## The Measurement of Numerical Aperture ANDREW D BOOTH D.Sc., Ph.D., F.R.M.S.

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-inth glass slide. The statishable 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 apartures less than unity. For this nurvose, 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 less 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 APERTOMPTER

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 bemisphere 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 class plate with bevelled back edge. From the time of Abbe semicrousar gass pairs with occurring taxas eage. From the line of Assessment 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 nurchase 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 automorphic perfactive index.

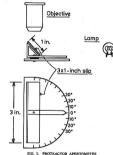
The Beck apertometer, too, is a thing of the past and the microscopist who wishes to make measurements of numerical specture may find himself in a difficult position. The purpose of this note is to describe two simple forms of apertometer, one of which consists basically of a 3 × 1-inch microscope glass slip and which suffices for numerical apertures up to 10, and the contract of the contract of



FIG. 2. TOLLES' ORIGINAL SKETCH FOR AN APERTOMETER

Apertures Up To 1-6 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 class 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 eveniece, 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



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 tabe. Alternatively, at low power reprises can be placed in position and the protractor readings take when the field of the epytice is blested by the bright and caused by the presence of the light on either side of the centre line when the microscope side is rotated. If a microscope, such as the old Powell & Lealand No. 1,



with a rotating stage is available the only apparatus required is the

3 X - Jonés silde which can be balanced on the stage in the manner shown in Figure 4. It is egule nancessary to focus the objective on the silde or in fact on anything at all. Users of the device one convince themselves that exacely the same readings are chained whatever the pointion of the objective (writink limits) with respect to the silde. This apertometer can be made for a leep proce, it is quite scientes and is capable of giving values of manufactual apperture with one accurate to there places of declarable of manufactual apperture with one accurate to there places of declarable or manufactual programs.

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 Abba spertometer can, however, he made to fit in the place of the elass side 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 14-inch long is cut. The back meriting of this section is removed to the dimension shown in Figure 5.



FIG. 5. PERSPEX-BLOCK APERTOMETER

A 45 degree havel is sown 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 ton surface of this small apertometer and is focusted on the ton 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. =149 so that, assuming that the block is used in the same manner as the slass disc in Abbe's annarutus, and sunnosing that the angles about the centre line at which disappearance of the light source from the back lens of the objective takes place are  $\theta_i$  and  $\theta_i$  the semi-angle of aperture in Perspex is then  $\frac{1}{2}(\theta_1 + \theta_2)$ and the numerical aperture is simply N<sub>a</sub> sin  $\frac{1}{2}(\theta_1 + \theta_2)$  or 1.49 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 Persney surface will obtrude. With care however numerical apertures can be measured which are accurate to the second place of decimals. For those who are adent at working with class it would be profitable to replace the Persney cylinder by a section from a rod of soda glass, the refractive index of which is 1-52.