

## **On the modes of measuring microscopic objects / by Harting of Utrecht.**

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## ON THE MODES OF MEASURING MICROSCOPIC OBJECTS.

BY HARTING OF UTRECHT.<sup>1</sup>

[FROM THE MONTHLY JOURNAL OF MEDICAL SCIENCE FOR MAY 1852.]

The determination of the size of objects seen through the microscope is, on various accounts, important. For when we examine bodies whose nature and distinctive characters are only revealed to us by properties appreciable by the sense of sight, every one of these properties becomes of consequence; and more especially does the determination of the dimensions of such bodies furnish some of their best specific characters,—being among the few which are totally independent of the subjective agency of the observer.

Some have expressed the opinion, that very accurate measurements—at least in the case of objects from the organic kingdom, which vary greatly in size—are not requisite; and that it is sufficient to make an approximation, and express it by a number which, by its simplicity, may assist our conception regarding the size of the objects, without, at the same time, pretending to furnish the precise result of their actual and accurate measurements. This opinion, that micrometry is inapplicable to organic objects, is assuredly quite unfounded. It is true that of a hundred Negro skulls, or of a hundred European skulls, no two might be found of the same exact size; and if both series were compared, skull by skull, it is very likely that some of the former might be discovered larger than some of the latter. But if the mean numbers, obtained from the sum of the dimensions of each series divided by 100, were compared, undoubtedly the *mean* of the first would be found to be smaller than the *mean* of the second series. This, then, would justify the general proposition, that a Negro has a smaller skull than a European.

With the elementary parts, of which the organised tissues are composed, the case is similar. They all vary in size, but within certain limits which can be learnt; for if a sufficient number of measurements of these elementary parts be made, the mean result of each series expressed in numbers will constitute a certain value, which, abstracting the probable amount of error inseparable from such results, may be regarded as a standard, furnishing one of the best specific characters for each object.

In order that the *mean* shall be just, each separate measurement must be taken with exactitude; and this is especially necessary, when it is proposed to use these mean numbers as the foundation for observations on the progressive development of the different tissues at different periods of life.

The best micrometric method is therefore that which gives the most accurate indications. But micrometry, like microscopic vision, has its limits, and the utmost that can be expected of any micrometric method is, that it shall put it in our power to attain results, in which the probable amount of error is less than the dimensions of the smallest object which can be seen through the microscope. We

<sup>1</sup> Translated from *Het Mikroskoop*, vol. ii., p. 287, *et seq.*

shall presently see, that there are certain micrometric methods in which this extreme limit of accuracy is observed.

It is not enough that a micrometric method should give results of the highest possible degree of *relative* accuracy. These results should be *perfectly* accurate; and, in order to make them so, they should represent fractions of some generally received standard. That no micrometer has been constructed to fulfil this condition, I have satisfied myself by a careful examination<sup>1</sup> of a number of such instruments made by the best artists; and the observed differences in their indications are by no means so small that they may be disregarded without error; for they sometimes amounted to  $\frac{1}{20}$ th, and in one instance to  $\frac{1}{8}$ th, of the values of the same quantity on different micrometer scales.

But as the indications of these instruments should be comparable with each other, it is desirable to possess a method, by means of which the absolute dimensions of each micrometer scale may, with sufficient accuracy, be contrasted with those of others. If it be once discovered how much the divisions of a micrometer exceed or fall short of absolute truth, it is not difficult to reduce the results of observation to accuracy.

#### *Mode of Constructing a True Standard.*

The method of doing this was, in principle at least, pointed out more than a century ago by Jurin. It consists in winding a thin wire round a thicker metallic rod, and reckoning the number of turns and the length of the whole coil. The latter quantity divided by the former gives as quotient the thickness of the wire.

To construct a true standard by this method, it is, however, necessary to observe certain precautions.

1. The thin wire to be wound round the other must have throughout an equal thickness, and this point must be ascertained by observation. If a copper harpsichord wire be used—and this is very suitable for the purpose, and may be procured from any ironmonger—it must be considered, that the thickness measured on the convex aspect—*i.e.*, the side of the coil next the eye—is always found to be greater than the measurement taken at right angles to this surface. If, then, the thickness be determined by counting the turns of the coil, all subsequent measurements must be referred to the dimensions of the same aspect of the wire.

2. In using the copper wire, it is well to twine it round a straight piece of iron wire of considerable thickness—*e. g.*, of 6-8 millimetres (one-fourth to one-third of an inch nearly)—for the contrast in colour renders it easy to ascertain, by means of a lens of sufficient power, or, still better, by means of a compound microscope magnifying forty or fifty diameters, whether the turns of the coil are accurately in contact, on which, of course, the nicety of the resulting measurement in great part depends.

3. The winding is best performed by means of a turning-lathe, more especially as by counting the turnings of the upper wheel the turns of the coil may be best reckoned.

4. Finally, it is self-evident that the length of the whole coil must be measured with the utmost exactitude; for the degree of accuracy finally attainable by all this manipulation bears a direct ratio to the number of turns embraced in this preliminary measurement.

By way of example, I may introduce the description of a determination by myself of the thickness of a copper harpsichord wire. 1048 turns were required to cover a space of 140 millimetres—5·512 inches—which had been first care-

<sup>1</sup> The results of this examination have been published in detail in *Recherches Micrométriques sur le Développement des Tissus et des Organes du Corps Humain, précédées d'un examen critique des différentes méthodes micrométriques.* Utrecht, 1845.

fully measured<sup>1</sup> and marked off upon the thicker wire. The thickness of the harpsichord wire was consequently  $\frac{140}{1048}$  millim. = .13359 millim. = .0052595 inches; and as the total turns of the coil were counted with certainty to within one-half of a turn, the greatest possible error could not amount to more than  $\frac{1}{2000}$ th of the whole length of the coil, nor consequently affect the observed diameter of the wire by more than  $\frac{1}{14000}$ th of a millim. = about  $\frac{1}{350000}$ th of an inch. The figure found might hence be regarded as quite correct even in the fourth decimal place. The difference<sup>2</sup> between the diameters of the outer surface of the wire and of the surface at right angles to it, measured successively with a screw-ocular-micrometer, proved to be .00299 millim. = .000118 inches.

If, now, the thickness of such a wire be used as a standard, not only can we compare with it the values of the divisions of glass and screw-micrometers, but we may even assume, with much probability, that similar results are true of all micrometers made by the same workman with the same dividing-machine. If, then, those who record their micrometric observations take care to mention the source from which they obtain the instrument employed, or the standard on which their measurements are based, the true measurements may be deduced from their figures by multiplying them by a certain co-efficient, determined by previous examination of a micrometer by the same maker. Thus a careful testing of the following micrometers has taught me that their indications must be corrected by multiplying by the fraction which follows each, in order to deduce the true value as given by the metallic thread above-mentioned:—

Screw-micrometer, by Schiek, .....	0.937
Glass-micrometer, by Oberhæuser, .....	0.959
Screw-micrometer, by Powell, .....	0.967
Glass-micrometer, by Ch. Chevalier, .....	0.969
Glass-micrometer, by Dollond, .....	0.981
Screw-micrometer, by Plöszl, .....	0.991
Glass-micrometer, by Plöszl, .....	1.067

—Vol. ii., p. 293.

#### *Micrometric Nomenclature.*

[In his remarks on this subject, Harting proposes the use of a standard scale in which  $\frac{1}{1000}$ th of a millimetre, or one *micromillimetre*, is regarded as unity. This method would certainly have the advantage of enabling us to dispense with the zeros prefixed to the significant decimal places in observed measurements, and would give a more distinct relative idea of the size of an object than is attainable by the use of any other nomenclature. But in this country the English inch is usually employed as the standard, and as we write chiefly for the instruction of our English readers, who are not likely to change a system which has been long used among them, for another which, though more convenient, is neither more accurate nor more generally understood, it is unnecessary to reproduce here the arguments by which Harting endeavours to recommend his new method.]—*Trans.*

#### *On the Instruments used in Micrometry.*

The number of micrometric methods is considerable; but in the following sections we shall content ourselves with the description of those at present in general use, reserving for the last volume of this work the description of those

<sup>1</sup> To measure this I used an iron *metre*, which had been tested by Swinden at Paris, and is now preserved in the museum of objects pertaining to natural philosophy at Utrecht.

<sup>2</sup> This difference of thickness, which occasions a certain degree of flattening of the wire, is here, as in other cases,—for example, the hair of the head,—the cause of the curling (*krullen*) of such a fibre.

which have been used by only one, or, at most, by a few observers, and have consequently more of scientific than of practical interest. In the first place, then, we proceed to offer our remarks upon,—

#### *Glass Micrometers.*

The art of ruling finely-divided scales on glass with a diamond has been carried very far. Every manufacturer of microscopes supplies his customers with glass plates, divided, according to their wishes, by lines, into hundredths, five hundredths, or even smaller fractions of a millimetre. These glass micrometers differ from each other not only in the degree of minuteness of the divisions, but in the mode in which they are laid down. In some, for instance, the strokes are all of the same length, and differences in the minuteness of the divisions are only indicated by the presence of different groups, in each of which the divisions have a definite value. A better arrangement is that of a common standard measure, in which the coarser divisions are marked out by longer and projecting lines. Finally there are scales divided throughout in one uniform manner, but in which the intersections of two sets of lines, ruled at right angles to each other, form minute squares. Each of the two last constructions has its special advantages; that resembling a common rule being best adapted for simple measurements, while the square micrometer enables us with greater ease to count the number of objects embraced in a certain fraction of the field, and to prepare delineations of objects.

In proving a glass micrometer, three circumstances must be attended to:—

1. The amount of *relative accuracy* of like divisions of the scale. This is in fact the most indispensable requisite in a micrometer, and one which few possess so thoroughly as could be wished. It is not unusual to find, even in micrometers by the best makers, differences between like portions of the same scale amounting to  $\frac{1}{25}$ th, or even  $\frac{1}{7}$ th. It is obvious that such differences, however large, cannot, in consequence of the minute division of the scales, be detected by simple inspection. The counting of the divisions embraced by the field, as successive portions of the micrometer are brought into view, can only point out considerable deviations from accuracy; for the extreme margins of the field can but seldom fall accurately upon the lines. To ascertain small differences exactly, the divisions of the micrometer must be each separately measured by one of the most accurate micrometric methods, to be presently explained.

2. A second circumstance to be taken into account is the *absolute length* of the scale laid down on the glass. That the length attributed to it by the manufacturer cannot, without thorough testing, be regarded as correct, we have already (p. 3) had occasion to remark. If we have procured a metallic wire of known dimensions, by the process above explained, a portion of it may be laid on the micrometer, and it may then be seen how far its breadth corresponds with the intervals between the ruled lines. However, in this case too it will be found that the edges of the wire seldom coincide with the lines denoting certain dimensions; and it is consequently in general better, when we desire to contrast the size of the wire with the divisions of the scale, to make use of comparative results of great accuracy, obtained by different methods.

3. The thickness, too, of the lines ruled on the glass must be taken into account. In a stage micrometer these lines should be as delicate as possible, and should have a smooth, not a splintered, outline. If the micrometer be intended for an eye-piece scale, the lines ought to be, and must be, considerably stronger, so as to make an impression on the eye when viewed through the microscope.

In measuring the intervals, it must be recollected that each extends, not between adjacent margins of two lines, but between their centres, or between their respective right or left margins.

Glass micrometers may be applied to the measurement of objects in various ways. In the first place, the object may be laid on the glass plate, and both may then be brought simultaneously into the field of the microscope. This method is objectionable on several accounts. For, firstly, it can only be accurately applied to dimensions not less than the smallest divisions of the scale; and

as from physical causes the divisions cannot be carried beyond a certain degree of minuteness, it is in vain to seek, by this procedure, such delicate measurements as are otherwise attainable. Besides, the results of this method are affected by all the faults in the construction of the micrometer; while, in most instances, and especially when high powers are used, the object and the lines (which never can be in the same precise plane) cannot be both exactly defined at the same time.

The last objection is not applicable to another mode of using the glass micrometer, consisting in adapting to opposite sides of an eye-piece two screws, terminating in fine points, which project into the field of vision, and can be approximated to, or removed from, each other. In measuring an object, the points are screwed apart till the margins of the object correspond with their points; the object is then withdrawn, and a glass stage-micrometer substituted, in order to measure the distance between the points.

This method is somewhat preferable to the former. Not only are the margins of the object, the divisions of the scale, and the screw-points, seen with equal distinctness, but by bringing successive portions of the scale into the field, and taking the mean of the indications observed, the error in the workmanship of the micrometer may be made to vanish. An objection of some consequence, however, is, that time is lost in constantly shifting the object. Besides, the limits of possible accuracy are as speedily reached by this as by the former method. The finest micrometric divisions on glass which the best microscopes yet made can distinguish are those of the ninth group of Nobert's test-plate,<sup>1</sup> in which the intervals between the lines measure  $\frac{1}{1520}$  millimetre = '000026 inches. This, consequently, is the utmost limit of accuracy attainable by the glass micrometer, and it may even be assumed that the diffraction of the extreme rays will prevent this degree of exactitude from being easily reached.

But the glass micrometer may be used in another way, which is in some respects preferable to the methods already described. Instead of being used as a stage-object, it is introduced into the ocular. A Ramsden eye-piece<sup>2</sup> is the best, for when the micrometer is placed between the lenses of an ordinary Huygenian or negative eye-piece, its image is too much distorted by spherical aberration.

The first advantage of this method is, that in taking measures of the same delicacy by it, one may employ a glass scale more coarsely divided than when a stage micrometer is used. The reason is, that in the former case the divisions of the scale are magnified only by the eye-piece. If this magnifies *ten diameters* and the micrometer is divided into *twentieths of a millimetre*, then each division projected upon a surface at the usual distance of distinct vision (ten inches) will be equal to *half a millimetre*, and if the magnifying power of the whole microscope at the same distance be *five hundred diameters*, then will the value of each division on the field be  $\frac{1}{1000}$ th of a millimetre. Now, as greater magnifying powers and more finely divided micrometers can be used, it is not difficult to measure in this way, directly and accurately, spaces of  $\frac{1}{2000}$ th to  $\frac{1}{2500}$ th of a millimetre = '00002 to '000016 inch; and this is greatly facilitated by the more considerable breadth and depth of the lines ruled on the eye-piece micrometer, which may be filled up with black-lead, and thus rendered more distinct.

The chief advantage of the method consists in the fact, that the error in the workmanship of the scale vanishes. Thus, if it has been ascertained that a division is equal to  $\frac{1}{1000}$ th of a millimetre, then if the relative difference of any two divisions is equal to one-seventh of their diameter (and this is the largest difference which I have ever observed), the results of measurements cannot vary at different parts of the scale by more than  $\frac{1}{7000}$ th of a millimetre,—a quantity which may be regarded as beyond the power of vision in most cases.

<sup>1</sup> More recent observations with Ross's microscopes have shown, that distances not exceeding  $\frac{1}{81600}$ th of an inch can be resolved satisfactorily. See Queckett on the Microscope, 2d edition, p. 476.—[*Trans.*]

<sup>2</sup> Two plano-convex lenses, with their plane surfaces turned outwards, and their convex surfaces opposed to each other within the eye-piece tube.—[*Trans.*]

The method has, however, its disadvantages. In the first place it gives no direct measurements, but requires a special calculation of the value of the divisions of the scale for every object-glass which is employed,—for this value is of course diminished constantly as the magnifying power is augmented. For this calculation any object of known diameter may be used—the best is the metallic thread above-mentioned, or a second glass micrometer may be used as an object, care being taken to compare different portions of its scale, and use the *mean* in making the calculation.

A more important objection to the instrument is the difficulty of distinguishing the lines in the eye-piece, when several objects lie mixed together in the field of the microscope. It will serve with great convenience for measuring blood-corpuscles and similar isolated well-marked objects; but to the measurement of ganglion-cells when entangled in their surrounding tissue, of fat cells which are crowded together, etc., it is either with difficulty applicable, or totally inapplicable.

#### *Screw Micrometer.*

When a screw is turned upon a piece of metal, we can give it a forward or backward motion, varying in amount according to the number of turns cut upon a given length of the cylinder. If there are five turns in a millimetre, then each revolution of the screw will give a forward or backward movement through one-fifth of a millimetre; if, then, one extremity of the screw be provided with a circular index-plate, divided into one hundred parts, each fraction of a revolution corresponding to such a division will indicate motion through  $\frac{1}{500}$ th of a millimetre. If, besides, there be applied to the index a *nonius* or vernier, capable of estimating one-tenth of each division, the screw may be made to indicate motion through  $\frac{1}{5000}$ th of a millimetre.

On this property of the screw depends its application to micrometry. The construction commonly followed is, to make a female screw, and fix it in a position not admitting of change, and within it to place a male screw, to impart a slow motion to the object. The screw micrometers now in use are of two kinds,—the *stage-screw* and *ocular-screw* instruments.

The purpose of the stage-screw is, to impart to a portion of the stage, and consequently to the object, a degree of motion by which the length of the object can be measured by bringing its extreme margins successively into contact with a thread stretched across the ocular. The length is estimated by the number of turns, or fractions of turns, imparted to the screw.

It has been already (p. 3) remarked, that the accuracy of measurements thus obtained cannot be altogether depended upon, and that one who is acquainted with the mode of cutting these screws, and the sources of error apt to affect their construction, need not be surprised to find variations amounting to  $\frac{1}{100}$ th or more in their indications. To ascertain the exact value of the scale of a screw micrometer, it is therefore absolutely necessary to test it with a fixed standard, such as the metallic thread already mentioned. The enumeration of the screw turns in a given space, though recommended by many authorities, gives a very uncertain result, in consequence of the shortness of the screw. A glass micrometer can only be used as a standard when the value of its divisions has been previously absolutely ascertained, and the error from inequalities in its divisions made to vanish, by taking the mean of a number of measurements of different portions of its scale.

In examining a screw micrometer, other considerations likewise must be taken into account. In ill-made instruments a certain amount of so-called *dead motion* is observed—*i. e.*, the index-plate may be moved without simultaneous motion being imparted to the object. This error, in course of time, comes to affect the best made screw instruments after frequent use, in consequence of the wearing of screw-thread; hence such instruments should only be used in making measurements, and never for the mere purpose of altering the position of the object in the field. If this fault exists, and the apparatus is otherwise perfect, the results of observation may be correctly obtained by bringing one edge of the image

of the object into apparent contact with the thread in the ocular, and then steadily turning the screw forward in the same direction till contact between the thread and opposite margin of the object is obtained; any to-and-fro motion of the object must in this case be avoided.

Inaccuracies in the cutting of different portions of the screw should also be ascertained and kept in view. In instruments by the best makers these are, it is true, but trifling; still, when exactitude is required, they must not be neglected. All different parts of the same screw do not furnish identical measurements, but when the amount of difference is known, the requisite corrections may be applied.

The thread stretched across the ocular is commonly a spider-web thread, although other threads and hairs, and likewise lines ruled on glass with a diamond, are sometimes substituted. It is of little consequence that the thread should be very thin, the great requisite being the accurate definition of the *margin* with which the image of the object is brought into contact. Another error, inseparable from all measurements with the screw micrometer, is the influence of diffraction upon the margins of the object, rendering it difficult to tell exactly when they are in contact with the thread. To remove this source of error, Mohl<sup>1</sup> has advised the substitution of a delicate needle for the thread in the ocular, and the bringing of its fine point into the middle of the field of view. The difficulty is not in this way got rid of, for the diffraction of the rays of light is as considerable between the object and point as between the object and spider hair. The only mode of getting rid of the diffraction is to make the observation by reflected light, and this can of course only be done in the case of certain objects, and under moderate magnifying powers.

To ascertain the amount of accuracy attainable by the use of the screw micrometer, Mohl and I have made some experimental measurements, and calculated the probable error of each separate observation, and of the mean of all. Mohl found in measuring objects of different sizes, with a screw micrometer by Plöszl, that the mean error of ten observations of the same object was  $\frac{1}{25000}$ th to  $\frac{1}{52000}$ th of a millimetre,—*i. e.*, about  $\frac{1}{13700}$ th of a millimetre =  $\frac{1}{342000}$ th of an inch. By measuring a blood-corpuscle with screw micrometers by Plöszl and Powell, I obtained the following results from a like number of observations:—

	Greatest diff. of two measurements.	Probable error of mean.	Probable error of each measure.
Powell, ...	$\frac{1}{492}$ millim.	$\frac{1}{5010}$ millim.	$\frac{1}{2300}$ millim.
Plöszl, ...	$\frac{1}{760}$ "	$\frac{1}{10100}$ "	$\frac{1}{3000}$ "

Hence it is obvious, that although the mean probable error of a number of measurements of an object is so minute, that it may in general be neglected; yet a single measurement may, when the object measures less than  $\frac{1}{10000}$ th of a millimetre, lead to an error of  $\frac{1}{4}$ th to  $\frac{1}{3}$ d of the total diameter, which may even exceed this amount, however carefully the manipulation is conducted.

The reason why the best made screw-micrometers of this sort cannot give more accurate results, is chiefly this,—that however steady the whole stage of the microscope is, the pressure of the fingers used in moving the screw causes some alteration in the position of the object, independent of the movement of the screw, so that when the opposite margin of the image comes into contact with the thread, one is not quite sure that the space which it has traversed is the diameter which it is wished to measure. It should be added, that all sources of error, whether connected with the contact with the thread, or with the workmanship of the screw, are increased in amount with the magnifying power employed.

For these reasons, the stage screw-micrometer must yield in accuracy to the ocular-screw instrument. The most useful variety of the latter is that applied to astronomical telescopes; consisting of a Ramsden eye-piece, in the field of which are placed two parallel threads, one of which is fixed, while the other can be moved to and fro by means of a screw. In using the instrument, the margins of the objects are brought within the threads. The value of each turn, and fraction

<sup>1</sup> Linnæa, 1842, p. 502.

of a turn, indicated by the index of the screw, is ascertained by methods already mentioned, and of course varies with each object-glass employed.

It is plain that, by using the screw in this way as a micrometer, much greater accuracy is secured. An eye-piece screw of five turns in a millimetre, which can only move an object  $\frac{1}{5}$ th of a millimetre at each turn, will correspond at each revolution, when the object-glass magnifies 100 diameters, to  $\frac{1}{500}$ th of a millimetre referred to an object on the stage. If the index be divided into 100 parts, each division will measure spaces of  $\frac{1}{50,000}$ th of a millimetre = about  $\frac{1}{1,250,000}$ th of an inch. Such delicacy of measurement, which may even be carried further, by means of a *nonius*, is to be regarded as quite superfluous; and hence the ocular screws are commonly made thicker, and with a smaller number of turns in a given space, which enables the workman to finish them with greater accuracy than the more delicate stage-screws.

With an eye-piece screw instrument by Dollond, in which, with a magnifying power of 435, each division of the index corresponded to  $\frac{1}{10,630}$ th of a millimetre, and with a power of 820, to  $\frac{1}{19,600}$ th of a millimetre, I have made a series of ten measurements of each of a few objects, with the following results:—

Object.	Power.	Greatest Difference.	Probable Error	
			Of the Mean.	Of each Measurement.
Blood Corpuscle, .....	435	$\frac{1}{2040}$ millim.	$\frac{1}{31200}$ millim.	$\frac{1}{9940}$ millim.
Glass Micrometer space of .05 } Millimetres, .....	435	$\frac{1}{2040}$ ...	$\frac{1}{30000}$ ...	$\frac{1}{9560}$ ...
Ditto, .01 Millimetres, .....	850	$\frac{1}{2700}$ ...	$\frac{1}{57000}$ ...	$\frac{1}{18200}$ ...
Intervals between lines on the } smallest scales of <i>Lepisma</i> , }	850	$\frac{1}{8250}$ ...	$\frac{1}{86000}$ ...	$\frac{1}{27000}$ ...

Hence it appears, that, while the probable error of the mean of each series of observations is far beneath the limit of size at which the eye can distinguish an opaque body through the microscope, the error of individual measurements trenches very closely upon this limit. This is the utmost degree of accuracy attainable by any micrometric method, and in this the ocular-screw instrument surpasses all other contrivances.

The only objection to its general use is its high price; for, to assist the micrometer itself, which of course is a rather expensive article, the stage of the microscope must be fitted with a moveable table, in order to bring the edges of the object into rough adjustment with the spider hairs, which cannot be done with sufficient accuracy by the unassisted hand.

#### Other Micrometric Methods.

Among the modes of measuring microscopic objects, there are some which have this in common, that they consist in projecting the magnified image upon some surface, where it is measured. The dimension thus obtained being divided by the linear magnifying power, gives as its quotient the diameter of the object.

This procedure is followed in using,—1st, *Various modes of dioptric and catoptric projection*; 2d, *The solar microscope*; and 3d, *The method by double vision* (*dubbelzien*).

All these methods, therefore, require, in the first instance, an accurate acquaintance with the magnifying power employed. I may add, that in order to calculate the number which should express the magnifying power for such purposes, we must not use the ordinary standard distance of distinct vision,<sup>1</sup> but must inquire,

<sup>1</sup> This is commonly estimated in this country at ten inches, and in France at 250 millimetres; and linear magnifying power is in general determined by dividing the measured diameter of an object projected on a scale at ten inches from the eye, by its true diameter. Thus if  $\frac{1}{100}$ th of an inch occupy two inches on the scale, the magnifying power must be 2 divided by .01 = 200.—[*Trans.*]

what is the amount of magnifying power referred to the fixed distance of the surface on which the image is projected? Of course this problem must be, in the first place, very accurately solved; and in every following observation in which the figures thus obtained are used, care must be taken that the image is projected to exactly the same distance.

Further, one must, when using these methods, remember the influence of the distortion of the image on the apparent magnifying power, and examine the errors from this source at different parts of the field. These may, however, be got rid of by placing a ring in the centre of the ocular, and before every measurement, bringing the object into such a position, that the margins of its image are in contact with the circle.

In the second place, the diameter of the image must be measured with the utmost exactitude,—*i.e.*, at least to tenths of a millimetre.<sup>1</sup>

In making measurements in this way, it is almost indifferent which of the different catoptric or dioptric modes of projection is followed. The different sorts of Camera Lucida and Sömmering's mirror, I have elsewhere described. All, however, require either that the microscope should be directed longitudinally, or that the pencils of light should be made horizontal by transmission through a right-angled glass prism; for images projected on a vertical plane cannot, on account of the unsteadiness of the hand, be readily measured with the requisite nicety. If the image be projected upon a common slate, which is well adapted for the purpose, care must be taken always to use one of the same thickness.

When all necessary precautions are attended to, very exact results may be obtained in this way, as the following account of some experiments with a Sömmering mirror will show:—

Object.	Power.	Extreme Difference.	Probable Error	
			Of Mean of 10.	Of each Measurement.
Blood Corpuscle, .....	740	$\frac{1}{1430}$ millim.	$\frac{1}{24100}$ millim.	$\frac{1}{7630}$ millim.
.05 Millimetre ruled on Glass,	369	$\frac{1}{530}$ ...	$\frac{1}{7812}$ ...	$\frac{1}{2730}$ ...

The probable amount of error is in these two cases rather different, and this is accounted for by two causes. The amount of accuracy increases with the magnifying power. It is true that a power of 740 does not give so sharp a definition as one of 369 diameters; still the amount of error committed in measuring an object under the latter power will be about double of that committed with the assistance of the higher magnifying power. Another explanation of the observed discrepancy, is the different size of the objects subjected to experiment. The size of the blood corpuscle was .0063 millimetre,—*i.e.*, about one-eighth of that of the micrometer-space afterwards examined; the image in the first case was consequently about four times smaller than in the second. Now the smaller an image is, the more easily can its opposite extremities be at the same time seen by the eye, and the more certain, consequently, is its measure with the compasses; but if an image occupies a large portion of the field, the difficulty of simultaneously seeing both its ends increases with its length, and the chance of error in measurement is thus multiplied. In this point of view, then, these methods are inferior to the use of the screw-micrometer, which measures objects, both great and small, with equal accuracy.

Different kinds of *picture-microscopes*<sup>2</sup> may be used for taking measurements; but as, in their common forms, they require to be employed in a dark room, they

<sup>1</sup> Harting has invented a kind of double calipers for this purpose.

<sup>2</sup> Literally *beeldmikroskopen*, including solar microscopes, and all those in which the image is projected on a screen, and viewed by the unassisted eye.

are far too inconvenient. The portable solar microscope, especially that which I shall afterwards describe and figure in another part of my work, will be found more serviceable. In it the image is received upon a ground glass plate,<sup>1</sup> and then measured, with the precautions above mentioned.

The following are results of measurements made with this apparatus:—

Object.	Power.	Extreme Difference.	Probable Error	
			Of Mean of 10.	Of each Measurement.
Blood Corpuscle, .....	593	$\frac{1}{1500}$ millim.	$\frac{1}{8400}$ millim.	$\frac{1}{7840}$ millim.
·05 on a Glass Micrometer,...	593	$\frac{1}{840}$ ...	$\frac{1}{13900}$ ...	$\frac{1}{4400}$ ...

It is obvious that in this way too we may measure an object very exactly. The accuracy diminishes with the magnifying power, as in the other methods already described. The diameter of the object also has a certain influence, though less than when Sömmering's mirror is used; but this inaccuracy of measurement with the *picture-microscope* is not, as in the case of the mirror, caused by the size of the object, but is connected with the superior sharpness of the image about the centre of the screen.

#### *Measurement by Double Vision.*

The last method to which we need direct attention is that by which an object seen with one eye, is by the help of the other projected and measured upon the stage-plate.<sup>2</sup> The precautions to be attended to in conducting measurements by this method may be here enumerated:—

1st. During the measurement the axis of the eye must undergo no change of direction, and should be kept steadily fixed in one direction.

2d. In order that the extremity of the compasses used for measuring may always be kept at the same distance from the eye, the compound microscope should be

<sup>1</sup> The essential parts of the instrument here alluded to are, an ordinary compound microscope, strongly illuminated by a mirror reflecting the direct solar rays, and a black funnel-shaped tube, the lesser end of which is fixed in the place of the eyepiece, or immediately above it, while its greater and upper end carries the ground glass plate or screen to receive the image. As little external light as possible should be permitted to fall on the upper side of the screen during the observations.

<sup>2</sup> The projecting of the images seen by one eye upon a surface seen with the other, is termed *double vision* (dubbelzien). If an opaque body, a finger for example, be held at a certain distance before one eye, so as to hide from it an object a little way off, the other eye will still see the object, and in certain circumstances will, as it were, see it through the finger. This same experiment may, after a little practice, be successfully made with the microscope. If, while one eye distinguishes an image in the microscope, the other be directed to some object held alongside of the instrument—to a pencil, compasses, or the like—both will simultaneously be perceived in the field of vision. If, for instance, we look with the left eye into the tube, while a piece of paper is laid to the right of the instrument, the microscopic field will seem, if both eyes be kept open, to be projected on the paper, and we may even draw on the latter outlines of images present in the former.

The power of double vision must be acquired by practice, but is so easily attained, that I cannot sufficiently recommend it to young observers, not only as the simplest mode of projecting the image, but as the only mode in which it can be done without loss of light. This property is especially valuable, as it affords the only means of perceiving the minuter parts of an object on a projected image. There need be no difficulty in effecting this by *double vision*; and the only precaution to be attended to, in order to make the illusion perfect, is, to give to the surface on which it is desired to project the image a colour nearly corresponding with that of the field of view, and for this purpose papers of different tints may be placed upon the stage.—Vol. i., pp. 259, 260.

provided with a large stage, on which the legs of the compasses<sup>1</sup> may rest. If the stage, as is the case with many microscopes, be too small, it may be widened by attaching to the spring-clips a piece of flattened card-board. In using the simple microscope, or a compound one of small height, the compasses may be made to rest upon the table, or upon the box to which the instrument is screwed.

3d. Upon the surface on which the image is to be projected, a piece of paper should be laid, possessing as nearly as possible the colour of the microscopic field; this will greatly assist the illusion, causing the object and compasses to be both distinctly visible at the same time.

4th. Great care must be taken that the distance between the eye and the plane where the measurement is made shall always remain alike. Hence the slides on which the objects are placed, and the paper itself which we lay on the stage, should always have the same thickness.

Other precautions alluded to in former sections of this article must, of course, be attended to.

A series of measurements made in this way gave the following results:—

Objects.	Power.	Extreme Difference.	Probable Error	
			Of Mean of 10.	Of each Measurement.
Blood Corpuscle .....	579	$\frac{1}{1940}$ millim.	$\frac{1}{27000}$ millim.	$\frac{1}{8330}$ millim.
Ditto .....	332	$\frac{1}{1660}$ ...	$\frac{1}{22100}$ ...	$\frac{1}{7040}$ ...
·05 of Micrometer .....	579	$\frac{1}{828}$ ...	$\frac{1}{13200}$ ...	$\frac{1}{4000}$ ...
Ditto .....	332	$\frac{1}{556}$ ...	$\frac{1}{8850}$ ...	$\frac{1}{2800}$ ...
·01 of Micrometer .....	910	$\frac{1}{2280}$ ...	$\frac{1}{34500}$ ...	$\frac{1}{10700}$ ...
Intervals between lines on smallest scales of Le- pisma .....	910	$\frac{1}{4350}$ ...	$\frac{1}{75800}$ ...	$\frac{1}{24000}$ ...

It is evident from these numbers that here, too, the degree of accuracy increases with the magnifying power and the minuteness of the object. This method, therefore,—the simplest of all micrometric methods and the most generally useful, being alike applicable to the simple and to the compound microscope,—may be practised with a degree of accuracy surpassing all others, except the measurement with the eye-piece screw micrometer. Of course, this accuracy is only attainable after much practice; yet an observer who has acquired proficiency in the mode of *double-vision* will but seldom be obliged to have recourse to any other means of measuring objects. It is, besides, of consequence to practise this method, as it enables us to make measurements with the greatest rapidity,—a consideration of some importance when a number of observations are to be made and the mean calculated.

In order to make twenty measurements of the same blood-corpuscle by each of four different methods, the following time was required:—

Different screw micrometers, ... ..	26 to 30 minutes.
Portable solar microscope, ... ..	18 "
Sömmering mirror, ... ..	16 "
Double vision, ... ..	11 "

<sup>1</sup> A scale for each magnifying power of the microscope is convenient, and may be easily constructed in a few minutes. Let the image of a glass micrometer be projected and drawn upon a piece of paper laid on the stage-plate. To this scale measurements are subsequently referred by embracing the image of the object between the points of a pair of compasses, held at the same distance from the eye; the compasses being then applied to the scale, the included distance is at once read off. The same method is equally applicable to observations with the camera and with the Sömmering mirror.—[*Trans.*]

*Choice of a Micrometric Method.*

In making choice of a micrometric method, another consideration must have its weight,—viz., the degree of accuracy requisite for the particular species of observation. Of course, when a simple measurement enables us to determine the size of a body with all possible accuracy, this most exact of methods must be preferred. For example, to determine the relative size of an air-bubble, and of the image of an object beneath it, when it is wished to deduce the index of refraction of the fluid surrounding the air-bell, we would prefer the ocular-screw micrometer to all other instruments. But in the case of objects from the organic kingdom, when we have to do with a mean number deduced from a series of observations of *different specimens*, another circumstance suggests itself,—viz., the amount of probable error always connected with this mean, and which in certain cases may even exceed the error probably introduced by the use of a less exact micrometric method. To discover how far this consideration is reasonable, I have made the following measurements with the ocular-screw micrometer, and calculated the probable amount of error in the mean results:—

Objects.	Number of Measurements	Mean Diameter.	Probable Error.
Fibres of areolar tissue .....	10	$\frac{1}{909}$ millim.	$\frac{1}{17000}$ millim.
Blood corpuscle of a man ...	15	$\frac{1}{130}$ ...	$\frac{1}{6000}$ ...
Primitive fasciculus of mus. } gastrocnemius of a child }	20	$\frac{1}{173}$ ...	$\frac{1}{4170}$ ...
Primitive fibre from me- } dian nerve (man) .....	15	$\frac{1}{60}$ ..	$\frac{1}{980}$ ...
Primitive fasciculus from } psoas of an adult .....	21	$\frac{1}{19}$ ...	$\frac{1}{500}$ ...

Hence it seems that, in proportion as the diameter of organised elementary parts is large, the probable error is great, and that in the case of objects measuring from  $\frac{1}{80}$ th to  $\frac{1}{20}$ th of a millimetre =  $\frac{1}{1500}$ th to  $\frac{1}{500}$ th of an inch, it may, even when the mean is deduced from a far larger number of observations, exceed the probable error of manipulation or observation in any of the micrometric methods above described. Consequently, in measuring objects of this sort, it is, in point of accuracy, quite indifferent which of these methods is employed.

But it is otherwise with minuter objects, blood globules, the elements of fibrous tissue, tendon, muscle, etc. Here our choice is more limited, for we should exclude all those methods of which it has been shown that their probable error can have a marked influence on the mean result. This is true, not only of the different species of glass micrometers, but in a less degree of the stage-screw instrument, for their probable error deduced from measurement of one and the same blood-corpuscle amounts to more than double of the mean error deduced from the measurement of several different blood-globules.

When the diameter is yet smaller, we must, for the same reason, betake ourselves to methods of the utmost delicacy,—of these the screw-ocular micrometer is without doubt the most exact. It is, however, nearly equalled in precision by the camera lucida, the Sömmering mirror, and, above all, by the method of *double vision*.