

# Leaf Structure as Environment Indicator: Activities

## Activity 1: Estimating the thickness of epidermal layer of leaf

### Aim

The aim of activity 1 is to estimate the thickness of the epidermal layer of the leaf sections in slides 1, 2 and 3.

### Method

Start with leaf section in slide 2, the *Nymphaea*. Select the appropriate slide from the light box and view it with the x20 objective. Check you can recognise the epidermal layer cells. The section may not be perfectly flat, so it is better to focus in some parts than in others. You may want to move around the slide to find an area where the epidermal layers are more clearly visible.

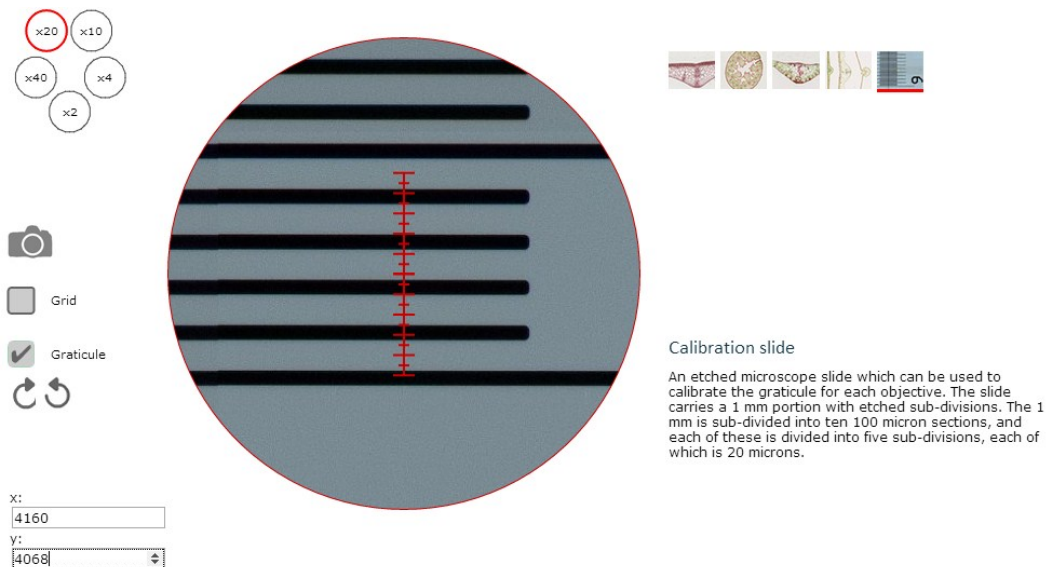
**Epidermal layer thickness:** Choose a clearly defined epidermal layer and measure its width. They may be more than one cell thick. Indicate whether it is the upper or lower epidermal layer. To do this you need to use a graticule, which is a fixed scale etched onto a piece of glass that you can insert into the eyepiece of a microscope in order to measure items. In the digital microscope, simply click on the box labelled "Graticule" and a tick will appear. You will see a red line divided into 10 large (or 20 smaller) divisions. You can rotate the graticule in order to line it up with the item you wish to measure. Click on one of the two circular arrows beneath the "Graticule" checkbox to rotate it.



**Figure 1** A screen shot of the digital microscope showing the epidermal layer of a leaf section of *Nymphaea*, leaf viewed with a x20 objective lens. The graticule is overlain on the image and correctly orientated to measure the thickness of the upper epidermal layer. The correct measurement in this instance is 2.0 large units on the graticule.

Record the thickness of the epidermal layer as the number of large units along the graticule. Repeat this measurement choosing four other different locations along the epidermal layer, and then calculate a mean of your five measurements. You may choose to make the measurements using a different objective lens (e.g. x10), but it is important to be consistent and make all five of your measurements using the same magnification.

To convert your mean to an actual length, it is necessary to calibrate the graticule at the magnification you used for your measurements. To do this, select the calibration slide from the light box and line up your graticule with the scale. The etched microscope slide can be used to calibrate the graticule for each objective. The slide carries a 1 mm portion with etched sub-divisions. The 1 mm is sub-divided into ten 100 micron sections, and each of these is divided into five sub-divisions, each of which is 20 microns. Estimate as accurately as you can the length of one large graticule unit, noting that there are ten large graticule units.



**Figure 2** A screen shot of the digital microscope showing the calibration slide viewed with a x20 objective lens. The scale labels on the calibration slide represent millimetres. The vertical graticule is shown overlain on the right-hand of the scale, with its bottom end aligned with the long bar labelled “6.” The top end of the graticule is approximately at “6.045” on the scale. The length of the graticule is 0.045 or 90 microns. Given the graticule is composed of 10 large units, the length of a single large unit is 90/10 or 9.0  $\mu\text{m}$ .

## **Activity 2: Classifying the three leaf specimens assigned A, B and C in slide 4**

### ***Aim***

The aim of activity 2 is to list any other three anatomical structures in leaf, apart from the thickness of epidermal layer, that differentiate the three leaf specimens in slides 1, 2 and 3.

### ***Method***

Start with leaf section in slide 1, the *Nymphaea*. Select the appropriate slide from the light box and view it with the x20 objective. Check you can recognise the different leaf anatomical structures in the specimen. You may want to move around the slide to find an area where the different structures are more clearly visible.

*List at least three other differences in leaf anatomical structures:*

Repeat activity 2 for *Ammophila* and *Zea mays*

### ***Analysis***

Using the calibration described in Fig 1.1, one large unit on the graticule is 9.0  $\mu\text{m}$  and therefore the thickness of the epidermal layer shown in Figure 1.0 is  $2.0 \times 9.0 \mu\text{m} = 18.0 \mu\text{m}$ . Using your own data, calculate the mean thickness of the epidermal layer at the four other different locations you chose.

### ***Task***

Repeat the method set out above to compare your value for the thickness of *Nymphaea* epidermal layer, for *Ammophila* and *Zea mays*. Once you have calculated your final result, compare the values for the three species. Use the values as well as results from activity 2 to identify the three leaf sections A, B and C in slide 4. Consider the implications for the survival strategy of the hydrophyte, mesophyte and xerophyte.