

Although all of this can be done by hand, there are machines available to ease the grinding and polishing labor.

Electro-polishing a metal to obtain a flat surface has other advantages, because this technique etches some metals with a clean open atomic surface showing orientation or phase contrast mechanisms.

Ion-beam etching can also provide clean open atomic surfaces but one has to be aware of artifacts.

The need for flat surfaces is usually for x ray analysis, WDX more then EDX, good BSE study where small phase differences are to be recognized, and CL to create a shorter route for white light to be seen.

Composite commercial products can be very complex and precise in their content alone. The best method found to date where the contents are not generally disturbed is the reciprocating diamond wire saw. This is a tungsten wire with diamonds embedded along it's length, spooled at both ends and moved from one spool to the other by a polarity changing electric motor. The sample sits between the spools against the wire and is cut.
from two directions. One spool sits partially in a liquid coolant which is picked up on the wire. The coolant serves to lubricate and also clean the wire. The cut can be so good from this device that sections can be made of some products where no further grinding or polishing is necessary.

**Soft samples: Cross/Thin sections**

Samples that require sectioning to give out information should fulfill two requirements to guarantee success. These are, reasonable consistency in hardness and solid enough for metal (sledge) blade or diamond knife sectioning. If the latter is not so it can be overcome by embedding the sample in a resin material to consolidate the sectioning. The hardness consistency is necessary to prevent dragging-out effect of the harder parts that can damage or contaminate the sample, which is undesirable especially if x-ray analysis is to be employed.

These techniques are referred to as microtomy, macro with the sledge type metal blade and micro with the diamond or glass knife. The macro method can be used on, combination materials, embedded materials, plastics, paints, rubbers, wood and paper. Ultra-microtomy is an original TEM technique which for SEM would possibly only be used on heavily metal stained (Os/Pb/U) embedded biological samples to produce sections for negative BSE observation. Low temperature (Cryo) methods are not usually employed as for TEM, unless differing hardness phased polymers are to be investigated. With these technique the surface finish is usually quite smooth, allowing no rough topography to disturb the result.

A further method of sectioning, although somewhat crude but effective, is the scalpel blade. This can be used when the result is not wholly reliant on the topographical surface but the composition, BSE?.

**Dispersions.**

These are some of the most difficult sample types to either prepare correctly onto the substrate, or metal coat with any reasonable success. Observation is usually a battle against charge phenomena as changing bright and dark areas, and or image drift that just will not go away.

This category can be split again into two types for simplicity, wet dispersions (in suspension), and dry dispersions (powders/fibers).

**Wet dispersions: Suspensions**

Polymers, paint pigment and biological cultures can be presented in this manner. The nature of technique will depend on the density of suspensions the size range of the material confined therein, and the importance of overall separation of particles. There are other methods to present biological cultures, depending on the importance of detail
structures because this technique involves air-drying which is not always suitable due to collapse of cell structures.

Dilution.

The correct dilution of a suspension can only be found by making several levels of concentration and testing them all, having logged their liquid to suspension ratio at each step. The liquids used for suspensions are usually:

- Distilled or De-mineralized Water
- Absolute alcohol, Ethanol
- Acetone
- Isopropanol

The choice depends on the chemical properties of the sample material, and or the suspension liquid already present. Acetone can be destructive to equipment used with these methods, therefore extra care needs to be employed if this solvent has to be used.

At each step of dilution the sample should be agitated in an ultra-sonic bath for at least 10 minutes to ensure separation of particles. Testing can be done by pipetting the solutions onto 10 mm die. (alcohol-cleaned) coverslips mounted on aluminum stubs. After the test-samples have thoroughly dried they can be coated with a metal (Au, Au/Pd). When they are observed a correct suspension can be seen to, firstly, not have covered the entire surface of the coverslips with particles, and secondly agglomeration should not be more than 2 - 3 particles high.

A choice of pipetting or atomising the suspension then can be made based mainly on the size and agglomeration effects seen on the test-sample. The size range for pipetting is far larger, sub-micron to hundreds of microns, where atomising is more limited from sub-micron to a few tens of microns. Size tends to dominate the choice. Agglomeration of material that will be pipetted can be further agitated in the ultra-sonic bath to separate particles Be aware that excessive use of ultra-sonic agitation can lead to sample damage if the sample is multi-structured. Atomising helps to disperse agglomerates of fine size, but it is always useful to ultra-sonic the suspension beforehand.

**Pipette**

This method is a matter of drawing up sufficient suspension immediately after agitation and placing 1 or 2 droplets on a prepared substrate, allowing it to dry, and then coating by sputtering. A new pipette should be used for each sample so not to cross contaminate samples. The result will be a ring of grouped particles showing varying density over the ring and
separated or small agglomerates within the ring.

**Atomize**

This involves more equipment and an artistic temperament rather than a scientific one. Atomising pens or Airbrushes are commonly used by commercial artists to create color pictures for advertising. These have the advantage of allowing size change at the dispersing nozzle (aperture) so that a particular size range of particles can be sprayed. After agitation the suspension is loaded into a small bottle attached to an atomising pen (Airbrush). Spraying directly onto a substrate is not advised as this creates a fast build-up of particles and possible agglomeration.

Spray at a 45° angle into a vertical tube approximately 10cm dia across by 30cm long, with the sample substrate sitting at the base of the tube, ready to collect gravity induced spray. The length of time to spray unfortunately is rather trial and error once more, but with 2 - 3 sprays this usually can be established. It is vitally important to thoroughly clean out the pen spray nozzle with before changing sample, but samples can be loaded into separate bottles for attachment. **Acetone should not be used with an atomizer, as dissolvable parts may be in use.**

**Sandwich technique**

As some sample materials from suspensions are difficult to observe due to charging problems, a sandwich technique can be employed to trap the material between two conducting layers. A substrate 10mm coverslip is washed in alcohol, dried and placed with adhesive onto an aluminum stub. The substrate is then sputter coated with gold so that a thick uniform layer is laid down. The substrate is then exposed to a shorter coating but at higher current so as to crack the first uniform coating. This leaves a layer of gold with deep cracks of very small width. The sample suspension is dispersed over the gold and allowed to dry, capillary action draws the particles down to the surface due to the cracks in the gold. A second coating of any sputtered metal can now by laid down onto the sample to sandwich the particles and improve their conductivity. The second coating should be thinner and with less current than the first two coatings to retain sample integrity.

**Separating size: Cascade Impactor**

Sometimes it is simpler to separate the size ranges of material suspensions by other methods than when they are already placed in the SEM, where complex image analysis techniques may have to be employed. The Cascade Impactor is a device with many capture positions for particles size, starting
at the largest and ending with the smallest. A vacuum is drawn through the device at the bottom end and a dispersion is sprayed into the top end, at each size position there is an aperture backed by a collecting substrate, and the relative size particles are trapped through the aperture. As the apertures range from large to small, the largest particle size stay at the top of the device and the smallest size at the last a position. Any combination of progressively smaller or intermediate apertures can be added to give even finer separation of the ranges.

**Dry dispersions: Powders/fibers**

Non-metal materials that come as powders when mounted for the SEM are well conducting. Choosing the adhesive well in the first place can help to prevent unnecessary charging later. This is very important if x-ray analysis is to be used.

Metal powders are not usually a problem, but adhesives such as double sided tape should be avoided as it has been known to contain traces of Zinc carbonate, which can be picked up by reflection and show on the spectrum. Fibers pose a more difficult problem due to their loose conductive connections to each other and to the substrate. By anchoring one end of the fibers to the stub with adhesive, allowing to dry, and then pulling the fibers over the stub and repeating the procedure, a reasonable sample can be made. The excess fiber can be trimmed away. There are special aluminum stubs (2 parts) made to hold fibers. It is wise to use carbon or silver paint with these stubs just to be sure of good contact. Choosing a good background may be important as fibers of all kinds usually have a sub-structure. Organic fibers favor low Z backgrounds such as carbon. Sputter coating should be done with care as comes especially organic fiber can be damaged easily by thermal radiation.

**Adhesive choice**

Since the introduction of the sticky carbon disc (tab) mounting powders of fine sizes is much easier.

By dispersing the powder on a clean surface and pressing the carbon disc mounted stub down onto the powder, and trapping the excess away. a suitable sample can be made. Coating can now be performed. If the powder is likely to out gas the previous technique can be employed but care should be taken to degas the powder well before coating, so that the coating is well applied and all around. Carbon paint is better in these circumstances as the particles are held firmer and into the adhesive. After outgassing a sputter coating has more chance of bridging the area between particle and adhesive.
Filters

Collection of powder or fiber material on filters is not the best method for the SEM, as out gassing and conductivity can be at its worst with this combination. If they have to be used then cellulose acetate filters (nuclear-pore) are preferred over fiber types as they are easily dissolved, if necessary, and when coated give a more uniform conductivity. To help in difficult situations where charging is known to be serious, first carbon coating can be applied to set up a bond between the sample and the metal coating later sputtered on top.

Live material.

Insect

Insect samples, if fresh need to have their fats removed before they can be placed into a vacuum. The sample should be immersed into ethyl-alcohol for at least 1 - 2 weeks to dissolve the fats. It then should be washed in fresh ethyl-alcohol to remove surface fats and allowed to dry on a filter paper. If this preparation is not done fats will be observed on the surface of the insect, and in the worst case the insect could explode within the SEM chamber.

When mounted (carbon paint), first a coat of carbon to help bond the surface, followed by a heavy coat of gold (Au) or gold palladium (AuPd).

If one is just interested in looking at a fly for a presentational exercise, then search for a spiders web and allow the dry out method be done by a spider. It is quite effective and will be ready in 1 - 2 days.

Plant

Fresh plant material should be looked at after cryo treatment and at cryo temperatures within the SEM to give accurate observation of the surface structures.

For a show time (15 minutes) one can observe plant material at voltages less then 1kV, with no coating. After that time the structures collapse due to the vacuum. Carbon discs are very handy for this crude technique.

Replicas

Where necessary?

Replicas are necessary when either the bulk of the object is too large for the SEM and cannot be cut, the material is bad for the vacuum or cannot be exposed to the beam. Further, holotype (geological) samples generally cannot be coated and replicating is the only option.
There are two main methods using different materials, simple cellulose acetate sheet replication and two part silicone rubber replication.

**Cellulose Acetate sheet**

This is used mainly on polished and etched metal and ceramic surfaces. It can be used to extract material for X-ray Analysis such as precipitates on grain boundaries. The surface to be replicated must be able to withstand Acetone as this is used to allow the cellulose to flow into and around all structures. Acetone is flooded over the surface of the sample and the cellulose acetate sheet is lowered from one side onto the surface so as to slowly force the acetone across it. The soft sheet is allowed to solidify before it is slowly pulled from the surface. This forms a negative replica which can be cut to size or area of interest and coated with a metal or carbon. This type of replica can be highly detailed and have been used for high magnification.

**Silicone rubber (Silica-set)**

Silicone rubber can be used on most surfaces, it is only slightly exothermic but should not affect plastics or paint surfaces. It has be used to great effect in geological research for replicating micro-fossils and casts. The method uses a two component silicone rubber, silicone rubber with plasticizer and the catalyst liquid. Firstly, the object or surface to be replicated should be free from dust or loose particles, then coat it with a thin layer of the silicone component application can be with a tine artists paint brush. Secondly, mix the two components as recommended by the manufacture of the product and then when well mixed apply to the already wet surface. Leave to set for the recommended time. A thin layer of silicone rubber will cure slower but too much bulk is not preferred for the vacuum of the coating system. Coating with carbon first is recommended as silicone is not easy to place sputtered metal directly on. Although this method is capable of high detail. it is generally used for low magnification work.

**General mounting media**

The most common object used in scanning electron microscopy is the mounting stub of one or another shape and fitting. Further more this is usually made of aluminum although other materials such as carbon and copper are used for specific observation and analysis techniques. The reasons for tile widespread use of aluminum mounting stubs are base on it's good conductivity, low cost and easy shaping. The three main material substrates Al, C, and Cu can be characterized with their applications.
Aluminum

The aluminum stub can be utilized in most general SEM modes of observation and analysis. Other than the standard 15mm and room diameter types there are available pre-angled stubs (30, 45 and 60 degree), or with clamping screws to hold a thin object in cross-section, or with a ring to trap thin fibers or fabrics. and of course, pre-coated with adhesive for convenient use.

Usage:
Secondary imaging Backscattered imaging (Composition and Topography)
Cathodoluminescence imaging
Semi-quantitative EDX
Image analysis

Of course care should be taken not to unnecessarily expose the aluminum to the beam of electrons, especially in the analytical modes, to avoid spurious values entering the statistics.

Carbon

Carbon mounting stubs are used for particles or powders, which need xray analysis, but only contain elements above carbon in the periodic table. This prevents interference which would be present with a metallic substrate.

Usage:
EDX X-ray analysis

A further use of the carbon stub is for it's low atomic contrast shown in the backscattered mode, where material mounted upon the surface can be isolated quite easily from any background effects. Carbon atomic contrast appears black in backscattered mode.

Usage:
Backscattered imaging (Composition)
Image analysis

Copper

This type of substrate usually has a specific cup like shape for containing a temperature or beam sensitive sample, and holding it at a low temperature. The copper stub is cooled down by immersion in liquid nitrogen and then, with the sample mounted on a separate metal block is placed in the vacuum of the SEM. This method can be used successfully on some samples such as polymeric or biological, but can suffer from ice-up if the time taken between leaving the liquid nitrogen and entering the vacuum is too long. This is no substitute for proper Cryo techniques which allow regulated low temperature conditions.
to be used within the SEM.

Usage:
Secondary imaging
Backscattered imaging (Topography)
Cathodoluminescence imaging
EDX, X-ray analysis

Further Analytical mounts

Analytical 1.0 and 1.25 inch mounts. These are made from either 2-part conductive or non-conductive epoxy resin (cold setting), or Bakelite conductive or non-conductive (hot or cold setting). Whichever is used the sample is embedded within the resin or Bakelite which is molded into the necessary diameter. The upper surface is ground and polished flat to possibly 0.2s micron of fatness. The thickness of the resulting block will depend largely on the sample dimensions. This technique has been used for many years in the metallurgy field but has also been found useful with the analysis of ceramics, composite materials, glasses, paint, powders and whole IC devices amongst others. Because of the possible non-conductivity of these substrates, conductive coating is usually needed. The advantage of having samples mounted in this manner for ray analysis, especially WDX, is that all samples can be brought to a common working distance with an analytical sub stage for accurate quantitative analysis.

Usage:
Backscattered imaging (Composition)
EDX, X-ray analysis
WDX, X-ray analysis
Image analysis

Note:
Stub mounts made from brass should be avoided if possible due to their tendency to oxidize and some porous, with later consequences of out gassing during use.

Mounting adhesives

Adhesives (liquid / semi-solid)

There are an overwhelming number of adhesives available for general or specific purposes and rightly so the preparation for SEM samples demands specific types to allow an electrically conductive connection between substrate and sample surface. The adhesive must also firmly fix the sample so as not to allow mechanical drift when tilted or thermal drift when irradiated by the beam. Finally the adhesive must be able to withstand the vacuum of the SEM wisdom out gassing continuously.

Many household adhesives are therefore not suitable, and care should be taken not to use
advanced materials such as Super-Glues as these can out gas carcinogens which may find their way out the rotary pump exhaust.

Notice should also be area of the solvent in the adhesive as this may prolong outgoing or affect the sample materials or also be detrimental to health.

The more common solvents generally used in well known adhesives can vary between Toluene, Xylene, Benzene, Dichloro-ethylene, Amyl-acetate, Acetone. Alcohol, Pentane, Ketone, and water plus others that little is published about for one reason or another.

**Double-sided tape**

The convenience of double-sided tape for holding samples caused its widespread use in microscopy, unfortunately when it comes to the SEM there are some problems associated with this medium that limit its use. For instance it is non-conductive and does not lend itself to being coated correctly (charging problem). Further it does not hold material securely (tilt problem). Left over time it can affect the sample material due to the solvent contained within. It outgasses continuously, and some types contain mineral deposits (background for X-ray analysis). Some are thought to be carcinogenic.

**Metallic tapes**

Metallic tapes generally have the same adhesives as double-sided tape (non-conductive), but exposure to the beam is usually minimal. The need for securing, usually irregular shaped samples, or a conductive path to the sample holder and the sample surface promote the use of metallic tapes. Use of these tapes are usually a matter of convenience then good practice.

**Conductive metallic tapes**

Metallic tapes should not be confused with "conductive" metallic tapes. which have a conductive adhesive as well as being of copper or aluminum. The latter are more frequently used for forming the all important earth connection and a single piece can be repeatedly applied to successive samples.

**Wax or oil based mounting media**

Paraffin wax, Sailor's wax and Canada balsam are used widely for mounting samples for light microscopy. Under white light these mediums have little problem, but under the electron beam they can be highly unstable and are likely to contaminate parts of the microscope. Wax and oil based mounting media should be avoided.
Plastercine ® may appear convenient but intact is oil based, and under vacuum will start to dry out. If one needs to use this type of mounting medium choose the Carbon plast as a well proven alternative.

**Mounting Adhesives**

The various mounting media used in scanning electron microscopy can be found in most small item suppliers catalogs for microscopy. The most common are:

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<thead>
<tr>
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</thead>
<tbody>
<tr>
<td>Gold paint</td>
<td>ketone/pantane</td>
<td>permanent</td>
<td>conductive</td>
<td>small items /high resolution</td>
</tr>
<tr>
<td>Silver paint</td>
<td>ketone/pantane/amy1-acetate</td>
<td>permanent</td>
<td>conductive</td>
<td>general samples /imaging modes</td>
</tr>
<tr>
<td>Copper paint</td>
<td>ketone/pantane</td>
<td>permanent</td>
<td>conductive</td>
<td>large items /imaging modes</td>
</tr>
<tr>
<td>Carbon paint</td>
<td>ketone/pantane</td>
<td>permanent</td>
<td>conductive</td>
<td>Xray analysis /low Z background</td>
</tr>
<tr>
<td>Colloidal carbon</td>
<td>Water</td>
<td>temporary</td>
<td>conductive</td>
<td>Aqueous solvent only</td>
</tr>
<tr>
<td>Carbon plast</td>
<td>???/semi-solid</td>
<td>temporary</td>
<td>conductive</td>
<td>Xray /irregular sizes and shapes</td>
</tr>
<tr>
<td>Carbon tabs/disc</td>
<td>???/semi-solid</td>
<td>Temporary with bulk samples</td>
<td>conductive</td>
<td>Xray analysis / bulk / powders</td>
</tr>
<tr>
<td>Microstick</td>
<td>dichloro-ethylene</td>
<td>temporary</td>
<td>non-conductive</td>
<td>Micro particles /powders /imaging</td>
</tr>
<tr>
<td>Tempfix</td>
<td>???/solid &lt;40° C</td>
<td>Permanent &lt;40° C</td>
<td>non-conductive</td>
<td>Non-sinking media powders /delicate samples</td>
</tr>
</tbody>
</table>
Background

To give a guideline indication of the type of background available considering the materials likely to be used the list is composed as follows.

<table>
<thead>
<tr>
<th><strong>Background material</strong></th>
<th><strong>Best method of application</strong></th>
<th><strong>Texture smooth-middle-rough</strong></th>
<th><strong>SE Contrast light – gray - dark</strong></th>
<th><strong>Magnification range / Detection method</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td>Aluminum stub</td>
<td>polish surface to 1 um</td>
<td>low mag - circles high mag smooth</td>
<td>high Kv – light low K – gray</td>
<td>10x – 20 kx SE/ BSE/ CL</td>
</tr>
<tr>
<td>Glass cover slip 10 – 15mm dia.</td>
<td>wash with alcohol Au sputter coat</td>
<td>fine coat – smooth heavy coat - middle</td>
<td>high Kv – light low K – light /gray</td>
<td>10x – 50kx CE / CL</td>
</tr>
<tr>
<td>Glass cover slip 10 – 15mm dia.</td>
<td>wash with alcohol Carbon coat</td>
<td>fine coat – very smooth</td>
<td>high Kv – light (Si) low K – dark (C)</td>
<td>10x – 100kx SE / BSE / low voltage</td>
</tr>
<tr>
<td>Silver paint</td>
<td>mix well, apply with stick, allow to become tacky</td>
<td>rough / flakes</td>
<td>all Kv's - light</td>
<td>10x – 250 kx SE / CL</td>
</tr>
<tr>
<td>Carbon paint</td>
<td>mix well. apply with stick, allow to become tacky</td>
<td>middle / flakes</td>
<td>all Kv's - dark</td>
<td>10x – 1000x SE / BSE / CL / EDX / Ia</td>
</tr>
<tr>
<td>Carbon tab/disc</td>
<td>sprinkle or press sample onto surface</td>
<td>smooth</td>
<td>all Kv's – dark (can charge at high Kv)</td>
<td>10x – 5-kx + SE / BSE / CL / EDX / IA / low voltage</td>
</tr>
<tr>
<td>Microstick</td>
<td>paint on substrate fast evaporation</td>
<td>as substrate surface</td>
<td>as substrate, or as metal coating</td>
<td>10x – 1000x SE / BSE / low voltage</td>
</tr>
<tr>
<td>Tempfix</td>
<td>dissolve at 40 °C onto substrate</td>
<td>very smooth</td>
<td>Au coat – light C coat – dark</td>
<td>10x – 50kx + SE / BSE / EDX / IA / low voltage</td>
</tr>
<tr>
<td>Fiber (fiber)</td>
<td>sample material absorbed onto surface</td>
<td>rough</td>
<td>uncoated – charge Au coated- gray</td>
<td>10x – 150x SE / CL / low voltage</td>
</tr>
<tr>
<td>Fiber (neclear)</td>
<td>sample material absorbed onto surface</td>
<td>middle / smooth</td>
<td>Uncoated – charge Au coated – light C coated - gray</td>
<td>10x – 1000 + SE / BSE / CL / low voltage</td>
</tr>
</tbody>
</table>
This is a subject that is generally forgotten until it is too late, but with a little forethought one can produce the best available background to suit the bad subject at hand. Correct choice of the mounting stub or the adhesive can be very important to determine the contrasting background. Various simple materials or metals can help to produce good quality photograph.

Coating with metals or carbon can change the final contrast and detection modes possible, nonconducting backgrounds are therefore recognized as possibly being coated in the chart.
Secondary Electrons (SE)

To help with decisions made about the preparations of your own particular samples, here is a list of Materials which are frequently presented to FEG-SEM. The list is not meant to be specific in each field but to give a general guide as where to begin

**Secondary Electron (SE) Chart for FEG-SEM**

<table>
<thead>
<tr>
<th>Sample type</th>
<th>200 volts</th>
<th>1</th>
<th>2</th>
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<th>15</th>
<th>20</th>
<th>25</th>
<th>30kV</th>
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<td></td>
<td>low voltage</td>
<td>nonconductors</td>
<td>conductors</td>
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<td>Metals</td>
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<tr>
<td>Rocks</td>
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*Note: The chart includes a list of materials such as Metals, Ceramics, Rocks, Silicon, Glass, Semiconductors, Bio-Medical samples, and Powders. Each category contains subcategories with specific materials and preparation methods.*