

# How to use a Stage Micrometer

## Introduction

Whenever there is a need to make measurements with an eyepiece graticule, there is also a need to ensure that the microscope is calibrated. The use of a Stage Micrometer to check the divisions and measurements on the eyepiece reticle is the best way to achieve this calibration.

There are a number of factors that can influence the true calibration:

- Any inherent inaccuracies of the objective lenses
- Variations in the optical system itself (such as effective tube length)
- Inaccuracies in the reticle scale

These combined errors need to be taken into account and quantified if measurements taken with the microscope are to be considered accurate within defined limits of uncertainty.

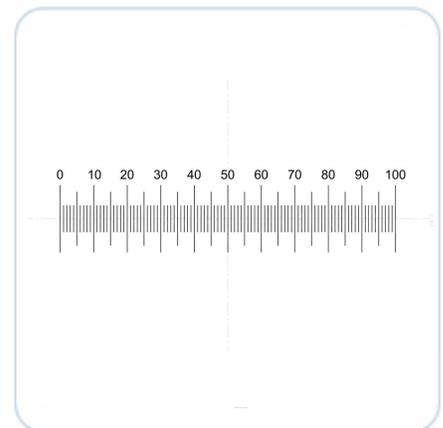
## What is a Stage Micrometer ?

A Stage Micrometer is simply a microscope slide with a finely divided scale marked on the surface. The scale is of a known true length and is used for calibration of optical systems with eyepiece graticule patterns. This is particularly important when alternating between objectives on one microscope or when using the same graticule in different microscopes.

## What is an Eyepiece Graticule?

A microscope may be equipped with a linear measurement scale fitted into one eyepiece. This eyepiece scale is called a reticle or graticule. The terms reticle or graticule are interchangeable.

The reticle can be used to measure any planar dimension in a microscope field, since the ocular can be rotated in any direction and the object of interest can be repositioned with the stage manipulators.



# How to use a Stage Micrometer

Using the procedure explained below a 'conversion factor' can be derived. This will enable the user to convert the apparent size of a subject as seen through the eyepiece scale, into a real world dimension.

An accurately derived conversion factor will compensate for any of the errors discussed earlier.

To measure an object length, note the number of divisions spanned by the object then multiply by the conversion factor for the magnification used.

The conversion factor is different at each magnification. Therefore, when using a reticle for the first time it is necessary to calibrate the scale by focusing on a second micrometer scale (Stage Micrometer) placed directly on the stage.

## Conversion Factor

To get the conversion factor, you are required to calibrate the microscope and its associated reticle with a Stage Micrometer.

The following is an example procedure:

- 1 Select your normal or lowest magnification.
- 2 Ensure the eyepiece reticle is in sharp focus.
- 3 Place the Stage Micrometer on microscope stage. Position and focus so the stage scale is clearly visible.
- 4 Rotate the eyepiece reticle and position the stage in the field of view so that the two scales appear parallel, one positioned above the other.
- 5 Adjust the alignment of the scales so that the zero values correspond. With the zero values aligned, the Stage Micrometer scale will either appear longer than the eyepiece scale or alternatively it may appear the same length or shorter than the eyepiece scale.

# How to use a Stage Micrometer

- **If the stage scale appears longer than the eyepiece scale**

Count the number of Stage Micrometer divisions that cover the eyepiece graticule scale.

## Calculating the conversion factor

*Using a Stage Micrometer with 100 divisions of 0.1mm having a 10mm total length (Graticules Optics model S1 or PS1)*

### Example 1 : Selected objective magnification X4.

Suppose the full length of the reticle scale covered 25 divisions of the Stage Micrometer, then the full length of the reticle scale is equivalent to  $(25 \times 0.1\text{mm}) = 2.5\text{mm}$  long. For an eyepiece reticle with 100 divisions, each division will measure  $25\mu\text{m}$  at the stage for this magnification.

### Example 2 : Selected objective magnification X10.

Selecting the X10 objective and repeating the exercise above would show that the reticle scale now covers 10 divisions of the stage scale. Then the full length of the reticle scale is equivalent to  $(10 \times 0.1\text{mm}) = 1\text{mm}$  long. For an eyepiece reticle with 100 divisions, each division will measure  $10\mu\text{m}$  at the stage for this magnification.

**In summary, you can apply these conversion factors to state what each division of the eyepiece reticle is measuring for a selected magnification.**

X4    1 division =  $25\mu\text{m}$

X10   1 division =  $10\mu\text{m}$

- **If the stage scale appears the same length or shorter than the eyepiece scale**

In some cases, the Stage Micrometer scale may appear shorter than the eyepiece scale. In that case, note how many eyepiece divisions match the full length of the Stage Micrometer scale.

## Calculating the conversion factor

*Using a Stage Micrometer with 50 divisions of 0.002mm having a 0.1mm total length (Graticules Optics model S12 or PS12)*

### Example 3 - Selected objective magnification X40.

The full length of the stage scale only spans the first 40 divisions of the reticle scale. Then the first 40 divisions of the reticle scale is  $0.1\text{mm}$  long. So each reticle division is equivalent to  $(0.1\text{mm} / 40) = 2.5\mu\text{m}$ . For an eyepiece reticle with 100 divisions, the full length of the reticle scale equals  $250\mu\text{m}$  ( $0.25\text{mm}$ ) at the stage for this magnification.

# How to use a Stage Micrometer

## Real Case

The examples above give ideal perfect world results. They do not exhibit calibration discrepancies that occur due to any inherent inaccuracies of objectives and the optical system, or the inaccuracies in the eyepiece reticle scale. These errors are more noticeable at higher magnifications using Stage Micrometers with the finest divisions.

The following is such an example using the same Stage Micrometer as example 3 and a X100 Oil immersion objective.

### Calculating the conversion factor

*Using a Stage Micrometer with 50 divisions of 0.002mm having a 0.1mm total length (Graticules Optics model S12 or PS12)*

#### Example 4 - Selected objective magnification X100.

The full length of the stage scale only spans the first 95 divisions of the reticle scale. Then the first 95 divisions of the reticle scale is 0.1mm long. So each reticle division is equivalent to  $(0.1\text{mm} / 95) = 1.05\mu\text{m}$ . For an eyepiece reticle with 100 divisions, the full length of the reticle scale equals  $105\mu\text{m}$  (0.105mm) at the stage for this magnification.

Other factors should be taken into consideration before taking this figure of  $1.05\mu\text{m}$  per division as a clear-cut certainty.

Human error can be introduced in taking the measurement, as alignment of the scales can be a little difficult at high magnifications. It is good practice to reduce the effects of human error by taking a number of measurements and averaging the results.

Mathematically calculating that each division =  $1.05\mu\text{m}$  is OK, but what is the microscope limit of optical resolution? Assuming a resolution of  $0.5\mu\text{m}$  or so using oil immersion, how could you report that an object covering 2.5 divisions in the reticle measures exactly  $2.625\mu\text{m}$ ?

Additionally, how can you be sure that the Stage Micrometer is in fact 0.1mm long, with divisions of  $2\mu\text{m}$ ? Even if the micrometer is as precise and accurate as the PS12 used above. Just how precise is it, and how do you prove it?

This is of particular importance if the measurements you are taking are critical to your process and required to be part of an ISO quality procedure. This is where a traceable calibration certificate of the Stage Micrometer may be required.

# How to use a Stage Micrometer

## Stage Micrometers & Calibration Certificates

Graticules Optics Stage Micrometers, calibration standards, long scales and grids are used all round the world for calibrating microscopes, imaging systems, co-ordinate measuring equipment and metrology instruments.

All Graticules Optics calibration products are produced to exacting standards with high-resolution imaging to ensure well defined line edges, uniformity of line thickness and excellent pattern integrity. These products are guaranteed to satisfy the requirements for traceable calibrations.

## Calibration Standards

Where customers have the need for traceability of their calibration, Graticules Optics offer certificates of calibration, traceable to International standards, and re-calibrations of customers' own products.

Calibration certificates from UKAS and NPL offered by Graticules Optics will satisfy the requirements of NIST, DIN, DKD, Cofrac, Accredia, NABL and all other National Metrology Institutes across the world, and provide compliance for ISO/IEC17025 and ANSI/NCSL Z540-1. The ASTM B487 standard demands a calibration uncertainty of 0.2um for Stage Micrometers; we are pleased to offer calibration certificates from NPL that will satisfy this requirement.

We strongly recommend that you use our PS range when traceable calibrations are required, as these have a unique engraved serial number.



# How to use a Stage Micrometer

Here are the three types of Calibration Certificate we can offer:

## Calibration by NPL

The National Physical Laboratory carries out measurements at selected points on the scales and grids and issues a certificate of calibration.

## Calibration by UKAS Accredited Laboratory

A UKAS accredited laboratory carries out measurements at selected points on the scales and grids and issues a calibration certificate.

## Measurement by Graticules Optics

For applications that do not require a traceable certificate or the accuracy provided by calibrations carried out by NPL or a UKAS accredited laboratory, Graticules Optics can provide a Graticules Certificate of Comparison. The scale or grid is compared with NPL calibrated in-house standards and a statement is provided on the accuracy of the item with respect to these standards

## Physical Description of Graticules Optics Stage Micrometers

Pyser have two distinct Stage Micrometer product ranges, the 'S' and the 'PS' series.

### 'PS' series

If you require traceable calibration to satisfy NIST or ISO, then you need to have a Stage Micrometer with a unique engraved serial number. In which case you require our 'PS' series Stage Micrometers. These differ from the 'S' series, because the glass disc is mounted in a stainless steel slide with an engraved, unique serial number. Each slide is supplied in a polished wooden presentation/storage box to distinguish it as a traceable standard of high value.



### Features

- Unique engraved serial number
- Stainless steel slide mount
- Supplied in polished wooden box
- Various scale lengths from 0.1mm to 20mm
- Negative & Positive images for transmitted or reflective light
- High quality image
- Available with Internationally traceable certificates of calibration

# How to use a Stage Micrometer ?



## 'S' series

The 'S' series intended for general everyday routine calibration. The scale is centred on a glass disc mounted in a black anodised aluminium slide 75mm x 24mm x 2mm thick.

## Selecting the Right Scale for Your Application

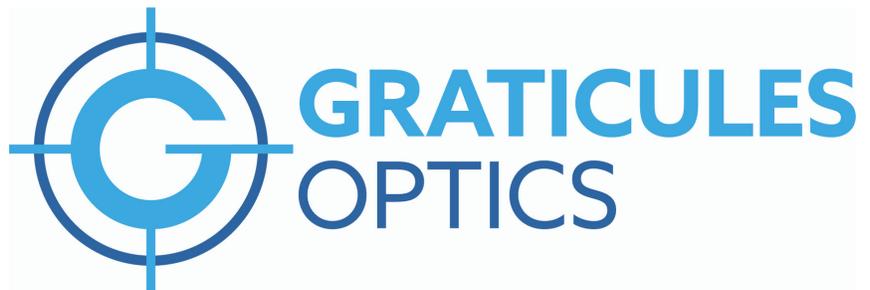
### Transmitted (Brightfield) Light

Objective Magnification	Measured Size Range	Eyepiece Graticule (Length/Divisions)	Stage Micrometer (Length/Divisions)
0.5	0.1mm - 10mm	NE5 5mm/0.05mm	PS1 10mm/0.1mm
0.5	0.01mm - 1mm (10 - 1000µm)	NE28 1mm/0.01mm	PS8 1mm/0.01mm
1	0.1mm - 10mm	NE1 10mm/0.1mm	PS1 10mm/0.1mm
1	0.01mm - 1mm (10 - 1000µm)	NE28 1mm/0.01mm	PS8 1mm/0.01mm
2	0.1mm - 10mm	NE5 5mm/0.05mm	PS1 10mm/0.1mm
2	0.01mm - 1mm (10 - 1000µm)	NE5 5mm/0.05mm	PS8 1mm/0.01mm
4	0.01mm - 1mm (10 - 1000µm)	NE5 5mm/0.05mm	PS8 1mm/0.01mm
10	0.01mm - 1mm (10 - 1000µm)	NE1 10mm/0.1mm	PS8 1mm/0.01mm
10	0.002mm - 0.1mm (2 - 100µm)	NE28 1mm/0.01mm	PS12 0.1mm/0.002mm
16	0.01mm - 1mm (10 - 1000µm)	NE1 10mm/0.1mm	PS8 1mm/0.01mm
16	0.002mm - 0.1mm (2 - 100µm)	NE5 5mm/0.05mm	PS12 0.1mm/0.002mm
20	0.01mm - 1mm (10 - 1000µm)	NE1 10mm/0.1mm	PS8 1mm/0.01mm
20	0.002mm - 0.1mm (2 - 100µm)	NE5 5mm/0.05mm	PS12 0.1mm/0.002mm
40	0.01mm - 1mm (10 - 1000µm)	NE1 10mm/0.1mm	PS8 1mm/0.01mm
40	0.002mm - 0.1mm (2 - 100µm)	NE5 5mm/0.05mm	PS12 0.1mm/0.002mm
60	0.002mm - 0.1mm (2 - 100µm)	NE1 10mm/0.1mm	PS12 0.1mm/0.002mm
100	0.002mm - 0.1mm (2 - 100µm)	NE1 10mm/0.1mm	PS12 0.1mm/0.002mm

# How to use a Stage Micrometer ?

## Reflected (Incident) Light

Objective Magnification	Measured Size Range	Eyepiece Graticule (Length/Divisions)	Stage Micrometer (Length/Divisions)
0.5	0.1mm - 10mm	NE5 5mm/0.05mm	PS1R 10mm/0.1mm
0.5	0.01mm - 1mm (10 - 1000µm)	NE28 1mm/0.01mm	PS78 1mm/0.01mm
1	0.1mm - 10mm	NE1 10mm/0.1mm	PS1R 10mm/0.1mm
1	0.01mm - 1mm (10 - 1000µm)	NE28 1mm/0.01mm	PS78 1mm/0.01mm
2	0.1mm - 10mm	NE5 5mm/0.05mm	PS1R 10mm/0.1mm
2	0.01mm - 1mm (10 - 1000µm)	NE5 5mm/0.05mm	PS78 1mm/0.01mm
4	0.01mm - 1mm (10 - 1000µm)	NE5 5mm/0.05mm	PS78 1mm/0.01mm
10	0.01mm - 1mm (10 - 1000µm)	NE1 10mm/0.1mm	PS78 1mm/0.01mm
16	0.01mm - 1mm (10 - 1000µm)	NE1 10mm/0.1mm	PS78 1mm/0.01mm
20	0.01mm - 1mm (10 - 1000µm)	NE1 10mm/0.1mm	PS78 1mm/0.01mm
40	0.01mm - 1mm (10 - 1000µm)	NE1 10mm/0.1mm	PS78 1mm/0.01mm



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