

4 Microscopy

The basics, experience and patience are important in microscopy. A microscopy image can be obtained quickly, but only with the correct settings of magnification, resolution, contrast, depth of field, illumination and brightness can very good microscopy images be produced.

The following does not describe all the basics of microscopy, but only those that are necessary for the correct use of the microscope. The following should always be taken into account when using microscopy:

- The total magnification is multiplicative of the magnification of the objectives and eyepieces.
- The objectives produce an intermediate image which is an enlarged representation of the object. Object structures are resolved and magnified.
- Eyepieces enlarge the intermediate image produced by the objective. Only the object structures that have already been resolved are enlarged. Eyepieces do not increase the resolution.
 - This means that if an objective could not resolve a structure, then an eyepiece cannot magnify it either, no matter how powerful the eyepiece is.
 - This means, for example, that the combination of 20X objective and 10X eyepiece shows more object structures and therefore a better microscope image than the combination of 10X objective and 20X eyepiece.
- If you want to use a microscope with a higher magnification, it is always advisable to increase the magnification of the objective first.
- The magnification is always changed from **small** → **large**. That means:
 1. First, the 4X objective is selected and the object is focused and centred in the field of view.
 2. Then the 10X objective is adjusted and the object is refocused using the fine adjustment. This is repeated until the desired magnification is achieved.
- You should always use objectives from the same supplier, as they usually have a parfocal distance from each other. This means that when changing the magnification (turning the revolving nosepiece), only small adjustments to the fine drive are necessary to bring the specimen back into focus.
- Only the lamp control is used for brightness control.
- Although the brightness can be changed by adjusting the aperture or field diaphragm, the correct settings of the aperture or field diaphragm are important for resolution, contrast, depth of field and illumination.
- If the brightness is incorrectly controlled by the aperture or field diaphragm, the microscope image will be poorer.
- For a good microscope image, the condenser position, aperture and field diaphragm must be correctly adjusted.
- For each change in the objective, the condenser position, aperture and field diaphragm must be correctly adjusted again.
- Without adjusting the condenser position, aperture and field diaphragm, the quality of the microscope image will be impaired.
- The dioptre compensation on the eyepieces does not change the sharpness of the microscope image.
 - To bring the microscope image into focus, it must be focussed using the coarse and fine adjustment.
- The specimens should be prepared carefully. Experience is helpful here.
- If there is much more sample between the slide and cover glass than necessary, much more substrate will be microscoped than necessary.
 - For example, when examining blood, the blood cells may stack up and form large clumps, or when examining aquatic life, the image may look foggy.