The Measurement of Numerical Aperture

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Summary
Two simple devices for the measurment of numerical aperture are described and also a device for the same purpose which consists merely of a 3 ×1-inch glass slide. The attainable accuracy of measurment is in all cases at least one per cent.

Introduction

UP TO the time of Abbe, microscopists were concerned only with the measurement of numerical apertures less than unity. For this purpose, the apparatus generally used consisted of a microscope barrel mounted horizontally on a protractor as shown in Figure 1. With the objective in position, the microscope tube was moved to the two extreme positions in which the light source was just visible in the back lens of the objective. The angle between these gave the angle of aperture (of the dry objective) and from this the numerical aperture, if desired, could be calculated. With the coming of water immersion and oil immersion objectives something

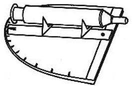

FIG. 1. CLASSICAL FORM OF APERTOMETER

more sophisticated was needed. The American optician Tolles suggested using the apparatus of Figure 1 but arranged in a vertical plane and with a glass hemisphere in immersion contact with the front lens of the objective as shown in Figure 2. By this means, knowing the refractive index of the glass of the hemisphere, the numerical aperture of objective could be calculated even though this exceeded unity.

Abbe devised his well known form of apertometer, first as a rectangle of glass with a bevelled back edge, and later with the more familiar semicircular glass plate with bevelled back edge. From the time of Abbe until about 1940 Abbe apertometers were fairly readily available, although at a considerable price, and any microscopist who wished to ascertain the numerical aperture of his objective could, if he desired, either borrow or purchase one of these instruments. Since the second world war apertometers have no longer been available, although for a time Beck supplied a simplified form in which a scale was observed through a glass block of appropriate refractive index.

The Beck apertometer, too, is a thing of the past and the microscopist who wishes to make measurements of numerical aperture may find himself in a difficult position. The purpose of this note is to describe two simple forms of apertometer, one of which consists basically of a 3 × 1-inch microscope glass slip and which suffices for numerical apertures up to 1·0, and the other is a simple device which may be fabricated from a small piece of Perspex and which will suffice for measurements up to 1·4.

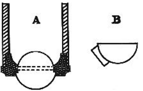

FIG. 2. TOLLES' ORIGINAL SKETCH FOR AN APERTOMETER

Apertures Up To 1·0

The reason for the glass block in the Abbe apertometer is basically to enable apertures greater than 1·0 to be measured. For apertures less than one the glass is unnecessary and all that is needed is a reflecting plane at 45 degrees to the optic axis of the microscope and a means of defining the limit of angular vision through the objective. The apparent stumbling block is, of course, the graduations which enable the direct measurement of numerical aperture to be made with Abbe's device. Many years ago, however, the eminent microscopist E. M. Nelson suggested that the graduations were unnecessary and that with microscopes possessing a graduated rotating stage a more accurate measurement could be made without using the graduations on Abbe's device. Nelson's suggestion was that moveable stops on the Abbe apertometer be removed and that the device be set up on the stage of the microscope with a small source of light some distance away. Looking down the barrel of the microscope with the objective in place, and without the eyepiece, a bright spot can be seen. Initially, the Abbe apertometer is set up with its straight edge at right angles to the direction from the objective to the source of light. The microscope stand is then set in such a position that the rotating stage reads 0 degrees. On rotating the stage with the apertometer in position the bright spot seen through the objective moves along a curve and eventually disappears from the back lens of the objective. At the point of disappearance, the position of the circular stage is read and the same is done again on the opposite side of the centre line. The sum of the two readings gives the total angle of aperture of the objective. In the case of Abbe's device, the angle so measured is the angle in glass and to find the angle in air for dry objectives account must be taken of the refractive index of the glass. Before explaining how this is done, however, we show in Figure 3 a simple device based on the Abbe apertometer and which is

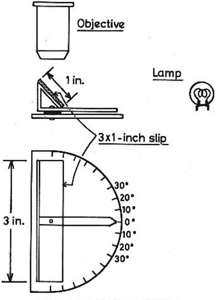

FIG. 3. PROTRACTOR APERTOMETER

applicable to the measurement of the numerical aperture of dry objectives in microscopes not possessing rotating stages. It consists essentially of a 3 × 1-inch slide mounted at an angle of 45 degrees to an aluminium pointer which can be turned about a protractor scale. The device is mounted on the stage of the microscope with the objective over the centre of rotation of the circular protractor scale. The device is used in exactly the same way as the Abbe apertometer and microscopists of a classical bent who desire a fuller description of the use of the latter apparatus can refer to the 1901 edition of Carpenter's book. In observing the image of the lamp in the back lens of the objective a cap perforated with a small central hole should be placed over the microscope tube. Alternatively, a low power eyepiece can be placed in position and the protractor readings taken when the field of the eyepiece is bisected by the bright band caused by the presence of the light on either side of the centre line when the microscope slide is rotated. If a microscope, such as the old Powell & Lealand No. 1,

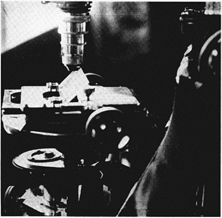

FIG. 4. POWELL & LEALAND MICROSCOPE WITH A 3 × 1-IN. SLIDE FOR MEASURING NUMERICAL APERTURE

with a rotating stage is available the only apparatus required is the 3 × 1-inch slide which can be balanced on the stage in the manner shown in Figure 4. It is quite unnecessary to focus the objective on the slide or in fact on anything at all. Users of the device can convince themselves that exactly the same readings are obtained whatever the position of the objective (within limits) with respect to the slide. This apertometer can be made for a few pence, it is quite accurate and is capable of giving values of numerical aperture which are accurate to three places of decimals, considerably better than the cheaper glass block form of the device.

Numerical Apertures Up To 1·5

The simple device just described will not work for oil immersion objectives except in the unlikely event that the user has a small glass hemisphere of the type proposed by Tolles. A very simple Abbe apertometer can, however, be made to fit in the place of the glass slide of the previous device.

It is shown in Figure 5 and is constructed by taking a small section of one inch diameter Perspex rod, which can be bought cheaply from hobby shops. The outer surface of this rod is well polished by the manufacturers. To make the apertometer, a slice of rod about $\frac{1}{4}$-inch long is cut. The back portion of this section is removed to the dimension shown in Figure 5.

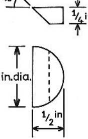

FIG. 5. PERSPEX-BLOCK APERTOMETER

A 45-degree bevel is sawn, filed to a good finish, and then polished, using fine emery and eventually Bluebell metal polish. It can easily be shown that, if a microscope objective is arranged to be in oil immersion contact with the top surface of this small apertometer and is focussed on the top surface of the Perspex plate then, in the horizontal plane, the image of the point of focus will always lie on the centre of the original cylinder. The refractive index of Perspex for sodium light is $N_d = 1{\cdot}49$ so that, assuming that the block is used in the same manner as the glass disc in Abbe's apparatus, and supposing that the angles about the centre line at which disappearance of the light source from the back lens of the objective takes place are θ_1 and θ_2 the semi-angle of aperture in Perspex is then $\frac{1}{2}(\theta_1+\theta_2)$ and the numerical aperture is simply $N_d \sin \frac{1}{2}(\theta_1+\theta_2)$ or $1{\cdot}49 \sin \frac{1}{2}(\theta_1+\theta_2)$. This apparatus is not quite so simple to use as the previous one since it is important to line up the centre of the block with that of the protractor or of the rotating stage of the microscope so that all centres coincide with the optic axis of the instrument. If this is not done error due to refraction in the Perspex surface will obtrude. With care, however, numerical apertures can be measured which are accurate to the second place of decimals. For those who are adept at working with glass it would be profitable to replace the Perspex cylinder by a section from a rod of soda glass, the refractive index of which is 1·52.