NEATH PORT TALBOT COLLEGE
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School of Maths & Science
Science Practical

Preparation of biological material for optical (light) microscopy

◆ Aim

To prepare temporary slides of plant material.

◆ Introduction

Biological specimens, living or preserved can be observed with the optical (light) microscope provided there is sufficient **contrast** for the structural differences of the specimen to be appreciated. “Contrast” is the highlighting of tiny differences in the structure of cells. In biological material which is too transparent, contrast is achieved by staining the material. Stains are taken up by the cells and colour particular structures within cells, e.g. haematoxylin / eosin is a double staining technique used for animal material: nuclei and cytoplasm stain blue and pink respectively.

◆ Safety

Control Measures

- The wearing of **safety goggles** and a **laboratory coat** at all times will be sufficient to take account of most hazards and significant risks.
- When working with stains or dyes, avoid contact with skin or clothing.
- Take care when using sharp dissection equipment, and when it is placed on the bench besides you.
- The compound microscope is a precision instrument, handle it with care and protect it from dust by storing it under cover.
- Always lift the microscope by the arm, support the base on palm of hand.
- Do not tilt – mirror or eyepiece may fall out.
- Keep lenses clean – use lens tissue.
- Do not allow lenses to get wet.
- Always watch from side when moving tube downwards with coarse adjustment, otherwise may smash slide and objective.
- Always change to low power objective before removing slide from stage, otherwise you may chip objective.
- Use only clean slides and cover slips.
- When you have finished using the microscope leave the low power objective in position.
- Keep stage dry.

Control Measures – continued

- Take great care when using electrical equipment such as bench lamps.
- You are reminded of the need for good laboratory practice in order to maintain a safe working environment.

Hazards

Irritant **Iodine solution**

Do not use direct sunlight to illuminate the mirror of the microscope, since sunlight when focused on to the retina may damage the light sensitive cells there.

Take care when focusing, particularly with the high power objectives – do not focus downwards without looking. Always focus with the low power objective first, before using the high power objectives.

◆ Materials & apparatus

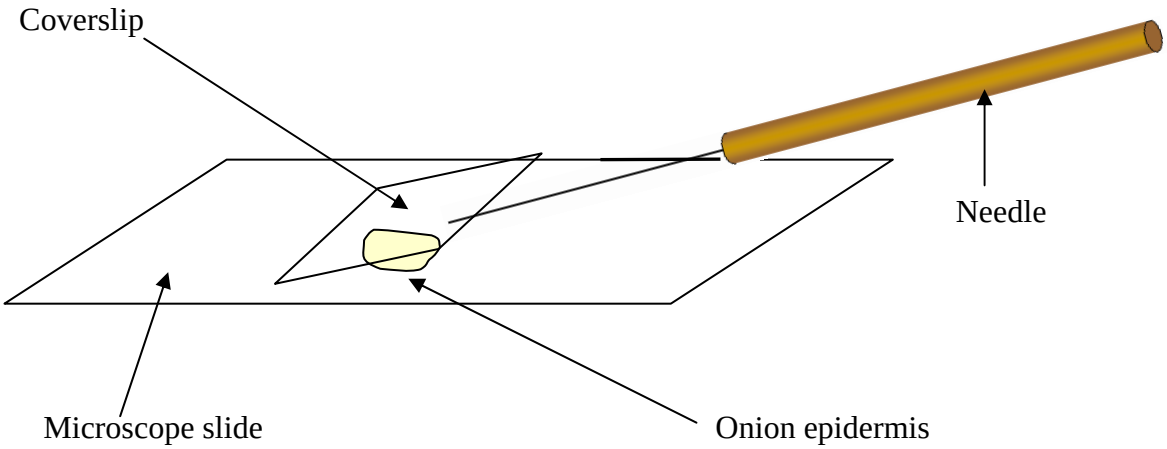
The following items are required:

Microscope	Elodea
Lamp	Distilled water
Slides & coverslips	Pasteur pipettes & bulbs
Small scissors	Forceps
Dilute iodine solution	Onion bulb
Needle	White tiles

◆ Procedure

1. Strip off a piece of epidermis from the inner lining of one of the fleshy scales of an onion. The strip of epidermis is placed on a microscope slide and a drop of iodine solution is added to the slide. A coverslip is then placed over the stained specimen (wipe away excess stain with a tissue). The slide is now ready for viewing under the microscope.

Figure: Diagram to show the preparation of temporary slides.



Note:

Make sure onion epidermis does not fold on itself.
Try not to create too many air bubbles when placing coverslip over specimen.

2. Observe the slide under low power.

3. Observe the slide under high power.

*Observe the **cytoplasm** surrounding the clear vacuole. The **nucleus** is located in the cytoplasm close to the cell wall.*

4. Obtain a single leaf of pondweed, *Elodea*. Place on a slide and add a drop of water. Place coverslip over leaf.

5. Observe slide under low and high power.