A typical eyepiece graticule looks like this:

```
0 1 2 3 4 5 6 7 8 9 10
```

It is 10mm in length and each mm is divided into 10 parts
So each small division = 0.1mm = 100µm

The eyepiece graticule is located at the primary image of the microscope. So it is in focus with image of the specimen.

The specimen is magnified onto the graticule by the magnification of the objective. So, the actual amount of specimen superimposed on the graticule is:

\[ \frac{0.1\text{mm}}{\text{Objective magnification}} \]

Typically this gives the following calibrations (with an eyepiece graticule 10mm in 100 parts):

<table>
<thead>
<tr>
<th>Objective Magnification</th>
<th>Each small division Equals (in mm)</th>
<th>Each small division Equals in microns (µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>4x</td>
<td>0.025</td>
<td>25</td>
</tr>
<tr>
<td>10x</td>
<td>0.01</td>
<td>10</td>
</tr>
<tr>
<td>20x</td>
<td>0.005</td>
<td>5</td>
</tr>
<tr>
<td>40x</td>
<td>0.0025</td>
<td>2.5</td>
</tr>
<tr>
<td>100x</td>
<td>0.001</td>
<td>1</td>
</tr>
</tbody>
</table>

TO CALIBRATE

HOWEVER – There may be small differences between one microscope and another. So to be certain of measuring the true distance it is necessary to calibrate the microscope. This is done using a stage micrometer. These have an accurate scale on them on 1mm divided into 100 parts.

So each small division on a stage micrometer = 0.01mm = 10µm

To calibrate the eyepiece graticule with a particular objective the stage micrometer is viewed through the microscope. The image will look something like the following with a high power objective:

```
```

This process should be repeated with each objective.

CAUTION – Some microscopes have a viewing head with an adjustable interpupillary distance which changes the tube length of the microscope. Usually each eyepiece tube can be focused to adjust the tube length (for parfocality between objectives). When calibrating it is essential to ensure the same settings on the viewing head are used for calibration and subsequent measurements.