## VIII. — The Relation of Aperture and Power in the Microscope.\* By Professor Abbe, Hon. F.R.M.S.

## (Read 10th May, 1882.)

## I.-General Considerations as to Wide and Narrow Apertures.

The question of the relative values of high and low apertures has been much obscured by the one-sidedness with which it has been One party of microscopists-the "wide-aperturists"treated. having recognized that high apertures are capable of exhibiting minuter details than low apertures, conclude therefrom that all microscopical work must be done with very wide apertures, and that low-angled systems are worthless. Another party, relying upon the fact that there are many cases in which low or moderate apertures perform decidedly better than wide ones, generalize this experience and deny that there can be any essential benefit in very wide apertures, asserting that all observations, with the possible exception of resolving diatom striæ, can be done as well with lowangled objectives. The premises of both these views may be said to be true, but true under conditions only; and by disregarding these conditions both parties arrive at conclusions which are equally remote from a proper estimation of the requirements of scientific work with the Microscope. My view of the question † is based on the following considerations :----

1. Every given degree of minuteness of microscopic detail requires a given aperture in order to obtain a complete (or perfect) image, i. e. an image which is a *true* enlarged projection of the structure, exhibiting all elements in their true form and arrangement. The minuter the dimensions of the elements the wider an aperture is *necessary*—the larger these dimensions the narrower an aperture is *sufficient*. Structures whose smallest elements are measured by considerable multiples of the wave-lengths of light are perfectly delineated with low or very moderate apertures, and their examination with wide apertures does not improve their recognition. ()n the other hand, if we are dealing with objects whose dimensions (or structural elements) are equal to a few wave-lengths only, even the

\* The paper (received 8th April) is written by Professor Abbe in English.

<sup>+</sup> As some suggestion appears to have been made when the above paper was read as to my views having undergone a change, I beg to remind my readers that the views above explained are those which I have professed since 1873—the date of my first paper on the subject. My advocacy of wide apertures for minute objects appears to have been interpreted as an advocacy of wide apertures for all purposes – a misapprehension which I am at a loss to account for, as nothing I have ever said or written could justify any such a supposition. All the catalogues of Mr. Zeiss issued since 1872 give practical evidence of

All the catalogues of Mr. Zeiss issued since 1872 give practical evidence of this, as the objectives there specified (and stated to be constructed according to my principles and under my direction) include no low and medium powers, except with low or very moderate apertures.—E. A. widest apertures hitherto obtained will not afford complete or strictly true images, but will show these objects more or less incomplete or modified. This general principle holds good in regard to objects of every kind, regular or irregular, isolated particles or composite structures, because the physical conditions of microscopical delineation are always the same.

The obvious inference from this principle is that the widest possible apertures must be used for the observation of objects or structures of very minute dimensions, low and moderate apertures for relatively large objects.

It may perhaps be said that the objects of microscopical research do not justify such a distinction of large and minute, since the works of nature are always elaborated to the minutest details, all coarse objects being composed of smaller elements, and these of still smaller ones, &c. This is quite true in regard to the objects considered as natural things, but not as objects of scientific research. The interest of research is not always directed to the ultimate elements, but is as often confined to the consideration of the coarser parts, and in such cases the observer is not only allowed but sometimes compelled, to disregard everything which is not connected with the scientific aim of his investigation. To observe every object in nature throughout, from alpha to omega, is the privilege of dilettante microscopy only, which has no distinct aim. There are many lines of the most valuable scientific research (e.g. the greatest part of all morphological investigations) which have not to deal with very minute things. This kind of work can be completely done with low or moderate apertures.

To recommend the application of wide-angled objectives for every branch of microscopy, as has been, in fact, done by excited wide-aperturists, is no more to be supported than it would be to recommend the use of a magnifier to a painter for inspecting the tree which he proposes to delineate.

According to what has just been said, the only benefit of greater aperture is that it is capable of delineating *minuter* things. Now minute dimensions require high amplifications in order that they may be enlarged to a visual angle sufficient for distinct vision. Low figures of amplification cannot render visible (at least not distinctly visible) details which are beyond a certain limit of minuteness. Even if they are delineated by the Microscope they would remain hidden to the eye for want of sufficient visual angle. It follows therefore that wide apertures will not be utilized unless at the same time there is a linear amplification of the image, at least sufficient for exhibiting to the eye the smallest dimensions which are within the reach of such an aperture. On the other hand, a high amplification will be useless if we have small apertures which delineate details of dimensions only capable of being distinctly seen in an image of much lower amplification. We have here an empty amplification, because there is nothing in the image which requires so much power for distinct recognition. In the first case (deficiency of power) the large aperture cannot show more than a smaller one; in the other case (deficiency of aperture), the high amplification shows no more than a lower would do. Consequently:—

Wide apertures when high amplification is required; low or moderate apertures when low or moderate amplifications are sufficient or cannot be overstepped.

2. The utilization of a given aperture depends in principle on the amplification of the ultimate image which is projected by the entire Microscope to the observer's eye. Now one and the same amplification may be obtained in very different ways since it is the resultant of three distinct elements, (a) focal length of the objective, (b) focal length of the ocular, and (c) length of the tube. Any definite number of diameters (say 1000) can be obtained with a low power objective (say a 1-inch) as well, from a mere dioptrical point of view, as with a higher power (say  $\frac{1}{8}$ -inch), by applying a sufficiently deep eye-piece and a sufficient length of the tube. It is, however, well known that there is a great difference in the optical qualities of images which are produced under these different conditions. Forcing a high amplification from a low-power objective is always connected with a considerable loss of sharpness of definition of the image, owing to the magnification of the residuary aberrations, which are inherent even in the most finished construc-It is, therefore, a well-established practical rule that a certain tions. amount of amplification requires a certain power of the *objective* higher amplification a higher power (shorter focal length)-in order to obtain the image under those favourable conditions which are necessary for their full effectiveness. This considered, the inference of the foregoing paragraph may be expressed in these terms :-

Wide apertures with objectives of short focal length; low and moderate apertures with objectives of low and moderate power.

As a detailed discussion of this subject will be found in the second part of this paper, it will be sufficient here to point out some notable facts of experience by way of example only.

With objectives of say 1 inch, and  $\frac{1}{2}$  inch, focal length, the lower and medium eye-pieces in use will yield 40-80 and 80-160 diameters only. In order to obtain 150 and 300 respectively, very deep oculars (or an extra length of the tube) would be required. So far now as such objectives are intended for the lower powers mentioned above, an aperture of about 0.15 (18°) in the case of the 1-inch, and of 0.3 (35°) in the case of the  $\frac{1}{2}$ -inch, are at all events more than sufficient for showing every detail which can possibly be recognized by the eye under these amplifications, and therefore wider apertures are useless. In point of fact, no observer will see anything more or anything better with similar objectives of say 0.40 (48°) and 0.75 (96°) respectively, than with the narrower angles indicated above, as long as the low and medium oculars are in question only. These latter apertures would require for their full utilization, i.e. for convenient observation of the minuter details which are within their reach, amplifications of much more than 150 and 300 diameters. With well-made objectives of those apertures, such figures may be realized indeed, and details may be shown by means of deeper eye-pieces, which remain quite invisible with the lower angled systems; but no microscopist can deny the inferior quality of the images obtained in this way if compared to those of equal amplification, which are obtained with these same apertures when the objectives have double the power and the oculars the half only. Structures of so simple a composition as diatom striæ may perhaps be tolerably displayed under such forced amplifications of low-power objectives, but with objects of somewhat irregular and complicated structure the deterioration of the image attendant upon a considerable enlargement of the residuary spherical and chromatic aberrations by deep eye-pieces, becomes at once obvious even with the most finished objectives. In point of fact, no experienced histologist will ever use in ordinary work even an ocular amplification of the amount necessary for obtaining 100 diameters from a 1-inch objective or 200 from a 扌-inch. He would be unwise if he troubled himself with inferior images whilst good images of the amplifications required could be obtained with equal, or even greater, convenience with objectives of the same apertures but half the focal length.

The above is an example of waste of aperture, or lack of useful power; waste of power and lack of aperture are exemplified by every objective of excessively short focal length, e. g.  $\frac{1}{50}$  inch. Such a lens, even if immersion, cannot be made with an aperture of much greater numerical value than  $1 \cdot 0$ , in consequence of the technical obstacles arising with such very short focal lengths. Now the limit of an aperture of that amount is entirely exhausted, at all events with a power of 1000 to 1200 diameters, inasmuch as nothing of the real attributes of an object can be seen with that aperture under a higher amplification, which could not be as well recognized under A  $\frac{1}{50}$ , however, will yield 1500-2000 diameters with the lower. the lowest eye-pieces which are usually employed. The lowest attainable power is therefore an empty power already, and every useful amplification available with the aperture in question could be obtained under favourable conditions and with much less inconvenience by an objective of half the power, or even less.

3. The preceding shows that wide apertures can only be utilized in the observation of minute details, under high amplifications obtained with objectives of short focal length. Wide apertures are therefore useless when those conditions are not fulfilled, because in this case the same result could be obtained as well with low-angled systems. But as abundance *primâ facie* is no detriment, the foregoing considerations do not enforce any positive objection to the use of wide apertures for every kind of work. There are however other points of view from which it becomes obvious that the application of wider apertures than can be utilized is not merely superfluous but is a decided disadvantage, inasmuch as they prevent the utilization of some really valuable benefits which are the privilege of low and moderate apertures.

The first disadvantage results from the reduction of the depth of vision (or the "penetration" of the Microscope) which is connected with wide apertures. I have given in another place\* a discussion of the circumstances on which penetration depends, and the formulæ which afford an approximate numerical estimation of the depth of vision in microscopic observation. These theoretical suggestions show (in accordance with the experience of practical microscopists) the reduction of penetration with increasing aperture under one and the same amplification, and especially when the amplification is not restricted to very small figures. Now there are many objects of microscopical research which do not require. and, indeed, do not even admit of high powers, but demand for effective investigation as much penetration as possible. This is always the case where the recognition of solid forms is of importance, and therefore a distinct (at least, a tolerably distinct) vision of different planes at once must be possible, whether the observation is assisted by stereoscopic devices or not. The greater part of all morphological work is of such a kind, and in this line of observation therefore a proper economy of aperture is of equal importance with economy of power.

Whenever the depth of the object under observation is not very restricted, and it is essential that the depth dimension shall be within the reach of direct observation, low and moderate powers cannot be overstepped, and no greater aperture should therefore be used than is required for the effectiveness of these powers—an excess in such a case is a real damage. High powers and correspondingly wide apertures are restricted to those observations which do not require any perceptible depth of vision, i. e. to two different cases (1) when the objects are quite flat or exceedingly thin; (2) when preparations of greater depth are sufficiently transparent to admit of an *indirect* recognition of their solid structure \* See this Journal, i. (1881) p. 689.

by means of successive optical sections through successive focussing of different planes. For the latter method of observation the loss of penetration with increasing power and aperture is no drawback, but rather an advantage, because it enhances the distinct separation of the sectional images at successive foci. A disregard of these natural restrictions in the use of wide apertures is obviously the origin of the opinion that aperture per se is antagonistic to good definition. It is quite true that there are many even very delicate objects which are much better seen under a given amplification with a system of very moderate than with one of very wide aperture, the former giving a clear view of the whole structure, the latter showing perhaps some distinct points, but as a whole veiled in haze. Provided, of course, that we have well-corrected objectives, the fault here is not on the part of the lens, but on the side of the object, which requires for proper recognition a greater range of depth than is reconcilable with a wide aperture. The theoretical suggestion which has been brought forward in support of the notion that different parts of the clear area of an objective produce dissimilar images, and that therefore the resultant image must show increasing confusion with increasing aperture, cannot apply to the delineation of a plane object. In a well-corrected objective the partial pictures received through the various parts of the aperture-area are always strictly similar so far as one plane of the object is concerned. The confusion suggested is nothing else but confusion of the images of different depthslack of penetration, but not lack of "definition" in any reasonable sense of that term. Provided the objectives are properly corrected and the objects are fit for the delineation of an image, undisturbed by interfering confused images from other planes, the "defining power" of an objective is always greater with greater aperture for every kind of objects, inasmuch as under all circumstances the wider aperture admits of the utilization of higher amplifications than can be obtained without perceptible loss of sharpness (with the same objects) by lower apertures.

There is therefore no drawback in principle to the use of a large aperture when the objects are suitable. But the considerations above lead to the conclusion :—

Wide apertures (together with high powers) for those preparations only which do not require perceptible depth of vision, i.e. for exceedingly flat or thin objects, and for transparent objects which can be studied by optical sections. Moderate and low apertures when a wide range of penetration cannot be dispensed with.

4. There is still another point of view, and one of special practical importance, which shows the positive damage connected with the use of *unnecessarily* wide apertures. The increase of

aperture is prejudicial to the ease and convenience of microscopical work in two essential respects.

1stly, It necessitates a progressive reduction of the working distance of the objective. Owing to the rapid increase of the anterior aberration with increasing obliquity of the marginal rays (particularly in the case of dry lenses), perfect correction of a system cannot be obtained unless the layer of low refraction between the object and the front lens (i.e. the working distance) is reduced to a certain fraction of the focal length of the system. which fraction is necessarily diminished in a rapid proportion as the aperture becomes greater and greater. Whilst there is no objection to retaining as working distance  $\frac{1}{10}$  of the focal length for an aperture of 30°, if the aperture is  $60^{\circ}$  not more than  $\frac{3}{10}$ can be allowed, and with an aperture of 116° really good correction is not reconcilable with a working distance exceeding  $\frac{1}{10}$  of the focal length. It is therefore an obvious disadvantage to use aperture angles of 60° and of 116°, when the power which is required or available can be obtained with 30° and 60° respectively.

2ndly, Increase of aperture is inseparable from a rapid increase of sensibility of the objectives for slight deviations from the conditions of perfect correction. The state of correction of an objective depends on the thickness of the refracting film between the radiant and the front lens, represented by the cover-glass and that portion of the preparation which is above the actual focus. This is a variable element independent of the objective itself. In order to avoid large aberrations which must result from the change of that element, its variation must either be confined to narrow limits or must be compensated for by a corresponding change in the Now there is a great difference in regard to this objective. requirement between the objectives of low and of wide aperture, in particular with the dry system. An objective of a few degrees is almost insensible, it may be focussed to the bottom of a trough of water without any loss of performance. With 30° differences in the cover-glasses within the usual limits are still inappreciable, and an object may be seen at the depth of a drop hanging on the under surface of a cover-glass. With 60° a deviation of the coverglass from its standard thickness by not more than 0.1 mm., or a corresponding increase of the depth of the preparation above the actual focus, will introduce perceptible aberrations and a visible loss of definition if not compensated for. With an aperture exceeding 100° in a dry lens, the same result will arise from a change of thickness of 0.02 mm only. To preserve always the best correction in such a system would necessitate a change of the correction-collar for almost every change of focus in the inspection of successive layers, unless the preparation is exceedingly thin.\*

So far as the necessity of obtaining a certain amount of amplification in an efficacious manner requires a certain aperture, the above-mentioned restrictions and difficulties in the proper management of the objectives cannot be avoided. But all restrictions in regard to the objects, and all the trouble taken in the adjustment of the objectives, is quite for nothing when the same result can be obtained with a lower aperture. If for the sake of convenience the precautions required in the use of wide-angled lenses should be disregarded in working with the lower powers of wide aperture, the performance of such lenses is always worse than that of much narrower apertures under the same amplification. The best wideangled system, if not carefully adjusted when in use, is not better than a bad low-angled lens, for the tolerably sharp image, which could be still obtained through the central part of the aperture alone (even under the imperfect state of correction) is disturbed by the coarse dissipation of light from the ineffective marginal parts of the aperture.

The amateur who likes the Microscope for his amusement may not much object to some extra trouble connected with the use of

\* The reduction of this sensibility in somewhat large apertures is one of the great practical advantages of the immersion-method. The extreme increase of that sensibility which is met with when the aperture of dry lenses approaches the maximal value of a for air (1 N.A.), is in my opinion a strong objection to the construction of such lenses with greater apertures than 0.80-0.85. Not only in this case must the working distance be reduced to an intolerably small amount in order to obtain proper correction, but the preservation of that correction in the practical use of the systems is almost impossible, notwithstanding the correction-collar, whilst at all events the very slight benefit of optical performance is not worth speaking of in comparison to the large increase obtained with the immersion-method under so much more favourable conditions.

I need scarcely point out here that the claim of a special insensibility of certain lenses in regard to differences of the cover-glass (as has been sometimes made) is, to say the least, either great thoughtlessness or simple self-delusion, just as are similar claims of special penetration in favour of certain objectives. The aberrations in question, as well as the dissipation-circles from difference of focus, originate outside the Microscope. The particular construction of the objective cannot possibly therefore influence their amount in a cone of rays of given aperture, and the degree in which both become visible in the ultimate image of the Microscope must be strictly determined by the same elements which determine the visibility of any real object of given dimensions at the same plane of focus. There is no room left, therefore, for special properties of different constructions.

It is, however, true that an *apparent* insensibility, as well as an apparent depth of focus, is sometimes found, viz. in *badly* corrected objectives. When a system has no distinct focus at all, it is quite evident that the dissipation-circles arising from different thicknesses of the cover-glass, and from the difference of focus of different levels, may become much greater before the deterioration of the indistinct image becomes visible. Well-corrected objectives *must* be sensitive in both respects in strict accordance with their aperture so far as one and the same system of construction (dry or immersion) is in question. wide-angled low-power lenses, which he admires as brilliant specimens of optical art. For those, however, who *work* with the Microscope, the economy of labour to which they are obliged will be expressed by the rule:—

Never use wider apertures than are necessary for the effectiveness of the power, because excess of aperture is always waste of time and labour.

5. A few remarks about another point of practical interest. By those who plead in favour of large apertures in all cases, it has been sometimes suggested as a rational plan for reconciling opposite demands, to have all objectives constructed with relatively wide angles, and to reduce them by stops or diaphragms when smaller angles are desired. The greater penetration and insensibility of the low apertures may of course be attained thereby : but nevertheless this device is only a makeshift, and the result is inferior to that obtained by objectives *originally* arranged for a lower aperture. It is not merely that the stops cannot increase the working distance (which will always remain at the point corresponding to the full aperture of the lens), but that the low-angled lens which is made out of a good wide-angled one by means of a stop, is in optical respects a relatively bad objective-not nearly as well corrected as the same power would be if carefully adjusted for the lower angle. The reason will be readily understood from the following consideration.

The best correction of an objective of given aperture depends on the proper distribution of a certain amount of residuary aberration, which cannot be eliminated with our present means. The greater the aperture the more aberration must be intentionally left at the central part of the system in order to prevent an obnoxious accumulation in the marginal zone. It is obvious, therefore, that with an aperture-angle of say 90° the inmost cone of 45° cannot be so well corrected as it might be if the marginal zone could be left out of account. The effect is by no means inconsiderable, particularly in regard to the colour corrections. Owing to the chromatic difference of the spherical aberration the central portion of a somewhat wide aperture must always, even in a well-arranged objective, be perceptibly under-corrected chromatically, and in using this central part alone (the compensating influence of the over-corrected marginal zone being done away with), we have the performance of an inferior lens. In point of fact, no intelligent optician would ever make an objective of 30° aperture on the same formula as one of  $60^{\circ}$ , or one of  $60^{\circ}$  on the same formula as another of 100°, though this could be done by merely reducing the clear diameter of the lenses.

There cannot, therefore, be a reconciliation between the pleasure of exhibiting mere optical accomplishment and the interests of the working microscopist. Bad lenses will certainly not meet the demand for low and medium powers affording the utmost possible economy of time and labour in scientific work. This can be done only by systems in which all advantages attendant upon the lower apertures are fully realized by constructions specially aiming at the *best* which can be obtained under the actual conditions of the case.

The progressive increase of aperture in the higher powers, formerly within the capabilities of the dry system, and at a later period by the development of the immersion method, is, without any reasonable doubt, the most important feature of the modern advance of microscopical optics. It has rendered possible the successful extension of microscopical research to minuter and minuter objects, which otherwise would have been impossible by the ineffectiveness of all increase of amplification beyond certain low figures. The appreciation of that progress and the recognition of its true basis has led to a tendency to increase more and more the aperture of every kind of objectives. The fact has been disregarded that it is an entirely different thing whether the object is to promote the performance of the Microscope in the whole at the limits of its power, or to promote its performance for aims beyond these limits. The opinion has thus arisen that what is a benefit for one kind of lenses must also be a benefit for every other kind. Objectives of low and medium powers (1-inch to  $\frac{1}{4}$ -inch) of 15° to 60° are proclaimed at this time by many microscopists as old-fashioned and worthless things; 45° to 100°, or even 60° to 140°, are wanted for the same powers. Now as from a purely technical point of view, it is an accomplishment when the delineating power of an objective cannot be exhausted even with the deepest eye-pieces, opticians (notwithstanding the total bootlessness of such a superabundance) of course take pleasure in making such "superior" lenses, and the natural consequence is that the lower apertures required for useful scientific research are likely to be esteemed as second-rate work, no longer worthy of high technical art.

This opinion is a fatal mistake, and its practical effect, if not counteracted, will be a decided retrogradation of microscopical optics. Nobody, of course, can have the least objection to the construction of lenses of any descripition whatever for the personal pleasure of this or that microscopist. Strong opposition should, however, be made against all tendencies of captivating microscopical optics, in favour of such predilections, at the cost of the general usefulness of the instrument.

Scientific work with the Microscope will always require not only high-power objectives of the widest attainable apertures, but also carefully finished lower powers of small and very moderate apertures.