Book of Tutorials and Abstracts

European
Microbeam Analysis Society

University of BRISTOL

Mineralogical Society

EMAS 2018

13th EMAS Regional Workshop

MICROBEAM ANALYSIS IN THE EARTH SCIENCES

4 - 7 September 2018

University of Bristol, Wills Hall, Bristol, Great Britain

Organised in collaboration with:
Mineralogical Society of Great Britain and Ireland
and
University of Bristol

PREPARATION OF EARTH SCIENCE MATERIALS FOR MICROBEAM ANALYSIS

J. Wade

University of Oxford, Department of Earth Sciences
South Parks Road, Oxford OX1 3AN, Great Britain
e-mail: jon.wade@earth.ox.ac.uk

1. INTRODUCTION

Although often overlooked, sample preparation often plays a key role in an analytical campaign, determining both data quality, and the analytical technique suitable for the prepared sample. An obvious example being where a sample, initially prepared in resin for SEM and EPMA analysis, subsequently appears promising for SIMS based light element isotope analyses. If the resin used is unsuitable for high-vacuum devices and is prone to outgassing, the sample may never make it past the load lock. Additionally, the outgassing resins – hydrocarbon based – will limit the detection limit, accuracy and precision of the resultant analysis. Outgassing resins are unlikely to get the user on their favourite SIMS lab's Christmas card list!

Before starting any sample preparation, it's worth pausing to consider a few things ….

- How valuable are these specimens? In other words, must they be returned to a museum in a pristine state or can you do as you please with them?
- What do you hope to do with the sample? If your 'quick look' on the SEM reveals a really interesting feature, will you then wish to use another analytical technique to investigate it further?
- Do your samples fit inside the instrument chamber, or if they are on glass slides, will these fit the instruments sample holder?
- Do the samples need mounting in a media for preparation? If so, is that media – often an organic resin of some sort – compatible with the analytical instrument?
- Do you wish to merely look at the samples, or will you require some form of quantitative analysis – in which case do we need flat, polished sections?
- If we can coat the samples, what type of coating (e.g. carbon/gold/other metal) is appropriate?

In short, don't do something to your sample you may later regret …..!

2. PRISTINE MATERIAL

In many cases the primary goal is to image a sample surface without any alteration i.e., in its raw state. This may be done either because of conditions attached to loan of specimens – frequently the case with some fossil material, or because sample cleaning may change the nature of the material you want to look at. A big consideration here has to be whether you will observe the sample in a vacuum chamber and if so is it low vacuum (or variable pressure - VP) or high vacuum (HV) or even ultra high vacuum (UHV) mode. For techniques that rely on a charged beam, such as imaging in an electron microscope, this is determined by the electrical conductivity of your sample and if an insulator, whether it can be coated with an electrical conductor. In other words – will you be coating the specimen?

If the sample is to be imaged in a low vacuum mode, surface cleaning sometimes significantly improves the signal – this can be done by using ultrasonic baths and then oven drying. Use of a variety of solvents is possible, but these can be fairly aggressive to some materials. It is often only necessary to use a gas duster, ensuring the sample does not end up in the sample graveyard of the lab floor.

3. *SAMPLE MOUNTING*

For analysis in an SEM, there are a variety of mounts available, depending on size of the sample (Fig. 1). Use the appropriate holder – this is particularly important when samples are to be tilted to high angle, such as in EBSD analysis. Each instrument design will have a different limit of working distance and maximum tilt angle to ensure the stage does not collide with other components in the chamber. By using the correct holder – either a pre-tilted holder or one that 'stands-off' the sample from the stage, these (expensive!) incidents can be avoided.

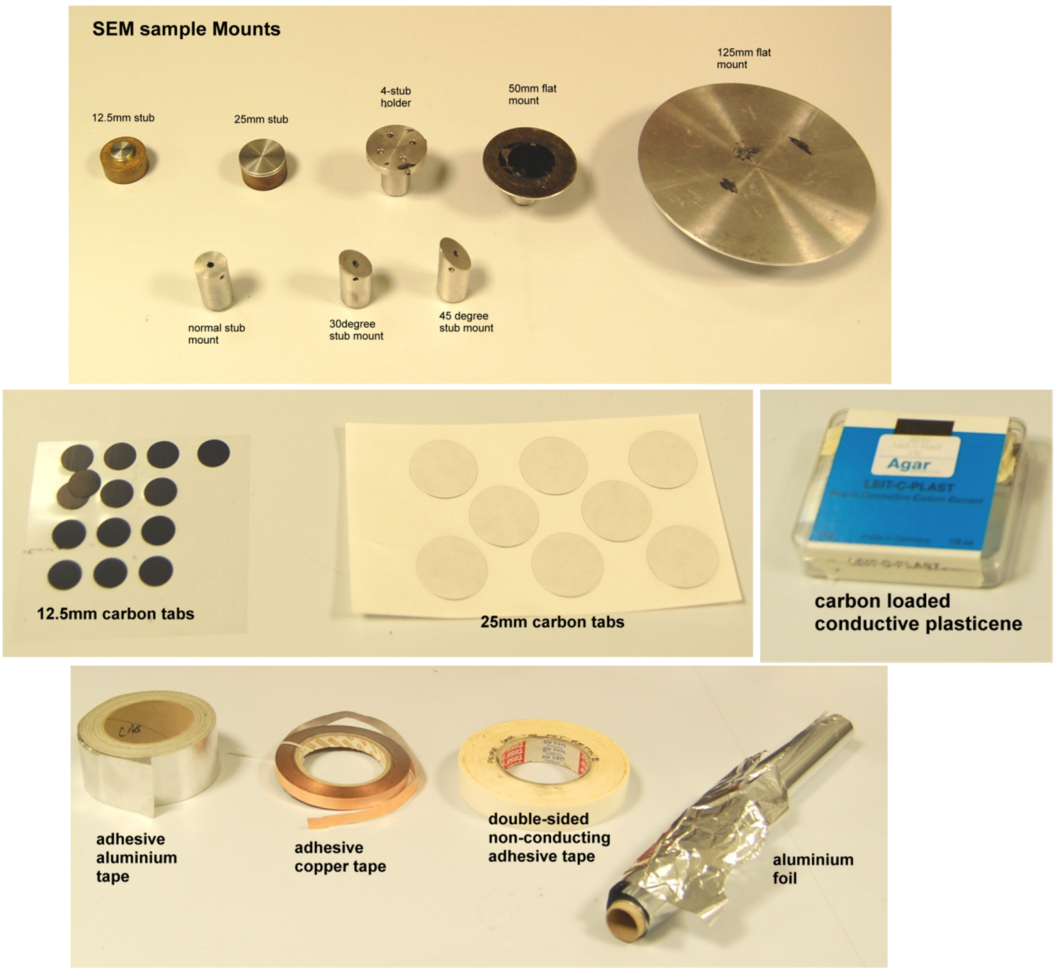


Figure 1. A variety of sample mounting materials is available to ensure sample is properly presented to the electron beam.

For mounting the sample to a stub or holder, double sided tape or carbon tabs will improve the adhesion of the sample to the mount but these are not always necessary. If the sample is to be coated then it is essential that the coating path is continuous from the part of the sample you intend to observe through the holder down to ground potential through the instrument. Any break in continuity will result in charging problems. Often when using pristine dielectric material it is beneficial to wrap it in conductive foil to aid charge neutralization even when using VP mode. When mounting very small particulate matter, it is a good idea to make sure all the excess, un-stuck, particles are removed prior to placing in the chamber – a gas duster is often sufficient. Apart from potential damage to the turbo pump (the filter prior to the turbo pump is usually only a coarse gauze so as not to inhibit pumping), there is an outside chance of loose particulates puncturing any ultra thin windows of for example, an EDS when the chamber is vented.

4. SURFACE PREPARATION

For many quantitative techniques, e.g., EPMA or SIMS, as well as SEM-BSE work, samples must be both flat AND polished (one does not imply the other). For example, for quantitative techniques that utilise X-rays, such as EPMA, a rough surface implies that the X-rays must follow an unpredictable path length before exiting the sample's surface (Fig. 2). If the sample surface is uneven, either from a lack of preparation or poor polish (scratches, voids, etc.), the path length of the X-ray through the sample is different to that in a sample with a flat surface. Importantly, the matrix correction routine by which raw counts from both sample and standard are converted into a quantified concentration, assumes that the path length within the sample and standard is similar. Additionally, as the surface tilt increases, so the proportion of electrons that are backscattered from the sample increases, decreasing the number X-rays generated. Finally, if the sample is polished but not flat (i.e., tilted towards or away from the detector), the path length for the X-rays to exit the sample is, obviously, greater or lesser than that for a flat sample. This change in path length has the greatest effect on light element X-rays, which are preferentially absorbed by the matrix. Poor or rough surfaces may therefore significantly alter the relative intensities of the various X-ray peaks (e.g., Fig. 3). In short, the matrix correction routine is predicated on a number of assumptions, namely that the sample and standard are geometrically similar, or in other words, flat and polished.

If a WDS or EDS analysis is 'bad', frequently the finger can be pointed at the sample preparation. Most samples are presented to the electron probe either as 'thin' sections mounted on a slide (check the slide size first – they are **not** the standard biological size slides) or as a resin mounted polished block. Obviously, if small (~1 to 10 μm) particles are being analysed, depending on incident beam energy, some electrons may penetrate the substrate, whilst electrons may also exit the sides of the particle – this will produce a low yield of X-rays (and hence low analytical totals) originating from the particle and a significant number originating from the substrate. In this instance, optimisation of the analytical conditions may be required.

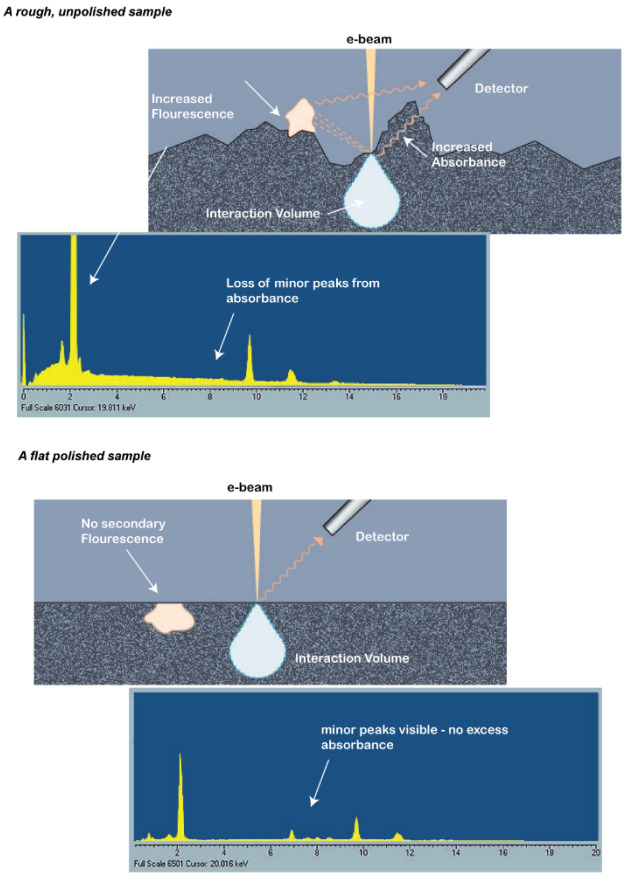
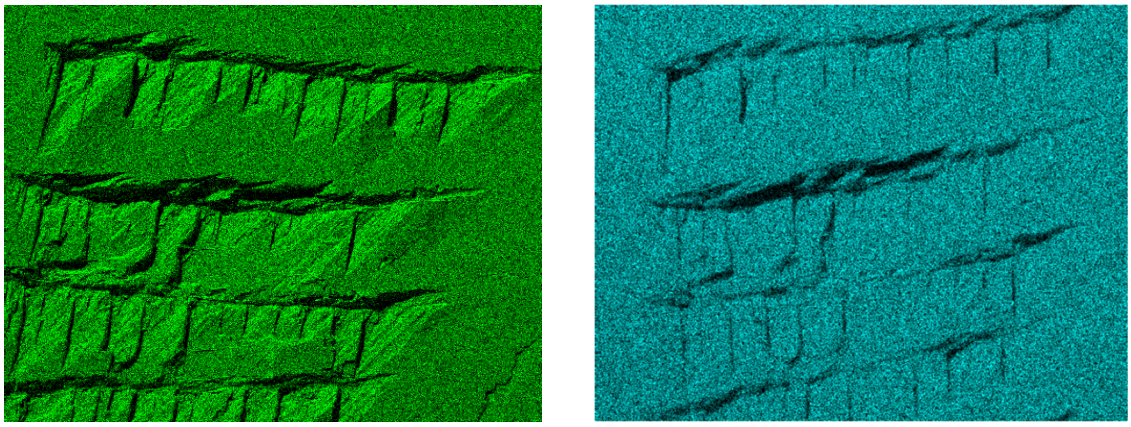


Figure 2. Rrough versus polished samples, showing excess absorbance and fluorescence arising from the rough sample (Figure taken from Oxford Instruments).



A B

Figure 3. EDS image of oxygen Kα (A) and Fe Kα (B) X-rays in homogenous calcite. The darker shadowed areas in this case obviously result from a gouge on the samples surface with the EDS detector orientated on the backside of the feature. Orientation of a feature facing the detector would result in apparently high O (the cleavage plane trending NE-SW in the oxygen image), rather than the low seen here. Note this is not visible for higher energy iron X-ray). This demonstrates the importance of a flat surface when interpreting x-ray images. Similar topographic effects are also commonly seem in ion-probe images, where edge effects are prominent.

Poor sample preparation also causes significant issues for SIMS users. Unlike an SEM or EPMA, SIMS instruments run at significantly better vacuums, such that resins that exhibit significant outgassing, either through incomplete resin/hardener mixing or the presence of bubbles. Topography from bubbles on the surface results in an distorted extraction field (10 kV over a distance of 4.6 mm), while outgassing may generate an arc-over of the e-beam (Fig. 4). This nanosecond arc results in significant damage to the specimen, drilling a hole into the epoxy, and the surrounding grains vanish somewhere into the instrument and extraction lens is coated with epoxy. At best 2 days lost. At worst, a grain ends up-stream in the instrument , in a bad position and charges up. The further into the instrument they go, the more work involved in its removal and the less popular the user. The cross-section (below) shows its electron beam damage goes deep within the sample resulting from this nanosecond arc.

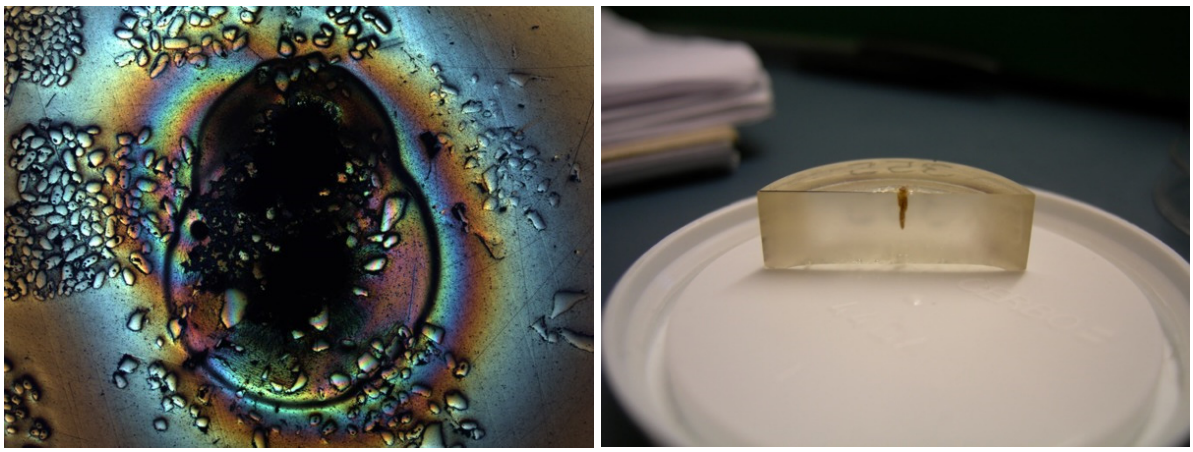


Figure 4. Left - sample damage and grain removal in the ion probe resulting from an e-beam arc-over, and right, cross-section showing the depth of damage (pics courtesy of Edinburgh Ion Probe facility).

5. MOUNTING RESINS

Many resins are available, and which one used may depend on the final use of the sample and also the type of sample. The various commercially available resins have a range of physical properties – such as hardness, thermal stability, curing time and shrinkage, as well as variable vacuum stability/suitability. Acrylic resins (e.g., Beuhler's Varidur) given good edge retention and are attractive to the time-pressed experimentalist as they cure very rapidly at room temperature – typically in the time it takes to make the tea. Unfortunately, mixing the correct hardener to acrylic powder is not easy, and the mounts are often contain bubbles making them unsuitable for anything other than low-vac, VP work.

Epoxy resins possess a range of cure times – Petropoxy is ideal for optical thin sections, cures within 20 minutes at 110 °C, and has excellent penetrating properties for back-filling voids in samples. It is, however, not suitable for sample destined for high and ultra-high vacuum devices (e.g., ion probe) due to its high out-gassing behaviour. The Struers/Beuhle**r** two-part epoxies are preferred for these purposes, but typically take ~ 24 hours to set. Ensure that hardener is well mixed with resin, and accurately measured. All epoxy resins readily absorb water which effects both hardness of the final block and outgassing behaviour (Fig. 5). If possible, ensure that the resins are mixed in a dry environment (less than 50 % r.h) and manufacturer's mixing times and quantities are adhered to.

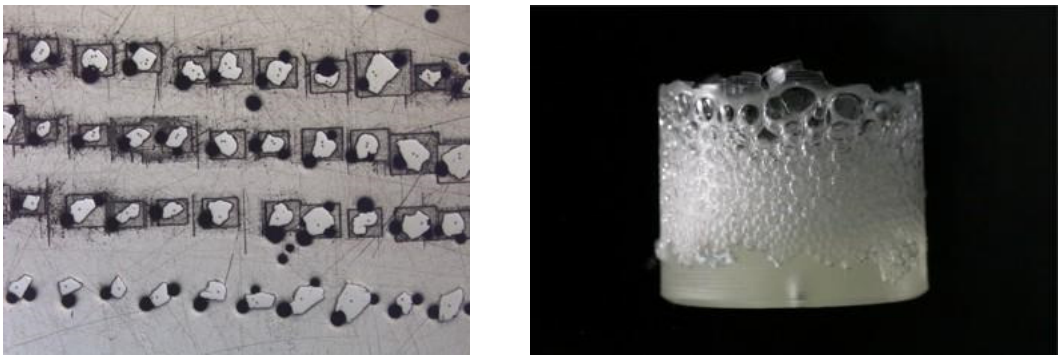


Figure 5. Left – a typical grain mount with pinhole bubbles next to each grain. This makes for a miserable afternoon in the lab as hitherto unseen oil will inevitably seep out from under each grain and perfectly coat the area of interest. Right, an example of over eager vacuum impregnation. Although vacuum impregnation can aid in encapsulating porous media in particular, care must be taken not to allow the resin to 'froth' when being pumped down. A gentle, controlled evacuation to ~250 Torr is sufficient. The preferred method of bubble prevention is curing under 4 bar pressure or N_2. Acrylic resins with excessive hardener also generate similar, frothy, mounts. (Images from Edinburgh ion probe labs).

To ensure that small grain mounts are not prone to gaps between the mineral and resin, clean the grains prior to mounting in an ultrasonic bath using acetone or ethanol. This ensures no residual grease/dirt is present on each grain's surface and reduces the risk of poor resin adherence and hence sample charging or 'muck' magically appearing on your lovingly prepared mount as soon as it enters the vacuum. However, ensure your samples are entirely free of solvent *before* mounting in epoxy – if not, the possibility exists that the glue of the mounting tape will dissolve and be efficiently deposited around the grains prior to the setting of the epoxy. This results in poor attachment of the epoxy to the grains, and their loss both in the ultrasonic bath, and worse, in the SIMS instrument when exposed to the UH vacuum, resulting in no analyses that week.

Of note - All resins are susceptible to softening by solvents; when cleaning a sample during polishing ensure the solvent (preferably water or pet-ether) is suitable for the resin. Acetone is

particularly aggressive and should be avoided. If you are intending to use an instrument with which you are unfamiliar, please ask the lab manager what mounting media they prefer – some instruments (such as the nanoSIMS) may have **very strict** requirements!

Where mounting resins are strictly prohibited – especially where you are trying to analyse elements present in the resin (e.g., H, C by SIMS), pressing polished samples into indium metal is a successful way to present a flat surface to the incoming beam.

Damage may also result from prior analyses. This is typically of Cathode luminescence (Fig. 6) analyses, which typically use an high beam current over the entire grain, and results in significant raster beam damage to the surrounding epoxy. This is suboptimal, both from the perspective of subsequent analyses (not flat, no ion-probe analyses or significant charging in the EPMA) but also deposits epoxy on the detector.

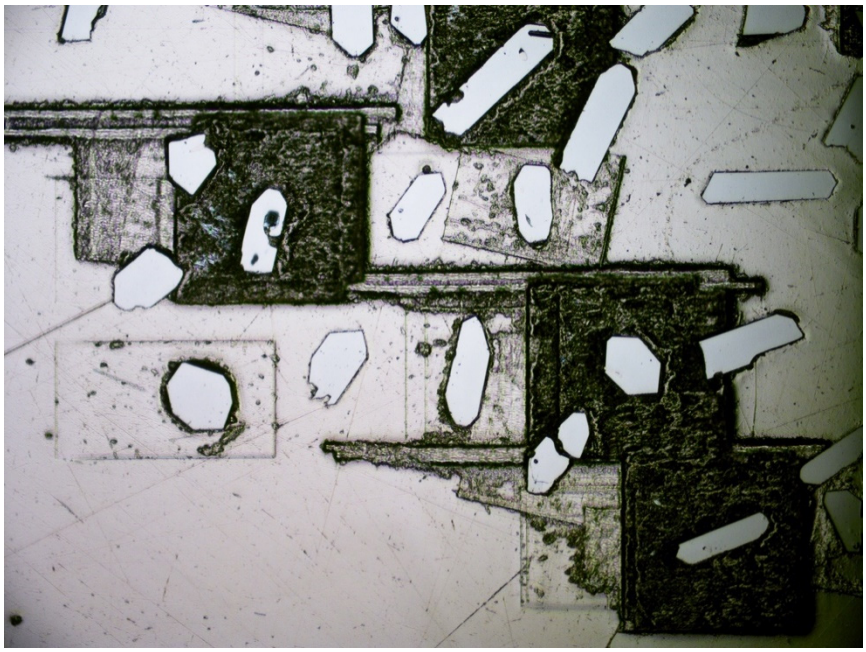


Figure 6. Excessive raster pits around grains resulting from CL imaging. These present neither a flat surface required for SIMS, nor a conductive path for e-beam instruments, and also result in grubby detectors in the SEM.

6. SECTIONING AND POLISHING

Most samples destined for analytical work will require cutting to size, mounting in resin, grinding away of excess resin and polishing. For most users, this typically involves the time honoured process of grinding on SiC papers and polishing on diamond or alumina laps. Here, samples are best sectioned using a wafering saw – preferably after mounting in resin.

Some tips on using the wafering saw:

- Blades are expensive – cheap blades start at ~£ 60, however, the thin cut blades (mm cut thickness) start at around £ 200. They also get 'better' as they get older so it is worth preserving them. **Never** present a sample or preparation stick to the blade by hand – more often than not it will break the blade. Always use the accessories/clamps to hold your sample.
- Always use a cutting fluid – either water or proprietary cooling fluid. This prevents the blade overheating and clogging and results in a finer cut, whilst prolonging blade life.
- Make sure you use the biggest collet to support the blade as possible – this helps to stop the blade bending during use. Additionally, don't use excessive weight on the blade or blade speed (rpm) – it does not speed things up!
- Always remove the blade after use (stops it being accidentally bent) and empty water from the cutting tray and wipe the saw down after use (stops the saw accessories going rusty).

7. GRINDING/LAPPING OF SAMPLES

May be done by hand or automated using a decreasing grit size of abrasives. The aim of all polishing is to remove the deepest scratches left by the previous process. For grinding/lapping, start using a 600 grit SiC abrasive (usually SiC paper – again, use water if possible or oil to lubricate the paper) and try to ensure that whilst grinding you do not introduce a tilt to the sample – ensure top and bottom of the mount remain parallel. If possible use an automated jig to help prevent this, but if grinding by hand, remember to occasionally rotate the sample to prevent preferentially grinding one side. Decrease grit size (increasing paper number – see table 1) when desired depth is reached 600 grit -> 800 grit-> 1,200 grit (approx.. a 25 µm 'polish'). Finer grit papers are available (e.g. 2000 and 4000) but often not required when polishing silicates – they may be useful for metal preparations.

8. POLISHING

After lapping ensure that the sample is cleaned, preferably using an ultrasonic bath and resin suitable solvent. Polishing laps are surprisingly costly, and also get better with use (especially those used with diamond pastes). However, the main reason for discarding a lap is cross contamination from poorly cleaned samples – you can bet your last bean that the only 15 µm grain of grit on your 1 µm lap will always find and scratch your sample!

The aim of polishing is to produce a polished, scratch-free sample, with the minimum of topography. Although there is no set procedure to guarantee a good polish there are some good guidelines ….

Table 1. USA/European Equivalency Grit Guide - FEPA* and ANSI/CAMI** and mean grainsize (table taken from Buehler Buyer's Guide 2005/2006).

FEPA	Grainsize	ANSI/CAMI	Grainsize
P60	269 µm	60	268 µm
P80	201 µm	80	188 µm
P120	127 µm	120	116 µm
P180	78 µm	180	78 µm
P240	58.5 µm	220	66 µm
P320	46.2 µm	-	-
P400	35 µm	320	34.3 µm
P600	25.8 µm	-	-
P800	21.8 µm	400	22.1 µm
P1000	18.3 µm	500	18.2 µm
P1200	15.3 µm	600	14.5 µm
P2500	8.4 µm	1,200	6.5 µm
P4000	5 µm	-	-

Over polishing will produce unwanted topography on the sample, especially is samples containing materials of differing hardness. Polish for the minimum time necessary.

Napless or low nap clothes are normally used with diamond polishing media. Hard cloths minimise surface relief while high nap, soft cloths produces a better polish.

9. POLISHING MEDIA

Alumina slurries – alumina powder (~.3 to .005 µm) is mixed with water to produce a slurry and this applied to the cloth. This produces a rapid polish allowing the sample to go from grinding to final polish in one step. However, due to the high loading of alumina in the slurry, polish for the **minimum** time required to avoid excessive relief. Secondly, alumina will not polish all materials – especially micas and aluminium.

Diamond – must be polished in multiple steps, usually grinding -> 15 µm -> 8 µm -> 3 µm -> 1 µm, with ~ 5 to 10 minutes on each, and a thorough **clean** (ultra-sonic) between steps. Once again, the aim is to remove the deepest scratches introduced by the previous step. Because of its hardness, diamond tends to produce less topography than alumina, and less deformation to the surface layer of the sample.

Other polishing media exist, including silicon carbide, boron carbide, colloidal silica and chemically enhanced methods

10. OTHER PREPARATION TECHNIQUES

Surface sensitive analysis such as EBSD, requires that no disruption to the samples crystallography results from the preparation process. Typically, the sample is prepared in a similar fashion to a typical EPMA mount, but the final polishing stages are optimised to prevent, or at least minimise, polishing induced lattice distortion. This may involve slow vibro-polishing, or chemical polishing – colloidal silica, such that the top surface is 'etched' and removed.

11. ION BEAM PREPARATION

Other techniques available include ion cross beam polishing, where the sample is masked and cut using an ion beam. A more refined version of this is focused ion beam (FIB) polishing. Here a sample is typically prepared as before and the region of interest identified and removed using a FIB (e.g., Fig. 7). This is achieved by placing down a protective 'strap' or mask of platinum using the ion beam, and then removing material either side of the area with the ion beam. The small, micron sized, wafer can then be removed by a micro-manipulator and either mounted to a TEM grid or processed further into a needle of atom probe use.

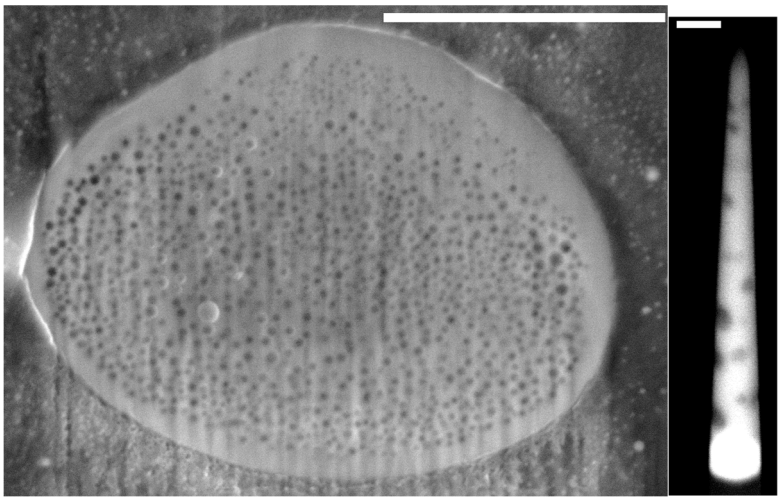


Figure 7. Left - Typical diamond anvil cell (DAC) metal:silicate experiment, prepared using a focussed ion beam. Shown is the metallic part (light grey), containing nanosized blebs of oxides and the surrounding silicate (scale bar 3 μm). Right, FIB prepared needle of metal shown, for use in the atom probe (scale bar 300 nm).

Ion beam cross-section polishing (Fig. 8) uses, typically, an argon beam to mill or polish cross sections of virtually any material. Unlike FIB preparation, cross beam polishing can prepare relatively large samples, with diameters up to ~5 mm efficiently milled and sample sizes up to 50 mm in diameter accommodated in the polisher. Typically, the sample is initially either mechanically ground or ion beam milled to expose the region of interest, and subsequently polished using a lower energy beam. Ion beam cross sectioning and polishing is particularly useful where water sensitive materials are being investigated or where there a significant changes in hardness present in the sample (e.g., high pressure experimental runs). Because there is no mechanical removal of material, it is also extremely useful when sample porosity is present and needs to be preserved.

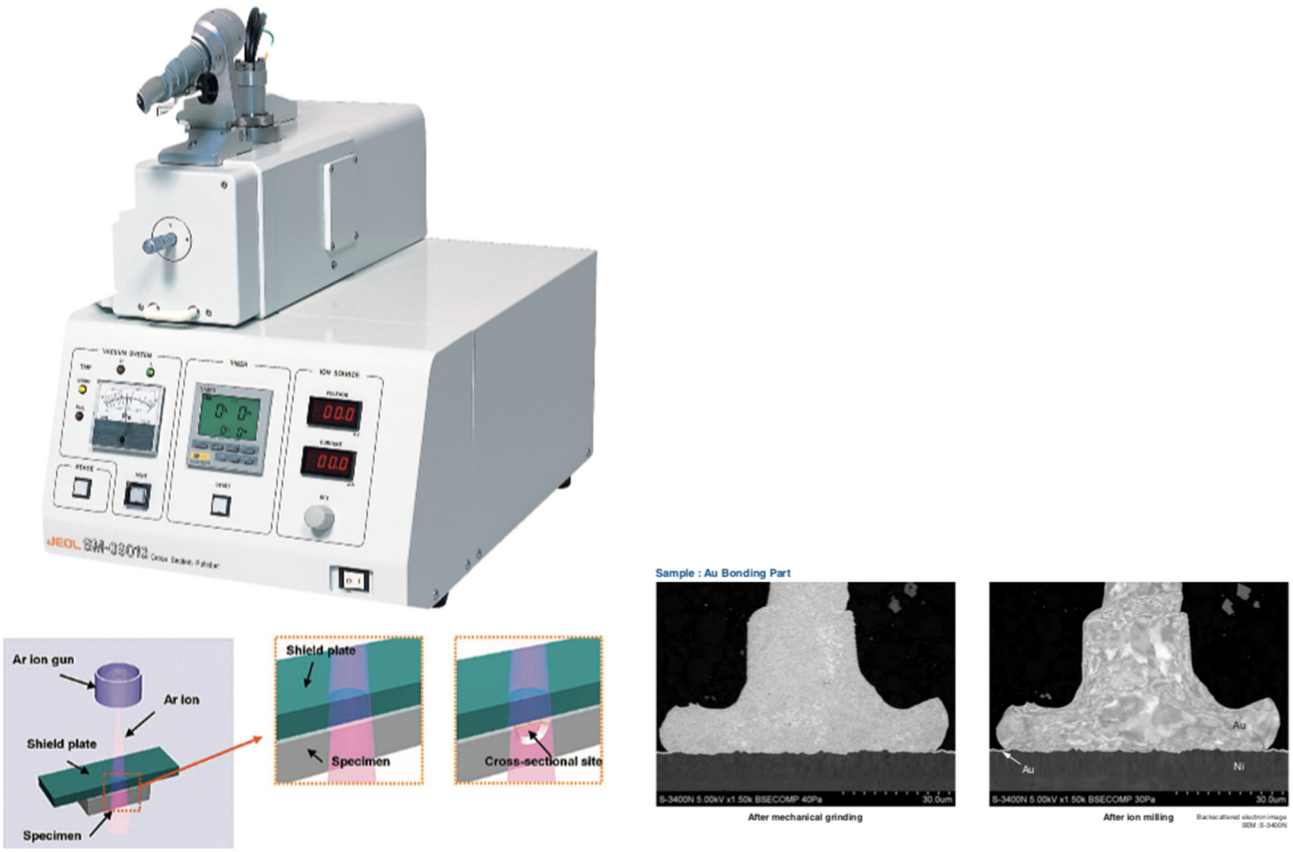


Figure 8. Left (top) JEOL ion beam cross section and (below) schematic of sample, mask and ion beam. Unlike the FIB with its highly focussed beam, the sample in the cross-section polished is physically masked with a shield plate and exposed to a wide ion beam. To minimise stripes on the sample, the sample is typically rocked back and forth. Below, sample of gold solder prepared using both mechanical grinding ion milling (Hitachi).

12. SAMPLE MACRO IMAGING

Nearly all microanalytical techniques operate best at high magnifications. Before coating your sample or putting it into an instrument are you going to be able to find that square millimetre of sample that you need to target? The easiest way of doing this is to take a low mag image of the sample. Methods available are simple digital photography, low mag microscopy (e.g., Leica EZ4), low mag Pol light microscopy, or particularly useful – a flat bed scanner; some photographic systems on phones can give a useful result and are better than nothing. Given the move by institutions to charge economic rates for instrument time, being able to find your sample rapidly makes financial sense. It is also worth spending time talking ample pictures at varying scales on, for instance, the SEM if the sample is going to be subsequently analysed by EPMA or SIMS, or prepared by FIB techniques. SEM time, by virtue of software optimised for imaging and instrument costs, is significantly cheaper – realising your samples need more preparation on a local SEM is considerably less upsetting than on a distant ion probe or beam line!

13. SAMPLE COATING

Most geological samples are non-conducting dielectrics and therefore prone to charging under the electron or ion beam.

As with most things in life, 'do as little as possible' is not a bad mantra for sample preparation, especially with valuable samples! Modern ESEM's allow a non-conducting sample to be imaged without any coat, but there is a contaminant decrease in image resolution as well as 'beam-skirt' issues associated with EDS analysis. Even with non-coated samples, it is beneficial to provide the shortest path to earth for any charge deposited on the sample surface – this can be done by covering the sample with aluminium foil and only exposing the area of interest. Wire grids may also be used for this purpose.

Because of the beam skirt effect, EPMA analysis requires high vacuum operation and thus non-conductive samples need to be conductive to prevent charge build-up. But, again, some thought may be required before coating.

Two commonly used materials are used to coat samples – gold and carbon. Gold is used for its high secondary electron yield and good sample coverage but is very difficult to remove from a sample, whilst its high z nature makes subsequent X-ray analysis difficult, especially if light elements are to be investigated. Carbon is the preferred coat for EPMA due to its low z number (low absorbance of X-rays) but has poor vertical sample coverage (due to the type of evaporation coater typically used). Secondly, if we were investigating carbon concentration in a metal (say, steel), we may wish to use a different coat to carbon. Other metals can be used in an evaporation coater, such as Mo and Al, but are not commonly used.

13.1. Carbon coating

Samples are placed in an evaporation coater beneath a pair of spring loaded carbon rods. One rod is sharpened, whilst the other flattened - once an adequate vacuum is reached, a current is passed through the rods heating the carbon to incandescence and evaporating a thin film. The thickness of this film can be controlled by monitoring the colour of a polished brass block, the same height as the sample (from [1]):

Orange	15 nm
Indigo-Red	20 nm
Blue	25 nm
Blueish-green	30 nm
Green-Blue	35 nm
Pale Green	40 nm
Silver Gold	45 nm

The EPMA software (typically ….) assumes a carbon thickness on both standards and unknowns of around 15 to 20 nm.

13.2. Gold coating

In conventional SEM sputter coating a gold (gold-palladium, or platinum) target is bombarded with heavy gas atoms (usually argon but air is a fair substitute). Metal atoms ejected from the target by the ionised gas cross the plasma to deposit onto the any surface within the coating unit including the specimen. A low vacuum environment is used (0.1 to 0.05 mbar), which with one of the modern low voltage sputter coaters, enables metal to be deposited at up to 1 nm/s**.** Unlike evaporation coating, sputter coating is affected less by sample topography. Coating is typically quick – it only takes a few minutes to achieve vacuum and run the coating process which typically is fully automated.

tip - if your sample is not flat – for instance a fossil tooth – and/or you are particularly interested in feature in the sides, it is a good idea to use an inclined sample holder and a shorter coating time. Run through the coating process more than once but rotate the sample between runs to adequately coat the sides.

Try not to overcoat your sample - gold is not cheap (obviously), and you may lose some of the fine surface detail with too thick a gold coat.

13.3 Other coating materials

Although carbon is still the most common EPMA coating, the advent of low voltage analysis has led to other metallic coats being explored. These are favoured to minimise absorption of a low energy line, when carbon is being analysed, or when 'thermal' damage appears excessive. A variety of sputter coated metals may be used, including iridium and silver.

Other techniques demand other coating materials – the high electric field of the atom probe results in insulators typically behaving poorly during analysis and rupturing. Coating the sample with a metallic coat may improve the run-time of the sample, however, the favoured coating material and its thickness may vary for each material.

14. CONCLUDING REMARKS

For fear of this sounding like one of my school reports, here's a few concluding remarks:

- "**If in doubt, do as little as possible**". Polishing, in particular, can be overdone, generating excess relief and starting again.
- **"Think first!"** If your sample turns out to be the next Nature or Science paper if only you had a few ion probe data points, nothing would disappoint you more to realise you mounted in acrylic because it was quick and close to hand.
- **"Have some ideas where you're going".** Take lots of images at different scales. Being lost in your sample because the image in the instrument looks nothing like it did in your badly drawn sketch is a good way to lose your hair.
- **"Label your samples".** If using a resin mount, always label your samples with an engraving tool. Someone once ran 30 experiments and labelled them all with an indelible pen, and then stored them in a 110 °C oven to keep them dry before analysis. When that someone then opened the oven on the day, the indelible pen had ineffably disappeared, and the hand drawn sketches made of the samples in his lab book (see above) all, weirdly, looked the same.
- **"Ask first!".** Good labs have extensive experience of a range of materials, and will know their instruments best. Lab managers will have preferred materials to use – epoxies, mounting systems etc., and usually for good reason. You will endear yourself no end to the ion probe lab manager if your sample would not make it through the load lock because it is outgassing like a leaky balloon. More importantly, you would not get any data.

15. REFERENCES

[1] Reed S J B 1996 *Electron microprobe analysis and scanning electron microscopy in geology*. (Cambridge, UK: Cambridge University Press)