

BIOL1003 High Distinction Notes

2016 Semester One

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Introduction to the microscope

Positioning and handling of microscope

- Handle the microscope by handling the arm in upright position
- Position it 5cm away from the edge of the bench
- Report and check for any defects before use, don't fix it.

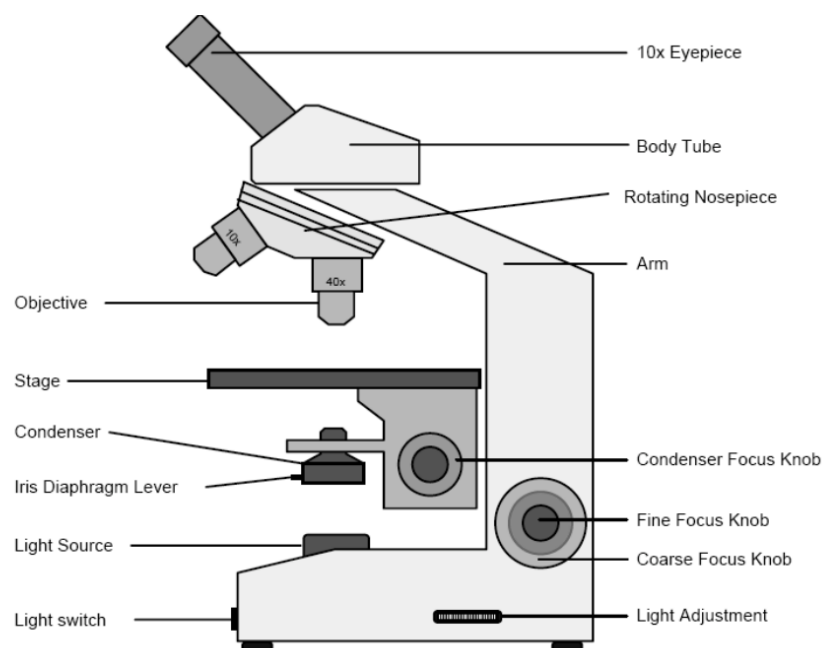
Cleaning the microscope

- Use lens cleaning tissues and solutions (70% ethanol) to clean the lens
- If no clear images are obtained, clean the eyepiece and objectives, moisten lens cleaning tissue with cleaning solutions and wipe the lens
- Rotate the eye piece and if the image moves, then the eye piece must be dirty
- Move the objectives, if the dirt disappears then it is the objective lens that are dirty

Microscope skills needed

- Set up and use the microscope correctly
- Make observations with the microscope
- Accurately record observations, by drawing or writing descriptions

Parts of microscope



Setting up the microscope

Focusing the light

1. Swing the objective into 10x and put the slide on the stage (with cover slip on)
2. Turn on the light and adjust light intensity
3. Use mechanical (coarse focus) knob to raise to stage 5mm away from slide
4. Focus the specimen by bring down the stage
5. Use fine focus knob to sharp focus the specimen
6. Open the condenser and iris diaphragm fully

Maximise resolution

1. Look down on the eyepiece and close the iris diaphragm
2. A white spot in grey circle can be seen
3. Open the diaphragm until the white spot fills $\frac{3}{4}$ of the grey circle
4. Place back the slide
5. Refocus the specimen using fine focus
6. Focus as much as possible
7. Turn into high power, adjust diaphragm and fine focus

Microscope theory

- Uses a system of lens to produced enlarged image of an object
- It is both the magnification power and also resolution power that allows to see sharp images
- Minimum resolved distance = minimum distance two objects can be distinguished as distinct
- Magnification is the enlargement of the image

How the condenser works

- Controls the focus of light by moving up and down
- Iris diaphragm controls the amount of light reaching the objective
- Condenser focuses light and control how much light enters the lens

Iris diaphragm

- Varies the angle which light passes the specimen (contrast and resolution)
- If too wide, then light scatters and not focus
- If too narrow, it will be too dark with poor resolution and high contrast

Trouble shooting of microscope

1. Can't see an image? – microscope plugged in, light on/off, objective lens in position
2. Can't find specimen? – centre specimen over the light source, use lower objectives
3. Dirt obscuring specimen? – is the dirt on the lens or specimen
4. Can't focus? – specimen too thick, coverslip on top, iris diaphragm opened/closed too far
5. Too dark or bright? - check light intensity and opening or iris diaphragm
6. Poor contrast? – close down iris diaphragm
7. Poor resolution? – adjust condenser, open up diaphragm

Producing a scale bar drawing (relate to the size of the object)

A scale bar is the representation of the ratio between the size of the object in reality and the drawn size of the same object

A = actual size of the object in real life (eg. pencil in real life is 18cm long)

D = drawn size of the object (eg. Pencil is drawn 7.5cm on paper)

a = length the scale bar is representing (eg. X cm on scale bar = 5cm in real life)

d = actual size of the scale bar (eg. Scale bar is measured at 5cm)

Therefore

$$\frac{D}{A} = \frac{d}{a}$$

Once we have D , A and a , we can calculate d (the length we have to draw the scale bar).

i.e.
$$d = \frac{D \times a}{A}$$

Example: producing a scale bar of 18cm long pencil

Actual length (A) = 18.0cm

Length of drawing (D) = 7.5cm

Theoretical bar length (a) = 5cm

Scale bar length (d) = $7.5 \times 5 / 18.0 = 2.1\text{cm}$

Hint* = scale bar units should be in whole numbers, and the bar should represent $1/3$ or $1/4$ the drawn length (eg. 2.1cm is roughly $1/4$ the drawn length of 7.5cm)

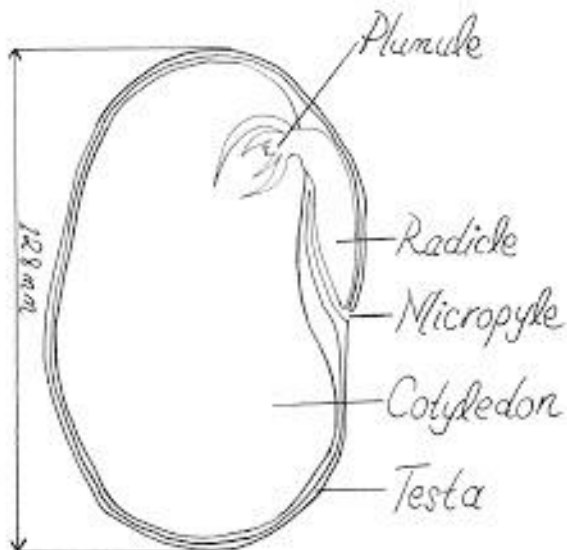
Estimating the size of the objects using a microscope

- Place a grid under low power with 0.1mm each

- Count how many 0.1mm sides squares fit across the field of view
- If 10 fits, then $0.1\text{mm} \times 10 = 1\text{mm}$ diameter of the field of view
- Count how many times an object can fit across the field of view (eg. If 4 can fit under, then the size of object is $1\text{mm} / 4$ or $1000\text{microns}/4 = 250\text{microns}$ per object)

Biological drawings

- Include a title of the drawings
- All drawing must be done on unlined paper with lead pencil
- Do not colour in or shade in the drawing
- Drawings should be simple, done in clear, definite lines that join other lines
- Animal drawing should include the anterior or dorsal towards top of the page
- Plants should be drawn the way they are oriented
- Label in pencil without arrow heads, must be parallel and not cross with each other
- All drawings should include scale bars



Longitudinal Section of a bean
seed (X12.8)

$$\begin{aligned} \text{Magnification} &= \frac{\text{Length of Drawing}}{\text{Length of Specimen}} \\ &= \frac{128 \text{ mm}}{10 \text{ mm}} \\ &= \underline{\underline{X12.8}} \end{aligned}$$