PHOTOMICROGRAPHY BY INCLINED ILLUMINATION*

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ABSTRACT

Differences in the indices of refraction may be the only physical property distinguishing different materials in some thin sections. This can be illustrated photographically by inclined illumination but the entire area photographed should be equally illuminated. Modifications of the illuminating system of a microscope by which this can be accomplished are described, and illustrated.

A method here described for making photomicrographs by means of strongly inclined illumination was found to be particularly useful for presenting the properties of the Alamogordo glass. The Alamogordo glass, except for a minor part containing copper, is completely colorless and isotropic in thin section, and so a difference in the index of refraction is the optical property which serves to differentiate the various materials and to emphasize the range in their optical properties.

The methods commonly used for measurement of indices of refraction by means of inclined illumination involve the introduction of a half shadow at some plane in the optical system of the microscope. This results in brilliant illumination of part of the microscope field, while the other part is in partial shadow. These lighting arrangements, which are entirely suitable for index-of-refraction determinations, and which have the advantage of quick change from parallel to inclined illumination, are not suitable for photographic presentation of differences in indices of refraction. For these reasons, a description of the illumination system used in the petrographic laboratories of the United States Geological Survey to overcome these difficulties will probably be of general interest.

A diagram (Fig. 1) illustrates the lighting system as applied to a petrographic microscope, but the same method could also be applied to a biological microscope. The essential feature is a reflecting surface (*D*) placed below the stage of the microscope. This may be constructed as an attachment which will clip over the upper condensing lens of the microscope or be inserted in place of that lens. A glass mirror may be used, but a stainless steel mirror is preferable. This mirror should be constructed so that the reflecting surface makes an angle of about 10 degrees with the horizontal when the carrier for the upper condensing lens, which supports it, is turned into full position. It is well to construct the attachment so that the reflection angle is slightly too great for optimum results when the carrier is in full position. Then the system can be adjusted for greatest

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contrast by turning the mount carrying the reflecting mirror slightly back from normal position, and adjusting the vertical position by means of the screw which controls the vertical position of the condensing system.

A convenient light source is one which allows a rapid change from ordinary transmitted light to reflected light, and to inclined transmitted light. One light source serves all these uses, and is placed in a housing well

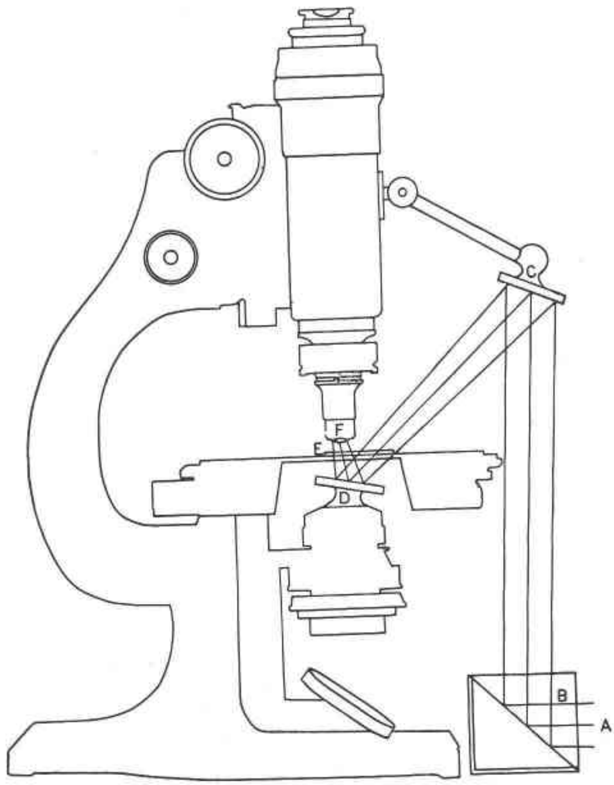

FIG. 1. Diagram of microscope equipped for microphotography by inclined illumination.

back from the microscope, where it is out of the way of all manipulation. A small mirror (C) is permanently attached to the tube of the microscope about 10 centimeters above the front lens of the objective by means of a short extension rod. This has a hinge where attached to the microscope which allows it to be turned up against the tube when not in use, and another hinge above the mirror to permit the reflection of the beam into the proper position.

A light source which just covers the mirror of the microscope (about

5 centimeters in diameter) is desirable for the usual microscopic observations, but for reflected or inclined transmitted light a narrower and more concentrated beam is needed (about 1.5 centimeters in diameter). For this purpose a condensing lens of suitable focal length is used with the reflecting surface (*B*). A small concave mirror, such as that used by dentists, is attached at (*C*). The light beam in this system is reflected three times, and travels four paths before it enters the microscope, but this does not introduce undue optical complexities. The light leaves its source as a horizontal beam of parallel light, is reflected into a vertical beam, and due to the condensing lens travels as a slightly convergent beam to the mirror (*C*). Here it is reflected again and travels toward the microscope stage at an angle of about 45 degrees. It passes through the slide (*E*) and strikes the inclined mirror (*D*) where it is reflected again and enters the microscope objective (*F*) as an inclined beam, which strongly accentuates differences in indices of refraction. The vertical beam between (*B*) and (*C*) has the advantage that the microscope follows it during focusing.

The light is transmitted through the slide, and so this must have fair transparency; large colored or opaque areas tend to cast a shadow which would be noticeable in photographs.

The same equipment, but without the mirror (*D*), is useful for many other purposes including the examination of thin sections for identifying such materials as leucoxene, distinguishing pyrite from magnetite, or observing partial or complete alteration to hematite. If desirable, a film of polaroid may be placed above the reflector (*B*) giving almost complete polarization so that the equipment can be used in index of refraction measurements.

This method of inclined illumination gives best results with objective having a 16 or 25 millimeter focal length, but it is also good with the objective having a 40 millimeter focal length. Lenses with a very short working distance cut off the light from the inclined beam; the objective with a focal length of 9 millimeter is usable but does not give contrast equal to that produced by the others. This means that the usable magnifications range from about 25× to 80×, but at 150× the results are not so good.

The mirror (*C*) may be replaced by a direct light source. Either a vertical illuminating system such as that of Bausch and Lomb or the universal microscope lamp of the American Optical Company is suitable. The light should be fairly brilliant and a 20 to 25 candle-power lamp should be used.