

Manipulation
of the Microscope

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MARINE BIOLOGICAL LABORATORY.

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MANIPULATION
OF THE
MICROSCOPE

BY
EDWARD BAUSCH

ILLUSTRATED

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FIFTEENTH THOUSAND

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PREFACE TO FIRST EDITION.

It may seem to some persons an act of presumption for a maker of microscopes and microscopic accessories to enter the field of authorship and attempt to supplement the valuable labors which in recent years have made the use of the microscope an indispensable aid in the advancement of science.

To such, if any, I submit, that being a producer of microscopes and their accessories, I have had opportunity to become acquainted with the lack of general knowledge of the fundamental principles of the instrument and the best method of technique, even among owners of microscopes. Indeed, with so many complications, with almost unlimited powers and uses of the instrument, the beginner cannot fail to feel the need of a guide and adviser.

In order to accomplish the greatest good, I have started out in this little Manual with the supposition that the purchaser, or owner, is a beginner, and absolutely ignorant of the microscope and everything which pertains to it, and therefore have attempted to convey, step by step, in as simple language as I could command, information which will, I trust, lead to ease of manipulation and give both pleasure and profit to those for whom it was specially written.

With these, its purposes and hopes, I beg for my self-imposed labor a friendly reception.

EDWARD BAUSCH.

PREFACE TO SECOND EDITION.

The demand for this book having considerably exceeded the expectations of its author and the comments on its utility having been so favorable leads to the view that it fills a gap in microscopical literature.

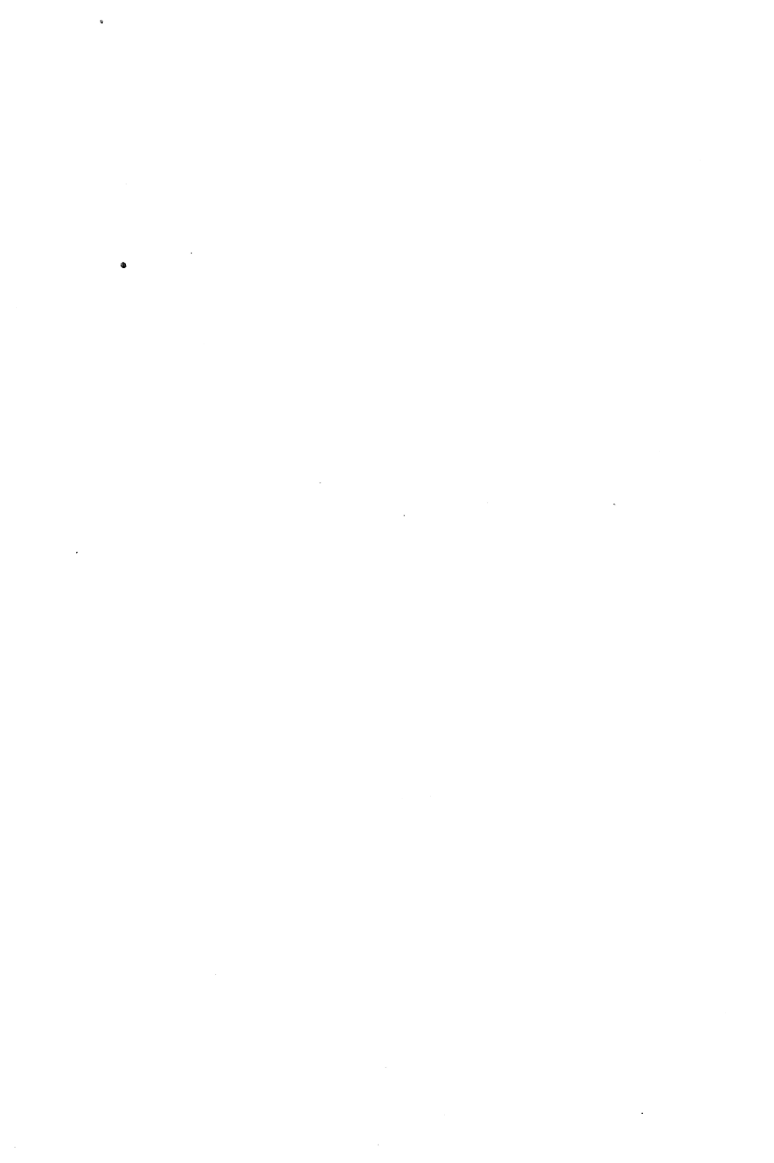
In preparing for a new edition an opportunity has been given for enlarging on some of the subjects and rewriting others, so as to make them conform to the changes which the last five years have brought about in the construction of apparatus.

While it may be true that many of the subjects might be treated much more extensively, the writer has purposely refrained from doing so, because he has considered it beyond the province of his intention and because books giving more extensive information are available.

An intending purchaser of a microscope finds it more or less difficult to make a suitable selection and while it is always best to consult an experienced microscopist, the writer has endeavored to convey information which, he hopes, will aid in this direction.

THE AUTHOR.

May, 1891.



PREFACE TO THIRD EDITION.

The past demand for this little volume makes extended remarks superfluous, the new edition appearing as evidence that it is considered of some value.

This edition has been almost entirely rewritten to bring it in accord with the advance which has been made in the construction of microscopes and accessories and while it is not expected to be a complete guide, it is, nevertheless, hoped that it will make the labor of the beginner more easy.

Since its first issue there have appeared two books covering the same purpose: "The Microscope and Microscopical Methods" by Prof. S. H. Gage of Cornell University and "Microscopical Praxis" or "Simple Methods of Ascertaining the Properties of Microscopical Accessories" by Dr. A. C. Stokes, both of which are heartily commended to the microscopist. Neither should be wanting in a microscopical library. The writer is also pleased to acknowledge the suggestions of an enlarged scope for this book, which he has obtained by a perusal of them, as well as from the admirable work of Dr. W. H. Dallinger in the latest issue of Carpenter, "The Microscope and Its Revelations"

which may be commended to those who wish to study more deeply the principles of the microscope and learn its history and development.

The writer trusts that omissions will be pardoned, as the only time which it has been possible to devote to this work has been the spare moments of a busy life and hopes sufficient information may be obtained to give full compensation for such defects.

THE AUTHOR.

March 1, 1897.

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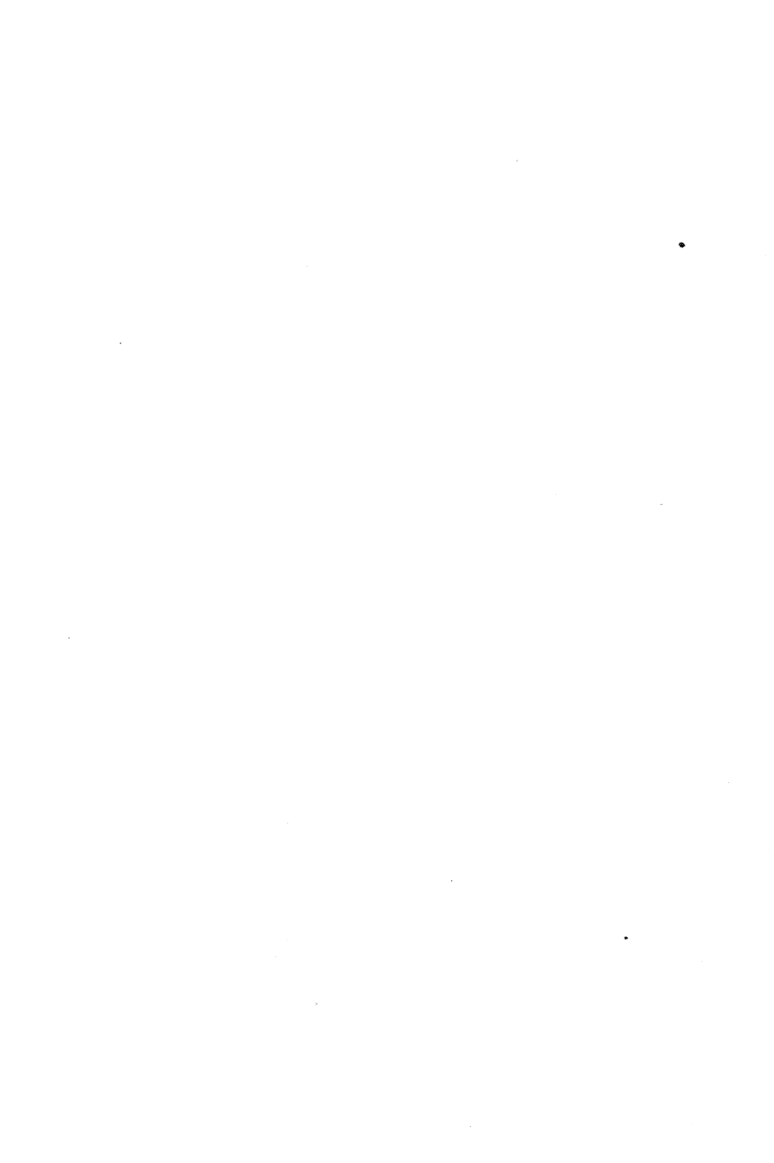
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INTRODUCTORY.

The knowledge which this volume attempts to convey in the proper use of the microscope, may be gleaned by following several methods.

First.—*By reading carefully all contents from first to last.* This is strongly recommended, as an effort has been made to make the contents progressive and it is hoped that in this manner a general knowledge of terms and optical principles involved will be acquired, which will greatly aid in intelligently and with greater facility and pleasure carrying out the instructions in the chapter “How to Work.”

Second.—*By reading the general principles and studying the various terms and omitting some of the instructive processes,* as “How to Judge Chromatic and Spherical Aberration,” “How to Measure Angle,” “How to Measure Working Distance,” etc., which may be carried out after some familiarity with manipulation of the instrument has been acquired.

Third.—*By studying the parts of the stand and then referring to chapter “How to Work.”* This is not in any case advised, but may be done when

there is a pressing requirement for the use of the instrument, or great anxiety to operate it. The previous chapters should then be studied later on.

Fourth.—If the reader does not possess a microscope and desires to purchase one, study the parts of the stand, general principles and “How to Select a Microscope.”

OPTICAL PROPERTIES OF LENSES.

Purpose of the Microscope.—The microscope is an instrument which magnifies near objects, so that we are better able to examine their structure than is possible with unassisted vision.

Simple and Compound Microscopes.—Microscopes are divided into two classes—*simple and compound*—the difference between the two being as follows: With the simple microscope the object is viewed directly and the magnified image shows the object erect, or in its real position. It must consist of one lens, but may consist of more, which however are in close contact. With the compound microscope a magnified image is observed and this in a reversed position from the true one of the object, so that what is right in the object is left in the image. *This must consist of at least two lenses* with suitable distance between them.

Lenses.—As microscopes depend upon the action of lenses it seems fitting that these, as well as the action of light passing through them,

should receive attention. Every person has unquestionably observed that when a spoon is placed in a tumbler of water it is apparently bent at the surface of the water, or when looking at an object lying in the bottom of a dish, it is apparently at a different point than when viewed from the side. This is caused by the great principle of actually bending the rays of light as they pass from one transparent medium into another of greater or less density and is called *refraction*. The amount of refraction increases as the difference in the density of the two media becomes greater. We also know that in viewing objects through a glass prism, they apparently lie in a different direction than their real one. The amount of this deviation is absolute and depends for the one factor upon the density of the glass composing the prism and for the other upon its shape.

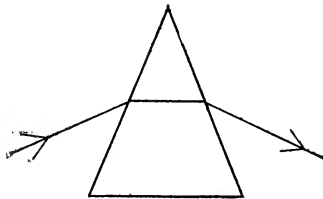


Fig. 1.

Fig. 1 represents a cross section of a prism and shows how the ray, as it touches the first surface of the prism, undergoes refraction and on

its emergence from the prism, its second refraction takes place; it will also be noticed that the light is bent downward or *toward the base of the prism* and if the prism be imagined reversed with its base upward, the action of the prism on the light will be the same, but its course will be upward, *always toward its base*.

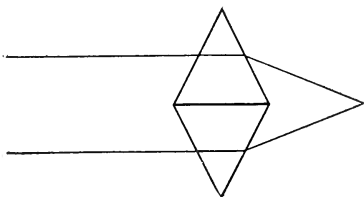


Fig. 2.

Now a lens in either of its two principle forms is nothing more in effect than the prisms showing as in Fig. 2, where the two bases are placed

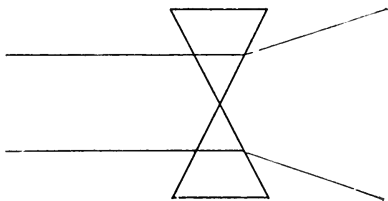


Fig. 3.

together, how the light is refracted toward the bases and thus converges; or how, as in Fig. 3, where the bases are above and below, the action of

the prisms on the light still remains the same, refracting the rays of light toward the bases, and in this case causing them to diverge or separate.

If the combinations of prisms be imagined with curved surfaces instead of flat ones, the action of light through them will be precisely the same and we have the two great classes of lenses, *converging or convex* and *diverging or concave*, as shown in Fig. 4.

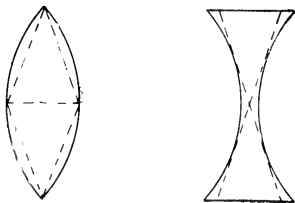


Fig. 4.

These two classes are again subdivided, as shown in Fig. 5, showing the three forms which

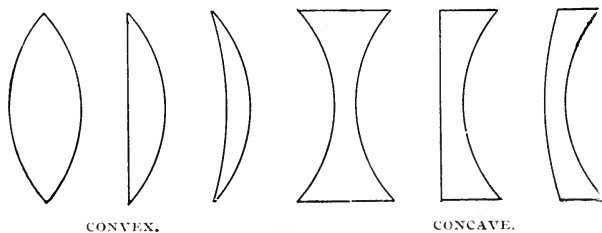


Fig. 5.

are made in each of the convex and concave types and which are called in rotation as drawn, *double*

convex, plane convex, convex meniscus and double concave, plane concave, concave meniscus.

In the convex lenses, parallel rays are converged to one point as shown in Fig. 6, in which c is

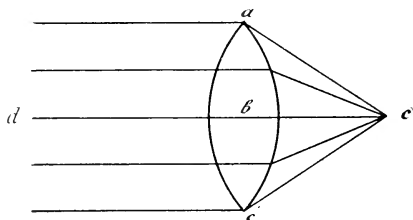


Fig. 6.

the *principal focus*, or *focal point* and the distance from b to c the *focal distance*. With the same lens, if a flame be placed at c , all the rays which strike the lens will be emitted in a parallel direction on the opposite side. The straight line $d b c$ which passes through the middle of the lens is called the *principal axis* and the line $a c$ the *radius*.

The radius determines the power or converging quality and as it becomes longer, reduces the power by making the lens longer focus. If in a double convex lens the radius of each surface is 1 inch, the lens has a focus of 1 inch and if in a plane convex lens, the convex surface has the same radius, the focus is twice as long, or 2 inches.

In a concave lens the action of the lens is oppo-

site; instead of converging the light toward the axis, it diverges the light from the axis as shown in Fig. 7. The imaginary extension of the

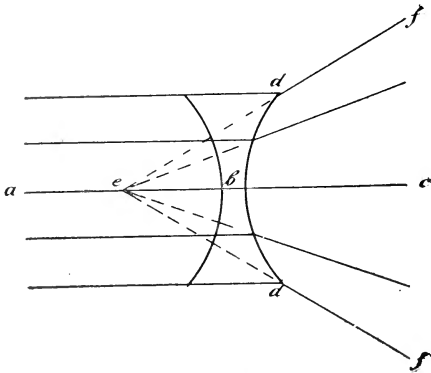


Fig. 7.

diverging rays should meet at *e* and the distance *e d* indicates the *virtual focus*, in contradistinction to the *real focus* in the convex lens.

To Determine the Focus of a Lens.—The focus of a lens may be quite accurately determined by the following methods: Taking the sun as an object, hold the lens toward it and bringing a piece of paper under it, move it slowly toward the lens. A large bright spot will appear, which, as the paper is brought nearer, will decrease in size but increase in intensity until it becomes quite minute and as the paper is brought still closer to the lens,

the image will be found to enlarge again. Return until the most intense point is reached again. By holding sufficiently long, especially if the paper be dark, it will be found to burn, due to the concentration of heat rays and hence is called the *burning point* or *focal point*. Or, in a room opposite a window or lamp, hold a rule against a white wall and placing the edge of a lens upon it, move it slowly toward the wall until a greatly reduced, but bright image of either object appears and read off the distance between the wall and edge of lens, which is its focus.

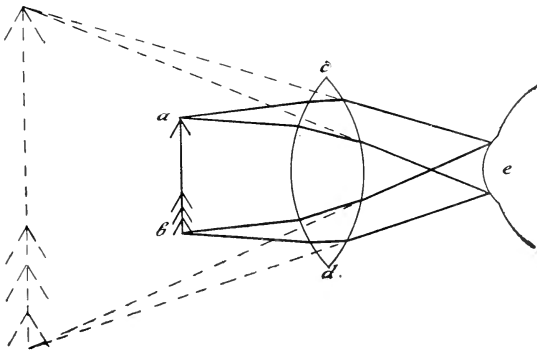


Fig. 8.

Magnifying Power.—Magnifying power of a convex lens depends also upon its convergence. In Fig. 8 *a b* represents the object, *c d* the lens, *e* the pupil of the eye. By following the

course of rays from the object it will be noted how they are converged and refracted by the lens and intercepted by the pupil of the eye. If now the lines between c and c' be considered elongated, they will be found to meet beyond $a b$ and there form a *virtual image*. It will be well to understand at this point the difference between a real and a virtual image. The *real image* is one which can be accurately seen and projected upon a surface, as in the magic lantern, or in the picture which is given by the photographic camera. The *virtual image* cannot be so shown. In a lens of less convexity or longer focus, there is less convergence of rays and the length of the virtual image is consequently reduced and thus the magnification is less.

Spherical Aberration.— In considering the refraction of light by a lens up to this point, we have purposely avoided mentioning another quality which is incident to it. In magnifying an object by a single lens it will be noticed that the virtual image as seen through its central portion is quite clear, while that near the margin or edge is quite indistinct. This is due to *spherical aberration* and the extent of this aberration increases with the power of the lens. It is due to the unequal refraction between those rays passing near the margin and those passing through the central portion, so that the rays instead of combining at the focal

point, come together at different intervals along the central line or axis. By reference to Fig. 9 it will be seen that the outer, or marginal rays are

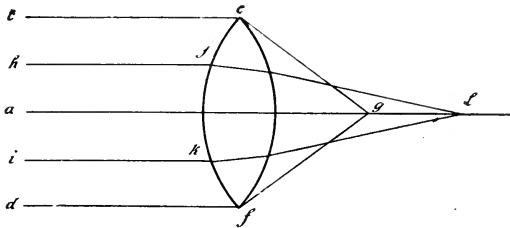


Fig. 9.

refracted at *c* and *f* so that they will combine at *g* and the inner, or central rays are refracted at *j* and *k* so that they will meet at *l*. In the same manner will the rays which enter between *c* *h* and *i* *d* come together at intermediate points between *g* *l* and those of the central portion between *h* and *i* will fall beyond *l*. The amount of spherical aberration depends upon the shape of the lens and in different lenses of the same focus, is greatest in the double convex form, less in the plane convex and least in the so-called crossed lens, in which the two surfaces are of different radii and in the proportion of 1 to 6, *on condition however*, that the surface of short radius is directed toward the object. This form of lens however, is seldom used, as it shows the greatest amount of aberration if used in the reversed position. The

most common form is the double convex and when considerable magnifying power is desired, the defects of spherical aberration are partially overcome by the interposition of an opaque plate with round opening of such size that it will shut out the marginal rays. This is called a *diaphragm* and when used the lens is said to be *stopped down*.

Chromatic Aberration.—In magnifying an object with a single lens it will be noticed that it has not only the defect of spherical aberration, but that the object is fringed with colors, predominantly violet and red, or if objects are viewed through a prism, we have not only an apparent change of position, but a decided appearance of so-called rainbow colors. This appearance is called *chromatic aberration* and is a result of refraction. It is caused by the dispersion, or *dispersive quality*, the separation of light into its primary colors, *violet, indigo, blue, green, yellow, orange* and *red* in the order given. As the light is refracted by a lens or prism on its emergence, the violet end being more refrangible will be principally affected and be brought to a focus within, and the red to a focus beyond the principal focus. This band of colors is called the *spectrum* and is nicely illustrated in the diamond. This has a very high degree of refractiveness and is polished or cut to make a many-sided prism in which each face or facet creates refraction with its consequent

dispersion and play of colors. To avoid to the greatest possible extent the spherical and chromatic aberration, has been for many years the study of opticians.

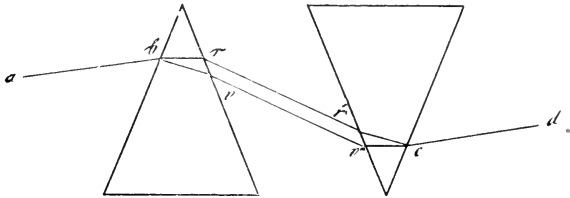


Fig. 10.

If in Fig. 10 the beam of light $a b$ is followed it will be found to separate at the first surface of the first prism and still more at its emergence from the second surface. The violet ray undergoing the greatest amount of refraction, takes the direction of $v v'$, and the red ray being refracted to a lesser extent, follows the course of $r r'$. To neutralize or correct this dispersion a second prism, identical with the first, but in a reversed position, may be placed to intercept these rays, so that they will be recombined, as shown in $r c$ and $v c$ and finally emerge as white light. Now while the dispersion can be corrected in this manner, it will be noticed that the emergent ray $c d$ from the second prism takes the same direction as the incident ray $a b$ and that this course corresponds

to the diverging rays of a concave lens. It will be remembered however, *that in order to have image forming rays they must converge.*

As dispersion depends upon two conditions, the angle of the prism or convexity of the lens and dispersive power of the glass, it will be seen that in two prisms of the same angle but made of glass of different dispersive power, the separation of the red and violet rays will be *greatest* in that having the *greatest dispersive power*. Or, in two prisms of the same density but one having one half the angle of the other, the dispersion will be one half as great in the prism of smaller angle. Again, in the prism of twice the angle but one half the dispersive power of another the dispersion will be the same. We also know that in a prism having a greater angle the refraction will be greater than in one of lesser angle and as a refraction must be maintained which will create convergence while at the same time neutralizing the dispersion, a solution will offer itself in using in conjunction with a prism of a certain angle and dispersion, another of lesser angle and greater dispersion. If the same principle be carried out in lenses, a convex lens of low dispersion would be combined with a concave lens of high dispersive power. Such a lens is shown in Fig. 11, and is called an *achromatic* or *corrected lens*. If the chromatic and spherical aberrations are both corrected it is called

aplanatic. The convex lens is made of *crown glass* and the concave of *flint glass*.

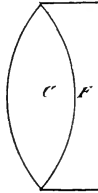


Fig. 11.

The corrected lens shown in Fig. 11 is an achromatic lens of the simplest form and while not absolutely corrected is generally used when the demands on it are not too great. Better correction is obtained when two or more of these lenses are used in combination and they are thus used in some of the lenses of the compound microscope. The variety of forms, due to the variety of glass from which combinations may be made, is almost infinite.

SIMPLE MICROSCOPES.

Simple microscopes are usually termed *magnifiers* and whether consisting of one or several lenses in close contact, *always remain simple*. They are made to be held in the hand or are fixed

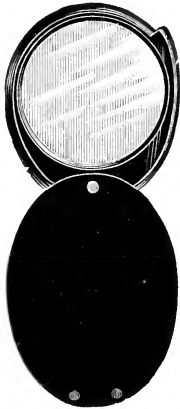


Fig. 12.

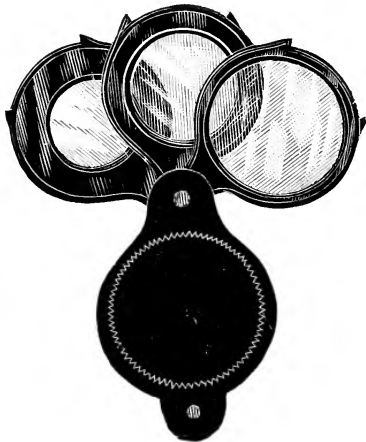


Fig. 13.

on a *stand* or *mounting*, with provisions for adjusting for focus, thus to give steadiness and leave the hands free for dissecting or moving the object

during observation. They are made in a large variety of forms, the difference appearing in their optical as well as mechanical construction. The most common are those with one or several double convex lenses and mounted in hard rubber or vulcanite, as shown in Fig. 12 and Fig. 13. Those containing several lenses are preferable, since they offer a variety of magnifying powers and in combination give the greatest magnification admissible with single lenses.

Coddington Lens.—A lens of greater efficiency than the ordinary magnifier is the so-called *Coddington lens*, Fig. 14. While this is also a single

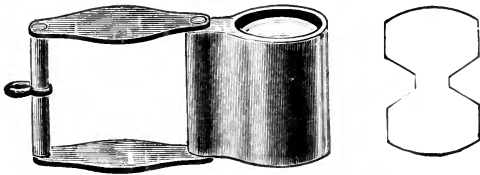


Fig. 14.

double convex lens it will be noticed that it has considerable thickness, being really the central portion of a sphere and provided with a circular incision at the middle, which is blackened and thus acts as a diaphragm, shutting out the marginal rays and correcting the spherical aberration, at the same time however, limiting the size of field.

Aplanatic Triplet.—The best type of magnifier is the *Aplanatic Triplet*, Fig. 15, being composed of three lenses cemented together in such a manner that they are virtually one. For many years

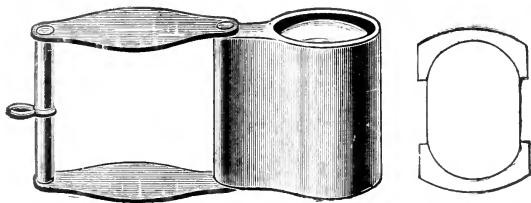


Fig. 15,

this form was, on account of cost, used only to a limited extent, but an increased demand has brought about a greatly reduced price. While it corrects both aberrations it gives a *large* and *flat field* and can be commended to those wishing a good magnifier.

Hastings Triplet.—This form of lens has been recently computed by Prof. C. S. Hastings of Yale University and is a modification of the Aplanatic Triplet, giving not only the highest spherical and chromatic corrections, but a considerably *flatter* and larger *angular field* and longer *working distance*; that is, the distance between lens and object is greater and is an important feature in higher powers, as it admits of better illumination.

In all of the better types of magnifiers the

vulcanite mountings are discarded for those made of metal, especially German silver, although many are used of pure silver and some even of gold.

Reading Glass.—When magnifying lenses reach a diameter of 2 inches or more they are usually termed *Reading Glasses* and are then provided with a handle. They are used to enlarge small printed matter, or to determine detail in engravings and photographs.

Bruecke Lens.—This is a combination of lenses named after the inventor which, while at one time a popular construction went almost into disuse, but in recent years has been found to have valuable properties and is now very generally used. A combination of achromatic lenses is superposed by an achromatic concave lens and gives a very satisfactory image. Much higher power can be obtained than would be possible in any other form of simple microscope. While it has the disadvantage of a small field, this is more than compensated for by a much larger focal distance than would be the case in the ordinary forms of simple or compound microscope, thus making dissection possible during observation and a further advantage over the compound microscope, in that the image is erect as in the simple form and not reversed as in the compound. A change of powers is made convenient either by changing the lenses or varying the position of the eye lens.

Stands and Holders.—There is a great variety of mechanical contrivances for holding magnifiers, to give steadiness as well as to leave the hands free for moving and working on the object and adjusting for focus. When provision is made to hold and adjust the lens the apparatus may be called a *lens holder* or *stand*, but when a platform called a *stage* is added, upon which the object can be placed, and a mirror for illuminating the object properly, it is called a *dissecting microscope*.

There are two forms of magnifiers which contain within themselves some properties of stands, one a *Tripod Magnifier*, Fig. 16, rests upon three legs

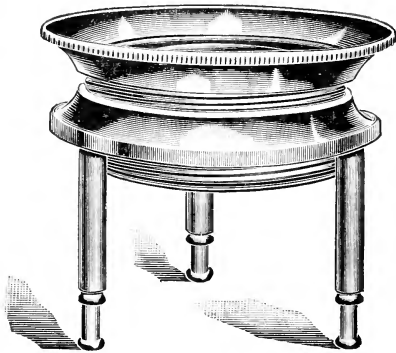


Fig. 16.

and has a screw for adjustment of focus. Its optical parts are two convex lenses, usually having a power of 10 diameters, and which are separated

by a diaphragm, giving a large, fairly flat field. It is especially used in primary botanical work.

Another is the *Lincoln Tester* (Fig. 17). It is made in various sizes, but the ordinary form has a

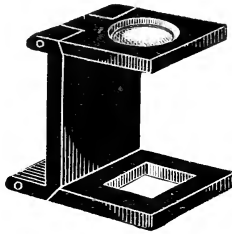


Fig. 17.

lens of 1 inch focus. It is arranged to fold into small compass for convenience in carrying and when opened for use, is placed over the object so that this comes into the square opening in the base. Its principal purpose is however, as its name indicates, its use in the textile industries for counting the number of threads which appear in the standard opening of $\frac{1}{4}$ inch.

The most simple form of holder is a base to which is fixed a stem or rod upon which the magnifier may be adjusted. Fig. 18 shows one with a white opal glass plate for placing the object upon. In this or other similar forms the advantage of holding the lens is counterbalanced by disad-

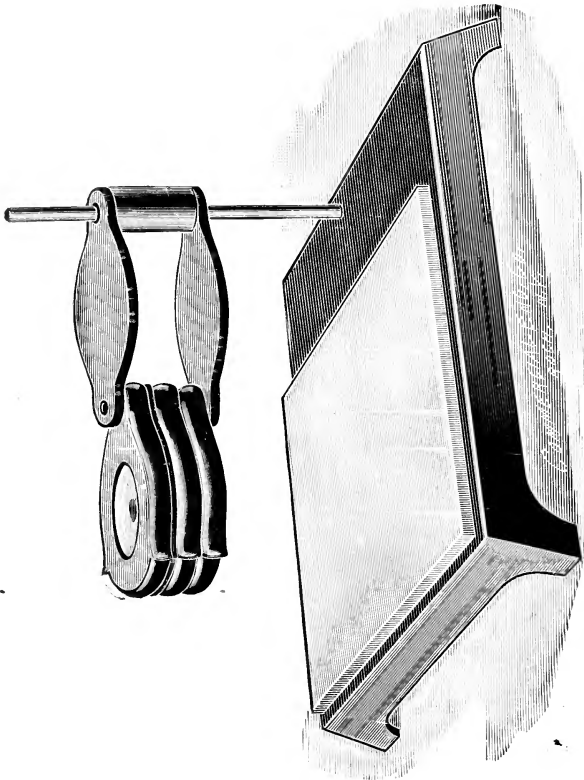


Fig. 1.

vantages, such as the necessity of holding the base when adjusting the magnifier and the danger to the eye from the projecting stem.

The Excelsior Dissecting Microscope (Fig. 19) can be recommended when portability is a desideratum and for school use in primary work, the dissecting microscope designed by Prof. C. R. Barnes. This meets many conditions of a dissecting microscope in a simple manner.

There is a variety of more complex forms, giving a number of advantages, such as stability, convenience of varied illumination, delicate means for and long range of adjustment, jointed arms for moving the lens conveniently over a large field, etc., to the various descriptions of which the reader is referred to catalogues.

How to use Magnifiers and Dissecting Microscopes.—It is generally admitted that the intelligent use of a magnifier is a great aid in microscopical studies and while its use is a simple matter, some words of advice may be of aid in obtaining better results, or lead to doing work with more comfort. In all work, whether with simple or compound microscopes, it is a good plan to start out with the principle *not to use a greater magnifying power than is necessary* to accomplish the results in view.

It should be made a habit at the outset to keep both eyes open.

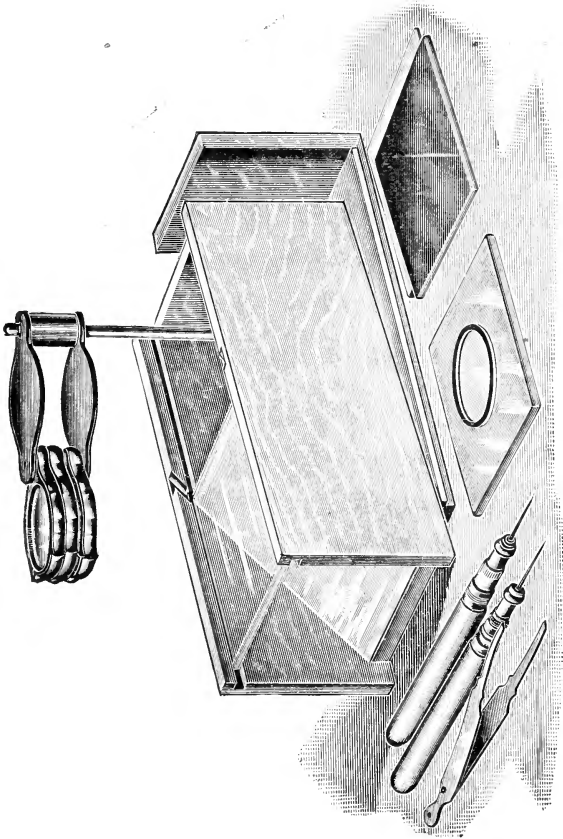


Fig. 19.

Keep the eye comfortably near to the upper surface of the lens, as the angular view is increased and there is the least spherical aberration and the focal distance is the greatest. This can be easily tested by gradually increasing the distance between the eye and lens, when it will be found that the lens must be brought nearer to the object. In single lenses the spherical and chromatic aberrations become more pronounced and the field smaller.

As magnifiers which are used on opaque objects—those which are not transparent and which are illuminated by reflected light not transmitted through the object—a position should be chosen toward a window or flame, which will allow the greatest amount of light to reach the object. If a hat is worn, place this back on the head so that the rim or shield will not cut off the light.

In holding the object in one hand, take the magnifier between the thumb and forefinger of the other and place the fingers of hand holding the lens in such a manner that they shall rest upon the other hand; this will insure steadiness between the lens and object and will add considerably to the comfort of working.

While it seldom occurs that magnifiers are made with other than double convex surfaces, single achromatic lenses are sometimes used which are plane convex. In these the plane side should

always be toward the eye. In the case of two plane convex lenses, they should be used with their convex surfaces toward one another.

In magnifiers containing several lenses, when these are used together, the one of highest power should be nearest the object. When reversed the angular field is greater, but presents considerable spherical and chromatic aberration, which it is advantageous to limit.

In simple dissecting microscopes like the Barnes, in which the mirror is in a fixed position, the microscope should be set squarely before the source of light. Diffused light, such as daylight, is always preferable to any artificial illumination.

It should always be sought to modify the light as much as possible and still have enough to see easily as the eyes are much less fatigued.

While in some classes of work it is perhaps unnecessary to have the very best magnifiers, such as the Aplanatic or Hastings Triplets, the latter can always be recommended when the means will permit on account of the higher results and greater degree of satisfaction and comfort derived from them.

Magnifying Power.—Unless a microscope, whether simple or compound, is known to come from the hands of a reliable firm, any claim as to magnifying power should be accepted with reserve. In former years, when the country was

overrun with cheap foreign productions, the most fanciful claims were made in this direction. Avoid strolling or street venders who, as a rule, not only make the most ridiculous claims as to magnifying power, but charge much higher prices than the same articles can be bought for from reliable opticians and generally offer articles of worthless or at best doubtful value.

Some precaution should also be used in reference to quality in purchasing a magnifier. As competition causes a downward tendency in prices, it unfortunately often involves a deterioration in quality. The ordinary forms are mounted in vulcanite; black horn is often palmed off as such, but is a poor substitute as it warps and cracks. The surfaces of lenses, instead of being perfectly polished are often scratched and unfinished showing small pit-holes or undulating surfaces. This is common among cheap Coddington lenses and naturally prevents obtaining a distinct image.

It is evident that a lens magnifies an object equally in all directions; this is said to be in *areas*, and is the square of the *linear*, so that if an object is magnified four times in the linear, it is sixteen times in area. The commonly accepted term to express magnifying power of simple, as well as compound microscopes, is in *diameters* (linear). A single lens of 1 inch focus magnifies about ten diameters; one of 2 inch focus, about five diameters;

one of $\frac{1}{2}$ inch focus, twenty diameters and so on. In a lens of high magnifying power, the focus is ordinarily about twice the diameter, so that if a lens is $\frac{1}{2}$ inch diameter its focus is about 1 inch.

To Determine Magnifying Power.—While the determination of focus in single lenses gives approximate magnifying power, it cannot be done in some of the forms which have been described. The following method, if carefully followed, will give very accurate results and is withall simple and interesting. Upon the upper or farther edge of a sheet of white paper, which rests upon a table with the light at the right or left hand, place a series of books of such height that when the magnifier is placed upon the inner edge or that nearest the body, of the upper book with the lens projecting over it, the distance to the upper surface will be exactly 10 inches and weigh down the mounting with an additional book or weight. Now place a pocket rule between the leaves of upper book so that when the edge is close to the magnifier the divisions on the rule will be exactly in focus, place the rule so that it shall not project much over the lens. It is immaterial what the divisions on the rule are, whether inches or millimeters, so long as they are reasonably fine. View the divisions with the right eye and open the left eye, when it will be found that the divisions are apparently projected upon the paper. Take a

pencil and outline upon the paper one of the spaces. By dividing the actual number of divisions on the rule into this enlarged space the exact magnifying power will be determined. Thus, if it is found that five spaces are contained in the one space on the paper, the magnifying power is five and the focus of the lens 2 inches, or if ten spaces, it is ten with a focus of 1 inch. One or several lenses in conjunction may be examined in this way. Some difficulty may or probably will be experienced in seeing the divisions on the rule and on the paper at the same time, but this will be overcome with a little practice. Indeed, it is well to point out at this stage that both eyes should be kept open in viewing objects through simple as well as compound microscopes, as continued work can be done with infinitely more comfort and while at first some difficulty may be experienced, it will be found that after a little earnest effort, both eyes unconsciously remain open and the prominence with which objects have appeared to the unoccupied eye is less, as the mind becomes intent upon the object it is viewing..

THE COMPOUND MICROSCOPE.

As has been stated a *magnified image* is observed in the Compound Microscope. Any two lenses, one of short, the other of long focus, placed sufficiently far apart, will attain this object and this was for years the method of its construction.

In any microscope, whether simple or compound, the difficulty of holding it or the object steady during observation, increases with the increase in magnifying power and in the compound form with only a moderately high power, it is utterly impossible to retain sufficient steadiness to make any reliable observation. Mechanical contrivances therefore became a necessity and were applied in the very earliest constructions of the microscope. Even when such a luxury as an achromatic lens was unknown they were all made to fulfill the following conditions :

A platform for holding the object.

A means of adjustment for properly focusing on the object.

Provisions for suitably illuminating the object.

From what may be called a crude attainment of these three purposes, the construction gradually became more complex. Many additions have

been made which have proven useful and have remained, while others have been discarded. As the first microscope was constructed in 1590 it has required nearly three centuries to bring the instrument up to its present general form and it is interesting to note that many improvements which have been introduced within the last forty or fifty years have been used and lost sight of within this time.

While certain parts are necessary to make up a modern instrument, no one design of construction is followed. The forms are innumerable, each maker following his own inclination in variety, design, number of parts and material. For the latter brass predominates, although bronze and iron are used to a considerable extent. The first two metals are usually highly finished and as they easily tarnish, are protected by lacquer, which is not only serviceable in this direction when well done, but offers a means of ornamentation. Iron is covered with a heavy coating of japan and being dark is on this account often recommended by instructors as being agreeable for the eyes. The entire apparatus, including the optical parts, is called a microscope, whereas, without them, it is termed a *stand*. Some people call it a "machine," but we earnestly protest against this harsh term as applied to an instrument of such precision as the microscope is.

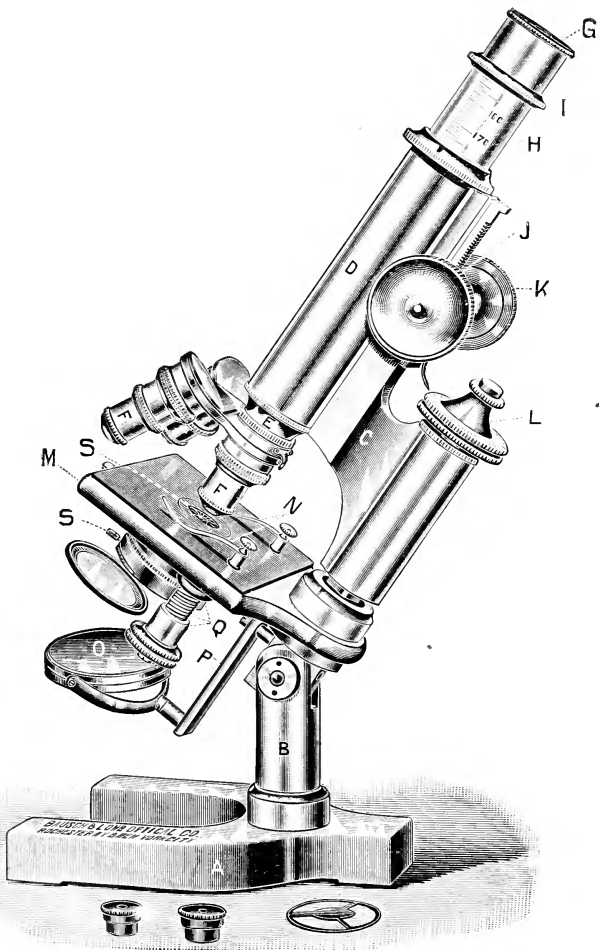


Fig. 20.

Description of Parts.—As it is necessary for the student to become conversant with the terms of the various parts and to understand their use, we give an illustration (Fig. 20) with letters and append a list giving the names and recommend that they be impressed upon the memory, as they are the basis of microscopical language.

A. Base.—This is the foundation of the instrument. It usually rests upon three points (or should do so) and is of such a weight that it keeps the instrument firm when it is in an upright or inclined position. The two principal forms are the tripod and horseshoe. The revolving plate, when this is provided, by means of which the upper portion of the instrument is revolved, without changing the position of the base, is considered a part of it.

B. Pillar.—It is the vertical column which is fastened to the base and carries upon its upper end the joint or axis which is provided for inclining the instrument. It generally consists of one piece either round or square but is often, in larger instruments, made in two columns.

C. Arm.—This supports all the upper working parts of the instrument and carries the provisions for adjusting for focus.

D. Body.—This is the tube portion to which the optical parts are attached.

E. Nose-Piece.—This is an extra piece which is attached to the lower part of the tube.

Society Screw.—This is a standard screw which is cut into the nose-piece and is called so from the fact that it was first established by the Royal Microscopical Society of London. It is also called the *universal screw* and is in general use in this country and England; it has lately been adopted by some firms on the continent of Europe.

F. Objective.—This is screwed into the nose-piece and is called so because it is nearest the object. It is the most important of the two optical parts (of the microscope proper) and upon its perfection the distinctness of the image and therefore the value of the instrument almost entirely depends.

G. Eyepiece or Ocular.—It is called so because it is nearest the eye and is the remaining optical part. It magnifies the image given by the objective. This and objective will be fully treated later on.

H. Draw-Tube.—This is that portion of the body which moves in the outer sheath and which receives the eyepiece. It is provided for the purpose of attaining different lengths and variations in magnifying power.

I. Collar.—This is a ring which is attached to the draw-tube and is usually provided with a *milled edge*.

J. Coarse Adjustment.—This is a provision for moving the body quickly back and forth for adjusting the focus approximately. It is done by a sliding rack and stationary pinion (not shown in cut) or a sliding body in an outer sheath.

K. Milled-Heads.—These are attached to the shank of the pinion, which is revolved by means of them and are usually large to give sensitiveness to the movement. They should be placed wide apart so that the fingers may be entirely free from the body.

L. Fine Adjustment.—This is slow moving and serves to get an exact focus. It is attained by a fine screw, provided with a milled head and acts upon the body, either directly or by levers. This as well as the coarse adjustment should be extremely sensitive and should not have the least side or lateral motion. The fact that either of them have it, is evidence of poor workmanship.

M. Stage.—This is the portion on which the object is placed for examination and is usually attached to the arm, although the arm is sometimes attached to the stage.

N. Clips.—These are two springs which are attached to the upper surface of the stage and serve to hold down the object.

Centering Screws.—These are provided for moving the stage in different directions to bring the center of its revolving motion in the center of the field.

O. Mirror.—This is used for reflecting and condensing light upon the object. As a rule two mirrors are used, one plane and the other concave. The first gives a comparatively weak light, while the second concentrates it and gives it more intensity.

P. Mirror Bar.—This carries the mirror and swings in a circle around the object in order to illuminate it from various directions.

Q. Substage.—This is a ring or attachment below the stage to receive various accessories which may be required. It is sometimes fixed to the stage but in the best instruments it is separated from it and is provided with an adjustment to vary its distance from the object.

Substage Bar.—This receives the substage and permits its adjustment. In modern American instruments this, as well as the mirror-bar, is on an axis in the plane of the stage, so that whatever position they may be in, relative to the object, the

distance from this to the substage or mirror does not vary, except when made to do so.

S. Diaphragm.—This is a provision for stopping down or regulating the amount of light which illuminates the object.

Optical Axis.—This is an imaginary line which passes from the center of the eyepiece through the centers of the body, objective, stage and substage to the mirror. Whatever lies in it is said to be *centered*.

Object.—That which is examined.

Slide or Slip.—This is a thin plate of glass upon which the object is placed or mounted, the prevailing standard being 3 inches long by 1 inch wide.

Cover Glass.—This is an extremely thin piece of glass, round or square, which is placed upon the object, either for flattening or preserving it, or both.

Classification of Microscopes.—Until recently microscopes were divided into two classes, the Jackson and the Ross models. While the latter was for many years very popular, particularly with the English makers, it has been almost entirely superseded by the Jackson form and with good reason. In the former the means of adjusting were provided, as near as consistent with the construction, to the body or tubes, whereas in the

Ross they are placed at the back or more distant point in the instrument, thus increasing by means of the connecting arm the faults which might exist in the adjustment.

A certain form of instrument which at the present is very popular and called the *Continental* pattern, from the fact that it was made originally by the manufacturers on the Continent of Europe, is a combination of both the Jackson and Ross models. Whereas the coarse adjustment, when consisting of a rack and pinion, is placed closely to the tubes, the fine adjustment is placed in the arm.

There is another direction, however, in which microscopes are divided into two classes, which is of far more importance, and affects their utility in a much higher degree. The writer does not know that instruments have been so classified by others, but knows that they can be with perfect propriety. The distance between the eyepiece and the objective is one of vital importance in the optical results and as there are several lengths of tube, the optical qualities of an objective are injuriously affected if it is used with a different length than that for which it was originally intended. It is therefore proposed to classify microscopes according to their tube length as *long tube* and *short tube* instruments.

Tube Length.—In the Continental form (Fig. 20), a short tube from 160.0 to 170.0 mm. (6.3 to 6.7 inches) is used, whereas in the English form, this

	Pts. included in Tube-length. See Diagram.	Tube-length in Millimeters.		
a-d	}	Grunow, - - - 203		
		E. Leitz, - - - 170		
		Nachet et Fils, - - 146 or 200		
		Powell and Lealand, - 254		
		C. Reichert, - - - 160 to 180		
		Spencer Lens Co., - - 235 or 160		
		W. Wales, - - - 254		
		b-d	}	Bausch & Lomb Opt. Co., 216 or 160
				Bezu, Hausser et Cie., 220
				Klonne und Muller, - - - 160-180 or 254
W. & H. Seibert, - - 190				
Swift & Son, - - - 165 to 228½				
C. Zeiss, - - - 160 or 250				
a-g	}	Gundlach Optical Co., 254		
		R. Winkel, - - - 220		
-d c-d	Ross & Co., - - - 254			
-e c-e	R. & J. Beck, - - - 254			
-f c-f	J. Green, - - - 254			
-f	Hartnack, - - - 160-180			
-f	Verick, - - - 160-200			
-g	Watson & Sons, - - 160-250			

Fig. 21.

being largely followed in America, the length is from $8\frac{1}{2}$ to 10 inches (216.0 to 250.0 mm.) The short tube of the European makers offers no optical advantages, but is mainly used to contract the height of the instrument to as great an extent as possible, as this is the vital point throughout its construction.

Until recently this subject was given little attention, each maker following a standard which he had adopted for himself. The pernicious influence of this diversity was not appreciated by the public, as it was not acquainted with the products of different makers, until Prof. S. H. Gage made it the subject of a paper before the American Society of Microscopists and as a result of his inquiries, tabulated the standards as followed by the different makers, which is published herewith.

As a result of his reports a committee was appointed to consider this subject, as well as that of eyepiece, objective and thickness of cover glass, to which we will recur farther on and reported in favor of the adoption of two standards for tube length.

Short standard 160.0 mm. (6.3 inches); long standard 216 mm. (8.5 inches); that the tube length shall be considered those parts between the upper end of the tube where the ocular is inserted and the lower end of the tube where the objective is inserted. While the European makers have paid

little heed to this standard, we believe that all of the American firms follow it and if it has accomplished nothing with the foreign producers, it is at any rate a mean between the standards of the leading makers and with the publicity which the matter has received, gives the user an opportunity to use his intelligence to obtain the best results. There are no optical advantages in the one or the other. The short length is almost a necessity however, in the Continental pattern of microscopes as compactness is the special desideratum, but while this subject will be given more extended attention and optically considered farther on, it might be here stated that when an objective, except perhaps in the very low powers, is constructed to be used with a certain length of tube, it should be used with this length only. This statement cannot be made too prominent and will bear repetition.

Stage.—This being the plate or platform on which the object is placed, it should be of a strength to stand the weight of the finger under considerable magnification. This depends upon the material of which it is made and its thickness, but since the material is virtually the same in all instruments, we must depend upon the thickness for rigidity. Absolute rigidity is practically impossible when considerable force is exerted, as can easily be determined in the best instruments and

it is a mistake to condemn an instrument for this cause as is sometimes done. If the object will remain in focus under a high power with a fair amount of exertion above that which is required in moving the object about, the stability may be considered ample. In fact in this, as well as other directions, the writer considers it unwise for a user of a microscope with, in most cases, a necessarily limited knowledge of its construction, to make assertions, as is often done, as to supposed defects against the long experience of the microscope maker, although it is the purpose of this book to aid the reader in judging of defects when such exist. In older instruments the fault often occurred of making the stage unnecessarily thick, which interfered with the proper use of substage accessories and the mistake has in recent years been made by some makers of having it too thin, so that a slide could not be moved except with the utmost care without depressing it and thus putting the object out of focus. At the present time, however, the writer believes that in instruments of the best makers the thickness is ample, although in cheap foreign ones he knows that it is sometimes not the case. It is of the greatest importance however, that the surface of the stage should be square with the tube in all directions.

The lower surface is generally blackened and the upper surface brightly polished and lacquered

or blackened. From the constant friction of moving the slide around, the lacquer or artificial blacking becomes worn and in time the stage assumes a shabby appearance. To overcome this, glass has been used with more or less success, but in recent years vulcanite has come into use and has proven very successful. The peculiar gritty feeling due to small particles of dust between the stage and slide is not so noticeable as on a metal surface and it is not affected by acids or alkalis and will therefore retain its neat appearance almost indefinitely.

Revolving Stage.—While in the largest number of instruments the stage is fixed and either round or square, there are others which are *revolving*, that is, may be revolved around the optical axis. These are absolutely necessary in the examination of crystals and rock sections and are then arranged with *graduations* around the edge—a series of divisions reading to degrees or fractions of them—by means of which the angles of the objects are measured. As a slight deviation of the center of revolving motion from exact coincidence with the optical axis will cause the object to swing out of the field, *centering screws* are provided by which this error can be quickly corrected. It sometimes occurs as the stage is revolved, that an object at the edge of the field, which is in focus, gradually becomes indistinct, showing poorest at

the half revolution and as the stage is brought around to the first point again comes into focus. This may be due to poor fitting of the parts, or to the fact that the stage is not square in all directions with the body; in either case a serious defect.

Glass Stage.—This with the slide carrier (Fig. 22) is a device for moving the object more

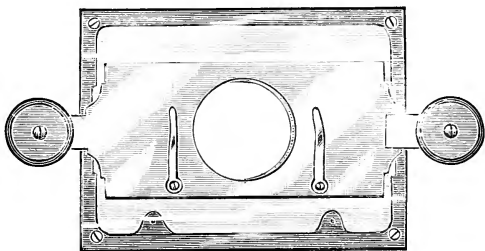


Fig. 22.

steadily and smoothly than can be done directly on the stage. It is made detachable and consists of a glass plate in a metal frame upon which rests, on four points, the slide carrier. At its ends the spring arms are passed around the glass plate and press against its lower surface, thus offering the minimum of friction, with sufficient resistance to make an easy movement.

Mechanical Stage.—This is a very important form in which the movements are mechanical in two directions and at right angles to one another,

being transmitted through the milled heads by a rack and pinion or screw movement. If pecuniary means will permit it, it is a most useful addition and by it work can be done systematically and with the assurance that every portion of the field

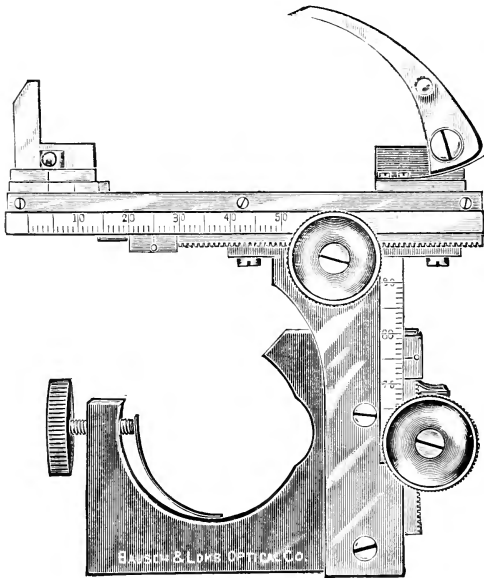


Fig. 23.

has been covered and withall with a degree of comfort which must be experienced to be appreciated. For instance, in a bacteriological or urinary specimen, where one is searching the field

for certain appearances, it is the only reliable means of covering each portion of it. This stage is especially valuable for blood counting and plankton work. While not many years ago it was spurned as a toy by many scientists, it is now generally accepted as an invaluable part of a microscope. In order to be so however, it must be of the most perfect workmanship, which is difficult to attain on account of the necessarily small parts of which it is composed and the hard usage which it must bear. The movement must be smooth and easy and on reversing the milled heads, must not show any lost motion or dead point.

There is a variety of forms, but two principal types, one of which is built onto the stage, usually the revolving one, the other attachable (Fig. 23) which may be removed when not wanted, leaving the ordinary stage intact. They are made with graduations, usually divisions in millimeters, by which one may read off the amount of space which is covered.

In using the mechanical stage it should first be determined how many spaces, or how much of one space is contained within the limits of the field ; then begin at one edge of the specimen and with lateral movement make the object pass across the field. Move the object forward with the other movement the amount of space which has been previously determined and by a return motion

of the lateral movement, bring it across again and thus through the entire specimen, or until the object is found or the specimen searched over.

Nose-Piece.—This being the lower end of the body, to which the objectives are attached, it is important in so far as it must be accurately made. As stated it has the society screw. Previous to 1857 each maker followed a standard of his own and this to a great extent is still the case on the Continent. The Royal Microscopical Society of London appreciating the inconvenience of this diversity, recommended a standard thread of 36 to the inch with an external diameter of 0.8 inch, which was finally adopted in England and this country. The Society supplied to the makers a so-called standard hob or tap with which to gauge the thread. Unhappily, however, these taps are not to a standard, as there is a variation in those which are sent out by the Society, so that while the public is under the impression that there are fixed dimensions there is a diversity in the products of different makers, so that it often happens that the objectives of one maker will not enter the stand of others. The writer in 1884 read a paper on this subject before the American Society of Microscopists and as a result a committee was appointed to bring about a better state of affairs. It failed, however, in obtaining the co-operation of the Royal Microscopical Society, the main reason

being the expense involved, so that we must contrive to suffer until some concerted action is taken by the manufacturers themselves, which we trust will not be too far distant.

Double Nose-Piece.—In changing one objective for another to obtain another power, time is consumed and it is often inconvenient; besides, there is danger of dropping the objective or disturbing or destroying the object. To avoid this the *double* (Fig. 24) the *triple* and *quadruple nose-pieces* are offered, to the first of which two, to the

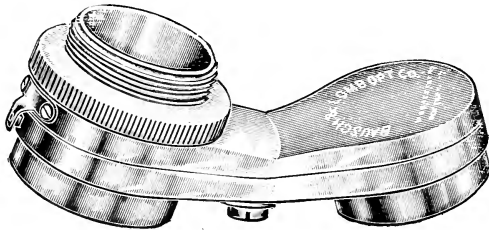


Fig. 24.

next three and to the quadruple four objectives may be attached in such a manner that, when fixed to the nose-piece of the tube, each objective may be in turn brought into use by swinging the nose-piece and in such a manner that each is centered and will be in focus, if not exactly, at any rate very closely. Of all the conveniences in accessories these are the most useful and in most common use, the writer knowing from his experience as

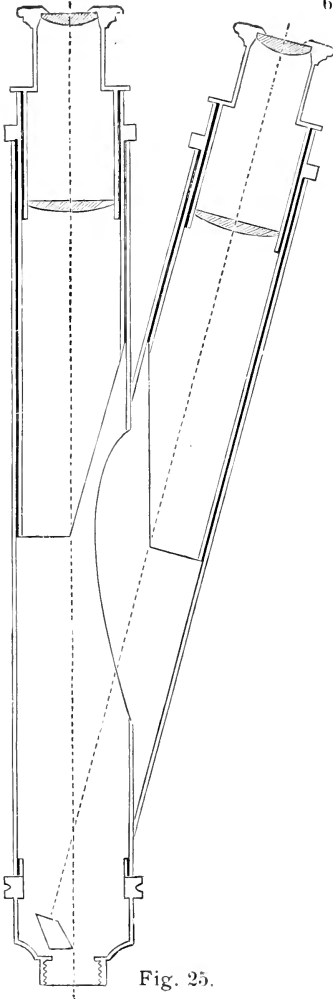


Fig. 25.

manufacturer that 80 per cent. of the instruments sold for personal use are supplied with a double nose-piece when two objectives are used, and the triple when three are taken.

Bodies or Tubes.

These are divided into two classes, *monocular*, having one body with which observation is made with one eye, which may contain one or two draw-tubes and *binocular* for observation with both eyes, in which the tubes are fixed together at the nose-piece and gradually separate until they reach the pupillary distance. The first would be a *monocular microscope*, the second a *binocular microscope*.

While the methods for transmitting the rays from the objective to the binocular tube vary, the construction in most common use is that introduced by Mr. Wenham.

By reference to Fig. 25 it will be seen that the rays from one-half the objective are transmitted uninterruptedly to the vertical tube, while the prism intercepts the rays from the other half and by reflection induces them to pass into the oblique tube. The result is an image in each eyepiece thus giving stereoscopic vision. This gives a perception of depth, a peculiar faculty of being able to look into an object and conveys to the mind the impression of roundness or separation of the object into different planes which it is impossible to obtain with monocular vision. Its use, however, is limited to the lower power objectives.

Coarse Adjustment.—In providing this adjustment, two methods are followed. The most simple form is that by a sliding tube in which the tube which carries the nose-piece at the lower end and draw-tube at the upper end, is moved up and down in an outer sheath, which is fastened to the arm. The milled ring, which is provided, is grasped by thumb and fore and middle fingers and pushed down or drawn up by a spiral motion. It is not to be commended except for economical reasons, as it lacks firmness, wears out quickly from the considerable friction, endangers the object and the

objective from the liability of sudden or jerking motions and does not well permit the application of the double nose-piece. While the clamping ring is provided in some instruments, which will fasten the tube in a fixed position, especially to permit the use of double nose-piece, this again has its disadvantages and is cumbersome. Therefore it is strongly recommended not to purchase an instrument of this kind if it can be avoided.

The *rack* and *pinion* adjustment is by far preferable in every respect and has stood the test of many years, although efforts have been made to introduce other methods, all of which, however, have become obsolete. To be satisfactory and lasting, it must be exceedingly well made and it is safe to advise that any instrument with this adjustment, which does not work well at the outset may be regarded as a poor one. In late years the pinion with spirally cut teeth and the rack with diagonal ones has come into common use and is better than the older form of straight cut teeth. In order to make the pinion operative, bearings are provided for it in the arm and its teeth engage in the rack, which is fastened to the slide, which also has its bearing in the recessed vertical length of the arm, as shown in Fig. 26.

This adjustment must meet the following conditions and if it does not, the instrument may be safely condemned as faulty.

It must work with the utmost smoothness and with not the least perceptible jar or grating.

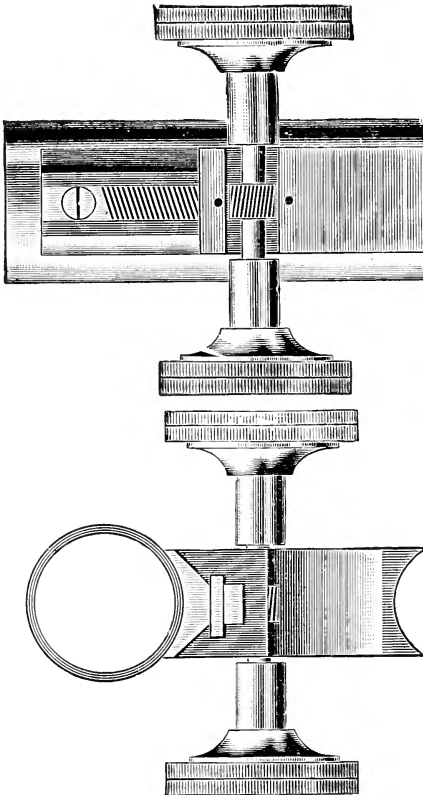


Fig. 26.

Cross Section of coarse Adjustment showing arrangements of bearing surfaces. Diagonal Rack and Pinion used in Coarse Adjustment and in substage construction.

It must be free from lost motion when working with the highest powers.

The slide must be so perfectly fitted, that it shall show no play when the tube is moderately forced from one side to the other.

Fine Adjustment.—While this is constructed in numerous ways it depends in all upon a screw for the propelling power. It is sometimes called the *slow motion*, as one revolution of the screw seldom gives more than $\frac{1}{50}$ inch motion. This screw is also called the *micrometer screw*. The brass button is called its *head* and when it is provided with equal divisions upon its upper surface, it is the *graduated head*. In this case a stationary *index* is fastened to the arm. A fine adjustment which will not fulfill the following conditions may be considered as faulty.

The screw must work freely and smoothly and without any side motion or play.

The adjustment should act promptly in the forward and backward motion without the least particle of hesitation or lost motion.

There should not be the slightest displacement of the object in the field when the screw is worked forward and backward.

While the fine adjustment, even more than the stage, will show displacement with moderate magnifying power by a slight exertion of force

against the tube, it should return to its position when released.

If a new instrument does not meet the conditions here set down for testing a fine or coarse adjustment, it may be put down as of faulty construction, no matter by whom made or how well made it may appear in other respects.

Draw-Tube.—While this part of the instrument may be an advantage when judiciously used, it may have a pernicious influence when abused. It will give both short and long tube standards and should be provided with a mark to indicate each length, or should have divisions by which the standard can be read off. It should not be overlooked, that when a double nose-piece is used the height of this is added to the optical length and suitable allowance should be made for it. In the cheaper instruments the draw-tube slides in the inner surface of the outer tube, but in the better instruments, a special sleeve is provided in which the draw-tube operates. As both of these have the defect incident to the sliding tube adjustment, a cloth lining is preferable as the movement is smooth, while firm and will remain so for an unlimited time. Care should be used in moving the draw-tube as a too sudden movement upward may draw the main tube with it and thus injure the rack and pinion, or downward, may force the objective onto the object or by the compression of

air in the tube, may force out the eyepiece. To slide it properly, hold the main tube with one hand and with the other grasp the draw-tube and move it up or down by spiral movement.

The draw-tube may be used to vary the magnifying power, but unless used judiciously may be the cause of more harm than good. While this feature will again be touched upon in another chapter, showing the optical effect, it will suffice at the present to state that it should be used only with objectives of low power or when with high powers, only under well defined conditions.

The draw-tube usually has at its lower end a diaphragm to prevent reflection from the inner surfaces of the tubes and this is sometimes also utilized as a society screw for attaching some very low power objectives or accessories.

Base.—The judicious form and weight of the base adds greatly to the stability of the microscope and it is a too common fault that in many instruments, even from reputable makers, this essential feature is sacrificed from wrong motives of economy, portability or compactness. While it can hardly be expected that when the arm is inclined to the horizontal position it shall be firm, as it is never used in this position except for photography and must then be clamped to the table, it is but reasonable to demand that when the instrument is upright or slightly inclined

under ordinary manipulative operations, it should not be required that the base be held with one hand, while the other makes the adjustments. We can imagine nothing more aggravating than a lack of stability. A considerable weight directly under the pillar is of little value, or a great expansion of the resting points with extreme thinness is little better. There should be a combination of both qualities and if suitable proportions are not maintained, an excess can hardly be called a fault, whereas too little would certainly be.

A favorite and good method to obtain increased weight within reasonable dimensions is, to load the base with lead and while some dealers, from interested motives, point this out as a defect, a purchaser need not hesitate on this account if the instrument is in other respects acceptable.

Joint for Inclination. — This should work smoothly but firmly and the arm should remain in any position in which it is placed. If it has a gritty sensation, the two parts are liable to “eat” and finally reach a point where they cannot be moved.

Besides the above qualification a good joint should work without the slightest back-lash, when the arm is worked quickly back and forth over a small space.

When the arm comes against the stop for upright position it should not lean forward.

Mirror and Mirror-Bar.—The proper illumination of an object is an important feature and although there are numerous accessories for properly accomplishing this, which will be spoken of later on, the mirrors alone are effective agents when properly constructed and applied, particularly when no high magnification is used. The plane mirror is generally used with very low powers and reflects light in about its original intensity. The concave mirror, however, is intended to concentrate the light so that all the rays which strike its surface are reflected and come together at some point above and the rays from the surface being contained within a comparatively small space, cause an increased intensity. This point is called the *focal point* and is usually arranged to coincide with the opening of the stage, when parallel rays such as from the sky are used. When the source of light comes considerably nearer to the mirror, as for instance from a lamp and the rays are diverging, the focal distance becomes considerably longer. Some of the intensity is lost in consequence, as well as the degree of convergence. For this reason some mirror-bars are so arranged that the distance of the mirror from the stage may be varied to accommodate the variation in the location of the source of light. While this is of considerable aid, there is in some instruments not sufficient room for a complete accommodation,

with the result that, under certain conditions, the utmost effectiveness of the microscope is not obtained.

Diaphragm.—This is provided for regulating the amount of light. While the mirror should work to its utmost capacity, it very often occurs that for certain investigations a profuseness of light is more harmful than otherwise. When too much light exists, objects are said to be drowned in it and thus often makes it impossible to determine structure. An intelligent use of the diaphragm is of great service.

Besides the *revolving diaphragm* there are other forms which may be said to be better—for instance, the so-called *cap diaphragms*, which require a separate piece for each aperature and which are held by a special substage receiver; then the *dome diaphragm*, which is a new application of the ordinary revolving diaphragm. It consists of a substage fitting having a dome, to which is fitted a curved revolving diaphragm.

The ideal regulator of light is the *iris diaphragm* consisting of a series of overlapping blades placed around a central opening, size of which may be varied by means of a lever or milled edge. In the ordinary revolving form the aperture is of necessity at some distance from the object and does not fully control the light on account of the stray rays, which the other three forms accomplish. The

distance of diaphragm from the object is one of considerable importance. The best position is just below the surface of the stage, but as this is not always possible, it should be as near as conditions will permit. The nearer it is, the more intensity will be maintained with suitable moderation by the limit of opening. Very recently it has been possible, to so construct the iris diaphragm, that it passes up through the opening of the stage and is flush with its upper surface.

OBJECTIVES AND EYEPIECES.

In taking up this subject we would say at the outset that it is fraught with difficulties, as almost all of the features are based on scientific facts which can be explained by mathematical formulæ, but as it is our purpose to give intelligible explanations to those who may not be conversant with algebraic expressions, many of the statements and descriptions will appear rather dogmatic. We can but advise those who wish to study the subject further, to consult such books as contain more explicit information.

As has already been said, the compound microscope is composed of two lenses, the upper one of which magnifies the image which is formed by the lower one. We know that the purpose is to give a greater magnification than can be obtained with the simple microscope. The defects of chromatic and spherical aberration will however become more pronounced than they would in the simple form, to such an extent as to nullify the benefit which might be derived from the magnifying power only. In fact, magnifying power in itself is of very little value without the attributes obtained from the

chromatic and spherical corrections and their qualities which will appear as we proceed. The advent of achromatic lenses was the first decisive step in advance and has been the foundation of all later improvements and the high standard of the best productions of the present day.

It is a matter of pride to Americans to note that two of our countrymen, now deceased, were influential in furthering the progress to a considerable extent and that their memories should always be honored by the microscopical world. They should be remembered with feelings of gratitude, particularly as the compensation for their efforts was extremely limited. The pioneer in microscopical optics in this country was C. E. Spencer, who was followed by R. B. Tolles, and while both men did a great amount of original advance work, it is the latter particularly who, by his wonderful achievements, created a great discussion in European circles, by enabling results to be obtained which for a long time it was claimed could not be accomplished.

Of inestimable value to the scientific world have been the labors of that most capable and genial gentleman, Prof. E. Abbe of Jena, to whom, while best known to the general microscopist for some of his more insignificant improvements, such as the Abbe condenser and Abbe camera lucida, we are much more indebted for his profound disclosure of

the principles of microscopical optics, as well as to the combined efforts of himself and Dr. Schott for their labors in the art of glass making and to the large variety of glass which they have placed at the disposal of opticians, who by this means have been able to accomplish much higher results than would otherwise have been the case. In this connection it is opportune to state that the production of this glass, generally termed *Jena glass*, has been taken advantage of by unscrupulous parties in creating the impression that the bare fact of using this glass gives in itself much better results. Such is not at all the case. The merit of the production consists mainly of the large variety of glass with different ratios of refractive index and dispersive power.

As defects of the objective have thus far been specially mentioned, it will at this point be well to state that the single lens of the eyepiece has also been found unsuited and while an achromatic lens is unnecessary, a supplementary lens below and near the upper lens has been beneficial in so affecting the image, by collecting the rays, that it can be viewed at one glance and without spherical and chromatic aberration which the single lens would show. It is for this reason called the *field lens* or *collective* and the upper lens the *eye lens*. Both of these lenses are mounted so as to form one part of the microscope with fixed relations and is

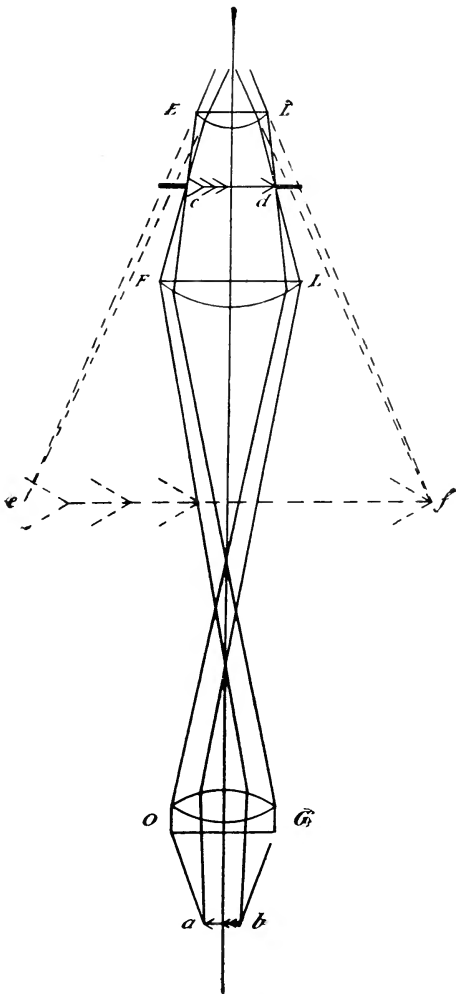


Fig. 28.

then called *eyepiece* or *ocular*. In the diagram (Fig. 28) the course of rays from the object through the objective and eyepiece is shown. OG represents the objective, FL the field lens and EL the eye lens of the eyepiece. As the rays from the object pass through the objective they are seen to cross before reaching the field lens, are converged as they pass through and further converged by the eye lens EL . At the point cd they form a real image of the object, which can readily be seen by placing a ground glass or piece of oiled paper at this point. It is an interesting experiment and one which we recommend trying. The eye lens enlarges this image and forms a greatly magnified virtual image at cf . From this diagram several changes with consequent results can be noted.

If the objective is of short focal length, a larger real image is formed at cd .

If the distance between objective and eyepiece is increased, an enlarged real image at cd results.

If the eyepiece is of higher power, an enlarged virtual image is formed at cf .

In the same manner a reduced magnifying power may be obtained by reversing these conditions.

As has already been stated a 1 inch lens with a distance of 10 inches between it and the image gives a power of 10 diameters, and the eyepiece

multiplies the virtual image by the extent of its power. From this it can be easily computed that with a 1 inch objective used with a tube length of 10 inches and a 1 inch eyepiece, a magnifying power of $10 \times 10 = 100$ will be obtained; or the same combination with a tube length of 5 inches will give one-half this power or 50.

Objectives.—These are divided into two classes, *dry* and *immersion*. In the dry there is no intervening medium other than air between the bottom lens of the objective and the top of the cover glass. In the immersion a liquid fills up this space. From this fact it is easily seen that liquid can only be used in objectives which are quite close to the cover and therefore short focus or high power and so on the other hand can objectives of long focus or low power not be immersion. We will see later that the purpose of the immersion is to obtain higher optical results, is in fact a necessary condition, and an objective which is constructed as a dry one cannot be used as an immersion, and vice versa. Although there have been objectives constructed which can be used both as immersion and dry, they have gone into disuse as they must suffer when used in one or the other direction or in both.

Water was for many years used as immersion fluid, but cedar oil was discovered and as by means of this better results are obtained, it has largely taken its place. Its optical properties are almost

identical in refraction and dispersion with those of crown glass and it is for this reason often termed *homogeneous immersion fluid*, but for brevity objectives which are constructed to be used with it are called *oil immersion*.

Tube Length.—Objectives are constructed and their aberrations corrected for the length of tube with which they will be used and as has been shown in a previous chapter there are now two generally accepted standards. They are corrected for either the long or short tube and specially marked by progressive firms and it is hoped that this will become a universal custom in time. When an objective is not marked the purchaser should require to know the tube length with which it is to be used.

Nomenclature, or Rating of Objectives.—While for many years objectives, or rather the mountings, were marked arbitrarily by the makers and differently by each maker, it is now customary to mark the objectives so that the figures shall indicate the true optical value; on the continent of Europe in millimeters and in England and this country in inches. The objectives are rated according to a single lens, which the combined value or equivalent focus of the lenses contained in the objective, shall equal. This is also true of eye-pieces. So, if two combinations equal in magnifying power a single lens of 1 inch focus they are

marked 1 inch, or if a collection of four lenses will be equal to a lens of $\frac{1}{12}$ inch focus it is so marked.

The powers increase in proportion to the decrease in focal length, so that a $\frac{1}{12}$ for instance will give a real image twelve times larger than 1 inch or a real magnification of 120 times.

Powers.—According to their powers, objectives are called *low*, *medium* or *high* and are classified by Carpenter as follows :

Low powers, 3 inch, 2 inch, $1\frac{1}{2}$ inch, 1 inch, $\frac{2}{3}$ inch, $\frac{3}{4}$ inch.

Medium powers, $\frac{1}{2}$ inch, $\frac{4}{10}$ inch, $\frac{1}{4}$ inch, $\frac{1}{5}$ inch.

High powers, $\frac{1}{6}$ inch, $\frac{1}{8}$ inch, $\frac{1}{10}$ inch, $\frac{1}{12}$ inch, $\frac{1}{16}$ inch, $\frac{1}{20}$ inch.

It might be stated that such powers as $\frac{1}{20}$ and $\frac{1}{25}$ inch are very rarely constructed at the present time and that the $\frac{1}{16}$ inch may be considered the maximum, although seldom used. The $\frac{1}{12}$ inch is the highest which is ordinarily used and will give all the optical advantages, while the higher powers involve so many mechanical difficulties as to increase the cost of production very considerably and as a rule detract from the optical qualities. A modern objective of the highest capacity may be considered a work of art and there are few productions of the human hand which exact so much untiring application, ingenuity and skill.

Systems.—An objective is said to consist of systems which may vary in number from one to

four and five. Two are generally used in low powers, three and sometimes four in the medium powers and four to five in the high powers. They are the individual portions consisting of one, two or three lenses, which, when composed of more than one lens, are cemented together and make up the objective. An achromatic single system objective may consist of two or three lenses and a three

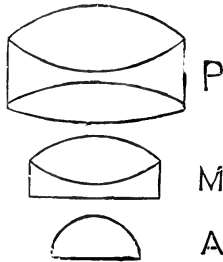


Fig. 29.

or four system may consist of as many as eight or ten lenses. The systems are called in their order : *anterior* or *front*, *middle* and *posterior*. When one consists of two lenses it is called a *doublet*, when of three lenses a *triplet*. Thus in Fig. 29. A is the anterior, M the middle and P the posterior system ; thus also A is a single system, M a double and P a triple one.

The various features which must be considered as determining the quality of an objective are :

Angular Aperture,
 Achromatism,
 Resolving Power,
 Flatness of Field,
 Penetration,
 Working Distance,
 Magnifying Power.

Although these attributes may be considered separately, some of them go hand in hand. The presence or extent of one necessarily involves or precludes another.

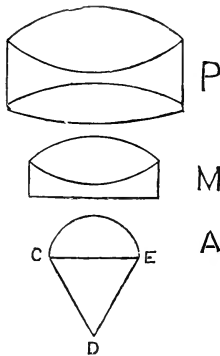


Fig. 30,

Angular Aperture.—The angle which the most extreme rays, which are transmitted through the objective, make at the point of focus, is called its *angular aperture*, or in short its *angle*, the

extent of which is expressed in degrees and of all the qualities in an ideal objective, this is the most important. Thus in Fig. 30. D is considered the point of focus, and CDE the angular aperture. The above definition has its limitations however. While in objectives of proper construction it holds true, there are many in which it is not the case. For instance, an objective may be so constructed that it may transmit a considerable number of rays in excess of those which combine to form an image and it is evident that as they do not aid in forming an image, they serve no purpose and therefore have no value in consideration of angular aperture.

As there are many objectives of the same power but of different angular aperture, there are again others of varying power but of the same angle.

Light is radiated by an object equally in all directions and the more of the rays which can be collected to form an image, the more distinct will it become and to a greater extent can we see detail. If in two objectives one receives on its front surface and transmits a larger number of rays than another of equal power, we have a case where power would indicate that we should see equally well, but we will find that there is a difference, due to the amount of angular aperture, in favor of the wider angle. Or, in the case of two objectives in which one has one-half the power of the

other, but which takes in the same amount of rays, it would appear that if the power alone were to indicate the visibility of the object, the higher one should show more detail, whereas they in reality show equally well.

Aperture, without the defining word angular, indicates a very important feature in an objective and designates the beam or pencil of light which passes out through the rear lens of an objective, or in other words is the effective diameter of the rear lens. Many objectives are made in which the rear lens is larger in diameter than the beam of rays which, coming from an object, can be transmitted through it and while not particularly detrimental, has no value except perhaps to lead to a wrong conclusion in reference to angular aperture when this is measured, as the excess of image forming rays, called stray rays, may indicate a greater angle than the objective really possesses.

Cover Glass.—At this point we must introduce another feature which we have not yet considered but which is important in understanding this, the most important property of an objective. As we have shown, in describing the principle of refraction, rays which fall from air upon a surface of glass, are bent out of their course or refracted. The cover glass which is used on the object during examination, or for its preservation, although

extremely thin, has a marked influence on the optical performance of an objective and this influence increases as the magnifying power of the objective increases.

For the purpose of illustration, we will imagine a cover glass of considerable thickness (Fig. 31).

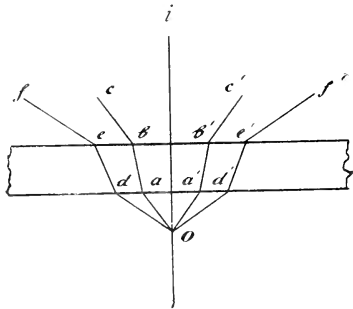


Fig. 31.

o represents the source of light, or in this case the object. While the rays from it are emitted in all directions, we need only consider those coming from the upper half, and for simplicity we will select from them two pairs. As oa and oa' strike the lower surface of the cover glass they are refracted toward the axis oi and on their emergence from the upper surface of the cover glass are again refracted away from the axis in the direction of bc and $b'c'$, which is the original direction of the rays. The same action will take place with the extreme rays od and od' , which will emerge as shown at ef and $e'f'$.

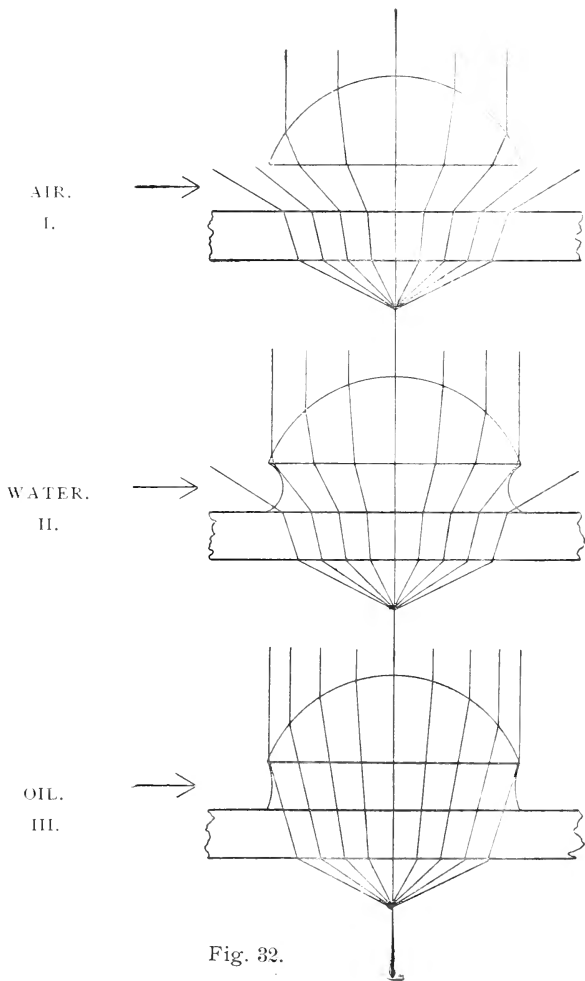


Fig. 32.

If we now bring the front lens of an objective close to the cover glass we can study its influence and for this purpose will use the simple but intelligible illustrations of Prof. Gage to show the same phenomena.

I shows a dry objective in which the intervening medium between the top of cover glass and front of objective is air.

II is a water immersion objective in which this space is filled with water.

III is an oil or homogeneous immersion objective in which the space is filled with oil of cedar.

From what we have said in regard to the laws of refraction we know that as the medium becomes more dense the refraction becomes greater and for the same reason it is clear that as the difference in density between the two media becomes less the refraction is proportionately less. As water has a greater density than air, but less than glass, refraction between these two media is less than between air and glass. As the homogeneous fluid has the same density as glass, or as we have stated, is virtually fluid glass, no refraction between these two media takes place.

If we now refer to the diagrams and will bear in mind the course of rays from an object through a cover glass, we can follow out their action. In the case of a dry objective, we see that some of

the emerging rays coming from the top of the cover glass are so refracted that they fail to touch the front lens of the objective and are therefore lost. In the water immersion, however, the rays being less bent, more of them reach the front lens and are passed through the objective. With the oil immersion the only refraction is that which takes place at the lower surface of the cover and from that point the rays are carried uninterruptedly through the cover, fluid and front lens as far as the convex surface, where they are refracted and carried through the objective. From a view of the diagram it might appear that if the lens were enlarged in diameter more of the extreme rays which are lost might be utilized, but as the radius of the front lens would have to be increased we know that its focus and magnifying power would be decreased. This, however, would be detrimental, inasmuch as, according to the law which has been determined and promulgated by Prof. Abbe, the capacity of an objective is determined by the ratio between its focal length and the diameter of the emergent pencil at the point of its emergence, from the back of the objective, or in other words, it is "the sine of half the angle of aperture multiplied by the refractive index of the medium between front of objective and cover." This has been called by Prof. Abbe the *Numerical Aperture*, and is now commonly used to designate the effi-

ency of an objective. When applied to objective mountings or used in tables it is abbreviated to N. A. The formula by which computations are made is $N.A. = (n \sin u)$ in which N.A. is numerical aperture, n the index of refraction of front medium and u one-half the angle of aperture. Since the media are air, water or oil it is necessary to know the refractive index of each, which is 1.0 for air, 1.33 for water, 1.5 for oil and for the ordinary crown glass the index is the same as for oil.

To illustrate this formula better we will take an example. A dry objective $\frac{1}{4}$ inch focus has an angular aperture of 100 degrees. By reference to a logarithmic table we find that one-half of the sine of 100 degrees is 0.766 and we know that n , the medium in front of the objective being air, has a value equal to 1.0. The formula then is, in figures, $N. A. = n \sin u$ or $1.0 \times 0.766 = 0.766$.

To make this computation we have all figures except that of angular aperture at hand. This must be determined and the method for accomplishing it will be given in a succeeding chapter.

As a rule the designation as to power and numerical aperture engraved on the mounting of an objective from a responsible firm can be relied upon as being quite close, the variations seldom being greater than is incident to accurate human handi-work and such variations as do occur, have little influence on the optical capacity. It is mani-

festly difficult for opticians who produce large quantities of objectives to measure and suitably mark each individual product, particularly when the differences are at best slight.

It is easy to learn the numerical aperture of an objective after the angular aperture has been determined, as the various values for different angles have been computed and are issued in tables. Such tables can be found in the pages of the *Journal of the Royal Microscopical Society* and in the catalogue of the Bausch & Lomb Optical Co.

Before the time when the numerical aperture was so thoroughly elucidated, it was known that an increase of angular aperture gave higher results, but just why this was so was not appreciated. So also was it difficult to understand that a dry objective of 180 degrees, a water immersion of 96 degrees and an oil immersion of 82 degrees had the same effective aperture.

How to Measure Angular Aperture.—In instruments of the American type in which the axis of the mirror-bar is in the plane of the stage and in which the circular part is graduated, the matter of measuring angular aperture is quite simple. It was first recommended by Mr. Tolles and has been carefully worked out by Dr. George E. Blackham of Dunkirk, N. Y. After the object has been focused upon, incline the body of the microscope to a horizontal position, remove the

mirror with its bow from the socket and place therein, or by rubber band attach to the end of the bar, a toy candle at such height that the flame will be in the optical axis. The mirror-bar is now swung to the right and left to such an extent that one-half the field shall be illuminated, provided the object shall still be well defined, or to such a point where the definition of the object shall appear impaired without regard to the illumination of the field. These limits will mark the efficient beam of light which passes through the rear system of the objective. As we have stated before, in some objectives the rear system is larger in diameter than the effective beam of light transmitted from the object, which will permit stray rays to reach the eyepiece, but which have no value in forming the image and therefore are not to be considered. In instruments which have no graduated mirror-bar the matter becomes more difficult and less accurate. The following is called Lister's method.

After the objective has been focused, place the body of the microscope in a horizontal position and in front and some distance from it, a candle or lamp, if the latter, with the narrow edge of the wick toward the instrument, but level with the tube. Move the lamp on each side of the axis to a point as described in the foregoing method. Indicate the position of the center of the lamp at each

extreme on the table and beneath the instrument also a point situated vertically under the object. Connect these points and with a protractor measure the angle.

A very accurate method and one which can be carried out on all instruments is that suggested by Prof. Abbe and for which the firm of Carl Zeiss supply an apparatus which is called the *apertometer*, which consists of a semi-circular disk of glass having at its straight edge a beveled surface which reflects the light through a perforated disk into the objective. Two strips of brass which act as stops are placed on the arc and thus indicate the limit of aperture, which can be read off on the scale as well as the corresponding numerical aperture.

In the foregoing we have gone to some length in stating the importance of angular and numerical aperture and laid stress upon the influence which these factors have upon the efficiency of the objective. We shall now endeavor to explain what these attributes are.

Resolving Power.—First of all qualities in an objective is the resolving power, which is the ability to show intricate structure and minute detail, it being of course understood that the objective is properly constructed so that defects shall not detract from this quality. It is of course clear, that no matter how great the numerical

aperture may be, its effectiveness may be injured by the presence of chromatic or spherical aberrations or defective work and it is a matter of no uncommon occurrence that in objectives of the same power and aperture there is a noticeable difference in resolving power, or in objectives of different numerical aperture, the one of less aperture will have a greater resolving power than the other. If we could accept the statements of makers as true ones, a portion of this work and a vast amount of literature would be unnecessary, but the writer has occasion to know that this is not the case and that there is a constantly increasing danger and tendency to allow small defects to pass, in spite of the fact that the general efficiency of the microscope has increased in late years.

To resolve a structure is to make it visible and the resolving power is in direct ratio to the numerical aperture and can be mathematically calculated. It can be studied from the aperture tables already mentioned. It will be seen that an objective with a numerical aperture of 0.50 will show one-half as many lines as one with a numerical aperture of 1.0. This, it will be seen, refers only to the resolving power of an objective and makes absolutely no reference to its magnifying power. Now, as we know that the purpose of the compound microscope is to give magnifying power

and that there is certain structure which is not visible, how can we reconcile this with the fact that numerical aperture only does it? Why, if this is true, should we use a higher power objective with a certain aperture in preference to a lower one of the same aperture?

The normal eye can see about 200 lines to the inch, or structures which are $\frac{1}{200}$ th of an inch apart and it is therefore evident that any structures under the microscope must be separated at least to this extent in the virtual image, in order that they may become visible. While we have shown that magnifying power may be increased

- by increasing power of objective,
- by increasing power of eyepiece,
- by increasing tube length,

we are limited in the last direction mainly by such length of tube as has been found convenient in the construction of the stand; in the case of eyepiece on the one hand by convenience in use and on the other by the fact that too great an increase causes an indistinctness, so that although variation in eyepiece within narrow limits is admissible, we are compelled to select an objective which, with a medium power eyepiece, will give such separation to structure and fine detail, that it will be visible to the eye without any undue strain.

Chromatic Aberration.—Up to this point we have spoken of the correction of chromatic aberration by suitable use of flint glass lenses and for the purpose of not making the subject too complex, have purposely refrained from stating that *entire freedom from color is impossible* in the ordinary combinations of flint and crown glass. The correction is for only two colors of the spectrum, the red and violet, leaving as a residue the other colors which appear as green and purple. These are called the *secondary spectrum* and on bright objects can easily be discerned. For all ordinary purposes it is not prejudicial to the performance of an objective. It becomes more pronounced in dry objectives of large aperture and when high power eyepieces are used. In properly corrected low and medium powers of medium aperture and high power immersion objectives it is most noticeable, but certainly not to any extent to be objectionable, except when oblique light is used.

Great care should be used in judging an objective by its chromatic correction and one should not be led to false conclusions by the amount of color which an objective shows. It has been a common experience with the writer to have had objectives complained of which were properly corrected and which were excellent in every respect except that they showed the *secondary colors*, so that it may safely be said, remembering that wide aperture

involves a greater amount of color, that an objective showing the proper colors of green and violet and having proper resolving power may safely be accepted, or in the choice of objectives between one showing no color but having no resolving power, the other having color and resolving power, the latter is certainly the preferable one.

When colors of green and violet are not sufficiently pronounced on an object when the mirror is in central position, they will become more apparent when the mirror is swung to an oblique position, using for an object a coarse diatom mounted dry. If the mirror is swung to the left the object will be fringed on the right side by yellow-green and on the left by violet color, or if the mirror is swung to the right the conditions are reversed.

When the objective is not properly corrected it is said to be chromatically *under-corrected* or *over-corrected*.

Under-correction may be judged by central light, after the object has been focused, by slightly increasing the distance between objective and object, when the latter will show a blue color and by decreasing the distance an orange color. Or, will show over-correction, if on increasing the distance the orange will appear and by decreasing the distance the blue. The appearance will also become more pronounced by using oblique light.

Under-correction will show when the mirror is swung to the left and when the object is fringed with orange on the left and the blue-violet on the right. Or, over-corrected when the colors are reversed.

Another method is that recommended by Naegeli & Schwendener in their work on the microscope. The right half at the front or back of the objective is covered by black paper or tin-foil so that only the other or left half remains optically effective; take for an object a line or dot of light which can be easily produced by blackening one surface of a slide with chimney soot and drawing a line across it with a point.

Under-correction shows when the image has on the right side a violet or blue border and on the left a red or orange colored border.

Over-correction shows on the other hand when the left side appears violet and the right red or orange.

Spherical Aberration and Cover Glass.—As we have stated there is a residue of chromatic and spherical aberration in all ordinary achromatic lenses and objectives. The use of cover glass influences the spherical correction and while not appreciable to any extent in low powers, it is very sensible in the medium and high powers. If we refer to Fig. 33 it will be remembered that the rays from the object do not uninterruptedly reach

the surface of the objective front but are changed in their course. If we make use of the same illustration and extend the refracted rays downward as shown by the dotted lines $e c$, $f d$, $f' d$, $e' c$ until they meet at the axis, these points will be the apparant location of the object and will appear to meet in the planes $c a$ and $d b$ instead of at o . To neutralize this condition the objective will require to be spherically

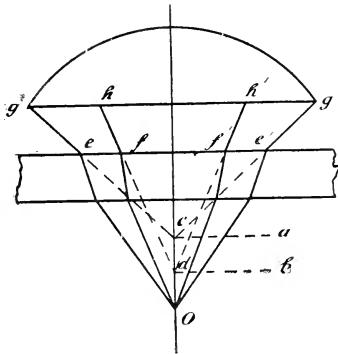


Fig. 33.

under-corrected, by which is understood that the marginal rays will emanate from a point near the objective and the central rays at a greater distance from the objective, or as is shown in the diagram, appear to come from exactly the same points, which are the apparant positions of the object or $g c$, $h d$, $h' d$ and $g' c$.

If a thicker cover is used the objective will require to be more under-corrected, or if a thinner

one, less under correction is needed, so that if an objective is corrected for a definite thickness of cover, its correction will be disturbed if greater or less thickness be used, and since resolving power depends, as first condition, upon the proper correction of the two aberrations, it will be *entirely lost* if the variation from the normal thickness is considerable.

It will be well to bear this phase in mind, as it has a great influence on the efficiency of the objective and will appear repeatedly in the instructions to be given later on.

There are several methods by which corrections may be made. In objectives with fixed mountings, such as are ordinarily used, in which the lenses have fixed relations, which correct the spherical aberration for a definite thickness of cover glass, no change can be made in them and therefore cover glass should be used, which is very close in thickness to that which was used as a standard. Or, correction can be made within narrow limits by varying the tube length.

For a *thick cover* the tube must be *contracted*; for a *thin cover* the tube must be *extended*.

Objectives are also constructed in which the distances between lenses may be varied and then are called *adjustable*.

For thick covers the lenses are *brought together*, or the adjustment is *closed*.

For *thin covers* the lenses are *separated*, or the adjustment is *open*.

It is a misfortune that cover glasses are not made of the same thickness, but the difficulties in producing them of equal thickness are almost unsurmountable, and it is also to be deplored that opticians have not agreed upon a definite thickness to which they correct the objectives; but since this is not the case, all aid should be given to the microscopist to obtain the best possible results. The list herewith given, which has been prepared by Prof. Gage will show the variations in thickness used by different opticians as standard.

$\frac{2.5}{100}$ mm.	}	J. Green,
		J. Grunow,
		Powell & Lealand,
		Spencer Lens Co.,
		W. Wales.
$\frac{2.0}{100}$ mm.		Watson & Sons,
$\frac{1.8}{100}$ mm.		Klonne & Muller,
$\frac{1.7}{100}$ mm.	}	E. Leitz,
		R. Winkel,
$\frac{1.6}{100}$ - $\frac{2.5}{100}$ mm.		Ross & Co.,
$\frac{1.6}{100}$ mm.		Bausch & Lomb Optical Co.,
$\frac{1.5}{100}$ - $\frac{2.0}{100}$ mm.		C. Zeiss,
$\frac{1.5}{100}$ - $\frac{1.8}{100}$ mm.		C. Reichert,
	}	Gundlach Optical Co.,
$\frac{1.5}{100}$ mm.		W. & H. Seibert,
		R. & J. Beck,
$\frac{1.2}{100}$ - $\frac{1.7}{100}$ mm.		J. Zentmayer,
$\frac{1.0}{100}$ - $\frac{1.5}{100}$ mm.	}	Nachet et Fils,
		Bezu, Hausser et Cie,
$\frac{1.0}{100}$ mm.		Swift & Son.

The writer may be pardoned for introducing at this point extracts from a paper which he read a number of years ago.

“The cover glass may truly be called a necessary evil ; for, while absolutely required in microscope investigations, there is no adjunct to the microscope that has been and is productive of so much evil, and has retarded the utilization of benefits made possible by the advance in the construction of objectives, so much as it.

“It must be remembered that the majority of objectives will always be dry, and especially so when improvements, which we hope are still to be made, are accomplished. It is an unfortunate circumstance that with this class of objectives the influence of variation in thickness of cover glasses is most apparent ; but since it is so, we should, if possible, provide an agency which, eliminating the personal factor of efficiency, will give, under all conditions, results closely equal to those under which the objectives were originally corrected.

“It is surprising to see how little attention is paid to this subject in the large majority of the standard works on the microscope. Almost all books give carefully prepared illustrations and descriptions showing the effect on the course of light of the interposition of the cover glass, and after giving conclusive evidence of its disturbing

influence, still, in a general way, say it is of little moment. * * *

“The system which I have devised to aid in overcoming these difficulties depends in the first instance upon a micrometer for measuring the thickness of cover glass. (Fig. 34.)

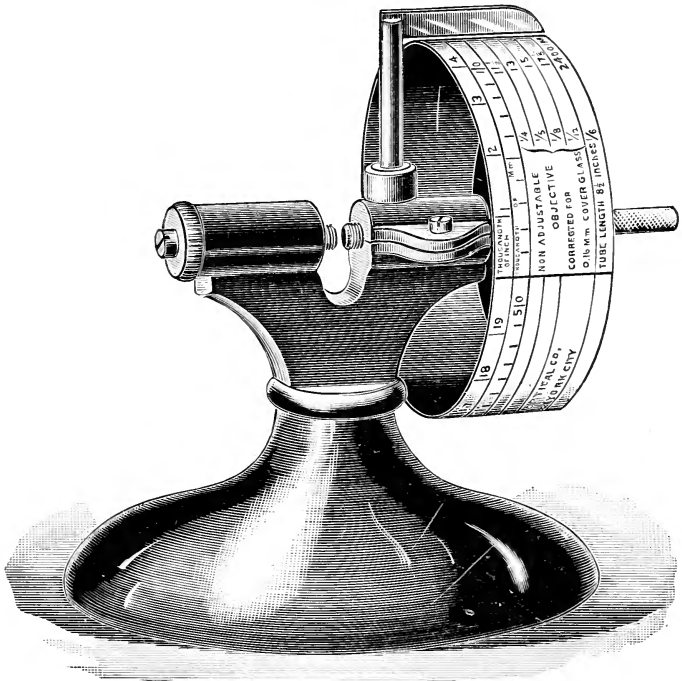


Fig. 34.

“In objectives provided with cover correction the graduation is so arranged as to read to $\frac{1}{100}$ mm. No matter what the power of objective, the number gives proper correction for a thickness corresponding to it. Thus, with a cover glass of 0.20 mm. the collar of such an objective need merely to be set at 20 to give the proper correction and, consequently, the best results.

All the other scales give the correct tube length in inches and millimeters for covers corresponding to them, and in this manner offer a ready and definite means of correction. The tube lengths required for the thinnest and thickest covers are so extreme that probably no convenient means for obtaining them can be practically arranged, but they can be so, approximately if not entirely. At any rate, the micrometer will detect the requirements before using the covers, and those deviating considerably from the normal can be used on objects for use with low powers only, in which case the effect will not be very appreciable.

“In this system I do not overlook the fact that variation in tube length involves a variation in magnifying power; but, except in cases where micrometers are used, I consider this of secondary importance, as it always is in comparison to results obtained in resolving power.

“ This system involves four conditions :

First—That all cover glasses be measured before using them, and that the thickness be noted on the preparation.

Second—That for convenience all draw-tubes be marked in inches or millimeters, or both.

Third—That adjustable objectives be corrected according to this scale.

Fourth—That the same tube length and cover glass thickness be used in all original corrections of objectives.”

Penetration.—Penetrating power is the quality which enables us to look into an object—to observe different planes at one time. In the mind of the writer, it is of no special importance, or at any rate not as much as is claimed for it, and if desired is easily attained. It depends upon magnifying power and angular aperture, and decreases with the increase of either of these. Objectives are generally not constructed with any reference to it ; it is a natural consequence of certain conditions.

Penetration and resolving power are antagonistic, or at any rate in an inverse ratio, and can only be combined to a certain extent. In two objectives of the same power and aperture, one cannot have penetration as a special feature and the other resolving power ; they will be almost similar in these qualities provided that they are

similarly corrected. However, if they are not similar in their angular aperture the one of small aperture will have more penetration than the other. In objectives of the same angle but different power, the one of low power will have in itself more penetration ; it will be similar in its action to the eye, which, when an object is close to it, can distinguish but one portion of it distinctly, while, as its distance to the eye is increased, can distinguish various parts of it lying at different distances, and will finally see other objects outside of it. By looking at an object at 5 feet distance, only this can be seen plainly ; but, at 10 feet, others quite a distance in front or back of it can be seen as well.

Low power objectives have a proportionately greater penetrating power than medium or high powers. In an object of considerable thickness, different planes can be observed at one time without focusing on them and thus obtain an appreciation of form which is impossible in the higher powers, as in these the adjustment for different depths is required.

Furthermore the accommodation of the eye is also a factor as it varies with different persons and thus, to a certain extent, is a matter of individuality.

Flatness of Field.—The field in a microscope is that portion which is observed in the eyepiece, and its flatness may be observed when focused on

a flat object—preferably a micrometer. It is said to be flat when all portions of the object are seen over the entire field at once without further focusing. When not flat, it will be found that as the image approaches the edge of the field it becomes more and more indistinct, and that the objective must be correspondingly adjusted; in many cases it remains indistinct or blurred, and this may be considered the most serious fault. In the case of looking at straight parallel lines, such as in a micrometer, they will appear to become more curved as they near the edge, as shown in Fig. 35.

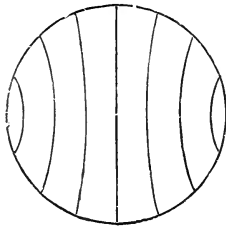


Fig. 35.

Flatness of field mainly depends upon the correction of the spherical aberrations and as under the best conditions the latter cannot be entirely eliminated, it is impossible to attain absolute flatness. It may also, however, be due to a faulty eyepiece; in this case it can be determined, by observing whether it shows equally in different objectives. With beginners, especially, it is usually most complained of, owing probably to the fact

that it is most easily noticeable. It is a desirable equality and indicates to a considerable degree the quality of objectives. It is *impossible to obtain absolute flatness of field*, in objectives of sufficient angular aperture to meet the requirements of the present day.

Working Distance.—This term, strictly considered, is an invariable quality of the objective and is the distance between the front lens in the objective and the object, when the objective is in focus and is corrected for that object. All objectives require a certain amount of projecting metal to protect the front lens and this, with a certain thickness of cover glass, lessens the working distance. In objectives with fixed mountings this may be, and with thick cover glasses is, considerable. It is comparatively unimportant however, for the working microscopist to know the working distance *per se* of his objectives, but of considerable moment to know what is the actual space between the objective and cover glass.

In objectives of low and medium power, it is of little consideration; but where it must be expressed in $\frac{1}{100}$ or $\frac{1}{1000}$ inch, it becomes a matter of importance.

Working distance is spoken of as being *long* or *short* and varies with the power and angular aperture. Generally the working distance decreases with the increase in numerical aperture and be-

comes greater as the aperture becomes smaller. It was for a long time considered that these two qualities varied according to a fixed rule, but this at the present time is not considered to be the case. While in objectives of the same aperture it may vary considerably, it may in others of different aperture be so that the higher one may have the greater working distance. The skill of the optician must to a considerable degree determine the amount of it.

It will be seen from the above that working distance stands in no direct relation to the focal distance of the objective and it may be added, that it is *never as great as the focal distance of a single lens* of the same magnifying power.

As may be imagined, there is a variety of opinions as to what constitutes long or short working distance in a certain objective. No definite rule can be laid down for this, as it is conditioned by the skill and requirements of the manipulator. It has several times occurred in the experience of the writer, that objectives were complained of as having no working distance (that the objective could not be focused) when on investigation it was found that window glass or a slide had been used for a cover.

To Measure Working Distance.—The actual or available working distance is of little moment except in objectives of medium or high power and

then for two reasons. As we will show later how to focus an objective, it may be of value to the student to take no risk in endangering the object and in the case of oil immersion objectives, where the working distance is exceedingly small, to know what thickness of cover glass may be used. In instruments having a graduated micrometer screw, bring the front of the objective just in contact with the top of the cover glass. This can best be done with no danger to either objective or cover glass, by grasping the instrument beneath the base and raising it so that the cover glass is level with the eye. By looking toward a window the slightest space can be seen. In the case of a heavy instrument lower the head to the level of cover glass. Note the division and by an upward focusing turn of the screw, bring the object in focus and read off the distance traversed. In the case of an oil immersion objective follow the same process by bringing the objective in contact with the cover glass dry, then separate slightly and inclining the instrument allow some of the oil to drop into the space, then focus and read off.

In the case of a micrometer screw without graduations the matter becomes more difficult, but can be done in the same manner although not so accurately, by marking the milled edge of the head of screw with a wax pencil or ink, in the two positions, by taking the middle of the arm as a fixed

point and by knowing or determining the pitch of screw ascertain the value of space traversed.

It must however, be borne in mind that cover glass of normal thickness be used, particularly in oil immersion objectives, since if unusual thickness is used, working distance may be entirely eliminated.

Magnifying Power.—This is a question of vital importance in a microscope, not so much as a quality in itself, as in connection with the resolving power. The inquiry should not be simply, how many diameters an instrument will magnify, but what the precision and extent of its definitions are under a certain magnifying power. If a high magnifying power is all that is desired, this may be obtained to an almost unlimited extent by means of simple lenses which may be procured at a small pecuniary outlay ; but these do not give a distinct image nor do they make structure visible which, be it remembered, it is the purpose of the microscope to do.

The normal eye can distinguish from 200 to 250 lines to the inch and in a microscope such magnifying power should be used as will apparently bring the structure which is sought after at least up to this figure. To illustrate take a $\frac{1}{4}$ inch objective of 0.77 N. A. and a 2 inch eyepiece. An objective of this kind properly corrected, resolves the test-object *Pleurosigna angulatum*, in which the

lines average 60,000 to the inch. With the above eyepiece it is utterly impossible to see them, while if it is replaced by a $\frac{3}{4}$ inch or $\frac{1}{2}$ inch eyepiece, they can easily be distinguished. This is not owing to any peculiar quality of the eyepiece, but merely to the fact that by increasing the magnifying power, the dimensions of the object have been increased to such an extent that the lines have apparently been separated and become visible to the eye.

Beginners as a rule are apt to use too much magnification or *amplification*, and often attempt to view a large surface with an objective which will show but a small part of it. It must not be forgotten that the apparent field of view is decreased as higher powers are used and that a low power will give a better impression of a large, coarse object and its relative parts, because it makes a larger surface visible.

In objectives of the same power, but of different angular apertures, the magnifying power and field will always be the same.

The following table will probably be of assistance to the beginner. After he has become better acquainted with his instrument his judgment will dictate to him what to do.

A power of 25 diameters will show a surface of about $\frac{1}{5}$ inch diameter.

A power of 50 diameters will show a surface of about $\frac{1}{10}$ inch diameter.

A power of 100 diameters will show a surface of about $\frac{1}{20}$ inch diameter.

A power of 500 diameters will show a surface of about $\frac{1}{100}$ inch diameter.

A power of 1000 diameters will show a surface of about $\frac{1}{200}$ inch diameter.

This table is approximately correct with a Huyghenian eyepiece.

As we have already shown, magnifying power may be obtained by increase of power in objective, eyepiece, or increase of tube length and have also pointed out that *the objective should be relied upon to obtain this increase.*

Objectives of the same angular aperture, but of different powers, will give identical results by bringing them up to the same magnifying power, unless the difference is considerable. In both objectives and eyepieces the lenses decrease in size with the increase in power and consequently give less light and while this one objection exists in the objective, an additional one occurs in the eyepiece, in that the eye must be brought closer to the eye-lens and must be kept more strictly in the optical axis, which at a long sitting, becomes fatiguing.

Choice of eyepiece should be determined by requirements and individual preference. All responsible manufacturers and dealers make up such outfits of stands, objectives and eyepieces, as ex-

perience has taught them are most generally useful.

It is safe to follow in all work on recognized forms (objects of which the structure is known) the rule, *not to use a higher power than is necessary to properly study them.*

Apochromatic Objectives.—All objectives of what may be termed the ordinary corrections have a residual chromatic and spherical aberration, the former being called the *secondary spectrum*, but Prof. Abbe has in this direction also effected a notable improvement, which with a uniform correction of the spherical aberration, corrects for *three colors*, thus resulting in a closer concentration of image-forming rays which, with the greater numerical aperture made possible by these conditions, results in a higher resolving power. It has been found however, that in high powers of wide aperture, even with these objectives, there is an outstanding error which by itself cannot be corrected. This is balanced in the eyepiece, which is correspondingly over-corrected and is called the *compensating eyepiece*. It has therefore been necessary to make the low powers under-corrected as well, so that they might be used with the same eyepieces, for it is evident that neither these objectives with the Huyghenian eyepiece, nor the compensating eyepiece with the achromatic objectives will give satisfactory results.

The proper combination of apochromatic objective and compensating eyepiece gives a beautiful image with the maximum of resolving power, unapproached by any of the achromatic combinations. They furthermore have the advantage that they are exactly suited to photomicrography, there being exact coincidence between the photographic and visual image, which in the achromatic objectives is not the case, as these must be either corrected for photography, when they are not satisfactory for ordinary purposes, or an allowance must be made for the difference between two images in the camera, which is very difficult.

One objection to the high powers has been the liability of deterioration in some of the materials used, which has happily been overcome by Prof. C. S. Hastings of New Haven, Conn., in the apochromatic objectives, which have been computed by him for the Bausch & Lomb Optical Co.

Eyepiece or Ocular.—The purpose of the eyepiece is so to refract the diverging rays coming from the objective that they will reach the pupil of the eye and while the most simple part of the optical combination is, withall, an important part of it. In fact it may be said that, owing probably to its simplicity, it has been neglected and very often eyepieces are furnished which are not at all commensurate with the quality of the objectives.

They are divided into two classes :

Negative eyepieces in which the focus is within the eyepiece itself (at the diaphragm).

Positive eyepieces, in which the focus is outside and below the field lens and which can be used as magnifiers.

Huyghenian Eyepieces.—This is named after Huyghens, who is said to have first used it. It is a construction which is in most general use, although made up in a variety of mountings. It is negative and consists, as we have already stated, of an *eye lens*, nearest the eye and *field lens* or *collective* which is the large lens nearest the objec-

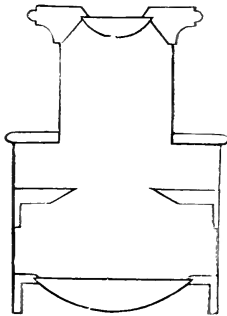


Fig. 36.

tive. Between them and placed as the focus of the eye lens is a perforated, blackened disk, called a *diaphragm*, which limits the size of field and

shows it within a sharply defined border. It is made up in two forms :

The *English type* as shown in Fig. 36, which has a large tube fitting into the microscope tube and a neck or smaller tube which is usually arranged with a cap to slide over the eye lens.

The *Continental type* has a straight tube which drops entirely into the tube of the microscope and rests upon the mounting of the eye lens.

Solid Eyepiece.—This is also a negative eyepiece and is the invention of the late R. B. Tolles. It is called solid, from the fact that instead of being composed of two lenses, it consists of one piece of glass, which is cut to a cylindrical form and on the ends of which the proper curvatures are ground and polished. The diaphragm is made by cutting a circular groove into the glass at the proper distance between the two surfaces, which is then filled up with an opaque pigment. (Fig. 37.)

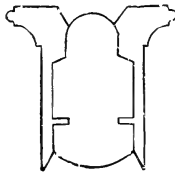


Fig. 37.

These eyepieces are only made in high powers, as optical glass is usually not of sufficient homo-

geneity to make low powers, and their cost would be too considerable, without a corresponding advantage. They are usually only made in powers of $\frac{1}{2}$ inch and stronger and for this reason have but a limited use.

Ramsden Eyepiece.—This is a positive eyepiece and is constructed of two plane convex lenses with the convex surfaces toward one another. They are especially useful as *micrometer eyepieces* in microscopes and telescopes, as they are used as a magnifier on the fine divisions of micrometer and at the same time give the virtual image.

Periscopic, Orthoscopic and Kellner Eyepieces.—These are also positive eyepieces which

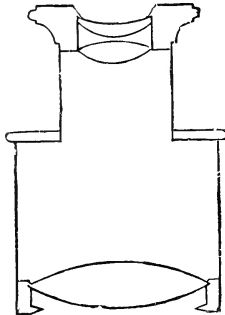


Fig. 38.

are achromatized by making the eye lens a doublet or triplet and by its correction permits the use of a larger and flatter field. (Fig. 38.)

There are a large number of other eyepieces called by a variety of names which can be found in catalogues, but for which there is very limited use. They, therefore, require no special mention in this connection.

Compensating Eyepiece.—In this place it is also proper to speak of the new compensating eyepiece as made and suggested by Prof. Abbe. These eyepieces compensate the residual errors in the apochromatic objectives and are of no value except when used with them and for projection, as they are highly over-corrected, as can be seen in the edge of the field, which has a strong orange color, whereas the Huyghenian has a blue color.

In this list of eyepieces there is a *searcher* which is of low power and intended to find objects. The higher powers are for general work, some of them being negative others positive. One more kind is the *projection eyepiece* which is intended for projecting an image on a photographic plate, or on a screen or wall.

Rating or Designation.—Until within recent years the rating of eyepieces was arbitrary, each maker following a system of his own and while this is still done, the general method followed is to name them as in objectives, according to a single lens, which the equivalent focus of the eyepiece lenses will equal and do this in inches or millimeters. This conveys an idea of the extent to

which an eyepiece magnifies the real image. Thus a 2 inch or 50.0 mm. eyepiece magnifies five times, one of 1 inch or 25.4 mm. focus ten times and so on.

Flatness of Field.—Although this depends mainly upon the objective, the absence of it may be owing to a faulty construction of the eyepiece. If it is so prominent as to be easily noticeable and to the same degree with a number of objectives it may be ascribed to the eyepiece. It must, however, be remembered that an absolutely flat field has not yet been obtained; it may be closely approached by decreasing the diameter of field to less than its normal size.

Size of Field.—Quite a general but erroneous idea prevails that the size of the tube has an influence on the size of the field. Except in eyepieces of very low power, or with tubes with smaller than usual dimensions, this is not true. It must be remembered that a Huyghenian eyepiece admits of a definite size of field and this is regulated by the opening in the diaphragm; the same size of opening is used in all of the same power, whether it is an eyepiece with a large or small diameter.

Defects.—As has been stated many eyepieces are carelessly constructed and possess defects which interfere with obtaining a distinct image. These defects do not show easily in low power objectives, but can readily be seen with high

powers. The most frequently occurring fault is the lack of perfect grinding and polishing of the lens surfaces. If the former, it will show itself as spots in the field and if the latter, as a series of streaks and shadows, usually circular in form as if the lens had been wiped with greasy fingers. This may be the difficulty, and before passing judgment the lenses should be carefully cleaned.

Another defect may be in the glass itself, in the so called *striae*, which will be indicated by dark and light streaks across the field.

Care must be taken not to confound small particles of dust which are apt to fall upon the field lens and which at times are very prominent in the field, with imperfections of the surface. These can be distinguished from other defects only by wiping, or using a camel's hair brush, and even with the utmost care some particles are liable to remain.

The eyepiece often fits too closely in the tube and when making observations with high powers, a change of eyepiece is apt to disturb the object. It should enter without any friction and still so closely that it drops slowly into its place from the compression of air in the tube when objective is attached.

Eyepieces are now generally made *parfocal*, that is the equivalent foci are made to correspond,

so that a change in eyepiece does not materially effect a change in focus and no further adjustment than a slight turn of the micrometer screw is required.

HOW TO WORK.

To Attach Eyepiece.—As the exterior surfaces of the eye lens and field lens are exposed, they are apt to become dusty and should always be carefully cleaned before use. If there are two or more eyepieces, always use the lowest power first. Eyepieces should be so loosely fitted that they will drop into the tube as far as the collar *by their own weight*. They do this slowly when the objective is attached, as an airtight compartment is formed and air to the extent of the dimensions of the eyepiece must first be expelled from the tube. To fix in position however, it may be hastened by gently pushing the eyepiece downward, but not to such an extent as to push in the draw-tube, or force down the coarse adjustment. In fact, care must be used in applying the eyepiece, or sliding the draw-tube, as the objective may be forced against the object and thus destroy it, or the focus may be disturbed.

To Attach Objective.—Take a low power objective first and after it has been removed from the box, see that its front lens is clean; elevate the tube by means of the coarse adjustment so

that the nose-piece shall be at least two inches from the stage and place the upper end of screw of objective in the thread of the nose-piece, hold the lower end between the fore and middle fingers (left hand) palm upwards, so that it is in line with the tube and gently pressing upward, revolve the objective with the thumb and forefinger of the other hand (right hand) by the large milled edge at its upper end, until shoulder sets against shoulder. To properly attach an objective is not always simple and *cannot be done too carefully*. One danger lies in the fact that the objective may be dropped onto the object and thus injure or destroy one or the other or both and another that the thread is started wrong by holding the objective sideways and the threads may thus be destroyed.

In this connection we draw particular attention again to the convenience of the double, triple and quadruple nose-pieces. The convenience which is obtained from their use, freedom from danger to objective and object and saving of time, commend them in all cases where two or more objectives are used.

Finding an Object.—The slide upon which the object is mounted is placed upon the front of the stage and slipped under the two spring clips to a point where the object comes as nearly as possible in the center of the opening of the stage.

The slide should pass easily under the clips which however, is not the case when the edge of the clips are too bluntly rounded and when the springs are too stiff. In either case the clips may be bent so that the slide will work easily. Some persons prefer to work without clips, but this can only be done after considerable experience has been acquired and only when the instrument is in an upright position.

With the low power objectives, which are used on coarse and usually large objects, it will be found after properly focusing, that a portion of the object will show itself in the field and by moving it, can easily be brought to the center. In this connection it must be remembered that the image in the eyepiece is in a reversed position from that of the object and that a movement of the object to the left gives an apparent movement to the right in the field. This will create some confusion at the outset, but after a little practice the movement becomes involuntary. In the case of a small object which is not found after the objective is known to be in focus and which is easily recognized by the mounting medium or small particles of dust on the cover glass, move the slide about on the stage by grasping one end with the thumb and forefinger, when it can usually be recognized by the shadowy outlines of the object as it flits across the field. The difficulty of

finding an object or a particular spot in it becomes more difficult with the increase in power and even in experienced hands becomes quite vexatious. Recourse may be had to two methods :

First.—By using a low power eyepiece.

Second.—By using a low power objective as a finder.

The object is thus found and after moving to the center of the field the objective is removed and the high power attached, or in case a revolving nose-piece is used, swing the high power objective in position, care being taken not to touch the slide, and focus in the manner to be described. A low power eyepiece in this connection is also very useful. The object may not be in the field, due to a slight variation in the centers of the objectives, but it will certainly be very close and ought to be easily found.

The Mechanical Stage in either the fixed or attachable form will be found to greatly facilitate work in this direction, particularly if the object is minute and if in an important investigation one desires to be absolutely convinced that the whole field of the object has been covered, as for instance in the search for bacilli.

To Illuminate the Object.—This is an extremely important feature and should always be carefully done, as one may easily fail to obtain the

best results, may be led to wrong conclusions, or may injure the eyes.

The mirrors of the microscope are usually plane and concave. As it is clearly inconvenient or impossible to hold the microscope toward the source of light, they are provided to reflect the light upward to the object when this is transparent. The plane mirror reflects the light in the initial intensity of its source and is used with low power objectives. The concave mirror concentrates the rays on the object and thereby gives intensified illumination and is used with medium and high power objectives, except when substage condenser is used, which subject is left for future consideration.

The sources of light are either daylight, or artificial light from a lamp. In the former the light from a northern sky is preferable and in the latter a flat-wick oil lamp, or a Wellsbach gas flame. An ordinary gas flame should not be used on account of the difficulty of obtaining equal illumination and the constant flickering which is very injurious. When using the flat-wick lamp the narrow edge of the flame should be used, as this is more intense than the broad side, as can be easily determined by experiment.

When using daylight, place the microscope as near as possible directly before a window and when a lamp is employed have it on the table within easy reach.

Light is either *transmitted* or *reflected*. When the former, it is used on transparent objects and passes through the objects from below the stage into the objective. In opaque objects this is impossible and reflected light is required, when it is directed onto the object from above and illuminates its upper surface. In the following instructions it is assumed that transmitted light is used unless otherwise stated.

The concave mirror converges the light and therefore has a focal point and it is evident that if its focus is of such length that with parallel rays (daylight) it will fall on the object, the focus will be longer with the diverging rays (lamplight) and when no provision is made in the instrument to adjust the mirror to meet these two conditions, it becomes difficult and sometimes impossible in critical work to obtain the best result. For this and other advantages an additional illuminating apparatus, called a *condenser*, is now commonly used.

Before lighting an object make certain that the mirror-bar is in exactly central position and set the mirror at such an angle to the light, that it will be directed upon the object, which can be done most quickly at the outset by observing the object direct, keeping the head at one side of the tube. Now remove the eyepiece and observe the light through the objective. It should be central and of

equal intensity, which with daylight is sometimes difficult, as the sash of the window may be reflected and show itself in the field as dark bands, or in the case of lamplight the blue portion of the flame may appear as a dark spot. These are only preliminary directions but will suffice for the beginning. There will be little difficulty in obtaining proper illumination at the outset, if one will bear in mind the three necessary conditions when looking at the back of the objective :

- Central illumination,
- Even illumination over the entire field,
- Mellow illumination.

Defects in illumination which may not be apparent will show when the eyepiece is again attached, and defective lighting will be indicated,

- When dark points or shadows appear in the field,
- When a shadow is thrown at one side of the object,
- When the object appears to lie in a glare of light.

In the first two cases the correction can be made by suitably adjusting the position of the mirror. In the latter by reducing the amount of light, (which it may be said in passing, is seldom necessary with medium or high power objectives), by the use of a diaphragm.

When first setting up the instrument the largest opening of the diaphragm should be used and if the light is found too intense, it may be reduced by bringing into position a smaller stop, or in case of the iris diaphragm, by reducing the size of opening. The iris diaphragm is infinitely superior to the revolving or cap diaphragm as these have fixed openings, whereas with the former any intermediate gradation may be obtained.

It is now generally conceded that observations may be made with the microscope to any extent without any detrimental results to the eyes, provided however, that the conditions of light are just right. It is a good rule to follow, to use the least illumination which will show the structure being studied and it may also be safely accepted that if the eye tires or feels uncomfortable, that the light should be moderated.

Illumination is either—

Central or axial, when the center of the mirror is in the optical axis, or

Oblique, when the mirror is swung to one side which, in objectives of wide aperture, will disclose structure which cannot be seen with central illumination.

How to Focus.—A safe method to follow and one which is generally in vogue by all careful manipulators, is to *focus upwards*—that is, bring the objective closer to the object than its working

distance and, applying the eye to the tube, elevate the objective by the coarse adjustment until it is in focus. In the lower power objectives, in which working distance is considerable, this may appear unnecessary, but is nevertheless strongly recommended, principally to retain the systematic method. In the medium and high powers it is very important as the working distance is small. In these, the front of the objective is brought down nearly in contact with the cover glass and the distance should be judged by holding the head down to the level of the stage and observing the distance as the objective is brought downward. The rack and pinion is infinitely superior to the sliding tube, as the distance can be traversed with absolute certainty without any liability to slip. Then apply the eye to the eyepiece and rack upward, slowly however, that the point at which the focus is reached may not be overlooked, which may easily be the case in transparent and faintly colored objects. After a better acquaintance with the working distance of the objective the proper distance can be judged quite closely.

After the focus has been found the fine adjustment should be brought into action. This should only be used after the focus has been obtained with the coarse adjustment, in order to focus through the different planes or depths of the object. Its range of movement is necessarily short and at one

end of the screw comes to a stop, and at the other goes beyond the limit of the movement and becomes inoperative. It should always be kept as near as possible *at the medium point*. During observation of the object, the milled head of the fine adjustment should always be kept between the thumb and forefinger of one hand (right) turning the screw in either direction to focus in different planes of the object, while the other hand (left) moves the object about.

Which Eye to Use.—The right eye is generally used for observations, but while the manipulator may from habit be inclined to use this, it may be possible that in some cases the left can be used to better advantage and with less fatigue. It is a fact well known to oculists and opticians that many eyes are defective, of which fact the possessors may not be aware. Short or long sightedness has little or no influence in viewing an object, except to require a different adjustment, but so called *astigmatism*, by which lines in a certain axis cannot be seen distinctly, may influence the best results. If this error is corrected by wearing glasses and these are used while making observations, either eye can be used. But, in order to determine whether a defect exists, of which the possessor is not aware, observe closely with both eyes, preferably an object with fine striations, such as *Diatomacae*, to learn whether with one eye a

better view is obtained than with the other and use the one giving the best results.

As already stated, make it a habit at the outset to *keep both eyes open*.

There is one point over the lens called the *eye-point* at which the rays cross within the smallest compass and *this is the proper position* for the eye, as the largest number of rays enter it. *When above or below this point* the size of field will be either reduced, or shadows or colors will appear in it. In low power eyepieces the eye-point is farther than the lens; in high powers quite close—in fact, in some so close that the eyelashes may rest upon the lens and may sometimes appear to be in the field as dark bars. Generally speaking the best point is where the entire field is seen and its margin (diaphragm) sharply defined.

What Objects to Use.—Suitable objects to test the capacity of objectives are also valuable in leading the student to a skillful use of the instrument and in giving him proper judgment.

Low Powers—*Proboscis of blow-fly*. This should be flat and transparent. For 1 inch, $\frac{2}{3}$ and $\frac{1}{2}$ inch objectives the scales from *Lepisma sacharina*.

Medium Powers—*Pleurosigma angulatum*, dry, stained Bacteria and Micrococci.

High Powers—Oil immersion $\frac{1}{10}$ inch and $\frac{1}{12}$ inch objectives, *Amphipterua pelucida*, *Surirella gemma*

in balsam or styrax. Stained Bacteria and Micrococci.

Test Plate.—This will be an excellent acquisition for all those who can meet the pecuniary outlay. It consists of a series of 20 diatoms arranged according to the coarseness of the lines. They are furnished mounted both in balsam and styrax. Below is a table giving the numbers, names of the various diatoms and divisions on their surfaces to $\frac{1}{1000}$ inch. A specimen of *Eupodiscus argus* begins and ends the series :

	Stiriae in $\frac{1}{1000}$ inch.
1. <i>Triceratium favus</i> , Ehrbg., - -	3.1 to 4.
2. <i>Pinnularia nobilis</i> , Ehrbg., - -	11.7 to 14.
3. <i>Navicula lyra</i> , Ehrbg. var., - -	14.5 to 18.
4. <i>Navicula lyra</i> , Ehrbg., - -	23. to 30.5
5. <i>Pinnularia interrupta</i> , Sm. var., -	25.5 to 29.5
6. <i>Stauroneis phoenicenteron</i> , Ehrbg.,	31. to 36.5
7. <i>Grammatophora marina</i> , Sm., -	36. to 39.
8. <i>Pleurosigma balticum</i> , Sm., -	32. to 37.
9. <i>Pleurosigma acuminatum</i> (Kg.) Grun., - - - -	41. to 46.5
10. <i>Nitzschia amphioxys</i> , Sm., -	43. to 49.
11. <i>Pleurosigma angulatum</i> , Sm., -	44. to 49.
12. <i>Grammatophora oceanica</i> , Ehrbg. <i>G. subtilissima</i> , - - - -	60. to 67.
13. <i>Surirella gemma</i> , Ehrbg., - -	43. to 54.
14. <i>Nitschia sigmoidea</i> , Sm., - -	61. to 64.
15. <i>Pleurosigma fasciola</i> , Sm. var., -	55. to 58.

- | | | | |
|--|-------|-----|--------|
| 16. <i>Surirella gemma</i> , Ehrbg. | - - - | 64. | to 69. |
| 17. <i>Cymatopleura elliptica</i> , Breb., | - - - | 55. | to 81. |
| 18. <i>Navicula crassinervis</i> , Breb. Frus-
tulia saxonica, Rabh., | - - - | 78. | to 87. |
| 19. <i>Nitzschia curvula</i> , Sm., | - - - | 83. | to 90. |
| 20. <i>Amphipleura pellucida</i> , Kg., | - - - | 92. | to 95. |

Whatever opinion one may have in reference to the study of diatoms, the fact cannot be gainsaid that they have been a great aid in the improvement of objectives, are used by opticians to judge their various characteristics and offer a reliable standard for testing the resolving qualities, such as no other objects can. The writer particularly recommends that the test *P. angulatum*, dry, form a part of every outfit, not so much as a test for resolvability as to use it for study and to acquire quickly a knowledge how to judge the phenomena of spherical aberration. It may be used on powers $\frac{1}{4}$ inch and higher and after it has served its purpose may be put on one side to work over objects which come under the particular branch of study which one is following.

The writer wishes to counteract as much as possible the opinion, which is too prevalent, that the use of diatoms indicates microscopical play and is unworthy of consideration in histological and biological work, but the fact that the optician deems them necessary for determination of optical qualities ought at least to indicate that they are a valu-

able adjunct and certainly will aid in giving greater manipulative skill.

At this point it is considered advisable to add some suggestions from Carpenter.

“The correctness of the conclusions which the microscopist will draw regarding the nature of any object from the visual appearance which it presents to him, when examined in the various modes now specified, will necessarily depend in a great degree upon his previous experience in microscopic observations and upon his knowledge of the class of bodies to which the particular specimen may belong. Not only are observations of any kind liable to certain fallacies arising out of the previous notions which the observer may entertain in regard to the constitution of the objects or the nature of the actions to which his attention is directed, but even the most practiced observer is apt to take no note of such phenomena as his mind is not prepared to appreciate. Errors and imperfections of this kind can only be corrected, it is obvious, by general advance in scientific knowledge; but the history of them affords a useful warning against hasty conclusions drawn from a too cursory examination. The suspension of the judgment, whenever there seems room for doubt is a lesson inculcated by all those philosophers who have gained the highest repute for practical wisdom; and it is one which the micro-

scopist cannot too soon learn or too constantly practice. Besides these general warnings, however, certain special cautions should be given to the young microscopist with regard to errors into which he is liable to be led even when the very best instruments are employed."

How to Set Up the Instrument.—Draw the instrument from the case by grasping the base, free it from dust with a large camel's hair brush, 1 inch or $1\frac{1}{2}$ inch wide, or by wiping carefully with a chamois skin, or old linen handkerchief. Place the instrument with proper relation to light on the work table, which should be of such height that observations can be made with the utmost possible comfort without straining the neck or compressing the chest. Bear in mind always to sit as upright as possible.

Bring the draw-tube to standard length by drawing the draw-tube to the length for which the objectives are corrected. Do this by grasping the milled edge of the draw-tube and give it a spiral motion while holding the main tube with the other hand, which will facilitate its easy movement. The objection being, however, that in any but cloth-lined sheaths the polished tube will soon be scratched, especially if not kept very clean. In stands without the graduated tube, a mark or ring is, or should be, provided on it, which should be made to coincide with the upper end of main tube.

Where the graduated draw-tube is provided bring the proper figure, either 216.0 or 160.0 mm., in line with the upper end of main tube, in accordance with the tube length to which the objectives are corrected.

Attach low power eyepiece.

Attach low power objective.

Place object on stage.

Illuminate object.

Focus on object.

If the instrument is used in the upright position, place the base close to the edge of the table; if inclined, it may set farther in. Rest the arms, as much as the height of the instrument will permit, upon the table.

How to Work.—It is now supposed that the instrument is ready for work. To start, it is well for the beginner to provide a few prepared specimens, as these will help him considerably if it is his intention, as it should be, to prepare them later himself. If only a portion of the object can be seen and it is desired to see a larger surface, the length of tube may be contracted by means of the draw-tube. In this case the object will be placed out of focus and another adjustment becomes necessary. If a higher power is desired the draw-tube may be extended. Observe whether the field is well illuminated and if not bear in mind what has been said about properly adjusting the

mirror. Now use the micrometer screw, remembering to grasp the milled edge between the thumb and forefinger, and work to the right and left, to reach different depths, and note carefully the beautiful structure which is open to view.

Opaque Object.—We will now suppose that the object is a mineral or plant. Place this upon a slide and slip under the clips. In this case the low power objective is used for two reasons; because a general view is sought, involving low magnification and large field with light-giving power and if it should be desired to use a higher power this cannot be done on account of the short working distance. The light may and undoubtedly will be found insufficient to distinguish the object clearly. If the instrument is of the American type, swing the mirror-bar upon its axis around the stage to a point above it so that it will be at an angle of about 45° to its surface. If a lamp is used and is in the same position as when used for transmitted light, it is probable that the tube of the instrument will obstruct the light and it is then well to move it toward the front. Using the concave mirror, adjust it so that the light will be concentrated upon the object, by watching it directly, and then observe through the tube. If it is not sufficiently illuminated continue to adjust the mirror; also vary its distance from the object and swing the mirror-bar to a higher or lower point.

Medium Power Objective.—After sufficient time has been devoted to study with the low power objective, exchange it for the higher power and replace the object by the slide *P. angulatum*. Focus upon this, being mindful of the suggestions previously given and do not fail to observe what has been said in regard to well illuminated field. Observe now whether any lines can be seen upon the surface of the diatoms. If not, vary the distance of the mirror from the object, if adjustment is provided for; or, if lamp light is used bring the lamp closer to, or remove it from the instrument in one line, so that the illumination will not disappear. If this does not bring out the lines, swing the mirror-bar from the central position into an oblique one, on the side opposite to that of the light and readjust the mirror. Grasp the ends of the mirror-fork between the thumb and middle finger and move the mirror by the first finger. If the field cannot be evenly illuminated, it is evident that the mirror is beyond the limit of angular aperture of the objective and it must therefore be brought back until the light appears equally well over all parts of the field. It must here also be noticed that if the diaphragm is still attached to the instrument and does not swing with the mirror, it may also be the means of cutting off light. The largest opening should be used or in the case of the cap dia-

phragm, remove it. By means of the micrometer screw carry the fine adjustment back and forth beyond the plane of the object and observe closely whether any lines can be distinguished. It is very probable that they will show; but if not, the cause should be determined. It may be that the magnifying power is not sufficiently great and in this case a higher power eyepiece should be used, or the cover glass may be more or less than the normal thickness, which would cause a spherical over or under-correction in the objective. In this case the lines would appear when the diatom is not in focus. If the objective is a non-adjustable one, the proper correction may be approximately reached by means of the draw-tube. If the lines appear over the plane of the object, it shows over-correction and the length of tube should then be decreased, or contrary when the lines show below or beyond the plane of the object. If the above directions have been followed, the lines cannot fail to be seen with a moderately good $\frac{1}{4}$ or $\frac{1}{3}$ inch objective; but if they are not, the trial should be repeated. Again, be careful to have no obstruction between the course of rays from the mirror to the stage; get good illumination on the object; observe well; keep the instrument in such a position that the object is not illuminated from any other direction than from the mirror.

When the diatoms are *resolved* in this manner the lines will appear to be diagonal in some, longi-

tudinal or transverse in others, according to their position and if the resolution is very good, these lines will further resolve themselves into minute beads of a hexagonal form.

It will now be well to bring the mirror more nearly to a central position. Do this at intervals of about 10° and note the appearance at each decrease of obliquity. It will be found that as the mirror approaches the optical axis the lines will appear to become more faint and may disappear before central illumination is reached; in this case it will be well to begin again. An endeavor should be made to make each attempt give better results than the preceding one. Repeated trials will not only impress the various phenomena upon the mind, but will cause a notable improvement in manipulative skill and thus a better performance in the objective.

To Judge Spherical Aberration.—This is a matter of experience based upon a knowledge of the principles involved and after having been studied, will be found to be of the utmost value in utilizing the capacity of a microscope. To judge spherical aberration by the use of histological or biological objects without a previous knowledge, acquired from objects which are more suited, is extremely difficult. One may be aware, by the unsatisfactory appearance of the image, that

something is amiss, but will probably not know how to correct the deficiency.

Using an objective $\frac{1}{4}$, $\frac{1}{5}$ or $\frac{1}{6}$ inch and the test object *P. angulatum*, select a diatom which is flat and move to the center of the field. Focus carefully so that the margin of the object will be sharply defined and observe the markings. If they show in the same plane without any further focusing, the spherical correction may be taken as being correct. If, however, the lines appear to lie in a higher plane and it is necessary to focus upward so that the margin of the diatom is out of focus, it indicates spherical over-correction and the remedy is found in the contraction of the tube length. This should be done progressively in spaces of about $\frac{1}{2}$ inch and after each change, carefully focus again until proper correction is obtained.

When the lines appear to lie below the plane of the object, it indicates spherical under-correction and can be corrected by increasing the tube length. If there are two or more eyepieces, results can be obtained quicker with the higher powers.

If the markings cannot be seen, it may be due to abnormally thick or thin covers, a not uncommon occurrence, thus destroying the resolving power. This may be judged by using slight oblique illumination. If too much is used the nice differences will be lost.

Chromatic Aberration.—This may be judged as described under “Chromatic Aberration” in a previous chapter.

Cover Glass.—We have thus far not considered the cover glass, except to show its influence on the optical performance of objectives. In preliminary examinations of solid objects with low powers it may be dispensed with; but where fluids are used, whether with low, medium, or high powers it should always be used. A drop or small quantity of fluid placed upon a slide assumes a spherical form and, on viewing it with a low power, it will be found to give a distorted field and will cause disagreeable reflections and shadows.

In medium and high powers, the front lenses will be so close to the water, urine, blood, etc., that capillary attraction will often cause an adherence to the front surface of the objective; besides this, there is such a considerable depth to the fluid that it obstructs the light, requires a great change in adjustment for the various planes and is usually in such vibration that a sharp focus becomes impossible. By merely dropping a cover glass upon it all these objections are overcome.

The covers are commercially classified as No. 1, No. 2 and No. 3, but there is a variation within the

limits of different numbers. The variation is about as follows :

No. 1, $\frac{1}{150}$ to $\frac{1}{200}$ inch, or 0.16 to 0.13 mm. thick ;

No. 2, $\frac{1}{100}$ to $\frac{1}{150}$ inch, or 0.25 to 0.16 mm. thick ;

No. 3, $\frac{1}{50}$ to $\frac{1}{100}$ inch, or 0.50 to 0.26 mm. thick.

According to the prices of cover glasses, when purchased by weight, the No. 1 gives the greatest number and No. 3 the least. It may for this reason be thought that the purchase of No. 1 is most advantageous, but it must be considered that there is a greater amount of breakage by cleaning, as they are very thin and sensitive. Considered from the optical standpoint, the thinner of No. 2 and the thicker of No. 1 should be used, as these come within limits which are adopted by opticians as standard and to which medium and high power objectives are corrected. Test objects which are prepared to test the resolving power of objectives and consist of diatoms, are generally covered with these thicknesses.

An excellent means of determining the thickness of cover glass, as well as of studying spherical and chromatic aberration, is the Abbe test-plate. This consists of a series of cover glasses ranging in thickness from 0.09 mm. to 0.24 mm., silvered on the under surface, with lines cut through the film, and cemented to a slide, each cover being marked with its thickness.

Spherical aberration is corrected when the bands or lines show distinctly without any nebulous fringe, and thus indicate the proper thickness of cover to use.

Chromatic correction may be judged by the character of color bands which show with oblique light. If the bands are narrow and the complimentary colors of the secondary spectrum, yellow-green to apple-green on one side and violet on the other, indicates the best correction. For determining the thickness of cover glass and best correction for spherical aberration we refer to the cover glass gauge designed by the writer.

If the cover glass is first measured with this before the object is mounted, the tube length is given for proper correction for each thickness and can be read off on the drum. The scale provides for $\frac{1}{4}$, $\frac{1}{5}$, $\frac{1}{8}$, and, $\frac{1}{1\frac{1}{2}}$ inch objectives of the long tube length and for the $\frac{1}{6}$ inch of the short tube length. It also gives the thickness of cover in millimeters and inches, and thus indicates the true position of adjustment-collar in adjustable objectives.

Dry, Adjustable Objectives.—Adjustable objectives, or objectives with *collar correction* are those in which there is a provision to vary the distance between lenses, in order to make proper correction for spherical aberration with facility. They involve a high degree of mechanical perfection and are therefore more expensive than objectives with

fixed mountings. They may, however, be recommended to microscopists who have acquired some experience in handling objectives and even to beginners who will use judgment in their use, as they certainly give excellent results and quick means for obtaining the utmost limit of efficiency in objectives, a fact best appreciated by those who use them. In the objectives as at present constructed by the best makers, a milled collar is provided, which in turning imparts a rectilinear motion to an interior tube, carrying the posterior systems of the objectives, thus varying the distances between them and the front system, which remains stationary. The screw collar is graduated in such a manner that the figures indicate the correct point for the proper thickness of cover, thus 10 indicating proper correction for a cover of 0.10 mm., 16 for 0.16 mm. and so on. When set at the highest figure, for thick covers, the lenses are in the closest position and the adjustment is said to be *closed*. When for the thin covers, are farthest apart and is then *open*.

In objectives of older construction and in some produced at the present day, the figures are arbitrary and serve no other purpose than an index for reference.

Close the adjustment before attaching the objective as its front may otherwise come in contact with the cover before the focus is reached. For

practice with this objective use *P. angulatum*. Focus carefully and observe whether any lines can be seen ; if not, grasp the milled edge of the adjustment collar between the thumb and first finger of the left hand, keeping the fingers of the right hand upon the micrometer screw. Turn the collar slightly toward its open point and as this will place the object out of focus, move the fine adjustment correspondingly. Continue to turn the collar little by little and do not cease to observe closely ; also, after each movement, focus above or below the plane of the object, so^o that this will be distinct, and look for the lines. Possibly after a little they will begin to appear faintly ; but, if not, continue to bring the collar toward the middle point. The lines must now soon make their appearance and when they do, it will probably be above the plane of the diatom. This is an indication that the objective is approaching its correction for the cover. Now keep the lines in focus, while the correction collar is being gradually turned, until the lines and the outline of the diatom lie in one plane. The objective is now said to be corrected for cover. Observe which number corresponds to the index, and again return the collar to its closed point and go through the same proceedings as carefully as at first. When the best point is again reached look for the number and see whether it agrees with the first ; very likely it does not, which is

owing to a want in the faculty of perception, due to a too slight acquaintance with the phenomena. These trials should be repeated until the proper sensitiveness of feeling in making the adjustments is acquired and until they can be made to correspond with a certainty to at least within two divisions. When it is found after repeated trials that sufficient skill has been acquired, mark the number upon the slide. For future examination of the same slide, this will facilitate work and give the assurance that the best results are thus obtained without further trial by simply referring to the recorded number.

On stained Bacteria and Micrococci, focus briskly with the fine adjustment to either side of exact focus. There will be an expansion of the outline of the object both when within and without the focus. If the greater expansion or coma is within the focus, or when it is necessary to raise the objective, there is spherical over-correction and the adjustment must be closed. When the proper point of correction is reached the expansion of outline is the same in both directions.

Immersion Objectives.—As has been stated before, immersion contact between the objective and cover glass is made by either water or homogeneous fluid. Distilled water only should be used and kept in a suitable bottle. Cedar oil is used for the homogeneous fluid and is thickened

for convenience in use. A small bottle is generally supplied with each oil immersion objective. *Great care should be used in keeping it free from dust*, as it often happens that an objective fails to give satisfaction, due to a small particle of dust which may float in the fluid before the front lens. Great care should also be exercised in applying oil to the front lens and after the application, it is strongly recommended to examine it *with a magnifier*, that there may be no air bubbles present. These as well as dust may seriously interfere with obtaining satisfactory results. If bubbles are present the oil should be removed and a fresh quantity applied. Care should also be taken *not to apply too great a quantity*. After the stopper has been withdrawn from the oil, allow the oil to run down the rod or brush until the last natural drop has separated from it and apply the remainder, or less than a drop, to the front of the objective.

Attach the objective and lower it until the fluid comes in contact with the cover and observe this by lowering the head. Focus as with dry objectives. The use of immersion fluid in itself involves a certain amount of inconvenience, but the observance of fixed rules will materially help to overcome some of the disagreeable features. Extreme cleanliness should be observed with it. After the work has been completed the objective should be removed from the stand and its front as well as

the slide should invariably be cleaned. The fluid may be removed by a moist piece of soft linen and the front then cleaned with a dry piece or with lens paper. Chamois skin is not suitable, as it does not absorb the fluid.

Immersion Objectives on Test Plate.—From the fact that in oil immersion objectives the fluid has the same refractive index as the cover and front of the lens, it is not sensitive to variations in thickness of cover, although many of the most expert manipulators prefer adjustable mountings in order to obtain the highest results, if any disturbing element should be present.

To determine the highest capacity on test objects, ordinary daylight is not sufficient; a flat wick oil lamp is best suited. If the right hand is used on micrometer screw, place the lamp at the right side of the instrument, about ten inches from it, with the edge of the flame turned toward the mirror.

Place the test plate upon the stage and as the diatoms in balsam are very transparent and therefore very difficult to find, a lower power objective may be used as a finder; bring No. 1, or Tricentrum favus into the center of the field and after the objective has been removed, attach the immersion objective, which we assume to be a $\frac{1}{4}^2$, in the manner prescribed. Get the best possible illumination with the mirror at the central point and move the test plate from diatom to diatom until it

reaches No. 11, *P. angulatum*, but observe closely the structure of each one as it comes into the field. Next, see whether the objective is corrected. If the lines and outlines, or middle rib, do not appear to be in one plane, adjust the collar in adjustable and the tube length in non-adjustable objectives until they are and then continue the advance toward the higher numbers until one is reached on which no lines can be seen. Swing the mirror-bar to an obliquity of 20° to the left side and readjusting the mirror, observe the effect. It is very probable that the lines will show and if so, continue the advance; if they do not, give 10° or 20° more obliquity and after the structure comes out, again go forward. A point may thus be reached, where with the greatest obliquity which can be given and with the best possible illumination, the objective seems to have come to the limit of its performance. From the claims which have been made for it, it ought to do better. What is the cause of failure? Possibly the mirror is not correctly focused, or the adjustment collar may not be correct for oblique light; perhaps the eyepiece does not give sufficient magnifying power to distinguish the *striæ*. It may be any one of these causes or all combined. As to the eyepiece, the manipulator must remember the amount of separation of lines in the last object which was resolved and from the gradation in the coarser specimens must judge

whether the power is sufficient ; it should be added that for any over No. 14 and under No. 18 a 1 inch eyepiece should be used and for those above No. 18 a power of $\frac{3}{4}$ inch will probably be necessary. After this condition has been complied with, look to the correction collar of the objective. To obtain the highest results it very often occurs that a different adjustment is required for oblique light from that for central light. Note the number at which the collar stands and then work it back and forth, watching carefully for results. If this has no influence, return it to its number or to a point where the outline of the object appears most sharp. Now look to the illumination ; vary the distance of the mirror to the object, or if this cannot be done, vary the distance of the lamp to the instrument and watch the effect of the change through the eyepiece. If neither of these changes give any improvement, recourse must be had to another expedient. Place a bull's-eye between the lamp and mirror with the plane side of the lens toward the lamp and close to it, so that the light is thrown on the mirror. It should be properly concentrated, so that the circle of light will not be larger than the mirror, which can be determined by placing the hand or a piece of paper back of it. Adjust when necessary by moving the lamp or bull's-eye. Keep it a little below the line of the top of the stage, so that the beam from the bull's-eye will not

strike it on its upper surface and as little as possible on its lower surface. If the direct light from the bull's-eye reaches the object, it destroys to some extent the effect of the oblique illumination from the mirror. Great care should be given to this point, as it is very important.

If all of these suggestions have been followed, a great difference will undoubtedly be noticed in the performance of the objective ; but if it still does not come up to the standard, patience must not be lost. The slightest change in the position of the mirror, bull's-eye, or lamp, or a touch to the correction collar or micrometer screw, is sometimes followed by astonishing results. The beginner should sit down with the expectation that he will fail at the first trial. At each succeeding trial he can easily notice his improvement in manipulation and a corresponding gain in the results. He should be able to bring the performance of the objective up to the claims made for it, if it has come from the hands of a reliable optician and should not rest until this is accomplished.

The writer has often recommended sunlight with generally successful results where ordinary means of illumination have failed. The light is of course intense and great care will have to be used to modify it by properly using the mirror, but success is often attained and then creates confidence.

It is, however, only recommended for this purpose and not for general use.

Stained Bacteria and Micrococci also make excellent objects for immersion objectives. The mode of illumination is the same as with dry objectives.

ILLUMINATION WITH CONDENSER.

Up to this point the matter of illumination has been treated in its most simple form with mirror only, as most instruments are of the simple type, but we must now consider the substage condenser, which is a most valuable adjunct to the microscope. While skillful treatment of the microscope will go far toward obtaining good results, it will in many investigations not suffice. Furthermore the condenser will give results with ease which without it involve effort and skill.

Purpose of the Condenser.—The purpose of the condenser is not only as its name implies, to condense light, but is more especially to illuminate the object with a cone of light having an angular aperture equal to that of the objective used and which is absolutely unattainable with a mirror only, as well as to provide means for controlling the amount and character of the illumination to suit the various conditions of work.

Abbe Condenser.—The history of substage condensers is very unique and interesting and shows, how from having been the subject of no end of condemnation, which for many years it

received, it is now generally accepted as a necessary adjunct to a complete outfit, in fact should always accompany an oil immersion objective. From single lenses, compound, non-achromatic and achromatic, the use of eyepieces and objectives as condensers with any number of devices for regulating the light, the generally accepted

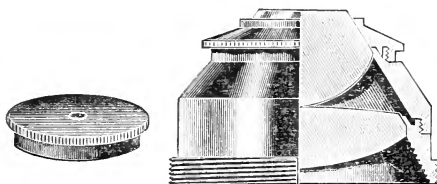


Fig. 39.

Optical part of condenser 1.20 Aperture.

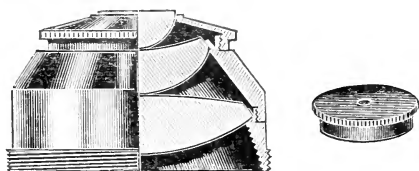


Fig. 40.

Optical part of condenser 1.42 Aperture.

forms have come to be those devised by Prof. Abbe. One of them with a numerical aperture of 1.20 consists of a combination of two lenses (Fig. 39) and the other with an aperture of 1.42 of three lenses (Fig. 40). A third is made achromatic with an aperture of 1.0 which, however, is considerably more expensive.

The most simple form, largely used for instruments for laboratory and everyday work is one which has attached to it below an iris diaphragm for regulating the amount and angle of light and to which is attached a swinging arm to receive

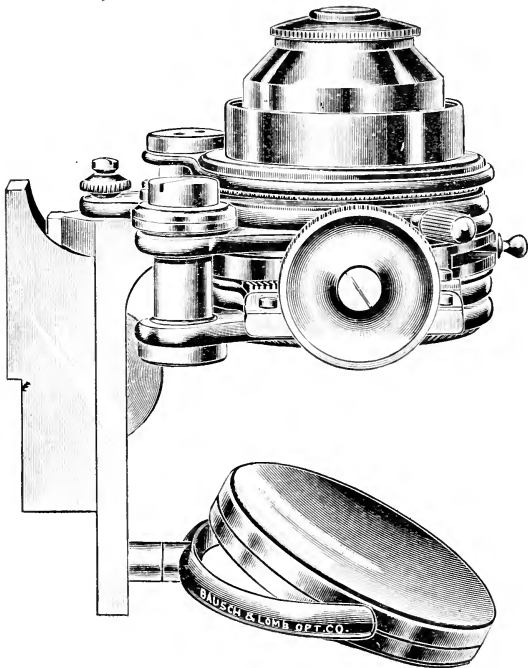


Fig. 41.

blue glass for moderating light, or stops for dark ground or oblique illumination. A screw motion gives a serviceable means of adjustment.

The most complete form is that shown in Fig. 41, which has adjustments for obtaining every modification and character of illumination, with rack and pinion for vertical adjustment and swinging out of the way the condenser and iris diaphragm if it is not desired to use these.

A condenser while useful may be abused so as to do more harm than good and we deem it proper to give some instructions in its use, which we trust may be of service.

Use only plane mirror with the condenser.

Centering the Condenser.—Every condenser should have a centering arrangement, so as to bring its axis coincident with that of the objective. In the simple form of microscopes in which the character of work is not critical, the condenser is sufficiently centered for all ordinary requirements, but even here it should be possible to center if conditions should demand it.

In the more complete apparatus a pin hole cap to aid in centering the condenser and fitting over the upper part of the mounting should be supplied, and after focusing upon the opening in the cap with the low power objective, bring into the center of field with the centering screws. Closer adjust-

ment can be made after the high power objective is attached.

In the simple form, pin hole cap is usually not provided. To verify correct centering two easy methods may be followed.

1. *Use a 2 inch objective and focus through the condenser onto the diaphragm, which is reduced to its smallest opening.*

2. *Use a $\frac{2}{3}$ or $\frac{3}{4}$ inch objective and bring to its focal point; remove eyepiece and look through the tube, keeping the eye in center, when the small opening of diaphragm will be seen sharply defined.*

This opening in diaphragm may be enlarged to the full aperture of objective so that the margin of opening in the diaphragm will just coincide with that of the back lens of objective.

Centering the Illumination.—This is fully as important as centering the condenser, for it is of little avail to have a centered condenser when the projected beam of light is not centered. This depends upon the source of light and position of mirror and when it is remembered that the mirror may be so adjusted as to give all gradations of oblique illumination from the central to the limit of aperture, the necessity for accurate adjustment becomes apparent.

Attach $\frac{2}{3}$ or $\frac{3}{4}$ inch objective; open diaphragm to full extent and focus upon the minute image of

flame ; adjust mirror so that the image will be in the center of field.

To Focus Condenser.—Since the condenser is nothing more than a combination of lenses similar to those in an objective, but used in a reversed position, it has the same properties of angular aperture, focus and working distance. But, as it must work through the thickness of slide, its working distance is proportionately large to the angular aperture and owing to the variation in thickness of slides, its focus is variable, falling in some slides above and in others below its effective position. In the simple mountings the condenser is generally so placed that its upper surface is just below the surface of the stage and for the low and medium powers this will be generally found sufficiently close, in fact at this point it may often be found that the illumination is too intense and may then be reduced either by reducing the aperture of the diaphragm, or by removing the condenser farther from the object.

In all objectives with a numerical aperture less than 1.0 the condenser may be used dry, without oil. In immersion objectives the top of condenser should be brought in fluid contact with the slide. The same phenomena take place by refraction of the light from condenser through the slide, as from an object through the cover glass to the objective. To make contact, place a drop of oil

on the top of condenser, drop the slide upon the stage, first throwing the clips to one side. With immersion objectives the proper focusing of the condenser becomes a matter of nice distinction to obtain best results and can only be reliably accomplished by considerable practice and experience.

To obtain best position use a $\frac{2}{3}$ objective ; focus upon the object, after the slide has been brought in fluid contact with condenser and then adjust condenser until image of flame will fall in the same plane with the object.

Relation of Aperture of Condenser to Objective.—As has been stated the condenser may be the means of doing more harm than good, depending mainly upon the angle of illumination used. Too much illumination decidedly injures definition by obliterating detail. In the study of Bacteria and Micrococci, with which the objectives used are of wide aperture, it is sought to have them stand out boldly in a bright field, which is accomplished by bringing the diaphragm to its full aperture.

In all dry objectives the aperture of the condenser should be less than that of the objective.

Little experience is required to judge when the condenser has its proper opening. When correct, the image will stand out sharply defined without any appearance of fogginess and as the aperture is

reduced, it will be noticeable by the decrease in the amount of light. By removing the eyepiece and looking at the back of the objective the relative aperture of the condenser to that of the objective may be easily seen, as the outlines of the diaphragm are sharply defined. If the opening in diaphragm appears to have the same opening as the back of objective, the condenser has the same angular aperture. By experience the following conditions have been found to give most satisfactory results :

In oil immersion objectives on bacteria use the full aperture of condenser.

On diatoms reduce the aperture to about two-thirds opening in objective.

In histological and other dense objects use an aperture equal to about one-half the opening of back lens in objective.

Oblique Light With Condenser.—Oblique light may be obtained by setting the mirror in such a position that the light reflected from it shall enter the condenser only at one side, leaving the balance of it unused. This, however, is only advisable when the condenser mounting has no other provision for obtaining oblique light. In the mountings however, having such provision, it is obtained by reducing the opening in diaphragm so that it shall correspond in size to the back

lens of objective and then moving this opening laterally across the bottom of condenser to such a point as will bring out the structure of the object, or to the limit of the aperture of the objective. When beyond the limit of aperture the field will appear to become dark. The amount of illumination may be modified, but in a general way it may be said that the best results with oblique illumination are obtained by reducing the amount of illumination to its minimum practicable amount.

In objects with striated structure, the illuminating rays should be brought to a position at right angles to the striæ, either by rotating the object to the proper position, or by swinging the diaphragm.

HOW TO DRAW OBJECTS.

To be able to make a correct drawing of the enlarged image of an object is very important and while for some lines of work the photographic camera is called into requisition, drawings are nevertheless, largely relied upon. The apparatus requisite for this purpose is the *camera lucida* and while there is a variety of forms, all are based upon the principle of causing the image of the object to appear projected upon the paper, where it may be drawn.

The most simple form is that shown in Fig. 42.

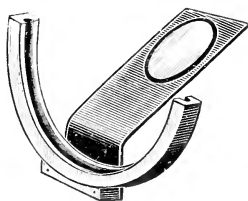


Fig. 42.

With this the microscope must be considerably inclined and the camera lucida attached to the eyepiece. The emergent pencil is reflected by the

thin film of glass into the eye and apparently projected through the glass upon the paper.

Another simple form is the *Wollaston camera lucida* (Fig. 43), which is attached to the eyepiece in the same manner as the foregoing, with the

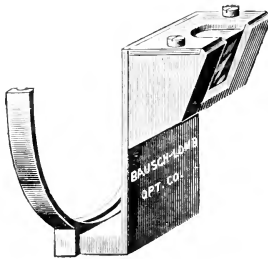


Fig. 43.

instrument also inclined. This consists of a quadrangular prism, which reflects the emergent pencil through a small opening into the eye. The eye is placed over the edge of the prism in such a manner that the rays forming the image enter only a portion of the pupil, while its other portion views the paper. In using this however, the eye must be kept quite steadily in the proper position.

While there are some other forms, they are not in general use. The very best is one designed by Prof. Abbe, and which goes under the name of the *Abbe camera lucida* (Fig. 44). It is fixed to the tube of the microscope, permitting the mounting which contains the revolving prism to be swung

back, so that the eye may be applied to the eyepiece. It is composed of two rectangular prisms, cemented together at their diagonal surfaces, but having between them a perforated film of silver.

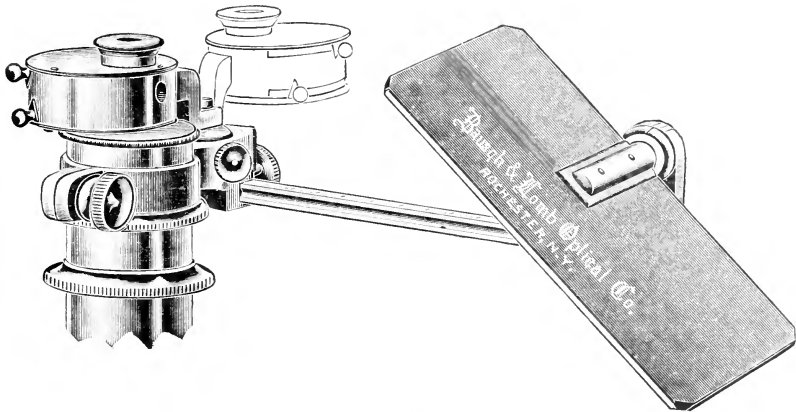


Fig. 44.

At one side of the prism is a large mirror which reflects the image of the paper and pencil onto the silvered surface of the cemented prism and this in turn to the marginal portion of the pupil, while the axial portion of the eye views the image through the perforation of the silvered surface, thus giving both views coincidentally.

A number of precautions are to be taken to obtain good results. First of all it must be remembered that if spectacles are necessary in

reading, these will be required in order to see the pencil point clearly.

The pencil point should be well sharpened in order to closely follow the minute detail.

The drawing board should be at *right angles to the axial portion* of the projected image, otherwise there will be an elongated picture.

The relative illumination of field and paper varies with magnifying power and distance of eyepiece from paper, but should be made as nearly alike as possible. In low powers the light from the objective is stronger than that from the paper and should be modified, in the simple and Wollaston forms by using the plane mirror or by covering it with white tissue paper, and in the Abbe form by the use of dark tinted glass, which is provided with the apparatus. In the higher powers the paper is brighter in which case it should be shaded by a screen placed between it and the source of light, or a substage condenser should be used to give increased illumination.

Special instructions with reference to the use of the simple and Wollaston forms are as follows :

Focus the objective upon the object ; then incline the microscope, preferably to the horizontal position ; raise the microscope by underlaying with a block or books. The size of drawing may be varied by change of eyepiece or objective, but only to a limited extent ; or by varying the dis-

tance between eyepiece and table, it being apparent that as the distance becomes greater the magnification will proportionately increase. The usual standard for distance is 10 inches, although this may be varied to suit requirements. Attach the camera lucida; place the paper upon the table and fasten with drawing tacks; apply the eye to camera lucida; refocus carefully with the fine adjustment and endeavor to obtain equally sharp view of image and pencil point. In these two forms it is quite difficult to see the pencil point clearly and some care will be required in modifying the light or shading the paper so that the image and pencil point may be sufficiently distinct at the same time to follow the outlines.

In the Abbe camera lucida the optical results are considerably better, since a direct view of the image is obtained and the equalization of illumination is easily accomplished.

While with it, the microscope is intended to be used in an upright position, the reflecting mirror is close to the eye so that, if the image is projected vertically, a portion of the field is cut off by the stage or base of the microscope and if the mirror is so inclined as to bring the full field upon the paper, it will not be round, but elongated or elliptical and thus also elongate the image. This can only be corrected by correspondingly raising one end of the drawing board so that it shall be at

right angles to the axis of projected cone, or until the field appears round. A convenient method of accomplishing this, with the further advantage of being able to elevate the drawing board and thus produce a variation in the size of drawing, is the drawing table made by Bausch & Lomb Optical Co. Care must also be used in attaching the camera lucida to the tube, so that the opening in the prism shall be in the optical axis. This can be accomplished by the centering screws and observing whether any of the field is cut off. If the field appears smaller with the prism than by directly looking into the eyepiece, the prism is too close, or too distant from the eye lens and should be properly adjusted.

No particular skill is required in drawing as it is simply a question of copying, but the lines should be light so that any irregularities may be corrected after the work with the camera lucida is completed. To determine the standard distance of 10 inches, measure from the optical axis to the axis of mirror and from this point to drawing paper.

If it is desired to determine the amount of enlargement of the image on paper, this can be done by replacing the object by a stage micrometer and drawing its spaces over the object. If the divisions or spaces are 0.01 mm. and it were found that 10 spaces covered the object and the actual

measurement was 30.0 mm. the enlargement would be 30 times 100 divided by 10, or 300. If a standard of 10 inches is maintained in all drawings and the amount of magnification with certain objectives and eyepieces be previously determined by means of the stage micrometer, a standard is established for each and further measurements will not be required. If however, variations from this standard distance are made, the actual magnification should always be determined.

To Determine Magnifying Power.—While the magnifying power may be known from tables accompanying the microscope, these are only approximate, as there is more or less variation in eyepieces and objectives and furthermore the microscope may be used under different conditions. There are three requisites.

A camera lucida.

A stage micrometer ruled in divisions of inches or millimeters.

A pocket or foot rule in inches or millimeters, according to the stage micrometer which is used.

If a low power objective is used, place the stage micrometer with divisions of 0.1 mm. or 0.01 inch upon the stage and focus. After attaching the camera lucida, place the microscope in exactly the same position as for drawing, maintaining the standard distance of 10 inches from optical axis to

drawing paper ; mark the spaces of the micrometer as projected upon the paper and determine how many of the divisions of the rule are contained within one or more spaces on the paper. If the values are in millimeters and it should be found that 25.0 mm. on the rule are contained in one space on the paper, the magnification would be 250. If 18.6 on the rule are contained within three spaces on the paper, the magnification would be 63 times.

To Measure the Size of an Object.—One of the most valuable possibilities of the microscope is to be able to accurately measure the actual size of a minute object. Computations may be made in inches or millimeters by figuring 25.4 mm. equal to 1 inch. It may be done by several methods, two of which are generally followed.

The first of these give satisfactory results on coarser objects and wherever the most accurate results are not required, although it is somewhat inconvenient. The requisites are

A camera lucida.

A stage micrometer.

The object is placed upon the stage and after focusing, the camera lucida is attached and the instrument set up exactly as for drawing. On the drawing paper the outlines of object, or that portion which is to be measured are marked, without

in the least disturbing any of the conditions of tube length or distance from the paper. Remove the object and replace it by the stage micrometer, focusing only with the fine adjustment and, it may be added, there should be very little variation in thickness between the two slides. Move the micrometer so that one of its lines shall exactly coincide with one end of the drawing on the paper and then measure off how many spaces are covered by the object. Thus if 0.001 inch are the values of the micrometer spaces and the object covers one space, its size will be 0.001 inch, or covering 7 spaces will be 0.007 inch.

A variation of the distance of the camera lucida from the paper, or a change of power in eyepiece or objective does not vary the result so long as objective and micrometer are used under exactly the same conditions.

The second method is with the eyepiece micrometer which in its simple form is a micrometer mounted on a plate which is placed in the focus of the eye lens. It is usually graduated to 0.10 mm., seldom exceeding 0.05 mm., as closer lines cannot well be distinguished with the magnification obtained by the eye lens. The plate is dropped onto the diaphragm, where it is in focus, or enters slots which are provided in the tube of the eyepiece. A better form however, is the eyepiece micrometer (Fig. 45) in which the eyepiece and

micrometer form a complete apparatus and a lateral adjustment of the scale across the field is given by a screw.

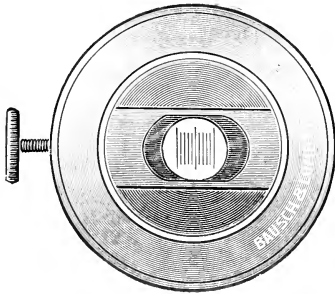


Fig. 45.

In either of these forms the ruled lines appear to lie directly on the image of the object, but while we have on the one hand the actual value of the micrometer we have on the other only the image of the object. The valuation of the eyepiece micrometer in the value of the stage micrometer must be first determined. While the optician can do this, it should be done by the manipulator on account of the varying conditions of tube length, power of objective, etc. A stage micrometer divided into the same spaces as the eyepiece micrometer is necessary.

Focus the eye lens on the eyepiece micrometer and the objective on the stage micrometer, being careful to bring the first line of the former coinci-

dent with a line of the latter, using care to see that they are parallel. As the lines of the stage micrometer will appear to have a certain amount of thickness, make the first line of the eyepiece micrometer correspond with one edge of a line on the other. Now read off how many of the lines are contained in one space of the stage micrometer and note this. We will assume that it is 8 divisions. Replace the stage micrometer by the object to be measured and bring one edge of the object coincident with the first line of the eyepiece micrometer, being careful to leave all the conditions unchanged. Note how many divisions are required to cover the object and divide by the figure first obtained with the stage micrometer. Thus, if there are 40 spaces which we know are 0.1 mm. divisions the real size of the object will be 0.5 mm., or forty tenths divided by eight.

If measurements are made under exactly the same conditions of tube length, with same objectives, it will be unnecessary to repeat the operation with eyepiece and stage micrometer, as the proper ratio may be marked on a card, as it remains constant.

The most efficient apparatus however, for obtaining accurate results is with the *filar* or *screw micrometer* (Fig. 46). This consists of a metal case to the upper surface of which is fitted an adjustable Ramsden eyepiece. Within is a frame carrying

one or several delicate spider lines, which are moved across the space to be measured by an

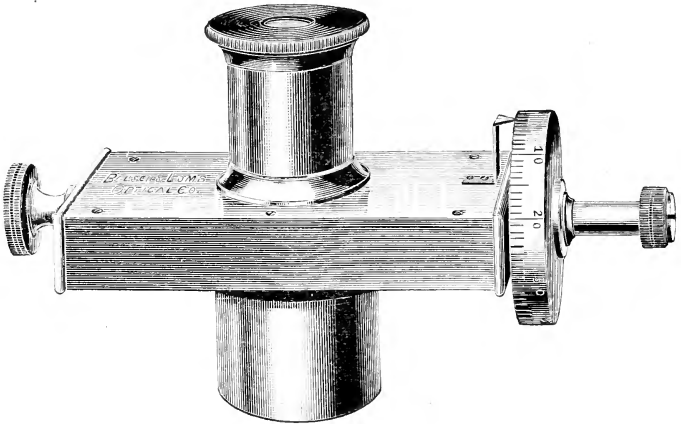
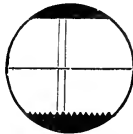


Fig. 46.

accurate screw of either 0.5 mm. or $\frac{1}{50}$ inch pitch, which has at its end a graduated disk divided in 100 or finer spaces, thus giving a definite value for



Field of Large Filar Micrometer
showing cross hairs and recording comb.

each space. An adapter is also provided for attaching to the tube of microscope.

TO SELECT A MICROSCOPE.

When a person has concluded to obtain a microscope, a suitable selection is a matter of considerable importance to him. The varieties are innumerable, prices without end, all sorts of claims made for them.

The variety of special lines of investigation involves nearly as great a variety of requirements. The amount of money to be expended ; what shall be the stand ; what the objectives ; shall the entire outfit be purchased at one time or little by little ; are all questions of paramount importance, which the writer does not expect to solve, but hopes to give sufficient information so that a more intelligent selection may be made than might probably be done otherwise.

If one has a friend or teacher, who is generally accepted as an authority, it will be well to consult him or her and obtain suggestions as to the most suitable selection for the intended work and such advice will always be gladly given. Or, if advice is asked of a reputable firm, the writer is convinced that it will be honestly and disinterestedly given.

When means will permit, the outfit for immediate requirements should be obtained complete and as Prof. Gage says, "the best that can be afforded should be obtained," and further, "even when all the optical parts cannot be obtained in the beginning it is wise to secure a stand upon which they may all be used when they are finally secured." The writer agrees entirely with this advice. Even though the stand be plain, it should be good, with the necessary adjustments and capable of receiving and fully utilizing such further accessories as may be obtained later on.

Stand. American or Continental?—First of all, choice will have to be made between the two types and while one's sense of the æsthetic may be a factor it is mainly the practical utility which must govern the decision. Whether large or small must largely be determined by the future use to which it is to be put. One rule may apply to all however, and that is, that the instrument shall be so balanced, that it will be absolutely steady during manipulation in the upright or inclined position. In general the low stand is preferred as it permits of resting the arms upon the table while moving the object and the comfort of looking through the tube whether the instrument be upright or inclined.

Tube Length.—In the matter of tube length the optical results are the same in both, so that a

conclusion must be reached in so far as it effects the height of the instrument.

Base.—The base is an important feature and while not over heavy should insure steadiness by the proper form and disposition of metal; it should not rest on more than three points, with the rear one fairly distant from the pillar.

The Joint for Inclination of Arm.—This, without question is an advantage and while it is an inexpensive addition it will add considerably to the comfort of working and should invariably be present, if pecuniary means do not absolutely prohibit it.

Coarse Adjustment.—Almost all instruments for reliable work are provided with both fine and coarse adjustments. The choice of the latter lies between the sliding tube or rack and pinion. The former has only the advantage of economy and is a decided disadvantage in the hands of students in injuring objectives and preparations. Further than this, it is almost impossible for the maker to center the nosepiece with the tube, so that a change of objective usually throws an object out of the field and requires that it be looked for anew with each change. With the rack and pinion the nosepiece has an unvarying relation to the tube and is not liable to this difficulty and offers a steady and agreeable adjustment. The advantages of the rack

and pinion seem to be generally appreciated in this country and there are few instruments sold and used without it. Dr. Stokes speaks of the sliding tube adjustment as follows :

“This is a very inconvenient and undesirable arrangement. It is awkward, since the friction is often so great that the whole stand will move out of position before the body will budge, and frequently, more frequently than not, even when the foot is heavy enough to keep the instrument firmly on the table, both hands are needed to manipulate the body. It is dangerous too, since under circumstances, the body has the obnoxious habit of suddenly slipping further than the microscopist intends, stopping only when it crashes against the slide, where it usually grinds and crunches cover glass and objective with apparently fiendish glee. A stand without a coarse adjustment by rack and pinion is a good stand to be permanently left with the optician. No fine microscopical work can be done with an instrument whose body slides through a friction collar. That arrangement may be cheap, but it is also a torment and a peril.”

Rack and Pinion.—This should be absolutely smooth with no back-lash or lost motion throughout its entire length, which can be determined by holding the main tube and working the pinion buttons very slightly but quickly back and forth. It should be perfectly fitted in its bearings, so that

there will not be the least side motion and this should be tested under the magnifying power of an objective. There should be no sensation of the individual teeth coming in contact. It is safe to assume that if the rack and pinion shows either of the above defects, the instrument is faulty in other directions as well.

Fine Adjustment.—Nothing in the microscope will cause more aggravation than a faulty fine adjustment. It should work absolutely smooth with no side play in the screw. The body should respond promptly, when moving the milled head rapidly forward and backward and should not cause any swaying of the image during observation. The micrometer screw should be back of the pinion, not at the front of the tube nor under the stage.

Metal.—Whether an instrument shall be of japanned iron or lacquered brass is probably largely determined by the amount of money to be expended. So far as the intrinsic suitability of the metals is concerned there is no difference. Brass however, offers a maker a better opportunity for displaying his mechanical skill and while it is no doubt true, that many highly finished instruments are of poor workmanship in their working parts, it is also a fact that a well made instrument is always nicely finished.

Size and Weight.—The size of instrument is worthy of consideration. If an instrument is to remain stationary in a practitioner's office or laboratory, it may be large without being cumbersome. If, however, it is intended to be carried about it should be of the smaller and more contracted pattern.

Working Space Below Stage.—Another important consideration is the space between the stage and base, or table. While it is advisable to have the stage low on account of the convenience in manipulating a slide, there should still be sufficient space for the convenient attachment of sub-stage accessories. In this respect the American instruments, whether of the American or Continental type, are superior as they are built for the better accommodation of accessories.

Stage.—A variety of stages is offered on instruments of similar construction. The plain, flat stage while preferred by some, offers no advantages over the ordinary round one, unless specially made for examining specimens on larger slides than the standard 3 by 1 inch. Those stages, covered on top with vulcanite, offer many advantages. The spring clips are usually of similar construction, although varying in detail and curves. Properly constructed clips should have such thickness of

metal and be so bent as to allow specimens to be brought under them without resistance and keep them properly in place, without too much pressure and consequent friction.

A Glass Stage and Slide Carrier may be considered a good investment, as it admits of the convenient manipulation of the slide, without the grating feeling which usually accompanies the direct movement of the slide on the stage.

The Mechanical Stage, while an absolute necessity in petrographical and other work where a systematic search, as for bacilli or in blood counting, over the entire surface of object is required, it will also be found a most useful accessory. The obstacle of considerable cost which prevailed until the present time, is now removed and good mechanical stages may be obtained at a very reasonable cost. They are supplied in two forms,

Fixed mechanical stage, in which the mechanical movement is built on a stage.

Attachable mechanical stage, which can be attached to the Continental stands having plain stage. This has advantages, since it may be removed, leaving the stage plate free but, of course, cannot be revolved. Either form involves the most delicate work and while the parts are necessarily small should be built with a view to strength and durability. They should work with the

utmost *precision* and *smoothness* and with *absolutely no lost motion*.

Revolving Stage.—This is also a great convenience in all work, while being a necessity in some directions, and when provided with centering screws may be used to some extent as a mechanical stage with only a limited movement however. It should work freely with the rolling motion of one finger, without any side play and without throwing the object out of the plane of focus during revolution.

Substage.—This is an absolute necessity in a modern microscope, except perhaps for students use in primary work. It should have a vertical adjustment and preferably with rack and pinion. If possible select the complete substage attachment.

Substage Condenser.—If means will permit, purchase this, as it is in all work most convenient and in some directions, like bacteriology, absolutely necessary. The Abbe condenser is the cheapest form giving good results and one with numerical aperture of 1.20 is sufficient in all cases unless oil immersion objectives of the greatest aperture are used.

Objectives and Eyepieces.—It is hoped that the information given of the various qualities in an objective will aid to make a suitable selection of

the optical parts. Since the stands have been classified as of long and short standard tube lengths, the first quality to look for is, after the stand has been selected, the suitability of objective and eyepiece to it and to the work. As a variety of powers is obtained by a suitable combination of eyepieces and objectives and while power in itself can be obtained by increasing the power of eyepiece, this is not advantageous as we have shown.

Eyepiece.—Select the Huyghenian eyepiece and of no higher power than $\frac{3}{4}$ inch. In catalogues many outfits are made up of one eyepiece and two objectives, but this is only for economy; it is always advisable to select two eyepieces, preferably the 2 inch and 1 inch and insist that they be parfocal, as this will be found extremely convenient and will not disturb the optical standard length. If for any work $\frac{1}{2}$ inch or higher powers are desired, the solid eyepiece may be recommended. With the apochromatic objectives use the compensating eyepiece only. Every eyepiece should be marked with its equivalent focus.

Objectives.—For all ordinary student and professional work, not involving bacteriological investigations, the $\frac{3}{4}$ inch 0.24 N. A., and $\frac{1}{2}$ inch 0.62 N. A. for long tube; and $\frac{2}{3}$ inch 0.25 N. A., and $\frac{1}{8}$ inch 0.82 N. A. or $\frac{1}{8}$ inch 0.85 N. A., for short tube have generally been accepted as best suited.

Bacteriological investigations absolutely require an oil immersion objective in which the $\frac{1}{2}$ inch of 1.32 N. A. is generally employed.

Botanical work necessitates a 2 inch or 3 inch in addition to the regular outfit.

Objectives of Wide Aperture.—It will be noted that objectives of the lowest price and lowest in the scale of efficiency have been recommended as ample for ordinary use, but it is well to bear in mind or study the advantage, which is obtained by objectives of larger angular aperture. These advantages are absolute and unquestionable, but whether commensurate with the additional pecuniary outlay, must be left mostly to the judgment of the purchaser. That he may be somewhat guided, we may say that the selection of higher or highest grade objectives is not by any means exceptional, but general, and would undoubtedly be more common but for the barrier of expense.

In these days of competition, prices alone are too often made the object of inducement, without any reference to quality. Be distrustful of all such objectives and if contemplating their purchase, always reserve the right of having them examined by an expert. Have a distrust especially of all "nameless" objectives. It is safe to assume that if the maker cannot attach his name he is doubtful of their quality.

It is sometimes found that dealers offer the same objectives of different quality at different prices. Too great care cannot be observed in such cases, as the very fact of the admission of a difference in quality indicates that they are made by an unreliable maker. This mode of offering objectives was in vogue many years ago when the principles of optics and facilities for making were limited and when a higher price was asked for those which might be termed a happy combination. There is no excuse however, at the present day, for anything of this kind, because every conscientious optician has his standard for every objective.

In purchasing a microscope a beginner may be easily misled by the enticing appearance of an object, which may be due not so much to the instrument as to the object itself and if the optical parts are inferior, it will require but a short experience to become convinced of it—usually as soon as a comparison can be made with reliable work. The investment in one of these objectives is not only a source of disappointment, but usually proves to be a pecuniary loss, as it is generally followed by a fresh outlay in responsible work.

It is of ordinary occurrence that such objectives as have been spoken of are sent to the writer's firm with the request to examine them and rectify the faults ; but an examination almost invariably proves that the cost of doing so is considerably

greater than purchasing a new objective of the same power and it would not even then be equal to the latter.

Accessories.—We have already stated in the body of this book which accessories are considered useful. Some of them are absolutely necessary in some special lines of work, in which case however, the student is generally conversant with the requirements and may make a suitable selection, but for all general purposes some accessories are necessities where others are only conveniences and we append a list of such which, unless prohibited by necessity, should accompany each outfit.

Abbe substage condenser, preferably the complete substage attachment giving all adjustments.

Double, triple or quadruple nosepieces according to the number of objectives accompanying the microscope.

Abbe camera lucida.

Revolving or attachable mechanical stage.

Eyepiece and stage micrometer.

Mounted objects, Proboscis of Blow-fly and *P. angulatum*, dry.

Pocket magnifier, preferably Aplanatic or Hastings triplet.

Cover glass gauge.

Flat-wick oil lamp.

Dissecting stand or dissecting microscope.

Besides these there are other requirements such as slides, covers, mounting media, forceps, etc., the necessity of which, however, can be better determined from books devoted to this purpose. There are other articles which in some directions are necessities, but are general conveniences, among which may be mentioned :

Adjustable drawing table.

Polariscope.

Photomicrographic camera.

Live box or compressors.

Eye shade.

Turn table.

Revolving microscopical table.

Cabinet for objects.

CARE OF A MICROSCOPE.

Besides acquiring the ability to properly use an instrument with its accessories, it is important to know how to keep it in the best working condition. It may be said without reserve that an instrument properly made at the outset and judiciously used, should hardly show any signs of wear either in appearance or in its working parts, even after the most protracted use ; and further than this, every good instrument should have a provision for taking up lost motion, if there is a likelihood that this may occur in any of the parts.

Especial care should be given to the optical parts, in fact such care, that they will remain in as good condition as when first received. Accidental injury may of course occur to them, but if a systematic manner of working is followed and a special receptacle for each part is provided, this may usually be avoided.

To Take Care of a Stand.—*Keep free from dust is one of the first rules to be observed.* This may be done in a manner formerly prescribed. If dust settles on any part of the instrument remove

it first with a camel's hair brush and then wipe carefully with a chamois skin, wiping with the grain of the finish of the metal and not across it, as in the latter case it is likely to cause scratches. Keep the working and sliding parts absolutely free from dust, as this grinds and will thus soon cause play.

Use no alcohol on any part of the instrument, as it will remove the lacquer. As the latter is for the purpose of preventing oxydization of the metals, it is important to observe this rule.

To use the draw-tube impart a spiral motion.

To lubricate any of the parts, use a slight quantity of soft tallow or good clock oil, or paraffine oil.

If the pinion works loose from the jar incident to transportation or long use, which sometimes occurs to such an extent that the body will not remain in position, increase its tension by tightening the screws on pinion cover.

In using a screw-driver, grind its two large surfaces so that they are parallel and not wedge-shape and so it will exactly fit in the slot of the screw-head.

In inclining the stand always grasp it by the arm and never by the tube, as in the latter case it may loosen the slide or tear off some of the parts.

When repairs or alterations are necessary, always have these made by the manufacturers, who can,

from a system of duplicated parts, not only do it cheapest, but best.

To Take Care of Objectives and Eyepieces.—The utmost cleanliness must be observed with objectives and eyepieces. When indistinct, dark specks show in the field, the cause may usually be looked for in the field lens of the eyepiece, although sometimes in the eye lens also. The dust may be removed by a camel's hair brush, but when this is not sufficient use a well washed piece of linen, such as an old handkerchief. From its fine texture chamois skin is desirable, but as it is fatty it should never be used until after it has been well washed.

The same method applies to cleaning objectives. *Clean an immersion objective immediately after it has been used*, first by removing the fluid with a moist linen and then by using a dry piece, or by means of Japanese lens paper. *Never separate the systems of objectives*, even if they can be unscrewed by the fingers; it is always dangerous as they are liable to become decentered.

Keep the objectives especially in a place where they are not subject to extreme and sudden changes of temperature, as the unequal expansion and contraction of glass and metal may cause the cement between the lenses to crack. Also keep them from direct sunlight.

Screw them into the nosepiece and unscrew, by grasping the milled edge.

Avoid any violent contact of the front lens with the cover glass. Usually the latter suffers, but it is as liable to injure the former.

Above all, the owner should make it a rule that no one except himself handles the microscope and accessories. One person may be expert in the manipulation of one instrument and still find it difficult to work with another. The fine adjustment particularly causes the greatest difficulty, as in some instruments it corresponds with the movement of the micrometer screw, while in others it is contrary and thus the objective as well as the object is endangered.

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