

Simple sample preparation

To see detail in a specimen there are two important concepts:

- The microscope must have controlled illumination
- The sample must be prepared in such a way that the detail can be illuminated

Generally, the smaller the detail the more control and preparation is needed. However, many natural history specimens can be observed with relatively simple procedures. These will be divided into those needed for whole sample observation, such as a dead fly, and those required for observing specific areas in detail, such as the structure of the antenna of the fly.

Whole sample observation

The microscopes used for this will have modest magnification, usually given by an objective of $\times 1$ – $\times 4$ with eyepiece magnifications of $\times 10$ giving total magnification of $\times 10$ – $\times 40$. The microscope may be a stereo microscope, where light from the specimen passes to the eyes using two different light paths so the image is seen with stereoscopic vision. This gives the perception of depth and is essential if the specimen needs to be manipulated. The alternative microscope is a compound microscope where the light from the specimen passes to the eyes along a single light path which is often then directed to the eyes via a binocular system.



Stereo microscope



Simple compound microscope

The other thing to know is that the microscopes can be set-up so the light is either reflected by the specimen or passes through the specimen. In the latter case at least some of the specimen must be transparent!

If the specimen is thick or opaque it must be viewed by reflected light. A compound microscope usually used for transmitted light can be fitted with a small light to direct light onto the specimen with $\times 5$, $\times 10$ and often $\times 20$ magnification objectives. The specimen must be as flat as possible and some samples can just be supported on Blu-tack® on a slide and viewed. Others can be prepared by using a glass slide or cover slip to flatten them. The slide of a dead butterfly wing was made like this.



The next step

There are lots more techniques in mounting specimens. For example some specimens may need to be dried, cut into thin sections and stained before mounting in a resin which will provide a permanent mount.



There is lots of information available on these more advanced methods. See for example the following book and websites:

Practical Microscopy: J.E. Marson 72 pages from Brunel Microscopes

The Micscape website – <http://www.microscopy-uk.org.uk>. Check out the index for lots of information on different mounting media for the amateur and different methods for preparing lots of types of specimens.

The website of the Quekett Microscopical Club www.quekett.org

A mount of a whole specimen

A lamp is used to direct light onto the specimen so we observe the light reflected from the surface. The specimen is supported on a flat surface, a glass plate or a piece of card, but it might be difficult to see some parts because they are hidden by other sections. It needs to be orientated to the microscope lenses (and probably to the illuminating light). If the specimen is supported on a small piece of Bluetac®, it can be tilted so different areas can be illuminated and observed.



If a specimen is small and needs to be examined in reflected light more than once it can be permanently mounted. There are special slides as shown below. In the ring mount the specimens (possibly sand grains) can be shaken to show different sides, in the grid slide several specimens may be mounted to show all sides.



To observe transparent or semi transparent specimens, such as the small beasties in pond water, special glass plates – microscope slides – are needed. There are special glass slides called cavity slides or ring slides where there is a small well to trap the drop of liquid.



Alternatively flat slides where a drop of water is trapped under a smaller, thinner piece of glass – the cover slip – can be used. Of course small live animals in pond water will often move around so they are difficult to see in focus using the microscope.

They can be slowed down if a few fibres of cotton wool are incorporated with the drop of pond water or a small amount of a chemical which temporarily anaesthetises them is added to the water. An anaesthetic throat spray is often used.

Observation of detail (small structures) in a specimen

The most common way of looking at detail is by transmitted light microscopy with a compound microscope. The techniques used are based on putting the thin specimen onto a microscope slide. It is then surrounded with a mountant (usually applied as a liquid) and then covered with a cover slip. Most microscope are standardised to work best with microscope slides that are 3"×1" in size and 1mm thick with cover slips of thin glass (often designated No 1 or 1½) which may be square or circular. The mountant surrounds the specimen excluding air and providing contrast at the edges of the transparent structures. It does this by having a different refractive index to the specimen. Water can be used as a mountant but for many specimens glycerine or special oils are used. Of course, if a mountant is a liquid it can leak out from below the cover slip but can be trapped for a modest amount of time by painting round the edge of the cover slip with a lacquer (nail varnish works well). A more robust mountant is glycerine jelly.

The best way to introduce the mountant is to put the dry specimen on the microscope slide, put a drop of the mountant as a line along one edge of the cover slip, then place that edge of the coverslip at an angle on the slide and let it drop gently onto the specimen. The mountant will flow over the specimen pushing out the air ahead of it.



The microscope will usually have objective lenses giving magnifications of ×10 to ×40. With standard ×10 eyepieces the total magnification range is ×100 to ×400. It is now critical that the specimen is as flat as possible and does not move around too much.