

JOURNAL  
OF THE  
ROYAL  
MICROSCOPICAL SOCIETY;

CONTAINING ITS TRANSACTIONS AND PROCEEDINGS,  
AND A SUMMARY OF CURRENT RESEARCHES RELATING TO  
ZOOLOGY AND BOTANY  
(principally Invertebrata and Cryptogamia),  
MICROSCOPY, &c.

*Edited by*  
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*and Professor of Comparative Anatomy and Zoology in King's College ;*

WITH THE ASSISTANCE OF THE PUBLICATION COMMITTEE AND  
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FELLOWS OF THE SOCIETY.

FOR THE YEAR  
1890.

Part 1.



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**WILLIAMS & NORGATE,**  
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## MICROSCOPY.

## a. Instruments, Accessories, &amp;c.\*

## (1) Stands.

**Mirand's and Klönne and Müller's Microscopes with revolving stages.**—In 1880 we described† the Microscope of MM. Klönne and Müller with a revolving stage for eight slides, which had then been issued. Subsequently we heard of a similar instrument by M. Mirand in which, however, the stage had also a motion from back to front, so that three objects could be observed on each of the slides, and in 1883 we described‡ this as an extension of the principle of MM. Klönne and Müller's Microscope. It is now stated§ that M. Mirand's instrument was exhibited at the Paris Exhibition of 1878, and was therefore the original form, so that in place of the French makers having devised a "modification mieux comprise," it is the German makers who are responsible for an "imitation mal comprise."

**Nobert's Micrometer-Microscopes.**—Fig. 1 shows an early form of Microscope devised specially by the late F. A. Nobert for fine measurements by stage-micrometer.

The chief peculiarities in the design are (1) the application of the stage-micrometer, and (2) the arrangement of the fine-adjustment.

(1) The stage-micrometer is a permanent attachment of the stage, the micrometer-screw acting upon a travelling stud fixed beneath the upper plate, causing it to traverse the field of view laterally. The screw is actuated by a large radial wheel, the spokes being of such a length that a very small movement can be effected. The radial wheel is removable, when the Microscope can be used for ordinary observations.

(2) The fine-adjustment is effected by a screw passing through the standard from the back and pressing against a bar or arm about 2 inches in length, extending downwards at the back of the stage. The stage is suspended on the standard on coned screw-pivots fitted in a fork-piece, and is easily detached by releasing the pivots by the milled heads shown on either side of the standard. The fine-adjustment screw pressing against the bar beneath the stage causes the latter to incline upwards from the horizontal, and so to approach the objective; with the reverse motion of the screw the stage inclines the opposite way by gravitation. This system of fine-adjustment was (we believe) first devised by Herr Nobert, and has been largely adopted in Germany for low-priced Microscopes.

Fig. 2 shows the improved form of Micrometer-Microscope as exhibited by Herr Nobert at the Exhibition of 1862.||

(1) The stage-micrometer with its graduated drum and vernier is carried by the stage, whilst the screw is actuated by a large milled

\* This subdivision contains (1) Stands; (2) Eye-pieces and Objectives; (3) Illuminating and other Apparatus; (4) Photomicrography; (5) Microscopical Optics and Manipulation; (6) Miscellaneous.

† This Journal, 1880, p. 144.

‡ This Journal, 1883, p. 897.

§ Journ. de Micrographie, xiii. (1889) pp. 523-4.

|| Vide Reports of the Juries, Class XIII., p. 25.

head (instead of the radial wheel of fig. 1), which is supported on a separate standard and connected with the screw by a Hooke's joint. We believe this plan of providing a separate standard for the milled

FIG. 1.

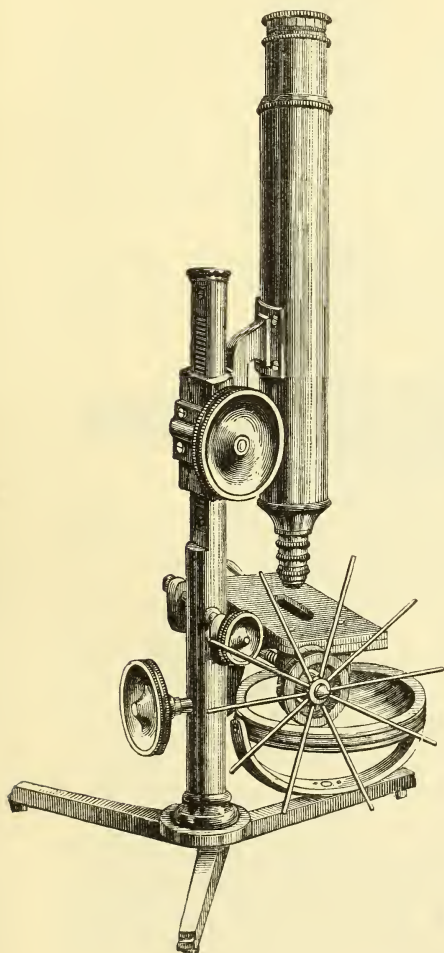
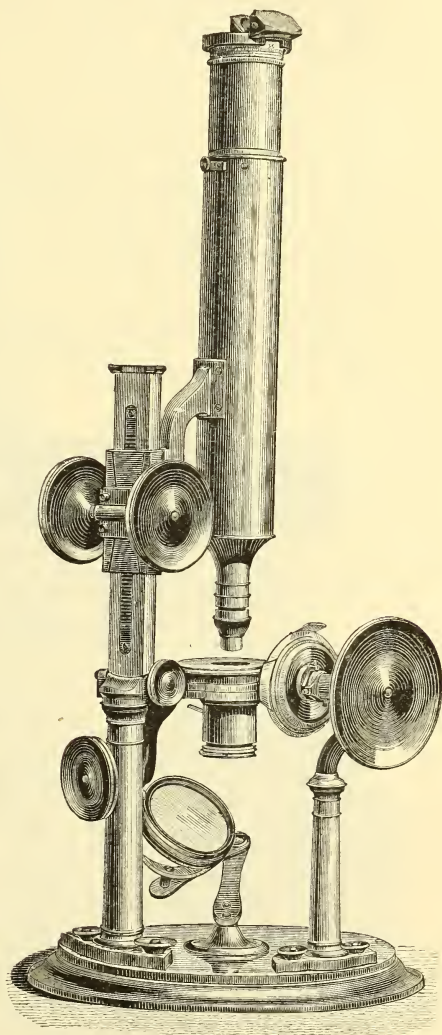


FIG. 2.



head and actuating the screw by means of a Hooke's joint was devised by Herr Nobert to avoid the tremor of the hand being communicated to the micrometer, and yet to utilize this peculiar fine-adjustment acting upon the stage.



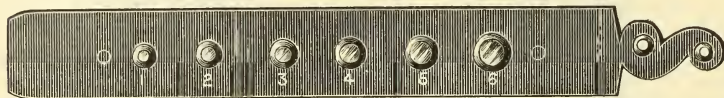
(2) The base is double; the upper plate, carrying the standards of the Microscope and of the micrometer, rotates on the lower on a centre in the line of the optic axis.

(3) The mirror is attached to a rotating cylinder in the centre of the lower base-plate by two elbow joints set at an angle, by which it can be adjusted at any angle in altitude beneath the object, whilst maintaining very nearly an equal distance from the object in all positions. The mirror can thus be rotated radially upon the object, or *vice versa*.

The example of this second form of Nobert's Micrometer-Microscope in Mr. Crisp's collection is furnished with a mechanical stage (with glass surface) in which the rectangular movements are effected by means of a single plate. Fine micrometer-screws are applied to project from two right-angle edges of the stage and pass through fixed shoulder-rings; each screw has a spiral spring encircling it and pressing against the shoulder-ring; milled nuts exterior to the shoulder-rings act on the micrometer-screws, giving very smooth and delicate rectangular movements to the stage.

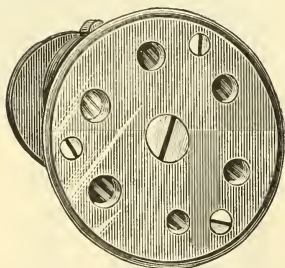
**Old Microscope with nose-piece for rapidly changing objectives and mirror formed of a silvered bi-convex lens.**—The nose-pieces of which so many were brought out a few years ago for rapidly changing

FIG. 3.



the objectives were generally considered to represent an entirely modern idea, though our forefathers had placed objectives in a long dove-tailed slide like fig. 3, or more commonly in a rotating disc like fig. 4.

FIG. 4.

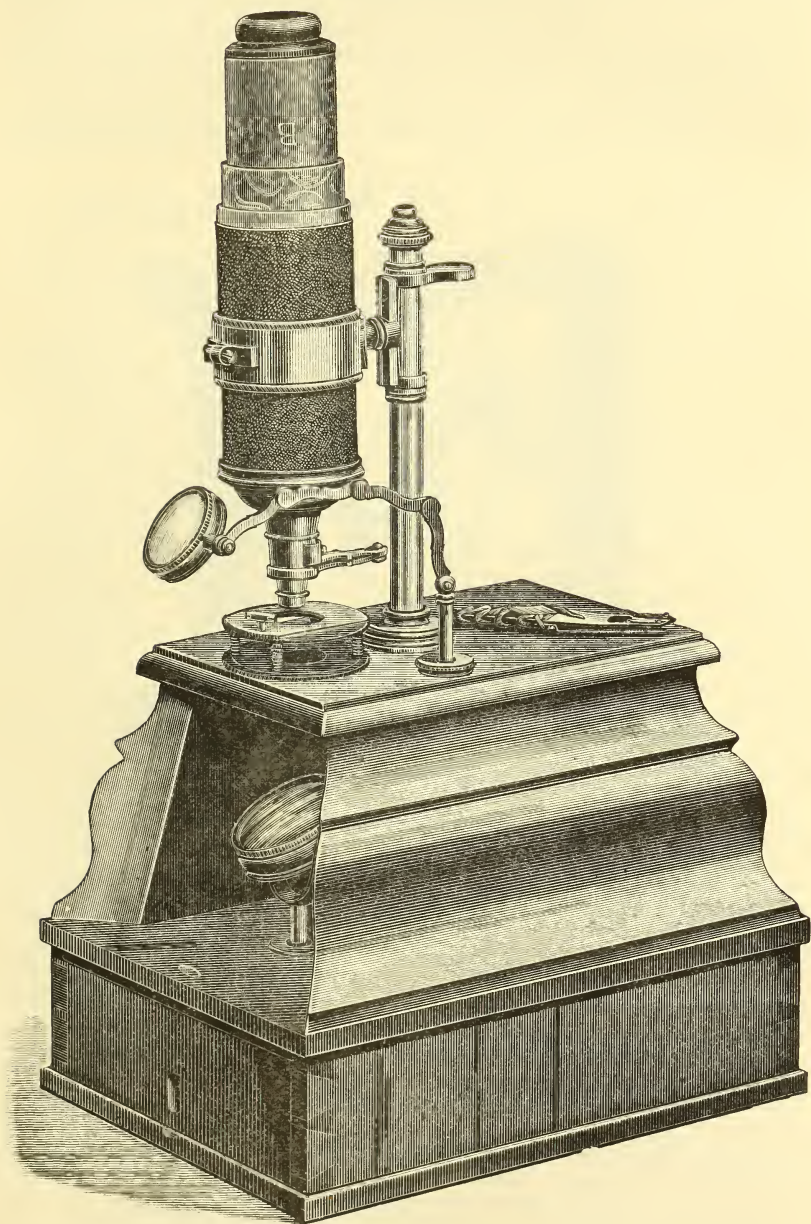


An old Microscope recently acquired by Mr. Crisp (shown in fig. 5) has an arrangement which is doubtless the earliest of its kind. To the body-tube is screwed a nose-piece, to which is attached a short arm, on which pivots a second arm with a "cell" at the end, into which the objectives drop. To change the objective the second arm, which has a slight amount of "spring," is depressed and then swung away from the body-tube, the objective lifted out of the cell and another inserted

in its place, and the arm turned back again. The cell, which is about 1/8 in. deep, fits over the end of the nose-piece, and thus keeps the objective in position.

The Microscope is apparently of Augsburg make, probably by G. F. Brander, whose career as a mechanic and optician was comprised between the years 1734 and 1783, when he lived at Augsburg. The mirror is a biconvex lens silvered—a device which has been reinvented more than once during the last ten years!

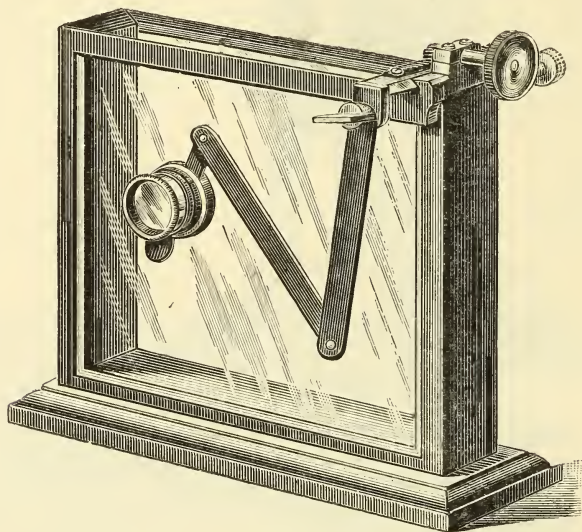
FIG. 5.



**Rousselet's Simple Tank Microscope.**—Mr. C. Rousselet exhibited at a recent meeting of the Society a small tank Microscope (fig. 6) designed for the purpose of rapidly looking over pond water and weeds collected at a day's excursion and placed in a small parallel-sided window aquarium.

One of Zeiss' aplanatic lenses is carried on a jointed arm, which moves parallel to the side of the tank, and the lens is focused by means

FIG. 6.



of a rack and pinion, the whole being fixed to the upper left-hand corner of the tank by means of a screw clamp.

The following points, Mr. Rousselet considers, will recommend themselves to those who are in the habit of looking at their captures with the pocket-lens in the ordinary way:—

When an object of interest is found it can be followed with the greatest ease and taken up with a pipette, both hands being free for this operation. It frequently happens that a minute object is lost simply by removing the pocket-lens for an instant to take up the pipette; in the above apparatus the lens remains in the position in which it has been placed. The definition of these aplanatic lenses is excellent; the lowest power has enough working distance to focus through tanks of moderate size, and the magnification (6 diameters) is sufficient to permit of the identification of all ordinary rotifers, and anything uncommon or new is at once recognized. Such delicate creatures as the Floscules, which are all but invisible with the ordinary pocket-lens, are seen without difficulty, and the whole contents of the tank can be ascertained with a great saving of time.



Mr. Rousselet also specially recommends these small window aquaria to those not already acquainted with them, as affording the very best means of examining pond water for microscopic life.\*

In Mr. Rousselet's original figure the lens-carrier was clamped on the vertical side of the tank; we have ventured to show the apparatus clamped on the top of the tank, where we think it will be found safer in practice.

## (2) Eye-pieces and Objectives.

**New Objective of 1.63 N.A.**—This objective is further described by Dr. H. Van Heurck† (see also the paper by Dr. S. Czapski, *supra*, p. 11), who says that "its advantages have surpassed all that could be hoped for."

The design of the objective was started by Prof. Abbe four years ago, but it was only in August last that he was able to complete the preliminary calculations and to commence the actual execution of the objective, which was finished on the 17th September.

The objective is 1/10 in., apochromatic, and with an aperture of 1.63 N.A. A special compensating eye-piece 12 removes the last traces of colour. The cover-glasses of Dr. Van Heurck's objects have a refractive index of 1.72, and the slide is approximately of the same index; both are of flint glass. The diatoms are melted in the cover-glass. The mounting medium has an index of 2.4, and the immersion fluid (monobromide of naphthaline) an index of 1.65. The aperture of the immersion condenser is 1.60 N.A., and the upper lens is of flint, for utilizing the most oblique light. Monobromide of naphthaline is used here also as the immersion fluid.

The lenses of the objective are thus disposed:—

- (1) Front lens (more than a hemisphere) of flint. Index 1.72.
- (2) Achromatic lens.
- (3) Crown-glass lens.
- (4) Achromatic lens.
- (5) Correcting lens (three glasses).

Three of the lenses are in fluorite.

Prof. Abbe considers that the difference between the indices of the cover and the immersion liquid notably favours the resolution.

In regard to its management, Dr. Van Heurck has used it daily for two months, and for hours together, and he finds it in every way as practical as other objectives of large aperture.

"In oblique light *Amphipleura* is entirely resolved in beads, as clearly as we see *Pleurosigma* with the best existing objectives, the beads being much closer than the previous imperfect resolutions had led one to believe. Repeated measurements of photographs show that it has 3600 transverse and 5000 longitudinal striæ per millimetre. It is not, therefore, surprising that there has hitherto been so much difficulty in showing these beads.

It is only for these beads that oblique light is required. All the other difficult tests, such as *Frustulia saxonica*, *Surirella gemma*, and

\* Journ. Quek. Micr. Club, iv. (1890) pp. 53-4 (1 fig.).

† Cf. Bull. Soc. Belg. Micr., xv. (1889) pp. 69-71; Journ. de Microgr., xiii. (1889) pp. 527-8.

even the transverse striæ of *Amphipleura*, are resolved with axial light. *Pleurosigma angulatum* shows new details which have still to be studied. On examining it without the eye-piece eleven spectra are seen, i. e. five new intermediate spectra. Some bacteria have also shown new details.

The illuminating power of the objective is very great. Strong photographs of the beads of *Amphipleura* have been obtained in six minutes with a magnification of 1500 diameters with monochromatic solar light, whilst with the ordinary apochromatics at least ten minutes was necessary with only 1000 diameters and ultra-oblique illumination.

Only three objectives have yet been made, two for the Continental tube and one for the English. One of the former is in the hands of Dr. Koch, the Berlin bacteriologist, and will, it is hoped, give some interesting results.

Dr. Van Heurck considers that "the new objective forms an honourable practical crowning of the long theoretical labours of the illustrious Prof. Abbe, who for fifteen years has so happily led the Microscope into new paths, and who has with indefatigable patience realized practically all that theory indicated."

**Semi-apochromatic Objectives.**—Mr. E. M. Nelson read the following note at the December meeting:—

As these new semi-apochromatics are "Students'" lenses, let me briefly trace their development. The earliest form of student's lens was a combination of three "French buttons" or doublets; almost the whole of the medical and students' work, both here and on the Continent, was carried on by means of these lenses. The one I am exhibiting to-night is an example of the favourite form in this country, the sale of which, as I am credibly informed, must be counted by thousands. This lens gave way to the Hartnack, which consisted of two doubles and a single. The Hartnack was an immense advance over the French button, but looking at them from a present day point of view, we should say that while some picked specimens were good, the bulk of them were very mediocre. Somewhat later came Seibert of Wetzlar. His lenses, in form not differing greatly from those of Hartnack, were decidedly superior to them in finish; at the same time, his angles were low for the most part. Of his lenses two even now justly have world-wide celebrity. I allude to his No. III. and his water-immersion 1/16.

Before leaving Seibert let me point out that a Seibert No. III., unscrewed from its brass mount, constitutes the best high-power pocket-lens ever made. One mounted like a Coddington would be a useful appendage to a microscopist's outfit, as it has fully 1/8 in. working distance, which the Coddington has not.

One other point. You are all aware that on the Continent almost nothing has been done with low-power lenses. Seibert alone of all the Continental makers produces a No. 0, which is a first-class 1½ in. With this lens Mr. Rousselet and myself have seen the cilia on *Volvox*. An example of this lens is on the table. The Hartnack was superseded by the Zeiss achromatic, a lens much of the same form, i. e. two doubles and a single, but altogether of superior workmanship. Zeiss also, by making each class of lens both wide and low angled, suited all tastes. To illustrate this period of lens I have brought a D D or 1/6 in.

I have now come to the time when English opticians made students'



lenses; they adhered to the usual form of two doubles with a single front. Among the first and most successful was Swift; an early example is on the table. This lens is a  $1/5$ , and was sold for half the price of the English  $1/4$  of that day.

A new competitor appears on the field, viz. Reichert of Vienna. The example before you is one of the first batch of his lenses that came to this country; it is a  $1/7$  of  $0.84$  N.A., very well corrected and very well finished. Its price was  $2\text{L.}$ , and at that time there was nothing made here that would at all compare with this lens for three times that sum. Next in order comes Leitz of Berlin, who was mainly known by his  $5\text{L.}$  oil-immersion, and now as I have brought up the history to recent times I will give no more particulars with regard to achromatics, but go straight to semi-apochromatics.

I call these lenses "semi-apochromatics" because, while they are not "apochromatic," they possess a higher degree of achromatism, due to the employment of Jena glass in their construction, than previously possible with the old glass. Among others the most remarkable instance of the capabilities of this Jena glass will be seen in the production by Leitz of two lenses, a  $2/3$  of N.A.  $0.26$  and a  $1/8$  of  $0.88$  N.A. The  $2/3$  is a remarkably fine lens which has no achromatic rival, even though it consists of only two pairs, and the  $1/8$ , which to my fancy is rather too high in power for its aperture, by far surpasses any similar achromatic lens. Now when we remember that the price of these two lenses together is only the modest sum of  $2\text{L. } 8\text{s.}$ , we are in a position to realize the great strides the manufacture of Microscope lenses has made quite recently. I hope to show a blow-fly's tongue under one of these presently.

In this country Swift has made use of Jena glass in the production of "Students'" lenses with great success; some dry  $1/6$  and  $1/8$  may be specially noted as having eclipsed every similar lens, and this without entailing any extra complication in construction.

Last week Mr. Baker sent me a new Reichert  $5\text{L.}$  oil-immersion  $1/15$  of  $1.25$  N.A., which on measurement I found to be a true  $1/12$  of  $1.24$  N.A. Under this lens I am exhibiting the secondaries of *Coscinodiscus Asteromphalus*.

This lens is the finest oil-immersion I have ever seen, excepting only the apochromatics. The spherical aberration is beautifully balanced, as can be seen by the large cone of illumination used, viz. the full aperture of the Zeiss achromatic condenser, with bull's-eye. Beyond this, however, the lens falls off. I know of no similar lens that will stand such a severe test. The object I have chosen has thick intercostal silex, and therefore is especially one to show up any colour left outstanding. The thicker the silex the stronger the colour (hence an excellent means of determining roughly the thickness of diatomic structures). Most lenses show this same object deeply coloured. With another object such as a *Navicula Rhomboides* (Cherry-field) in balsam, the silex on either side of the raphe will appear as very pale lilac. The lens also shows admirably a difficult test such as the secondaries on *Aulacodiscus Sturtii*. Such a lens cannot fail to play an important part in the microscopy of medical and science schools.

At present it is a short-tube lens, but by slightly closing the lenses it could be made into an objective for the long tube.

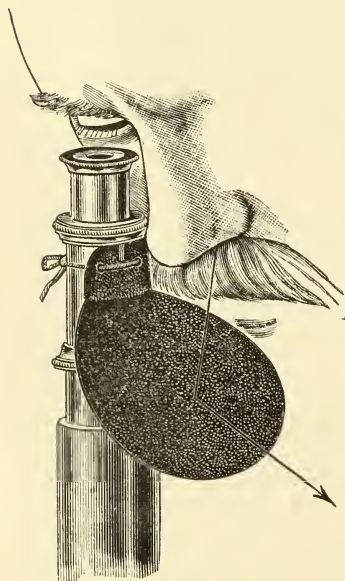
In conclusion let me ask you to go back to the microscopy of 1879, and then you will be better able to appreciate the great advance that has taken place in the improvement of the optical portion of the Microscope during the past ten years.

### (3) Illuminating and other Apparatus.

**Improvement in Abbe's Camera Lucida.\***—Dr. H. W. Heinsius has devised the following alteration in Abbe's camera, so that it can be readily removed from the eye-piece and replaced, and that without disturbing the coincidence of the two images. The special advantage of the arrangement is that it allows the details of the preparation to be directly inspected from time to time, an advantage which any one who has tried to draw with a camera of any sort will appreciate.

A ring of blackened brass of the same dimensions as the lower part of the camera, is connected by means of a joint to the arm which carries the mirror, and at the place where this is screwed to the mount of

FIG. 7.



the prism. The three binding screws pass through the new tube instead of the old one, and thus clamp the instrument to the Microscope. One slight alteration is necessary in order to prevent the neutral-tint glasses from falling out when the camera is turned up. The frames of these glasses are turned so that the latter are pushed in at the front and not at the top.

**Breath-screen.†**—In snub-nosed persons, says Dr. P. Schiemenz, the expired air tends to pass down parallel to the tube during a microscopical examination. The deposit of moisture, especially in winter, is sometimes annoying, and to obviate this the author recommends the adoption of a screen. This (fig. 7) may be made of a piece of stiff paper, the principal part of which is nearly circular (diameter about 8 cm.). The smaller portion is pierced by two holes, through which passes a string by which the apparatus is attached to the Microscope-tube.

This breath-screen can of course be easily fixed in or moved to any position.

**Siphon Apparatus for cultivating living organisms under the Microscope.‡**—Dr. J. af Klercker describes an apparatus which he has used for some time for observing living organisms under the Micro-

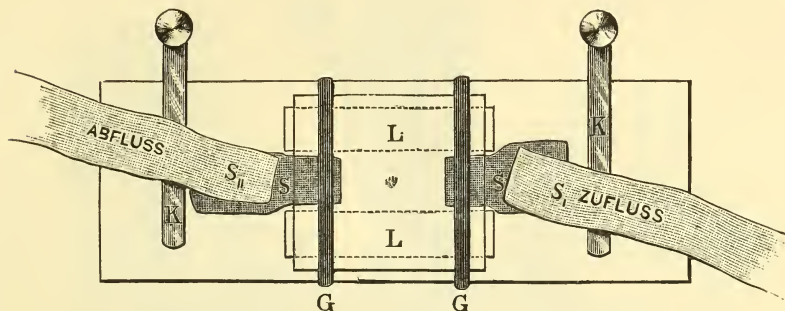
\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 36-7.

† T. c., pp. 37-8 (1 fig.).

‡ T. c., pp. 145-9 (3 figs.).

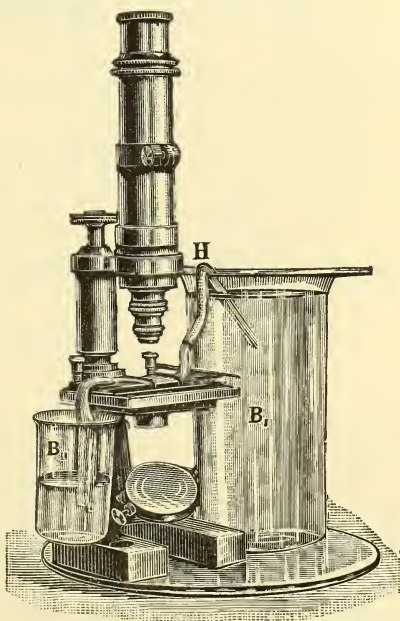
scope (figs. 8-10). The current of water for keeping the organisms alive is maintained by siphon action. Two oblong strips L of cover-glass (0.14 mm.), 28 mm. long and 6 mm. broad, are fixed parallel to

FIG. 8.



each other on a 3 by 1 in. slide, and at a distance of 8 mm., by means of Canada balsam. Within the channel thus formed is placed the object to be examined in a largish drop of water, and then a fairly large cover-glass superimposed. Should the object not be exactly in the centre, it can easily be pushed into the desired position by inserting a bristle or glass thread under the cover-glass. At each end of the channel a short piece of linen S is pushed under the cover, which is fastened to the slide by a couple of rubber rings G. A second slide is laid underneath the first, and the two connected by means of wax. The pair of slides are then fixed to the stage in the usual manner by the clamps K. A large glass vessel B<sub>1</sub>, the lip of which projects over the Microscope for about 5 cm., is filled with water, and in this hangs a doubly bent siphon H. Within the siphon is placed a strip of linen S<sub>1</sub>, the free end of which lies on the short strip S. By this arrangement a constant and regular inflow of water is assured. The volume of water passing through the siphon is easily regulated by jamming the linen strip S<sub>1</sub> into the siphon tube more or less tightly. The outflow is managed by means of another strip of linen S<sub>2</sub>, one end of which rests upon the short

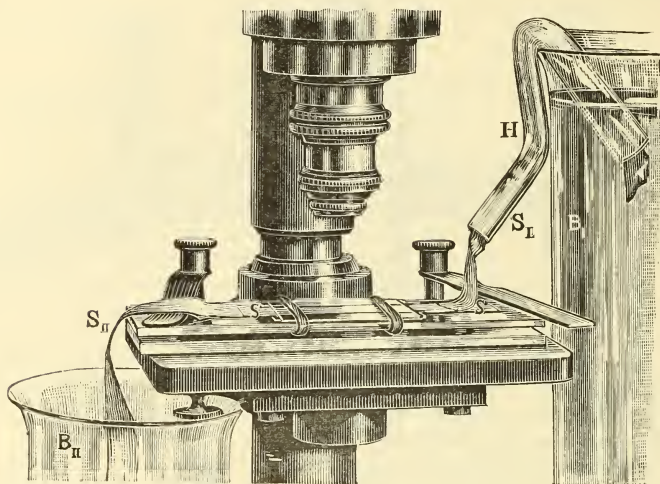
FIG. 9.





strip S, while the other dips down into a tumbler B<sub>II</sub> placed below and underneath the Microscope stage. With this apparatus nearly 50 cem.

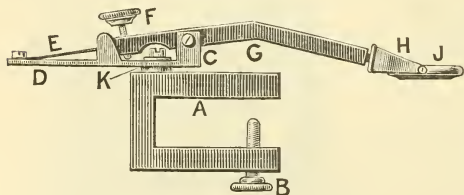
FIG. 10.



of water flows across the stage in the twenty-four hours, this being at the rate of about 3 cm. a minute.

**Schulze's Compressorium.\***—The object of this apparatus (fig. 11) is to apply pressure to an ordinary slide when upon the stage.

FIG. 11.



AB is a clamp to be attached to the side of the stage, carrying the piece CD, which rotates on the pin at K. The bent lever G is supported at C, and can be raised and lowered against the spring E by the screw F. At the other end of G is the fork H, in

which screws a ring J, with an opening of 12 mm.

The apparatus is screwed on the stage so that the opening of the ring lies on the cover-glass of the preparation, when by turning the screw F pressure can be applied as desired.

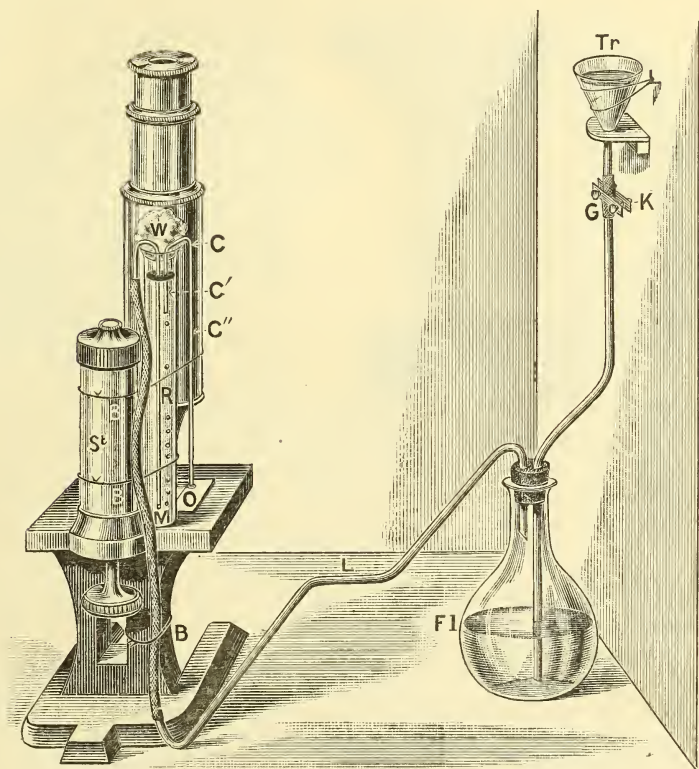
**Apparatus for examining the developmental stages of Infusoria under the Microscope.†**—Dr. L. Rhumbler devised the following apparatus for examining *Colpoda cucullus* and *C. Steinii* in hay infusion. To the vertical bar of the Microscope St (fig. 12), is fastened a medium sized

\* Behrens, Kossel, and Schiefferdecker, 'Das Mikroskop und die Methoden der mikroskopischen Untersuchung,' Band i., 8vo, Braunschweig, 1889, p. 53 (1 fig.).

† Zeitschr. f. Wiss. Zool., xlv. (1888) p. 549. Cf. Zeitschr. f. Wiss. Mikr., vi. (1889) pp 50-1.

test-tube R filled with sterilized hay infusion. Two very fine capillary glass tubes C C' C'' bent to a U shape are then placed in the test-tube. The longer leg of one rests close to the cover-glass on the slide O. This allows a very small quantity of water to flow down by capillary

FIG. 12.



action, and if it should be too much the excess may be removed by means of strips of blotting-paper placed on the other side of the cover-glass.

The rest of the arrangement is merely to supply air to the test-tube, as the infusoria tend to run towards the edge of the cover-glass if air be deficient. The water in the funnel Tr presses the air from the flask through the tube L out of the capillary point M in the test-tube.

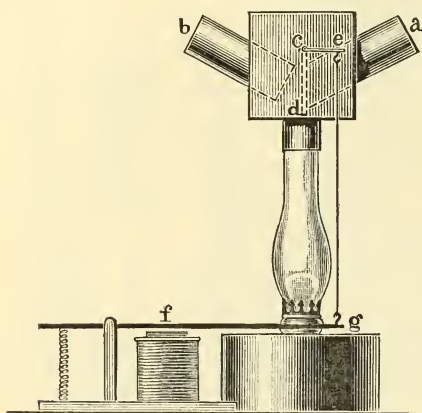
**Thermostat with Electro-magnetic Regulator.\***—N. Sacharoff has devised an instrument which is at the same time a modification and combination of Sahli and Scheibler's regulator (fig. 13). Upon the chimney of a mineral-oil lamp is placed a tin box, from opposite sides of which

\* Protokoll. d. Kaiserl. Kaukas. Med. Gesell., 1888, p. 111 (Russian). Cf. Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 49-50 (1 fig.).



project two tubes *a b*, the ends of which nearly meet inside, and the direction of which is obliquely upwards. Above these internal openings is a transverse bar *c*, from which is suspended the valve *cd*, and this when hanging free covers the mouth of the tube *a*, and the heat escapes through *b*. When, however,

FIG. 13.



*ce* is pressed down, the valve *cd* closes the aperture to *b*, and then the heat from the lamp escapes through *a*. If, therefore, a thermostat be connected with *b*, it can be warmed or cooled by the action of this valve. This action is governed by the electro-magnet. When the current is closed and the bar *f* drawn down by attraction, the latter pulls on *ge* and the valve closes *b*.

The opening and closing of the current is effected by means of a vessel filled with 500 g. of mercury. This vessel, which is placed within

the water-mantle of the thermostat, has a narrow tube let into it. Within the narrow tube, and also in the vessel of mercury, are placed two platinum wires; these are connected with magnet and battery. When the temperature rises the mercury ascends in the narrow tube and reaches the platinum wire. Hereby the current is closed and therefore the access of heat. As the thermostat cools the action of the valve is reversed and the heat again enters. The apparatus regulates to about  $1/2^{\circ}$ .

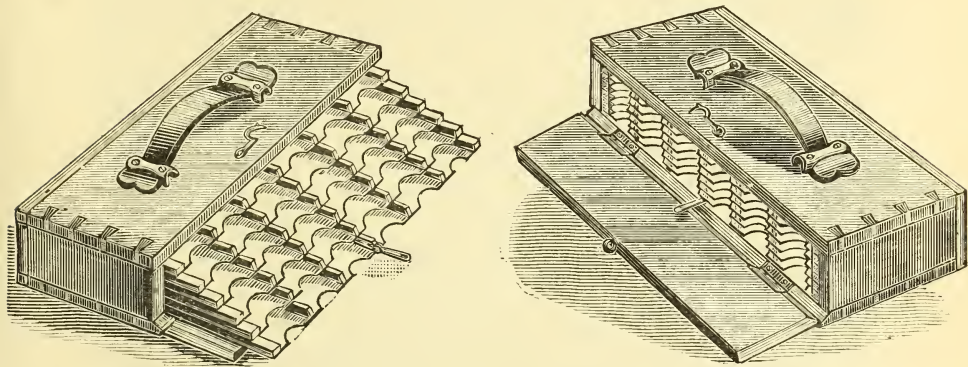
**Krutickij's Microspectroscope.\***—Herr P. Krutickij describes a micro-spectroscope which he invented sixteen years ago. Placed between the stage and the mirror it throws an objective spectrum on the slide, which is seen at the same time as the preparation examined. In order to facilitate the employment of any magnification which may be required, it is necessary to narrow the slit of the spectroscope in proportion to the magnification of the object. This is effected by screwing to the spectroscope an objective of the same power as that with which the object is observed. The spectroscope itself consists essentially of a combination of three prisms (two of crown-glass, the middle one of heavy flint-glass). The light is thrown from the mirror on to the slit (which is protected by a glass plate and can be narrowed to any extent by a screw), and is then concentrated by a lens on the prisms, dispersed by them, and thrown through the objective as a microscopically small spectrum upon the slide. The apparatus is provided with a divided ring, by turning which the slit is brought into the focus of the objective; and a contrivance to move the spectrum in a horizontal position in the field of view.

\* Script. Bot. Hort. Univ. Imp. Petropolitanae, ii., pp. 35-40, 1887-8. See Bot. Centralbl., xl. (1889) p. 10.

**Moseley's Object-box.**—This (fig. 14) is a new form of object-box invented and provisionally protected by Mr. E. Moseley.

The special feature of the box (which was exhibited at the November meeting by Messrs. W. Watson and Sons) is that, by drawing forward

FIG. 14.



the bottom tray all the others follow in series, displaying the labels of the slides. In the old form of object-box each tray has to be removed to pick out any object, but in Mr. Moseley's the object can be at once seen without any trouble. The box occupies no more space than the old form.

**Maddox's Simple Substage Condenser.\***—Dr. R. L. Maddox writes:—"On the supposition that the following remarks may be of interest, I beg to offer them to your notice. They are founded on the application of a rather novel kind of substage condenser for the Microscope, which has furnished me with some rather unexpected results, both visually and photographically. Whatever may be its real value, it has one claim which cannot be questioned, and that is its cost can be placed at zero. No doubt many of your readers have perused Professor Lowne's interesting article on "Interference Phenomena in Relation to True and False Images in Microscopy," reported in the Journal of the Quekett Microscopical Club for April of this year. Prof. Lowne suggests also a new theory for the formation of the diatom-image when it is brighter than the field, and that 'the cause of the positive image is that the diatom is illuminated from above, not from below. It is illuminated by reflected light from the upper surface of the front lens of the objective'; and the Professor cites an experiment showing the 'great illuminating power of the back of the front lens of an objective.' This surface of emergence of the front lens is a concave mirror, which condenses the reflected pencil upon the object. That such is the case to a certain extent is correct; but the following experiment will, I think, show it does not entirely suffice to form a bright image of the object in the case of diatoms. Having suggested to an eminent microscopist and photomicrographer the use of a cylindrical lens of short focus for a certain

\* Brit. Journ. of Photography, xxxvi. (1889) pp. 812-3.

purpose, and even hinting at a trial with a small piece of thermometer-tube retaining the mercury-column, under the supposition of the correctness of the argument used by Prof. Lowne, thinking it might be possible that the bright, reflecting, flat surface of the mercury within the tube would aid the object in view by producing the desired image, I determined to test the same. At the time I was too unwell to carry out my suggestions, but I did so at the earliest moment, and my object in this article is to state the results.

The only properly constructed cylindrical lens I possessed was of too long a focus for the purpose, which was to try and render evident some doubtful markings, dots, lines, or areas on the diatom *Amphipleura pellucida*. To construct a short-focus cylindrical lens means more time and trouble than I could give, so I cut off a piece of a thermometer-tube  $\frac{1}{2}$  in. long and from  $\frac{1}{6}$  to  $\frac{1}{5}$  in. in diameter, and having sealed in the small column of mercury, I mounted it centrally in a thin flat piece of ebonite, as the first thing to hand. It was let into a slot diametrically, cut exactly to fit the tube lengthwise, keeping its surface parallel to the surface of the ebonite. The tube was thus held longitudinally at its widest diameter, the flat face of the little mercury-column showing above and beneath. It was in this extemporized setting fitted on the top of the brass tube of a substage condenser without its lenses, but having its own rackwork, and being capable of rotation in the centering of the substage of the Microscope. Here I had a kind of cylindrical lens formed round one axis of revolution, the central portion being blocked out by the small column of mercury. No time was lost to now test its value as a simple substage condenser for use on lined objects, and also as it appeared to me useful to test Prof. Lowne's theory. After duly centering the mercury column, I placed on the stage a slide with *Pleurosigma balticum*, and by aid of the plane mirror and daylight, using the  $\frac{1}{5}$ -in. objective and No. 1 eye-piece, I noticed that the bright reflecting surface of the mercury in the little tube did not suffice to give by its own light, reflected from the back of the front lens, more than a very faint image of the diatom; but the moment the small tube was decentered, so as to place the mercury-column to one side, or just out of focus, I had a very beautiful image of the object, and could by rotation of the tube round the central axis of illumination easily bring out, separately, either the short horizontal lines or the longitudinal ones by alteration of this substage adaptation, or both, showing the markings or areas in squares. Another objective was tried, as Zeiss E, equal to about one-ninth. Here the image was more perfect, only from its larger numerical aperture there was less difficulty to separate the striation. The next trial was to go over the same ground again, using simply the divergent rays of the Microscope-lamp, and with the same result. The divergent rays were next made parallel by a bull's-eye condenser, also by a crossed lens before reaching the small tube, which rendered this image very bright.

Having thus far satisfied myself, I next cut a small piece from a solid glass rod of about the same diameter. This was mounted more carefully, and upon testing its use in the same manner, I was greatly surprised at its efficiency when used to illuminate the same object, and also other diatoms. The extreme brightness of the images with a  $\frac{1}{12}$  water-immersion made by Gundlach, and selected for me years since by



Mr. Winspear, optician, Hull, for photomicrography, when focused on *Pleurosigma formosum*, led me to test its value photographically. Unfortunately, I had to fall back on some old slow quarter-plates, and being without any guide as to exposure, I simply made use of my small camera arrangement, described in one of your almanacks, and attached it to the draw-tube of the Microscope, using for illumination a large paraffin lamp, and a crossed lens as a condenser. At the first trial a very fair image was obtained, using the developing solution described in the present 'British Journal of Photography Almanac,' 1890. It seemed evidently worth while to try another more magnified image, so I managed to centre a quarter-plate camera by means of a blackened card with a central dark-lined paper tube fitted to the draw-tube of the Microscope, and made to fill up the lens aperture in the camera. As soon as ready, I took a photomicrograph of *Pleurosigma formosum*, using the 1/12-in. bull's-eye condenser and lamp, the little rod being set parallel to one set of lines on the diatom. The result I inclose for your notice, as it possibly may be one of the first negatives you may have seen produced under such conditions. Unfortunately it is a trifle over-developed, but with the naked eye, or better with a lens, you will see the effect that can be obtained by such a simple piece of apparatus.

The value of the little rod as a condenser appears to me to rest chiefly in giving linear illumination of a convergent character, which can be directed in any position as regards the striation of lined objects. Some very curious effects can be brought out by keeping the eyes fixed on the object at the same time that the rod is gently rotated round the axis of the Microscope, and it is just possible some of the peculiarities in the structure of striated diatoms may be better brought out than with an all-round convergent illumination. There is one point that must be carefully observed to obtain the best result, which is to be careful to use it at its own focus, otherwise the image is pale or fogged. It is not pretended to offer this plan for anything more than a *costless* substitute for a *costly* piece of apparatus. It possesses a certain value, but is not intended to compete, in general excellence with a first-rate achromatic substage condenser. You will be able to judge for yourself. I should have liked to have tested rods of coloured glass, but could not put my hands on any suitable; and there remains yet to try the rod with a right-angle prism, instead of the plane mirror or parallel light by means of the bull's-eye condenser. To find the best position of the rod requires a little trouble."

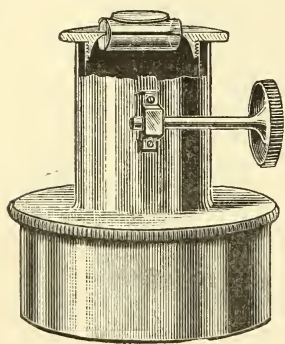
**Maddox's Small Glass Rod Illuminator.**—Dr. Maddox refers to the preceding as follows:—"In this age of rapidly advancing microscopy may I for a few moments crave the attention of the Fellows of the Society to the claims of a small piece, 1/2 in. long, of solid white or blue glass rod, about 1/5 or 1/6 in. in diameter, when used as an illuminator, and substituted for an ordinary substage achromatic condenser. I ask this permission as several errors have crept into the pages of a weekly contemporary journal, through the incorrect statements of a writer who noticed an article on the use of the white glass rod, contributed to the 'British Journal of Photography' of December 13th, 1889.

A few days since I mounted a piece of blue glass rod in the same manner, i. e. by fitting it horizontally at its widest diameter into a thin cell, which screws on the top of a substage fitting which has its own

rackwork, and this, when inserted into the substage support, is capable of being rotated; thus the centering, focusing, and position—matters of importance—are secured. By both plans I have been enabled to examine many lined objects, whether the appearance be due to ridges, or areas as elevations or depressions, and I have reason to suppose, if employed properly and patiently, either will render visible any markings any objective, at least of the old school, is capable of revealing.

The rod has been used with the  $1/2$ ,  $1/5$ ,  $1/9$ , and  $1/12$  in. water-immersion, the latter photographically, the radiant being a small paraffin lamp, and between it and the rod a large No. 1 eye-piece, or a crossed lens, or a bull's-eye condenser. After numerous trials, preference was given to the first.

FIG. 15.



The white rod has also been made into an immersion illuminator by fixing on the top horizontal edge a small cover-glass (fig. 15). Two of the negatives accompanying this were taken by it used thus, as an immersion illuminator; the others by the blue rod, dry.

I have been rather surprised at the minutiae either will reveal, as small bars of silix extending into the large areas in some of the fragments from the Oamaru deposit; the secondary markings in *Triceratium*, &c., are remarkably well shown by

either rod, and they well define the areas in *Navicula rhomboides*, &c. It appears to act as a narrow convergent central line of light, which, by careful manipulation, yields at certain points of rotation, excellent definition.

Possibly what I have said will be called in question, but it must be understood I do not claim for the rod more than has been stated, and trust others, if induced to try it, may find it has not been exaggerated."

#### (4) Photomicrography.

**Photomicrography.\***—Dr. Th. Kilt reviews the history of photomicroscopy and the present condition of its technique. He describes in detail the apparatus of Zeiss and Klönne and Müller, light-filters, and orthochromatic plates; the various copying methods are thoroughly discussed. The specimens of photography given by the author show the great superiority of this method over drawing, and it is safe to prophesy that if the improvements of this art can be continued, it will soon sweep the field for bacteriological and histological illustrations. Although, from motives of economy, photozincography, which only imperfectly reproduces the delicacy of the negative, was selected, the illustrations given are extremely clear and sharp.

**Silver Combinations of Eosin.†**—Dr. E. Zettnow finds that the orthochromatic power of eosin-silver plates is due to the erythrosin or its silver combinations, and not to the eosin. The erythrosin plates

\* 'Encyclopädie d. gesamm. Thierheilkunde u. Thierzucht,' Wien u. Leipzig, 1889. Cf. Zeitschr. f. Wiss. Mikr., vi. (1889) p. 193.

† Zeitschr. f. Wiss. Mikr., vi. (1889) p. 193, from Photogr. Correspondenz, 1889.



are moreover extremely sensitive to yellow, and as long as this kind of light predominates, the excellence of their delineation is unsurpassable. By their aid sharp negatives can be obtained with a mineral oil lamp and the ordinary low-power objectives, and this without a filter. With sunlight, or as soon as the light becomes impregnated with many blue rays, they fail.

#### (5) Microscopical Optics and Manipulation.

*Amphipleura pellucida* and *Pleurosigma angulatum*.—Dr. H. Van Heurck sent for exhibition at the December meeting some remarkable photomicrographs of *Amphipleura pellucida* and *Pleurosigma angulatum* taken with the new Zeiss's 1/10 in. objective of 1.63 N.A.

Monochromatic sunlight was used. Medium for the preparations 2.4. For those of *Amphipleura* moderately oblique light was used with magnifying powers of 2000 and 3000 diameters. The *Pleurosigma* photographs were taken with strictly axial light and a small aperture of the diaphragm, the magnification being 3000, 10,000, and 15,000.

The photographs of *Amphipleura* show the valve completely resolved in beads (cf. *supra*, p. 91), while those of *Pleurosigma* show details not hitherto observed.

Dr. Van Heurck considers that his "conclusions as to *Pleurosigma* are now complete and positive and may be summed up as follows:—

(1) The alveoles of *P. angulatum* are hexagonal, at any rate in the place where the two layers of the valve unite.

(2) The intermediate beads are produced by bad focusing of the angles of the alveoles."

The following is a translation of a communication made by Dr. Van Heurck to the Belgian Society of Microscopy:—

"I have the pleasure to submit a new series of photographs of *Pleurosigma angulatum* obtained with the 2.5 mm. objective 1.63 N.A.

On studying this diatom attentively, I observed a very singular appearance; the alveoles or beads showed themselves in the form of very minute points, and were surrounded by a ring of six secondary beads when each alveole was viewed separately. If, however, the whole valve was viewed it was seen that the secondary beads were really intermediate between two principal adjacent layers of the valves.

I thought at first that this appearance of structure was new, but later I saw that a similar appearance existed on the margin of the valve photographed by Dr. R. Zeiss (5000 diameters), and which is figured in his 'Atlas of Photomicrography.'

Photograph No. 1 reproduces the above appearance. No. 2 shows it under a power of 10,000 and with an exact focus. In No. 3 the focus was purposely altered so as to show the secondary beads better.

How is this structure to be explained?

If the photograph No. 2 is attentively observed it will be seen that the alveoles are not round as has been generally believed in modern times, but that they present sensible angles."

An absolutely exact focus (photograph No. 4) shows that the opinion of the old microscopists was well founded and that the alveoles are really hexagonal.

This hexagonal form being admitted, an easy explanation is obtained

of the secondary beads, which are produced by the imperfect focusing of the angles of the network, that is, by the places where two lines run into one another. To verify this hypothesis I have studied with the same objective a great number of large diatoms where the structure allows of no doubt, and I have found in *Coscinodiscus excentricus* the confirmation of my assertion.

The structure of this diatom is well known. With low-power objectives it is seen to present large hexagons. The valve is very convex, and by regulating the focus suitably we can obtain at the same time all the appearances from the real hexagons to the isolated point surrounded by six illusory intermediate beads. It is this which is shown in photograph No. 5.

The last photograph, No. 6, shows that the valve of *Pleurosigma* is formed of two layers, and that the alveoles are hollowed out in the substance of the valve. The photograph shows a valve where the lower layer bears, on a part of the surface only, a fragment of the upper layer. The hexagonal form of the alveoles is seen round the median nodule. It may therefore be considered that the form of the alveoles is hexagonal at the point of union of the two layers, and that each alveole terminates above and below very gradually in the form of a dome. This is what my latest researches seem to demonstrate. I hope to be able, as soon as I have leisure, to send photographs in support of this view.

List of photographs:—

1. *Pleurosigma angulatum* W. Sm.  $\times 3000$ .
2. Ditto—exact focus on the intermediate illusory beads—about  $\times 10,000$ .
3. Ditto—out of focus, to show the intermediate beads better—about  $\times 10,000$ .
4. Ditto—focus on the hexagon—about  $\times 15,000$ .
5. Ditto—showing partially the two layers of the valve.
6. *Coscinodiscus excentricus*."

**Structure of Diatom Valves.**—Dr. Van Heurck also sends us the following paper:—

In my 'Synopsis of Diatoms' I showed that in the large *Cryptoraphides*, for instance, *Coscinodiscus*, we can clearly distinguish three parts: an upper membrane, a lower membrane, and an intermediate layer, and these may be identified when isolated, either wholly or partially, in certain gatherings.

We know from Prof. Abbe's theory that the Microscope *alone* does not enable us to determine with certainty the structure of minute forms. But though technical means fail us, we can still make estimations by analogy as we do in most of the sciences. The study of large forms authorizes us to infer that the structure of the more delicate forms may be identical or at least very similar.

It has frequently happened that the examination of favourable fractures has enabled me to confirm these views, and they have also been confirmed by careful observers, such as Deby, Cox, and others. The portion of the intermediate layer photographed by Mr. T. F. Smith, and figured as No. 5 in his note on the *Pleurosigma* valve, is a case in point that may be considered quite conclusive.

The more recent and powerful optical means placed at our disposal by the house of Zeiss, of Jena, allow us to go a step further in the study

of the valve, firstly, in producing an optical image which is more complete and hence more real, and further by reducing more nearly to a mathematical plane the portion of the valve that can be seen with one and the same focal adjustment.

The new results obtained and confirmed by photography—which all serious observers now regard as the best criterion—still further simplify our opinions, and enable me to summarize them as follows:—

(1) Diatom valves consist of two membranes or thin films, and of an intermediate layer, the latter being pierced with openings.

The outer membrane, which is often very delicate, may readily be destroyed by the action of acids in cleaning, or by friction, &c. It may be also that this membrane exists only in a very rudimentary state. Specialists on this subject are generally agreed in supposing that these membranes may be sufficiently permeable to allow circulation by end-osmose from the interior to the exterior of the valve, though they have no real openings during the life of the diatom and whilst it remains intact.

(2) When the openings of the interior portion are arranged in alternate rows, they assume the hexagonal form; when in straight rows, then the openings are square or oblong.

The hexagonal form, which occurs so frequently in nature, seems to be the typical form of the openings in the interior portion, and this form obtains mostly in large valves, which are not otherwise provided with strengthening ribs. Even in the forms having square openings we frequently perceive deviations, and the tendency to recur to the hexagonal type on certain portions of the valve. It may be that the interior consists of several layers superposed, formed successively and very closely joined, but so far I have not met with any form exhibiting superposed layers differing from each other in type.

This description seems to me to represent in broad outline the structure of diatom valves. But this structure may appear complicated, either by the presence of secondary internal valves ("Regenerations-hülle"), or by deposits of silica on various parts of the valve. These deposits originate the "thorns" met with in divers forms (such as *Triceratium*), the rings found on the under membrane of certain forms of *Coscinodiscus*,\* and the anastomosed ribs of *Navicula aspera* Ehr. (*Stauroneis pulchella* W. Sm.†) &c.

All these deposits are merely secondary silicious products which have nothing to do with modifying the general structure of the valve in its primordial elements.

#### *Description of the Plates.*

Plate II.—(1) *Amphipleura pellucida* Kütz. resolved into beads  $\times 2000$ . The insufficient magnification shows a good general view, but the beads are not so sharp in the print as in the negative.

\* I have observed the rings in *Pleurosigma formosum* referred to by Mr. T. F. Smith, but the new objective (1.63 N.A., medium 2.4) when employed on valves that were purposely broken, shows them lying flat on the under membrane, precisely as in *Coscinodiscus*. Possibly these rings are portions of secondary valves. I have not been able yet to determine the point.

† The valves of *Navicula aspera* Ehr. appear at first sight very complicated, and they have hitherto been erroneously figured by all writers. My latest examinations would show that the appearances observed are due to the mixing up in vision of more or less distinct views of ribs or thickenings regularly anastomosed so as to form rings more or less alternate.

2. *ib.*  $\times 3000$ .

3. *ib.*  $\times 8000$ , upper part of the valve, showing square beads identical with those of *Amphipleura Lindheimeri* Grun.

4. *Amphipleura Lindheimeri* Grun.  $\times 2500$ .

5. *Surirella gemma* Ehr., about  $\times 1000$ .

6. *Pleurosigma angulatum*, in hexagons, about  $\times 10,000$ .

7. *Van Heurckia crassinervis* Bréb. (*Frustulia saxonica* Rabh.)  $\times 200$ .

8. *Van Heurckia crassinervis*, Bréb., about  $\times 6000$ .

In all the photographs the focus was upon the intermediate layer, and here and there in most of them the gradations of form are shown between squares and hexagons.

Plate III.—*Pleurosigma angulatum* W. Sm.  $\times 2000$ . On the right of the centre the illusory intermediate beads are seen at the same time as the real beads (the openings), of hexagonal form.

The photographs were all produced with Zeiss's new apochromatic 1/10 in. of 1.63 N.A. Monochromatic sunlight. Compensating eye-piece (special) 12. Condenser 1.6 N.A.

The preparations were all in a medium of 2.4. Cover-glass and slides of flint, 1.72. Diatoms melted into the cover-glass softened by heat.

Ilford dry plates, developed with hydroquinone and eosine solution as supplied by Mercier, of Paris.

**Resolving Power a "Superfotation."**—The following extract from M. A. Zune's 'Traité de Microscopie' (1889) should be interesting to microscopists.

"Resolving power. We regret not to have the necessary authority to erase this word from the dictionary of microscopists, since it appears to us to constitute an entire superfotation. To say of an objective that it has resolving power is, according to most authors, to attribute to it the power of isolating so to say one from another the finest details of structure on the surface of a transparent object such as striæ, fibrillæ, depressions, reliefs, &c.; but an objective which defines well in the complete sense of the word, ought it not to resolve perfectly?"

This carries a long way further the error on which we commented in the case of the Quekett discussions, where, however, it was not proposed to abolish the term "resolving power"! As we explained then, and shall probably have to repeat again, an objective may have perfect defining power, and yet, by reason of its want of aperture, it will be unable to show particular markings. It defines all that it can take up, but cannot define what is not imaged by it.

It would be possible, no doubt, to arrange that "definition" should be considered to include "resolving power," but nothing would be gained by confusing the two terms, especially as we have already the term suggested by Prof. Abbe—delineating power—to denote the combination of the two qualities, an objective having large delineating power when it both defines well and has large aperture.

The author's views are in other respects peculiar, as he is of opinion that "an objective of large angle, *well constructed*, will—all other things being equal—show details in depth as well as it will show those on the surface."



## (6) Miscellaneous.

**Paris Exhibition, 1889.**—The following English opticians obtained rewards at the last Paris Exhibition, though not necessarily for Microscopes alone:—

*Grand Prize.*—Messrs. Ross & Co.

*Gold Medals.*—Mr. J. H. Dallmeyer, Mr. J. Pillischer, and Messrs. Watson & Sons.\*

**Carlisle Microscopical Society.**—The official report which we have received embodies a *résumé* of the work done by this vigorous provincial Society since its foundation in the year 1881, and especially since its affiliation with the Royal Microscopical Society in 1883. The Society was inaugurated by a public address delivered to a large audience by its first President, the Rev. Canon Carr, who afterwards gave a series of educational papers on Vegetable Histology. Papers have been read at successive meetings by various members of the Society on such subjects as the adulteration of food, water, coal fossils, trichina, diseases of plants, animal physiology, photomicrography, the Microscope in manufactures, slide-mounting, and others too numerous to mention. Frequent excursions have been made by the Society to collect material for microscopical examination. Two public lectures have been delivered to crowded audiences by the Rev. Dr. Dallinger, and one by Sir Robert S. Ball, Astronomer Royal for Ireland, who has also promised to give another in the course of the present session. The aim of the Society has been eminently a practical one, and much earnest work has been done. The President of the Society is C. S. Hall, Esq., and the Hon. Vice-Presidents, Prof. Pasteur and the Rev. Dr. Dallinger.

**Orthography of the Microscope.**—There is no word which is so variously spelt as "Microscope" or (with "microscopical," &c.) so often misspelt by printers.

The form "Microscope" occurs times out of number.

The Germans, apart from the standard form of "Mikroskop," also spell it "Mikroskop," "Microskop," and "Microscop."

"Microscope" appears in Proc. Amer. Soc. Micr., 1886.

"Mikrospischen" is found in Stenglein's 'Anleitung,' 1887.

"Mirosopical" in Amer. Mon. Micr. Journ., viii. (1887) p. 49, and this Journal, 1887, p. 1039.

"Microscop" in 'The Microscope,' 1888, p. 108.

"Mikrokopiker" in 'Flora,' 1888, p. 39.

**Mr. Crisp and this Journal.**—The 'Athenæum' says:—"Microscopists, abroad as well as at home, will hear with great regret that Mr. Frank Crisp is about to resign the office of Secretary to the Royal Microscopical Society, which he has held for twelve years. During that period the character and position of the Society have been greatly improved, and the numerous microscopical societies which have sprung up elsewhere have come to regard it as their common parent; the number of its Fellows has been doubled, and its Journal has been converted into one of the most useful aids to research which are now put into the hands

\* Cf. Journ. d. Microgr., xiii. (1889) pp. 481-93, and Mr. J. Mayall, junr., this Journal, 1889, pp. 851-2.



of working biologists. For twelve years this Journal has averaged a thousand pages in each volume, and its circulation is understood to be more than one thousand copies. This result, it is generally known, has only been obtained by the yearly expenditure of a sum of money larger than the annual income of the Society; Mr. Crisp's banker alone, in all probability, knows how large that sum is. But Mr. Crisp has not only given money; he has also devoted a large amount of time to editing and improving the character of the Journal, and by his own contributions and criticisms has done a great deal in making intelligible to microscopists the modern theories of the Microscope. His retirement from, no less than his election to, the office which he holds marks a critical period in the history of the Society. But though his legal duties are so much increased as to leave him no choice, he will still be intimately associated with the Society, as he is willing to act as its Treasurer, and we may be sure that his interest in it is in no way abated." \*

### β. Technique.†

#### (1) Collecting Objects, including Culture Processes.

**Cultivation of Actinomyces.‡**—Dr. Kischensky inoculated blood-serum and agar to which 6 per cent. of glycerin had been added with actinomyces granules. The next day evidences of growth were observed. In the course of a few days filaments associated with coccus forms were seen under the Microscope, and after two or three weeks the ends of the filaments were observed to possess bulb-shaped expansions (involution forms). In cultivations on potato the fungus grew in the form of yellowish granules. In gelatin at 39° C. the filaments seemed to grow in a radiate way, and sometimes showed bulbous expansions at their ends. The filaments were easily stained by Gram's method.

Whether these cultivations were really pure cultivations of actinomyces seems doubtful at present, as inoculation experiments were not tried.

**Pure Cultivation of Actinomyces.§**—For some months past, says Dr. O. Bujwid, "I have easily obtained pure cultivations of actinomyces, and have further ascertained the important fact that it is an anaerobic fungus."

The method adopted by the author was to take some of the granules from the abscess-pus of a person suffering from actinomycosis and cultivate them in ordinary gelatin, ordinary and glycerized agar, sterilized milk and potato at a temperature of 36° C. For some of the tubes 10 per cent. pyrogallie acid was used to absorb the oxygen (Buchner's method).|| In these anaerobic cultivations the points inoculated were observed to have swelled in about 48 hours, while the rest of the tubes only showed copious growth of *Staphylococcus aureus*, *S. albus*, and some sort of rodlet.

\* Athenæum, 1890, Jan. 11, p. 53.

† This subdivision contains (1) Collecting Objects, including Culture Processes; (2) Preparing Objects; (3) Cutting, including Imbedding and Microtomes; (4) Staining and Injecting; (5) Mounting, including slides, preservative fluids, &c.; (6) Miscellaneous.

‡ Arch. f. Experimentelle Pathol. u. Pharmacol., xxvi. (1889) p. 79.

§ Centralbl. f. Bakteriöl. u. Parasitenk., vi. (1889) pp. 630-3 (2 photos).

|| See this Journal, 1888, p. 1039.

The granules increased in size and in a few weeks had become pretty large yellowish-white grains which penetrated within the substance of the agar so that they could only be removed with difficulty. Microscopical examination afterwards showed that the colonies grew radiately, forming branches and mycelia like some moulds do, whence it would seem highly probable that *Actinomyces* is a sort of mould fungus. This appearance does not resemble that of the club-shaped elements found in human pus and in the fresh pus and nodules of ruminants, but is identical with that of the nodules found in human organs or in human pus after treatment with caustic soda.

In older agar cultivations bulb-shaped expansions formed on the ends of the filaments, but whether these were gonidia or an involution form the author cannot yet say.

Two photographs from sections magnified 340 and 840 times were obtained as follows:—An agar cultivation three weeks old was cut up into small pieces and placed in absolute alcohol for 48 hours; then for 24 hours in 1 per cent. and afterwards for six hours in 5 per cent. photoxylin solution. From these pieces sections were made in the usual way and removed from spirit to a slide, and when they were dry (20–30 minutes) were stained by Gram's method. The photographs were taken with Zeiss's apparatus by sunlight. Zettnow's light-filter was used and Attout-Tailfer's isochromatic plates.

**Cultivation of Typhoid Bacillus in Sewer Water.\***—M. L. Olivier states that sewer water is a very favourable medium for the cultivation of typhoid bacilli; they develop in it quite as well as in bouillon.

## (2) Preparing Objects.

**Preparation of Cells for showing the Division of Nuclei and the Formation of Spermatozoa.†**—For examining cell-fission, and the formation of semen in the hermaphrodite glands, Dr. G. Platner recommends immersion in the strong Flemming's mixture. Fresh pieces of gland cut up small, if necessary, are placed in the mixture for an hour; the fluid is then diluted with three or four times its bulk of water, and allowed to harden further for 24 hours longer. They are then thoroughly washed in the usual manner, and afterwards transferred to spirit of increasing strength. The best nuclear stain is logwood, and the author recommends Apáthy's modification of Heidenhain's hæmatoxylin (hæmatox. crystals 1, absolute alcohol 70, aq. dest. 30, the fluid to be kept in dark-coloured bottles).

The objects were stained *in toto* for 24 hours, and afterwards acted on by a 1 per cent. alcoholic solution of bichromate of potash. This solution is made by mixing 10 parts of bichromate with 300 of distilled water, and then, when required, diluting 30 ccm. of it with 70 ccm. of strong spirit. This gives the proper colour after acting for 12 hours. If a lighter stain be desired, it must work for 24 hours. The objects are then transferred to 70 per cent. spirit, and kept in the dark for one or more days. After this they are dehydrated in absolute alcohol, and

\* Comptes Rendus Soc. Biol. Paris, 1889, No. 27; Centrabl. f. Bakteriöl. u. Parasitenk., vi. (1889) p. 519.

† Arch. f. Mikr. Anat., xxxiii. (1889) pp. 125–52 (2 pls.).

then soaked in cedar oil. They are next soaked in paraffin for 20 minutes. The series of sections are stuck on the slide with castor-oil collodion, and after the removal of the paraffin with xylol, mounted in balsam.

For studying the division of the nuclei in the Malpighian vessels of *Dytiscus marginalis*, the author used Kleinenberg's picrosulphuric acid for hardening. This was found specially advantageous in that it decolorized the dark-brown granules which beset the cell-plasma.

By staining with borax-carmin and then treating with acidulated alcohol, a beautiful colour was obtained.

**Preservation of Mucous Granules in Secretory Cells.\***—Mr. J. N. Langley advises the following method for preserving mucous granules in secretory cells. The animal is killed by bleeding or decapitation. A small piece is then snipped off a salivary gland, the piece having been previously pierced with a threaded needle. The piece of gland is suspended by the thread in a bottle, which contains some 2 per cent. osmic acid. The thread is fixed between the stopper and neck of bottle, and the piece of gland hangs just above the level of the fluid. The object is hardened in about 24 hours. It is then removed, washed for a few minutes in water, and then for 15 minutes apiece in 30 per cent. and 50 per cent. spirit. Next, for half an hour apiece in 75 and 95 per cent. alcohol; finally for one or two hours in absolute alcohol. The preparation is then soaked for half to one hour in benzol previously to being imbedded in hard paraffin. The series of sections are fixed on the slide with albumen stained with methylen-blue, and mounted in balsam. Or the paraffin may be dissolved out by means of benzol or turpentine. This method is said to give good results with mucous cells from the mucosa of many of the lower vertebrata.

**Removing the Jelly and Shell from Frogs' Eggs.†**—The method for removing the coverings from frogs' eggs recommended by Prof. F. Blochmann is essentially the same as that previously advocated by Prof. C. O. Whitman.

The author employs eau de Javelle, a solution of hypochlorite of potash, while Whitman used sodium hypochlorite. The ova which have been preserved in chrom-osmium acetic acid, and been well washed in water, are placed in the solution, twice or thrice diluted, and then shaken up by inverting the vessel. The eggs, freed from their gelatinoid coat, sink to the bottom in 15 to 30 minutes. They are then very carefully washed in water, and afterwards transferred to strong spirit. If the eggs be kept in the dark the chromic acid is removed more effectually. The author recommends borax-carmin for staining. Hæmatoxylin is not suitable.

**Carbonate of Ammonia for demonstrating Sarcolemma.‡**—Prof. B. Solger recommends a cold saturated solution of ammonia carbonate for demonstrating the sarcolemma of frog's muscle. In this solution the muscle is placed for 3 to 5 minutes, and having been teased out, examined under the Microscope. The reaction is more complete if the animal be previously kept for several weeks in captivity.

\* Journal Physiol., x. (1889) pp. v. and vi.

† Zool. Anzeig., xii. (1889) p. 269.

‡ Zeitschr. f. Wiss. Mikr., vi. (1889) p. 189.

**Demonstrating the Neurokeratin Network of Nerve-fibres.\***—Dr. G. Platner advises the following procedure for demonstrating the neurokeratin network.

Thin fresh pieces of nerve, freed from connective tissue and fat, are placed in the following solution—Liquor ferri perchloridi 1 part, distilled water or rectified spirit 3–4 parts. In this the pieces of nerve are left for days to weeks. The iron chloride is then to be thoroughly washed out so that no trace of iron can be chemically detected in the water or spirit. After this the pieces are to be kept till wanted in spirit.

The best stains for nerves thus manipulated are “Echtgrün,” a dinitroresorcin which in combination with the iron still remaining in the tissues gives a green colour, and alizarin, which imparts a deep violet hue.

To use dinitroresorcin, a supersaturated solution of the solid pigment is made in 75 per cent. alcohol. In this solution large pieces of tissue require to lie for several weeks. When thoroughly freed from iron the immersed pieces gradually become dark green, but the fluid itself exhibits no trace of green. After having been dehydrated, the pieces are imbedded, and sections, both longitudinal and transverse, made. In transverse section, the axis cylinder is stained a dark emerald green, and from this radiate outwards to the medullary sheath, numerous green delicate filaments, the neurokeratin network. In longitudinal section the same network is shown.

The stain is fairly resistant to acid and alkaline reagents, and the different methods of hardening do not exclude the use of the perchloride solution.

**Preparing the Silk-glands of Araneida.†**—Dr. C. Apstein, in making a macroscopical examination of the living animal, opened the body under water and then removed the heart, intestine, liver, and organs of generation. An addition of some drops of sublimate to the water imparted to the previously glass-like spinning-glands a milky appearance. Alcohol-material was prepared under 35 per cent. spirit. For sectioning the author prepared the animals with hot water, boiling them from 1/2–3 minutes, according to size, and then imbedding in paraffin, after passing them through turpentine or chloroform. The author cautions against using cedar-oil, as it is a poor solvent of paraffin. Borax-carminé, and after-staining with hæmatoxylin, are recommended for staining.

The statement that the silk-threads of the glandulæ pyriformes consist of a double substance is interesting, since the secretion from the upper part of the gland forms a solid non-staining cord, while the cells from the lower parts of the glands secrete a tubular filament which is clearly stained. The author verified this in different species.

**Preserving Actiniæ.‡**—Dr. J. P. McMurich recommends the collector of Actinians who has not the time to properly carry out the narcotizing methods to act as follows. After noting general characteristics, place the animal in a jar just wide enough to allow of its complete

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 186–8.

† Inaugural-Dissert. Kiel, 1889. Cf. Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 199–200.

‡ Journal of Morphology, iii. (1889) pp. 2–3.



expansion, and with just enough water to cover it when fully expanded. A glass syringe filled with Perenyi's fluid should have its nozzle quickly inserted into the mouth of the Actinian, and the contents should be rapidly injected. At the same time, if possible, a quantity of the same fluid should be poured over the animal. After half an hour the animal should be treated successively with 50, 70, and 90 per cent. alcohol, care being taken to inject a considerable quantity of the spirits into the interior at each change. Though considerable contraction and loss of colour ordinarily follow the use of this method, the parts are satisfactorily preserved for future histological study.

**Demonstrating Cyclosis in *Vallisneria spiralis*.**\*—Prof. S. Lockwood says that Mr. F. W. Devoe is able to show the circulation in this plant to the best advantage. "Having selected a bit of a leaf, not too mature, he shaves off one side with a sharp knife, although a razor is better. It is then put on a slide, the shaven side up. A drop or two of clean water and a cover-glass of medium thickness with good illumination follows, Mr. Devoe using a prism illuminator. Begin with a 6/10 objective, and continue to a 1/6 or a 1/10, and a vision is got of a startling clearness. The vivid individuality of each bioplasmic molecule and the mystic, almost solemn movement of this pellucid stream of infinities of life, form a sensational picture of which the beholder never tires."

**Cleaning Diatoms from Sand.**†—Mr. Norman N. Mason communicates the following method:—

After removal of the organic matter with acid by the usual methods, add to the diatoms and sand in a large bottle, thirty, forty, or fifty times the quantity, by measure, of water, and gently shake until they are mixed. This water, with the diatoms and sand kept suspended by an occasional shake, is slowly poured in a small stream upon the upper end of a strip of clean glass, 3 ft. long by 3 in. wide, and securely supported. The upper end of the glass should be from 1/8 to 1/4 in. higher than the lower end, and the glass should be level transversely. Beneath the lower end place any convenient receiver. The water and diatoms will pass into the receiver. The sand, which will form little bars on the glass, must be removed occasionally, as it gradually creeps towards the lower end of the glass, and there would eventually pass into the receiver.

The loss of diatoms will be very small. Usually one pouring is sufficient for cleaning. The sand can be re-washed if necessary, or a little clear water run over the sand on the glass strip will carry forward almost the last diatom; but this will scarcely pay for the trouble. A short piece of glass will cause a failure, and too great an incline will be found almost as bad.

**Preparing Crystals of Salicine.**—Dr. F. L. James a few years ago ‡ described a phenomenal class of crystals produced from salicine. The process is now stated § to depend on bringing a saturated solution of salicine made with distilled water in contact with cold below the

\* The Microscope, ix. (1889) pp. 327-8.

† Journ. New York Mier. Soc., v. (1889) p. 116.

‡ See this Journal, 1887, p. 507. § Amer. Mon. Mier. Journ., x. (1889) p. 214.

freezing-point, and the explanation is, that the rapid congelation of the water interferes with the usual arrangement of the crystals, producing a wonderful series, which are entirely unlike any forms resulting from crystallization at the ordinary temperature.

### (3) Cutting, including Imbedding and Microtomes.

#### Dextrin as an Imbedding Material for the Freezing Microtome.\*

—Mr. T. L. Webb says that by taking an aqueous solution of carbolic acid (1 in 40) and dissolving therein sufficient dextrin to make a thick syrup, a medium is obtained which is superior to the time-honoured gum and sugar in three ways. It freezes so hard as to give a firm support without being too hard. It keeps better than gum. It is much cheaper, costing only 4*d.* a pound, whilst powdered gum acacia costs 5*s.* Dextrin dissolves but slowly in cold water, so that a gentle heat is advisable when making the mucilage.

**Imbedding in Celloidin.**†—Dr. A. Florman recommends the following procedure for imbedding pieces of animal tissue in celloidin so as to obtain thin sections. After hardening the tissue in absolute alcohol, pieces about 3 mm. thick are placed for some hours in absolute alcohol, and after this in a test-tube containing a mixture of 3 parts ether and 1 part alcohol. In a couple of days some celloidin solution is added until the mixture is about as thick as a thin syrup. Herein the pieces remain for 14 days or longer, when more celloidin is poured in to make a thicker solution. After 4–8 days the contents of the test-tube are turned into a shallow glass capsule, wherein the celloidin solution must form a layer of 10–12 mm. thick over the preparation. The pieces having been arranged in the desired position, the capsule is covered with a glass plate, a cover-glass being interposed so as to allow of slow evaporation of the celloidin solvents. In 2 or 3 days' time a consistent mass free from air-bubbles is obtained, and from this the pieces are cut out so that each is surrounded by a layer of celloidin at least 3 cm. thick. When removed their under surface is to be daubed over with a thick solution of celloidin, so as to make all the surfaces of the same width. The pieces are replaced in the capsule to allow the new layer to become consolidated by evaporation of the ether and alcohol. Pieces thus prepared will have the consistence of cartilage, and sections from a block the sides of which are 1.5 cm. can be made 0.015 mm. thick, and if the area of the surface be decreased, still thinner.

**Manipulation of Celloidin.**‡—The failures that some microtomists experience when dealing with celloidin are due, says Dr. S. Apáthy, to the neglect of a few slight artifices. Commercial celloidin in plates or in shavings should be first of all thoroughly dried in the air, whereby it is rendered hard, transparent, and yellowish. Pieces of celloidin thus hardened are put into an air-tight vessel and just covered with a mixture of equal parts of sulphuric ether and absolute alcohol. After having been allowed to stand for some time with frequent stirring, the supernatant fluid is decanted off. This may be called the original or No. 1 solution. Some of this, diluted with an equal volume of equal

\* The Microscope, ix. (1889) pp. 344–5, from the 'National Druggist.'

† Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 181–6.

‡ T. c., pp. 164–70.

parts of ether and alcohol, forms solution 2, and some of solution 2 similarly treated forms solution 3. The preparation is placed for 24 hours or longer in each of these solutions successively. For consolidating the celloidin flat glass capsules are to be used. In these the objects are placed, and the capsules filled to the brim and covered over for some hours with a glass plate, in order that by preventing the surface from becoming hardened any air-bubbles may be allowed to escape. The glass plate is replaced in the course of some hours by a bell-jar, and in 6-24 hours, when a hardish film has formed upon the celloidin surface, the capsule is filled up with 75 per cent. spirit. In 24 hours the celloidin is fit for sectioning. From the glass capsule the pieces are cut out and stuck with a *thick* solution of celloidin on elder-pith. The celloidin block should be broader than high, and the under surface scratched with a needle. The elder-pith and celloidin are to be firmly pressed together, and then placed in 70 per cent. spirit.

For cutting sections from these blocks the knife should be smeared with yellow vaseline, and during the act of sectioning moved as nearly parallel as possible.

#### (4) Staining and Injecting.

**Benzoazurin and Benzopurpurin Stains for Microscopical Purposes.\***—Dr. Martin employs benzoazurin in watery dilute solution. The sections are overstained (1-4 hours, according to thickness of section or strength of solution). The sections are then decolorized with spirit acidulated with 1/2-1 per cent. hydrochloric acid. If a nuclear stain be desired, this effect may be counted on if the section be withdrawn when the celloidin is blanched. If the tissue elements are also to be dyed, then the decolorizing action must be interrupted earlier. A beautiful blue nuclear stain is thus obtained, and this is quite as distinct and sharp as that from carmine or logwood. This pigment seems, from the author's account, to be very useful for epithelial cells, where it brings out the nucleus and the contour of the cell, and also for most connective-tissue elements.

This dye seems to possess two valuable properties. The first is that old spirit-preparations are stainable with comparative ease; this is very difficult with other pigments, especially logwood, and the second is that preparations containing picric acid are little or not at all affected. Benzo-purpurin seems to be most suitable for double staining with hæmatoxylin or benzoazurin.

**Hæmatoxylin Staining.†**—Dr. S. Apáthy advises serial sections to be stained with a solution of 1 part hæmatoxylin crystals dissolved in 100 parts 70 per cent. spirit. They are then to be transferred to 70 per cent. spirit, to which a few drops of a 5 per cent. aqueous solution of bichromate of potash have been added. The hæmatoxylin solution is allowed to act for 10 minutes; the sections are then mopped up with blotting-paper and placed in the bichromate spirit in the dark for five to ten minutes, when they assume a bluish tinge, the celloidin remaining unstained. A double stain for differentiating the nervous and connective

\* Deutsche Zeitschr. f. Thiermed. u. Vergleich. Pathol., xiv. (1889) pp. 420-2.

† Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 179-1.

tissues of the Hirudinea is also given by the author. The object is placed for half an hour in a half per cent. watery solution of hæmatoxylin, then having been quickly washed in distilled water, it is transferred for two hours to a 1 per cent. aqueous solution of bichromate of potash. It is then again washed and imbedded in celloidin. The sections, which should show a pale yellowish staining, are after-stained in a weak aqueous alum-hæmatoxylin solution.

**New Method of Hæmatoxylin Staining.\***—Dr. N. Kultschitzky advocates the following procedure as being more simple than Weigert's method for staining nervous tissue. Pieces of brain or cord are hardened in Müller's or ERLITZKI's fluid, and imbedded in celloidin. The sections are then placed in the following hæmatoxylin solution:—1 grm. of hæmatoxylin dissolved in a little alcohol is added to a mixture of 20 ccm. saturated watery solution of boracic acid and 20 ccm. distilled water. Before using this solution a little acetic acid is added (two or three drops to a watch-glassful). The sections require some few hours (to 24) for staining. The medullated nerve-fibres are stained blue, the rest of the tissue yellow, or yellowish-red. If the sections are then placed for twenty-four hours in a saturated watery solution of carbonate of soda or lithium the nerve-fibres become dark blue, while all the rest is almost uncoloured. Then alcohol, mount in balsam.

A still more simple hæmatoxylin solution, which gives the same results, is 100 ccm. of 2 per cent acetic acid, and 1 grm. of hæmatoxylin dissolved in a little alcohol.

**Simplification of Weigert's Method.†**—Dr. U. Rossi, who says that Weigert's method is unnecessarily complicated, recommends the following simplified procedure:—Pieces of spinal cord or brain are fixed at the ordinary temperature, or in a thermostat at 35°, in the following solution:—distilled water 100 ccm., chromic acid 0.75–1 gramme, acetate of copper 5 grammes. The time required for hardening the human cord is six to eight days; cord of dog, three to four days; for the entire brain of the dog fifteen to eighteen days, and so on according to the size of the pieces. In the thermostat the fixing process is completed in half the time required for doing the same thing at the ordinary temperature. The pieces are next transferred to rectified spirit 24 to 48 hours, and afterwards to absolute alcohol. When properly hardened they are imbedded in celloidin and sectioned. The sections are placed for staining in a vessel containing about 30 ccm. of rectified spirit, to which has been added 7 or 8 drops of a hæmatoxylin solution made as follows:—absolute alcohol 20 ccm., hæmatoxylin 1 gramme. In less than 2 or 3 hours the sections become dark, and they are then placed in some of the following solution:—absolute alcohol 100 ccm., hydrochloric acid 8 drops. Herein they assume a brick-red hue, and when the grey and white matters become differentiated they are removed to distilled water, wherein they quickly become blue. After this they are to be well washed again to remove all traces of acid, then dehydrated, cleared up, and mounted in balsam.

In addition to the foregoing stain, the author says a double stain

\* Anat. Anzeig., iv. (1889) pp. 223–4.

† Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 182–4.



with a weak solution of Grenacher's borax-carminé adds to the effect. The use of xylol is to be avoided, as the stains are thereby weakened.

**Staining Animal Mucus with Anilin Dyes.\***—Various mucoid secretions, such as saliva, synovia, &c., have been examined by Dr. Süssdorf in order to show that basic anilin dyes have a specific staining action, both on free mucus and while it is still in the secreting tissue.

As may be supposed, there seems to have been no difficulty in staining free mucus by the simple method of dropping the staining solution in the secretion, and then looking at it under the Microscope.

For showing the existence of mucus within the cells of tissues and organs, the author used sublingual, submaxillary, and parotid glands of the horse, and also the intestinal and tracheal mucosa of the horse and cat. These were well hardened in alcohol, osmic acid, and chrom-osmic acid. The sections were stained with methyl-violet, methylen-blue, or fuchsin in one per cent. solution for a few minutes only. They were then washed in alcohol or spirit acidulated with one per cent. hydrochloric acid until the dye was no longer given off. Some of the sections were also stained with borax-carminé. In the latter the nuclear and plasma-elements of the cells were stained by the carminé, while the mucinous parts were dyed by the anilin pigment. In the single-stained preparations the mucinous parts alone were coloured. Some more experiments on salivary glands by the method of double-staining seemed to the author to support Haidenhain's division of the salivary glands into serous and mucous.

**Use of Colouring Matters for the Histological and Physiological Examination of Living Infusoria.†**—M. A. Certes says that anilin black dissolved according to circumstances in sea or fresh water possesses striking advantages for the study of living organisms. After filtration the solution, though loaded with pigment, will keep quite a long time without forming a precipitate even on evaporation. The effect produced resembles that obtained by Nachet's dark-ground illumination method, with the special advantage that high powers and homogeneous-immersion objectives can be used.

Anilin-black is in no way toxic to Infusoria, for they will live therein and multiply for weeks together. The contractile vesicle and other anatomical details as observed by this method are particularly interesting.

**Staining Actinomyces bovis.‡**—Dr. A. Florman states that he has made very successful preparations of Actinomyces by the following method which, though complicated, shows the club-shaped elements as well as the filaments. The sections used were 0.008 mm. thick. These were stained for 5 minutes in a solution of saturated alcoholic solution of methyl-violet 1 part, water 2 parts, aqueous (one per cent.) solution of carbonate of ammonia 2 parts. They were then washed for 10 minutes in water, and after this placed for 3 minutes in the iodine solution, iodine 1 part, iodide of potassium 2 parts, water 300 parts. After being carefully washed they were decolorized for 20 minutes in

\* Deutsche Zeitschr. f. Thiermed. u. Vergleich. Pathol., xiv. (1889) pp. 345-59 (3 figs.).

† Bull. Soc. Zool. France, xiii. (1888) pp. 230-1.

‡ Zeit.-chr. f. Wiss. Mikr., vi. (1889) pp. 190-1.

fluorescein-alcohol (i. e. until no more dye was given off). The fluorescein was washed out in 95 per cent. alcohol. Then anilin oil for some minutes. The anilin oil was removed with oil of lavender, then xylol, and finally balsam.

**Decoloration of Osmized Fat by Turpentine and other Substances.\***

—Dr. W. Flemming gives the results of experiments on fat stained with osmic acid, and afterwards acted on by various substances. Turpentine decolorizes in  $1\frac{3}{4}$  hours, ether in 4 hours, xylol in  $5\frac{1}{2}$  hours. Canada balsam dissolved in turpentine and thinned with xylol in  $4\frac{1}{2}$  hours, dammar dissolved in turpentine and chloroform in 3 hours; balsam dissolved in xylol, no action observed; chloroform, no action; oil of cloves, no action.

Hence xylol is much to be preferred to turpentine. But chloroform and oil of cloves are obviously safer.

**(5) Mounting, including Slides, Preservative Fluids, &c.**

**Manipulation of Paraffin-imbedded Sections.†**—Prof. H. Strasser, who keeps on devising alterations in the technique of paraffin imbedding, describes a new procedure, the chief feature of which is the manipulation of the section on a “provisional” or temporary slide.

The provisional slide is thin well-sized paper, one side of which has been smeared with a gum solution containing 10 per cent. by volume of glycerin. The section is then stuck on with a solution of collodium simplex 2, castor oil 1—and then fixed down by brushing over the upper surface with collodium conc. dupl. 2–3, castor oil 2.

The preparation is then removed to turpentine to dissolve out the oil and the paraffin, and also set the collodion. The plate, i. e. the imbedded section plus the paper, is then placed in an aqueous or watery-spirituos fluid for staining or other purposes. During the water stage the gum is dissolved, and the section in its collodion case thereby set free. The next step is to put this into turpentine again, after which it may be mounted in a resinous medium on a temporary or permanent slide.

Owing to the fact that the paper, i. e. the provisional slide, which plays the principal part in this procedure, becomes dyed in its transit through the staining solutions, the method, as the author confesses, is at present somewhat imperfect.

**New Method for Fixing Sections.‡**—Dr. W. M. Gray who describes the following method, says that it is identical in its procedure with the “gum arabic process,” provided the tissue from which the sections are cut has been successfully stained in mass. “The process is as follows. Dissolve one part of gold label gelatin in one hundred parts of warm distilled water; after the gelatin has dissolved, filter and add a crystal of thymol, to prevent the formation of fungi. If, on standing, the gelatin coagulates, warm slightly and use the fixative in the same manner as the gum arabic solution, or in other words, flow a small quantity on the perfectly clean slide, place the object on the fluid, and heat gently until the sections or series of sections are flat and free from

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 178–81.

† T. c., pp. 150–63.

‡ The Microscope, ix. (1889) pp. 325–6.

wrinkles, taking special care not to melt the paraffin surrounding the preparations. As they are perfectly flat, drain off the superabundant fluid, and stand the slide on end to dry. The best results are obtained if the slide is allowed to stand over-night to dry spontaneously. After the sections are dry, the whole is immersed in turpentine or other solvent to remove the paraffin, then into alcohol to remove the turpentine, thence into a two per cent. solution of potassium bichromate for five minutes, which renders the gelatin fixative *insoluble*. After washing the slide in water to free the section from bichromate) which, by the way, will not injure the most delicate tissue or interfere with any staining process) the section may be stained as desired. For sections stained in mass the soaking in bichromate is unnecessary, but if, after mounting, the stain should prove insufficient, the sections may be readily restained by removing the cover-glass, soaking off the balsam with a suitable solvent, transferring to alcohol and then rendering the fixative insoluble by soaking in bichromate before immersing in the stain. This process is especially valuable in staining tissues for bacteria, as it admits of extremely thin sections being placed on the slide free from wrinkles, and does not blister by prolonged soaking in aqueous solutions, as frequently happens in Schällibaum's clove-oil-collodion process, the method in general use for staining sections on the slide."

**Use of Oil of Cloves.\***—Mr. W. Hatchett Jackson points out that sections to which oil of cloves has been added and which have turned milky are not, as is often supposed, useless. If a small quantity of oil is poured on the sections and the whole gently warmed for a short time, the milkiness disappears. If it does not disappear at once the oil in the slide should be poured off, fresh oil added, and the heating repeated. The milkiness is due to a combination between the essential oil and a small residuum of water, and this is readily soluble by the aid of warmth in an excess of the essential oil. Repeated soaking in absolute alcohol effects the same end.

**Cement for fixing down Glycerin Preparations.†**—The cement recommended by Dr. S. Apáthy is said to be hard, without brittleness, and not to run under the cover-glass.

It is made of equal parts of hard paraffin, melting-point 60° C., and commercial Canada balsam. The mixture is heated in a porcelain dish until it assumes a gold yellow hue, and a resinous odour is no longer perceived. When cold the mixture forms a hard mass, which requires to be heated for use and to be laid on with a glass rod or brass spatula. The metal spatula is then heated and run round the edge to finish it off.

#### (6) Miscellaneous.

**Detection of Blood-stains.‡**—Dr. C. Charles remarks that, according to Linossier, the most sensitive spectroscopic reaction of blood is that given by reduced hæmatin.

The blood-stain is dissolved in water and examined for the spectrum

\* Zool. Anzeig., xii. (1889) pp. 630-1.

† Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 171-2.

‡ Amer. Mon. Micr. Journ., x. (1889) p. 236, from 'The Dosimetric Medical Review,' July 1889.

of oxyhæmoglobin. A drop of freshly prepared hyposulphite of soda is now added, when the spectrum of hæmoglobin appears at once; finally, a couple of drops of a concentrated solution of soda are added, which decomposes the hæmoglobin into globulin and reduced hæmatin, the spectrum of the latter consisting of two absorption-bands situated between D and b, the left one lying between D and E and being well marked; indeed, this intense band is the only one to be distinctly observed in dilute solutions, and it ought to disappear if the solution is heated to 50° C. without stirring, or agitation, and reappear on cooling; it ought further to disappear when shaken in the air, and reappear on the addition of a drop of hyposulphite of soda. This test applies even to putrid blood. Should the blood-stain have become insoluble in water, we are directed to dissolve in ammonia and reduce by adding one or two drops of a solution of ferrous sulphate and tartaric acid.

Jaksch's 'Clinical Diagnosis of Disease by Bacteriological, Chemical, and Microscopical Examination.'\*—The fact that within two years Dr. R. von Jaksch's book on the diagnosis of disease has gone through two editions, and that translations into several languages are in preparation, shows that it supplies a want.

While this second edition is an improvement on the first, and is not a mere reprint thereof, yet there are several small points which are either errors of omission or commission. For example, there is no mention of Fraenkel and Netter's researches on the diagnosis and prognosis from a bacteriological examination of pleuritic fluid. On the other side, the Finkler-Prior bacillus seems to be regarded by the author as the bacillus of Cholera nostras.

These and similar deficiencies apart, the work may be considered very useful, and fairly up to date.

Israel's 'Pathological Histology.'†—Dr. O. Israel's introduction to pathological histology seems to be biassed by his views on the staining of microscopical preparations, which he scornfully designates coloured mummies. In other respects the work does not seem to differ materially from the usual run of text-books on this subject, and it is well got up.

Insects in Drugs.‡—At a meeting of the Chemists' Assistants Association some rather disquieting specimens were lately exhibited, demonstrating the existence of "insects and germs" in sundry pharmaceutical preparations and drugs. The first was a fair-looking sample of crushed linseed recently obtained from a large wholesale firm, and kept in a wooden cask with a wooden cover. The exhibitor gravely asked what would be the effect of applying a poultice containing "thousands of insects" to an open wound, especially if the poultice be made with hot instead of boiling water. The other specimens, from aconite root, nuxvomica, and cantharides, are perhaps of less importance, as these substances are not employed in the crude state. In the present anxiety to detect microscopic germs and to render them innocuous, it is worth considering whether we are not in danger of overlooking more obvious sources of infection. In the hunt for small deer a different lens is employed, and mental vision is thrown out of focus for larger game.

\* 2nd ed. enlarged, Vienna and Leipzig, 1889, 8vo, 438 pp.

† Berlin (A. Hirschwald), 1889.

‡ Lancet, 1889 (ii.).



**Brownian Movement.**—The President of the New York Microscopical Society informed\* the members at a recent meeting that the specimen of gamboge rubbed up in water which he had prepared on Aug. 3rd, 1874, and which had until recently showed very active movements, seemed at last to have ceased its activity, a leak having developed in the inclosing cell, and evaporation having ensued in consequence. He thought the subject of interest, as fourteen years was probably the longest period during which the phenomenon had been under observation.

We recently purchased a number of the 'Philosophical Magazine and Annals of Philosophy' for 1828, which contained (pp. 161-73) the original article of Robert Brown on the existence of active molecules in organic and inorganic bodies, and at the beginning of the article was inserted a MS. letter addressed to "Rev'd. Dr. Buckland, Christ Church, Oxford," and signed "J. H. C.," which, we understand from a relative of the late Dr. Buckland, to be the initials of the Rev. John Henry Conybeare, Anglo-Saxon Professor at Oxford, brother of the Dean of Llandaff.

Of Brown's views he writes as follows:—

"Touching Brown's theory that all matter consists of live mites, I don't believe a word on't. I don't wish to regard our own planet as rotten cheese any more than the moon as cream cheese. If you suspend particles of matter in a fluid for microscopical observation, a thousand circumstances, may generate motion, and to this I attribute his facts; if, however, they should be confirmed, I know nothing inconsistent with the received philosophical notions as to the intimate corpuscular structure of bodies in them. Biot, if I remember, in the optics of his Nat. Phil., has some curious speculations on the subject. He states it to be possible that solid bodies may be composed of systems of moving molecules, representing in small what the planetary systems do in large. I would only add one supposition more; that these molecules are inhabited, and have philosophers among their population who, having observed the motions of some half-dozen molecules in their neighbourhood and ascertained their laws, believe they have developed the system of the universe."

**Method for Transmitting Microscopic Objects.**†—Prof. G. O. Sars describes the following method for transmitting microscopic creatures from a distance:—

On March 14 a quantity of mud was gathered from a freshwater lake in the northern part of Australia. This was dried and sent to Christiania, where it was received on the 29th of October, in masses so hard and stony that they were broken with difficulty. The weather was so cold that the experiments were not begun until the last of May, the mud and its contents having been in a dried condition for more than a year. It was finally placed in an aquarium consisting of a large cylindrical glass vessel, where a great number of the various orders of the Entomostraca were hatched out from the "winter eggs" dormant in the gathering, and in many cases studied through several generations. The method is a suggestive one, and in the hands of others may be followed with as successful results.

\* Journ. New York Mier. Soc., v. (1889) p. 46.

† *Fordhandlingar i Videnskabs-Selskabet i Christiania*, 1887. Cf. *The Microscope*, ix. (1889) p. 349.

**Microscopical Examination of Paper.\***—Mr. Herzberg, who has charge of the examinations of paper at Charlottenburg, has just published a very exhaustive work upon the subject, with numerous reproductions of microscopic preparations. He brings specially into prominence the peculiarities of certain fibres for rendering them easily distinguished.

The author uses a solution of iodine for recognizing the various fibres, which, according to their origin, assume various colours: (1) Wood-wool and jute are coloured yellow; (2) straw, "cellulose," and alfa do not change; (3) cotton, flax, and hemp are coloured brown.

For disintegrating the paper, Mr. Herzberg does not employ the processes in common use. Mechanical appliances, either needles or a mortar, do not remove the size, starch, and weighing substances which in part conceal the structure of the fibres and render the examination of them difficult. He recommends that a small quantity of the paper to be examined be submitted to ebullition for a quarter of an hour in a 1 to 2 per cent. solution of soda. In this way the foreign substances are got rid of and the fibres set free. The presence of wood-wool will be ascertained, during the boiling, by the paper becoming yellow.

After this treatment the whole is poured upon a brass strainer with fine meshes, and is washed with pure water. The washed residuum is reduced to a homogeneous paste in a porcelain mortar.

In the case of coloured paper the colouring matter must be removed if the boiling does not effect the removal. To this end, hydrochloric acid, chloride of lime, &c., is used according to the chemical nature of the colouring matter. When the paper is not sized, nothing but water is used for the boiling. If the presence of wool in the paper is suspected an alcoholic solution, instead of an alkaline one, is used, as the latter would dissolve the wool. The solution of iodine in iodide of potassium may be more or less concentrated. The colour produced varies in depth according to the concentration. The author generally uses the following formula:—Iodine, 18 grains; iodide of potassium, 30 grains; water, 5 drachms.

For spreading the paste upon the object-holder of the Microscope he employs two platinum needles. The object-holder is placed upon a white ground, so that the fibres will stand in relief more prominently. The paste is covered with a glass, and the excess of water is removed with blotting-paper. For the determination of the fibres a magnifying power of 300 diameters is best adapted, but for ascertaining the relative proportion of the fibres, one of 120 diameters, that permits of taking in a wider surface, is preferable.

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\* Amer. Mon. Micr. Journ., x. (1889) pp. 274-5, from 'Guttenberg Journal.'

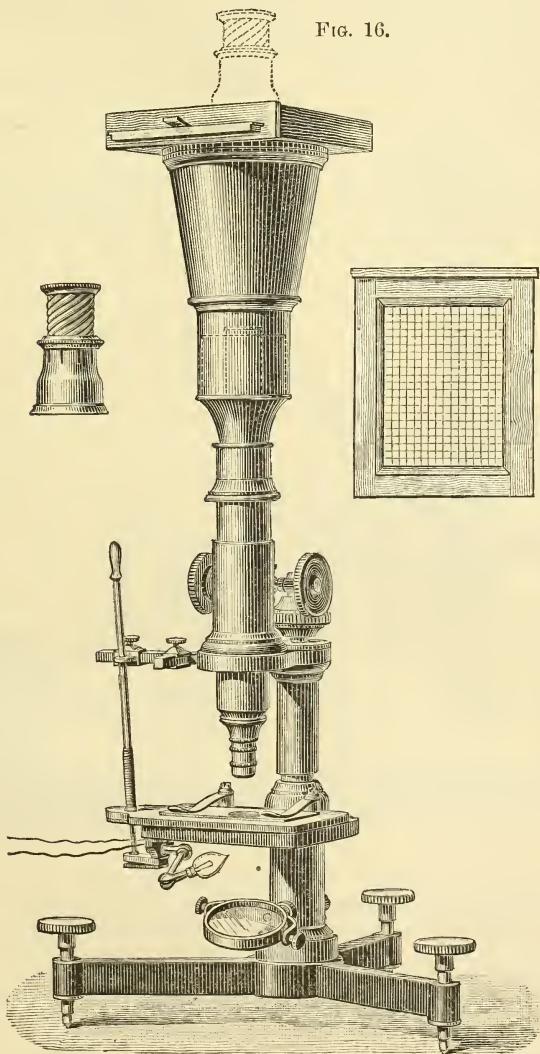
## MICROSCOPY.

## a. Instruments, Accessories, &amp;c.\*

## (1) Stands.

**Duboscq's Photographic Microscope.**—The Microscope shown in fig. 16 was devised by M. Jules Duboscq of Paris for obtaining photographs 8 cm. in diameter.

FIG. 16.



\* This subdivision contains (1) Stands; (2) Eye-pieces and Objectives; (3) Illuminating and other Apparatus; (4) Photomicrography; (5) Microscopical Optics and Manipulation; (6) Miscellaneous.

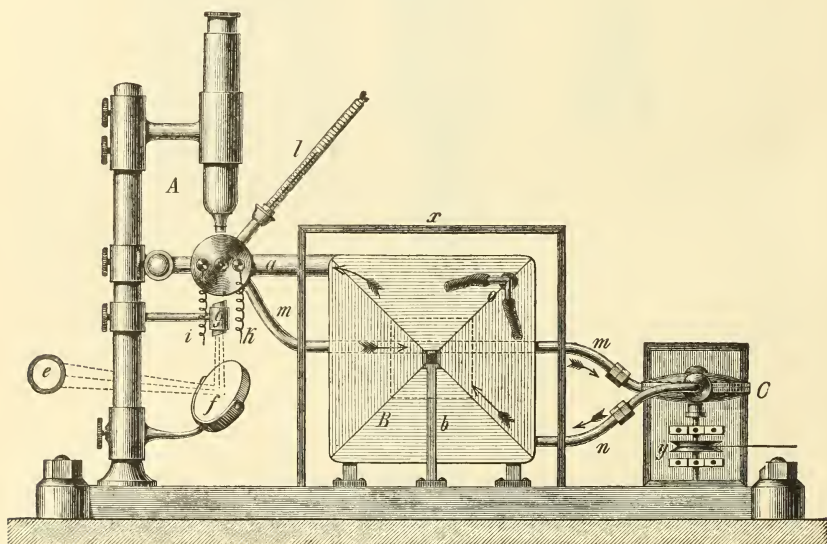
A camera, as will be seen from the fig., slides over the top of the body-tube. The tripod feet of the Microscope are provided with levelling screws, while the movements of the stage are effected by a White lever, which is made of extra length so that it may be close to the milled heads of the coarse-adjustment. A special support for the lever is attached to the cross-arm. A small electric incandescent lamp is attached beneath the stage.

The small fig. on the left is the focusing glass (shown in position by dotted lines on the top of the camera). The fig. on the right is the ground-glass plate, which is divided into spaces.

The instrument is also made with the photographic part independent, and mounted on a slide fitting, supported by a strong cast-iron base.\*

**Lehmann's Microscope for heating objects at definite temperatures.**†—Dr. O. Lehmann has found the Microscope shown in figs. 17 and 18 very serviceable where it is desired to heat an object at definite temperatures, a regular stream of hot liquid being kept up through the vessel containing the preparation by means of a pump. Dr. Lehmann

FIG. 17.



first made use of an ordinary air-pump, in combination with a spacious reservoir, which was put in motion by a gas motor, but in the later form a centrifugal pump is used, as represented (somewhat diagrammatically) in fig. 17. A is the Microscope, whose stand is fastened to the wall in order to avoid oscillations due to the action of the pump; B is the reservoir for the liquid, and C the centrifugal pump. Out of the reservoir B, in which equal distribution of temperature is effected by

\* Cf. *La Lumière Électrique*, xix. (1886) pp. 217-9 (2 figs.).

† 'Molekularphysik,' Band i. (1888) pp. 151-2 (2 figs.).

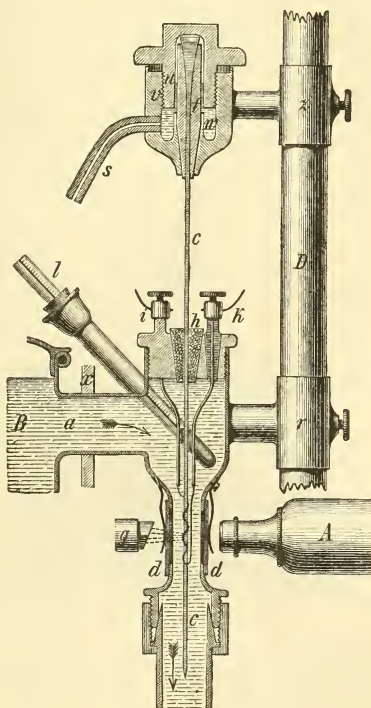


a stirrer worked by the arm *b*, the liquid passes through a short wide tube *a* into the observation tube, which is shown in section in fig. 18. This tube is contracted and flattened out opposite the objective, and the side walls are cut away and replaced by plate glass *dd*, through which the light from the lamp *e*, after reflection at the mirror *f* and passage through the nicol *g*, can enter into the body-tube A.

The substance under observation is contained in a thin capillary tube *cc*, which passes through a cork *h* in the cover of the observation tube. It is closed below, but terminates above in a funnel-shaped opening. *i* and *k* are two wires, attached to two binding-screws, which transmit the electric current through the fine wire spirally wound round the capillary tube at the position of observation. By the passage of the current the temperature is locally and for an instant considerably raised. One wire *k* is insulated from, while the other is directly connected with, the cover. A thermometer with small bulb close to the capillary tube serves to register the temperature. The hot water or paraffin passes from the tube *a* (fig. 17) into the exhaust tube *mm* of the pump, and from this through the tube *n* back into the reservoir B. To protect the observer from the hot gas-flame, the reservoir is surrounded by a screen of asbestos *x*. The reservoir is provided with a Reichert's temperature-regulator O, which automatically keeps the gas-flame at the right height.

The wheel *y* which drives the pump is put in motion by a small gas motor stationed on the ground away from the wall on which the Microscope is mounted. By means of a strap the gas motor also drives the stirrer. For examining under high pressures the capillary tube *cc* can be put in connection with a Cailletet pump by means of the capillary tube *s* (fig. 18), which passes into the small metal reservoir *v*, closed above by the cover *u* screwed on air-tight, and below by the stopper *t*, in which the capillary tube *cc* is fixed with shellac. *t* is so high that it can be fastened after removal of the cover *u*. The open space *w* contains glycerin from the Cailletet pump. The whole is supported by the clamp *z*, sliding on the rod D, which is provided with a second clamp *r* for holding the observation tube, while it is itself

FIG. 18.



supported by a clamp of the Microscope-stand or is fixed to the wall which supports the whole apparatus.

**Lehmann's large Crystallization Microscope.\***—Dr. O. Lehmann describes the large Crystallization Microscope which he designed for use where great stability is required. (A more portable form was described in this Journal, 1885, p. 117.) The instrument is shown in figs. 19–21.

For stability the base of the whole is formed of a large heavy cast-iron plate *bb* (fig. 20), which for convenience in height is let into an opening in the table, and rests by means of four levelling-screws upon two strong ledges strengthened by cross-pieces. It is pierced by several holes provided with screw-threads in which fit the different stands and apparatus. On the same ground of greater stability the movement of the body-tube is effected in quite a different manner to that of the ordinary Microscope. The socket which carries it is rigidly connected with the base-plate by a  $\Gamma$ -formed holder (fig. 21). This allows the cross-arm to be very long, which renders more convenient the handling of the object on the stage. Since experiments at high

FIG. 19.

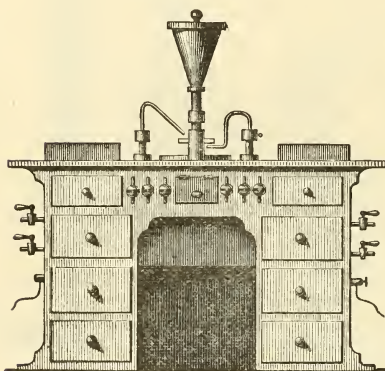
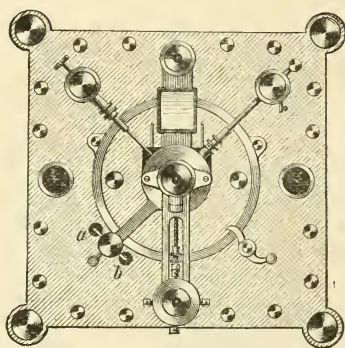


FIG. 20.



temperatures, in which a large flame is used, require a considerable raising of the stage, and consequently a greater height of the body-tube than could be attained by a simple movement in the socket, the  $\Gamma$ -formed holder consists of two parts, viz. the tube, firmly screwed into the base-plate, and the holder proper, which slides tightly in this, and can be fixed in definite positions by means of a steel pin, which is inserted into holes bored through the holder at regular intervals equal to the distance through which the body-tube slides in its socket. The nut is then screwed on to the lower end, and the whole is as firmly fastened as if it consisted of a single piece. For the coarse-adjustment the body-tube simply slides in the socket, but the fine-adjustment is effected by means of a second socket, consisting of two parts, in which

\* 'Molekularphysik,' Band i. (1888) pp. 126–33 (3 figs.).

the first moves, though not quite freely. The first socket is provided with a screw-thread, upon which works a collar fastened to the second socket by pins. By turning the collar the first socket is slowly raised or lowered as the adjustment requires. Two spiral springs on the second socket, fixed below, and having their upper ends in contact with a long plate attached to the first socket, prevent backlash between the screws. (Later this was effected by means of a single screw attached to the cross-piece of the holder.)

Another peculiarity of the instrument is the stage, which is not connected with the body-tube, but is carried by a special foot. In the middle of the base-plate is let in a conically-turned toothed ring, which can be rotated by a small toothed wheel. On this ring is screwed a divided circle, which is itself surmounted by a cast-iron plate of smaller diameter, provided with two parallel slits, in which slide the two sides (tapered below) of the horse-shoe which forms the foot of the stage. This motion is effected and measured by means of a micrometer-screw provided with a divided head. The ring as well as the foot are provided on their under side with screws and caoutchouc rings for avoiding backlash. The foot carries a pillar, which supports a plate *a*, bored through, and having on its upper side a conical projection. On this cone rotates a second plate, provided with a slit in which slides a metal piece, bored through and tapered below, to which are attached two short pins, which support the thin circular disc forming the stage proper. The form and size of the stage varies according to the experiment. When a high temperature is required the lenses of the Microscope are protected by a thick copper diaphragm, provided with a small hole. In certain cases this is made hollow and kept cool by a stream of cold water flowing through it. In fig. 20, *a, b* are the holes through which pass the tubes conveying the water.

The burner *R* for heating the object, and the rod carrying the screen of glass or mica for moderating the temperature, are attached at right angles to a hollow metal column, which communicates by a branch tube below with the gas supply, and contains a smaller tube, reaching as far as the attachment of the burner, which conveys from a gas-holder the air necessary for the non-luminosity of the flame. The gas supply is cut off from the burner by a screw stopper at the top of the column. The latter is not screwed into the base-plate, but fits conically into it, and is fastened by a nut and caoutchouc ring only so firmly that it may be easily turned by the hand about its axis. The contact of two arresting pins during the rotation automatically effects the correct adjustment of the burner exactly beneath the opening of the stage.

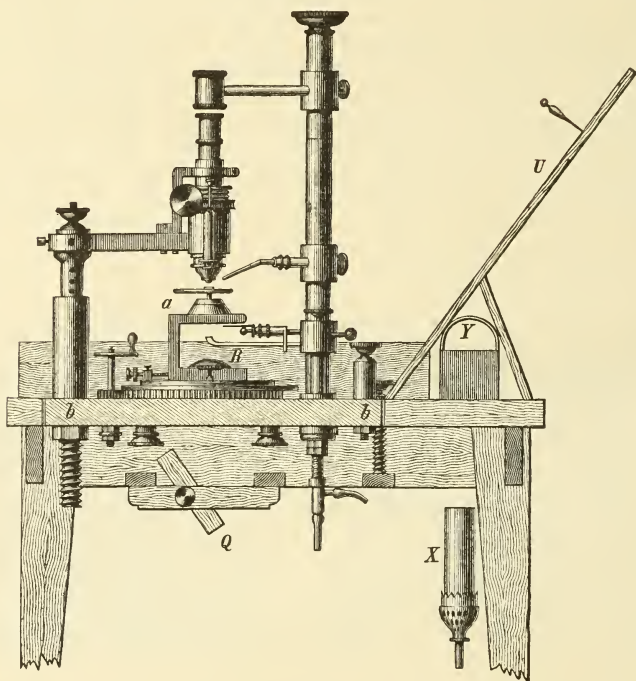
Polarization can be effected in several ways, either by a bundle of glass plates *Q* (fig. 21) reflecting to a condensing lens the light from the gas-lamp *X*, the smoke from which passes off by the chimney *Y*; or by means of a concave mirror; or finally by a bundle of plates, which receives its light from an adjustable plane mirror. The analysing nicol is carried by an arm which slides on a vertical pillar, and can be clamped in any position above the eye-piece. The pillar in the lower part of its length is hollow, and forms a tube which at about the middle projects outwards at right angles, and is then bent downwards towards the stage with gradually diminishing section. This



tube serves to convey a stream of air for cooling the preparation. The tube is closed, and consequently the air cut off, by means of a conically pointed screw on the opposite side of the pillar.

For measuring extinction angles and angles of crystals, &c., the Microscope is provided with cross wires. In order that the axis of rotation of the stage should pass exactly through the centre of the cross-wires, the cross-piece of the  $\Gamma$ -formed arm is not rigidly connected with the vertical column, but only by means of a nut screwed

FIG. 21.



firmly on. The opening through which the spindle of the screw passes is somewhat larger than it, and the latter is square below. Against the faces press four screws set in the cross-piece at angles of  $90^\circ$  to each other. As is seen from fig. 20, the required adjustment can be effected by suitable movement of these. In order to be able to fix the form of the object a drawing-board *U* (fig. 21) is set up obliquely behind the Microscope, a small three-sided prism being fixed to the eye-piece. Two raised portions on the table serve to support the arm during drawing and observation.

**Konkoly's Microscopes for the Cameras of Telescopes.\***—Dr. N. v. Konkoly uses a compound Microscope for focusing the image produced

\* Central-Ztg. f. Optik u. Mech., x. (1889) pp. 229–32 (6 figs.).



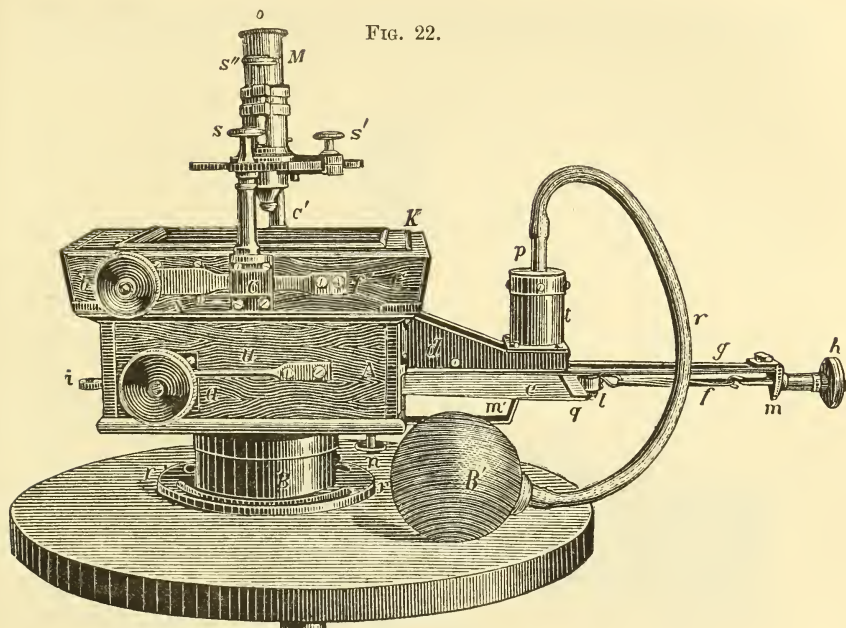
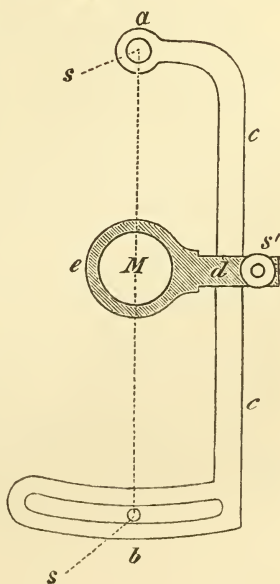


FIG. 22.

by a telescope on the plate of a camera, "as" (he says) "I venture to designate as illusory any other mode of focusing." Such an arrangement is new to us. In the "Universal Camera" the Microscope *M* is connected with the apparatus, as shown in fig. 22, and the following arrangement is adopted for viewing the different parts of the picture on the plate. To the plate-holder are attached two pillars *C C'*, which carry an arm *abcc* (fig. 23) secured in position by two milled heads *ss*. This arm supports the holder *de* for the Microscope, *de* slides on *cc* and can be clamped in any required position. At *b* the arm has an arc-shaped slit whose centre is at *a*. The movement of the Microscope along the arm *cc*, combined with the rotation about the centre *a*, enables the observer to cover the whole field of view. In order to bring the focal planes of Microscope and telescope into coincidence, a limited fine motion is communicated to the eyepiece *O* of the Microscope by means of the screw *s''* (fig. 23).

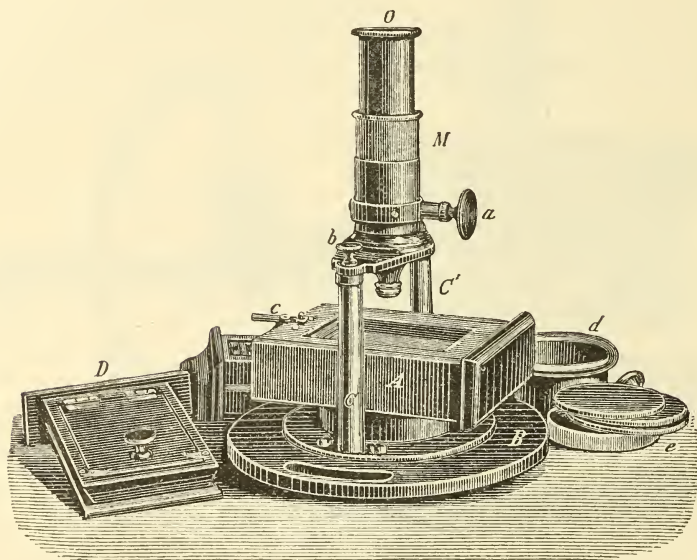
In the small camera for small tele-

FIG. 23.



scopes (fig. 24) the focusing is also effected by a Microscope. The two pillars  $C\ C'$  carry the fixed bridge  $b$  in which the Microscope  $M$  is

FIG. 24.



screwed. The objective is a dividing doublet, so that greater play has to be given to the movement of the eye-piece, as one or both are used, and for this purpose a rack and pinion  $a$  is applied.

**Boys' Microscope Cathetometer.\***—Mr. C. V. Boys, in his experiments on the elasticity of quartz fibres both to stretching and to torsion, devised the apparatus shown in fig. 25.

The apparatus (made by Hilger) consists of a Microscope cathetometer shown in the figure at  $M$ , which can be made to traverse a vertical slide by means of a fine screw having a micrometer-head, the divisions of which are capable of being read directly to the  $1/1000$  mm. To the end of the Microscope farthest from the eye-piece is attached the vertical tube  $T$ , which carries at its lower end an adjustable arm  $A$ , fitted with a clamp  $C$ . To the end of a separate bracket is fixed the block  $a$ , which supports, by means of a knife-edge, the beam  $B$ , which is weighted with a gravity-bob  $W$ , and carries on a second knife-edge  $b$  the micrometer-scale  $D$ , the opposite end of the lever being counterpoised by the adjustable weight  $P$ . The fibre to be tested has attached to it a pin at each end to facilitate its being fixed in the apparatus, it being stretched vertically between the scale  $D$  and the clamp  $C$ .

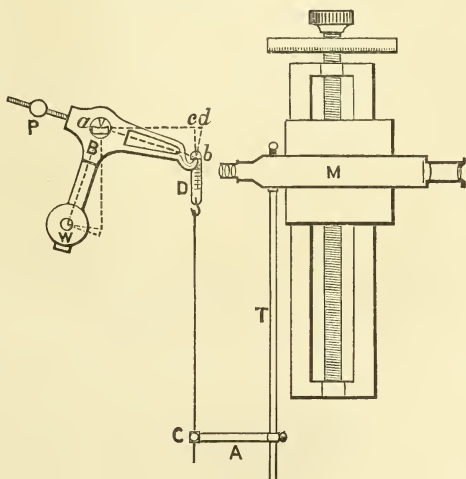
When the micrometer-head is turned, the cathetometer  $M$  is lowered, carrying with it the tube  $T$ , and thereby putting a tensile strain on the fibre, which draws down the lever  $B$ , being itself stretched under the

\* Journ. Soc. Arts., xxxvii. (1889) pp. 833-4 (1 fig.).

increasing pull of the lever. The extension of the fibre is measured by the movement of the scale D across the field of the Microscope, and the deflection of the lever B is a measure of the force that is being applied to the fibre, which is obtained by subtracting the amount of extension of the filament from the distance traversed by the Microscope, which latter may be determined with the greatest accuracy by the readings of the micrometer-head.

In adjusting the instrument, the slide is first made vertical by levelling screws, the accuracy of the levelling being determined by means of a spirit-level placed in different azimuths on the top of the micrometer-head. The counterweight P is next adjusted until the knife-edges at *a* and *b* are both in the same horizontal plane, and this adjustment is made when the scale D and the upper attachment pin of the fibre are in their proper position, and the Microscope is focused so as to give a sharp definition of the divisions on the scale. The fibre having been attached to the upper supporting pin and suspended in its place, the length of the arm A is so adjusted that the lower supporting pin of the fibre hangs freely in the axis of the clamp C, which is then tightened, and thus perfect verticality at the commencement of the pull is insured.

FIG. 25.



The micrometer-head is then slowly turned, readings being taken as each division of the scale D traverses and coincides with the cross wire of the Microscope, and the force which thus extends the fibre by each increment of  $1/20$  mm. is determined in the following way.

If the adjustments of the instrument have been made in the manner described above, the moment due to a vertical pull is proportional to the cosine of the angular displacement of the beam, while that due to the gravity-bob and the other portions of the beam varies as the sine of that angle, the actual tensile force applied at D being proportional to the tangent of the inclination of the beam. The vertical distance *cb* is a measure of the sine of the inclination, and when the angular displacement is small this distance is practically the same as the tangent of the angle, and it may be corrected to measure the tangent if very great accuracy be required. The true value of the force corresponding to various values of *cb* may, however, be more easily found by attaching weights to D, and observing by means of the Microscope and scale the weights which produce corresponding deflections.

In this instrument there are two apparent sources of error, which,

however, do not in any way affect the accuracy of the measurement. In the first place, it is evident that as the beam is deflected the point *b* becomes more and more distant from the Microscope, and the pull on the fibre ceases to be vertical, but it must be also noticed that in doing so the scale *D* is carried out of the focus of the Microscope, which has in consequence to be adjusted by being moved forward to the exact amount which the scale had receded by the movement of the beam, and thus the arm *A*, carried by the end of the Microscope, is moved forward to an equal extent, the scale comes again into focus, and the fibre becomes again vertical.

Again, in the case of the tube *T* being very long, it might happen that the spring of the tube and of the arm *A* might cause the fibre to appear more stretched than it really is, but the error due to this cause can be perfectly eliminated by finding, in the course of the experiment, the force that is being applied to the fibre, and afterwards placing weights on *C* until a pull of the same amount is obtained. As a matter of fact, however, with ordinary fibres the further movement of *D* under these circumstances is not observable.

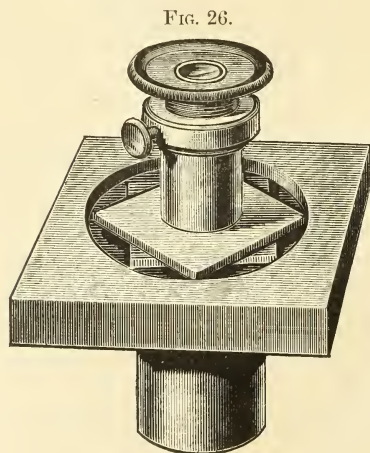
POLI, A.—Note di Microscopia. III., Il condensator nei Microscopi. (The condenser in microscopy.)

*Rivista Scient.-Industr.*, XXI. (1889) Nos. 18, 19, p. 217.

#### (4) Photomicrography.

**Bourdin's Photomicrographic Apparatus.\*** — After describing Duboseq's large Microscope for photomicrography,† M. J. Bourdin advises microscopists not to neglect the very simple method of producing

photomicrographs by means of a small camera, applied in the body-tube when the eye-piece is removed, large enough to permit the use of glass plates of 3 cm. square. The exposure of the sensitive plate is very short, and the magnification being low, it becomes necessary to employ an enlarging apparatus with which one may readily obtain transparent positives as large and as sharp as may be desired, especially by the use of Cowan's chlorobromide plates which are developed with a solution of 30 grm. citric acid and 20 grm. carbonate of ammonia in 100 grm. distilled water.



The small camera in question is shown in fig. 26, and is stated to be recommended by M. Luiz de Andrade Corvo, who is engaged on special investigations of the phylloxera. It is shown together with a focusing lens made by Starsnic, the successor of Véric.

\* *La Lumière Électrique*, xix. (1886) pp. 217-9 (2 figs.).

† *Supra*, p. 231.

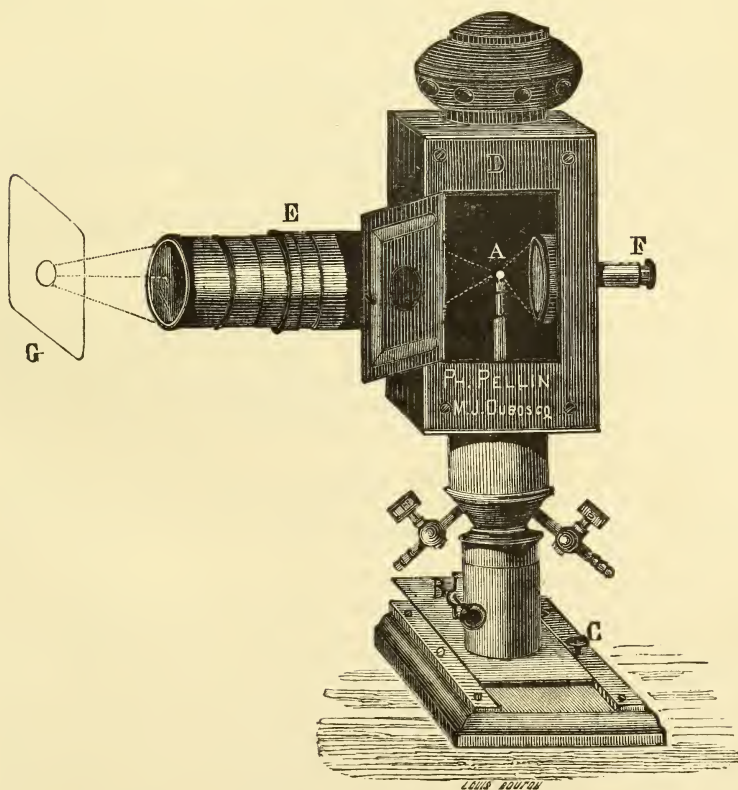


The focusing lens is adjusted over a glass square, on the underside of which is fixed a fragment of a fly's wing, and the image produced in the Microscope is focused on the plane of the fly's wing. The focusing lens and the glass square are then removed, and a sensitive plate is substituted, being covered by a small cap of obvious construction; the incandescence lamp is then set in action as required, care being taken that no actinic light strikes the sensitive plate except during the required exposure in the Microscope.

**Roux's Lantern for Photomicrography.**—Dr. Roux has devised the lantern for photomicrography shown in fig. 27.

A small ball of magnesia, 5-6 mm. in diameter, is placed in the lantern D, which is rendered incandescent by an oxy-hydrogen jet. A

FIG. 27.



condenser E concentrates the light on the stage G of the Microscope. Behind the ball is a mirror adjustable by the rod F. The screws at B and C enable the lantern to be centered vertically and horizontally.

Care is required in heating the ball, which must be brought to 1890.

incandescence gradually, but when attention has been paid to this point it will last from 60 to 70 hours.

**Photomicrography at the Photographic Jubilee Exhibition at Berlin, 1889.\***—Dr. R. Neuhauss, who gives his impressions of the photographic exhibition, awards the first place in the microscopical class to the photographs exhibited by the Berlin Hygienic Institute. These are principally the work of Dr. Koch and Dr. Pfeiffer. The latter showed the flagellate micro-organisms, some of which have appeared in the 'Atlas of Bacteriology' of Fraenkel and Pfeiffer. The Institute also showed a very interesting series illustrating the progress of microscopical photography.

Schultz-Henke showed two photographs taken from the same preparation—one with the ordinary dry plate, the other with the eosin-silver plate (spinal cord  $\times 30$ ). The latter photograph showed more details, but it is possible that the dry-plate process was not shown at its best.

Max Hauer exhibited a series of photographs from his 'Atlas of Vegetable Anatomy.' The photographs, which were very large, had been taken with relatively low powers. The size had been attained by means of a large camera or subsequent enlargement of the negative. The defect of this procedure is that the photographs show diffraction lines, a defect possibly inseparable from the method.

The foregoing afford a good illustration of the exhibits, but there are several others mentioned by the author, including an album of his own work.

#### (5) Microscopical Optics and Manipulation.

**Method of Detecting Spurious Diffraction Images.†**—Mr. E. M. Nelson writes:—In a previous paper I gave as my opinion that certain alleged diatom gratings, of double fineness and either above or below the original structure, were spurious, because they were caused by the action of an over- or under-corrected Microscope objective on the diffraction-spectra.

I now show how a test may be applied to determine whether these structures are entities or only diffraction-ghosts. The test will suit equally well other objects which yield a similar arrangement of interference conditions.

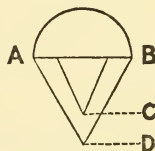
Set up the Microscope and adjust tube-length, &c., so that the best view is obtained of say the upper fine grating in *Pleurosigma formosum*, the reality of which is required to be tested. By means of the fine-adjustment the distance between this fine grating and the original coarse grating is accurately measured. The draw-tube of the Microscope is now lengthened one inch or more, and the distance between the two gratings is again measured. If this last measure agrees with the former measure, the grating is in all probability an entity, but if the measure with the long tube exceeds that with the original adjustment, then the fine grating is an optical ghost. In the case of an under-corrected objective, with a fine grating below a coarse grating, it will be necessary

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 273-7.

† Note read 10th April, 1889. Cf. also Journ. Quek. Micr. Club, iv. (1890) pp. 55-6 (1 fig.).

to shorten the draw-tube before the second measure is made. The increase in tube-length need not be so great in the case of a short tube as in a long. In the actual measures I made of the distance between Mr. Smith's upper fine grating and the ordinary well-known grating on *P. formosum* I found the distance between them, at the original adjustment, was  $1/8000$  in.; an additional inch of tube on the long body increased that distance to  $1/5000$  in., thus making a difference of nearly  $1/13,000$  in. A reference to the subjoined figure will make the case abundantly clear. Let AB represent the front lens of an over-corrected objective, C being the objective. Now when a *P. formosum* is placed at C, the spectra of the focus for the central, and D the focus for the marginal portion of the first order only are recombined at the objective conjugate, consequently only the original coarse structure is seen. Let, however, the focus be raised so that the valve is placed at D, then the spectra of the second order only are united at the conjugate, and a grating of double fineness is seen. The movement of the objective gives, of course, an irresistible idea that the fine grating is above the coarse. The effect of lengthening the tube is to increase the over-correction of the objective, i. e. the distance between C and D, and consequently the distance between the fine and coarse gratings.

FIG. 28.



Let me put it in another way: when there is, say, an over-corrected objective on the Microscope, it is just the same as if there were two separate objectives in a rotating nose-piece: the one, a narrow-angled lens of short focus, which is only capable of resolving the coarse grating of, say, 25,000 per inch; the other, a wide-angled longer-focused objective with its centre stopped out, which exhibits the same grating as possessing double fineness, viz. 50,000 per inch. In such a case no one would have been led astray, as an alteration of focus would have been expected, but because all this occurs in one and the same objective, a confusion has arisen.

It is for those who deny the effect of diffraction in the production of the Microscope image, and those who insist on the reality of structures which are not consonant with that theory, to explain why an alteration of one inch in tube-length nearly doubles the distance between the gratings.

**Method for measuring the Spherical and Chromatic Aberration of Microscope Objectives.\***—M. C. J. A. Leroy remarks that the principle of his method is similar to that which enabled Foucault to put in practice his method of "retouches locales." The surface of the objective, observed through a small hole in a screen is seen illuminated only in the part traversed by the rays isolated by the hole. When the object is a monochromatic luminous point, and the small hole is on one side of the axis, the part illuminated is on the same side or the opposite, according as the corresponding rays cut the axis in front of or behind the plane of the small hole. If the latter be displaced transversely the limit of the bright zone will be displaced, under the same conditions, in

\* Comptes Rendus, cix. (1889) pp. 857-9.

the same direction (direct) or in the opposite (inverse). Accordingly, if the observer move along the length of the pencil, so long as he is in front of, or beyond the extreme points of crossing of the rays, the path of the light in the whole extent of the displacement will be either direct or inverse. On the other hand, throughout the extent of the zone limited by these extreme points, there will be simultaneously direct and inverse paths for a certain number of positions of the small hole during a transversal displacement.

The study of chromatic aberration was made in the same manner, the bright regions presenting the colour of the rays isolated by the small hole when white light was used.

The longitudinal aberration proving very troublesome to measure, owing to its enormous extent, the author confines himself to the transverse.

The object was a luminous slit, traced with a sharp razor in a silver layer deposited on a glass plate, and had a width of from 0.005 mm. to 0.0025 mm. The slit was covered by a cover-glass. The small hole, having a diameter of 0.8 mm., was displaced perpendicularly to the slit, and the displacement was read on a scale giving  $\frac{1}{10}$  mm. The Microscope was adjusted so that the image was clearly defined in the plane of the small hole, whose distance from the slit, placed on the stage, was always the same, 0.20 mm.

The image was generally decomposed into two parts, one with rays coming from the marginal zone of the objective presenting an aberration in a determinate direction, the other, corresponding to the central zone, presenting an aberration in the opposite direction. In subtracting from the total displacement (between the limits of appearance and extinction of the light) the displacement due to the central zone, the same result was always obtained as in taking account of the dimensions of the slit and hole measured directly.

The objectives of the best makers of France and Germany were studied, and gave measurable spherical aberration varying from tenths of a millimetre to several millimetres. In measuring the spherical aberration, a coloured glass was placed over the small hole; for the chromatic aberration white light was used, and its value was judged by the intensity of the variations of the tints. Nearly all the objectives had sensible chromatic aberration, but many were found in which it was scarcely perceptible, and in some, constructed simply of flint and crown glass, it was quite inappreciable.

The author concludes, therefore, that the problem of achromatism may be considered as solved, but that that of aplanatism is far from being so. For the improvement, therefore, of objectives, the correction of the spherical aberration must be chiefly kept in view.

#### (6) Miscellaneous.

**Dr. Hudson's Presidential Address.**—The 'Times' of the 18th February contained the following leading article on this Address.

"An address such as the President, Dr. C. T. Hudson, delivered before the Royal Microscopical Society at its Annual Meeting last week is as surprising as it is delightful. All science has a tendency to grow more and more technical and elaborate. So complicated become



its processes, that they exceed the powers of all who cannot devote their lives to it. The details increase so constantly in number and variety that it is compelled to partition itself into infinitesimal provinces. Its mere language and vocabulary distract by their immensity and peculiarities. None but professed students are any longer able to master the diction and the classifications, which are perpetually being changed. The literature on any and every department follows the same rule of crabbedness and bulk. Its publications are too costly for the majority of pockets, and particularly for those which have other demands upon them. Intending naturalists have the authority of the President of the Microscopical Society for believing that all these impediments to the pursuit are superfluous and erroneous. The instrument from which the association derives its name might have been supposed to be principally responsible for the modern banishment of a popular character from inquiries into natural history. Dr. Hudson protests against the injurious suspicion, and deprecates earnestly the practices of professors of science in which it has originated. Specialists, he declares, are enemies against whom war should be waged. Natural history does not need, in his judgment, the uncouth terminology which is the bane of monographs. A host of creatures would, he is persuaded, live more comfortably within the common species, in which of old they congregated, than penned, as now, into separate little enclosures. The one essential for a naturalist is joy in the investigation of the wonders of life; and the freest cultivation of the propensity to indulge the pleasure is equally requisite for the progress of the true science of natural history. Other sciences are backed by their utility. On them arts are founded which ward off material dangers, or serve material interests. The structure of civilization, and most of the conveniences of human existence depend on the principles they embody. Dr. Hudson sees little of this practical bearing in the study for which he pleads. Though a few branches, especially researches into the minutest forms of life, may, he acknowledges, be of the profoundest consequence to human health and prosperity, in general the knowledge of natural history must be its own final reward. For the attraction of recruits to its camp it will, as hitherto, have, he thinks, to rely chiefly on the delight it yields. He is seriously afraid that the emotion may be choked, stifled, and killed before it has had a chance of maturing into a habit, by the exasperating resolve of specialists to encompass the whole subject with an atmosphere in which none but themselves can breathe.

This is a very grave indictment to proceed from the learned recesses of the Royal Microscopical Society. Its President based his remarks on the stumbling-block interposed by the caprices of classification, the addiction to technical terms, and the multiplication of species, to the enrolment of volunteers in the army of naturalists. Their real importance rests rather upon the degree to which the disposition he attacks is adverse to the advancement of science itself. Specialists would not be afflicted if they were left alone in their pursuit. They are inclined to resent and not to court the company of amateurs. They feel the eager inner enjoyment of their study which Dr. Hudson regards as the mainstay of the whole. To them the changes in classification are substantially necessary. Every fresh subdivision for which they can invent a

plausible excuse is for them an absolute enlargement of the territories they severally occupy. To a certain extent they probably could defend themselves successfully against their present critic. Definitions and frontier lines laid down a generation ago have been superseded and over-ridden by the fruitful discoveries of late years. The kingdom of nature has been found to be an agglomeration of a vast multitude of realms within realms. The new technical vocabulary in which it is described has had to be expressly manufactured to name orders of existence revealed only after the elder terminology had encrusted itself with a confusing significance. Partly it has been rendered indispensable by the demand of fellow workers in different regions of the globe for a common tongue. Melancholy as is the conclusion, and reluctant as everybody must be to come to it, the ancient simplicity and stability of scientific nomenclature are, it is to be apprehended, gone beyond recall. It does not follow, therefore, that the ponderous intricacy and restlessness of the system installed in their stead, of which Dr. Hudson complains, can prove any sufficient justification. A cry has been raised for the establishment of a tribunal to create a fair and uniform standard of judicial pains and penalties. In the world of science a Court is as much wanted for the revision of the vocabulary and classifications introduced by a legion of discretionary scientific jurisdictions. Formerly, when the field of natural science was virtually undivided, the terminology had to submit to a measure of central control. A Linnæus or a Cuvier would sanction or disallow. At present the distribution into an indefinite medley of special groups has given to the workers on them an autonomy they are not invariably qualified to exercise. Though it is too much to hope for a return of the golden age when naturalists spoke in a tongue understood of the people, and species were not continually splitting off under the disintegrating operation of the Microscope, at least there ought to be some sort of warranty against a repetition in natural science of the experiences of the Tower of Babel.

That would be for the benefit of specialists themselves. The unlicensed fabrication of terminologies and classifications cannot be agreeable to any of them, unless when they are personally engaged in the process. For the sake of the outside commonalty of persons simply endowed with delight in natural history, to whom Dr. Hudson was addressing himself at King's College, it is much to be wished that his professed brethren would give more encouragement than they have given of late to the pursuit in its older form. Without disrespect to the physiological aspects of the study, it is to be regretted that the view which treated it as primarily observation of the ways and usages of the stages of animated nature below the human has fallen comparatively into neglect. The President of the Microscopical Society has exhorted its members to prepare themselves for the profitable employment of microscopical investigation by diligent attention to living animals, their beauty and their actions. Nothing can be more astonishing than that science, with all its toil, has as yet discovered so few of their characteristics as sentient and moving creatures. How they exist, the arts by which they catch their prey or elude capture, the secret of their confidence or spite, the laws of their affections, their amusements, their

sense of humour, and their humours, their cleverness and their stupidity, are problems still for the most part remaining for natural history to answer. Its students will find but scanty information on them throughout the entire stately library of science. The system thus inculcated was followed by Gilbert White. Old fashioned as it appears now, it may well be that the path it points out leads more directly than those which modern philosophy prefers to the solution of the deep mysteries of the gradations of animate being and intelligence. With relation even to utility, which Dr. Hudson is ready to give up as off the naturalist's beat, there are questions fully worthy of his consideration. Miss Ormerod has shown that natural science has its uses for agriculture. Beside her particular charges there are other insect pests in plenty of which the world could be rid if naturalists would take the trouble to learn their habits. Where, for instance, is there a martyr of science willing to devote himself to a thorough search into the manners and morals of black-beetles, the things they love, and the things they hate? A naturalist who taught London to understand, and rout and extirpate, them would deserve any metropolitan honours he chose to ask. The County Council might feast him as lavishly as the City Corporation, and not the meanest ratepayer would grudge the cost of the entertainment."

**New Italian Microscopical Journal.**—We welcome the appearance of the first and second fascicles of the *Bollettino della Società Italiana dei Microscopisti*, the organ of the Italian Society of Microscopists. The Society, which embraces the whole of Italy, was founded on the model of the corresponding Societies in England and America; and its *Bollettino* will contain papers on the investigations of microscopists on animal and vegetable organisms, on petrology, on bacteriology, especially in its pathological relations, and on the structure of the Microscope and microscopical appliances. In addition to a number of minor articles and notes, the first number contains important papers on a new genus of green Algæ, and on two new genera of fossil Foraminifera, on a rock containing leucite from Etna, on the function of calcium oxalate in leaves, and two important contributions to bacteriology.

**Prof. Frey.**—The death is announced of the famous Zurich professor, Dr. Heinrich Frey, one of our Honorary Fellows since 1879, who after forty years of active work, retired only a few months ago. Frey was born at Frankfurt-on-the-Main, June 15th, 1822, and at twenty-five years of age had qualified, by brilliant preliminary studies, for the post of *Docent* in the University of Göttingen. In 1848, the Medical Faculty of Zurich nominated him Extraordinary Professor, and in 1851 Ordinary Professor. In 1855 he undertook the Professorship of Medicine in the Polytechnic of Zurich, and also the post of Director of the Microscopo-Anatomical Institute. From 1854 to 1856 he also filled the position of Rector in the "Hochschule." His researches in physiology were published in works which have been translated into nearly every European language, and are valued as models of lucid exposition. His book 'Das Mikroskop' has passed through eight editions, and was translated into English by Dr. G. R. Cutter. Prof. Frey was also an accomplished entomologist.



**Microscopy at the Paris Exhibition.**—The ‘Annales de Micrographie’ has concluded \* a series of brief articles on the Microscopes and apparatus at the Paris Exhibition of 1889, which, with those of Dr. Pelletan in the ‘Journal de Micrographie,’ and of Mr. Mayall in this Journal, constitute, so far as we know, the only record of this section of the Exhibition.

**Price of the new Objective of 1.63 N.A.**—We understand that the price of this objective is not 10,000 francs or 400*l.*, but 1000 francs or 40*l.* An extra nought seems to have crept into the original report on the subject.

### β. Technique.†

#### (1) Collecting Objects, including Culture Processes.

**Friedländer’s Microscopical Technique for Clinical and Pathological Purposes.**‡—Dr. C. J. Eberth has just published the fourth edition of C. Friedländer’s well-known work on microscopical technique. The author has not only revised the whole, but made considerable improvements. For example, Section II., which treats of the microtome, is much enlarged, and Section III., dealing with the methods of preparation, such as hardening, imbedding, &c., has evidently had a good deal of pains bestowed on it. Some of the sections, e. g. Section V., “Observing Living Tissues,” are unaltered, and some sections appear to contain views of doubtful value, but on the whole the work is one which can be recommended to the bacteriologist and the pathological anatomist.

**Artificial Cultivation of Ringworm Fungus.**§—Dr. H. L. Roberts’ observations, and his conclusions from a series of cultivation experiments made on *Trichophyton tonsurans*, are very interesting. A portion of scalp affected with ringworm was first cleansed with a 1:200 solution of corrosive sublimate. The broken hairs were then removed with forceps, and their bulbous ends having been snipped off, the pieces were dropped into flasks containing saccharine infusion of malt and alkalinized beef-broth, and incubated at 30° C.

The fungus was observed to have started developing in 24 hours, and in three or four days from the formation of the primary colony secondary deposits were visible. If the colonies rose to the surface, they speedily became covered with a white powder. On microscopical examination the mycelium was found to be regularly septate, and filled with a granular protoplasm. When development takes place in air, the mycelium becomes finer, the segments are small, and the terminal fruit-bearing filament may end in an ampulla. The spores are pear-shaped, are attached by their narrow end, and are sometimes seen to project from the ampullæ.

Inoculation experiments on guinea-pigs, and on the author’s own

\* Ann. de Micrographie, ii. (1890) pp. 168–71.

† This subdivision contains (1) Collecting Objects, including Culture Processes; (2) Preparing Objects; (3) Cutting, including Imbedding and Microtomes; (4) Staining and Injecting; (5) Mounting, including slides, preservative fluids, &c.; (6) Miscellaneous.

‡ ‘Friedländer’s Microscopical Technique,’ 4th edition revised by C. J. Eberth, Berlin, 1889. Cf. Centralbl. f. Bakteriologie u. Parasitenkunde, vii. (1890) p. 72.

§ ‘British Journal of Dermatology,’ i. (1889) pp. 359–65 (3 pls.).



arm, gave the usual characteristics of ringworm. The author concludes that *Trichophyton* is "a fungus able to vary its form and activity according to the physical and chemical properties of the soil on which it grows." As a saccharine medium has been found to be the most favourable soil, it follows that the animal skin is unsuitable; hence "the ringworm fungus vegetates, but does not develope" there.

BEHRENS, W., KOSSEL, A., U. SCHIEFFERDECKER, P.—*Das Mikroskop und die Methoden der mikroskopischen Untersuchung*. Bd. I. *Die Gewebe des menschlichen Körpers und ihre mikroskopische Untersuchung*. (The Microscope and the Methods of Microscopical Investigation. Vol. I. The Tissues of the Human Body and their Examination by the Microscope.)

Braunschweig (Bruhn), 8vo, 1889, 315 pp., 193 figs.

BONEVAL, R.—*Nouveau guide pratique de technique microscopique appliquée à l'Histologie et à l'Embryogénie*. Suivi d'un formulaire indiquant la composition des réactifs employés en anatomie microscopique. (New Practical Guide of Microscopical Technique applied to Histology and Embryology. Followed by formulæ for the reagents employed in Histology.)

8vo, Paris, 1889, 21 figs.

DAVIS, G. E.—*Practical Microscopy*. New ed., Philadelphia (Lippincott), 1889.

RAMON Y CAJAL, S.—*Manual de histología normal y de técnica micrográfica*. (Handbook of normal Histology and Microscopical Technique.)

Valencia (Ostega), 4to, 1889, 692 pp., 203 figs.

## (2) Preparing Objects.

**Mode of Studying Free-swimming Larvæ.\***—Dr. G. C. J. Vosmaer recommends that free-swimming larvæ should be put into a glass, the bottom of which is covered by a loose, thin sheet of collodium; to this they attach themselves readily. The spot to which a larva is attached can be cut out under water whenever required. The collodium is transparent and easily cut with the larva. If it is desired to examine the base, the collodium may easily be dissolved. The preservative fluid recommended is that which Kleinenberg used for *Lopadorhynchus*; this gives by far the best results, cilia, for example, being hardly shorter than in the living animal.

**Examination of Renal Organ of Prosobranch Gastropoda.†**—M. R. Perrier has used three methods in examining the renal organs of Prosobranch Gastropods. The examination of living tissues, teasing, and serial sections have been the methods employed. The great difficulty to overcome has been the extreme alterability of the tissues, and the delicacy of the renal cells has been noticed by all observers. Under the influence of whatever reagents, the epithelium becomes completely destroyed unless sufficient precautions are taken. Owing to the changes which are continually taking place it is necessary to at once arrest the secretion. Ordinary fixing reagents, and particularly osmic acid, are of no use for this purpose; indeed, they seem to make the secretion more active. The best results have been obtained with acetic or picric acid, or, still better, a mixture of the two; picro-sulphuric acid has also been of use. The organ must be cut out of the body as rapidly as possible, plunged for one or two minutes into a 1 per cent. solution of osmic acid, washed rapidly, and left for some hours in a mixture of picric and acetic acids. It must then be put in 70 per cent. alcohol for as long a period

\* Tijdschr. Nederl. Dierk. Ver., ii. (1889) p. 289.

† Ann. Sci. Nat. Zool., vii. (1889) pp. 71-9.

as may be wished, when it is ready for sectionizing. Sections were made with one of Dumaige's automatic microtomes, which gives the most excellent results. When fixed, the specimen was stained with picrocarminate of ammonia, which is the best of all; after one or two days in a solution of this material, the preparation must be gradually hardened in alcohol of 70°, 90°, and 100°—one day in each, fresh absolute alcohol being applied two days in succession. To this last fluid methylene-blue may be added, as it will stain the protoplasm and muscles, while having no influence on the nuclei. The object should next be successively placed in cedarwood-oil, paraffin with this oil, and pure paraffin. As the renal cells of Molluscs are very small, the sections should be extremely fine, and it is well not to have them more than 1/400 mm. in thickness.

When about to be placed on the slides it is well to make a limpid solution of 2 or 3 parts of gelatin in 100 parts of water; this, after careful filtration, should be placed on the slide, and the rows of sections will be found to swim in it; they can then be arranged as desired. The slide must then be placed on a plate warmed to about 40°, but not hot enough to melt the paraffin. At this gentle heat the sections become spontaneously extended in the gelatin, and all the creases in them will be found to disappear. The gelatin may now be left to dry. When the gelatin is dry, the paraffin may be easily dissolved away and the sections mounted in balsam.

**Mode of Preparing Ova and Embryos of *Blatta Doryphora*.\***—Mr. W. M. Wheeler used the following method in his studies on Insects' eggs:—The ovarian ova in all stages up to maturity were dissected out in normal salt solution, and hardened for fifteen minutes in Perenyi's fluid. They were then transferred to 70 per cent. alcohol, which was changed several times at intervals of an hour, and were finally preserved in alcohol of the same strength. When stained with borax-carminé and sectioned, the yolk retained none of the red stain, while the chromatin of the nucleus shone out as a glistening deep red spot. Perenyi's fluid rendered the chorion of the mature ovarian egg pervious to borax-carminé. Hardening in a saturated aqueous solution of corrosive sublimate gave good results with young ovarian eggs. Oviposited eggs were killed by placing the capsules in water slowly heated to 80°–90° C. The two lips of the crista of the capsule were then separated by the aid of fine forceps, and pieces of the walls torn away, till the eggs could be easily pushed out of the compartments formed by their choria. The ova thus isolated were either transferred directly through 35 per cent. (10 min.) to 70 per cent. alcohol, or they were left for 15 minutes in Kleinenberg's picrosulphuric acid, and after repeated washing in 70 per cent. alcohol, preserved in alcohol of the same strength. Both methods gave equally good results.

Though he has succeeded in dissolving the chitin of the ootheca with sodium hypochlorite, the method of tearing off the walls after heating to 80° C. gave such satisfactory results that he adhered to it through his work. He has found Grenacher's borax-carminé in every way the most expedient and reliable staining fluid. Eggs and embryos, up to the time

\* Journ. Morphology, iii. (1889) pp. 292–3.

when the cuticle develops, were stained before imbedding in paraffin; the sections of other embryos were stained on the slide after attaching them with Mayer's albumen fixative. Beautiful results in preparation were obtained by heating the eggs to 80° C. for 10 minutes in Kleinenberg's picrosulphuric acid (with 3 volumes of water), and preserving in 70 per cent. alcohol. By this process the envelopes, which in the fresh egg adhere closely to the yolk, dilate and stand off from the surface of the egg, and except in the very youngest stages can be rapidly and easily removed with the dissecting needles.

**Investigation of *Derostoma unipunctatum*.**\*—Herr K. Lippitsch found his specimens of this worm preserved in sublimate, osmic acid, or osmic-acetic acid. The staining reagents used were hæmatoxylin, picrocarmine, and alum-carminé; osmic acid is not a good preservative, as it causes deformations of the epithelium, but sublimate is, as with other Turbellaria, very good. After treatment with hæmatoxylin for two or three hours all the glands become very clear, and the same reagent is good for the nervous system when osmic-acetic acid has been previously used. Twenty-four hours' stay in picrocarmine is useful for the study of the epithelium, nervous system, musculature of the pharynx, and connective tissue. Alum-carminé is also to be recommended.

**Preparation of Horny Teeth of Batrachian Larvæ.**†—Herr E. Gutzeit preserved his larvæ in 0·2 per cent. chromic acid or in sublimate, and afterwards placed them in alcohol; they were stained *in toto* by hæmatoxylin or picrocarmine. Paraffin was generally, and soap only rarely used as imbedding material. The sections were attached by oil of cloves and collodion, and Canada balsam was added. Wickersheim's fluid or Müller's solution was used for macerating purposes, and preparations so made were preserved in glycerin-gelatin.

**Production of Colourless Spirit-preparations.**‡—Herr H. de Vries proposes the following process for this purpose:—By adding two parts by volume of strong hydrochloric acid to 100 parts of alcohol, the production is prevented of brown pigments in the parts of plants which are plunged when living into the mixture; and the preparations thus obtained are much more beautiful than by the ordinary method. Even plants in which the brown pigment is very conspicuous, such as *Orobanche*, become white in this mixture; the only case of failure was with *Aucuba*, older portions still retaining their brown colour, while younger portions became quite white.

**Observation of Nuclear Division in Plants.**§—Prof. D. H. Campbell recommends for this purpose the pollen-mother-cells of *Allium canadense* or of *Podophyllum peltatum*, taken from a bud. They should be crushed or teased out into a mixture of equal parts of acetic acid and water, when the pollen-mother-cells are at once recognizable by their thick colourless walls; if they are already in the required stage of division, they may be stained by acetic methyl-green or gentian-violet, made by adding a sufficient quantity of a saturated alcoholic solution of gentian-

\* Zeitschr. f. Wiss. Zool., xlix. (1889) pp. 148-9.

† T. c., p. 65.

‡ Ber. Deutsch. Bot. Gesell., vii. (1889) pp. 298-301.

§ Bot. Gazette, xiv. (1889) p. 199.



violet to a mixture of two parts of distilled water and one of acetic acid. If a drop of this mixture is added to the preparation containing the pollen-cells, the nuclei will almost instantly be coloured a deep blue-purple, while the cell-protoplasm remains colourless and entirely uncontracted. The staining fluid may now be removed by blotting-paper, and the preparation mounted in dilute glycerin. Specimens prepared in this way, especially when first made, show all the finest details of the structure of the nucleus.

**Fixing the Spores of Hymenomycetes.\***—Inasmuch as a solution of Canada balsam in turpentine-oil has a tendency to oxidize and become cloudy after having been prepared for a year, Prof. C. O. Harz now proposes to substitute lavender-oil or petroleum for the turpentine-oil.

**Direct Impressions of Plants.†**—M. Bertot obtains direct impressions of plants in the following way:—The plant is first saturated with oil by placing between pieces of paper soaked in oil, and an impression of it is then obtained in oil on white paper. The paper is now treated with graphite, and the oily places are thus turned black and a perfect impression of the plant obtained. The paper is now freed from excess of graphite by wood-ash. To fix the image, powdered colophony is mixed with the graphite, which sinks into the paper when slightly warmed. Spots which sometimes appear upon the paper may be removed by soaking the paper in an aqueous solution of tragacanth.

**Demonstrating Tubercle Bacilli.‡**—Dr. Bliesener recommends the following method as being very expeditious:—The cover-glass, having been dried in the air and passed thrice through a flame, is placed with the sputum layer uppermost on a metal plate about 5–6 cm. square fixed to a stand so as to keep it horizontal. Five or six drops of carbolic fuchsin are then dropped on with a pipette and the metal plate warmed until the fluid begins to evaporate. The flame is then removed, and then the cover-glass, after remaining on the plate for about a minute, is washed with water previous to its being dropped on the acid contrast fluid (methylene-blue 1·5, H<sub>2</sub>O 100, H<sub>2</sub>SO<sub>4</sub> 25). In about fifty seconds it is removed, washed in water, and examined.

The foregoing staining procedure, if combined with Biedert's method of examining sputum, is said by the author to be very satisfactory. Biedert's method consists in boiling the sputa with water to which some drops of caustic soda have been added.

**Agar-agar as a Fixative for Microscopical Sections.§**—M. A. Gervis, who recommends agar-agar as a medium for fixing sections, imbedded in paraffin, on slides, proceeds as follows:—Half a gramme of agar having been cut up into small pieces, is allowed to soak for some hours in 500 grammes of distilled water. When it has swelled up it is boiled for about a quarter of an hour in order to completely dissolve the agar.

\* SB. Bot. Ver. München, Nov. 11, 1889. See Bot. Centralbl., xl. (1889) p. 345. Cf. this Journal, 1889, p. 461.

† Bull. Soc. Linn. Normandie, ii., 1887–8 (1889) pp. 442–5. See Bot. Centralbl., xl. (1889) p. 285.

‡ Deutsche militärärztl. Zeitschr., xviii., pp. 406–9. Cf. Centralbl. f. Bakteriologie u. Parasitenk., vii. (1890) pp. 72–3.

§ Bull. Soc. Belge de Microscopie, xv. (1889) pp. 72–5.



When cold the liquid is filtered through a fine cloth and kept in stoppered bottles. A small piece of camphor to prevent the development of micro-organisms may be placed in each bottle. The slides must be perfectly clean, and should be boiled in water acidulated with hydrochloric acid, and, having been rinsed in distilled water, are dried with a perfectly clean cloth. Upon the slide is brushed over a layer of this fixative, excess of which is immaterial, as it is easily removed later on.

The sections are then arranged on the slide with a fine brush. Directly this is finished the slide is gently heated over a Bunsen's burner. The paraffin is to be softened only, and not melted. Any unevenness or folds in the sections at once disappear. As the slide cools the paraffin sets; and now if there be too much of the fixative, it may be removed by just sloping the slide so as to drain it off. The fixative must now be allowed to dry thoroughly, and it is best to leave the slides just covered from dust, &c., until the next day.

The paraffin is then dissolved in warm turpentine or in chloroform, and these last removed by means of a little strong spirit. If the preparations have been stained before imbedding, nothing remains to be done but to dehydrate the section in absolute alcohol, clear up in oil of cloves, and mount in balsam. If not stained, the slide is placed in the staining solution, and when withdrawn goes straight into spirit.

The advantages of this method are that the fixative is liquid at ordinary temperature, the sections are easily arranged, all folds and creases are completely removed, and no air-bubbles trouble the manipulator. As the fixative is an aqueous solution, the cells of vegetable preparations swell up in it to their original size. When properly dried, the fixative is insoluble in all reagents and alkalies, &c., except water, which causes it to swell up and tends to loosen it from the slide. Unless the agar-layer be thick, the fixative does not become coloured in the staining solutions.

The preparations may be mounted either in balsam or glycerin.

### (3) Cutting, including Imbedding and Microtomes.

**Florman's Method of Imbedding in Celloidin.\***—Dr. S. Apáthy raises several objections to the method of celloidin imbedding advocated and practised by Florman. The principles of the two methods are diametrically opposite. Florman advises imbedding in glass capsules in a thin solution of celloidin, and then solidifies by allowing the slow evaporation of the solvents, ether and alcohol. Dr. S. Apáthy's method consists in transferring the objects to solutions of celloidin of increasing thickness, and in only allowing evaporation of the ether-alcohol when the thickest solution has been reached. The objections to Florman's method seem chiefly to consist in the possibility of delicate objects being distorted, owing to the contraction of the celloidin, and also disarranged; in the long time required for imbedding; and in the fact that the undermost layer is usually left behind when the mass is extracted from the capsule.

But it is possible that the two microtomists are in the habit of dealing with different materials; the one with delicate objects, the

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 301-3.

structure of which becomes materially altered by the slow contraction of the imbedding mass, and the other with material which has been previously well hardened and is in itself dense, so that the defects alluded to are unperceived.

#### (4) Staining and Injecting.

**Kühne's Methylene-blue Method of Staining Bacteria.\***—This method is especially recommended for staining bacteria in sections of animal tissues, although it is equally applicable to cover-glass preparations made from fresh tissues. The usual differences in the method of staining cover-glass preparations and sections are to be observed.

The advantages to be derived from this method are found in its being applicable to all known forms of bacteria. It eliminates the use of special stains for certain micro-organisms where only their presence is to be demonstrated. It possesses superior powers of differentiation between bacteria and the tissue elements. The method as given by Dr. Kühne † is essentially as follows.

The sections which have been cut by the ordinary method (although Dr. Kühne recommends the freezing microtome for this purpose), are transferred directly from alcohol to a watch-glass containing carbol-methylene-blue. (1) The sections should remain in this staining fluid for about half an hour; some bacteria, such as the bacillus of leprosy, requiring a longer time, one to two hours. If the sections remain in the staining fluid for a much longer period, the differentiation between the germs and tissue-elements becomes more difficult.

After staining for the desired length of time, the exact period of which will have to be determined by test experiments for the different germs and tissues, the sections are rinsed in clear water and then placed in acidulated water (2) until they become a pale blue. They are then washed in a weak watery solution of carbonate of lithium (3), and again placed in clear water. This part of the procedure is very important, and to insure good results should be performed with much care. The time that the sections should remain in the decolorizing agents varies with their thickness, histological structure, and the intensity of the stain, making it impossible to give any definite rule to be followed. The degree of decolorization can be very nearly determined at any moment by moving the sections about in the fluid by means of a glass rod. If the section is very thin, or if there are other reasons why it should take up very little of the stain, a momentary immersion in the acidulated water is sufficient. In all cases where the staining process is completed the sections should have a pale blue colour, for if darker, the over-stained corpuscles and cell-nuclei of the tissue would obscure the bacteria. In cases where it is feared that too much colour has been removed in the acid a drop of a saturated watery solution of methylene-blue should be added to the lithium-water.

After the sections have remained in the water for some minutes they are dehydrated in absolute alcohol in which, in difficult cases, a little methylene-blue may be dissolved, and then transferred to a watch-glass

\* Amer. Mon. Micr. Journ., x. (1889) pp. 259-60.

† Kühne, 'Praktische Anleitung zum mikroskopischen Nachweis der Bakterien im tierischen Gewebe,' p. 15.

containing methylene-blue anilin oil (4). The sections can be dehydrated in the alcohol without injury to the stained bacteria. The sections are now transferred to pure anilin oil, in which they are rinsed and then placed in some essential oil, as turpentine, where they should remain for two minutes. In order that the sections should be perfectly cleared they are transferred from the turpentine to xylol, from which they are mounted in balsam. It is recommended that the sections should pass successively through two xylol baths in order to secure absolute elimination of the anilin oil. The xylol may be used for a considerable number of sections.

Dr. Kühne employs a glass rod for transferring the sections from one solution to another instead of the ordinary spatula or section-lifter. The end of a small glass rod is immersed in the fluid containing the section, which is allowed to fold itself over the rod, and in this position it is lifted from the fluid. The end of the rod is then gently immersed in the second liquid, where the section unfolds itself from the rod and floats upon the surface. In this way the danger of tearing the section is diminished and the time required for their transfer from solution to solution is much shortened. This is an important consideration where a large number of sections are to be stained.

(1) *Carbol-methylene-blue*.—This is prepared by grinding in a mortar 1.5 grams of methylene-blue with 10 ccm. of absolute alcohol until dissolved; 100 ccm. of 5 per cent. carbolic acid are gradually added and thoroughly mixed with the alcoholic solution. The resulting liquid is preserved in a well-stoppered bottle, until used. When only a small quantity is to be employed it is better to prepare only a half, or a quarter even, of the above quantity, as its staining power is diminished by long standing. It should always be *filtered* before using.

(2) *Weak acidulated water*.—To 500 ccm. of distilled water add 10 drops of nitric acid.

(3) *Lithium-water*.—To 10 ccm. of distilled water add from 6 to 8 drops of a saturated watery solution of carbonate of lithium. The saturated solution may be used as a decolorizing agent in sections with over-stained nuclei.

(4) *Methylene-blue anilin oil*.—About one-half gram of methylene-blue is ground in a mortar with 10 ccm. of pure anilin oil. When the oil is saturated with the colouring matter the entire mass is poured unfiltered into a vial, where the undissolved colouring matter will settle, leaving the saturated supernatant oil clear. To a watch-glass of pure anilin oil add a few drops of the saturated methylene-blue-oil until the desired degree of colorization is obtained.

#### (5) Mounting, including Slides, Preservative Fluids, &c.

**New Form of Clip for Balsam Mounting.\***—Mr G. H. Bryan says that there are few practical microscopists who do not admit that the spring-clips which have for so many years been used in mounting objects in balsam are a failure. The usual query which has been repeatedly asked is, "Why does air run in as soon as the clip is removed?" The answer is pretty obvious, viz. that the object yields to the pressure of the clip as long as it is subject to it, but as soon as

\* Journ. of Microscopy, iii. (1890) pp. 45-7 (1 fig.).

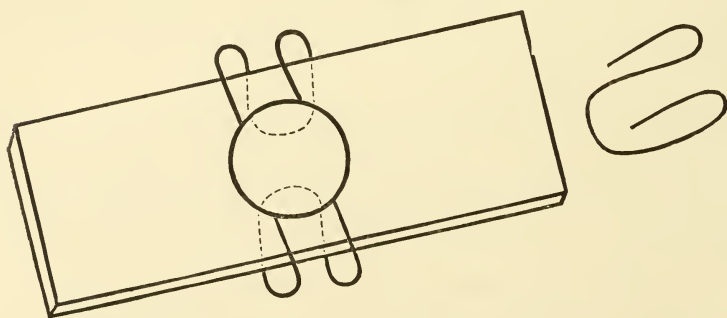
that is taken off, the elasticity of the specimen causes the latter to lift the cover up again, and what naturally happens? Why, of course the air runs in, because "nature abhors a vacuum."

Nor is this the only fault of spring-clips, for even a moderate amount of pressure is sufficient to damage many delicate specimens. Take the case of sections of stems of plants; the effect of squashing very frequently makes the cells and vessels in parts turn on one side, and where each cell should by rights be in its natural place, nothing is seen but a jumbled mass of tissue. Yet spring-clips are still frequently used in balsam mounting, the reason being that they fulfil a twofold purpose. One use of them is to produce pressure. This, as we have seen, is a bad purpose. Not but what a certain small class of specimens require flattening out, but this must be done before mounting them; it is too late to make the attempt when they are in the balsam. Their other use is to keep the cover in place while the balsam is hardening, and it is for this alone that they are usually used. They do not accomplish this end practically, for as a general rule, in applying the clip, the cover gets slightly shifted to begin with; moreover, they are almost certain to tilt the cover on one side or the other unless supports have been placed round its edges.

Nearly two years ago the idea occurred to the author that what was wanted was an arrangement that would hold the cover in its proper position by firmly gripping the edges instead of pressing down on the top of the glass. Since then he has mounted a number of slides, using these "pressureless edge clips" until the balsam has hardened, and with such success that now he "uses no other."

Fig. 29 shows one of the pressureless clips of the natural size, and how they are used for keeping the cover of a slide in its proper position.

FIG. 29.



It will be noticed that two clips are necessary, and when in use they firmly clip the slide only, their four points resting against the edges, not on the top of the cover-glass. In this way the cover is perfectly firmly held in position; it is impossible for it to slip out of place, while no pressure is applied to the object. In applying them to the slide, they are first clipped on anywhere, and then pushed up until their points touch the edges of the thin glass circle; this can generally be accom-



plished without shifting the latter perceptibly. The slide can then be handled with perfect impunity, no matter how soft the balsam may be, and a good deal of the superfluous balsam may be removed if care be taken not to displace the clips. The balsam may then, if advisable, be hardened under more or less heat; the top of a hot-water cistern is a first-rate drying-ground for the purpose. After about a fortnight in such a position, even slides mounted in ordinary balsam will generally be found sufficiently hard to be cleaned with perfect safety, but theoretically it is evident that the time taken to harden under the cover is the same as the time taken to harden in an open vessel by a layer of balsam whose thickness is one-quarter the diameter of the cover-glass. When the balsam is fully set the points of the clips will be firmly stuck down on the slides, but there is no difficulty in pulling them off; if necessary the wires might be heated, but this is not required.

Mr. Bryan now makes the clips of brass wire, the length required for each being about  $2\frac{1}{2}$  in. It is advisable to make the clips of different sizes, to accommodate the different sizes of cover-glasses, and, properly, the distance between the points of the clip should be about seven-tenths of the diameter of the covers for which it is made. For use with some mounts, it is convenient to bend the points of the clip inwards, while if the object be a very thick one the points turned down will be found very useful. Where neither of these things is done, the ends may be filed off at a suitable angle, so that they hold the edges of the cover more firmly.

**Quick Method of Mounting Microscopical Preparations.\***—K. Schilbersky, jun., finds that numerous micro-organisms can be permanently preserved by mounting them in an aqueous fluid (water or dilute glycerin) or glycerin-jelly, by means of the following simple device.

The object is (suppose) in water, and lying about the centre of the cover-glass. Any excess of water is then to be removed with bibulous paper, so that the edges of the cover-glass are quite dry; or this may be effected by evaporation. Before the edges are dried it is advisable to pass under the cover-glass a droplet of dilute carbohic acid, to prevent the development and settlement of schizomycetes, &c. When the edges are dry, the corners of the cover-glass are to be fixed with asphalt so thick that it runs with difficulty. Along the margins and corners of the cover-glass the asphalt is to be applied by means of a brush or glass rod, in such a way that the cover-glass is not moved. When complete, the ridge may be covered with Canada balsam.

If the object is in glycerin or other fluid not a solvent of asphalt, the procedure is quite similar, but extra care must be taken with glycerin to remove all traces of it outside the edge of the cover-glass, otherwise the asphalt will not stick. This is best done with a brush or strip of blotting-paper moistened with spirit. Instead of asphalt, balsam may be used, but it is not quite so serviceable.

If the object is to be mounted in glycerin-jelly, the following modification is adopted. The object (usually obtained by maceration) is placed under a cover-glass in water or glycerin, and the latter is then absorbed by means of a pipette or blotting-paper to one-third. The

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 273-83.

cover-glass is next carefully raised, and a small piece of glycerin-jelly put in the water or glycerin which remains on the slide. The slide is then heated to melt the jelly. Air-bubbles having been pricked out, the cover-glass is replaced in its original position. It is advisable to take too little rather than too much glycerin-jelly, as the deficiency is easily made good afterwards.

**Venetian Turpentine as a Mounting Medium.\***—Dr. J. Vosseler recommends Venetian turpentine for mounting specimens permanently, on the ground that it possesses all the advantages of the ordinary resinous media employed for the purpose; that in some respects it is superior to them; and that it is cheaper.

Venetian turpentine is obtained from the larch, and is found to consist of a resin and an ethereal oil; consequently it is to be classed among the balsams. In colour and consistence the raw material resembles honey, but is sometimes brownish from admixture with minute fragments of bark.

To obtain a suitable solution the author merely mixes equal volumes of the crude balsam and 90 per cent. alcohol in a tall glass vessel, the top of which is protected from dust, and allows this to stand in a warm place for three or four weeks. The processes may be hastened by increasing the heat in an incubator. A clear yellowish or sometimes greenish mixture is obtained, and this is at once ready for use, as the impurities have already sunk to the bottom. These impurities may be extracted with greater rapidity by filtration. If the filtrate should be of a brownish hue, it must be thickened anew until the yellow colour returns. If the balsam be applied in a too fluid form it may become milky: should this turbidity be not too great, it will be found to disappear in a day or two; if considerable, the balsam must be dissolved out in 96 per cent. alcohol, and the specimen be remounted. The ordinary consistence of Canada balsam is that most suitable for the solution of Venice turpentine.

Prepared in the foregoing manner, Venice turpentine mixes with the reagents constantly in use in histological technique—for example, ether, alcohol 100–96 per cent., chloroform, pure carbolic acid, creosote, xylol, benzol, toluol, and the ethereal oils. Preliminary clarification of sections or pieces of tissue is quite superfluous, although when an entire animal, e.g. a small arthropod, is to be mounted, it is preferable to pass it through turpentine or creosote first. Hence, with a few exceptions, specimens are to be transferred directly from 96 per cent. spirit to this medium.

The finer details of structure are better shown in the medium than in dammar or balsam, but it is remarked that these details may disappear shortly after mounting, to reappear again on the second or third day. The medium behaves towards staining agents in the same way as other resinous substances, and is perfectly suited for specimens and sections imbedded in celloidin or paraffin.

The only inconvenience connected with the medium indicated by the author is that it is as slow to dry as dammar; but when dry it is harder and less brittle than balsam or dammar.

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 292–8.

In order to examine specimens just mounted with an immersion lens, the author mentions the following device for preventing the cover-glass from slipping. A couple of needles, made hot, are laid along two sides of the cover-glass. This causes the resin to thicken from evaporation of the solvent. Of course, all the four sides might be banked up in this way, and the device is quite suitable for similar conditions of balsam or dammar.

**Fixatives for Diatom Preparations.\***—Herr E. Debes, after alluding to the isobutyl-alcoholic solution of shellac introduced by Dr. Witt, and which, though eminently suitable for a mounting medium, is equally difficult to make, proceeds to discuss two kinds of media convenient for mounting diatoms. These are resinous and gelatinous preparations. The resins are certain copals, and these are divisible into three classes according to their solvency in turpentine. To the first class, which is quite insoluble, belongs Zanzibar copal; to the second, Manila or soft copal—this is only imperfectly soluble; the third class includes those resins which, being quite soluble, are omitted.

The Zanzibar, or insoluble copal is made into solution with isobutyl-alcohol, after having been previously treated with turpentine to dissolve out any resinous matter that may be present. The filtrate is then dissolved in isobutyl-alcohol and again filtered. The solution is quite colourless and clear, and is at once ready for use.

The diatoms are fixed by placing on a 3-mm. cover-glass a small drop of the liquid, which spreads itself out all over the cover. The cover-glass is then put on a metal plate, heated by a spirit-lamp, and when the proper degree is arrived at, the diatoms are arranged. This degree is estimated by placing close to the cover-glass a small fragment of resin on a bit of cover-glass, and when the fragment is quite dissolved the correct degree of heat is indicated and the source of heat removed.

Another way of estimating the proper amount of heat is to place a small strip of white writing-paper on the hot plate, and when this turns colour (white to yellow or brown), the source of heat is removed. After having been heated, both these resins (Manila and Zanzibar) become less soluble, an inconvenience which, as may be understood, may cause disasters if not properly anticipated.

The gelatinous media are made from gelatin or isinglass. Two gm. of pure white gelatin are dissolved in 70 ccm. of glacial acetic acid (or 3 gm. of isinglass in 75 ccm.), the mixture being placed in a well-stoppered bottle. By frequent shaking, the solution is effected in three or four days. The process may be hastened by heating in a water-bath. If isinglass be used the solution must be filtered to get rid of fat and fibres. Five gm. of the solution are then diluted with a mixture of 3 gm. of ethyl-alcohol and 1.5 gm. isobutyl-alcohol. The mixture is made by squirting in small quantities of the latter through a pipette, and constantly shaking. If a cloudy or opalescent precipitate be formed, a little more acetic acid must be added.

The solution must be put in a well-stoppered bottle and kept in a cool dark place. The fluid, which keeps well, is put on the cover-glass &c., with a pipette; a small drop runs out peripherally to form a thin

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 283-92.

even layer. The diatoms are laid on dry, and fixed in position by merely breathing ever so lightly upon the cover-glass.

**Sterilization of Water by the Chamberland Filter.\***—M. L. Dor finds from several experiments that Chamberland's filter may be confidently relied on for removing bacteria, for by means of it the author has succeeded in rendering the water perfectly free of germs.

**Microchemical Test for Alkaloids and Proteids.†**—M. L. Errera finds, as the result of numerous and prolonged experiments between alkaloids and proteids, that alcohol acidulated with tartaric acid fulfils all the conditions required for a good test between these two classes of highly organized bodies; for the alkaloids are removed by means of the tartaric acid, and the proteids remain behind. Hence little difficulty will be experienced in distinguishing the one class of compound from the other. The experiments were made upon colchicine, pepton, mucor, ciguë, and lupin.

**"Air-gas" for Bacteriological Work.‡**—Dr. O. Katz, who had to work on Rodd Island, N. S. Wales, where the ordinary appliances of civilization were not available, made use of the "Alpha patent gas-making machine." This apparatus produces gas in the shape of a mixture of atmospheric air and the vapour of petroleum spirit (gasoline), the mixture being called air-gas. By means of weights atmospheric air is pumped through a drum into a chamber, where it is impregnated with the vapour of the volatile fluid. The mixture then passes into a gasometer, from which the burners are supplied automatically. The author used a 40-light machine capable of yielding 200 feet of gas an hour.

The author considers that air-gas makes an efficient substitute for coal-gas for ordinary lighting purposes, and he also used it with success for heating thermostats, and also for other bacteriological purposes.

#### (6) Miscellaneous.

**Changes in the Firm of Zeiss.§**—The firm of C. Zeiss, in Jena, has advertised by circulars a change in their management. Dr. Ernst Abbe, who has hitherto acted as general manager of the firm, was, on November 29th, 1889, admitted into the firm as partner with Dr. R. Zeiss, and has now undertaken the sole management of the company so formed. At the same time power of procuration of the firm has been given to Dr. Otto Schott, of Jena; and Dr. Siegfried Czapiski, of Jena, has also been authorized to represent the firm in all business matters.

**Correction, by Dr. H. van Heurck.**—Dr. H. van Heurck, Director of the Jardin Botanique, Antwerp, requests us "to correct an error, or rather an omission," which occurred in his note on *Pleurosigma angulatum*.||

He wrote:—"The last photograph, No. 6, shows that the valve of *Pleurosigma* is formed of two layers." In writing this phrase he states: "I had in view the upper membrane and the intermediate layer, which are

\* Lyon Médical, 1889, No. 23. Cf. Centralbl. f. Bakteriöl., vii. (1890) p. 75.

† Annales Soc. Belge de Microscopie (Mémoires), xiii. (1889) pp. 72-121.

‡ Proc. Soc. Linn. N.S.W., iv. (1889) pp. 328-30.

§ Zeitschr. f. Instrumentenk., x. (1890) p. 37.

|| *Ante*, p. 104.



seen in this photograph, the lower membrane which is beneath not being visible. My clerk omitted the two words "at least," thus completely altering the sense and placing me in contradiction both with the statements in my Synopsis, published in 1885, and with the note on the 'Structure of Diatom Valves' which I recently sent to the Royal Microscopical Society, in which I everywhere admit the existence of three layers."

**New Photograph of *P. angulatum*, by Dr. H. van Heurck.**—At the March meeting of the Society a photograph, by Dr. H. van Heurck, was exhibited of *P. angulatum*, produced with Zeiss's apochromatic objective of 1.6 N.A., in further elucidation of Dr. van Heurck's views on the structure of diatom valves.

The note accompanying the photograph was as follows:—

"I have the honour to submit to the Royal Microscopical Society a photograph of *P. angulatum*, made with the objective of 1.6 N.A., using strictly axial illumination. The fracture of the upper edge shows clearly that the "beads" are holes in the intermediate layer, and that the form of these holes (beads) is hexagonal, as maintained by Mr. Smith and myself. The form of the small bar on the extreme top, which is the part of the negative focused, shows that the "beads" cannot be round."

**The Formation of Images in the *Pleurosigma formosum*.**—Mr. E. M. Nelson communicated the following note to the Society at the meeting of the 19th March:—"It was stated at the January meeting of the Royal Microscopical Society that it was impossible to produce images in the markings on a *P. formosum*. Some years ago it was said that images formed by the primary structure of coarse diatoms, such as *Triceratium* and *Coscinodiscus*, proved that the markings were lenticular. With this opinion I did not agree, and was led to investigate the subject. I not only confirmed the experiment with regard to the coarse diatoms, but eventually succeeded in producing images in the *P. formosum*. I also produced images in minute holes punctured in a piece of tinfoil. This latter experiment shows that the production of images in diatom markings does not prove that they are lenticular. I have now made a photomicrograph of a *P. formosum* with images formed in the markings  $\times 2000$ . The images might have been made more distinct had more time been expended on the photomicrograph."

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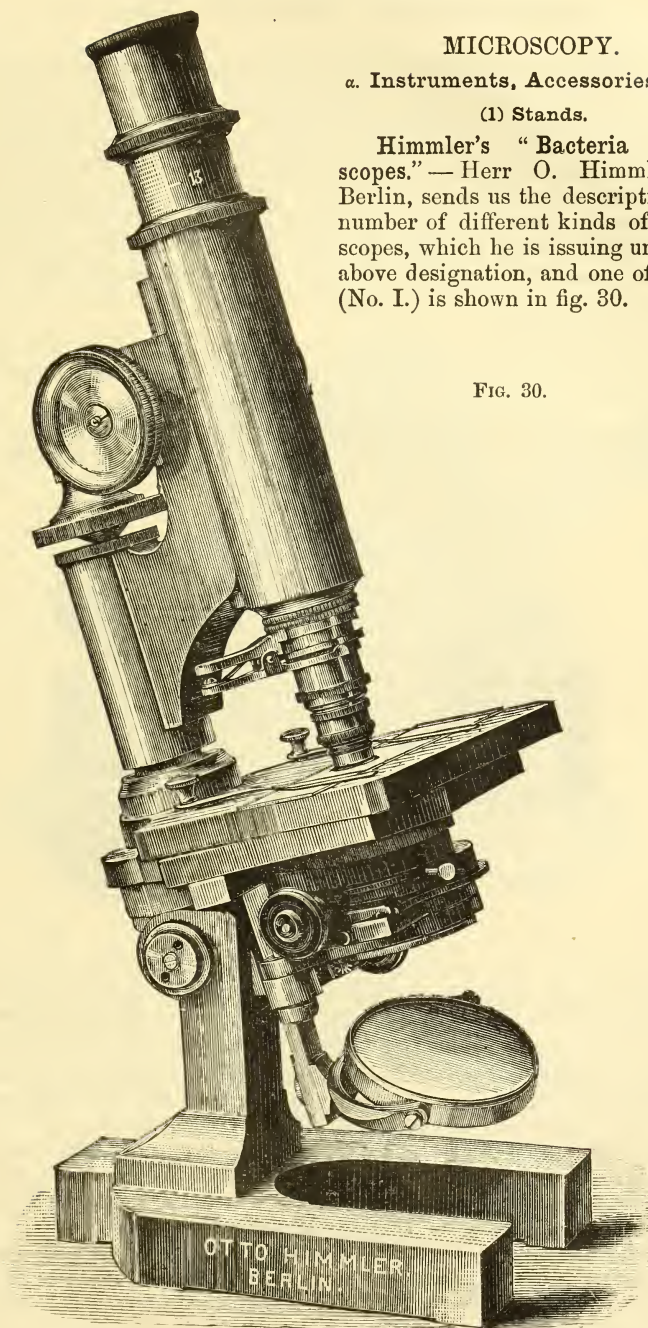
## MICROSCOPY.

## α. Instruments, Accessories, &amp;c.\*

## (1) Stands.

Himmler's "**Bacteria Microscopes.**" — Herr O. Himmler, of Berlin, sends us the description of a number of different kinds of Microscopes, which he is issuing under the above designation, and one of which (No. I.) is shown in fig. 30.

FIG. 30.



\* This subdivision contains (1) Stands; (2) Eye-pieces and Objectives; (3) Illuminating and other Apparatus; (4) Photomicrography; (5) Microscopical Optics and Manipulation; (6) Miscellaneous.

Generally, the instrument, as will be seen, is on the Hartnack model, but with an Abbe condenser. The condenser is on an arm with rack and pinion, so that it can be instantaneously moved in and out of the axis, by racking it down and turning it to the left. The maker claims that "this is a great advantage as against the instruments of other makers which have not such a contrivance." The claim as made is too wide, as we have seen German instruments which have had a similar arrangement in principle, though it might be more generally applied, as it is often very inconvenient to be obliged to alter the position of the Microscope, and slide out the illuminating apparatus when it is desired to work without the condenser.

For rapidly changing objectives, the instrument, as shown in fig. 30, is supplied with a Fuess clamp, the objective being released by pressing the end of the "tongs" together against the spring.

**Blackhall's Simple Microscope with Multiple Illuminator.** — In this little instrument (figs. 31 and 32) sent us by Mr. W. Blackhall, an

FIG. 31.

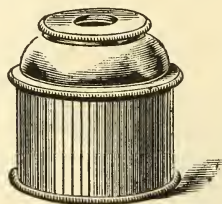
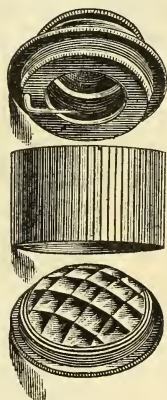


FIG. 32.



ingenious device has been made use of for illuminating the object, which is fixed on a pin in front of the simple lens. The bottom of the tube, in place of being closed by a convex lens, has a "multiplying glass," as shown in fig. 32, by the facets of which the light is thrown on the object.

**Heyde's Microscopes for Theodolites.\***—Herr G. Heyde has designed an instrument intended to unite the advantages of the screw Microscope with the convenience of the small Hensoldt scale Microscope. It has not generally been found possible to apply the screw Microscope to small theodolites, on account of the inconvenience for transport, &c., and yet their accurately divided scales deserve a better method of reading than either the Vernier or Hensoldt Microscope.

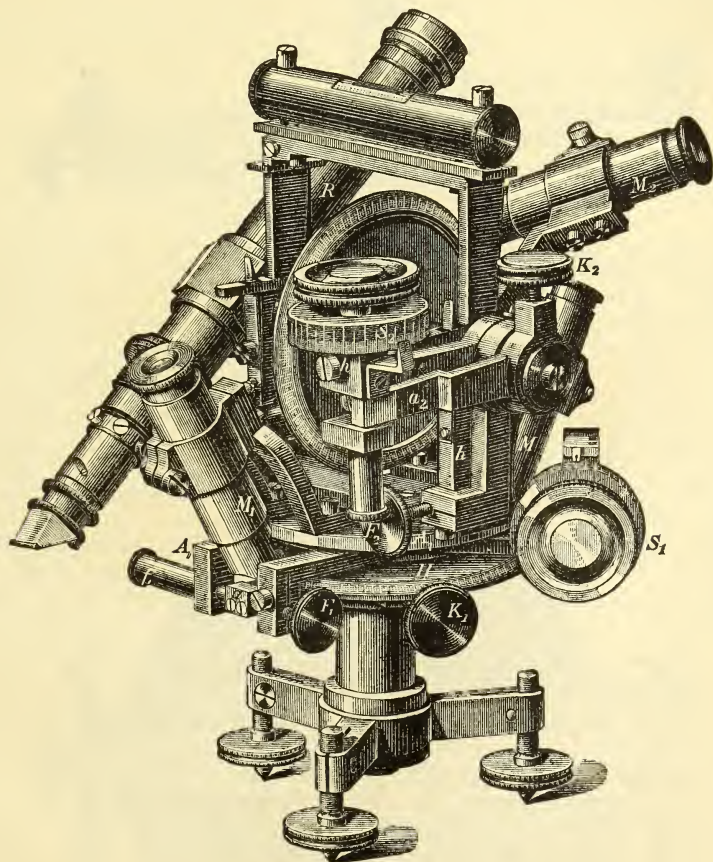
In the new theodolite M,  $M_1$  (fig. 33) are Microscopes with parallel wires for reading the horizontal scale H; they are attached to the arm  $A_1$ , which carries the supports of the telescope-axis. Below  $A_1$  is a second

\* Zeitschr. f. Instrumentenk., viii. (1888) pp. 171-6 (3 figs.).



arm  $A''$ , provided with two fine-adjustments; one at  $F_1$  with a spring  $f_1$  and clamping-screw  $K_1$  serves for the azimuthal adjustment of the telescope  $R_1$ ; the other  $S_1$  serves to turn  $A_1$  with respect to  $A''$ , and plays the part of the micrometer-screw in the screw Microscope. The Microscope  $M_2$ , which is used for reading the vertical scale, is connected in the same way with a micrometer screw  $S_2$ , which, after the telescope is adjusted to the right elevation, serves to measure the interval between the cross-wire and the image of the nearest division.

FIG. 33.



In an improved form of the instrument, the Microscopes  $M, M_1$  are attached to the arm  $A''$ , which is moved by the micrometer-screw, while the telescope supports are attached to the arm  $A_1$  which is adjusted by the fine-adjustment, so that in using the micrometer-screw it is only necessary to turn the Microscope-carrier through the small angle to be measured, while the telescope remains unmoved. The construction is



FIG. 34.

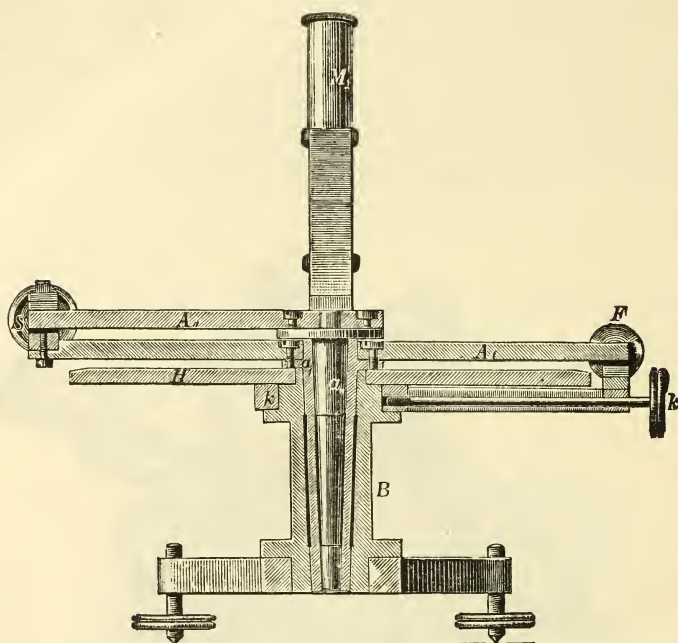
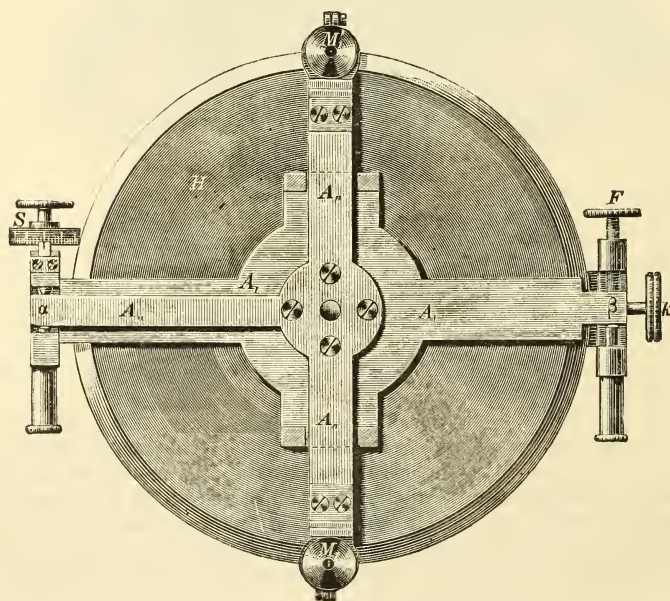


FIG. 35.



shown in figs. 34 and 35. B is the supporting column, and A, the principal axis, which contains a second axis  $A''$ , carrying the second arm  $A''$ , to which the two Microscopes  $M_1$  and  $M_1^1$  are screwed. The principal arm A, connected with the axis  $a$  and carrying the telescope, has two prolongations,  $\alpha$  and  $\beta$ , which extend beyond the divided circle.  $\beta$  is connected with the fine-adjustment screw F on the clamp  $k$ ; and  $\alpha$  carries the micrometer screw S. S serves to turn the Microscope arm  $A''$  alone; while F moves the principal arm with the upper part of the instrument, and at the same time the Microscope arm. A second arm is not necessary on the vertical circle, where it is replaced by the telescope carrier. The circles being divided into one-third of a degree, a complete turn of the drum is made to correspond to 20 minutes, and it is divided into 200 parts, so as to read to tenths of a minute.

## (2) Eye-pieces and Objectives.

**Binocular Eye-pieces.**—The late Mr. R. B. Tolles's binocular eye-piece has not yet been described in this country, and as inquiries are constantly made on the subject, the following description is reproduced with slight modifications from his original paper.\*

"To apply the binocular principle to the eye-piece of a Microscope or telescope, it is only necessary to make use of the erecting form of eye-piece, and to place the dividing prism at the point where the pencils composing the whole bundle of rays proceeding from the object cross the eye-piece, which is the point where, in any erecting eye-piece, the diaphragm proper is correctly placed.

If the theory of the erecting eye-piece of common form were generally understood, no demonstration that binocularity can be given to such an eye-piece would be necessary. Suffice it to say that, since any one pencil of light proceeding from any point of the object through the *whole area* of the object-glass *does* at this point equally fill the whole area of the diaphragm (that being of proper aperture) substantially in the same manner, therefore the division for binocular vision, if made here by the appropriate prism, must be a very nearly equal division of every particular pencil, and give a similar and satisfactory image of the entire field in each eye-tube. This is a sufficient expression of the whole theory of the binocular eye-piece.

It is, however, important in order to avoid pseudoscopic effects, to adopt the proper form of dividing prism; and this form is precisely that best suited to that kind of binocular Microscope in which the dividing prism is placed immediately above the objective. The natural presumption has been—contrary to this—that prisms of rectangular form would give the proper effects in the eye-piece, because of the pseudoscopic effects produced by their use in the Microscope of binocular body. But this is an error, inasmuch as the pencils proceeding to form the second image in the erecting eye-piece reach the small dividing prism under conditions suitable for correct vision of the object *were the eye placed there*, and, accordingly, the same false appearances obtain with the eye-piece of rectangular prisms, *having oculars above*, as if such division were made immediately above the objective; the effect being,

\* Silliman's American Journal of Science, xxxix. (1865) pp. 212-5 (1 fig.).

that the order in which the rays proceeding from the sides of the object or image viewed reach the eyes, as to right and left, is reversed from that which exists in natural vision; the left eye receiving a preponderating portion from the right side, and the right from the left side of the object.

It is to be noted, however, that the eye-piece with rectangular prisms, arranged after the first method of Prof. Riddell, does not uniformly produce conversion of relief, or that inversion of perspective which obtained in that first experimental arrangement for a binocular Microscope. Such a *binocular eye-piece* used in the Microscope upon transparent objects only occasionally gives the view in depth thus inverted. With low powers, and considerable thickness of the transparent object, the view is usually pseudoscopic. With medium and high powers, it is otherwise; and the effect is much controlled in this respect by the direction of the light upon the object.

When the binocular eye-piece with rectangular prisms is used in the telescope to view a landscape, the perspective is not *throughout* inverted, but portions of the field appear interposed between the eye and nearer objects in a singular and somewhat startling manner.

By arranging the compound rectangular prism so that the optical pencil is divided in the *plane of vision*, instead of vertically, the pseudoscopic effect is almost entirely obviated.

In constructing the binocular eye-piece, the prisms and arrangement of Nachet have been found to answer every condition and requisite of binocular vision. The dividing prism being placed, as before stated, at the point of crossing of the pencils in the erecting eye-piece, each pencil of light will enter the small dividing prism and impinge upon its reflecting surfaces in a manner similar to that illustrated in the Nachet binocular Microscope. The binocular eye-piece has greatly the advantage over the other arrangement. For when the prisms are placed *in the binocular body immediately above the objective*, their position, in order to secure a *proper division* of each transmitted pencil, should change with every change of objective used—which can be easily provided for in the case of low powers, but is rather impracticable with the higher numbers, it being very difficult to bring the prisms sufficiently near to the posterior combination of the objective. On the contrary, when the binocular arrangement is embodied *in the eye-piece*, the prism being once fixed in proper position, as before described, is correctly placed for every power of objective, and the eye-piece, thus binocular in form, is as applicable through the whole range of powers as if it were monocular. Applied to high powers, only one condition would be distinguishingly critical in the case of the eye-piece—that of the centricity of the central prism. The form of erecting *eye-piece* found most advantageous in this binocular adaptation is a duplication of the ordinary Huyghenian negative eye-piece, wherein the small dividing prism is very nearly at the eye-hole point of such a negative eye-piece as is ordinarily applied in the monocular Microscope. At a proper distance above this is placed another negative eye-piece, in which is formed a second image of the object viewed.

This form of erecting eye-piece gives less extension above the body of the Microscope than the positive form, and for that reason is preferred.



The annexed diagram (fig. 36) illustrates the division of pencils proceeding from the first image formed in the apparatus, and their general course to emergence at the two eye-surfaces. When the eye-piece is constructed of the form as here shown, the field is produced very satisfactorily, and of tolerable expansion; and does not necessitate more than 4.5 inches extension beyond the Microscope-body. The draw-tube can be as well withdrawn, and the eye-piece occupy its place, thus diminishing somewhat the total extent of the instrument. With proper modifications of the system of lenses placed before the prisms in the eye-piece, the whole binocular arrangement can be brought still nearer the objective, and retain also all the characteristics of the binocular eye-piece as contradistinguished from the binocular Microscope known and in use.

FIG. 36.

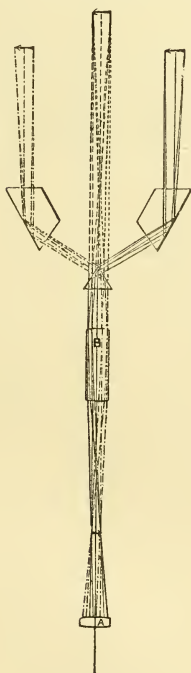
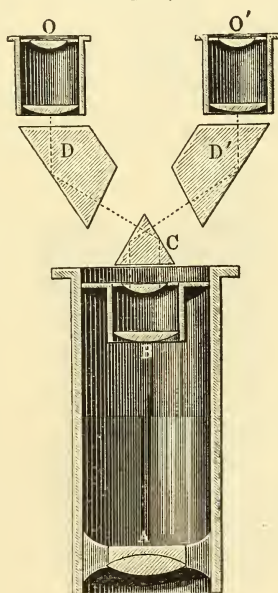


FIG. 37.



The objection that loss of light must occur on account of the additional front system of lenses pertaining to an erecting eye-piece (the lower system in the diagram), of course militates against the arrangement; but there are, on the other hand, incidental advantages in the use of the erecting form. . . . As the object is for the sake of efficiency with a high-power objective, to give as large an area to the transmitted pencil as possible at the point where it undergoes division in the small prism, therefore the power of the front system should be kept down, and amplification, as far as necessary in the eye-piece, be produced after the division has taken place. . . .



Having obtained by this means a pencil (or beam) transmitted through the eye-piece of the greatest possible dimension or area, at the point of binocular division, greater amplification in the eye-piece, as to its total power, might be advantageously effected by means of lenticular immergent and emergent surfaces of the upper prisms; the lower face of each prism to be *convex*, the upper emergent surfaces concave, giving achromatized refraction in each case. By this means a larger field, together with a minimum length of tubes above the prisms, would be secured.

By thus appropriating every surface of all the prisms not a reflecting surface, for the purpose of lenticular refraction, the greatest aggregate advantage appears to be secured."

We should mention that fig. 36 is not the diagram given with Mr. Tolles' original paper, but is one supplied by him shortly before his death, and drawn to scale, showing the path of the rays. In sending it, he remarked that without A the arrangement is a Nachet Binocular Microscope.

Fig. 37 shows one of the earlier forms of the instrument as combined with the eye-pieces, and is reproduced from Dr. Dippel's 'Handbuch der Allgemeinen Mikroskopie,' vol. ii. p. 598. Dr. Dippel points out that the eye-piece "either entails a very considerable lengthening of the body-tube or, if this inconvenience be avoided, considerably disturbs

FIG. 38.

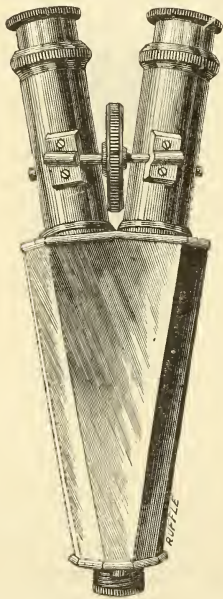
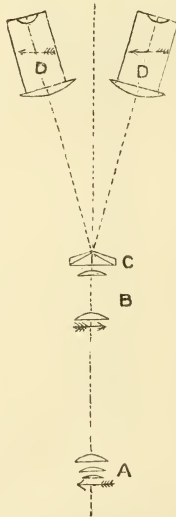


FIG. 39.



the optical effect of most objectives, because in this case they are used for a far shorter imaged distance, and consequently with an essentially different course of rays than in ordinary use."

*Prazmowski's* (figs. 38 and 39) is made entirely of brass, and being

of considerable size is inordinately heavy. A is the objective which forms an image near B, which is composed of two lenses, of which one is achromatic. This arrangement is essentially like a terrestrial or erecting eye-piece for a telescope, except that in the terrestrial eye-piece, the crossing-point (Ramsden circle) is *between* the component lenses, whilst in this binocular the crossing-point is situated higher up, in fact, just at the angle-edge of the achromatic prism C, where the pencil is divided. This arrangement gives a good field in each eye-piece. After division, the pencils pass on to the two eye-pieces D D, where they form images which are slightly unsymmetrical, by which the stereoscopic effect is obtained.

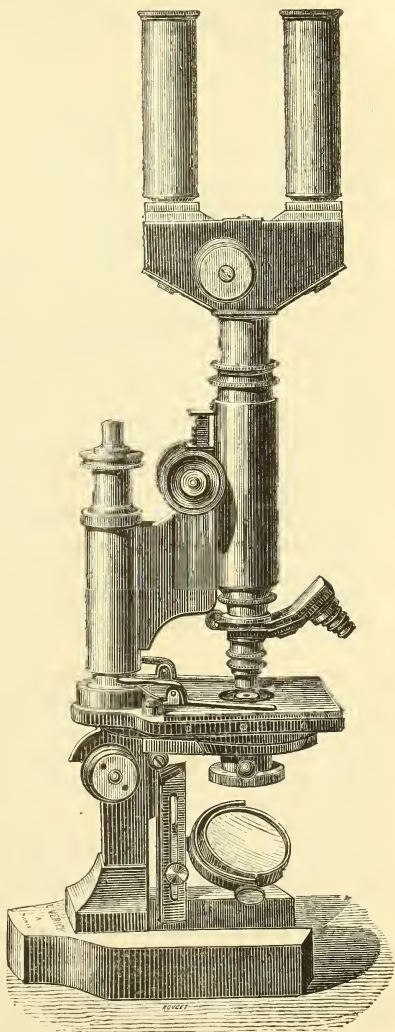
The diagram fig. 39 is from a drawing made for this Journal by the late A. Prazmowski.

*Vérick's* (fig. 40) is, so far as its essential optical arrangement is concerned, identical with that of Tolles, *having a central equilateral prism and two truncated ones at the sides*. The erector is, however, an achromatic combination formed of a lower plano-convex lens and an upper biconvex one. The central equilateral prism is also mounted with its lower face in a brass ring, having a circular diaphragm about  $\frac{1}{4}$  in. in diameter immediately above the upper lens of the erector. To secure its exact orientation in relation to the truncated lateral prisms, the brass ring is made to rotate partially in the horizontal plane; a portion of the cylindrical edge of the ring being provided with a "worm" on which acts an endless screw that can be turned by a small key whilst the observer views the image.

Fig. 41 shows the mechanism by which the lateral prisms (with the eye-pieces) can be separated to suit the width of the observer's eyes.

The sliding ebonite box-fittings in which they are mounted are attached respectively to the diagonal racked bars; the revolution of the toothed pinion (acted upon externally by the milled head shown by a dotted line

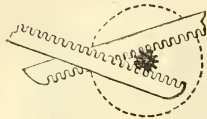
FIG. 40.



in fig. 41 and seen also in fig. 40), causing them to move exactly together outwards, or in the reverse direction.

A great advantage of the apparatus is that instead of being made of brass, and therefore very heavy, the eye-piece tubes are of aluminium, and the prism-box of ebonite; an admirably light eye-piece is thus obtained, which in that respect leaves nothing to be desired.

FIG. 41.



*Hartnack* also made some binocular eye-pieces, but we have not been able to obtain the materials for their description. Dr. Dippel gives \* the following description of two of them, but his woodcut of the first, which he describes as having four Riddell prisms, is obviously not correct, as it figures an eye-piece like that of *Vérick*, which will not take the Riddell prisms.

"In the older *Hartnack* binocular, which is inserted into the Microscope-tube by means of an adapter, the duplication of the image and halving of the pencil of rays takes place in the course of the pencil between the objective and the position of the real objective image. Riddell's arrangement of four prisms serves for this purpose, the two eye-pieces being rigidly connected with the two prisms, which direct upward the twice totally reflected rays parallel to the axis of the Microscope. The adjustment of the eye-pieces to suit the eyes of the observer is effected by mechanism put in motion by a screw-head. More recently, Dr. *Hartnack* has constructed a somewhat more complex binocular eye-piece, which gives splendid images with a small field of view, and, as far as I could ascertain, agrees in principle with the *Tolles* apparatus, inasmuch as a prism, over a lens-system in the lower tube acting as eye-piece, divides the image into two erect images, which are observed through two ordinary eye-pieces converging below and movable by rack and pinion in the direction of their long axes."

### (3) Illuminating and other Apparatus.

**Screw Eye-piece Micrometers.**—Much discussion has taken place in recent years on the subject of the relative accuracy of the different eye-piece micrometers, that is, between the fixed glass-plate micrometers on the one side, and the movable screw or spider's-web (filar) micrometers on the other.

In all these discussions the only screw-micrometer that has been referred to is the ordinary English form, with two spider lines, in which the optical part of the eye-piece is fixed immovably in the optic axis, while one of the spider lines traverses the field of view by the action of the screw. There are, however, some refinements of this apparatus to which attention may be called.

An important defect of the ordinary form of screw-micrometer arises from the fact that the measurements are not effected with the centre of the eye-piece alone, but use is made of the excentric parts of the lenses. The result of this is that the image of the object is subjected to more or less distortion, as its various parts are magnified differently, according

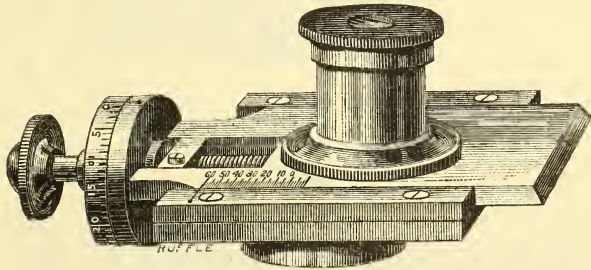
\* L. c., pp. 598-9.



to their lateral distance from the optic axis. Harting found that the image of ten divisions of a glass micrometer gave a result  $1/33$  less when it was measured as a whole than when the single divisions were measured and added together. The difference in magnifying power of the different parts of the field of view can of course be determined, but the process would be very tedious, and is not practically available.

Dr. H. v. Mohl\* therefore adopted the plan of moving the optical part of the eye-piece (with crossed threads) across the field. The image

FIG. 42.



is thus always observed only through the axis of the lenses, and the distortion found in the case of the ordinary micrometer is avoided. As will be seen in fig. 42, the tube containing the eye-lens and field-lens is attached to a slide which is moved by the screw so that the axis of the lenses may be displaced laterally in relation to the optic axis of the Microscope, or in other words, the eye and field lenses can be made to traverse the field of view. †

This micrometer, though it obviated the distortion caused by observation through the excentric parts of the eye-piece, gave rise to a some-

\* Arch. f. Mikr. Anat., i. (1865) pp. 79-100.

† Fig. 42 does not represent Mohl's micrometer as described by him (of which, indeed, we believe no figure is extant), but is taken from one sent us by Messrs. Merz, the makers of his original form. One important difference is that in the latter the eye-piece is not attached directly to the slide moved by the screw, but to a second upper slide which can be moved on the first by hand. The object of this is to adjust the lenses in the optic axis at the commencement of an observation without having to use the screw for that purpose. (We should be glad to be referred to a drawing or photograph of Mohl's original form if it exists.)

The following is a condensed abstract of Dr. Mohl's original description:—

"As regards the mechanical details of the instrument constructed for Mohl by Steinheil, the Microscope-tube is screwed into a horizontal plate fixed on a solid standard, and carries a Fraunhofer screw-micrometer which works in agate bearings; above the micrometer is an orthoscopic Kellner eye-piece with a short tube. The eye-piece tube is not fixed directly to the micrometer-slide which is moved by the screw, but to a second slide which moves between swallow-tail guides upon the upper surface of the first in a direction parallel to the length of the micrometer-screw; this slide itself is moved by a second screw of deep pitch. The stage and condenser are separated from the body of the Microscope, being carried by a bar which can be fixed to the stand of the instrument by means of two short arms. The eye-piece therefore is movable not only by the micrometer-screw together with the slide which is used for measuring purposes, but also when desired by the second slide (or "eye-piece slide") which moves horizontally upon the first, so that



what similar error of optical excentricity, the image observed by the eye-piece when not in the optic axis, being formed not by the central rays from the objective, but by the marginal rays. To obviate this

FIG. 43.

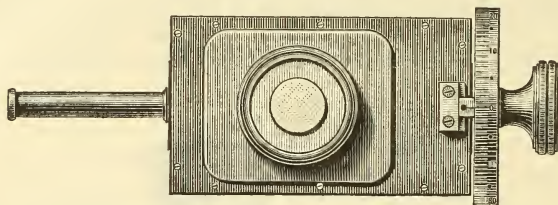
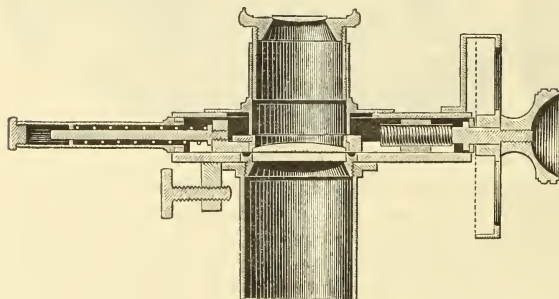


FIG. 44.



Prof. Abbe (see figs. 43 and 44) while making use of Mohl's device for moving the eye-piece across the field of view, added a second lens close

it can be made to traverse the whole of the fixed microscopic image. It is important to have some means of readily bringing the eye-piece back to the axis of the Microscope, and of noting the positions of the micrometer and eye-piece slides which correspond to this adjustment. For this purpose three diaphragms were employed, each perforated with a minute hole; one is placed over the objective, one in the middle of the body-tube, and the third immediately under the eye-piece, so that they only transmit rays along the axis of the tube; by this contrivance the eye-piece can be adjusted in the axis by bringing it into the position in which the cross-wires exactly divide the small circular openings of the diaphragm into four equal quadrants; the position is then noted by marking an index line upon the eye-piece slide and one of its guides, and reading the micrometer-screw, so that it may at any time be at once recovered without a fresh adjustment. If it is required to use another part of the screw for measuring purposes it is only necessary to adjust the eye-piece to the axis, note the position of the cross-wires upon an object, and then after moving the screw to the desired point, to bring back the cross-wires to the same position by means of the eye-piece slide. In attempting to make a preliminary series of measurements Mohl found that in spite of the solid stand employed in his instrument and the massive plate to which the tube and micrometer were fixed, the pressure of the hand upon the micrometer-screw produced such considerable deflections in the instrument that it was impossible to make accurate adjustments, and it was only by fixing the tube to the stand by means of a rectangular framework of brass plates that it could be made sufficiently rigid."

beneath the field lens which remains immovable in the axis, while the field and eye-lens move over it. The lower plane surface of the field lens has diagonal cross lines, as well as a double index-mark engraved on it, and the plane upper surface of the additional lens has a scale. The lens has a focal length of about 17 mm., so that its lower focal point lies approximately in the opening of the objective, and the principal rays are made parallel in front of the eye-piece as if their centre of divergence was at infinite distance. In consequence of this, in every position of the eye-piece, the point of the field under observation behaves as the centre of the field of view in the ordinary arrangement, that is, *all* the pencils are identical with the axial pencil, and the shifting of the eye-piece produces no optical excentricity.\*

**Winkel's Combination of Screw-micrometer and Glass-micrometer Eye-piece.**†—Dr. A. Koch writes as follows:—For fine microscopic measurements, in particular for the determination of the thickness of Bacteria, it appeared to me to be of advantage to possess an apparatus with which exact determinations could be made more easily than with the ordinary eye-piece micrometer. I found useful for this purpose an eye-piece with a thread such as has been in use for a long time in physical and astronomical instruments, and occasionally employed in Microscopes. In these eye-pieces a stretched thread or a mark on a glass plate can be moved parallel to itself by means of a micrometer-screw with divided head; for measuring, the thread is brought successively to both edges of the object, and its breadth is given by the number of turns of the micrometer-screw necessary to move the thread from one margin of the object to the other. The value of the divisions of the drum of the micrometer-screw is determined by an object-micrometer.

It is, however, inconvenient, especially with very strong magnifications and very small objects lying in great numbers in the field of view (such as Bacteria) to have to replace such a screw eye-piece by an ordinary micrometer eye-piece when it is desired to measure with less exactness the larger divisions of the object which we had previously been measuring with the screw eye-piece, e. g. the length of a *Bacterium*. Herr R. Winkel, of Göttingen, has, however, constructed a micrometer eye-piece in which the thread is replaced by a division on a glass-micrometer. This apparatus can therefore be used, as I have already mentioned in my work, 'Ueber Morphologie und Entwicklungsgeschichte einiger endosporer Bacterienformen,'‡ either as an ordinary micrometer eye-piece with fixed micrometer for less fine measurements, or for more exact determinations by using the micrometer-screw and successively adjusting one edge of a division on the margins of the object.

The mechanical details of the apparatus are shown in figs. 45 and 46, the latter fig. showing the internal arrangement after the upper part at A (fig. 45) has been unscrewed. In fig. 46 is seen the frame DE, which is moved by the micrometer-screw EF, which has a pitch of exactly  $\frac{10 \text{ mm.}}{100}$ ; the

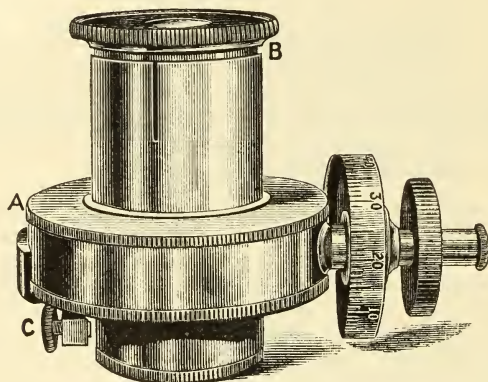
\* Cf. Dippel's 'Handbuch der Allgemeinen Mikroskopie,' 2nd ed., 1882, pp. 639-40 (2 figs.).

† Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 33-5 (2 figs.).

‡ Bot. Ztg., 1888.

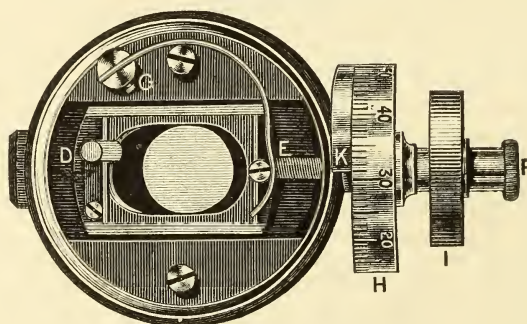
latter is held firm by the clamp at D, and on removing the upper part can be taken out for cleaning. Backlash of the micrometer-screw is completely avoided by the spring fastened at G, which extends to E.

FIG. 45.



On the micrometer-screw is the drum H divided into 100 parts, and the head I for clamping; the sharp end of the piece K screwed to the socket of the eye-piece serves as index. The upper lens of the eye-piece can be drawn out (B, fig. 45) for the exact focussing of the micrometer-

FIG. 46.



divisions. The whole is fixed to the body-tube by the screw C, though a slight shaking of the body-tube, owing to the movement of the micrometer-screw, does not injuriously affect the exactness of the measurements.

#### (5) Microscopical Optics and Manipulation.

**On the use of Fluorite for Optical Purposes.\***—Prof. E. Abbe has an article on this subject. Of the minerals which occur in nature, quartz and calc-spar are the only two which have been regularly used in

\* Zeitschr. f. Instrumentenk., x. (1890) pp. 1-6.



practical optics. The great transparency of these minerals for violet and ultra-violet light has recommended them for spectroscopic purposes, but their chief use depends on the specific property of non-tesseral crystals of double refraction, by which results are effected which are not to be attained with an amorphous substance, such as glass. Tesseral crystalline minerals which, so far as their optical properties are concerned, are like glass, have already been made use of for optical purposes. Brewster and Pritchard, within the last forty years, recommended and employed the diamond and other precious stones of unusually high refractive power in Microscope lenses. Attempts in this direction, however, have brought no lasting gain to optics, and must at present be considered as wholly given up, the simple Microscope formed of uncorrected lenses being relegated to a subordinate use. Quite different points of view are now kept under consideration in judging of optical resources for the continued improvement of the compound Microscope. For, in face of the refinements which, in recent times, practical optics has kept in view, the estimation of the materials used in lens-combinations has altered in direction; it no longer looks at the greater or less perfection of its fundamental effect, which depends of course on the refractive power, but it has turned to the consideration of the degree in which the properties of these materials facilitate and advance the neutralization of the unavoidable subsidiary effects—spherical and chromatic aberration.

From this point of view a material which, from the standpoint of the efforts of Brewster and Pritchard, would appear very unprofitable, viz. fluor-spar, becomes of special interest at the present time for practical optics. This is because it offers unusual advantages in respect to the neutralization of those subsidiary effects. Fluorite possesses an abnormally low refractive power; the index for sodium light is only 1.4338, and is thus considerably lower than that of crown glass; its use as a constituent of a lens-system is therefore, in respect to the fundamental effect, relatively disadvantageous. However, with many lens-combinations, such as those used for the Microscope, there must be a difference of refractive indices between media in contact and with equal curvature of the bounding surfaces in contact to remove the spherical aberration; it is on the amount of this difference that the compensating effect in respect to the spherical aberration depends. The lower the index for the first medium, the greater the amount of this difference, and the more perfect the compensating effect which is to be attained by the addition of a second medium of given refractive power. So, also, the lower the index for the first medium, the lower that of the second, when a certain given difference is to be maintained. If, for example, in a cemented double lens—as used in the Microscope—an ordinary crown glass of index  $n_D = 1.52$  serves as the one member, and the removal of spherical aberration requires a difference of refractive powers of 0.20 on both sides of the cemented faces, then the above consideration shows that there must be connected with that crown glass a second lens with index 1.72, and consequently one made of a very heavy, strongly dispersive, flint glass. Supposing, on the other hand, the first member to be a lens of fluorite, then the required excess of refractive power of the second member would be given by an ordinary flint glass of 1.63, which for



many reasons would be much more advantageous. That mineral therefore affords greater convenience in the choice of the kinds of glass to be used in obtaining perfect compensating effects for the removal of the spherical aberration in lens-systems.

Besides this advantage, which gains special importance in the construction of Microscope objectives of large aperture, fluorite possesses the further useful optical properties of an abnormally low colour-dispersion, and a relation to the partial dispersions for the different parts of the spectrum which is very serviceable for the removal of the secondary spectrum. For the three hydrogen lines  $H_\alpha$ ,  $H_\beta$ ,  $H_\gamma$  the differences of the refractive index are—

Material.	$N_\beta - N_\alpha$	$N_\gamma - N_\beta$	$N_D$	$\frac{\Delta n}{n-1}$	$\frac{N_\gamma - N_\beta}{N_\beta - N_\alpha}$
Fluorite .. .. .	0·00455	0·00255	1·4338	$\frac{1}{95\cdot4}$	0·561
Ordinary calc-silicate crown glass .. .. .	0·00860	0·00487	1·5179	$\frac{1}{60\cdot2}$	0·566
Aluminium-phosphate crown glass .. .. .	0·00737	0·00407	1·5159	$\frac{1}{70\cdot0}$	0·552
Borate flint glass .. .. .	0·01026	0·00582	1·5521	$\frac{1}{53\cdot8}$	0·567

If the interval from  $H_\alpha$  to  $H_\beta$  (C to F) be taken as a measure of the mean dispersion ( $\Delta n$ ), the above table shows that the fluorite, not only taken absolutely, but also relatively to the value of  $(n-1)$ , possesses a considerably lower colour-dispersion than the most advantageous glass hitherto produced; for while with the latter the so-called relative dispersion does not sink below  $1/70$ , with fluor-spar it is diminished to  $1/95$ . But the curvature which a compound lens of this medium must have in order to give an achromatic system of determined focal length when combined with a lens of greater relative dispersion, depends essentially on the amount of the relative dispersion of a medium. The smaller the  $\Delta n/(n-1)$  the less curvature suffices for achromatism under otherwise similar circumstances for a given focal length.

While thus a simple non-achromatic lens of fluor-spar, on account of its low refractive power, necessitates a much greater curvature for a determined focal length than one of crown glass, on the other hand, an achromatic lens with this material requires less curvature than one made of crown glass, supposing that the same flint glass is used for the compensation of the colour-dispersion.

Finally, the numbers in the last column of the above table show that the ratio of the partial dispersions in the two parts of the spectrum  $H_\alpha$  to  $H_\beta$  and  $H_\beta$  to  $H_\gamma$  has for fluorite, in spite of its very low dispersion, almost the same value as for an ordinary silicate crown glass with dispersion  $1/60$ . On the other hand, for the aluminium phosphate-crown, the most advantageous glass so far as the relative dispersion is concerned, the blue end of the spectrum is seen to be relatively

shortened, although the value of the  $\Delta n / (n - 1)$  is only diminished to  $1/70$ . Consequently fluorite may be said to offer special advantages for the simultaneous union of three rays of the spectrum, i. e. for the removal of the secondary colour-dispersion.

The above-mentioned phosphate crown glass, it is true, in combination with the above or a similar borate flint glass, also allows a direct achromatism for three different colours (not the three rays  $H_\alpha$ ,  $H_\beta$ ,  $H_\gamma$ , yet three rays within the less refrangible part of the spectrum). It therefore serves for the construction of a double lens with only tertiary colours remaining; but in this combination the curvatures are somewhat disadvantageous. This is due to the fact that the numbers for the relative dispersion  $\Delta n / (n - 1)$  in these two media— $1/70$  and  $1/54$ —show only a slight difference. If, however, fluorite be substituted for the above crown glass, then a combination is obtained which satisfies the condition of the union of three different colours, and at the same time gives a very considerable difference of the relative dispersions of the two constituents ( $1/95$  and  $1/54$ ). This difference still remains sufficiently large if the calc-silicate crown be substituted for the borate flint. The dispersion of this glass is, moreover, almost rigidly proportional to that of the fluorite through the whole visible dispersion. Accordingly, with these two media, a double achromatic lens of almost absolutely complete colour-union could be made; for there would be no tertiary spectrum remaining over. Having regard then to all the conditions which regulate the construction of a perfect lens-combination—the spherical aberration in systems of large aperture, as well as the chromatic aberration of first and second orders—fluor-spar affords more profitable relations than any material at present at our disposal in optics. The data on which the present conclusions are based, were made known long ago by the spectroscopic measurements on fluor-spar which Stefan published in the year 1871. The numbers given above are from the measurements of Dr. Riedel, of Jena, made in the year 1880 and later at the author's instigation, with the use of hydrogen lines, on different varieties of the mineral. They agree with the values found by Stefan within the limits of errors of measurement, so far as they concern the same parts of the spectrum. The characteristic optical properties of fluor-spar shown by these spectroscopic measurements are doubtless due to the specific effect of the fluorine which makes up fifty-six per cent. of the calcium fluoride. It might therefore be reasonably expected that if it were possible to introduce this element in considerable quantity into artificial fusions, kinds of glass would be obtained which, partially at least, would exhibit the valuable peculiarities of fluor-spar.

Experiments made in this direction by Dr. Schott in 1881 and the following year in the course of his work on the improvement of optical glass have to a certain extent realized that idea. By the use of fluorides in small quantity glasses were produced which, with lower refractive index, exhibited also a very diminished dispersion. These experiments, however, showed clearly at the same time (as Dr. Schott has already indicated) the extraordinary technical difficulties which stand in the way of the production of sufficiently homogeneous glass of such a composition. These difficulties at first appeared to be so great that it seemed

to be impossible to prepare practically useful kinds of glass with similar properties to those of fluor-spar.

This result has served to fix the author's attention on the use of the natural mineral for purposes of practical optics, and more especially for Microscope objectives; for previous experiments in the year 1881 had already shown that fluorite, in spite of its less hardness, is susceptible of being shaped like glass, although with some difficulty.

By using clear crystals and cleavage fragments, such as were then easily obtainable from mineral dealers, in the year 1884 the optical factory of Carl Zeiss in Jena first constructed, under the author's direction, Microscope objectives of different kinds, in which perfect correction of the spherical and chromatic aberration was effected by the use of lenses—one to three in each system—of fluorite instead of crown glass. With the introduction of the new Microscope objective, the "Apochromatic," the mineral has come into regular use in Jena, and has been further extended by other opticians in their imitation of the Zeiss construction. The calculations and technical details of these constructions have been rendered much easier by the introduction of fluor-spar in partial replacement of crown glass. Without its aid those lens-systems, for the same requirements in the construction, would have been still more complicated in composition, and more difficult in manufacture than they are at present.

In view of this use of fluorite for the Microscope now generally admitted, and considering the advantages which it offers for many other purposes of practical optics, it will be of interest to discuss the determining condition of its use, viz. the possibility of procuring this material in sufficient quantity and quality.

The inquiries which the author set on foot many years ago have hitherto led to no satisfactory result. Fluor-spar belongs, it is true, to the widely distributed minerals, and is found in very many places in transparent crystals. Most varieties, however, apart from the rarity of large clear pieces, are quite worthless for optical purposes. This is due to the fact that they show double refraction in a marked degree and owing to disturbances of the regular crystal growth. Until some years ago, tolerably large pieces, which were water-clear and in parts quite pure, could be obtained from mineral dealers. These were attributed to different, though principally Swiss, localities, and it seemed reasonable to suppose that this serviceable variety, free from double refraction, would be of quite general occurrence and consequently not difficult to procure. More exact inquiries, however, soon proved that all such specimens of fluorite, met with amongst dealers or in collections, are referable to one and the same locality in the Schwarzhornstock in the Bernese Oberland, and in fact, to a single find accidentally made there almost sixty years ago.

According to the communications of Herr E. v. Fellenberg, of Bern, and to information which the author obtained later at the place itself, a hole, out of which was obtained considerably more than 100 cwt. of large water-clear crystals and cleavage pieces of fluor-spar, was discovered above the Alp Oltscheren by Alpine shepherds from Brienzwyl, near Brienz, in the year 1832. This material was distributed amongst dealers in minerals in all directions, and after



dealers, collectors, and museums had been supplied, was sold by the owners to chemists for the preparation of hydrofluoric acid, or thrown away as worthless. A part is said to have come to Paris fifty years ago, and to have been used by opticians in lenses and prisms for experiments on heat radiation. The remnants presumably of this remarkable find, which included some water-clear crystals (cubes) as large as one's head, hidden away in cellars, &c., were purchased by the author in the preceding year from the grandchildren of the original finders, and were thus saved for optical purposes.

The precise locality of that old find had been forgotten. By means of the labels, however, found in the Bern Museum, Herr v. Fellenberg, who has assisted the author in these inquiries in the most friendly way, was enabled to fix it as the south-west slope of the Oltschihorn, the offshoot of the Schwarzhornstock towards the Lake of Brienz.

Chance investigations made with the help of some Oberland crystal-seekers proved the frequent occurrence of fluor-spar in the neighbourhood, but the old locality was not discovered, nor was further material with the characteristics of the earlier find obtained. The firm of Carl Zeiss therefore took up the quest, and during the summer of this and the preceding year caused regular excavations to be made by a large number of practised workmen under the direction of an agent. By this means, in July 1888, on a steep, almost inaccessible rock about 1900 metres above sea-level, the hole was discovered out of which came the find of 1832. It was found, however, to be practically exhausted. Further investigation of the mountain which—belonging to the upper Jura—is distinguished by massive schist formations with numerous precipices, fissures, and cavities, was then made. In this way semi-transparent calc-spar and fluor-spar, crystallized in large cubes, but so far as purity was concerned in no way comparable with that found in the old locality, were discovered in several places near that spot. Of several hundredweight collected, only some pounds were clear and suitable for optical purposes. In August of the present year the work was therefore discontinued, after all traces found by blasting had been followed up as far as they gave any indications of better results. It therefore appears beyond all doubt that the single locality which formerly afforded fluor-spar in large clear masses is now completely exhausted.

The employment of the mineral for Microscope lenses is hardly affected by this; for the comparatively small quantity required for this purpose is assured by the general occurrence of less perfect material, from which, with some difficulty, it can be picked out. On the other hand, the further extension of its use in optics will be dependent in every way on the discovery of new localities which afford large crystals or cleavage-masses of similar purity to that which was formerly found at Oltschihorn.

Perhaps this communication may help to make this mineral, so valuable to optics, an object of greater attention, and possibly to bring to light localities of it which have hitherto remained unnoticed.

J. M. M. writes,\* in reply to a correspondent "Prismatique," "Fluorite" is simply the Continental name for common fluor-spar, and,

\* Engl. Mech., li. (1896) pp. 205-6.



doubtless, if he will visit any of the Derbyshire spar or lead mines—for the mineral is a constant companion of lead veins—he will find crystals of it quite fit for optical work in overwhelming quantities. Perfectly colourless crystals are certainly not very common, neither are they very rare; but, at the same time, are they necessary? The most common colour the mineral assumes is a pale green, evident enough in large crystals; but in laminae thin as the lenses of an objective, scarcely, if at all, perceptible.

‘Prismatique’s’ experience of the deterioration of the new glasses from atmospheric influences is valuable. It is, at the same time, just what a chemist would expect from the composition of some of them, that is, if they are honestly named. It must not be forgotten that more than half a century ago our own Faraday in England, and Amici, in Italy, produced new glasses for optical work which possessed valuable properties, and offered great advantages, optically, over those in common use. Ross, in London, and Chevalier, in Paris, worked Microscope objectives from the new glass, and their performance was said to be a great advance upon that of lenses made from the ordinary material. It was, however, found that they deteriorated so rapidly that their manufacture was given up.

I have now in my possession a 1/10 in. made by Chevalier. The outer lenses of the combination, viewed by reflected light, are a bright steel-blue in colour, much like the screw-heads in a watch movement. The performance must have been phenomenal at the date of its production, for it will even now “dot” *angulatum*; but the field is filled with fog produced by the action of the decomposed surfaces upon the light, analogous in effect, but less in degree, to that produced by a very finely ground but unpolished lens. With this experience behind us, precisely equivalent to that of ‘Prismatique’ with the new glasses, it behoves one to pause before rushing to the conclusion that the optical millennium is here.”

Mr. Lewis Wright writes\* on the same subject in reply to some strictures by ‘Prismatique’ on the Jena glass and German opticians. “Apart from fluorite altogether, great improvement has been made by English and other opticians with the new glass alone; and German micro-objectives are now reaching this country superior to any made here at double the price. I speak from personal trials, of which I may perhaps say a few words another time. Zeiss undoubtedly used at first glass which would not bear exposure to the air; but these things were gradually discovered and remedied, though it is too soon yet to say if even present lenses will stand permanently. I believe I was, myself, the very first to utter a word of caution in these columns on that very point, though a glass may be useful in the middle of a triplet which will not stand atmospheric exposure.”

Jena Glass.†—Mr. A. Caplatzi thinks it will be of interest to give the list published in 1888, which brings the variety of glasses up to 63.

The first column contains the number, the second the factory number, the third the description, the fourth the refractive index for D, the fifth the medium dispersion C to F, and the sixth the specific gravity. At a

\* Engl. Mech., li. (1890) p. 222.

† T. c., p. 117.

little enhanced price pressed discs of the same glass having the approximate form of the desired lens can be obtained.

No.	Fabric No.	Description.	Refractive Index D.	Medium Dispersion, C-F.	Specific Gravity.
45	0.599	Boron silicate crown .. ..	1.5069	0.00813	2.48
46	0.337	Silicate crown .. ..	1.5144	0.00847	2.60
47	0.374	" " .. ..	1.5109	0.00844	2.48
48	0.546	Flint crown .. ..	1.5170	0.00859	2.59
49	0.567	Silicate crown .. ..	1.5134	0.00859	2.51
50	0.610	Crown of low dispersion ..	1.5063	0.00858	2.51
51	0.598	Silicate crown .. ..	1.5152	0.00879	2.59
52	0.512	" " .. ..	1.5195	0.00886	2.64
53	0.463	Baryta light flint .. ..	1.5646	0.01020	3.11
54	0.608	Crown of high dispersion ..	1.5149	0.00942	2.60
55	0.602	Baryta light flint .. ..	1.5676	0.01072	3.12
56	0.381	Crown of high dispersion ..	1.5262	0.01026	2.70
57	0.583	Baryta light flint .. ..	1.5688	0.01110	3.16
58	0.543	" " .. ..	1.5637	0.01115	3.11
59	0.527	" " .. ..	1.5718	0.01133	3.19
60	0.575	" " .. ..	1.5682	0.01151	3.15
61	0.522	" " .. ..	1.5554	0.01153	3.03
62	0.578	" " .. ..	1.5825	0.01255	3.29
63	0.376	Ordinary light flint .. ..	1.5660	0.01319	3.12
64	0.340	" " .. ..	1.5774	0.01396	3.21
65	0.569	" " .. ..	1.5738	0.01383	3.22
66	0.318	" " .. ..	1.6031	0.01575	3.48
67	0.266	" " .. ..	1.6287	0.01775	3.72
68	0.335	Dense silicate flint .. ..	1.6372	0.01831	3.77

**Lehmann's Molecular Physics.\***—The following review of Dr. Lehmann's treatise is taken from a recent number of 'Nature,'† where it appeared under the title of "The Application of the Microscope to Physical and Chemical Investigations":—

Very soon after the first invention of the Microscope, attempts were made to apply the new instrument to solve some of the remarkable problems of crystallogenesi. The early volumes of the Royal Society Transactions contain in the papers of Boyle, Hooke, and Leeuwenhoek, published between the years 1663 and 1709, many records of attempts of this kind; and the works of Henry Baker, which appeared between 1744 and 1764, are also largely concerned with the study of the process of crystallization under the Microscope.

In Germany, Ledermuller in 1764, and Gerhardt in 1780, showed the value of the Microscope in studying the internal structure of crystals; while in France a long succession of enthusiastic investigators, Daubenton, Dolomieu, Fleurian de Bellevue, Cordier, and others, were busily engaged in laying the foundations of the science of microscopical petrography.

Early in the present century, we find the English investigators once

\* 'Molekularphysik, mit besonderer Berücksichtigung mikroskopischer Untersuchungen und Anleitung zu Solchen, sowie einem Anhang über mikrochemische Analyse.' Von Dr. O. Lehmann, Professor der Electrotechnik am kgl. Polytechnikum zu Dresden. Leipzig (W. Engelmann), 1888-9, 2 vols., pp. 852 and 697 (624 figs. and 10 pls.).

† Nature, xlii. (1890) pp. 1-2.

more taking a leading part in applying the Microscope to the study of crystallized bodies. Between the years 1806 and 1862, Brewster published a long series of memoirs, dealing with the microscopical characters of natural and artificial crystals, and the inclusions which they contain. About the year 1850, too, Mr. Sorby commenced his important investigations on the subject, availing himself of the method of preparing transparent sections of rocks and minerals which had been, shortly before this time, devised by William Nicol. Mr. Sorby's epoch-making memoir, "On the Microscopical Structure of Crystals, indicating the Structure of Minerals and Rocks," made its appearance in 1858.

While one group of investigators, following the lines of the early work of Brewster and Sorby, have sought to make the Microscope an efficient instrument for the determination of minerals, even when present in rocks as the minutest crystals or fragments; others have no less diligently pursued the methods which the same pioneers in this branch of research have initiated for solving physical and chemical problems connected with the formation of crystallized bodies.

In the hands of Des Cloizeaux, Tschermak, Zirkel, Von Lasaulx, Fouqué and Michel-Lévy, Rosenbusch, and other workers, the Microscope has gradually been developed into a splendid instrument of mineralogical research; and the determination of the minutest particles of a mineral is now becoming no less easy and certain than that of the largest hand-specimens.

But, at the same time, Brewster and Sorby's early attempts to solve physical and chemical problems by the aid of the Microscope have not failed to exercise an important influence on subsequent workers in these branches of science. Link, Frankenheim, Klocke, Harting, and especially Vogelsang (whose early death was a severe loss to this branch of science), have done much towards establishing the science of crystallogenesi upon a firm basis of accurate observation; and their labours have been continued in more recent times by H. Behrens and Dr. Otto Lehmann, the author of the work before us.

As the well-known treatises of Rosenbusch, and of Fouqué, Michel-Lévy, and Lacroix give us an admirable *résumé* of the present state of determinative mineralogy, as improved by the application of the Microscope, so does the work before us contain a perfect summary of the contributions of the microscopist to the sciences of physics and chemistry.

It will only be possible, within the limits of an article like the present, to indicate briefly the plan of the very comprehensive and, indeed, almost exhaustive work, in which Dr. Lehmann has embodied the observations of himself and his predecessors in this field of inquiry.

The first division of the book deals with the construction and use of the Microscope; especial attention being given to forms of the instrument, like those devised by Nachet and by the author of this work, for the special purpose of studying crystallization and other physical and chemical processes.

The second division of the book treats of those physical properties of matter which are presented by all bodies, whether in the solid, liquid, or gaseous state. Such questions as the polarization and absorption of light, the conduction of heat, and the electric and magnetic relations of various substances are here dealt with by the author.

The next division relates to the peculiar properties presented by solids. Elasticity and plasticity are considered, and, under the latter head, the remarkable phenomenon of the production of twinned structures in crystals by mechanical means is fully discussed. Under the head of cleavage we find a treatment of such phenomena as the production of mathematical figures in certain crystals by pressure, percussion, &c.; while under the heads of "Enantiotropie" and "Monotropie" are classified the consequences which follow from heteromorphism among crystalline substances, and the tendency of the heteromorphous forms to pass one into the other.

The division dealing with liquids and their peculiar properties contains discussions on fluidity, surface-tension, diffusion, capillarity, and crystal-growth, with the origin of structural anomalies. The problems of solution and precipitation, with those of solidification and fusion, are also treated of in this part of the treatise.

The second volume of the work commences with the discussion of the properties of gases and their relations to solids and liquids. This division of the subject, which is very exhaustively treated, extends to 335 pages.

The work concludes with critical remarks upon different molecular theories. The chapters dealing with the theories of crystal structure, of allotropy, of heteromorphism, and of isomerism, with several others, in the same division of the book, are full of interest and suggestiveness.

A supplement of about 150 pages is devoted to what the author calls "crystal-analysis," or what is generally known to geologists and mineralogists as "microchemical analysis." Very minute particles of an unknown substance may often be determined by being treated with appropriate reagents and studied under the Microscope; in this way they are made to yield crystals of various compounds which can be recognized by their characteristic forms and habit. An admirable summary is given by the author of the work of Bôričky, Streng, Behrens, Haushofer, and others, who have gradually perfected this branch of research, and made the method one which is of the very greatest service to the students of microscopical mineralogy and petrography.

While the physicist and chemist will find in this work a perfect mine of interesting and ingenious experiments (many of which are suited to class-demonstrations by projection methods), the mineralogist and geologist will hail the appearance of the book as one that completes and supplements the well-known treatise of Vogelsang—a work that has exercised the most important influence on the development of petrological theory.

In conclusion, it may be pointed out that, not only are the numerous observations of the author on crystallogenesi that are described in memoirs in 'Groth's Zeitschrift' included in the work before us, but many others that have never before been published find a place in these volumes. The work is very fully illustrated both with woodcuts and coloured plates, and constitutes a complete synopsis of all that is known on a number of questions of great importance and interest to workers in many different branches of science.



## B. Technique.\*

## (1) Collecting Objects, including Culture Processes.

FOUREUR, A.—*Étude sur la culture des Microorganismes anaérobies.* (Study on the culture of anaerobic micro-organisms.)

Paris (Doin), 1889, 8vo, 73 pp. and 25 figs.

JEFFRIES, J. A.—A new method of making Anaerobic Cultures.

*Med. News*, 1889, p. 274.

## (2) Preparing Objects.

**Study of the Embryology of the Earthworm.**†—Mr. E. B. Wilson says:—"After testing many different hardening fluids, I have found none to compare with Perenyi's fluid, which gives uniformly the best results, both for sections and for surface-views of all stages, and is far superior to picro-sulphuric acid or corrosive sublimate. Flemming's mixture of osmic, chromic, and acetic acids gives very clear differentiation of the middle stratum of the germ-bands after staining with hæmatoxylin, but in most respects it is far inferior to Perenyi's fluid. The embryos were left in the fluid from 15 to 60 minutes, placed in 70 per cent. alcohol for a day, and kept permanently in 90 per cent. alcohol.

For permanent staining no method has proved so satisfactory as borax-carmin followed by hæmatoxylin. After being deeply stained in the carmine (12 hours), and extracted in acid alcohol in the usual manner, the embryos were treated with extremely dilute ammoniacal alcohol for a few minutes, to neutralize the free acid, and were then stained in very dilute Kleinenberg's hæmatoxylin (12 hours or more). In case of overstaining with hæmatoxylin, the colour may be again extracted with acid alcohol, after which the specimens are again treated with ammoniacal alcohol. This process, following treatment with Perenyi's fluid, gives beautifully clear preparations, which are specially favourable on account of the clearness with which the cell-outlines are shown. It has been found desirable to imbed the specimens for sectioning as soon as possible after hardening, and to reduce the time of immersion in melted paraffin to a minimum (i. e. not more than 10 or 15 minutes).

For surface-views of the germ-bands the borax-carmin stain should be very deep, and the hæmatoxylin very slight, so as to give the specimen only a purplish colour, not a dark-blue. The germ-bands are dissected off on the slide, in strong glycerin. This method has, in my experience, given far better results than that of osmic acid followed by Merkel's fluid, so successfully used by Whitman in the study of *Clepsine*.

For the study of entire specimens of the young stages I have found Perenyi's fluid, followed by alcohol, water, very dilute iodine solution, and glycerin, to give results superior beyond comparison to those attained by any other method. The iodine colours the protoplasm pale yellowish-brown, the cell-outlines are clearly marked, and the nuclei are stained deep brown. In time, most of the iodine is precipitated in the form of deep-brown spheres, which mar the clearness of the preparations, but

\* This subdivision contains (1) Collecting Objects, including Culture Processes; (2) Preparing Objects; (3) Cutting, including Imbedding and Microtomes; (4) Staining and Injecting; (5) Mounting, including slides, preservative fluids, &c.; (6) Miscellaneous.

† *Journal of Morphology*, iii. (1889) pp. 445-6.

such specimens may be afterwards stained with carmine, &c., sectioned, and mounted in balsam in the usual manner, and give perfect satisfaction, even after a stay of two years or more in the glycerin."

**Experimental Imitation of Protoplasm.\***—Prof. O. Bütschli has communicated to Prof. Ray Lankester a full account of the methods by which he attempts to imitate protoplasm.† A medium-sized watch-glass or flat dish must be filled with a thin layer of common olive oil and be placed on a water-bath or small cupboard at a temperature of about 50° C. The great point is to select the right moment at which the oil attains the proper degree of thickness and viscosity; this moment can, however, only be found by systematic trials. After three or four days a trial may be made. Should the drop not have become finely vesiculate and exhibit little or no streaming, the heating process must be continued and a trial made on the succeeding day. If the oil becomes too thick it will form frothy drops, and in such cases a small quantity of ordinary olive oil must be mixed with it.

The vesiculate drops are prepared thus:—In a small agate mortar a small quantity of dry carbonate of potash is ground to a fine powder. This must be breathed on till the salt becomes slightly moist, and then a drop of oil must be added; the two constituents should be mixed till they form a thickish paste. A few drops of it, about the size of a pin's head or smaller, are placed on a cover-glass, the corners of which are supported by small pegs of soft paraffin. Prof. Bütschli then places on a slide a drop of water, and puts the cover-glass over it in such a manner that the drops of paste are immersed in the water, but are not much compressed. The preparation is then placed in a damp chamber, and remains there about twenty-four hours, when the drops have a milk-white and opaque appearance. The preparation is then well washed out with water, which is supplied at one edge by a capillary tube and drawn out by blotting-paper at another.

If the drops have turned out well they will begin almost at once to move about rapidly and change their shape continuously. The water under the cover-glass must now be displaced by glycerin diluted with an equal bulk of water, when a vigorous streaming movement will be exhibited. The amœboid movements are generally more distinct if the drops are somewhat compressed. If the drops do not stream they can generally be made to do so by tapping the cover-glass slightly, by applying gentle pressure, or sometimes by breaking up the drops. It is especially interesting to see how fast and beautifully the drops creep to and fro in the water or in half-diluted glycerin, even when they are not compressed. The streaming movement, on the other hand, is better seen if the drops are somewhat compressed; this may be done by inserting under the cover-glass a piece of broken cover-glass of medium thickness, and then removing the paraffin pegs. This streaming movement is best demonstrated twenty-four hours after the addition of the glycerin, as the drops will then be thoroughly cleared and transparent. The movement and streaming are much more marked and distinct if the drops are examined on a stage warmed to 50° C.

\* Quart. Journ., Micr. Sci., xxxi. (1890) pp. 99-103.

† See this Journal, 1889, p. 731.

Prof. Bütschli adds:—"You must not doubt the correctness of the phenomena which I have described if the first trials do not give the desired results."

The student may also be referred to the account given by M. Yves Delage \* of his experiences in Prof. Bütschli's laboratory at Heidelberg.

**Method of Examining Network of Muscle-fibres.**†—Mr. C. F. Marshall adopted a modification of Mays' method of demonstrating nerve-endings in muscle. Mays used a mixture of 20 parts arsenic acid (1/2 per cent.), 4 parts gold chloride (4 per cent.), and 1 part osmic acid (2 per cent.), but this, while preserving the nerve-endings, disintegrates the muscle-fibre by the action of the arsenic acid. Mr. Marshall, after several experiments, used 20 parts acetic acid (1 per cent.), 4 parts gold chloride (1 per cent.), and 1 part osmic acid (1 per cent.). The muscle-fibre was placed in this solution for fifteen minutes after previous immersion in acetic acid (1 per cent.) for a few seconds; then in acetic acid (1 per cent.) again in a warm chamber for one or two hours.

**Mounting Spermatozoa of Salmonidæ.**—Mr. F. M. Walford, at the meeting on April 16th, said:—"Having occasion lately to examine the spermatozoa of the English brook trout (*Salmo fario*) and the American trout (*S. fontinalis*), I found it would be advantageous to have permanent mounts at my disposal. Mr. E. M. Nelson has suggested that the communication to the Royal Microscopical Society of a brief note descriptive of the method adopted might be of assistance to students of this branch of science.

The collection of the milt containing the spermatozoa flowing from a spawning fish presents no difficulties when a medium is used which will preserve the spermatozoa without coagulating them. Alcohol or acetic acid, even when dilute, coagulate the milt, and should be avoided. One part glycerin to five of water is a fairly good medium, but the aqueous solutions of phenol or corrosive sublimate of about 2½ per cent. are preferable.

The majority of text-books recommend glycerin and water for mounting spermatozoa, and hence this was one of the first media tried, but the resolution, even with 1/12 oil-immersion, was most unsatisfactory. The result of a number of experiments in staining may be summed up in the statement that the effect of staining is to make the heads more prominent and the filaments less visible. Specimens collected in 2½ per cent. and mounted in 1¼ per cent. solution of corrosive sublimate looked fairly well for a time, but after a few months the heads of the spermatozoa gradually dilated and showed signs of disintegration.

A suggestion was made that, as probably a medium of low refractive index was desirable, it might be practicable to mount the spermatozoa dry on the cover-glass. So far I have not succeeded in doing so, but future experiments in this direction may be productive of better results. I was told by a friend that at one of the hospitals Farrant's medium was used for human spermatozoa, and the idea occurred to me that, as working in Farrant often produced where not desired a plentiful crop of air-bubbles, it might be possible to take advantage of this peculiarity and show the

\* Arch. Zool. Expér. et Gén., v. (1889) pp. xliii.-xlvi.

† Quart. Journ. Micr. Sci., xxxi. (1890) pp. 73-4.



spermatozoa in the air-bubbles on the surface of the Farrant. The *modus operandi* is as follows:—A drop of Farrant is placed on the slip. A small quantity of the spermatozoa in  $1\frac{1}{4}$  per cent. corrosive sublimate is dropped from a pipette on the Farrant. The cover-glass is lowered horizontally on to the spermatozoa, and if there are no air-bubbles visible to the naked eye, the cover-glass is lifted and again allowed to fall flat on the spermatozoa. The superfluous fluid is drawn from the edge of the cover-glass with a piece of blotting-paper. The mount is placed in a drying cabinet for some hours until the Farrant is set quite hard, and is then secured by two coats of Hollis."

**Methods for making Permanent Preparations of Blood.\*—Dr. U. Rossi** communicates two methods by means of which he obtains permanent preparations of blood. (1) In a glass vessel is prepared a strongish and recently filtered solution of methyl-green. Another vessel is filled with one-third distilled water, one-third osmic acid (1 per cent.), and one-third of the foregoing solution. The mixture should be quite clear, and of an emerald-green colour. One drop of this mixture, which is at the same time fixative and staining, is placed on a slide. Then a glass rod just smeared with the staining solution is dipped in the heart's blood of a recently killed animal, and this drop of blood mixed with the drop of the methyl-green solution on the slide. The preparation, protected from dust, is left in a moist atmosphere for about half an hour. At the end of this time the preparation is treated with a minute drop of acetic acid, all the various ingredients being carefully mixed together with the quill-point which has carried the acetic acid. The preparation is then covered over, and glycerin in very small drops placed along the edge of the cover-glass, under which it slowly runs.

(2) Blood obtained directly from the heart of some small mammal is allowed to fall into a watch-glass containing osmic acid of  $1-1\frac{1}{2}$  per cent. The mixture having been well shaken up, is poured into a little tube and left for 24 hours. At the expiration of this time the blood is deposited at the bottom, and the osmic acid is then siphoned off or removed by means of a piece of cotton thread, one end of which dips into the fluid, but so as not to touch the blood, and the other into an empty tube. When the acid has been removed the blood is washed two or three times with distilled water, this being removed in the same way as the acid. The blood is then stained with alum-carmine to which has been added acetic acid in the proportion of 1 per cent. by volume of the carmine solution. The blood is then washed again, and next treated first with rectified spirit and afterwards with absolute alcohol. A drop of this blood is removed with a pipette to a slide, and when the spirit has evaporated is treated with carbol-xylol and then mounted in dammar.

**Effect of Galvanic Current and other Irritants on Protista.†—Dr. M. Verworn**, in studying the effect of galvanism upon certain

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 475-7.

† Pfleger's Archiv f. d. Ges. Physiol., xlv. (1889) pp. 1-36 (2 pls. and 6 figs.); xlv. (1889) pp. 267-383 (3 pls. and 5 figs.). 'Psycho-physiologische Protisten-Studien,' Jena, 1889, 8vo, 220 pp. and 6 pls. Cf. Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 496-50 (3 figs.).



Protista, employed the following apparatus. The fluid containing the organisms was placed in a rectangular cell, the long sides of which, composed of porous clay, were from 1-5 mm. thick and 20 mm. long, the shorter ends being composed of a cement made of a mixture of wax and resin. To the clay sides or electrodes were applied the brush electrodes. These were short glass tubes, closed at one end with clay and filled with a saturated solution of zinc sulphate. From the plug projects outward the brush, while at the other end projects inwards a zinc rod connected with the wires. Sometimes the extremities of the electrodes were made of porous clay and cemented down to the slide, so that their points were immersed in the fluid. The current was produced from a chromic acid battery of twelve elements, the cells of which were 17 cm. high and 11 cm. broad.

In this way the author found that various species responded differently to the two kinds of stimulus, some being affected by the positive current and others by the negative.

By the use of the porous points instead of brushes it was found that the galvanotropic effect was not confined to a small area near the electrodes, but was actively efficient even in vessels of 10 cm. contents over the whole mass of water. Hence the action tended to collect the organisms into aggregations. For example, in the same water *Flagellata* would accumulate about the anode, the *Ciliata* about the kathode.

When the movements of Protista are to be studied to ascertain the influence of light, it is important to remove all sources of disturbance. If a large drop of water should be used, the Microscope must be quite horizontal, oblique light must be cut off by surrounding the slide with black paper, and the warming power of the transmitted light obviated by the interposition of a layer of ice between the mirror and the slide. The Protista may then be examined either by placing them under the Microscope with all the just-mentioned precautions, and with the addition of first covering the mirror with black paper. The paper is then suddenly withdrawn, and the movements observed. Or a drop of water containing the organisms is placed on a cover-glass coated on the other side with black paper, in which a window 3 mm. square has been cut. The effect of coloured light can be observed by interposing solutions such as ammonia, copper, and bichromate of potash.

The effect of warmth can be studied in a similar way, that is, by means of first covering the mirror and observing the movements through the window of the cover-glass. It is of course necessary to first ascertain the degree of heat by previously focussing the light on a thermometer.

The effect of mechanical irritation was ascertained by shaking the slide either once or frequently. The continued vibration was attained by fixing one end of the slide and moving the other end up, and then allowing it to drop by means of a toothed wheel of four cm. diameter, and with the teeth 1 cm. apart.

In addition to the foregoing, the effects of local, acoustic and chemical irritants were also examined.

The behaviour of small pieces as compared with uninjured organisms was also observed. The pieces were obtained by crushing or cutting with a knife made by sharpening a needle into a blade.

**Effect of Hardening Reagents on Nerve-cells.\***—Dr. E. Sehrwald calculates that the large cells of the central nervous system become shrivelled to the extent of 21–26 per cent., owing to the effect of the hardening fluids necessary for producing Golgi's staining. The shrivelling is accompanied by warping, a result induced by the fibres and processes from the cells being incrustated with metallic salts. From the warpings and curves produced in the fibres, the author makes his calculation as to the diminution in size of the cells.

**Staining and permanent Preservation of Histological Elements, isolated by means of caustic potash or nitric acid.†**—Mr. S. H. Gage and Mrs. S. P. Gage point out the methods of checking completely the action of KHO and HNO<sup>3</sup> at will, so that the isolated elements may be permanently preserved in alcohol or glycerin, and also stained in the usual way.

30 to 50 per cent. solutions of caustic potash act with great rapidity on intercellular substance, and quite slowly on cellular elements, while weak solutions rapidly dissolve all the elements. The action of the strong solution may be checked at any time by means of a 60 per cent. solution of potassium acetate, or by the addition of sufficient glacial acetic acid to neutralize the caustic potash and form acetate of potash. Either fresh or hardened tissue may be used. The pieces should not exceed half a cubic centimetre in size, and fifteen to twenty times as much potash solution should be used as tissue. As soon as the elements separate readily the caustic potash solution should be poured off and replaced by a copious supply of a 60 per cent. solution of acetate of potash, to which one per cent. glacial acetic acid has been added. The isolated elements may be mounted in acetate of potash, in glycerin, or in glycerin-jelly. If the elements are to be stained, they must be soaked for twenty-four hours or more in a saturated aqueous solution of alum. They are then stained with hæmatoxylin, or alum-carmine.

Nitric acid is used in 20 per cent. solution, and the time required varies with the temperature. At the ordinary temperature, one to three days are required. If heat be used, the action may be completed in a few minutes. The action of the acid is suspended by immersion in water until the acid is quite removed. The fibres are teased out in water or in glycerin tinged with picric acid, and then mounted in glycerin-jelly. If the nuclei are to be stained, the Koch tubercle stain diluted 4–5 times answers well. The preparations are then mounted in balsam.

**Disintegration of Woody Tissues.‡**—Prof. G. L. Goodale recommends the following method of disintegrating woody tissues for microscopic observation. The tissue is soaked for a sufficient length of time in a ten per cent. solution of potassium bichromate, then quickly freed from the excess of the salt, by once rinsing in pure water, and immediately acted on by concentrated sulphuric acid. After the acid has acted for a short time, the tissue is to be placed in a large quantity of water, when it will be found to have undergone more or less complete disinte-

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 461–70.

† Proc. Amer. Soc. Micr., 1889, pp. 34–45.

‡ Amer. Journ. Sci., xxxix. (1890) p. 79.

gration, each structural element being separated from its neighbours, with little or no corrosion of the wall.

**Cleaning Diatoms.\***—Mr. Edward S. Nott recommends that—

(1) The material be completely disintegrated by continued boiling in a solution of sal soda.

(2) The disintegrated material should be sifted in a sieve made of bolt-cloth, removing all the fine earths and broken forms.

(3) The remainder may have the greater part of the sand removed from it by revolving it in an evaporating dish with water.

(4) The material, now mostly diatoms, should be boiled in acids: first, in muriatic, then wash; second, in nitric, then wash, and sometimes boil also in sulphuric acid.

(5) After washing all traces of acid away, boil once more in a solution of sal soda, wash, and sift in a fine sieve of bolt-cloth.

The object is to remove all the debris and waste material before using the acids, as the result will be better and the expenditure of time and labour less. The stock should be kept in alcohol, and in mounting, the best distilled water should be used.

#### (4) Staining and Injecting.

**Methylen-blue Staining for Nerve-endings.†**—The examination of nerve-endings by staining with methylen-blue, a method invented by Ehrlich, has received, since its introduction, considerable attention at the hands of physiologists, owing to the comparative simplicity of the procedure, and the satisfactoriness of the results—results quite equal to those obtained by the silver nitrate and gold methods. The method as recommended by Prof. S. Mayer, consists of two distinct parts, the first of these being the treatment with the blue pigment, the second that of its fixation by means of picrate of ammonia. The methylen-blue solution is made by dissolving 1 gram of the pigment in 300–400 ccm. of a half per cent. salt solution. The picro-glycerin solution is composed of a cold saturated solution of picrate of ammonia, diluted with an equal volume of pure glycerin.

The animals are injected through a blood-vessel with the blue solution, or pieces of fresh tissue are soaked in the solution, or the animal may be immersed alive in the fluid without danger to life.

Small pieces of the object are then immersed in the picro-glycerin and are at once ready for examination. If found suitable for a permanent preparation the cover-glass can be fixed down with a mass composed of equal parts of wax and resin.

If found desirable the injected or impregnated preparations may be kept for some time in the picro-glycerin.

The effect of the second reagent is to alter the colour of the stained parts, all shades of red, brown, black being seen in the axis-cylinders and the non-medullated terminal nerve-expansions. This disadvantage is compensated by the stain being fixed and the preparation cleared up at the same time, advantages not counteracted by any considerable changes in the tissues.

\* Proc. Amer. Soc. Microscopists, xi. p. 149. Amer. Mon. Micr. Journ., xi. (1890) p. 31.

† Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 422–38.



The simplicity and rapidity of the method is seen when pieces of tissue are to be examined. Small pieces as fresh as possible are placed for about ten minutes in the methylen-blue solution, they are then well washed in a half per cent. salt solution and then examined at once in the picro-glycerin, the time required for all the manipulation being about 30 minutes. The results as to the positive and negative pictures attained by this procedure are equivalent to those produced by the action of silver nitrate on fresh tissues. Hence this method has all the advantages without any of the disadvantages of the silver method.

**Technique of Golgi's Staining Method.\***—The many recent modifications of Golgi's method of staining nervous tissue have tended, says Dr. E. Sehrwald, either towards improving the excellence of the picture or towards rendering the preparation permanent. But in effect these modifications practically destroy the picture, the finer details, visible enough in the silver solution, being lost during the manipulations required by the various modifications.

The author proposes a method which leaves intact all the details of the original silver chromate deposit and allows the preparation to be soaked in warm paraffin, so that sections of any required thinness may be prepared. This method simply consists in saturating all the reagents employed in Golgi's method with bichromate of silver. The only precaution required is that the reagents should be saturated with the salt at a high temperature.

In this discursive disquisition no details are given, the author, after minutely describing a long series of failures, contenting himself with a piece of general advice and stating that if this method be adopted preparations will be obtained which, if they have any fault, are too full of detail.

**Method for Restaining old Preparations.†**—Mr. J. W. Gatehouse gives the following method by which it is possible to stain objects that after mounting in balsam have become so transparent as to be scarcely visible. Take filtered oil of turpentine and saturate it with picric acid, adding the acid gradually till a fine yellow colour has been obtained, and scales of the acid remain undissolved. To this solution add carefully crystals of resublimed iodine, taking care to add only a few at a time, as otherwise the chemical action set up may possibly produce sufficient heat to ignite the turpentine and cause even a slight explosion. With all due care even, a series of small decrepitations may be noticed as the iodine dissolves. Sufficient iodine should be added to change the colour of the solution from a light yellow to a distinct brown tint. Then place the slide in a dish containing turpentine, to which some of the stain has been added, and allow it to remain there until the balsam is softened and the stain has penetrated and done its work, when the turpentine can be replaced by more balsam. In this way the author has restained slides of embryonic tissues which had been mounted several years and which had become almost invisible except in special lights. After two days' soaking the whole of the structures were brought out splendidly, every detail being perfectly clear.

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 443-56.

† Journ. Microscopy and Nat. Sci., iii. (1890) pp. 113-4.



**Staining Elastic Fibres and the Corneous Layer of Skin.\***—Herr A. Köppen recommends the following method for staining elastic fibres and the corneous layer. The sections, freed from all foreign constituents, are to remain for 24 hours or longer in absolute alcohol; they are then placed in the following staining fluid:—Saturated alcoholic solution of crystal violet, 5; acid. carbol., 5; aq. destil., 100. In this solution, freshly made, the sections remain for 15–24 hours. They are then placed in iodine solution for two minutes (I, 1; KI, 2; H<sub>2</sub>O, 300), after this for five minutes in a 10 per cent. aqueous solution of common salt. They are then waved about for 15 seconds in 1 per cent. hydrochloric acid. Next they are decolorized in absolute alcohol. When sufficiently decolorized they are immersed first in turpentine and then in xylol, after which the sections are mounted in xylol balsam.

**Prevention of Surface Deposits in Golgi's Chrom-silver Method.†**—Pieces of nervous tissues which are treated by Golgi's method are frequently rendered useless, owing to the thick deposit which altogether prevents the details of the preparation from being examined. This inconvenience, says Dr. E. Sehrwald, may be avoided by enveloping the pieces in a substance which, while it penetrates into the cavities and adheres closely to the surface, yet allows the silver salt to permeate without hindrance. Such a substance is gelatin in 10 per cent. aqueous solution. This, when cold, forms a firm but plastic mass, and melts at a temperature below that of the body.

It is best manipulated by pouring it over the object placed in a box made by winding a strip of paper round a piece of cork. When cold the box may be immersed in the silver solution. A piece about a centimetre square is quite saturated in 24 hours in the cold.

Although fresh pieces may be imbedded in the gelatin before being fixed in Müller's fluid, it is much better to envelope with gelatin after the Müller.

When the silver reaction is complete the gelatin must be removed, at any rate if the object is to be imbedded in paraffin. This is done with warm water to which chrom-silver salt, as explained above (see technique of Golgi's method, p. 409), has been added to excess. The solubility of the gelatin is but little affected by the action of the silver salt or by light.

**Staining Paraffin Sections.‡**—Those who have used the paraffin imbedding method for serial sections have, doubtless, wished for some simplification of the process of staining. This may be done, according to Dr. Kükenenthal, by dissolving the colouring matter in absolute alcohol and dropping the solution into turpentine until the desired depth of colour is secured. Sections fixed to the slide with the collodion are kept in the oven until the clove oil has completely evaporated, the paraffin dissolved in turpentine as usual, and the slide brought into the dye. The staining is quickly effected. Over-staining may be corrected by placing the slide for a short time in a mixture of acid-free absolute alcohol and turpentine (equal parts). Turbidity of the colouring fluid may be corrected by adding a drop or two of alcohol; Meyer's carmine,

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 473–5.

† T. c., pp. 457–61.

‡ Amer. Mon. Micr. Journ., xi. (1890) p. 11.

methyl-green, methyl-blue, gentian-violet, safranin, Bismarck-brown, eosin, fuchsin, tropeolin, and malachite-green may be used in the above ways.

MAGALHAES, P. S. DE.—*Estudo geral das colorações em histologia.* (Use of staining-methods in histology.) Rio de Janeiro, 1889, 8vo, 89 pp.

FEIST, B.—*Ueber die vitale Methylenblaufärbung markhaltiger Nervenstämmchen.* (On methylen-blue staining of living medullated nerve-fibres.) Strassburg, 1889, 8vo.

KRYSINSKY, S.—*Beiträge zur histologischen Technik.* 5. Kupfercarmin. 6. Lithiumcarmin und Lithiumpikrincarmin. (Contributions to histological technique. 5. Copper-carmin. 6. Lithium-carmin and lithium-picrocarmin.) *Virchow's Arch.*, CXVII. (1889) pp. 204-6.

MARTINOTTI, G.—*Alcuni miglioramenti nella tecnica della reazione al nitrato d'argento nei centri nervosi.* (Some improvements in the technique of the silver nitrate reaction on the nervous centres.) *Atti del 12. Congr. della Assoc. Med. Ital.*, I., p. 179.

MONTI, —.—*Una nuova reazione degli elementi del sistema nervoso centrale.* (A new reaction of the elements of the central nervous system.) *Atti della R. Accad. dei Lincei—Rendic.*, V. (1889) p. 705.

WEIGERT, C.—*Neue Neurogliafärbung.* (New neuroglia-staining.) *Münchener Med. Wochenschr.*, XXXVI. (1889) No. 29.

#### (5) Mounting, including Slides, Preservative Fluids, &c.

**Mounting in Glycerin Jelly.\***—Glycerin jelly, says Mr. J. D. King, answers more purposes as a mounting medium than any other, but it is dreaded by many on account of the difficulty of getting rid of air-bubbles, a difficulty which may be avoided by the following method. Heat the jelly in a water-bath till the water boils, then, always working in a warm room, mount with it as you would with glycerin except dipping the cover in fluid, being careful to remove any stray air-bubbles under the dissecting glass before putting on the cover, for even very small ones cannot be depended on to disappear of their own accord.

Small or delicate objects can be arranged and kept in place by first covering the bottom of the cell with glycerin jelly and placing the objects in it, being careful to cover them well, and leaving them to harden. When hardened, apply additional jelly and put on the cover. After standing overnight in a cool place, if the jelly be of good quality it may be cleaned off under water with a small paint-brush and finished off with cement. It is the better way to use cells for glycerin jelly mounts, though it is not necessary to fill the surface or apply the cement before putting on the cover-glass, or even in all cases to have them as deep as the object is thick. A cell prevents the cover-glass from touching the slide at any point, and thus creating a liability of forming a vacuum by shrinkage, and it makes better work every way.

**New Mounting Medium.†**—Mr. H. Shimer has devised the following mounting medium, the use of which gives every satisfaction. It is a mixture of equal parts of glycerin jelly, Farrant's solution, and glycerin. The glycerin jelly is made as follows:—Gelatin, 30 parts; water, 70 parts; glycerin, 100 parts; carbolic acid, 2 parts.

Of this glycerin jelly, liquefied by the heat of a water-bath, pour 1 fluid oz. into a 4-oz. glass-stoppered bottle, add an equal volume of the Farrant's medium and of glycerin. A little gentle agitating in

\* Microscope, ix. (1889) p. 138.

† T. c., pp. 143-5.

the water-bath will soon insure a complete mixing. Into this bottle drop a small lump of camphor.

This medium needs a little warming (about 110° Fahr.) to make it fluid for use.

**Preserving Animals.\***—Dr. C. J. Cori, after trying various fixatives as reagents for rapidly narcotizing small invertebrate animals, such as hot sublimate, chloral hydrate, ethyl-alcohol, certain alkaloids, such as strychnia and cocain, found that ordinary wood-spirit or methyl-alcohol, since it has little action on albumen and possesses sufficiently satisfactory narcotic properties, gave the best results. The formula for the solution is:—Methyl-alcohol 96 per cent, 10 ccm.; water, 90 ccm.; sodium chloride, 0.6 grm. The addition of the salt prevents the too great maceration of the tissues.

For preserving and hardening the author found that chrom-osmium-acetic acid in the following proportions gave excellent results:—chromic acid 1 per cent., 25 vols.; acetic acid 2 per cent., 5 vols.; osmic acid 1 per cent., 1 vol.; water, 69 vols. The specimens are said not to become blackened, and stain quite well.

If objects contain lime salts, these neutralize the acids, an inconvenience which can be obviated by using large quantities of the solution and frequent renewals of the fluid. In the fluid the animals remain, according to size, from 2-48 hours; they are then washed in running water for 6-72 hours, then placed in 50 per cent. spirit, and finally in 70 per cent.

The osmic acid is dissolved in distilled water to which so much permanganate of potash has been added as gives it a faint rose colour. A little of the salt should be added to the solution from time to time, or when the colour is beginning to fade.

The osmic solution is best kept in yellow or black glass bottles with two grooves in the stopper, a device which allows large drops to be obtained without removal of the stopper.

**Agar as a Fixative for Microscopical Sections.†**—M. A. Gravis recommends agar as a medium for fixing sections to the slide. According to the author it possesses several conspicuous advantages. It is quite liquid at the ordinary temperature; the sections can be arranged on the slide with great ease. Air-bubbles never appear beneath the section. Vegetable cells, which often become distorted when imbedded in paraffin, resume their shape and original dimensions. When well dried this fixative is insoluble in all reagents, except in distilled water. The specimens may be mounted in either balsam or glycerin.

The fixative is prepared by soaking half a gram of agar in distilled water for some hours. It is then heated gently until it boils. It is then boiled for about 15 minutes so that the agar may be completely dissolved. When cold it is filtered through a fine cloth and preserved in stoppered bottles.

In order to make the fixative stick properly, the slides must be perfectly clean. It is best to boil the slides in water acidulated with hydrochloric acid, and then, having rinsed them in distilled water, dry them on a clean cloth. The fixative is put on the slide with a brush.

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 437-42.

† Journ. de Microgr., xiv. (1890) pp. 83-5.



Upon this layer the sections are arranged. Directly this is done the slide is gently heated over a Bunsen's burner in order to soften, but not to melt, the paraffin imbedding. Any excess of fixative may now be removed by merely draining it off. The fixative is now to be thoroughly dried, and as this requires several hours, the slides should be covered over with a bell-jar and left till next day. The paraffin is then dissolved out either with warm turpentine or with chloroform, and then the solvent extracted by running some strong spirit over the slide. If the preparations have been stained *en masse* the slide is then dehydrated in alcohol, cleared up with oil of cloves, and mounted in balsam. If the sections require to be stained, the slide is merely placed in the staining solution, and when withdrawn, rinsed with spirit, after which it is mounted in balsam. As indicated above, almost any reagent may be used, provided it be not purely aqueous.

**Use of Cajeput Oil for dissolving Canada Balsam.\***—Prof. O. Beccari recommends the use of cajeput oil, obtained from *Melaleuca Leucodendron*, instead of oil of cloves, for dissolving Canada balsam. It has the advantage of being soluble in dilute alcohol, and the object can therefore be transferred directly from the dilute alcohol to the oil, which is not the case with oil of cloves. In addition, objects placed in cajeput oil and alcohol take up methyl-green and retain it in Canada balsam.

**New Method of finishing Balsam Mounts.†**—Mr. F. N. Pease remarks:—"It is only a question of time, when balsam mounts thoroughly hardened and unprotected from atmospheric influence will be ruined, on account of the cover-glass becoming detached, especially during rough handling. Discoloration of the mounting medium often occurs previous to the more serious result above mentioned, proceeding from the margin inward. On the other hand, preparations in which the balsam, storax, or other resinous media are used, are often injured by the running in of the cement used for finishing the slide, when sufficient care is not taken.

A method has been adopted, which effectively obviates these objections, and at the same time renders it possible to mount and finish a slide at once, without the delay due to allowing successive coats of cement to dry before others are applied. The mounts need not be thoroughly hardened before finishing, provided the nature of the preparation does not require it.

The method used is as follows:—The object is mounted on the slide, applying the cover-glass in the ordinary manner, using either balsam, hardened balsam, balsam and benzol, storax, or dammar. The slide is then heated to drive off the solvent, or more volatile constituents, either gently in the water-bath or at a higher heat, even boiling carefully over a spirit-lamp when the nature of the object will permit.

When cold, the superfluous mounting medium, when present, is carefully removed, then a narrow ring of paraffin wax is applied in the following manner:—hard white paraffin wax (such as is used for imbedding) is heated in a suitable capsule until it is melted and quite limpid. With the aid of a very small camel's hair pencil, the melted paraffin is applied at the edge of the cover-glass, covering the exposed mounting medium and instantly solidifying. With round cover-glasses and a turntable, very neat narrow rings of paraffin wax can be readily and

\* Malpighia, iii. (1890) p. 410.

† Micr. Bulletin and Sci. News, vii. (1890) pp. 1-2.



rapidly applied. Whenever they are not satisfactorily symmetrical, a penknife may be used to bring them to the desired shape.

It is now necessary to apply a finishing cement. For this purpose Bell's cement has been found excellent, when modified as described below. The cement ring is finished at one application, enough being applied to produce a well-rounded ring. In a few hours the slide is ready for the cabinet. Bell's cement has been found at times to work unsatisfactorily, not flowing freely from the brush, and forming large bubbles in the ring, particularly in a warm room. The addition of a very little chloroform to the cement, and thorough mixing, produces a material that works smoothly, and dries with a satisfactory finish."

**How to mount Objects in Motion for Examination by Polarized Light.\***—Mr. George H. Curtis remarks:—"None of the manuals I have consulted give directions for preparing slides of objects in motion for polariscope. Rubber cells are best and they should be about 1/16 in. deep and preferably for a 3/4 cover. The medium I use is Canada balsam thinned with a not quite equal bulk of spirits turpentine. Stir well together, and when dissolved filter through cotton. Cement the cells to slide with something not acted on by turpentine, say shellac, or sealing-wax in alcohol. Le Page's liquid glue I think would answer, but I have not tried it. The fragments may be quartz, agate, sand, or anything not soluble in turpentine which polarizes well. One of the best is transparent gypsum or sulphate of lime. It is unnecessary to cement the cover on; set aside for a couple of days and the balsam will get dry enough to hold it. Should you wish to ring them with Brunswick black, size first with a coat or two of liquid glue made thin enough to flow, or the black will probably run in and spoil the slide."

**Glycero-gum as a Mounting Medium.**—Mr. C. C. Faris † finds a solution of gum arabic in glycerin preferable to Canada balsam or glycerin alone, as it is more transparent than balsam, with none of the objectionable features of glycerin. An object can be as well mounted in it without a cell as it can be mounted in balsam with a cell. The solution is made as follows:—Selected gum arabic, 2 oz.; glycerin and distilled water, of each 1½ oz.; thymol, 1 gr. Mix the glycerin and water, and dissolve the gum arabic in it by heating on a water-bath. After the solution has been effected add the thymol, and filter through absorbent cotton by the aid of a hot-water funnel. To have the solution perfectly clear the most transparent pieces of selected gum should be chosen. The solution will then be transparent and brilliant, and be found a successful medium for starches and pollen. It has shown no signs of deterioration after four months.

**Cleaning the Hands after working with Dammar Cements.‡**—A writer in the 'National Druggist' says:—"As everybody knows who has worked at mounting, it is no easy matter to get the gummy and resinous material off the hands. Ordinary soap is of no avail, benzoin is but little if any better, and aside from its costliness, benzoin burns and dries the skin. I have used with a good deal of satisfaction a liquid soap made as follows:—Castile soap, shaved fine, 15 parts; alcohol 95 per cent.,

\* *Micr. Bulletin and Sci. News*, vii. (1890).

† *Western Druggist; Microscope*, x. (1890) pp. 59-60.

‡ *The Microscope*, x. (1890) pp. 25-6.

10 parts; benzol, ordinary, 10 parts; ammonia water, 5 parts; glycerin, 5 parts. Dissolve the soap in the alcohol, add the ammonia water and benzol, and, after thorough agitation, the glycerin. After wetting the hands in plain water, the soap is smeared on with a bit of sponge over the patches of gum or cement, and well rubbed in. After washing and rinsing the hands, partly dry them on the towel, and finish by rubbing them over with a few drops of glycerin. The hands will not crack or chap in the coldest weather if the last precaution be taken. The soap will remain liquid during the summer, but solidifies in cold weather. It is, however, easily liquefied at all times."

(6) Miscellaneous.

'The Microtometist's Vade-Mecum.'\*—Mr. A. B. Lee's work, the first edition of which appeared in 1885, has been so fundamentally revised and rewritten to such an extent that it almost seems like a new work. While a great number of processes have been omitted or only briefly mentioned, other subjects, such as fixation and fixing agents, have received more attention. The methods of killing now occupy a whole chapter, and other chapters, such as those devoted to staining with coal-tar colours, on imbedding processes, the methods of cytology, and on the central nervous system, have been re-written and brought up to date.

The present edition is more suited to the wants of the zoologist than to those of the pathologist.

Demonstration of Bacteria in Tissues.†—Dr. V. D. Harris has translated and edited Prof. Kühne's small work, which deals with the question of how to stain bacteria in animal tissues, and the answer thereto is somewhat affected by the author's peculiar but not unpractical views.

In addition to running through the technique of preparing, staining, and mounting specimens, it gives a few very useful formulæ and some useful pieces of advice.

The translation, which is decidedly Germanesque in style, also bears evidence of want of revision. For example, Mastzellen are usually translated plasma-cells, not fat-cells (p. 10). The 50 per cent. carbolic acid solution (p. 38, No. 1) does not agree with the 5 per cent. mentioned on p. 14. On the whole, we think that if the work were rewritten it might possibly be useful to some student unacquainted with the German tongue.

RAWITZ, B.—*Leitfaden für histologische Untersuchungen.* (Introduction to Histology.) Jena, 1889, 8vo.

REMY, CH.—*Manuel des travaux pratiques d'histologie, des éléments des tissus, des systèmes des organes.* (Manual of Practical Histology.)

Paris, 1889, 8vo, 399 pp.

TYAS, W. A.—*Methods of Hardening, Imbedding, Cutting, and Staining animal sections, and methods of mounting the same.*

*Trans. Manchester Micr. Soc.*, 1888, p. 83.

ZUNE, A.—*Traité de microscopie médicale et pharmaceutique.* (Treatise on Medical and Pharmaceutical Microscopy.)

Bruxelles (H. Lamertin), Paris (J. B. Baillière et fils), 1889, 1 vol. sm. 8vo, 130 pp. and 41 figs.

\* 2nd ed., London (Churchill), 1890.

† 'Guide to the Demonstration of Bacteria' (Kühne), translated by V. D. Harris, M.D., London, 1890, 52 pp. and 7 figs.

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**JOURNAL**  
OF THE  
**ROYAL**  
**MICROSCOPICAL SOCIETY;**

CONTAINING ITS TRANSACTIONS AND PROCEEDINGS,

AND A SUMMARY OF CURRENT RESEARCHES RELATING TO

**ZOOLOGY AND BOTANY**

(principally Invertebrata and Cryptogamia),

**MICROSCOPY, &c.**

*Edited by*

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*and Professor of Comparative Anatomy and Zoology in King's College;*

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FELLOWS OF THE SOCIETY.



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VII.—*On a Simple Form of Heliostat, and its Application to Photomicrography.*

By THOMAS COMBER, F.L.S.

(Read 21st May, 1890.)

YOUR Secretary has asked me to give your Society a detailed description of the apparatus I use for photomicrography, and of my method of working; but it appears to me that it will be simpler and shorter, and at the same time answer every purpose, if I merely explain those features in which my mode of working differs from that which I believe is generally adopted by others, and is probably sufficiently well known. The general nature of the arrangement will be apparent from the woodcuts.

The two main objects that I have endeavoured to attain have been, firstly, a means of sunlight illumination, easily applied, quickly adjusted, and simple in construction so as not to be liable to get out of order; and secondly, an arrangement which admits of convenient and comfortable eye-observation, for the purpose of arranging the object and adjusting cover-correction, before the camera is attached to the Microscope.

So far as my experience goes, for high magnification—other things being equal, both as regards objectives and manipulative skill—better results can be obtained by sunlight than by any other kind of illumination. The photomicrographs produced by Mr. Nelson and other of your members by oxyhydrogen light may be superior to what others have produced by sunlight; but this is due to their superior optical appliances and greater skill as microscopists, which more than compensates for what I cannot help regarding as inferior illumination. The same operator, using the same lenses, will, I am confident, produce better results by sunlight than by any artificial illumination.

The reasons sunlight has been so little used in this country are probably (1) the uncertainty of our climate; (2) the fact that many of our microscopists work chiefly in the evening; and (3) the complicated nature of the heliostats obtainable, which renders them very liable to get out of order, and so difficult to adjust that, when sunlight is available, much time is lost in setting up the apparatus; and, consequently, before everything is in working order, the sun may too often become clouded. The last objection is aggravated by the heliostat being usually placed a considerable distance from the Microscope, and sometimes even outside a window; and, as any error in the action of the heliostat is increased in proportion to the distance, it has been found almost impossible to keep the illuminating beam unchanged by the motion of the sun.

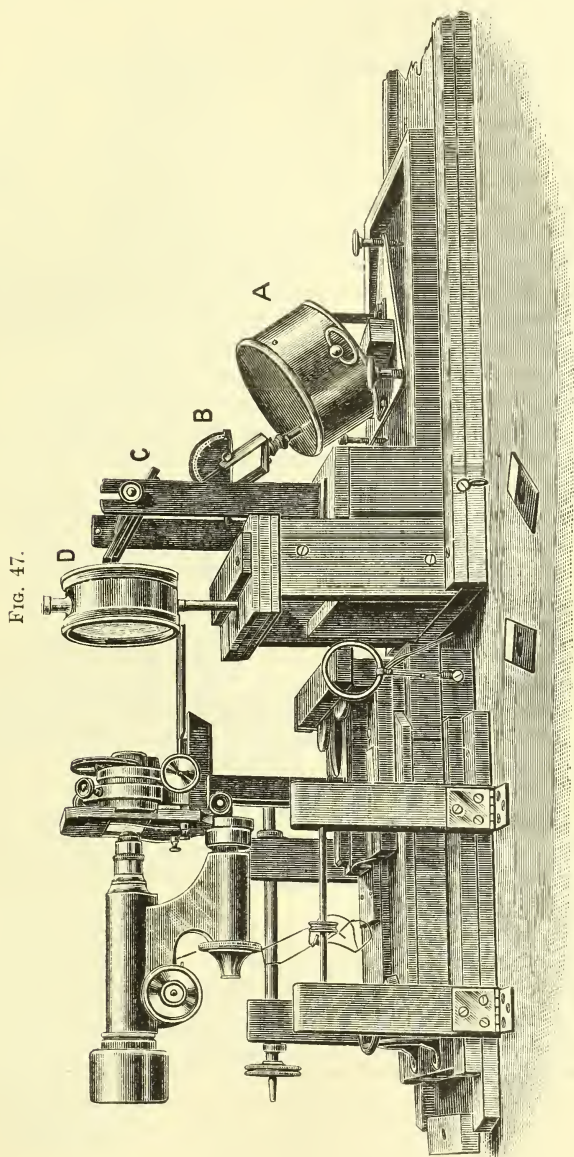
To avoid this difficulty, I place the heliostat inside the window,



and bring it quite close to the Microscope, so that it is within arm's length of the observer, and the sunbeam has so short a distance to pass before it reaches the substage condenser, that any slight error of the heliostat is of comparatively little consequence. The heliostat and all the accessories are fixed, once for all, on a wooden stand, so that they have not to be arranged each time they are used, but the stand has merely to be placed before the Microscope, and everything is in its proper relative position.

The heliostat itself is a brass time-piece A, fig. 47, to which is added an additional motion, causing the spindle, which need not be in the centre, to revolve once in twenty-four hours. It is mounted on a triangular brass plate, furnished with levelling screws, and is fixed at an angle to the horizon, corresponding to the latitude of the place in which it is to be used. When the point of the brass plate is directed due south, and the plate itself is levelled, by means of a spirit-level, in both directions, the clock is in the plane of the equator, and the spindle, at right angles to it, is parallel to the axis of the earth, and points to the North Pole of the heavens. The spindle is made slightly conical, and fitted to it, friction-tight, so as to be capable of easy rotation by the hand, is a small mirror B, with universal motion. The size of mine is two inches by one, which is ample. This mirror has to be set to reflect the light from the sun in the direction of the spindle, when the rotation of the spindle, corresponding exactly with that of the earth, only in the reverse direction, compensates for the apparent motion of the sun, and the reflected beam remains motionless. Where the reflected beam crosses the optic axis of the Microscope, there is placed a second fixed mirror C, inclined to the horizon at an angle equal to half the latitude, which reflects the beam in the axis of the Microscope. Between this fixed mirror and the condenser is placed an alum-cell D, to absorb the heat. In originally fixing the position of the mirrors, care has to be taken that the centre of the fixed mirror is truly axial with respect to the substage condenser and Microscope, and that, reflected in it when viewed through the Microscope, the spindle of the heliostat appears exactly end on, in the centre of the field. The heliostat will then be in its correct position, and the movable mirror can be placed upon it. All this may seem very complicated in the description; but once the position of the various pieces has been thus settled, all that has to be adjusted is the movable mirror, and its adjustment is no more difficult than that of the mirror which forms the ordinary adjunct of the Microscope. If the mirrors are of glass silvered at the back, the first gives a double reflection, which is again doubled by the second, and great loss of light is experienced. Glass silvered on the surface avoids this, but I found it tarnished quickly; so that I have had to adopt reflectors of speculum metal. These also are open to objection, for the light they reflect is distinctly reddish in tinge, and I believe there is considerable absorption of the rays of highest refrangibility.

The window at which I work faces about S.E., and has the sun from early morning until about two P.M., and, to ensure the apparatus



being placed due south, the end of the board upon which the heliostat stands is cut off at the angle corresponding to the glass of the

window, so that the table can be easily placed exactly in the required position.

The table itself (fig. 48) is heavy and solid, and stands upon three legs, so as to secure an equal bearing. It is at such a height that the horizontal Microscope-tube is at a convenient level for eye-observation, when the observer is seated, so that all the preliminary adjustments, as regards cover-correction, &c., can be comfortably made, and the illumination regulated, before the camera is attached. The base-board of the camera pivots on a steady tripod, and can, during this process of adjustment, be swung aside out of the way, but be brought round when required, and the anterior end of the base-board then fits to the edge of the Microscope table. The attachment of the camera to the Microscope is effected in the usual manner. For my own work, I find it most convenient to use a camera of fixed length, viz. one metre from eye-piece to sensitive plate; but a bellows body, capable of variable extension, can, of course, be substituted if desired. The focusing rod disconnects at the anterior end of the camera, sliding back off a square pin from the portion attached to the Microscope table. It works by means of a string, that passes round the milled head of the fine-adjustment (Fig. 49). The bar which carries the socket of the substage condenser has attached to it a small platform, upon which can be placed a screen of dark-blue glass, to subdue the glare for eye-observation, or a small cell containing ammonio-sulphate of copper or other solution, for producing monochromatic light.

So far, however, I cannot say that I have experienced any practical advantage from monochromatic light. It appears to me that when ordinary sunlight is used, the blue-violet rays are so prepotent in their actinic power that they do all, or nearly all, the work, and the other rays have not time to produce any material effect. The supposed advantages of monochromatic light are then practically attained without any special means, unless, indeed, some special method can be devised for working with rays of shorter wave-length than the blue-violet; and any suggestion for accomplishing this I shall be glad to receive, and to give it a trial.

The resolving power of our objectives depends not only upon their numerical aperture, but also upon the wave-length of the light used; and the high ultra-violet rays should therefore give a higher resolving power than the blue-violet; but I have not yet succeeded in making them operative in practice.

As regards general manipulation, the only special recommendations that I have to make are:—(1) That the cone of illumination should always be strictly axial. (2) That the image of the sun should be focused exactly in the plane of the object, so that it shows sharp and clear on the ground glass when the object is in focus. Clouds close to, or passing across the face of the sun, should be seen almost as if a landscape lens was being used. (3) That no unachromatized lens



should be introduced in any part of the system. I cannot, therefore, advise the use of a bull's-eye between the source of light and the sub-

FIG. 48.

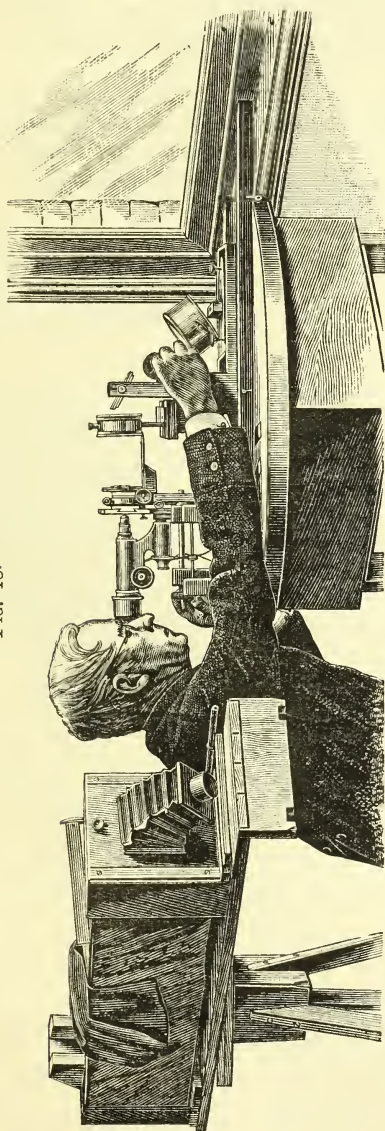
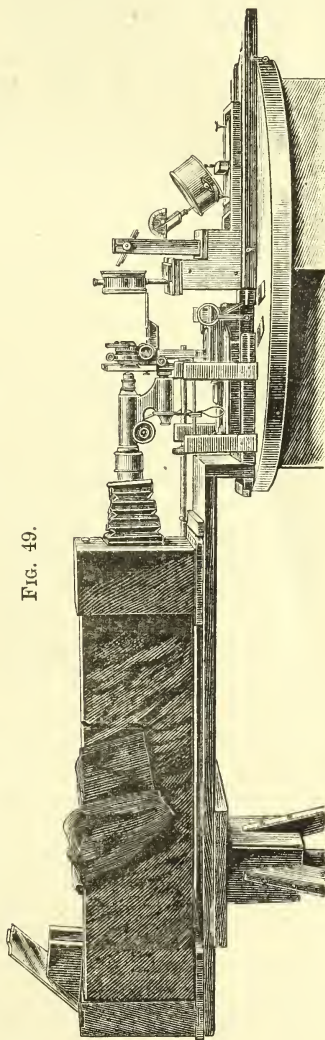


FIG. 49.



stage condenser. The angle of the cone of illumination which gives the best results, varies, I believe, not only with the object, but also with



the individual objective used. Too narrow a cone is apt to cause diffraction fringes, too wide a cone produces haze. I have not had much experience in photographing test diatoms, but so far as it goes, I find that my own 2 mm. Zeiss Apochromatic, 1.4 N.A., gives its best definition of such objects when about two-thirds of its back lens is filled by the dioptric beam.

I trust this description of my apparatus will enable others who may be desirous of using sunlight illumination to adopt it, and, I hope, improve upon it. I shall be pleased to answer any inquiries as to any point that may not have been made sufficiently clear.

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## MICROSCOPY.

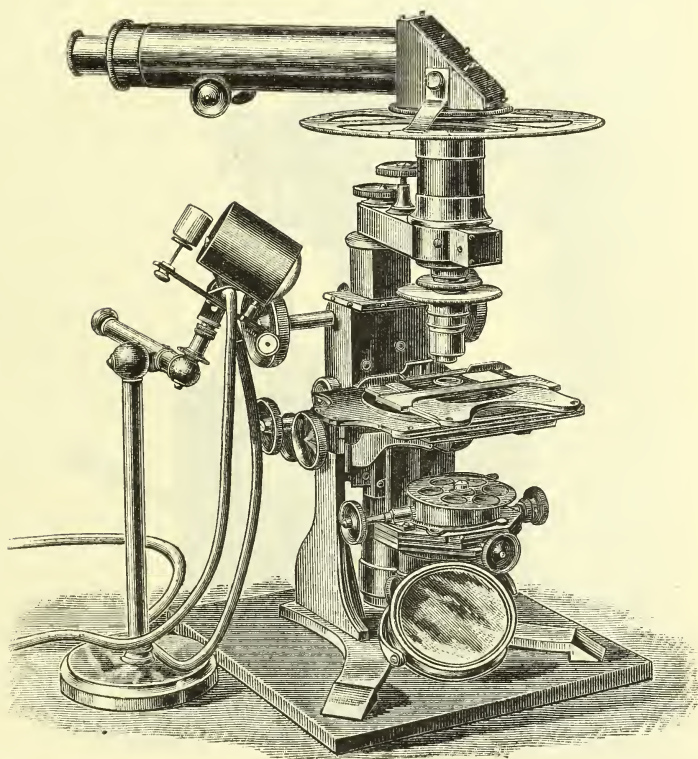
## a. Instruments, Accessories, &amp;c.\*

## (1) Stands.

**Braham's Universal Microscope.**—The following description has been communicated to us by Mr. Philip Braham, of Bath:—

“The original design of the instrument was based on the most improved Microscope, devised by the late Andrew Ross : but the modi-

FIG. 50.



fications I have made with a view to facilitating special investigations are considerable.

Fig. 50 shows my application of a rectangular prism, giving the

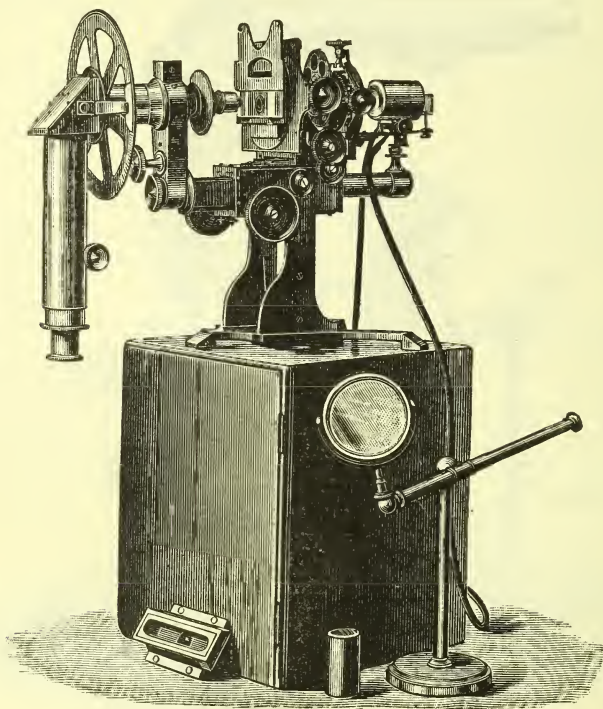
\* This subdivision contains (1) Stands; (2) Eye-pieces and Objectives; (3) Illuminating and other Apparatus; (4) Photomicrography; (5) Microscopical Optics and Manipulation; (6) Miscellaneous.

observer an easy position for examining objects in liquids, and the means of measuring the angles of crystals.

The angles of crystals are measured by cross-wires in the focus of the eye-lens, a divided circle attached to the body of the instrument, and a Vernier attached to the side of the brasswork, carrying the rectangular prism, which is adjusted by three screws, so that the hypotenuse is exactly at an angle of  $45^\circ$  with the optic axis. The adjustment is made by placing on the stage a slip of glass, ruled with fine cross-lines, which are made to coincide with the cross-wires in the eye-piece.

The divided circle can be turned by rotating the tube at right angles

FIG. 51.



to the optic axis. The magnified image of the crystal also rotates, the angles being measured by the coincidence of the sides of the crystal with the cross-wires in the eye-piece.

This arrangement is also useful in observing phases in polarization, the tube carrying the polarizing prism on the substage being rotated by clockwork, and four pins making electrical contact and ringing a bell,

by which every quarter revolution is marked, and attention called to the changes visible.

The limelight illuminator is shown in position for illuminating opaque objects, and a light from the mirror through coloured glass gives a good background for a variety of objects.

FIG. 52.

The limelight apparatus shown is conveniently clean and devoid of smell, and gives out very little heat. It can be used for oblique, opaque or transparent illumination, and can be varied in intensity. It consists of a diminutive limelight on a condenser stand, with an adjustable plano-convex lens in front. By varying the distance of the plano-convex lens in front of the limelight either convergent, divergent, or parallel rays can be obtained and projected in any direction.

Fig. 51 shows the instrument in position to project an image of an object on a sheet of paper on the table for sketching; the limelight being attached in the place of the mirror.

Fig. 52 shows the adjustment of the instrument in an inverted position. A board is attached to the box, and two struts are applied; the Microscope is then clamped to the upper part of the board, the feet fitting into corresponding notches in the board. This enables the observer to examine objects from beneath, whilst objects in liquids and tubes are seen free from cylindrical aberration by immersing the tubes in a cell shown on the table in Fig. 51.

The interior of crystals or gems can be microscopically explored by immersing them in equally refractive liquids.

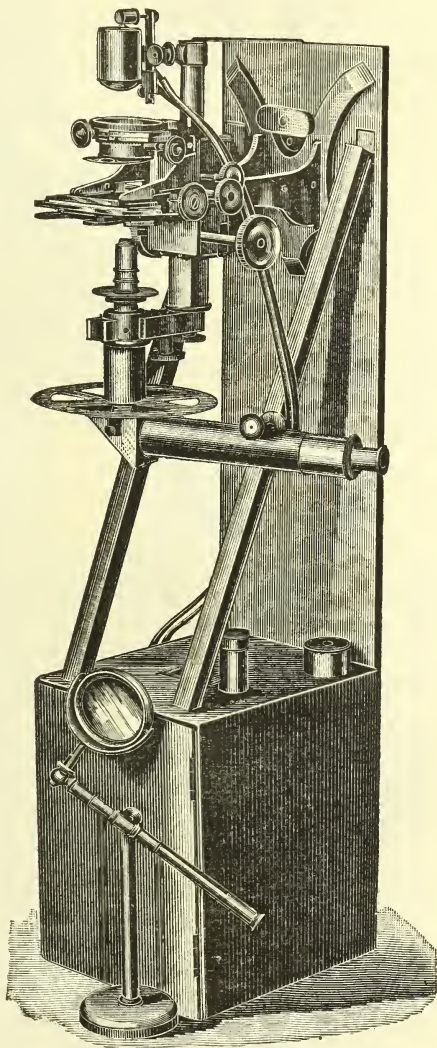
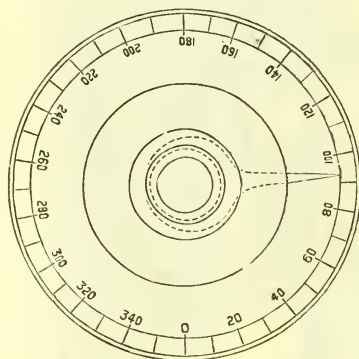
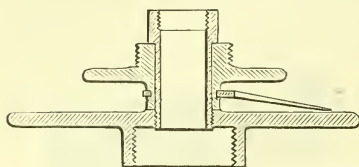




Fig. 53 shows my rotating nosepiece, consisting of a screw fitting to the objective end, and a divided circle fitting to the objectives and rotating. An index pointer sprung on to the nozzle shows the angle of rotation; the other end of the rotating tube is adapted to receive the analysing prism or a double-image prism, which can be used for measuring the angles of crystals by rotating the magnified extraordinary image round the ordinary. It can also be used in testing objectives."

FIG. 53.



the condenser carrier turns is fixed to a slight prolongation from the right posterior corner of the stage, which the author considers to be "a very great advantage; it constitutes for the hand that works the micrometer-screw a kind of natural support, and allows the fingers much ease and suppleness in using the screw."

M. Fabre Domergue refers to condensers as having been "completely neglected ten years ago!" The introduction of the Abbe condenser, he says, imposes upon constructors the necessity of modifying the old models of stands so as to allow of the introduction of condensers beneath the stage.

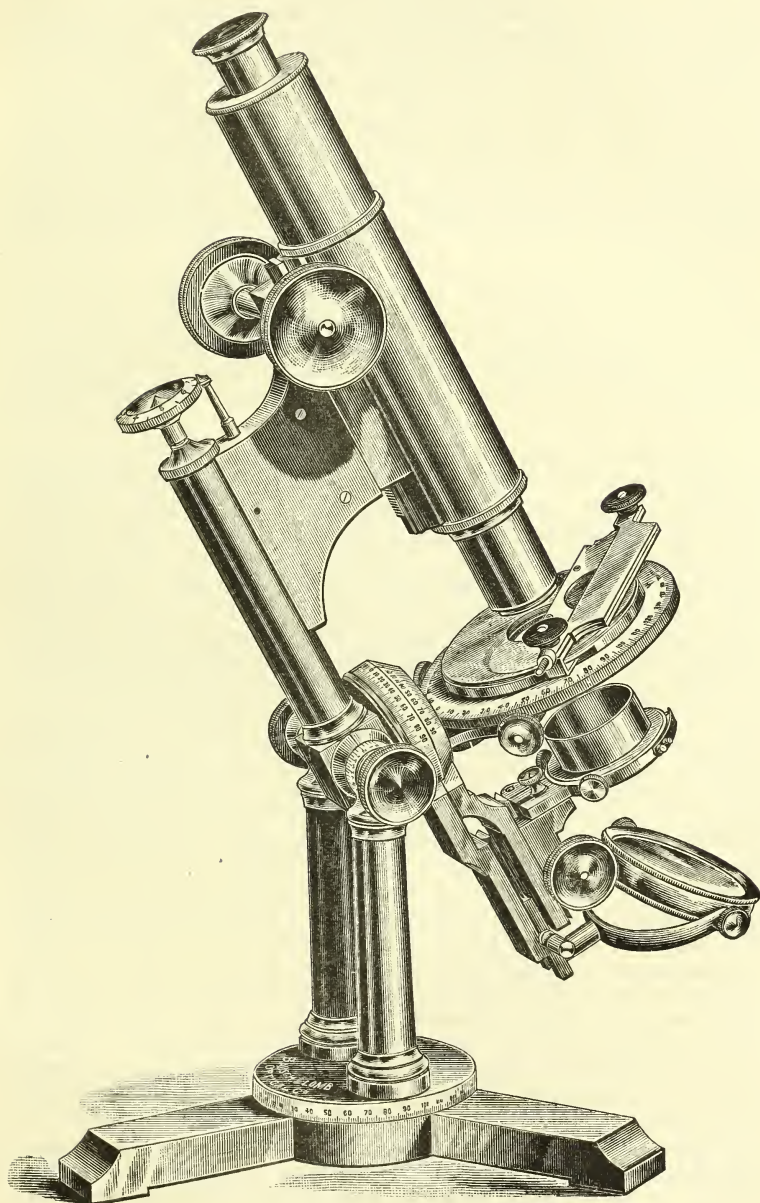
**Hart's Microtome-Microscope.**†—The following is the abstract of a paper by Dr. C. P. Hart, as printed in the Proceedings of the American Association for the Advancement of Science, under the title of "*A new, cheap, useful, and quickly constructed adjustable Microtome.*"

"This instrument is nothing more nor less than a Bausch and Lomb "Microscope-stand, converted into a microtome by the following changes: "—Having removed the substage, the slide-carrier clips, the objective "adapter, and the draw-tubes, a suitable razor-blade is permanently fixed "to the slide-carrier, so as to have a corresponding universal lever move-

\* Ann. de Micrographie, ii. (1890) pp. 164-7 (1 fig.).

† Proc. Amer. Assoc. Adv. Sci. for 1885 (1886) p. 356.

FIG. 54.



HART'S MICROTOME-MICROSCOPE.

"ment parallel to the glass stage. The imbedded substance is then carried "down the main tube of the instrument (which is placed in a horizontal "position) until it presses gently against the microtome-knife, when it is "fixed in position within the tube by means of the main draw-tube, the "diaphragm of which, either directly or by means of a small wooden "cylinder, is brought in contact with the distal extremity of the substance "to be divided, and this acts as a plug or follower to retain it in position "within the tube. Then, having moistened the knife, and, if necessary, "the substance to be operated on also, the slide-carrier, and with it the "microtome-knife, is made to pass through a sort of revolving cutting "motion, by which the sections are made. These sections may be made "of any degree of delicacy by means of the micrometer-screw attached to "the instrument."

The notion of converting such a Microscope into such a microtome seemed to be so unique in the novelty of its originality (it is difficult to hit on the exactly appropriate designation) that we imported the instrument from the United States, and give an illustration of it in fig. 54, which shows the razor-blade and slide-carrier in the form designed by Dr. Hart.

**Alterations in Nobert's Microscope.\***—Herr Kayser describes some alterations made on a Nobert Microscope. He particularly mentions the simple reading arrangement constructed by him, which is just as serviceable as a microscopical one composed of eye-piece and objective, but it does not invert the image. This arrangement consists of a small tube, containing only a thread and a plano-convex lens. Close to the eye comes the thread stretched horizontally, and then the lens, with convex side in front, at such a distance that the image of the thread is distinctly seen by the passage of the rays through the lens, and reflection at its plane silvered surface. A narrow strip of the silvering is removed in a direction passing through the centre, and at right angles to the thread. Consequently, when the distance of the tube is suitably adjusted, the eye can see a division through this central space. In order to have the division, but not the thread, more strongly magnified than in this, the simplest case, a second plano-convex lens of suitable focal length can be added immediately on the plane silvered face of the lens. Here two equal lenses of 10 mm. diameter and 25 mm. focal length are combined. On the thread end of the tube a white paper screen inclined at 45° with central aperture is fitted for the illumination of the thread. This small reading arrangement is fastened to the object stage, while an ivory rod with a range of 80 mm. divided into half millimetres, and fixed vertically on the Microscope-tube, can be displaced with the tube. A screw with large drum divided into 50 divisions serves to raise or lower the stage by slow degrees. Since the tenth of the division can be easily read, an arrangement is thus attained which, over a very large interval (80 mm.), gives an adjustment and a measurement which is exact up to 1/1000 mm. This is of importance, for instance, for microscopical measurements of the refractive indices of transparent plates. By means of the fine screw, the error of the divisions on the scale can be tested, and it is especially serviceable in

\* Schrift der Naturforsch. Gesells. Danzig, vii. (1890) pp. xi.-xii.



adjusting objectives of short focal length and immersion systems, which must otherwise be done by testing, and consequently with danger to the apparatus. In determining the refractive index of a transparent plane parallel plate, Herr Kayser proceeds as follows. The refractive index

is  $= \frac{D}{D-d}$  where  $D$  and  $d$  are given by three readings on the scale,

when the adjustment of the Microscope is made:—1. On the support of the plate. 2. On the upper face of the plate, after it has been put on the stage. 3. On the support as seen through the plate. The readings 1 and 2 give  $D$ , the readings 1 and 3  $d$ . This method is, however, not sufficiently precise. Another and more exact method, with experimental proof, will be given later.

### (3) Illuminating and other Apparatus.

**Mayall's "Jewelled" Fine-adjustment.**—At the April meeting of the Society, Mr. J. Mayall, junr., referred to an improved form of fine-adjustment constructed and exhibited by Messrs. Powell and Lealand, for the production of which he was himself chiefly responsible. He said that during the past ten or twelve years, several forms of fine-adjustment had been brought to the notice of the Society, but the principal aim in most of them had been economy of production or lowness of price, without regard to improving on the best existing forms. In the new form exhibited the chief aim had been to construct a fine-adjustment that should combine extreme sensitiveness of action with accuracy and probable durability, beyond what had previously been attained. With this view he had carefully considered every known form of fine-adjustment, and had selected that of Messrs. Powell and Lealand, as representing the highest type of construction yet devised, with which to test the possibility of improvement. The essential feature in the improvement was the application of what watchmakers would term a "jewelled movement." The whole of the contact surfaces by which the fine-adjustment was actuated consisted of polished steel and agate, the intention being to reduce the friction as much as was consistent with steadiness of motion. The perfection and durability of jewelled mechanism was a great feature in the highest class of clocks and watches; the most delicate parts of Nobe's ruling machine were jewelled, as were also the bearings in Dr. Hugo Schröder's feeling level for testing the accuracy of plane surfaces. Those who were familiar with Powell and Lealand's fine-adjustment, as previously constructed, would understand the extreme difficulty of improving the mechanism substantially, for it was the outgrowth of long experience and of the most conscientious devotion of expert mechanicians to the task of providing a perfect focusing movement. No other fine-adjustment had reached the same high standard of construction, which was probably due to the fact that during the fifty years that had elapsed since its first production, the makers had kept steadily to the same system, only varying the minor details of the mechanism as experience critically suggested in the direction of improvement.

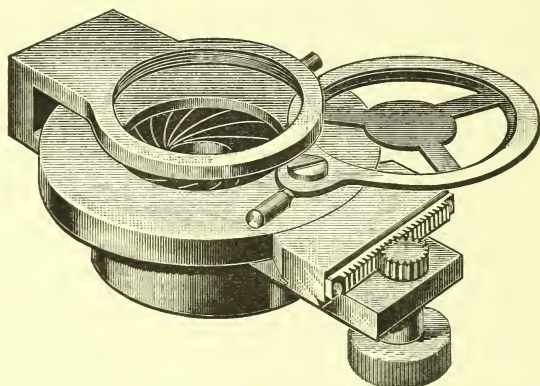
The application of polished steel and agate bearings throughout the mechanism was intended to reduce the friction, and thus render the



action more sensitive without introducing unsteadiness. The result attained was undoubtedly an improvement on the whole system, though the cost would probably limit the application to the few instruments required for very special and difficult investigations in microscopy. For high-class photomicrographic work, or where preparations had to be retained under observation for long periods of time, the new mechanism should be particularly useful, for the greater solidity of the general construction clearly pointed to greater precision of action and increased stability.

**Messrs. Bausch & Lomb's Condenser Mounting.\***—We give a figure of a condenser mounting with iris diaphragm recently designed by Messrs. Bausch and Lomb. This mounting provides a movement for the

FIG. 55.



diaphragm by rack and pinion. It has in addition a recess for receiving central stops and blue glass.

It can be attached to an adjustable substage or to a substage fixed to the stage, and may be used with the high and low angled Abbe condenser.

**New Stage Micrometers.**—At the May meeting of the Society, Mr. E. M. Nelson called attention to a new stage micrometer, produced by Messrs. Powell and Lealand, and so excellently ruled as to be worthy of remark:—"It comprises 100ths and 1000ths of an inch, and 10ths and 100ths of a mm., there being 10 divisions of each set; the finer divisions of  $\cdot 001$  in. and  $\cdot 01$  mm. being placed in the centre, the  $\cdot 01$  in. being on the one side, and the  $\cdot 1$  mm. on the other, respectively, a guiding line being ruled at right angles to them. The lines are fine,  $1/30,000$  in., and are blackened in, and mounted in balsam. The lines are straight, and evenly ruled. With regard to the spacing, I have made exhaustive comparisons with fine micrometers by Rogers and Zeiss, and some others not quite so perfect. Upwards of 240 screw micrometer

\* Amer. Mon. Micr. Journ., xi. (1890) pp. 25-6.

measurements were made, and the work carried on under hypercritical conditions. An account of these may be of interest. First, a magnification of 1200 diameters by means of a suitable immersion lens was employed for the finer ruling, and for the coarse a dry  $1/6 \times 600$  diameters; the screw micrometer was on an independent mounting. Care being taken with regard to the illumination, &c., a critical image of the lines was obtained. The order in which the lines were taken was from left to right, as seen in the instrument; each interval was then designated by consecutive letters of the alphabet. The intervals were then most carefully wired, and each value set down under its corresponding letter; when the ten spaces were finished they were meant.

It was then easy to see which interval differed from the mean, and to calculate how much. In the same way comparison can be made with any other scale, it matters not whether it is ruled in inches or mm. It is most important that both the instrument and the observer be tested. To this end I proceeded as follows. The screw value of 20 intervals on a badly ruled scale was written down as above, the paper was then put away, and the operation performed again.

On comparing the two papers, the screw values of seven intervals were identical, 12 different by one division, and one by two divisions. This error of two divisions occurred in the interval H, the first reading being 1033, and the second 1031. On careful re-examination of this interval, I came to the conclusion that the first reading was the bad one, and that the true value was 1031 or 1032. On substituting this last value in both sets of readings, the 20 intervals meant precisely alike, viz. 1038. As this forms a suitable illustration of the work, I append the two columns. With the exception, therefore, of the interval H, the screw readings may be taken as true to  $\pm 1$ . The point, therefore, we have to determine, is the value of  $\pm 1$ . The mean 1038 being the value in divisions of the screw-head, for  $50 \mu$ , the value of one division consequently =  $.000001897$  in., or less than  $1/500,000$  in.

This might be called 'the constant of the instrument, and observer.' We next have to find the greatest errors of the intervals from the mean; G is the greatest, and S the least. Calculation shows that G is  $1/20,000$  in. too large, and S  $1/40,000$  in. too small.

But, on returning to Powell's scale, we find a much closer agreement than this. Taking the  $.001$  in. first, we find the mean to be  $628.0$ . Three out of the ten intervals agree to that mean to  $\pm 1$ : this being 'the constant of the instrument and observer,' they are without sensible error. Four intervals agree to  $\pm 2$ , which is less than  $1/300,000$  in.; two lines B and H agree to  $\pm 3$ , which is less than  $1/200,000$ , and one interval G is  $+ 4$ , viz.  $1/157,000$  in., too large. Now, as we found that  $\pm 1$  was the limit of observation, we may say that the scale, with the exception of B, G, and H, has no sensible error. Practically speaking, G is the only interval that is out, and its error is small in comparison with other scales.

The next scale is the  $1/100$  mm.

The  $.01$  mm. is too small a quantity to treat in the above way; it must be left until we have objectives as perfect as those we have at present, but of double their power.

All that can be done is to take several of the divisions. Eight sets

of three each were measured on Powell's new scale: the variation from the mean was less than  $1/200,000$  in. Rogers' is a very well ruled scale; it is, however, difficult to observe, the lines being without pigment, and it is mounted dry. The lines under these circumstances present the usual black and white diffraction images. It is, on that account, very difficult to maintain an equable focus during measurement. In Rogers' scale, the greatest error is in interval G, where it amounts to four divisions, or somewhat less than  $1/100,000$  in. Thirteen out of twenty intervals have practically an insensible error. One cannot speak with the same certainty with regard to this plate as to the others, because of the focal difficulty. Different readings gave discordant results; therefore, in this case, more must be allowed for the 'constant of the observer and instrument.' With regard to the  $1/10$ ths of a mm. on Powell's scale, they were examined by a power of 600 diameters by a dry lens. The mean was 987; six intervals had no sensible error, but C and G had an error of three divisions, which is equivalent to  $1/100,000$  in.

Rogers gave a very similar result.

The error of the interval D, in the Zeiss scale, was  $1/30,000$  in.

I next compared the length of the mm. on the three scales, that is Powell's, Rogers', and Zeiss', with each other. I detected a slight but insensible difference of  $\pm 1$ . All that now remains to be done, is to compare the inch and the mm. scale on Powell's plate. By measurement, we found that  $30 \mu$  gave a screw value of  $741.25$ ; therefore, the value for  $.1$  mm. would be  $2470.8$ , and the value for  $1/1000$  in.

$$\frac{.001 \times 2470.8}{.003937} = 627.59.$$

The value actually measured was, as we saw above,  $628.0$ ; here again there is no sensible discrepancy. In conclusion, I feel sure that such an accurately ruled micrometer, and one so clear to read, will prove extremely useful to microscopists at large.

Before closing, I would like to bring to your notice a screw micrometer made for me by Mr. Powell, which contains some slight modifications from the usual forms, which practical experience has suggested to me.

First, with regard to the lens portion, I have substituted a compensating positive for the old form of Huyghenian or Ramsden. This yields far better images when making measurements with apochromatic and ordinary objectives. I have so arranged it that the compensating eye-lenses of different foci are interchangeable. In fact, no special lens is required, you use your ordinary working eye-piece, whatever that one may be. This is, of course, a great advantage: bacteria, for instance, require a high-power eye-piece micrometer, while such a power would be useless on an ordinary object.

Therefore, the ability to regulate your eye-piece power to the object to be measured, will meet a long felt want.

Next let me say that I entirely disapprove of having two movable threads; at the outset 'the constant of the instrument' would be doubled; moreover, I am confident that a movable zero is a mistake.

I have, therefore, considerably altered this portion of the instrument by making the screw portion, together with the fixed zero thread, movable in the other part, which might be aptly termed 'an eye-piece

adapter.' By this we secure the advantage of the double movable thread, without the additional error of the double movable thread, and this, moreover, without losing the convenience of a fixed zero.

This enables you to span your object at equal distances on either side of the optic axis, without disturbing the centricity of the eye-piece. A guiding line has been added, because an error might creep in unless measurements are made with precisely the same portion of the wires.

The divisions on the screw head have been made white on a black ground, on account of their being easier to read in a darkened room. A cap to protect the threads from dust and injury, &c., is provided, as the threads are no longer inclosed between the lenses, as in the Huyghenian form.

An iris diaphragm is placed below the threads and as close to them as possible.

In spanning the stage micrometer, it will be found better to take the readings from centre to centre of the lines, by doing which you avoid the diffraction which is always present at edges.

The measurement of all objects should be performed under a wide angled cone of illumination, so that the diffraction at the edges may be minimized as much as possible."

*Two Readings of Scale 50  $\mu$ .*

				diff.
A	1038	A	1038	0
B	35	B	36	+ 1
C	37	C	36	- 1
D	36	D	37	+ 1
E	30	E	29	- 1
F	37	F	37	0
G	65	G	64	- 1
H	32	H	32	0
I	29	I	30	+ 1
J	48	J	48	0
K	34	K	33	- 1
L	29	L	30	+ 1
M	40	M	40	0
N	45	N	45	0
O	38	O	37	- 1
P	44	P	43	- 1
Q	39	Q	39	0
R	40	R	41	+ 1
S	24	S	24	0
T	40	T	41	+ 1
20	760	760		0
	1038	1038		

H H altered from 33 and 31 respectively.

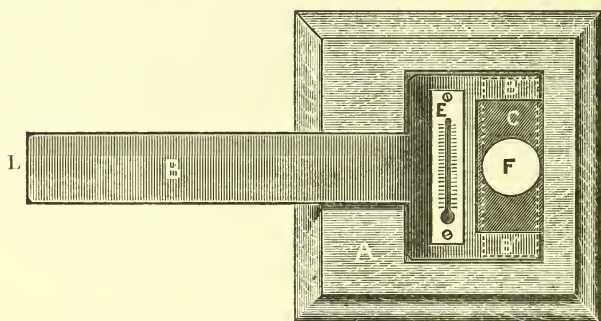
**An easily constructed Hot-stage.\***—A very simple and convenient hot-stage was exhibited by Dr. Robert Reyburn at a recent meeting of

\* Amer. Mon. Micr. Journ., xi. (1890) p. 1 (1 fig.).



the Washington Microscopical Society. This form is adapted from the more complicated and expensive forms used by microscopists, and is claimed to be especially useful from the fact that it can be made at a trifling cost by any one possessing a little mechanical skill. In fig. 56, A represents the wooden block or stage which is fastened upon the brass stage of the Microscope. A space is cut from the upper surface of this block, as shown by C, into which is fitted a piece of copper plate (B, B', B''). A round hole is also cut at F, the opening of the brass stage, to allow of the illumination of the object to be examined. The slide is placed on the copper bed with its ends resting at B' and B'', as indicated by the dotted lines. The heat is applied by a spirit-lamp at the end L of the copper plate B which

FIG. 56.



gradually transmits the heat by conduction to the slide. The temperature is registered by the thermometer E, which is screwed fast to the copper plate.

**Application of Apertometer to the Microscope.\***—Herr Kayser remarks that the narrowest perceptible distance of a wave-length stated by Fraunhofer and Nobert, is not the extreme limit which the newer Microscopes with oil-immersion systems have reached in the resolution of the structure of diatoms. The smallest recognizable distance  $\epsilon$  approximates to the expression established theoretically by Helmholtz

$$\epsilon = \frac{\lambda}{2 \sin a},$$

where  $\lambda$  denotes the wave-length and  $a$  the angle of divergence under which the extreme rays from the axis of the object fall upon the objective system. Since this angle can with an immersion lens be nearly a right angle, the numerical expression for the limit, taking  $\lambda = 0.00055$  mm. in the most general case, will amount to the half wave-length 0.000275 mm. According to the practical investigations of Abbe and Dippel, the resolving power of an objective system stands in very close relation to the magnitude of the angle of divergence. On this account makers are

\* Schrift. der Naturforsch. Gesells. Danzig, vii. (1890) pp. xiii.-xvi.

obliged to have regard to the greatest possible angle of aperture or to the highest "numerical aperture" of Abbe, of which the expression is  $a = n \sin u$  (where  $u$  is the refractive index). Accordingly they give in their price lists with dry systems the angle of aperture or the numerical aperture, and with immersion systems the latter. Whether these data correspond to the facts must be subjected to experiment. Herr Kayser received from a well-known firm an oil-immersion (1/16 in.), which he had required to be capable of resolving *Amphipleura pellucida* in oblique light. The system supplied did not answer to the requirements. The maker having ascribed the non-resolution to "badness of the preparation, defects in means of illumination, stand, &c.," it remained for a time doubtful whether these circumstances were really to blame. Herr Kayser was at that time engaged on the construction of apertometers. The apparatus resulting from his investigations, which serves for the examination of dry systems, has the following arrangement. Round a horizontal divided circle a vernier can be turned, and an upright, on which is fixed a Microscope directed horizontally, is set up in the centre. In front of the objective of the latter is a ring attached by a pin to the same upright. This ring can be rotated about the axis of the upright by means of side pieces which reach to the horizontal scale and carry a second vernier. With the plane of the ring at right angles to the axis of the Microscope, which passes through its centre, the reading on the second vernier is  $90^\circ$ , when the direction of the Microscope corresponds to the reading  $0^\circ$ . The system whose aperture is to be tested is placed in the ring. The Microscope is then displaced along its axis until the combined optical apparatus, which acts as a non-inverting telescope, shows the images distinctly. When by suitable turning of the whole apparatus the cross wires of the Microscope have been adjusted on an object not too near, the first vernier is displaced, without moving the second, both to right and left, until the image in each case just vanishes on the edge. The sum of the two angles read off is the angle of aperture. The angle thus given for an objective system, No. 7, of about 4 mm. focal length, was greater than the value given for it and found by the Abbe apertometer, in which the identity of an optician's systems of equal members is assumed. The author, attributing the magnification to his apparatus objective, tried the apertometer objective of Zeiss, specially made for the Abbe apertometer; but even with this the result remained unaffected.

The second apertometric apparatus constructed by Herr Kayser can be used for both dry and immersion systems. It consists simply of a glass plate of which one face is silvered and has scratched upon it a system of concentric circles which come into observation according to the dimensions of the apertures to be determined. The plate is laid on the stage with the silvered side downwards, and carries on its upper face in the middle of the rings, a small cover-glass, on the under side of which is a small mark. The Microscope containing the objective to be tested is first adjusted on this mark. Then without moving the body-tube the eye-piece is withdrawn, and again replaced in the tube when combined with the apertometer objective. The eye-piece is then adjusted so that the rings near the edge appear quite distinct; the extreme ring is counted, and if it does not exactly coincide with the edge, an estima-

tion in tenths of the following ring interval is made. A central portion of the silvering is removed and illumination by a mirror used in order to make the mark on the cover-glass visible. For the illumination of the rings, however, a white paper screen above the objective, and set obliquely to the incident light, is sufficient. The rings then appear dark on a white ground, and it is not necessary to have light incident from a mirror below. When an immersion system is to be tested, the observation is made in the same way except that, in this case, a drop of the liquid is first inserted between lens and cover-glass. To fix the diameter of the rings of this apparatus before they are actually scratched on the plate, a determination of the exact thickness of the glass plate and its refractive index must first be made. As found by the microscopical method, the first was 6.13 mm., the second = 1.525. The rings are arranged at intervals of 5/100 of the numerical aperture. The data, for example, for an aperture of 0.80 are as follows:—

$$0.80 = 1.525 \sin \chi,$$

whence the angle in the glass  $\chi = 31^\circ 38'$ , but

$$\tan \chi = \frac{r}{6.13},$$

from which is deduced the radius of the ring in question  $r = 3.777$ .

The angle of divergence  $\alpha$  in air, since

$$n \sin \chi = \sin \alpha$$

is

$$\alpha = 53^\circ 7'.$$

The double amount  $106^\circ$  is therefore the angle of aperture corresponding to the numerical aperture 0.80 mm. The radii for the numerical apertures up to 1 would be as follows:—

0.80	3.777 mm.
0.85	4.115
0.90	4.481
0.95	4.881
1.00	5.324.

The plate contains in this way rings increasing in diameter up to the aperture

$$1.40 \quad 18.820 \text{ mm.}$$

For greater distinctness, at certain intervals, two circles close together are drawn instead of one.

In testing the oil-immersion system previously referred to, the fifth reckoned from the ring corresponding to the aperture 0.80 fell on the edge of the field of view. It has, therefore, at most, the numerical aperture 1.00, whereas in the price list of the firm it was called 1.25. This was a great discrepancy, for if the system had really possessed the latter aperture, five more rings ought to have been seen. The numerical apertures necessary for the resolution of different diatoms are given in Dippel's text-book of general microscopy in the tables of comparison which have been established by exact scientific observations. On

reference to these tables, the data referring to 1.00 were found to be *Nitzschia curvula* and *Navicula rhomboides* (*Frustulia*) var. *saxonica* 36 striæ in 1/100 mm., while for the resolution of *Amphipleura pellucida* with 40–42 striæ, a system of 1.10–1.15 was found to be necessary.

Long before the use of the apertometric process, Herr Kayser had informed the maker of the system that he fixed the resolving power at 34 striæ from the fact that *Nitzschia curvula* was not resolved, and that *Frustulia* showed striæ first on the edges. The maker, ascribing the non-resolution to the mounting of the preparation, at the same time sent preparations which really were resolved. The striation of these, however, only amounted to 26 and 24 to 30 respectively, while *Amphipleura pellucida* was not forthcoming, because they were "at present not of good quality."

Dippel's work shows with what exactness the productions of microscopical forms can be apertometrically rated, in a way quite analogous to the determination of size by the scale. The action of an optician therefore who sells an objective system having a less aperture than it professes to have, must be compared to the behaviour of a tradesman who supplies goods deficient in quantity.

An advantage is now to be considered which the apertometer ring method possesses over that of Abbe. In the latter method a pointer is turned round on a polished glass cylinder until it appears to come on to the edge of the aperture. In this way the aperture is tested only in a certain diameter. By the author's method the whole range is seen at a glance, and any defects can also be noted. It is interesting that in the present dry system No. 7, the rings do not appear to be exactly concentric, but in a certain diametral direction on one edge there are broad intervals, on the opposite narrow ones, so that for the clear definition of the first, a further pressing in of the eye-piece is necessary. This asymmetry can be also recognized by the first method in the change in adjustment of the eye-piece, and out of the difference of the horizontal angle.

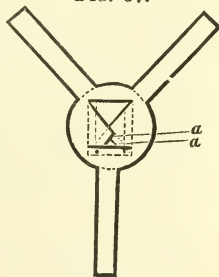
#### PLAXTON, J. W.—A Camera Lucida for nothing.

"The other day, after a morning's work, something went wrong with the prism of my camera lucida, and, do what I would, I could not bring it back to usefulness. At a loss for the moment, I cast about for a substitute, and in half-an-hour, with penknife and pencil, out of a piece of stiff paper and a square of thin glass, had turned out a fragile but efficient substitute for what is known in catalogues as 'Beale's Neutral Glass Reflector,' price 6s.

"This is how I did it—Describe a circle by standing the eye-piece of the Microscope on the paper and running a pencil round it; inscribe a square in the circle already drawn by drawing the pencil along the edges of the square of thin glass you intend to use; now lay down the diagonals of the square; draw three other lines within the square, each one parallel with a side of the square, and each, say 1/8 inch from the side; draw two other short lines (*a a* in the diagram, fig. 57) parallel to the diagonals.

Take the penknife and, following the continuous lines of the diagram, cut through the paper: you will have in paper what resembles a three-spoked

FIG. 57.





wheel without tire. The upper triangle of the four within the square falls away as useless; the lateral triangles open outwards, and stand at right angles with the plane of the circle; the little flanges on their lower edges are made by creasing the paper to support the thin glass. The base of the lower triangle answers the same purpose.

"Put the eye-piece in the Microscope, the circle of paper to the end of it; turn the spokes of the wheel back along the tube, and slip a tiny elastic band over them, or tie them with a thread; a little manipulation with the fingers, the thin glass is in place, and the thing is done.

"Need I say that any one can see that it would be almost as easy to use a piece of thin sheet brass or other metal as to use paper?"]

*Journ. of Microscopy*, III. (1890) pp. 40-1 (1 fig.).

#### (4) Photomicrography.

**Some Experiences in Photomicrography.**\*—"More than a dozen years have now elapsed since I made my first photomicrograph; at that time the successful workers in this country could be counted on one's fingers, and the old messy wet process and expensive appliances were regarded as indispensable for good results. The development of amateur photography following the general introduction of dry plates could not fail to influence photomicrography, since few microscopists viewed with indifference the placing at their doors of a ready means of recording observations. The experience gained during ten years of continuous work, covering almost every class of subjects, with amplifications ranging from four to four thousand diameters, has, naturally, emphasized many facts; the possibility that a brief statement of these conclusions may aid younger workers in this field must be my apology for the egotism of these remarks.

While appreciating fully and endorsing heartily the efforts of those working with no greater responsibility than their individual enjoyment, it is rather to those seriously engaged in endeavouring to produce the best and highest class of this work, that these suggestions are offered.

The three essential conditions for success in photomicrography are:—(1) Satisfactory apparatus; (2) Good illumination; (3) Suitable preparations.

Satisfactory apparatus by no means implies elegant appliances, but adaptation to purpose; so that the Microscope be very solid and firm, and supplied with a substage to which a condenser can be attached and satisfactorily adjusted, and that the camera be of sufficient length, it matters little as to exact form or detail; for high powers the mechanical stage is a convenience, and for extreme amplifications (2000 and over) well nigh a necessity. The most complete photomicrographical outfit to be had is, undoubtedly, the one made by Zeiss, of Jena. . . . Of greater importance, however, is the quality of the objectives, for only those of the most perfect correction will stand the severe test of photography. While there are many others, which personal use has shown to answer well, my experience would lead me to select the following as being especially satisfactory:—3 in. Crouch, 1½ in. Beck, ¾ in. Bausch and Lomb, 4/10 in. (BB) Zeiss, 1/4 in. "Photographical" Bausch and Lomb, 1/6 in. (DD) Zeiss, 1/12 in. oil-immersion apochromatic Zeiss. The 3 in. Crouch and the 4/10 in. (BB) Zeiss deserve especial mention,

\* By George A. Piersol, M.D., of Philadelphia. *Amer. Annual of Photography*, 1890.

as for crisp definition over an entire perfectly flat field they are unsurpassed; for high amplifications the new apochromatic oil 1/12 Zeiss is superb.

The question, whether to receive the image directly from the objective on the plate, or to employ some means to project the image, has received of late much attention. While fully appreciating the theoretical objections to the direct image, I confess that for low and medium powers I continue to use it by preference, as the photographs so obtained fully equal in every respect any which I have ever seen made by the indirect mode. With high amplifications (1000 diameters and over), the conditions are greatly changed by the approach to the limit both of the shortness of the focus of the objective and of the length of camera which can be advantageously used; my experience leads me to adopt the 1/12 in. objective as the one, and not over four feet as the other limit, since any given high amplification, say 2000 diameters, can be more satisfactorily and more conveniently obtained with a superior 1/12 in. connection with suitable optical means to increase the initial magnifying power of the objective than with an unaided 1/25 lens and the plate removed to a great distance. Until quite recently the various amplifiers offered the best means of increasing the power of an objective, but the introduction of the "projection-oculars" of Zeiss has given us an accessory for this purpose far superior to the older devices. These projection-oculars resemble the ordinary microscopical oculars, or eye-pieces, only in general form and in name, being optically a projection-objective in connection with a collecting lens. The new oil-immersion apochromatic lenses, in combination with these projection-oculars, form, undoubtedly, the most efficient equipment for high-power work, and have but one drawback—their cost. It is, unfortunately, as true for high-power photography as for microscopical observation in general, that the best results are to be obtained only with fine, and necessarily expensive, optical appliances. If for the satisfactory study of the intimate structure of a cell, or of a micro-organism, the most improved immersion lenses are necessary, it is to be expected that for the successful photographing of the same, tools at least as good are needed. The complicated mechanical arrangement for controlling the focusing adjustments from a distance, may usually be replaced with advantage by the simple contrivance of cords and weights, devised by the writer more than a dozen years ago, which has been so generally adopted in this country; during the extended continued use of this little device, it has never been found wanting, responding perfectly to the severe demands of the highest amplifications. A modification for the coarse-adjustment, having pulleys and very heavy weights, serves equally well when very low (2 to 5 in.) lenses are used. A very stiff spring in the fine-adjustment may sometimes require increased friction to prevent the cord from slipping, the necessary traction being obtained by heavier weights, or by taking an extra turn of the cord about the milled head of the micrometer screw.

My conclusions regarding the second of the necessary conditions—good illumination—are briefly stated; after many experiments with various kinds of artificial illumination, and after the examination of innumerable specimens of the best work of acknowledged experts, while, of course, admitting that good photographs can be made, under suitable

conditions, by these means, yet I am fully persuaded that sunlight is by all odds the best, and, for high powers, the only really satisfactory illumination by which to make photomicrographs that are satisfactory as photographs, as well as records of microscopical observations. That even by good lamplight fair impressions of objects under extreme magnification can be obtained, no one questions, but the negatives produced by such illumination seldom, if ever, possess the characteristics of a really good sunlight negative, where the sharpest details are combined with an exquisite softness and harmony of half-tones. That a photomicrograph should be a silhouette of deep shadows and chalky whites, is a proposition to which I could never subscribe. Sharpness and vigour are, of course, the first essentials in a photomicrograph, but there seems to be no reason that in such a picture all the qualities of a good photograph should not be represented. An almost identical opinion regarding the advantages of sunlight, has been reached by Dr. R. Zeiss,\* after a most exhaustive series of experiments with artificial illuminations of all kinds, stimulated by the hope of finding a satisfactory substitute for sunlight, the uncertainty of which, during the greater part of the year, is even a greater inconvenience in Germany than with us.

The third condition for good work—suitable preparations—though last, is by no means least, for all apparatus and illumination avail but little when proper preparations are wanting. Thanks to our present microscopical technique, these are readily obtained, since extremely thin and well-stained preparations of vegetal and animal tissues are now matters of everyday production. The thinness with which sections are now usually cut ( $\cdot 005$ – $\cdot 01$  mm.) often renders them, when stained with the staple carmine dyes, too actinically transparent to photograph well with very low powers. The interposition of some ray-filter readily overcomes this; during the last three years a screen of yellowish-green glass has been in constant use, with the most satisfactory results, yielding plucky pictures of objects entirely too transparent to produce sufficient contrasts in the negatives; the exposure, however, is increased about three to five times, but this, even when thus lengthened, seldom exceeds 20–25 seconds, on Carbutt's "B 12" plates. Where great differences of colour are present in the same preparation, or where certain unfavourable tints, as deep brown, prevail, the orthochromatic plates offer decided advantages; for, however, ordinary preparations with but one stain, the colour-screens, when judiciously selected, will yield equally good pictures, with a gain in economy, convenience, and certainty. The modified hæmatoxylin stains, producing browns and slate-blues, are very valuable for special purposes, but require some considerable technical experience for their successful production.

What has been written may appear to discourage the undertaking of this most fascinating branch of photography, where the primary object of instructive entertainment does not warrant the acquisition of the class of appliances above recommended; this should not be so, as the full force of these suggestions applies only to those whose work in this line necessitates the use of the higher amplifications, with the view of producing the highest possible results."

\* 'Special-Catalog über Apparate für Mikrophotographie,' Jena, 1888.



To the amateur, who has been using but lamplight for his exposures, it is suggested that he avail himself of some bright "off-day" to give sunlight a trial. If the mirror of the Microscope be of good size, it will be only necessary to make an arm on which to support the removed mirror outside some southerly exposed window, since it is desirable to have much more distance between the mirror and the stage than would be possible were the mirror attached in its usual place. Where the Microscope mirror is too small to be satisfactorily used, a rectangular wood-framed looking-glass is readily mounted with the aid of a few strips of wood, so as to turn about both axes.

The rays from the plane side of the mirror are passed through a condensing lens (of 8-10 in. focus, if possible), so placed that they are brought to a focus before reaching the plane of the object. The exact position of the condensing lens is a matter of experience; usually, however, the most favourable illumination is obtained at that point where the field is still *uniformly* illuminated, just before the rays form an image of the source of light; the nearer the rays are focused, the less disturbance from diffraction rings. Ordinary objectives will require the employment of monochromatic light—produced either by a deep blue solution of ammonio-sulphate of copper, or by the green glass screen already mentioned—since the optical and actinic foci do not usually coincide. Powers up to the  $\frac{3}{4}$  in. will require no further condenser; with the  $\frac{1}{4}$  or  $\frac{1}{6}$  objectives, the low power ( $\frac{1}{4}$  or  $\frac{3}{4}$  in.) serves with advantage as an achromatic condenser, when attached to the substage. The Abbe condenser, although so important for refined microscopical investigation, is not adapted to photography unless a very wide cone of light is desired, which, for the majority of preparations, is a decided disadvantage; a low-power objective, used as a condenser, will generally be found more satisfactory than the Abbe with a small diaphragm.

The simple apparatus indicated, when properly handled, will produce excellent work with such powers as the amateur is likely to employ; focusing the image by the monochromatic light, and avoiding over-exposure, being the points especially requiring experience. When it is remembered that seconds, with very slow plates, usually suffice for the minutes with rapid ones of an exposure by lamplight, the intensity of the actinic power of the sunlight will be somewhat appreciated. Some simple arrangement, by which the rays from the mirror may be cut off with sufficient rapidity, will suggest itself; an effective one is a small shutter, turning at one end on a screw and covering a circular opening in a board, through which the rays from the mirror pass; the rapidity with which the sun's image from a fixed mirror becomes decentered necessitates a readjustment of the light just before each exposure, but the patience thus exercised will be more than repaid in the character of the resulting negatives.

**Microphotographs of Wood Sections.**—An interesting communication on this subject was recently made by MM. Thil and Thouronde to the French Photographic Society. Microphotographs to the number of about four hundred were executed to the order of the Minister of Agriculture. M. Thil, Inspector of Government Forests, has, in very precise language, pointed out the reach of this application of photography, which permits of the classification of woods in families



and species, thanks to the comparison alone of the intimate structure of the fibres and cellular network. By this means we are enabled, with the help of simply thin cuttings, to give, so to say, a complete anatomy of each species, and to notice easily the essential differences which exist between woods of different species, although belonging to the same family; all the more, therefore, can we recognize classification in families. Microphotographic pictures, projected by the lantern, served to demonstrate clearly the truth of the propositions affirmed. This is a new example of the numerous services that photography may render to the sciences.

**The Coloured Screen in Photomicrography.\***—The following is an abstract of a paper by Professor Romyn Hitchcock :—

An ordinary gelatino-bromide plate is sensitive to the spectrum of sunlight from a point between the Fraunhofer lines E and F to about K. The maximum photographic action is about G. By considerably prolonging the time of exposure the limit of photographic action at the red end of the spectrum is greatly extended. In practice the light below the green of the spectrum may be regarded as quite inactive when we take photographs with ordinary plates.

By introducing a coloured screen—a plate of yellow glass for example—in the path of the light, we may absorb the more active rays, and prolong the time of exposure until the yellow rays have time to act upon the sensitive plate. In practice, however, it is found that there are two difficulties about this method of procedure; first, in obtaining a satisfactory screen, and second, in the long exposure necessary when working with the comparatively inactive rays.

With colour-sensitive plates, such as are now in general use abroad and gradually being introduced in this country, the range of photographic action towards the red is greatly extended. With such plates the yellow screen can be used with great advantage.

A few years since it was customary to work with monochromatic blue light in photomicrography, and the ammonio-sulphate of copper blue cell was much in use. When colour-sensitive plates were introduced yellow screens took the place of blue, because it was found that many specimens had yellow and red and brown parts which were not well photographed with blue light.

The colour and thickness of the screen both require attention. If it be too thin the blue light is not sufficiently cut off. In particular cases an almost monochromatic yellow light is desirable, as when it is desired to obtain sharp outlines of deeply stained objects regardless of structural details. But generally a rather broader spectrum range is desirable, for the light employed should correspond to the different colours or shades of colour of the object. It is owing to neglect of this consideration that we often see photomicrographs which are mere silhouettes, while the objects show much more structure to the eye. This is frequently observed in photographs of such structures as the tongue and sting of a bee, and legs of insects. In other preparations, in which the colour is a stain, brown or red for example, the fault lies partly in the exposure, which, in many cases, is insufficient to give more than

\* Amer. Mon. Mier. Journ., xi. (1890) p. 8.

outlines and blank interiors. This is frequently noticeable in photographs of bacteria.

By a proper choice of a screen, if a screen is required, a photograph should show any object as clearly as we can see it in the Microscope.

Colour-sensitive plates may be said to be indispensable in the photography of rock-sections with polarized light.

The yellow solution devised by Professor Zettnow, of Berlin, is used with much favour by many workers. It is composed as follows:—Copper sulphate, 175 grm.; potassic bichromate, 17 grm.; water, 1000 cem.

The true function of the colour-screen should be to give definition and detail, not to increase contrast between the object and the field, as many observers seem to believe.

#### (5) Microscopical Optics and Manipulation.

**Amplification in Micrometry.\***—My attention has quite recently been drawn to this subject in connection with the celebrated Dr. Cronin case. It may be taken for granted that one cannot measure what he cannot see. But how high an amplification is necessary in a given case is a matter of much importance. In the measurement of blood-corpuscles in medico-legal cases the late Dr. Richardson advocated the use of a very high power, viz. a  $1/25$  or  $1/50$  objective. In my own measurements of blood-corpuscles I have, out of respect to authority, always used a high power, from 1500 to 1800 diameters. Recent experience has, however, qualified my views upon the subject, and in the case of the comparison of the ultimate subdivisions of a micrometer, ruled on metal, I am now of opinion that practically the same result may be obtained by the use of a  $1/4$  objective as with a  $1/18$  or  $1/25$ .

In December 1885, I commenced the investigation of the  $1/100$  mm. spaces of "Centimeter A"; but was unable to finish it. Two series of measurements were then made with a Bausch and Lomb opaque illuminating objective, and a Bulloch filar micrometer. Recently I have measured the same spaces with a Spencer  $1/10$  and  $1/25$ , and with a Zeiss  $1/18$ . The results of these measurements are given in the table below, each correction being the mean of from three to twelve readings of the filar micrometer at each end of the measured space.

It will be observed that the agreement between the several series of the writer, and the results obtained by Prof. Hilgard is quite close, the discrepancy being practically insensible.

Provided the amplification is sufficient to render the object to be measured of a sensible size, and to render the difference between the sizes of two objects visible, my own judgment is that little, if anything, is gained by the use of a power so high as to impair the definition, even though such impairment be but slight. Quite as much, in other words, is lost by impairment of definition as is gained by increase of amplification. The practical conclusion then is that no higher power should be used than is consistent with perfect definition.

**Diffraction Rings and Diffraction Spectra.**—There appears to be still some confusion between the diffraction "spectra" of the Abbe theory and the diffraction bands or fringes and spurious lines seen

\* By Hon. Marshall D. Ewell, LL.D.

surrounding the outlines of all objects in the field of the Microscope, when the illumination is obtained by somewhat narrow but sufficiently bright beams of light, especially with high powers or deep eye-pieces.

The latter are true diffraction bands, originating from the diffraction of the light at the object, but the difference between the two phenomena is that the spectra represent the diffraction effect of the object at a very distant plane, conjugate to the posterior focus of the objective, whilst the "bands" or "fringes" show the diffraction effect of the same objects in a plane close by, i. e. in the neighbourhood of the objects themselves. Nägeli and Schwendener, it is true, deny that these fringes are diffraction phenomena, and explain them as interference phenomena in a somewhat complicated manner, but Prof. Abbe considers that he has established the incorrectness of their views on this point, except so far as they assert that the phenomena cannot be due to the diffraction effect of the lens opening, as had previously been assumed by Helmholtz and others.

#### (6) Miscellaneous.

**The 300th Jubilee of the Microscope.\***—"B. C." writes:—Natural science enters this year on a memorable anniversary, the 300th Jubilee of the Microscope, one of the most powerful of its resources. To this instrument is due in great measure the wonderful impulse given to science in the second half of this century. The importance to which the Microscope has attained in scientific investigations is well known. It has become an absolutely indispensable instrument to the zoologist and botanist, to the mineralogist and geologist, to the astronomer and the physician. The Microscope has effected a complete revolution, and has diverted the direction of study into the most varied channels. In fact it has created a new method of research, such as histology. On the healing art the Microscope has exercised a most beneficent influence; for while it explained the changes undergone by the finest tissues in the various diseases—it was on microscopic observation alone that Virchow founded his renowned system of cellular pathology—it pointed out at the same time the means of healing them. The Microscope has also been of wonderful service in technical matters. Before attaining its present high degree of perfection, the Microscope had to pass through a number of intermediate stages which it is of great interest to look back upon on this its 300th jubilee. . . .

It is strange how slowly the Microscope found its way into learned circles. It was only when Leeuwenhoek had by its aid discovered the infusoria that it became generally used in the scientific investigations of anatomists and physiologists. What it has accomplished since that time constitutes the glory of the natural sciences. The Microscope soon passed from the workshops of the spectacle-makers to those of the optician, by whose skill it has undergone, little by little, numerous changes, corrections, and improvements. Not to mention all of these, it will suffice to point out the arrangement of the transmitted light (1685), of the reflecting illuminating mirror (1715), and the use of achromatic and aplanatic objective lenses (1824). In more recent times the Microscope has received further improvements, which have cast into the shade all conceivable expectation; and unless appearances deceive us the finer

\* Central-Ztg. f. Optik u. Mechanik, xi. (1890) pp. 69-70.



mechanics of Microscope construction have not yet reached the limit of their capabilities. The latest acquisition of medical science, the bacteria, has put the greatest demands on the Microscope, and reveals to this instrument the deepest secrets of nature. Let it be the aim of science to gather in a still richer harvest by the aid of the Microscope!

**The Microscope banished.**—The following appears in the *Daily News* of the 9th April:—"An interesting paper by Mr. Bothamley in *The Photographic Quarterly* reminds us of the important part now played in education by the optical lantern which in the memory of so many among us was a mere toy for the entertainment of juvenile parties. The initiation and growth of the system is mainly due to Professor Miall, of the Yorkshire College, Leeds, in which important institution almost every department has its lantern, and such widely different subjects as biology and engineering, ancient history and textile industries are alike illustrated by this convenient means. In the biology lectures the lantern is said to have well nigh banished the Microscope, thereby effecting a great saving both in cost and time (!) The production of lantern slides is found to be most easily and rapidly done by photography. Original objects, drawings, large photographs, illustrations in text-books, can all be reproduced in the same way. At the Yorkshire College the number of slides required by the various departments is stated to be so large that the whole time of a special photographic assistant is occupied with their production, although the work is much facilitated by the ingenious copying camera devised by Professors Barr and Stroud. But perhaps the most remarkable fact in connection with this subject is Professor Miall's discovery of how the lantern may be used in illustrating lectures in a room illuminated by daylight."

**Miss V. A. Latham, F.R.M.S.\***—This lady has recently been elected to the chair of Demonstrator in Pathology in the University of Michigan. Professor Latham is the first lady who has held any office in the Medical Department of the University, and has our congratulations and best wishes for her success.

### B. Technique.†

**BÖHM, A., u. A. OPPEL.**—*Taschenbuch der mikroskopischen Technik.* (Handbook of microscopical technique.)

München (Oldenbourg), 1890, sm. 8vo, 155 pp.

**GORONOWITSCH, —.**—*Kurze Uebersicht über die Fortschritte in der mikroskopischen Technik im Jahre 1888.* (Short review of the progress in microscopical technique in 1888.)

*Medizinsk. Obzrenenije*, 1889, No. 8 (Russian).

**KAHLLEN, C. VON.**—*Technik der histologischen Untersuchung pathologisch-anatomischer Präparate.* Für Studierende und Aerzte. Ergänzungsheft zu Dr. E. Ziegler's Lehrbuch der allgemeinen und speciellen pathologischen Anatomie. (Technique of the histological examination of pathological-anatomical preparations. A supplement to Dr. E. Ziegler's Handbook for the use of Students and Physicians.)

6th ed., Jena (Fischer), 1889.

**POLI, A.**—*Note di microtecnica.* (Notes on microtechnique.)

*Malpighia*, III. (1889) June, August, December.

\* Amer. Mon. Micr. Journ., xi. (1890) p. 10.

† This subdivision contains (1) Collecting Objects, including Culture Processes; (2) Preparing Objects; (3) Cutting, including Imbedding and Microtomes; (4) Staining and Injecting; (5) Mounting, including slides, preservative fluids, &c.; (6) Miscellaneous.



## (1) Collecting Objects, including Culture Processes.

**Procuring and Preparing Protista found in the Stomachs of Ruminants.\***—To obtain Protista from the stomachs of oxen, says Dr. A. Fiorentini, it is merely necessary to open that viscus with a knife, and gather some of the gastric juice in test-tubes. In order to keep the animals alive it is advisable to keep the tubes immersed in water at a temperature of 30°–35°. To examine these Protozoa alive, it is necessary to make use of Schultze's or Ranvier's hot-stage, so that the slide may be kept at 35°. But the following method has the advantage of simplicity. Heat the slide over a spirit-lamp until it becomes warm. Then place thereon a drop of the fluid containing the animals to be examined, and cover with the cover-glass. Next with a pipette take some boiling water and drop it in lines on the slide, taking care, however, that it does not mix with the fluid under the cover-glass. This device will keep the preparation warm sufficiently long to examine the Protozoa alive. When cold a new preparation must be made.

For fixing the animals, the author used a 1 per cent. osmic, and for staining the nuclei and nucleoli fuchsin, alum-carmin, and alum-cochineal. Glycerin and Canada balsam were used for clearing up the preparations when osmic acid had blackened them or made them obscure.

**Useful Collecting Device.†**—Mr. J. Walker finding his collecting bottle, a modified Wright, somewhat cumbersome, "decided to use a smaller bottle, and have the strainer (I use bolting silk 10,000 to the inch) outside instead of inside. I therefore procured a bottle holding about 4 oz. A square bottle with a wide mouth is preferable, though a round one will answer well. I bored four holes opposite each other, 1 in. above the bottom and about 3/8 in. in diameter, and enlarged the openings in a direction parallel with the length of the bottle, until within an inch of the neck. Over these four oblong apertures I cemented fine bolting silk or other desirable material with shellac, and when dry, the bottle was ready for use. To those not having the tools needed for drilling glass, I would recommend a small tin can or box, such as that in which Colman's mustard is sold, or the common round pepper-box obtainable from the grocery stores, the lid making a good coarse strainer.

In working with it, the currents of water passing through the meshes of the strainer will cause fine debris to collect on the inside, which in this case is easily kept clean with a small brush, a piece of wood, or a stalk of grass. The concentrated material will be found at the bottom of the vessel, and can be transferred to another small bottle carried for the purpose."

**Collecting-bottle for Rotifers.‡**—Mr. A. Pell remarks, "Here is the 'boss' collecting implement at last. Take one of the new lard bottles which hold a quart, the mouth being about 4 in. across, with a metal cover that screws to the neck, and a handle by which it is readily carried. Make a tube of muslin or of linen, in any desirable

\* Journ. de Micrographie, xiv. (1890) pp. 15–6.

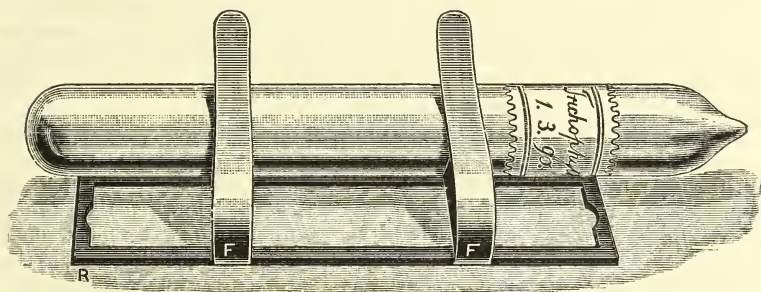
† The Microscope, ix. (1889) pp. 372–4.

‡ Op. c., x. (1890) p. 151.

fineness, and about 2 feet long, 4 in. in diameter at one end, and 2 at the other. Fasten a tin ring to the small end and attach the large end to the mouth of the bottle. Then put on your rubber boots and go to the pond. There pour the water into the small end of the muslin tube, holding it up for that purpose, the bottle hanging below. It will rapidly strain out the Rotifers, &c., which will finally get down into the bottle, and as the muslin tube has so large a surface the water will run through quickly, all solid matters collecting in the bottle. Less is lost by the use of the muslin tube than by a funnel-shaped strainer, and the cloth will not become clogged."

**Test-tube Holder for Microscopical Investigations.\***—Dr. D. von Sehlen has invented a test-tube holder, the advantages of which are mainly its stability and simplicity. Hence it will be found of great use in the cultivation of the various forms of Fungi, and also for photo-

FIG. 58.



graphic purposes. The apparatus consists of a flat oblong frame R which supports two uprights, placed equidistant from the ends of the frame. In these uprights a triangular piece is cut out in order to put the test-tube in, and the latter is kept in position by the two spring-clamps F. The distance between the two spring-clamps is enough to allow sufficient space for the objective to work in, and the length of the frame such that it is easily clamped to the Microscope-stage. It is hardly necessary to explain that the test-tube is easily moved round its short axis, and pushed up and down, so that when on the Microscope-stage it is easily illuminated from below.

**Preparation of Nutritive Agar.†**—Dr. V. A. Moore writes:—"The extent to which nutritive agar is employed in the cultivation of Bacteria renders it of much importance that its method of preparation should be made as perfect as possible. When it is prepared after the method recommended in works on bacteriology (which is practically the same as that first formulated by Koch for the preparation of solid culture media), a medium is obtained that favours the growth of most germs. In this respect the method is desirable, but in regard to the other

\* Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 17-20 (2 figs.);

† Amer. Mon. Micr. Journ., xi. (1890) pp. 115-7.

requisites of a satisfactory solid medium it is quite deficient. The objections to the method with reference both to the process itself and the character of the resultant agar are three in number. (1) The difficulties attending the filtration of the agar. This process alone often requires a very considerable length of time besides the use of a hot filtering apparatus that must be provided especially for this purpose. (2) The presence in the sterile agar of a flocculent precipitate that is invariably thrown down during the process of its sterilization, and which greatly interferes with its usefulness, especially in making roll and plate cultures. (3) The variation in the consistency of the agar. It is impossible to obtain this material of the same consistency, as the agar is only partially dissolved, even after long boiling, in the simple beef-infusion. The coagulation of the albumen ensheaths the stems of agar, floats them to the surface where they remain imbedded in the firm, albuminous coagulum. This property of the agar is worthy of consideration, for with the varying consistency of the medium a consequent change follows in the character of the growth of most germs.

For the purpose of securing a process for the preparation of nutritive agar that was free from the above mentioned difficulties I have reviewed carefully the method of Jacobi,\* Von Freudenreich,† and Cheesman,‡ in all of which I found difficulties that were equally as objectionable as those possessed by the original method.

The use of a solution of beef-extract in distilled water, instead of the simple beef-infusion made directly from the fresh meat, was also tried, but the agar thus prepared did not favour as vigorous a growth of many germs as when prepared from the fresh meat-infusion. So feeble was the growth of many germs upon this agar that the method was abandoned, although very satisfactory in other respects.

In the course of this experimental work it was found that when the stems of agar were cut into small pieces and boiled in a fluid containing no coagulate material, that it was entirely broken up and the soluble portion dissolved. The insoluble particles that remained suspended in the liquid were easily and completely removed by the addition of egg albumen, and subsequent boiling and filtering. From these facts a method for the preparation of nutritive agar was derived, which consists in first preparing the neutralized beef-infusion-peptone, and thus getting rid of all coagulable material before the agar is added. This process is effective in greatly diminishing the time and attention required for the preparation of this medium. The medium can always be made of the same consistency, as all of the agar that is added is dissolved. It remains free from precipitates when sterilized, and its nutritive qualities are as favourable to bacterial growth as when it is prepared after the original method.

(1) *The preparation of the beef-infusion-peptone.*—The method of preparing this liquid is practically the same as that already in use in most laboratories. Finely chopped or ground beef (freed from fat) is macerated in distilled water for from 12 to 18 hours in a cool place. The distilled water is added in the proportion of 200 ccm. to each

\* Centralbl. f. Bacteriol. u. Parasitenk., iii. (1888) p. 538.

† T. c., p. 797.

‡ American Naturalist, xxii. (1888) p. 472.



100 grams of beef. On the following day the liquid is separated from the meat by straining it through a coarse linen. The simple beef-infusion thus obtained should be equal in quantity to the amount of water added; if it is not the deficiency can be restored by the addition of distilled water. To the beef-infusion is added 1 per cent. peptone, 1/2 per cent. sodium chloride; and if it is desirable to make it alkaline, a sufficient quantity of a normal solution of sodium carbonate to give it a weak alkaline reaction. The liquid is then boiled for thirty minutes in a water-bath, cooled, filtered, and distributed in Erlenmeyer flasks plugged with cotton-wool. If only a small quantity of agar is to be made at once, 250 ccm. is found to be a very convenient quantity to put in each flask. It is then sterilized by boiling for one hour each day for three consecutive days. It need not be sterilized if it is desirable to prepare the agar at once. As the beef-infusion-peptone is also employed as a liquid medium in the cultivation of bacteria, very little time is lost in preparing an extra quantity of this liquid to be used in making the agar.

(2) *The preparation of the agar.*—To an Erlenmeyer flask (a glass beaker or agate or iron vessel may be used) containing beef-infusion-peptone, as prepared above, 1 per cent. of *very finely chopped* agar is added. The flask is then placed in a water-bath and boiled vigorously for two hours. At the end of that time the agar is dissolved, and the liquid is allowed to cool. When a temperature of 40–45° C. is reached, the white of egg is added in the proportion of one egg to 250 ccm. of the liquid. After the albumen is *thoroughly* mixed with the liquid agar it is returned to the water-bath and again boiled for two hours. It is of much importance that the albumen is evenly distributed throughout the mass before it is coagulated. It is now ready to be filtered. The egg albumen is coagulated in very firm masses, leaving the liquid perfectly clear. The coagulum is removed by filtering the liquid through fine Japanese filter-paper or a layer of absorbent cotton, as a 1 per cent. solution of the agar does not pass readily through ordinary filter-paper. Should a weaker solution of the agar (1/2 to 3/4 per cent.) be desired, its filtration can be accomplished by the ordinary method. A hot filtering apparatus is not necessary. The clear filtration is now ready for distribution in sterile cotton-plugged tubes.

The agar is sterilized by discontinuous boiling in a closed water-bath for three consecutive days. If small tubes have been used containing not more than 7 ccm. each, five minutes' boiling each day is sufficient. If larger tubes are used, they should be boiled for a longer time. Or it may be sterilized by steaming each day for from five to ten minutes after the agar has become liquefied for the same number of days. After its sterility has been tested by allowing it to stand in an incubator for several days, it is ready to be stored until required for use. It has been customary in this laboratory, in order to prevent the evaporation of the agar by long standing, to dip the lower end of the cotton-plugs in hot sterilized paraffin, and to store the tubes in a cool, moist chamber."



## (2) Preparing Objects.

**Preparation of Crustacea.\***—Dr. O. vom Rath gives an account of the method he adopted in his investigation into the structure of the Cymothoid Crustacean *Anilocra mediterranea*. The heads were cut off with a sharp pair of scissors and immediately placed in picric-nitric acid, picric-sulphuric acid, warmed absolute alcohol or chrom-osmic-acetic acid; the first of these reagents gives especially good results. The hardened heads were stained *in toto* in alum-carmin or borax-carmin. Paul Meyer was quite right in urging that the mere preservation in alcohol of Crustacea or other Arthropods with a strong chitinous membrane is quite insufficient.

**Modes of Studying Segmental Organs of Hirudinea.†**—M. H. Bolsius did not learn much by dissecting out the segmental organs and mounting them entire. It is better to cut sections of the entire animal, or, when it is large, of parts. Transverse, vertical, longitudinal, or horizontal longitudinal sections should be made. To prevent contraction of the body, large specimens should be anæsthetized before being killed. Small specimens should be placed in a 1 per cent. (or even weaker) solution of chromic acid. Passable results in the way of fixation were obtained by bichromate of potash, but bichloride of mercury is much more efficient. A saturated aqueous solution or Gilson's liquid may be used. In either case small individuals are placed in them for 15 to 30 minutes; larger pieces must remain a proportionately longer time. Excellent preparations were also obtained with a 2 per cent. solution of nitrate of silver; in this case staining reagents were not used, but with the others a picro-alum-carmin, the formula for which has not yet been published, but which is used at Louvain, was found to give excellent results.

**Mode of Investigating *Hydra fusca*.‡**—Herr K. C. Schneider recognizes that it is only possible to study the nervous system of Hydroids by maceration-processes. It is scarcely possible to recognize in sections the cell-boundaries of the ectoderm, to say nothing of distinguishing them from the separate subepithelial elements. The structure of the cells is considerably affected by the use of paraffin. As a maceration-medium, the author first used pure acetic acid from 1 to 10 per cent; but as this caused deformation of the elements, chloride of sodium was used, and was followed by various strengths and quantities of osmic acid. After some experiments, a mixture of one part 0·02 per cent. osmic acid with four parts 5 per cent. acetic acid was found to give excellent results. Pure osmic acid was found to give very different results from the mixture of osmic and acetic acids. Animals placed for eight days or more in glycerin were very useful in the study of the nervous system. Picrocarmin was found to be the best staining medium, but Beale's carmin and safranin were also of use.

**Microscopical Sections of Tooth and Bone.§**—It was with great satisfaction that we read Mr. J. Howard Mummery's notes on the prepa-

\* Zool. Anzeig., xiii. (1890) p. 232.

† La Cellule, iv. (1890) pp. 374-6.

‡ Arch. f. Mikr. Anat., xxxv. (1890) pp. 322-3.

§ Trans. Odontol. Soc. Great Britain, xxii. (1890) p. 207.

ration of microscopical sections of tooth and bone, in which he gives an account of some new and important discoveries in the structure of these tissues, for it was from this Journal,\* he tells us, that he obtained an account of Dr. L. A. Weil's method of carrying out the balsam process. "I prepared," says Mr. Mummery, "some sections according to these directions, and was so pleased with the results that I have since cut nearly two hundred specimens in this way." It should not be forgotten that this portion of the Journal is of great assistance to those who, like Mr. Mummery, have little time for searching the literature of microscopical technique.

**Preparing Sections of Teeth.**†—Mr. W. A. Hopewell-Smith remarks:—

"(1) The most satisfactory method, in my opinion, of preparing sections showing odontoblasts *in situ* is as follows:—The jaw, preferably the lower, of an embryonic mammal, such as kitten or pup, taken while still in a fresh condition, is carefully stripped of all the tissues covering it, except the oral epithelium and flange of gum, and is placed in the usual standardized solution of Müller's fluid, in order to harden its soft structures, the volume of fluid being about twenty or thirty times the bulk of the immersed tissue. The fluid must be changed every day for four or five days, and then every third or fourth day. The hardening process is to be completed by removing the specimen—which has remained in the Müller's for a fortnight—to alcohol or rectified spirit; and this is to be renewed occasionally until all the colouring matter has disappeared from the specimen and fluid. Vertical sections are then cut by means of a thin sharp knife, and these placed longitudinally on the stage of a Cathcart or Williams freezing microtome, and cut in the ordinary way. Best results are obtained from sections in the canine and bicuspid regions, as here the parts are less likely to be disturbed in the manipulations with the microtome. Imbedding in paraffin and wax, or celluloid, is of little service. The advantages claimed for this method are:—(a) The simplicity of its performance. It will be seen that the hard tissues are not softened by any decalcifying agent, which would materially affect the delicate soft tissues. The knife cuts quite easily the thin cap of semi-calcified dentine and bone, and the elements of the pulp are in no way disturbed in their relation to each other. (b) The odontoblasts are of large size, and easily observable at this period, as their formation of dentinal fibrils is at its highest stage of development. They can be isolated, if thought necessary, by separating with the point of a needle from the surface of the dentine papilla the cap of dentine to which in places they adhere. (c) This method affects little, if at all, the relative positions of dentine, odontoblasts and pulp; and I have found it to be extremely successful.

(2) I should advise your correspondent not to grind down sections of teeth of fishes *in situ*; but to decalcify the jaw and teeth with a 5 per cent. solution of chromic acid or 10 per cent. solution of HCl. After sections have been cut and stained they should be washed well in distilled water, dehydrated for three minutes in absolute alcohol, "cleared" in oil of cloves or xanthol, and mounted in Canada balsam.

\* 1888, p. 1042.

† Journ. Brit. Dental Assoc., xi. (1890) pp. 310-2.

Carmin is the best stain for fishes' teeth. If it is used, however, it is necessary before transferring to distilled water to pass the section quickly through weak  $\text{HC}_2\text{H}_3\text{O}_2$  as this "fixes" the stain. If gold chloride is used the specimens must be mounted in glycerin-jelly. . . .

(5) It is unnecessary to cut sections of enamel to demonstrate the prisms. After having softened enamel by immersion in 10 per cent. solution of  $\text{HCl}$ , remove by means of a needle-point or fine brush a small portion to a slide; put a drop of normal salt solution on to the top of the enamel, and press down cover-glass. Then run a solution of carmine or orange-rubine beneath the cover-glass, and draw off the excess with a little blotting-paper. Wash the stain away further by irrigation with weak  $\text{HCl}$ , or  $\text{HC}_2\text{H}_3\text{O}_2$ , and mount in this solution or acidified glycerin after Beale's plan.

**Examining Nuclei of White Blood-corpuscles.\***—The ordinary notion about white corpuscles, viz. that the majority are polynucleated, is, says M. Mayet, quite erroneous. By this the author does not mean that polynucleated corpuscles are not demonstrable, but that this condition is extremely rare.

To ascertain exactly the shape of the nucleus, glacial acetic acid must be intimately mixed with the blood in the proportion of three to one.

By this means the red corpuscles are rendered almost invisible, while the extra-nuclear part of the white is more or less dissolved, so that the nuclei are isolated and become very visible.

The nucleus then is found to be of very variable shape, and it is owing to this irregularity that various optical effects are produced, so as to give the appearance of more than one nucleus. The nucleoli are always multiple, there being one for each swelling of the nucleus.

When a white corpuscle is really polynucleated, it is just in the act of division, nucleus and extra-nuclear plasma as well, but this condition is rare.

**Studies in Cell-division.†**—Prof. D. H. Campbell recommends the following subjects as specially well adapted for showing the various stages of division in the plant-cell, and its modifications; the paper is accompanied by very good figures:—For cell-division where there is no definite nucleus—*Nostoc*. For division of a multinucleate cell, and division of the nucleus independently of cell-division—*Cladophora*. For cell-division accompanied by the division of the single nucleus—*Spirogyra*. If exposed to cold during the night, and brought into the laboratory in the morning, some of the cells will probably begin to divide almost immediately. An interesting modification of the process is shown by many desmids. For following the process in the living cell—the hairs on the filaments of *Tradescantia virginica*. It is well shown by removing the stamens from the young buds, and mounting the attached hairs in water or in a 3 per cent. solution of sugar. They may be stained without killing them by a weak aqueous solution of methyl-violet, dahlia, or mauvein. For easy demonstration of the process of karyokinesis—the final divisions of the pollen-mother-cells,

\* Comptes Rendus, cx. (1890) pp. 475-7.

† Bull. Torrey Bot. Club, xvii. (1890) pp. 113-21 (2 pls.).



especially of Monocotyledons as *Allium canadense*, or among Dicotyledons *Podophyllum peltatum*. The latter is especially favourable for showing the early stages, because of the small number (about ten) of the nuclear segments.

**Dehydration and clearing up of Algæ.\***—The following method, described by Dr. E. Overton, neither requires complicated apparatus nor demands a great expenditure of time, in obtaining a result more favourable than is usually expected when dealing with such delicate objects as Algæ, which shrivel or crumple up when transferred from one reagent to another.

The object, previously fixed and stained, is placed in a not too large quantity of 10 per cent. glycerin. Here it remains in an open vessel until the glycerin has given off nearly all its water. The objects are then transferred to absolute alcohol. Their further treatment depends on the nature of the clarifying medium. If turpentine, oil of cloves, or the like is to be employed, the object should be placed in a watch-glass, containing a 10 per cent. solution of the oil in absolute alcohol. The watch-glass is placed in a large covered vessel, on the floor of which are some pieces of calcium chloride to absorb the alcohol. In this way the objects are gradually impregnated with the pure oil, whereupon they may be transferred to dilute balsam. If before the objects be placed in the ethereal oil and alcohol mixture, they be passed through chloroform, this step will avoid the too great extraction of the staining by the spirit.

Should xylol be preferred for clearing up, then in the larger vessel pure xylol is placed as well as in the watch-glass. By a process of diffusion the inner vessel will ultimately contain almost pure xylol. By means of this method the most delicate algæ may be mounted in balsam without crumpling.

**Amplification required to show Tubercle Bacilli.†**—When properly stained and prepared, the bacillus tuberculi can be readily recognized with a good  $1/5$  objective and a 2-in. eye-piece, normal tube-length, or, roughly speaking, an amplification of 250 diameters. We do not think that it could be done much below this amplification, though the sharpness of vision of the observer, his acquaintance with the object, and the excellence of his objective would be important factors in settling the question. A  $1/4$  objective with a 2-in. eye-piece, normal tube-length, gives an approximate amplification of 200 diameters.

To be seen and diagnosed for certain, the bacillus tuberculi in urine or water must be prepared for examination by following the well-known technique in such cases (fixing, staining, bleaching, and mounting). No person who has any regard for his reputation as a microscopist would undertake to diagnose for certain bacilli of tubercle from other similar forms existing in water, urine, or any other medium whatever, whether with a magnification of 200 or 2000 diameters. The property of taking certain aniline stains, and retaining them so firmly that even nitric acid, diluted with only three volumes of water or alcohol, will not bleach them, is one peculiar to the tubercle bacillus, and shared, as far as we know, by the bacillus of leprosy only. This test, along with

\* Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 11-13.

† Amer. Mon. Micr. Journ., x. (1889) pp. 277-8; from 'National Druggist.'



isolation and pure culture, alone makes the recognition of bacillus tuberculi certain.

For search of tubercle bacilli and study of the same, we have found a 1/10 homogeneous-immersion objective with a 2-in. eye-piece (approximately 500 diameters) the most satisfactory and least tiring to the eye. A good 1/8, however, with the same eye-piece, should be quite sufficient.

GRANDMAISON, F. DE.—De l'emploi des solutions de chlorure de zinc pour la fixation des éléments anatomiques. (On the use of solutions of chloride of zinc for fixing anatomical elements.)

*Comptes rend. hebdom. Soc. de Biol.*, I. (1889) No. 39.

HOYER, H.—Ueber ein für das Studium der directen Kerntheilung vorzüglich geeignetes Object. (On an object particularly suitable for the study of direct nuclear division.)

*Anat. Anz.*, V. (1890) No. 1, p. 26.

#### (4) Staining and Injecting.

**Practical Notes.\***—Mr. H. M. Wilder writes:—*Picric Acid Staining*.—Picrocarmine is very easily washed out with water, at any rate the picric acid. I prefer for that reason to stand the slide on edge, in order to let it drain off, and finally touch the section (or what else) *on the edge* with blotting-paper or filtering-paper, but I do *not* put the blotter *on top*; even the best, and handled most carefully, will always leave fibres. I then allow the section to dry a little, and finally put on the medium. If in balsam I let the section dry thoroughly; the benzole balsam will soon clear it, without any alcohol or oil of cloves. That is for vegetable tissues.

*To mount Powders*.—In mounting powders I much prefer to breathe on the slide, press it on the dry powder, provided the firmness of the powder is tolerably uniform, give a few smart raps with the edge of the slide on the table, in order to get rid of superfluous powder, put on the cover-glass, with a pencil-brush dust off the surrounding powder, and let the medium *run under* by capillary attraction in the well-known way with a couple of drops on the side of the cover-glass. In this way I seldom have any air-bubbles to contend with.

*Silicate of Sodium* (soluble glass, water-glass) I would strongly recommend as a medium for vegetable sections and powders. It "sets" quickly, less than fifteen minutes after a mount is made; the slide can be cleaned with a nail-brush without fear of the cover-glass coming off. It clears well, and acts as its own cement, no ringing being necessary. Its disadvantages are: it does not agree with alcohol, ether, volatile oils, mucilage, acids (not even very weak), collodion; being alkaline it will colour lignified tissue yellow, and alter the shades of stains more or less (the bluish-purple colour of hæmatoxylin is turned sepia-brown). After some time it deposits "crystals," that is flakes, which, while they detract from the beauty of the slide, cannot well mislead any one; this tendency may, however, be largely obviated by using a mixture of four or five fluid parts of the silicate and one part of glycerin. This mixture is, of course, slow in drying.

*Note*.—Mucilage and water-glass do not well mix, because mucilage is always more or less acid; water-glass is very intolerant of acid.

\* *Micr. Bull. and Sci. News*, vii. (1890) p. 17.

**Staining of Vegetable Nuclei.\***—The following is the method employed by Mr. H. W. T. Wager in staining the nuclei in *Peronospora parasitica*, parasitic on the shepherd's purse (see p. 491). The sections were made by the Cambridge ribbon-section-cutting microtome. The fresh infected tissues of the host-plant were cut up into small pieces, and placed at once either in absolute alcohol or in chromic acid solution, where they were kept until thoroughly penetrated, and were then prepared for imbedding in paraffin-wax. The chromic acid specimens were thoroughly washed in 70 per cent. alcohol, then transferred to methylated alcohol, and finally to absolute alcohol. The pieces of tissue may then be stained *en bloc*, or the separate sections may be stained, when cut, on the slide. The latter gave the best results.

After being thoroughly dehydrated by alcohol, the pieces of tissue were transferred to turpentine for about forty-eight hours, and were then placed in soft melted paraffin-wax for about twenty-four hours, and finally transferred into hard melted paraffin-wax for about two days. They were then imbedded in small square blocks of paraffin, and very thin sections cut by the microtome. These sections were cemented to the slide by a solution of white of egg and glycerin, and the paraffin-wax melted by heating the slide on a water-bath, and washed off in turpentine. The slide was next placed in absolute alcohol, and afterwards transferred to a dilute solution of Kleinenberg's hæmatoxylin in water, made by adding a few drops of the strong hæmatoxylin solution to a beaker of water, until the whole was decidedly coloured. The sections were left in this until they were considerably over-stained, and were then placed in a dilute solution of acid alcohol, made by adding a few drops of strong hydrochloric acid to a beaker of 70 per cent. alcohol for a short time to reduce the stain. They were then washed successively in 70 per cent., 90 per cent., and 100 per cent. alcohol, and were next transferred for a few minutes to turpentine until quite clear and transparent, and were finally mounted in Canada balsam. The preparations thus obtained, which were in many cases only about 1/8000 in. in thickness, exhibited the structure of the nucleus clearly and distinctly.

**Nessler's Ammonia Test as a Micro-chemical Reagent for Tannin.†**  
—Mr. S. Moore writes: In most cases the presence of tannin is immediately shown by all the ordinary reagents used by the botanist for its discovery. This does not happen sometimes, however, as, for instance, in the tannin-cells found in the epidermis on the dorsal side of the leaves of some plants. As a good typical example the common primrose may be cited. Of all the ordinary tests, including iron salts, potassium bichromate, Möll's test (copper acetate and iron acetate), ammonium molybdate, and osmic acid in 1 per cent. solution, the latter alone acts immediately upon the tannin in the primrose leaf's epidermis. It may hence be worth while recording the discovery of a second reagent capable of acting rapidly and effectively; and one which is easily made and will keep for some time should be especially valuable. Such a reagent is Nessler's test for ammonia.

Nessler's test is made, as all the world knows, by saturating a solution of potassium iodide with mercuric iodide, and adding an excess

\* Ann. of Bot., iv. (1890) p. 131.

† Nature, xli. (1890) pp. 585-6.

of caustic potash. Ammonia gives with this a reddish precipitate; tannin a brown, and when in considerable quantity a deep black one; but if little tannin be present, the brown may tend towards purple. It goes without saying that much experiment must be undertaken before one can be sure of the substance giving the brown precipitate being really tannin. To be conclusive, such experiment should be carried out in four different directions:—

(1) The reaction ought to be given in all cases when the ordinary reagents make their presence immediately felt.

(2) Cells which will not immediately give the tannin reaction with ordinary tests, but which will do so with Nessler's test, must also do so under the former conditions if time be allowed.

(3) Tissues, which will not yield the reaction with Nessler's test, must not give it with any other reagent, even after the lapse of some time.

(4) Solutions of tannin must give a brown precipitate with Nessler's test.

Under the first of these headings may be mentioned growing shoots of the garden rose. On laying a radial longitudinal or a tangential section of this in Nessler's fluid, a copious black-brown precipitate is obtained, and the same thing occurs with the beautiful tannin-sacs of *Musa sapientum*. In all other instances, where tannin has betrayed its presence by the use of ordinary reagents, the brown colour has been obtained upon treatment with Nessler's test.

The primrose leaf may be again cited as an example of the time sometimes necessary to show up tannin with the usual reagents, of which it must here suffice to particularize ammonium molybdate. On laying in the molybdate a small piece of epidermis torn off the lower side of the leaf, one first sees a cell here and there coloured the characteristic and beautiful yellow given by this test: these coloured cells are usually situated among the elongated more or less rectangular cells overlying the vascular bundles. Re-examination after half an hour or so shows several more of the cells similarly coloured, but it is usually not till after a couple of hours that one can safely declare all the tannin-containing cells to have been stained. With variations in respect of time, and with the sole exception of osmic acid, all the other tests act in precisely the same way; even Möll's, preferred to all others by some of our Continental *confrères*, being as unsatisfactory as the rest. But sooner or later its characteristic colour is imparted to these cells by every reagent, thus proving tannin to be present.

For the negative experiment the absence of the brown colour from tissues treated with Nessler's fluid, and its absence from the same tissues when acted upon by ordinary tannin reagents, recourse was again had to epidermis. The experiment succeeded in all cases; among these may be cited *Fatsia japonica*, wallflower, box, *Stellaria media*, and *Pelargonium zonale*. In none of these did tannin show up, although twenty-four hours were allowed to elapse before the preparations were destroyed.

Lastly, Nessler's fluid gives a rich brown precipitate with solutions of tannin. Moreover, with gallic acid a grey-green one is thrown down, thus affording an easy means of distinguishing between these bodies.

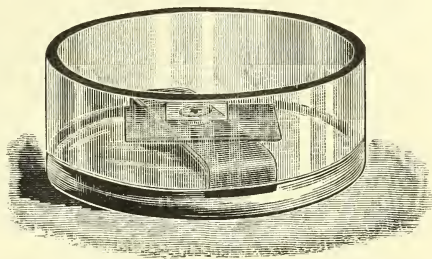


For these reasons, therefore, viz. the rapidity, certainty, and distinctness of its action; the ease with which it can be made; its permanence when made; and lastly, the difference in its behaviour towards tannin and towards gallic acid—for these reasons I am bold enough to anticipate the time when, to adapt a hackneyed expression, Nessler's fluid will be regarded as a reagent which no botanical laboratory should be without.

**Staining and Imbedding very Minute Objects.\***—The preparation of microscopically small objects is usually a very unsatisfactory procedure, but very good results may be obtained, says Dr. E. Overton, by adopting the following method:—Suppose the material is a hanging-drop cultivation on a cover-glass, as for example unicellular algae, Flagellata, pollen-tube, or the like. When the cultivation has reached the desired stage of development, the cover-glass is removed and iodine vapour allowed to stream over it. Iodine vapour is easily obtained by putting some crystals in a test-tube and warming them. Instead of iodine, osmic acid or its vapour may be used, but then manipulation is extremely difficult, not to say unsatisfactory.

By this method the objects are fixed at once, and then the iodine is removed by heating the preparation up to about  $40^{\circ}$  for 2-3 minutes. It is sometimes necessary to add a drop of distilled water during the evaporation of the iodine. The cover-glass, with the moist side still uppermost, is then put on a piece of elder-pith, about 3 mm. thick, and with a diameter rather less than the cover-glass. This, in its turn, rests upon a slide (Giessen size), which is placed in a glass capsule, the sides of which are about 2 cm. high. The slide does not lie on the bottom of the capsule, but is placed on a sort of little stool made of metal (see fig. 59).

FIG. 59.



To the preparation is added a drop of 20 per cent. alcohol and absolute alcohol in the capsule, the layer reaching half-way up the stool. The capsule is covered over and sealed up with vaselin. The vessel must be kept at an equal and moderate temperature, and not exposed to the sunlight. In a few hours the alcohol will have acted sufficiently upon the preparation. It is then removed and covered with a drop of collodion, or a solution of celloidin. When the celloidin has set a little, it is immersed in 80 per cent. spirit, wherein it becomes firmly set in about two minutes, so that the preparation may now be placed in any staining solution without fear of damage. The celloidin solution must be quite thin; the author uses the commercial solution diluted with six to ten parts of a mixture of equal parts of alcohol and ether.

\* Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 13-16 (1 fig.).



The best stains are carmine and hæmatoxylin, or eosin, iodine-green, and fuchsin. Other anilin dyes, as gentian-violet, are not suitable. The preparations should be dehydrated in 80 to 85 per cent. spirit, and then cleared up with creosote, or with a mixture of equal parts of 90 per cent. alcohol and creosote. They are then mounted in balsam, after having first passed through xylol.

Although this method may appear complicated, in reality it saves a great deal of time.

**Surface Deposits in Golgi's Method.\***—Sig. P. Samassa, in criticizing Schrwald's method for preventing surface deposits in sections treated by Golgi's method,† points out that in the original method of Golgi these surface deposits are considerably less. Hence, as in the latter no cover-glass is used, it is an obvious inference that the pressure of the glass sets up diffusion currents, whereby the precipitate is scattered over the section, and renders it often quite useless. The diffusion process is aided by the evaporation of the solvent. In the uncovered method, owing to the large area exposed to evaporation, these diffusion currents are not so likely to occur with such violence as when confined between two rigid layers.

**Staining Elastic Fibres and the Corneous Layer of Skin.‡**—Herr A. Köppen, in a continuation of the technique of staining elastic fibres,§ recommends a double staining, which may be either diffuse or nuclear.

For diffuse staining the following solution is used:—Carmin optim. 1·0 is dissolved in 50 ccm. cold water, then 5 ccm. liq. ammon. caust. is added, and the whole allowed to stand for two days. It is then filtered, and of the filtrate 1 drop is used to 20 ccm. water. The sections remain therein for twenty-four hours, and are then stained a diffuse red.

Staining of the nuclei and protoplasm.—(1) Weigert's picrocarmine stain is made by adding to the above solution 50 ccm. of a saturated aqueous solution of picric acid. This solution, which should be filtered before and after use, stains in from two minutes to several hours. (2) Grenacher's alum-carmine is made by boiling together for 15 minutes, and then filtering, carmine 1·0; alum 5·0; water 50·0.

The advantage of using these preliminary stains is that the subsequent decolorizing is extremely rapid.

**Decolorizing Preparations over-blackened by Osmic Acid.||**—The method of decolorizing objects over-blackened by osmic acid by means of peroxide of hydrogen was, says Dr. E. Overton, first introduced by Fol, but is so little practised that it merits a word in its favour. The following solution, which should be prepared every time, is recommended by the author:—Commercial peroxide of hydrogen 1 part; alcohol (70–80 per cent.) 10–25 parts. The removal of the osmium is completed in a few minutes, and the preparations stain excellently.

**Staining Sections of Botanical Preparations.¶**—Dr. A. Zimmermann gives a short description of some methods for staining botanical pre-

\* Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 26–8.

† See this Journal, *ante*, p. 410.

‡ Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 22–5.

§ See this Journal, *ante*, p. 410.

|| Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 10–12.

¶ T. c., pp. 1–8 (1 fig.).

parations, which he has found useful in the examination of chromatophores, crystalloids, and various cytoplasmic elements.

(1) Picro-fuchsin stain.—The sections are fixed to the slide, and the paraffin and its solvent xylol having been removed, are placed in a solution of acid-fuchsin, which is made by dissolving 20 grm. of the pigment in 100 ccm. of anilin water. In this solution, which should be gently warmed, the sections remain for 2–5 minutes, and are then washed in a mixture of 1 part of a saturated alcoholic solution of picric acid and 2 parts of water until no more dye is given off. After this the picric acid is to be extracted in absolute alcohol; then the sections are passed through xylol and mounted in xylol balsam.

(2) Acid-fuchsin staining, with subsequent washing out in flowing water.—This method is serviceable for staining thick sections made from living tissue and then fixed. After the fixative is extracted the sections are placed in a 0.2 per cent. watery solution of acid-fuchsin, in which they remain for 24 hours or longer. The excess of stain is then extracted in flowing water, and this is best done by means of Steinach's glass filter capsules.\* The capsules are placed in a receiver, over which is a pipe with a number of small taps, from which the water can be made to flow into the capsules. In adopting this method it is advisable to manipulate a large number of sections at once, and to examine them from time to time to ascertain the proper degree of decoloration.

(3) Iodine-green for staining chromatophores.—The sections are made from tissue previously fixed with an alcoholic sublimate solution, and then immersed for half an hour in a saturated aqueous solution of iodine-green. They are then washed in water and examined in glycerin or Hoyer's mounting fluid, or in balsam. If balsam be used, then dehydration must be effected by merely drying the preparation. Then xylol is added, and, when saturated with this, the xylol balsam. As a contrast stain for the rest of the tissue, a watery solution of Bismarck brown may be used.

(4) Ammonia-fuchsin for staining the chromatophores.—This stain is prepared by adding chemically pure ammonia to an alcoholic solution of fuchsin until the fluid assumes a bright yellow colour. The solution may be used at once, but will only keep a few weeks. The sections are fixed to a slide and some of the solution poured thereon and allowed to remain for some few minutes. They are then washed and examined in water or glycerin. Hoyer's mounting medium may be used, or even balsam. If the latter, then the sections must be dehydrated by drying them in the air.

**Staining Human Retina with Acid Hæmatoxylin.**†—Dr. J. Schaffer has been able to differentiate the outer and inner segments of the rod and cone layer of human retina by staining the tissue with the acid logwood recommended by Kultschitzky.

The sections, imbedded in celloidin, are taken from the Müller's fluid or alcohol in which they have been fixed and hardened, and left during the night in a 1 per cent. solution of chromic acid, which acts as

\* This Journal, 1888, p. 850.

† SB. K.K. Akad. Wiss. Wien, xcix. (1890) pp. 110–20 (1 pl.).

a mordant. After having been washed they are placed in the logwood solution for about twenty hours. The overstaining is removed by decolorizing with Weigert's borax and ferrocyanide of potash solution. The proper degree of differentiation is attained when the rod and cone layer alone remains of a dark colour, the rest of the layers having a brownish hue to the naked eye. The sections are then washed in water and mounted in balsam in the usual manner.

**Hæmatoxylin as a means for ascertaining the Alkalinity or Acidity of Tissues.\***—Prof. F. Sanfelice has found that the acid or alkaline reaction of tissues may be recognized by staining with Boehmer's hæmatoxylin (alkaline), or with the author's iodized hæmatoxylin (acid).†

In using this method as a test, two principal precautions must be observed. First it is necessary that the normal reaction of the tissue must not be interfered with, hence reagents such as chromic acid and its salts, Müller's fluid and Flemming's solution are unsuitable fixatives. The author used chiefly absolute alcohol for hardening and fixing, and also corrosive sublimate, the excess of which must always be carefully extracted with spirit. The second precaution is that the hæmatoxylin solution must have only a feeble reaction.

Among the instances of differential staining obtained by this method it is mentioned by the author that the protoplasm masses in the ovary and testicle of Selachians are coloured red when the whole of the tissue is treated with the alkaline solution—a fact which proves that the elements undergoing this form of necrobiosis acquire an acid reaction. Goblet-cells in the intestinal mucosa become coloured blue, while the rest of the tissue remains red. Hence the reaction of goblet-cells is alkaline, and this method might be usefully employed to ascertain the reaction of tissues or elements, and their products.

**New Method of Staining Central Nervous System, and its Results.‡**—Prof. P. Flechsig recommends the following method for staining the nerve-cells of the cerebral cortex and their prolongations. By means of it it was shown that the axis-cylinder process was the only prolongation from the cell which was in connection with a nerve-fibre; that the axis-process, which is not at its commencement medullated, divides like a T, i. e. dichotomously at a right angle. In the occipital lobe a trichotomous subdivision was the rule, although frequent subdivision was also remarked. In the neighbourhood of the central fissure some axis-fibres did not subdivide.

These results were obtained by hardening pieces in 2 per cent. aqueous solution of chromate of potash, and then making sections not exceeding 5/100 mm. in thickness.

After soaking in 96 per cent. spirit, the sections are kept for 3–8 days in a solution of redwood extract at a temperature of 35° C. The sections having been washed in distilled water are then decolorized in the following manner:—Each section is placed in 3 ccm. 1/4–1/5 per cent. solution of permanganate of potash until the solution have lost its

\* Journ. de Micrographie, xiv. (1890) pp. 21–2.

† See this Journal, 1889, p. 837.

‡ Berichte u. d. Verhandl. K. Sächs. Gesell. Wiss. Leipzig, 1890, pp. 328–30 (1 pl.).



bluish colour; it is then immersed in the decolorizer (distilled water 200, oxalic acid 1, hyposulphite of potash 1), until all traces of yellowness have departed from the section.

The redwood solution is made as follows:—1 gram of the pure extract of Japan redwood is dissolved in 10 grams of absolute alcohol, and then diluted with 900 grams of distilled water. To this are added 5 grams of a saturated solution of Glauber's salt and a similar quantity of a saturated solution of tartaric acid.

If this redwood method be combined with Golgi's sublimate staining, the sections, having been stained as above, are placed in a mixture of 20 cem. absolute alcohol and 5 drops of 1 per cent. solution of chloride of gold and potash, until the sublimate precipitate have become quite black, and the red nerve-fibres have assumed a bluish tone. They are then washed in 10 grams of distilled water, to which 1 drop of a 5 per cent. solution of cyanide of potash has been added, then dehydrated in absolute alcohol, cleared up in oil of lavender, and mounted in balsam.

BURCHARDT, E.—Eine neue Amyloidfärbung. (A new amyloid stain.)

*Virchow's Arch.*, CXVII. (1889).

Cf. *Fortschr. d. Med.*, VII. (1889) No. 23, p. 901;

*Centrabl. f. Klin. Med.*, XI. (1890) No. 4, p. 74.

DEKHUYZEN, M. C.—Ueber das Imprägniren lebender Gewebe mit Silbernitrat. (On the impregnation of living tissues with silver nitrate.)

*Anat. Anz.*, IV. (1889) No. 25, p. 789.

NICKEL, E.—Die Farbenreactionen der Kohlenstoffverbindungen. Für chemische, physiologische, mikrochemische, botanische, medicinische und pharmakologische Untersuchungen. (The colour-reactions of carbon-compounds. For chemical, physiological, micro-chemical, botanical, medical, and pharmacological investigations.)

2nd ed., Berlin (Peters), 1890, 8vo, 134 pp.

#### (5) Mounting, including Slides, Preservative Fluids, &c.

**Finishing Balsam Mounts.\***—Mr. F. N. Pease finishes balsam mounts as follows:—The object is mounted on the slide, applying the cover-glass in the ordinary manner, using either balsam, hardened balsam, balsam and benzol, storax or dammar. The slide is then heated to drive off the solvent or more volatile constituents, either gently in a water-bath or at a higher heat, even boiling carefully over a spirit-lamp when the nature of the object will permit. When cold, the superfluous mounting medium is carefully removed, then a narrow ring of paraffin-wax is heated in a capsule until it is melted and quite limpid. With the aid of a very small camel's hair pencil, the melted paraffin is applied at the edge of the cover-glass, covering the exposed medium and instantly solidifying. It is now necessary to apply a finishing cement. For this purpose Bell's cement has been found excellent. If this cement does not work satisfactorily the admixture of some chloroform makes it work smoothly. This cement ring is finished at one application, and in a few hours the slide is ready for the cabinet.

This method is intended to protect the mounting medium from becoming discoloured owing to atmospheric influences.

**A new Diatom Mounting Medium.†**—Mr. F. W. Weir writes, "C<sub>10</sub>H<sub>7</sub>Br + Resin of Tolu.—Dissolve 3 oz. of commercial balsam tolu

\* *Amer. Mon. Micr. Journ.*, xi. (1890) pp. 66-7.

† *Micr. Bull. and Sci. News*, vii. (1890) pp. 23-4

in 4 fluid drachms of benzine ( $C_6H_6$ ) at a temperature of about  $45^\circ C.$ , and strain. Add 4 fluid oz. of carbon bisulphide, agitate thoroughly, and allow to cool, when the tolu solution will separate and the carbon bisulphide with cinnamic acid in solution can be decanted. Add another portion of the carbon bisulphide and treat as before. Finally pour the tolu solution into a glass tray and evaporate the benzine.

Place in a  $1\frac{1}{2}$  oz. glass-stoppered phial 1 fluid drachm of naphthaline monobromide, and add gradually about three times its volume of the resin of tolu, or sufficient to make the mixture quite stiff when cold. The solution will be effected slowly at about  $45^\circ C.$  The above constitutes a mounting medium which is rather easier to use than Canada balsam.

Warm the medium at  $40^\circ$  to  $45^\circ C.$  until quite fluid, take up a minute quantity on a warm needle, place on centre of cover-glass and invert on slide. Use no pressure whatever, but warm the slide gently, when the medium will flow to edge of cover.

After a few days ring with a non-alcoholic cement. This method of treating balsam tolu does not remove an atom of resin, and does not allow an atom of cinnamic acid to remain.

The subsequent solution in naphthaline monobromide produces a medium of higher index (1.73) than the resin alone, permanent in structure and volume, and free from objections to which any medium in a volatile solvent is subject."

**Tolu and Monobromide.\***—Mr. H. L. Smith writes to the Editor of the 'Microscopical Bulletin':—

"I meant to reply to your letter before. The bromide medium will keep if *tightly sealed*, but almost all cements, and some coloured waxes, decompose it. I must say I am not satisfied, and would not advise any one to use it. The yellow medium can be made to keep, but I don't like the colour.

Mr. Weir, of Norwich, Conn., sent me a compound of monobromide of naphthaline and tolu, which is best of any of the high mediums yet—no crystals, easy to use, and very satisfactory.

He is about publishing the formula. I wish somebody—you or some one—would make it for sale, as he does not intend to do this. It has full as high index as monobromide, and none of its disadvantages.

It has consistency of ordinary balsam, and is used like that. It can be hardened by careful heat; or better, mount without heat, and in a day or so it will harden to allow asphaltting, or in a few more days will need no ring. It is going to do the thing, I *guess*.

Nothing could please me more than to have you make the bromide medium if I could advise it. It keeps perfectly well in the bottle. I have it two and three years old. No decomposition at all, but it acts so powerfully on all cements, that *this* prevents its usefulness. The index is considerably above monobromide, but the latter is high enough, and I am pretty well pleased with it."

**Fixing Sections with Uncoagulated Albumen.†**—Dr. J. Rabinoviez has found that albumen may be used for fixing sections to the slide by

\* Micr. Bull. and Sci. News, vii. (1890) p. 24.

† Zeitschr. f. Wiss. Mikr., vii. (1890) p. 29.

adhesion as well as by coagulation, and the method is as follows:—The sections are laid on the slide, covered with albumen, and pressed down with a brush. The slide is then put straight into toluol until the paraffin is dissolved. The time required for this varies with the quantity of paraffin (from one to five minutes). The specimen may then be mounted in balsam. If there be any glycerin, however little, mixed with the albumen, this must be removed by immersion of the slide in absolute alcohol for five to ten minutes.

This method has the advantage over others in that it is shorter, and that the albumen is not coagulated by heat or spirit.

(6) **Miscellaneous.**

**New Reaction for Albuminoids.\***—Herr C. Reichl proposes the following test for albuminoids, which, though not so sensitive as Millon's reagent, may yet be of service in micro-chemico-botanical investigations. Two or three drops of a dilute alcoholic solution of benzaldehyd, a moderate quantity of dilute sulphuric acid (equal parts of acid and of water), and a drop of solution of ferric sulphate, give a dark blue colour with an albuminoid. A light blue colour is brought out by the first two substances, which becomes deep blue by the action of the ferric sulphate. Concentrated hydrochloric acid may be used in place of the sulphuric, and a different soluble iron salt, for example the chloride, in place of the sulphate.

WHEATCROFT, W. G.—**Presidential Address to the Bath Microscopical Society.**  
*Journ. of Micr.*, III. (1890) pp. 48–52.

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\* SB. K.K. Akad. Wiss. Wien, Monatsheft f. Chemie, 1889, p. 317. See Bot. Centralbl., xlii. (1890) p. 367.

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6994. JOURNAL  
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ROYAL  
MICROSCOPICAL SOCIETY;

CONTAINING ITS TRANSACTIONS AND PROCEEDINGS,

AND A SUMMARY OF CURRENT RESEARCHES RELATING TO

ZOOLOGY AND BOTANY

(principally Invertebrata and Cryptogamia),

MICROSCOPY, &c.

*Edited by*

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*and Professor of Comparative Anatomy and Zoology in King's College;*

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## MICROSCOPY.

## a. Instruments, Accessories, &amp;c.\*

## (1) Stands.

ALTMANN, R.—Ueber die Verbesserungsfähigkeit der Mikroskope. (On the possibilities of improvements in the Microscope.) Part II.

*Arch. f. Anat. u. Physiol., Anat. Abtheil.*, 1889, H. 5, 6, p. 326.

ANDERSON, R. J.—A panoramic arrangement for the Microscope.

*Internat. Monatsschr. f. Anat. u. Physiol.*, VI. (1889) H 8, p. 289.

BERNARD, P.—Note sur un Microscope composé du 18<sup>e</sup> siècle. (Note on a compound Microscope of the 18th century.)

*Journ. des Sci. Med. de Lille*, XI. (1889) p. 1.

HEITZMANN, C.—Die Zukunft der Mikroskopie. (The future of Microscopy.)

*Wiener Med. Bl.*, XII. (1889) Nos. 37, 39.

OLIVIER, L.—Histoire des Microscopes. (History of Microscopes.)

*La Nature*, XVII. (1889) pp. 267, 314.

POLI, A.—Le Microscope et sa théorie. (The Microscope and its theory.)

*Rev. de Bot.*, VII. p. 20.

## (2) Eye-pieces and Objectives.

The Achromatic Object-glass.—J. Godfrey writes:† —“I am glad to find our correspondent ‘Prismatique’ coming to the front again, and as an amateur optician I write to ask his opinion upon a curious point. The glass I have always worked upon has been Chance’s hard crown and dense flint, and after all sorts of experiments with different combinations of curves, I have found that a very good combination is to make the crown lens equiconvex, and the curves of the flint in the proportion of ten to one double concave. Of course I am well aware that these curves are foundation curves only, and that delicate and final corrections are indispensable; the workman, so I find, can only select curves to work up to, and alter, according to his experience and manual skill. Now I find—and this is the result, not of theory, but of experience—that with these proportions of the curves the flint lens corrects the achromatism of the crown slightly more at the marginal zone than it does at the centre. For example, if the flat lens so far over-corrects the crown as to eliminate the irrationality of the crown lens with respect to the red of the spectrum for the outside zone, there then remains, as I have found by practice, a minute residuum of the secondary spectrum in favour of the crown lens in the centre of the object-glass, a faint trace of red, which of course is not obtrusive, but it is there. Now I want to eliminate this want of balance between the outside and centre of the object-glass. At present I am very busy working upon two very fine and massive discs of Chance’s hard crown and dense flint, and the object-glass will be 7 in. clear aperture; of course this is not the first glass I have made. My present 5 in. will show a curiously mottled and indented terminator upon Venus, and she is a terrible planet to define. Now I want ‘Prismatique’s’ opinion upon this point. I propose to make my crown-lens equiconvex, 27·5 in. radius, and the focal length of the flint to be 49·6 in. This will give the proportions of the focal

\* This subdivision contains (1) Stands; (2) Eye-pieces and Objectives; (3) Illuminating and other Apparatus; (4) Photomicrography; (5) Microscopical Optics and Manipulation; (6) Miscellaneous.

† Engl. Mech., li. (1890) p. 118.

lengths 1:1·8, which I have found a good proportion. But now instead of putting the radii of the flint as 1:10, or nearly so, I propose to take a plano-concave and make the radius 24·8. Then I should work the back of the flint, not to an absolute curve, but to an exceedingly long concave curve, nearly a plane. This would necessitate the flint lens being fitted a short distance from the crown, about two-tenths or so by experiment, and I think that this would give a better correction for achromatism. I am quite aware that this would give a slight excess of spherical aberration to the concave; but, if necessary, I propose to correct it after fixing the distance between the lenses that will best suit the achromatism. Now, I am rather fortified in my belief that this modification will answer, in that my proposed construction will closely approximate to the construction of the object-glass of the Lick telescope, in which the convex is an equiconvex, and the flint is a double concave with a long concave curve at the back, and the two lenses are separated and are not in contact. It appears to me that the achromatism would be improved by separating the lenses."

**The Jena Lenses.\***—The following is part of a letter from "F.R.M.S.":†—As statements have been made in former numbers of this paper impugning the good faith of MM. Zeiss, as to the new objective of 1·6 N.A., I wrote them requesting an authoritative statement on the subject. They have very kindly sent me a copy of a letter which their Prof. Abbe has written to Mr. Mayall in consequence of my communication to them. Their letter is dated 10th September, and in it Prof. Abbe says:—"Please to take notice of a formal assertion from *my* part that the objective has *not* undergone any alteration whatever, while in Jena; *that every lens and every piece of the mounting was in exactly the same state at the second departure to London in which it was at the first departure.*"

The italics are the Professor's, not mine.

Prof. Abbe further states:—"Though I have not myself looked up the lens all the time over, I am in a position to give this assertion quite *positively* on these grounds.

"(1) Nobody in the workshop had any sensible interest in making an alteration and *concealing it to me*. For *nobody* except myself was responsible for whatever defect of the objective. The computations had been made under my personal direction, and I had approved of the optician's work after execution. If a defect of any kind had happened to come out afterwards, the fault would have been *mine only*.

"(2) Nobody *could* try to change or improve the system without consulting me, because no other person was *au fait* with regard to that particular construction."

Prof. Abbe further states:—"The objective had not been tried *photographically* by us, neither Van Heurck's sample, nor the other one. I was therefore *quite prepared to admit* that a 'chemical' focus could exist, owing to an insufficient approximation in uniting the violet ray with the other rays (in our computation) under the condition always that in Van Heurck's sample *the same defect must exist*, as both objec-

\* Engl. Mech., Oct. 1890, p. 124.

† Published, however, without the authorization of Prof. Abbe.—Ed. J.R.M.S.



tives had shown the *same* degree of achromatism of the *visual* light. Though it appeared rather strange that Dr. Van Heurck should *not* have observed the fault, I supposed that he could perhaps have overlooked it, or had not found it hurtful, owing to his particular mode of illumination or photographic operation.

"In *this* spirit I advised Dr. Czapski to measure the residual difference of the chemical focus, and to compute a correcting lens, to be added to the system, in order to compensate for the expected difference.

"Having left the matter to Dr. Czapski, as I am not versed in photomicrography, I was *much astonished* to hear from him—a *short* time after arrival of the lens—that he could not find a difference of focus. In face of the positive assertion about the result of *your* trial, I felt doubtful about the accuracy of Dr. Czapski's observation, and I requested him decidedly to *repeat the trial* with all possible precautions, though *he* considered this as useless."

**Fluor-spar at Oltscheren.\***—Dr. E. v. Fellenberg gives a very full account of the occurrence at Oltscheren of fluor-spar, which is the subject of so much interest to microscopists at the present time. Fluorite is a mineral very widely distributed in the Alps. A locality long noted for the abundance of the pale-green variety is "Rauu," or more correctly "Runn," a wood near Giesbach opposite to Brienz. The first mention of this locality is to be found in G. S. Gruner's 'Versuch eines Verzeichnisses der Mineralien des Schweizerlandes,' Bern, 1775, and a further description is given in Höpfner's 'Magazin für die Naturkunde Helvetiens,' vol. iv., 1789, in an account of a journey made by General-Commissioner Manuel in the Bernese Alps. Green fluor was also obtained in the Jura limestone from the Vordendürschreunalp am Säntis and yellowish-brown and wine-coloured crystals from the Upper Jurassic limestone at Salève bei Genf. But by far the most remarkable and interesting occurrence of fluor is that at Oltscheren or Oltschialp, more exactly at Oltschikopf, south of the village of Brienzwyl in the Bernese Oberland. Here in 1830, according to a label on a specimen in the Bern Museum, Hans Fischer and Mitkaften discovered in a cleft of the mountain opposite Brienzwyl about 200 cwt. of fluor, of which 2 cwt. consisted of crystals. These men appear to have made considerable journeys with their treasure piled up in a cart in huge blocks, some of which, according to Prof. B. Studer, who purchased several specimens from them at the time, were a foot in diameter, and water-clear like blocks of ice. The precise locality of this remarkable find had been forgotten, when in 1886 Prof. Abbe began to make inquiries about the occurrence of water-clear fluor-spar. Many years before the author had sent to Herr Wappler, a mineral dealer in Freiberg, in exchange for Saxon minerals, some water-clear crystals of fluor from "das untere Haslithal im Kanton Bern." Prof. Abbe having seen these specimens was induced to visit the author, by whom he was referred to Herr Hamberger, the director of the pyrotechnic laboratory in Oberried, near Brienz, as well as to the hunter Caspar Blatter, as being the most likely persons from whom information could

\* 'Ueber den Flusspath von Oltschrenalp,' Mittheil. Naturf. Ges. in Bern, 1889, pp. 202-19.



be obtained of the occurrence of fluor-spar of similar quality at Oltcheren. The crystal seekers, M. Ott and C. Streich, of Guttanen, as well as the hunter Caspar Blatter, were at once commissioned by Prof. Abbe to make investigations in the neighbourhood, but it was not until the spring of 1887 that they succeeded in rediscovering the old locality of 1830. A new locality was also discovered, from which beautiful green crystals, varying in size from 1 cm. in diameter to one over 20 cm. in length, were obtained. The surface of most of these specimens was rough, many being covered with irregular holes, while others looked like ice which had begun to melt in the sun. These specimens were offered for sale by Ott and Streich without the knowledge of Prof. Abbe, and were purchased by the authorities of the Bern Museum. Of the material sent to Prof. Abbe at Jena very little was found to be fit for optical use. The authorities of Brienzwyler now took action and prohibited further search for useful minerals in the district under their jurisdiction. An agreement was then drawn up by them with a company of capitalists, at whose head stood the firm of Zeiss, in Jena, and Prof. Abbe, by which the exclusive right of search for fluor-spar in that district was granted to the latter. The stipulation was, however, made that all material unfit for optical purposes should become the property of the authorities. The company began work in the summer of 1888 under the directorship of Herr Kable of Jena, who was stationed in a hut on the Alp Bühlen. According to a letter of Prof. Abbe to the author the old find of 1830 came from two cavities on the south part of the mountain. The lower one was easily accessible, but the other, high above, could only be reached by a 72 ft. ladder from another projecting rock mass. Both were found to have been exhausted, and further search for fluor in the neighbourhood only met with indifferent success as regards quality. In conclusion the author describes the visit he himself paid to the locality under the guidance of Caspar Blatter and Herr Kable. Starting from Meyringen with Blatter he passed by Prasti, Schüttelboden, Laui-Vorsass, and Platten to Bühlen, where Herr Kable was installed. With the latter he then proceeded through the valley of Oltcheren to the upper Alp Oberfeld, whence could be seen the south slope of the Oltschikopf, with the two cavities, from which came the extraordinary find of 1830, plainly visible.

JOHNSTON, C.—The American Objective as compared with the German.

*Maryland Med. Journ.*, XXI. (1889) p. 130.

### (3) Illuminating and other Apparatus.

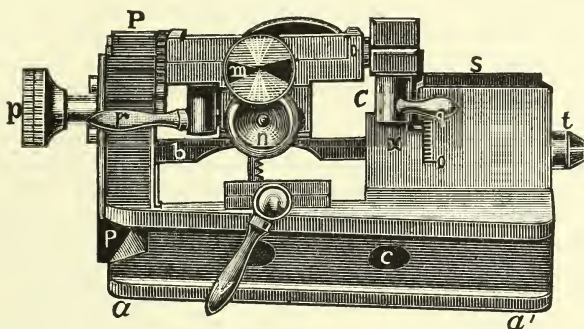
**Object-carrier with Vertical Displacement for the Jung Microtome.**\*—Prof. L. Koch points out that the object-carrier hitherto used only allows a comparatively slight elevation (3 to 4 mm.) of the object adjusted. This is due to a great part of the slide-way being occupied by the micrometer screw and the object-carrier. In fact, for the object itself, the displacement is only about a millimetre, since often more than a millimetre of paraffin has first to be removed, and, if the course

\* *Bot. Centralbl.*, xl. (1889) pp. 283-5.

of the knife is very restricted, the full extent of the slide-way cannot be utilized. In most cases this small displacement is not sufficient, so that it is necessary to dismount the object during the work and readjust it. This entails loss of sections, irrespective of the inconvenience of such a process.

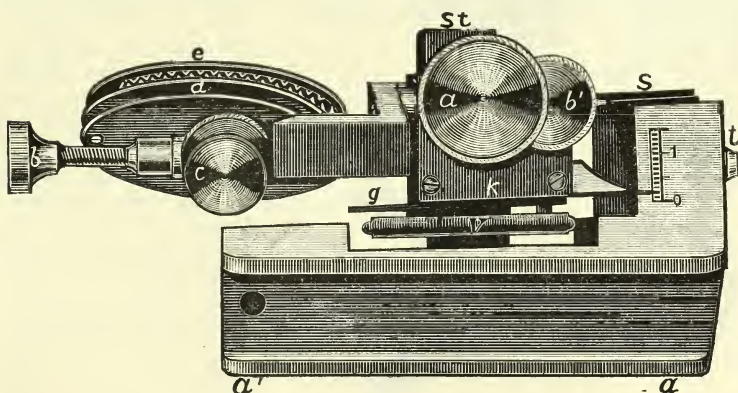
To get rid of this difficulty, Herr R. Jung of Heidelberg has, under the author's direction, constructed an object-carrier with vertical displacement. In one of these, represented in fig. 72, the frame O,

FIG. 72.



carrying the object-clamp, is movable in the vertical direction. This runs in a prismatic groove with so much friction that no fixing-arrangement is necessary to keep it in any given position. The

FIG. 73.



movement is effected by rack and pinion. The frame rests upon a steel base *b*, provided with a ratchet in which works a toothed wheel, set in motion by the lever *r*. One turn of the lever effects a rise of the frame, and consequently of the slide-way, of 1.2 cm.

To begin work, the lowest position is given to the frame carrying the object-clamp, the paraffin block is mounted somewhat high, and the

surface to be cut is raised, by means of the lever, up to the knife-edge. The removal of the paraffin is effected in the same way by means of the vertical displacement, but the cutting of the object itself is done exclusively by the use of the micrometer-screw. When the latter is turned to the end it is screwed back so as to bring the object-carrier into its original position, and the object is then again brought up to the knife-edge by means of the lever.

The object-carrier is especially serviceable in all cases in which the object is to be sectionized only at intervals determined by the development of lateral organs. The micrometer-screw is then used for the parts to be sectionized, and the vertical displacement for the rest. There is an index at  $x$  for measuring the intervals between two of the lateral organs to be cut. The object-carrier represented in fig. 73 is of simpler construction, but is quite satisfactory for most purposes, and is to be recommended for use with the small model of the microtome. The movable metal-piece  $k$  supports the projecting object-clamp, and runs in a prismatic groove  $st$ . It rests on a screw-plate V, by the rotation of which its rise and fall are effected. A binding-screw  $a$  fixes it in any position. The rise, exclusive of the slide-way, amounts to 1 cm.

**New Heating Apparatus for Mineralogical Investigations.\***—This piece of apparatus, designed by R. Brünnee, of the firm of Voigt and

Hochgesang, in Göttingen, can be easily fitted to any Microscope. It serves to raise solid preparations or liquids to a high temperature, and, since the flame burns directly beneath the object-carrier, observation can be made by polarized light during the heating. The apparatus has the following arrangement:—Beneath the object-stage B (fig. 74) is a piece bored through in four places. Round the lower, conically turned, part of the piece the arm A is fitted. The latter is movable on the cone, and is fastened to B by a screw  $c$ . Between  $c$  and B a ring-shaped space  $o$  is left,

FIG. 74.

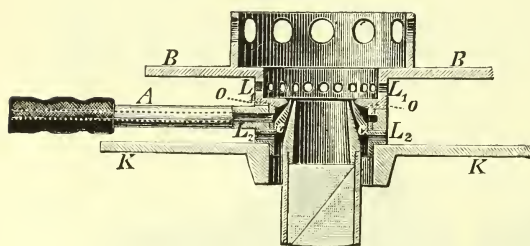
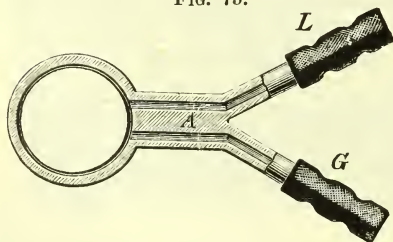


FIG. 75.



which is contracted internally to a fine slit. The gas and air required for the flame stream through the tubes L and G (fig. 75) into this space. The object-carrier B is provided with a row of outlets  $L_1$ . The openings

\* Zeitschr. f. Instrumentenk., x. (1890) pp. 63-4.



$L_2$  are for the admission of air. The tube  $L$  of the arm  $A$  is in connection with a reservoir of compressed air, which effects a quick cooling when necessary.

To connect this apparatus with a Microscope, the lower part of the screw-piece fits into the aperture of the Microscope-stage, so that the stage can be rotated while the arm  $A$  with the tubes remains fixed.

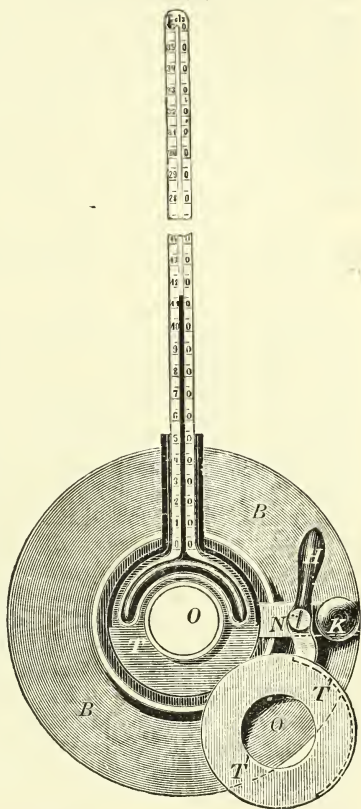
For heating up to  $360^\circ$  a drum (fig. 76), which carries a thermometer and the preparation, is added to the apparatus. This consists of two parts  $T$  and  $T_1$ . The lower part  $T$ , carrying the thermometer, is connected with the stage  $B$  by a screw  $K$ , while the upper part  $T_1$  can be turned to one side about the axis  $N$  by means of the lever  $H$ . The preparation is placed on a ring in the drum, and is kept at the same height as the thermometer.

The apparatus was exhibited at the Exhibition of the Heidelberg Naturforscher - Versammlung, and has been described in the 'Abtheilung für Instrumentenkunde.\* It has already met with considerable success and is particularly suitable for mineralogical-petrographical investigations.

**Bolting Gauze.†**—Mr. Charles M. Vorce writes that he has "done no microscopical work lately that has any novelty in it, unless it may be the measurement of an assortment of bolting gauze and other goods used for sieves, to ascertain the average and maximum sizes of the particles which pass through the same, and the relation of such size to the rating of the goods which is always by the number of meshes to the inch or centimeter. Bolting gauze of '200 meshes to the inch' will not pass particles of approximately globular form larger than about  $1/400$  in., and the *average* size of the particles passed will be considerably less, about  $1/450$ ."

**A Simple Turn-table.‡**—Mr. A. S. Elliott describes a simple turn-table. "Procure the frame and running gear of any cheap clock. Fifty cents will cover cost of all materials. Remove the main spring from its place and make the wheel carrying it firm on the shaft. Remove all

FIG. 76.



\* Cf. Zeitschr. f. Instrumentenk., 1889, pp. 359 and 478.

† Amer. Mon. Micr. Journ., xi. (1890) p. 106.

‡ T. c., p. 117.



projecting parts from both top and bottom of frame. Reverse the centre wheel, putting the larger end of shaft uppermost, and making all bearings tight and smooth without oil. Cut a brass plate (soft) 3 inches in diameter; find centre, bore, then bore two more holes  $1\frac{1}{4}$  in. from centre; make a pair of light bowed springs, solder to nail fitting such hole and fit tightly through plate, placing the clips in opposition to each other. Cut or scratch three concentric circles  $\frac{1}{4}$ ,  $\frac{1}{2}$ , and  $\frac{3}{4}$ , turning table rapidly. Fit the centre shaft firmly to plate without soldering.

The apparent disadvantage of using a cogged wheel in turning with the hand is more than counteracted by the greater ease and consequent steadier rotation, together with greater speed, attained by this table. Carefully made it will do as good or even better work than the ordinary form. If preferred the clips may be soldered fast to plate, but are rather unhandy.

The holes in the bottom of frame can be utilized to secure to firm base and hand-rest in any convenient manner to suit the requirement of the maker."

**Cheap Boxes for Slides.\***—Mr. Henry Shimer writes:—"W. P. Hamilton's slide-box described in the January number reminds me of a very nice arrangement. A box ready-made is more apt to be used than one made on purpose; for instance, the ordinary cigar-box, costing nothing. The flat ones are most suitable. They vary in size somewhat, but the ordinary one is about  $4\frac{1}{2}$  by  $8\frac{1}{2}$  by 2 in. inside. It can be filled with cardboard trays like Hamilton's, or with wooden ones made of cigar-boxes. The bottoms and lids will make bottoms for the trays, and the sides and ends sawn into narrow strips  $\frac{1}{8}$  or  $\frac{1}{4}$  in. wide and tacked on with brads, will make the margins. Each box will hold five trays. The bottom may be used instead of a tray by tacking a marginal strip on each end. Each of such boxes will store 70 short German slides, which by all odds are preferable, or it will hold 45 to 50 of the 3-in. slides. If we make the trays of cardboard, as per Hamilton, and a 3-in. holds 24, 2-in. holds 16 trays. Then 14 short slides to a tray gives room for 224 slides; 9 3-in. sheets to a tray gives 144 slides, or 7 to a tray will give 112 slides, and allow about  $\frac{3}{4}$  in. margin on the sides and a little less on the ends. Such boxes are neat, cheap, and convenient. The slides lie flat. These boxes can be numbered or otherwise labelled on the ends and stowed in bookcases."

BRAATZ, E.—Ein neues Mikrotom. (A new Microtome.)

*Illustr. Monatsschr. d. Aerztl. Polytechn.*, XI. (1889) p. 159.

GABRIEL.—Chambre claire du Microscope. (Camera lucida.)

*Progrès Méd.*, VIII. (1888) No. 51.

PETTIGREW, J. B.—On the use of the Camera Lucida.

*Trans. Manchester Micr. Soc.*, 1888, p. 80.

#### (4) Photomicrography.

**Mr. Pringle's Photomicrographic Apparatus.**—The two figures now given (plates XII. and XIII.) will, without further comment, supplement the description of Mr. Pringle's photomicrographic apparatus which was given on p. 543 of the Journal.

\* *Amer. Mon. Micr. Journ.*, xi. (1890) p. 106.

**Photomicrography by Gaslight.\***—Major Geo. M. Sternberg observes:—Those who have had much experience in making photomicrographs will agree with me that one of the most essential elements of success is the use of a suitable source of illumination.

Without question the direct light of the sun reflected in a right line by the mirror of a heliostat is the most economical and in some respects the most satisfactory light that can be used. But we cannot command this light at all times and places, and it often happens that when we are ready to devote a day to making photomicrographs the sun is obscured by clouds, or the atmosphere is hazy. Indeed, in some latitudes and at certain seasons of the year a suitable day for the purpose is extremely rare. The use of sunlight also requires a room having a southern exposure and elevated above all surrounding buildings or other objects by which the direct rays of the sun would be intercepted. For these reasons a satisfactory artificial light is extremely desirable.

The oxy-hydrogen limelight, the magnesium light and the electric arc light have all been employed as a substitute for the light of the sun, and all give satisfactory results. I have myself made rather extensive use of the "limelight," and think it the best substitute for solar light with which I am familiar. But to use it continuously, day after day, is attended with considerable expense, and the frequent renewal of the supply of gas which it calls for is an inconvenience which one would gladly dispense with.

These considerations have led some microscopists to use an oil lamp as the source of illumination, and very satisfactory photomicrographs with comparatively high powers have been made with this cheap and convenient light. But in my experience the best illumination which I have been able to secure with an oil lamp has called for very long exposures when working with high powers, and as most of my photomicrographs of bacteria are made with an amplification of 1000 diameters, I require a more powerful illumination than I have been able to secure in this way. And especially so because of the fact that a coloured screen must be interposed, which shuts off a large portion of the actinic rays, on account of the staining agents usually employed in making my mounts. The most satisfactory staining agents for the bacteria are an aqueous solution of fuchsin, or of methylene-blue, or of gentian-violet, and all of these colours are so nearly transparent for the actinic rays at the violet end of the spectrum that a satisfactory photographic contrast cannot be obtained unless we shut off these rays by a colour screen.

I am in the habit of using a yellow screen for my preparations stained with fuchsin or methylene-blue, and have obtained very satisfactory results with the orthochromatic plates manufactured by Carbutt of Philadelphia, and a glass screen coated with a solution of tropoline dissolved in gelatin.

But with such a screen, which shuts off a large portion of the actinic light and increases the time of exposure three or fourfold, the use of an oil lamp becomes impracticable, with high powers, on account of the feebleness of the illumination.

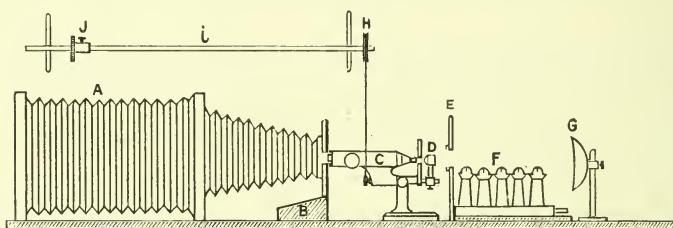
These considerations have led me to experiment with gaslight, and

\* John Hopkins University Circulars, ix. (1890) p. 72.

the simple form of apparatus which I am about to describe is the result of these experiments. I have now had the apparatus in use for several months, during which time I have made a large number of very satisfactory photomicrographs of bacteria from fuchsin-stained preparations with an amplification of 1000 diameters. My photographs have been made with the 3 mm. ol. im. apochromatic objective of Zeiss and his projection eye-piece No. 3. I use a large Powell and Lealand stand, upon the substage of which I have fitted an Abbe condenser. The arrangement of the apparatus will be readily understood by reference to the accompanying figure.

A is the camera which has a pyramidal bellows front supported by the heavy block of wood B; this can be pushed back upon the base-board which supports it so as to allow the operator to place

FIG. 77.



his eye at the eye-piece of the Microscope. When it is brought forward an aperture of the proper size admits the outer extremity of the eye-piece and shuts off all light except that coming through the objective. C is the Microscope and D the Abbe condenser supported upon the substage; E is a thick asbestos screen for protecting the Microscope from the heat given off by the battery of gas-burners F. This asbestos screen has an aperture of proper dimensions to admit the light to the condenser D. The gas-burners are arranged in a series with the flat portion of the flame facing the aperture in the asbestos screen E. The concave metallic mirror G is properly placed to reflect the light in the desired direction. I have not found any advantage in the use of a condensing lens other than the Abbe condenser upon the substage of the Microscope. The focusing is accomplished by means of the rod *i*, which carries at one extremity a grooved wheel H, which is connected with the fine-adjustment screw of the Microscope by means of a cord.

The focusing wheel J may be slipped along the rod *i* to any desired position, and is retained in place by a set-screw. The rod *i* is supported above the camera by arms depending from the ceiling, or by upright arms attached to the base-board.

I have lost many plates from a derangement of the focal adjustment resulting from vibrations caused by the passing of loaded waggons in the street adjoining the laboratory in which I work. This has been overcome to a great degree by placing soft rubber cushions under the whole apparatus.

**Position of the Light-filter in Photomicrography.\***—Since 1866 it has been the generally received doctrine, says Dr. R. Neuhauss, that the position of the filter for producing monochromatic light is of the greatest importance. This doctrine, laid down by Moitessier and followed by all other writers, states that the maximum of absorption is attained when the filter is placed before the collecting lens, and its minimum when inserted between the lens and its focus.

By experiments with a yellow disc placed in the position of the object on the stage and using an ordinary non-orthochromatic silver-bromide-gelatin dry plate covered with a silk-paper sensitometer in the one case, and inserting the yellow disc between the light and the lens in the other, it was found that the two images were exactly alike in every respect. For both the exposure was exactly 15 minutes, and in both negatives the numbers could be read when the layers of silk paper were not more than sixteen.

Similar results ensued from using a layer of a saturated solution of picric acid 3 mm. thick. Hence it is quite indifferent whether the filter be placed near the lens or its focus.

#### (6) Miscellaneous.

**The Microscope in Geology.**—A course of twelve lectures on the Microscope in Geology (with special reference to the structure and origin of the stratified rocks), is now being delivered by Professor H. Alleyne Nicholson in the British Museum (Natural History), Cromwell Road, on Mondays, Wednesdays, and Fridays, at 3 p.m., beginning 6th October and ending 31st October, 1890. Admission to the course is free.

#### B. Technique.†

##### (1) Collecting Objects, including Culture Processes.

**Cotton-wool as a substitute for Silk in Bacteriological Work.‡**—Dr. E. Braatz finds that animal products have a much greater affinity for mercury than vegetable, and for this reason advises that cotton-wool threads be used instead of silk threads in bacteriological work.

**Effect of highly concentrated Media on Bacteria.§**—Prof. H. Buchner replies to Metschnikoff's assertion that the inhibitive influence of the body fluids on micro-organisms is to be ascribed to the greater concentration of these fluids. The author first remarks that the germicidal property of serum is quite extinguished by heating it to 55° for half an hour, although its degree of concentration remains quite unchanged. He then gives the results of experiments made with highly concentrated media, viz. blood charged with 23 per cent., and also with 40 per cent. of cane sugar. In both instances, although there was at the very

\* Zeitsch. f. Wiss. Mikr., vii. (1890) pp. 20-2.

† This subdivision contains (1) Collecting Objects, including Culture Processes; (2) Preparing Objects; (3) Cutting, including Imbedding and Microtomes; (4) Staining and Injecting; (5) Mounting, including slides, preservative fluids, &c.; (6) Miscellaneous.

‡ Centralbl. f. Bakteriöl. u. Parasitenk., viii. (1890) pp. 8-9.

§ T. c., pp. 65-9.



outset a slight diminution of the Bacteria, they soon grew well enough.

Other two sets of experiments were made with 10 per cent. sugar and 10 per cent. pepton, each mixed with 10 volumes of blood. Both of these series as compared with a control series without sugar, showed that the addition made practically no difference.

Hence it is obvious that neither the concentration of the medium nor the too sudden transition of the Bacteria to an unaccustomed medium makes any difference to the result.

#### (2) Preparing Objects.

**Method of Preparing Mucous Gland of Prosobranch Molluscs.\***—M. F. Bernard found difficulty in obtaining reagents which were not either unable to coagulate the mucus or which were not too energetic, and so disformed the cells. He found, however, three mixtures which acted well, part of the gland being removed from the mantle as rapidly as possible. These were strongly acidulated picro-sulphuric acid; chloride of ruthenium of such strength that the solution is a clear red colour; this was the best of the reagents employed, but unfortunately the author was not able to get as much of it as he wished; it greatly aids dissociation with needles. The third mixture was made of 200 grammes of distilled water, 10 of alcohol at 90 per cent., 5 of glycerin, and 10 of acetic acid; this solution facilitates the staining of the elements with methylen-blue. Fragments thus fixed were teased in 38 per cent. alcohol, osmic acid at 1/10,000, or the acid mixture just mentioned. The last gave particularly good results with animals from Naples which had been already fixed by alcohol or various other reagents.

**Mounting Insect Eggs to study the Embryo.†**—Mr. E. A. Hill describes a method devised by himself, which he has used for two or three years past, in collecting and preparing the eggs of Lepidoptera for the microscopic examination of the embryo in its various stages of development.

In summer evenings, when working with the Microscope, the window being open, as is usually the case, moths frequently fly in attracted by the light; and when pursuing this line of investigation Mr. Hill has on hand a number of pasteboard pill-boxes (size is not important, but some which happened to be at hand were about 1 in. deep, and 3/4 in. in diameter). The moths are easily captured, after which each is placed in a separate box, with a reference letter on the cover. The next morning a number will usually be found to have laid eggs. These eggs are divided into as many equal parts as he anticipates there are days in the period of incubation, placing each portion in a separate homœopathic phial, the phials being about 1 in. high. The corks are marked with the reference letter entered in the record book, and, in addition, the phials are numbered consecutively from 1 upwards. The corks are inserted lightly, so as to allow air to enter the phials. Phial No. 1 is then filled at once with carbolic acid, filling No. 2 on the second morning, No. 3 on the third morning, and on the last day filling the phial containing the newly

\* Ann. Sci. Nat., ix. (1890) pp. 305-6.

† The Microscope, x. (1890) pp. 208-10.

hatched larvæ, entering in the note-book the time required for hatching. Meanwhile, if it is desired, and this is the better plan, the moth is mounted after the usual manner of entomologists, on an entomological pin, and preserved in a cabinet with the same reference letter, so that the species can be determined at leisure. The carbolic acid renders the eggs perfectly transparent, or at least does so in the cases which have come under notice, and hence the embryos can be observed in the various stages of development. Mr. Hill mounts in benzol-balsam direct from the carbolic acid, and to prevent the crushing of the eggs sometimes uses three supports for the cover-glass placed triangularly between it and the slide. Three are better than four, as three points afford a more uniform bearing for the cover than four, on the well-known principle of the three-legged stool.

For the supports either small beads are used, or, if special thicknesses are required for the supports, they can be made by drawing out a fine thread from a piece of glass tube by means of a spirit-lamp, after which small pieces can readily be broken off. Tin-foil also makes good supports. For example, cut a strip about 1 in. square, and roll it into a tight roll 1 in. long; it should then be flattened between two glass slides to a uniform thickness, when little square pieces can be readily clipped off with a pair of scissors and used instead of the beads. The thickness of the roll can be varied, and the little squares can also be reduced in thickness by removing one or more layers of the tin-foil until of the proper size.

Theoretically, a series of eggs beginning with No. 1 and running up consecutively should show a progressive development of the embryo, but practically there is not always as much regularity in the series as we could look for. Probably the eggs first laid develop first, and twelve hours' difference in the time of laying the first and last egg, if the whole period of incubation only amounts to a few days, may make some difference. When, however, we have several eggs in each phial, no trouble will usually be experienced in getting a good progressive series by making a judicious selection from each bottle, in which case the selected specimens may be mounted in proper order on a single slide.

**Preparation of Eyes of Lobsters.\***—Mr. G. H. Parker describes a method of staining nerve-fibres which he discovered while experimenting with Weigert's hæmatoxylin. The method consists in a cautious use of Schällibaum's fixative; the one employed consisted of three parts of oil of cloves, and one part of Squibb's flexible collodion; the mixture should be allowed to stand a week before being used. A moderate amount is applied to the slide, and the sections in paraffin are placed on it; the slide and the sections are now subjected to a temperature of 58° C. for fifteen minutes, and this is a point which must be carefully attended to. The slide must next, while warm, be thoroughly washed with flowing turpentine, which can be conveniently applied from a small wash-bottle; all the paraffin should be removed from the slide before it becomes cool. When the slide is cool the turpentine may safely be replaced by alcohol, 95 per cent., then 70 per cent, 50 per cent., and

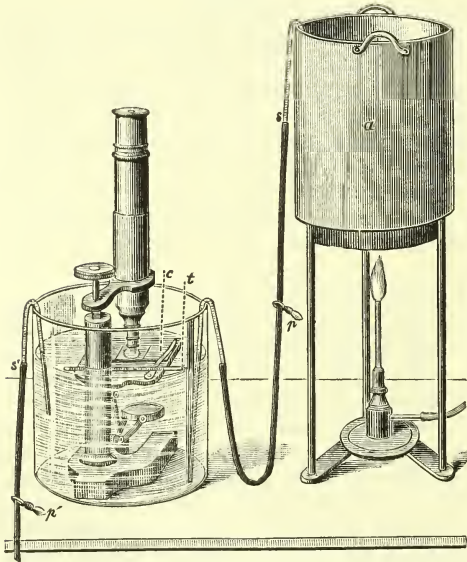
\* Bull. Mus. Comp. Zool., xx. (1890) pp. 3-4.

35 per cent., and finally it may be immersed in water. Sections of optic nerve mounted on slides and carried into water must be treated for about half a minute with an aqueous solution of potassic hydrate (1/10 per cent.), then thoroughly washed in distilled water, and transferred to Weigert's hæmatoxylin, in which they should remain for about three hours at 50° C. After distilled water and grades of alcohol they may be cleared in turpentine and mounted in benzol-balsam. Each nerve-fibre so treated has a distinct blue-grey outline.

**Methods of Recognizing Cysticerci of *Tænia saginata*.**\*—M. A. Laboulbène has a note on the means of recognizing the cysticerci of *Tænia saginata*, which are the cause of "measles" in veal and beef, and which are often so difficult to detect on account of the rapidity with which they dry on exposure to the air. He finds that meat which has become quite leathery will easily reveal the *Cysticerci* if it contain any, by being placed in water acidified with acetic or nitric acids, or in a mixture of water, glycerin, and acetic acid. By this means the parasites can always be detected, and if the meat be carefully heated to 50° or 60° C. it is always fit for human food.

**New Method for Examining microscopically the Elements and Tissues of Warm-blooded Animals at their physiological temperature.**†—This method, devised by M. L. Ranvier, essentially consists in

FIG. 78.



placing both the Microscope and the preparation to be examined in a bath of warm water (36° to 39° C.) But like most practical things the details are more important than the principle. Thus the Microscope must be of a simple model. As the preparation is to be examined under water an immersion objective with or without correction must be used. The preparation must be carefully protected from water by running paraffin round the cover-glass. Before using the objective it must be warmed up to 40° C., otherwise a thick fog will spread over the face of the lens. The Microscope is placed in a

flat-bottomed glass vessel about 0·12 m. high and 0·14 m. in diameter. This contains *distilled* water, heated up to 40°, in such quantity that its surface is from 0·5–1 cm.

\* Comptes Rendus, xxi. (1890) pp. 155–7. † Op. c., ex. (1890) pp. 66–9 (1 fig.).



above the level of the stage. A thermometer placed by the side of the preparation indicates the temperature of the latter.

The most convenient temperature for observations ranges from 37°–38°, and if observations are to be maintained longer than eight or ten minutes it is necessary not only to add warm water, but to remove the surplus in order that the original level may be maintained. This can be effected as in the illustration by means of two siphons, or by placing the glass vessel within one which is larger but not so high.

By means of this apparatus the author states that he has made more observations in a month than in the past twenty years with the old arrangements.

**Microchemical Tests for Alkaloids and Proteids.\***—M. L. Errera points out the want of a general test for the discrimination of alkaloids and proteinaceous substances. Although many alkaloids are readily detected by special reactions, yet Raspail's proteid-reaction (a red colour produced by sugar and sulphuric acid), and Millon's reaction, are both also produced by certain alkaloids. The best general distinctive tests for these two classes of substances are their different behaviour towards (a) absolute alcohol, (b) one gr. of tartaric acid in 20 ccm. of absolute alcohol, (c) 0.2 ccm. of hydrochloric acid in 5 ccm. of distilled water and 95 ccm. of absolute alcohol. In these three reagents all alkaloids are readily soluble, while proteinaceous substances are either entirely insoluble, or at all events leave a residue behind, even after very long treatment.

**Reactions for Lignin.†**—Herr R. Hegler discusses in great detail the various reagents used for the micro-chemical detection of lignified membranes. He divides those already in use into three groups, viz.:—(1) Those which react with vanillin, but not with coniferin,—thallin; (2) Those which react with coniferin but not with vanillin,—phenol-hydrochloric acid, thymol-hydrochloric acid; (3) Those which react with both vanillin and coniferin,—all the other reagents for lignin. Thallin,  $C_9H_6NOCH_3H_4$ , is an extraordinarily delicate reagent for lignified tissues, the vanillin assuming an intense orange-red colour. A new reagent recommended, with the same properties, is tolulendiamin,  $C_6H_3(CH_3)(NH_2)_2$ , used in a concentrated aqueous solution with a trace of hydrochloric acid; it stains lignified membranes a dark orange. Vanillin he regards as a product formed out of coniferin by the activity of the protoplasm; the process being of the nature of fermentation with secondary oxidation. The production of lignin,  $C_{18}H_{24}O_{10}$ , out of cellulose may be represented by some such equation as this:— $4C_6H_{10}O_5 = C_{18}H_{24}O_{10} + C_6H_6O_5 + 5H_2O$ ; the  $C_6H_6O_5$  may then be completely oxidized into carbon dioxide and water, or may pass over into such substances as tannins.

**Fixing and Staining of Leucoplasts and Protein-crystalloids.‡**—Dr. A. Zimmermann recommends a concentrated alcoholic solution of

\* 'Sur la distinction microchimique d. alcaloïdes et d. matières protéiques,' Bruxelles, 1889. See Bot. Ztg., xlviii. (1890) p. 232.

† Flora, lxxiii. (1890) pp. 31–61 (1 pl.). Cf. this Journal, 1889, p. 606.

‡ Beitr. z. Morph. u. Physiol. d. Pflanzenzelle, Heft 1, 79 pp. and 2 pls., Tübingen, 1880. Cf. *supra*, p. 617.



corrosive sublimate for fixing the leucoplasts, e. g. in the epidermal cells of the leaves of *Tradescantia discolor*; the leucosomes themselves not being in any way changed by the sublimate. Good results were also obtained—though not so good—with concentrated alcoholic solution of picric acid, and with alcohol alone. With small pieces this immersion in the sublimate solution is sufficient. To prepare for the microtome they should then be placed first in pure alcohol, then for twenty-four hours in a mixture of three parts xylol and one part alcohol, then as long in pure xylol, then in a solution of paraffin in xylol saturated in the cold, finally in pure paraffin. For staining, Altmann's method \* with acid-fuchsin was found to be the best; but a special modification of it is described in detail. Iod-green, cyanosin, and dahlia may also be used.

For fixing the cell-granules the author uses either a concentrated alcoholic solution of picric acid or 3 per cent. nitric acid. They may then be stained with acid-fuchsin by Altmann's method, which colours the granules an intense red, while the chloroplasts and nucleus are left quite colourless.

For staining the proteid-crystalloids, a method is employed termed by the author the acid-fuchsin method B. The section is first of all dehydrated by alcohol, and then placed in xylol or in xylol-Canada-balsam. The leucoplasts are fixed by picric acid or sublimate, and the section then stained with acid-fuchsin. While the nuclei and nucleoli remain perfectly uncoloured, the crystalloids take up an intense red. Good results were also obtained by the ordinary Altmann's acid-fuchsin method; also by fixing with concentrated aqueous or alcoholic solution of sublimate, aqueous or alcoholic solution of picric acid, 5 per cent. solution of potassium bichromate, or with Müller's fluid.

### (3) Cutting, including Imbedding and Microtomes.

**Imbedding Vegetable Preparations in Paraffin.**†—Herr L. Koch discusses at great length and with copious detail the proper method of imbedding vegetable preparations in paraffin. After a critical survey of various methods of paraffin imbedding, the author gives a general outline of his views on the subject, and then proceeds to give the minutiae requisite for obtaining a satisfactory result in special cases. His views, however, are tolerably simple, and do not seem to differ materially in practice from those of other people who apply themselves to vegetable anatomy.

The general proposition, on which much stress is laid, and the obvious inference therefrom, is one which occurs to any person after a very small amount of practice. It is that the imbedding mass must be made to penetrate into cells and intercellular spaces, and in order to do this the air and water must be thoroughly and completely removed. This is effected by immersing the objects in spirit, the strength of which is gradually increased up to absolute alcohol. The objects are then saturated with paraffin dissolved in chloroform. The saturation is effected by gradually increasing the thickness of the paraffin mixture; when a

\* Cf. this Journal, 1888, p. 147.

† Jahrb. f. Wiss. Bot. (Pringsheim), xxi. (1890) pp. 367-468.

suitable consistence is attained, the block is cut up with a medium sized Jung's microtome. The description of this well-known section-cutter seems somewhat superfluous. The sections, which vary from 0.03 to 0.005 mm., are fixed to the slide with the collodion and clove oil mixture and the paraffin dissolved out with turpentine. The turpentine is dissolved out effectually with alcohol, and this in its turn with water. The specimens are then mounted in glycerin or Kaiser's glycerin jelly. Staining and mounting in balsam are passed over in a very few words.

#### (4) Staining and Injecting.

**Laboratory Notes.\***—Mlle. Leclercq points out that it is advantageous to stain sections so as to show the micro-organism and the tissues as well. This can be effected by first staining with borax-carmin, and then using Gram's or Bizzozero's method. The steps to be followed are, first stain the section with borax-carmin, followed or not by decoloration with hydrochloric acid according to the result that is desired; washing in water; staining with Ehrlich violet; washing in absolute alcohol followed by the Lugol iodine solution; washing in absolute alcohol followed by decoloration in 1 per cent. chromic acid; then absolute alcohol, oil of cloves, and balsam.

For staining embryonic blood-corpuscles of birds, so as to distinguish them from other embryonic elements, the authoress gives the following method:—(1) Overstain with fuchsin; (2) moderate decoloration with 1/3 to 1/5 aqueous solution of acetic acid; (3) washing in water; (4) rapid staining with weak solution of malachite-green; (5) dehydration in absolute alcohol; (6) clearing up in oil of cloves or origanum oil according to the degree of staining; (7) mounting in balsam.

By this method the malachite-green combines with the fuchsin in the embryonic tissues, which become violet-coloured, while the blood-corpuscles and the karyokinetic figures are red.

The foregoing, although good for birds, is not successful for mammals, and for these the authoress adopts a method of triple staining, wherein she uses Congo red. This method consists (1) in staining for 10–15 minutes in a very weak solution of Congo red; (2) washing in water; (3) staining with Ehrlich's violet, followed by decoloration according to Gram's or Bizzozero's method; (4) staining with alcoholic eosin; (5) Dehydration in absolute alcohol, then oil of cloves and balsam.

In this case the blood-globules are stained an orange-yellow.

**Apparatus for Impregnating Tissues, &c., and for making Esmarch Tubes.†**—Dr. M. Herman describes an apparatus which is serviceable for histological, pathological, zoological, and bacteriological purposes. It consists of a water-wheel R (fig. 79) which revolves in a box. On one side of its axis is the handle M, and on the opposite side is an open metal case D, the latter being for the reception of a test-tube T, which is intended for the Esmarch cultivation method. The box rests on the

\* Bull. Soc. Belge Micr., xvi. (1890) pp. 61–5.

† Centralbl. f. Bakteriologie u. Parasitenk., vii. (1890) pp. 55–7 (2 figs.).

plate S, and this can be moved up and down by means of the screw V. The hopper E is divided into two compartments *a* and *b*, so that the water, which is introduced into the hopper through a pipe, may pass

FIG. 79.

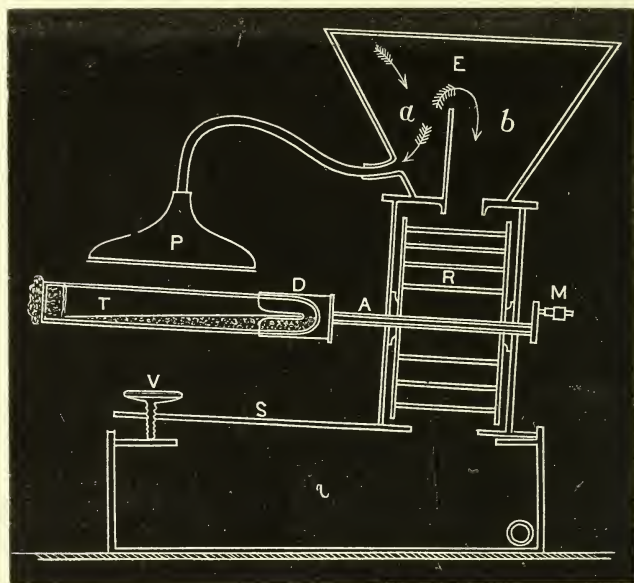
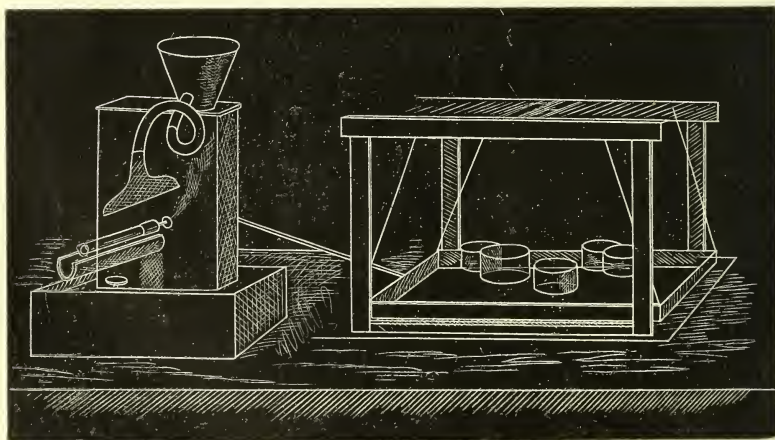


FIG. 80.



both on to the wheel R and through a tube P on to the test-tube. The surplus water collects in a reservoir, from which it passes out through an overflow pipe (not shown in the illustration).



By means of a thin metal rod, the handle M imparts a to-and-fro movement to a shallow rectangular metal tray, which is suspended by four wires to a wooden framework. The regular to-and-fro movement of the tray is effected by two small metal forks which act as guides. In fig. 80 is given a general view of the apparatus. In the tray are placed glass capsules to contain the pieces of organs or tissues which are to be stained, washed, hardened, or impregnated. In order to set the tray in motion, the plate S is levelled horizontally by the screw V, and water through a lead pipe is run into the compartment *b* of the hopper, so that it strikes against the wheel R and sets it in motion. The rapidity of the wheel's motion is regulated by different calibre of tube, &c.

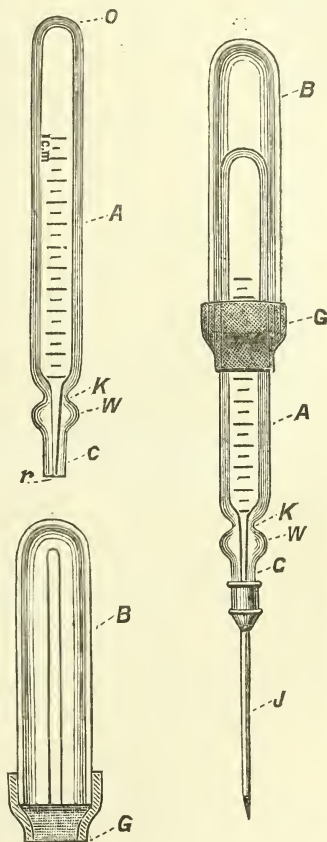
As an example of how the apparatus works, it is stated that sublimate solutions, &c., can be extracted in two days, the spirit only requiring to be once removed.

To make gelatin tubes for the Esmarch method, the screw V is made to give a greater or less inclination to the apparatus, according as there is more or less gelatin in the tube (see fig. 79). The water stream is then run into the *a* compartment, so that it first runs out through the pipe P to cool the test-tube T, and then passes over the barrier into the *b* compartment, and so sets the wheel in motion.

**Injection-syringe for Bacteriological Purposes.\*** — This syringe, which is the invention of E. Stroschein, consists of two glass tubes which are somewhat like ordinary test-tubes though smaller. The inner, narrower tube is prolonged at its front end to a conical point on which to fit the canula; at its posterior end is a small hole. The outer tube simply fits over the inner, and the two are connected with a caoutchouc band.

The syringe is filled by merely dipping the canula into the fluid to be injected, and then drawing the outer tube back as far as the elastic band permits, and so by creating a vacuum the fluid is sucked into the inner tube. Of course the fluid is injected by merely reversing the action. This little instrument, which is very moderate in price,

FIG. 81.



\* Mittheil. aus Dr. Brehmer's Heilanstalt, 1889. Cf. Centralbl. f. Bakteriöl. u. Parasitenk., vii. (1890) pp. 746-7 (3 figs.).

fulfils every requirement for bacteriological work as it is easily taken to pieces, and its constituent parts easily disinfected by dry or moist heat, or by chemical agents.

**Staining the Flagella of Bacteria.\***—Prof. F. Loeffler communicates a much improved method for staining the flagella of micro-organisms, the key to the procedure consisting in the greater or less acidity of the mordant. The quantitative differences in the reaction of the mordant are extremely slight, and vary with the different bacilli.

The best results were obtained from using 10 ccm. of tannin solution (20 + 80 water) to which had been added 5 ccm. of cold saturated ferrosulphate solution and 1 ccm. of aqueous or alcoholic solution of fuchsin, methyl-violet, or woolblack. This last pigment is used for dyeing wool without a mordant, and when dissolved in water is of a blue-black colour.

The foregoing solution, especially when made up with fuchsin, is to be regarded as the stock solution, and one which will stain the flagella of certain micro-organisms such as *Spirillum concentricum*, but for others the addition of an alkali or an acid is necessary. Thus, for typhoid bacilli 1 ccm. of 1 per cent. caustic soda solution is required, while *Bacillus subtilis* needs 28–30 drops, the bacillus of malignant œdema 36–37 drops, and so on. For cholera bacteria it is necessary to add 1/2–1 drop of sulphuric acid, for *Spirillum rubrum* 9 drops, to the 1 per cent. soda solution, the quantity of which is not however mentioned.

This is the mordant and it differs from that previously given by the author by certain omissions.†

The whole procedure now goes as follows. A small quantity of the pure cultivation is mixed up in distilled water, and with some of this the cover-glass is lightly smeared with a platinum loop. It is of the utmost importance that the cover-glass should be perfectly clean and free from grease or other impurities. The covers should be boiled in strong sulphuric acid, washed in distilled water, and having been immersed in ammoniated alcohol, dried on a clean cloth.

The bacteria, when spread on, are fixed in a flame. For staining flagella this is absolutely necessary, but it is also as important not to over-heat. The correct amount of heat may always be estimated by holding the cover between the thumb and forefinger, instead of using forceps; by this device overheating is avoided. While still warm, the mordant is applied. The cover-glass is then heated until it begins to vaporize (1/2–1 minute). It is then successively washed in distilled water and absolute alcohol.

The staining solution is then dropped on in quantity sufficient to cover the cover-glass, which is again warmed until the solution vaporizes and then the cover-glass is washed in distilled water.

The composition of the staining solution is ordinary neutral anilin water in which solid fuchsin is dissolved to saturation. To this as much of a 1 per cent., or still better 1 per thousand, soda solution is added as to bring it almost to the point of precipitation. Although it is not

\* Centralbl. f. Bakteriöl. u. Parasitenk., vii. (1890) pp. 625–39 (8 photographs).

† See this Journal, 1889, p. 711.

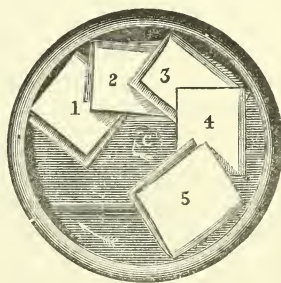
absolutely necessary to add the soda solution better results are thereby obtained.

With regard to the necessary addition of alkali or acid to the mordant, the author points out that there is some connection between this fact and the formation of acids and alkalies by certain bacteria: for the acid-forming bacteria required the addition of alkali, and the alkali-producers the addition of acid before they would stain.

Another interesting observation made by the author was that some bacteria possess tufts of flagella, and these are well demonstrated in the photographs accompanying this paper.

**Staining Spinal Cord with Naphthylamin Brown and Examining with the Dark-field Illumination.\***—For preparing serial sections of spinal cord, Herr O. Kaiser finds the following procedure useful. The sections imbedded in celloidin are removed from the knife with filter paper and placed at once in the following staining solution:—Alcohol, 100; water, 200; naphthylamin brown, 1. The sections folded up in filter paper are arranged in a glass capsule, as shown in the figure. Herein they may remain for some hours to two days. The sections when removed from the staining fluid are washed with 96 per cent. spirit and then placed on the slide. When the excess of alcohol is removed the sections are fixed to the slide by blowing ether vapour over them through a pipette bottle. As the sections become a little creasy, a few drops of absolute alcohol are run over them, after which the slide is placed in origanum oil, then in xylol, and the specimen finally mounted in balsam. Naphthylamin brown colours the chromophilous cells dark brown, while the chromophobous cells appear as bright objects on a dark ground. The blood-corpuseles are of a coppery red hue. In order to distinguish between the grey and white matter, it is necessary to use the dark-field illumination. This is easily done by inserting a stop in the Abbe condenser. The white substance now shows up as a bright yellowish-brown, while the grey matter is dark brown, all the finer details being quite clear. The blood-corpuseles are of a bright scarlet hue, so that the vessels seem injected.

FIG. 82.



**Staining the Endings of Motor Nerves with Methylen-blue.†**—Prof. A. S. Dogiel, after recommending this method, and alluding to the usual procedure, states that it may be simplified and improved in the following manner. The tissue removed from living or recently killed animals is placed on a slide or in a watch-glass containing some drops of aqueous or vitreous humour. To this are added two to three drops of a 1/15 to 1/16 per cent. solution of methylen-blue made up with physiological salt solution. In this condition the preparation is left exposed to the action

\* Zeitschr. f. Wiss. Mikr., vi. (1889) pp. 471-3 (1 fig.).

† Arch. f. Mikr. Anat., xxxv. (1890) pp. 305-20 (1 pl.).



of the air, but protected from the dust by a large watch-glass. The preparation may be examined from time to time under a low power to see how the staining is getting on. The effect varies extremely: thus the endings of motor nerves stain in 5-10 minutes, while the nerves in the retina require two or three hours or even longer.

As the staining disappears in a comparatively short time, it becomes necessary to fix the pigment. For this purpose picrate of ammonia is advised. This saturated aqueous solution precipitates the methylen-blue in a finely granular condition, rendering the rest of the tissue highly transparent. The length of time required for fixing the stain varies of course with the thickness of the tissue; some specimens are fixed in 20 minutes, while others require as long as 12 hours. The preparations are then mounted and examined in a mixture of equal parts of glycerin and distilled water.

Preparations which have been stained with methylen-blue may be hardened by immersion for 2-3 hours in a saturated spirituous solution of picrate of ammonia and then, having been imbedded in elder-pith or liver, sectioned with a razor. The sections are placed in glycerin. Or the stained tissue may be frozen and then sectioned.

By the foregoing method the author has obtained very excellent results, judging from the illustrations which accompany the text, from muscles of Amphibia and Reptilia. The procedure is less complicated than that where the stainings are obtained by injecting the vascular system with a solution of methylen-blue.

#### (5) Mounting, including Slides, Preservative Fluids, &c.

**Arranging Diatoms.\***—Mr. Cunningham states that he arranges selected diatoms by transferring directly from a strewn slide to the exhibiting slide by the aid of a "Kain's mechanical finger" attached to a Beck's 1/2-in. objective, the slide being manipulated by the left hand, and the bristle being directed into the field from the left-hand side. This method, he says, counteracts the effect of reversal of image, enabling every desired movement to be accomplished with ease and certainty. The right hand assists in racking the diatom from the slide high enough to clear the edge of the cover-glass upon which the diatoms are to be fixed. Very minute species are selected and isolated by this means.

**New Mounting Dammar.†**—A very superior mounting medium was accidentally discovered by adding by mistake liquor potassæ to a thick solution of benzol-dammar. After the lapse of some months, the jar, with a beautifully clear zone of some sort of gummy material superimposed upon a white one, was discovered. The clear zone, some 6 ounces, was drawn off and tested as to drying and other properties. It was found that it dried slowly, but ultimately set very firmly. Placed on a slide heated to a point that instantly vaporized water, it dried without forming a bubble. Used as a mounting medium on a hot slide, no bubbles were formed, and while in bulk the colour is somewhat darker than Canada balsam, in ordinarily thick mounts it is almost imperceptible.

\* Journ. N.Y. Micr. Soc., vi. (1890) p. 60.

† St. Louis Med. and Surg. Journ., lviii. (1890) p. 37.

**Alcoholic Method of Mounting Bryozoa.**\*—Miss V. A. Latham, when adopting the alcoholic method of mounting, first rings a cell of the brown cement and allows it to harden thoroughly; then, she says, "cover this entirely with balsam and benzol, and when dry again make it slightly sticky by a thin line of balsam which fastens down the cover-glass. Ring over all another layer of the last cement, and when dry use brown cement to completely seal the mount which, when dry, can be finished as the mounter wishes. Or, instead of the above method, after the organisms have been fixed and coloured, pass them through alcohol 30 per cent., 50 per cent, 70 per cent., and absolute, the last at least twice, and let them stand covered for 24 hours. Replace the spirit by pure benzol, remove about a tenth of the alcohol in which the organisms are placed with a pipette, and replace by the same amount of benzol; repeat this a number of times (about twelve) at intervals from 10 to 30 minutes. Great care must be taken that the benzol mixes thoroughly. After the last addition pour the fluid off and substitute pure benzol. At the end of 24 to 48 hours in the benzol, according to the size of the object, a fifth part of the Canada balsam dissolved in benzol is added; this is repeated at intervals of from a quarter to half an hour; the objects may now be preserved in the tubes till wanted, or mounted at once. In mounting, care must be taken that each drop holds in suspension a sufficient variety of the organisms. The method is not quite so tedious as it appears from the reading."

**Kaiser's Glycerin-Gelatin.**†—One part of the best French gelatin is macerated in six parts by weight of distilled water for about two hours. To these are added seven parts by weight of pure glycerin, and to every 100 grams of the mixture 1 gram of pure carbolic acid. The whole is then warmed for 10–15 minutes with constant stirring until all the lumps and flakes which form after the addition of the carbolic acid have disappeared. The decoction is then filtered through the finest glass-wool, which has been previously washed in distilled water, and placed still wet in the funnel.

\* *Microscope*, ix. (1889) p. 141.

† *Bot. Centralbl.*, i. p. 25. Cf. *Jahrb. f. Wiss. Bot.* (Pringsheim), xxxi. (1891) p. 400.

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# JOURNAL

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OF THE

## ROYAL MICROSCOPICAL SOCIETY;

CONTAINING ITS TRANSACTIONS AND PROCEEDINGS,

AND A SUMMARY OF CURRENT RESEARCHES RELATING TO

ZOOLOGY AND BOTANY

(principally Invertebrata and Cryptogamia),

MICROSCOPY, &c.

*Edited by*

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**WILLIAMS & NORGATE.**

LONDON AND EDINBURGH.



## (3) Illuminating and other Apparatus.

**Substages for Students' Microscopes.**—The following interesting correspondence appeared in the pages of the 'English Mechanic':\*—

"Under this heading Messrs. Watson and Sons reproduce, if I mistake not, a figure of the underpart of the stage of one of their Microscopes, which was published in the 'E.M.' a year or two ago. They appear to claim for it some originality of design, and bring it forward again, as if it wholly superseded the necessity for Swift and Sons' rival production, notwithstanding the fact that such rival production is recommended by the Secretary of the Quekett Microscopical Club (vide Report of Proceedings Q.M.C., 'E.M.,' p. 185).

Examining Messrs. Watson's figure with a somewhat experienced eye, I note that the substage is not mounted on a substantial tail-piece, but is merely connected with the stage proper by a screw, on which it pivots out of the axis, the axial position being roughly secured by a stop-pin; the centering is then effected by the two projecting screws on the circumference of the substage acting upon a movable inner ring or tube in which the condenser or other apparatus is applied.

This system of pivoting the substage has long been in use on the Continent for all classes of Microscopes, from the most elaborate down to the commonest types of so-called students' instruments. It is a bad system—bad in the Johnsonian "leg-of-mutton" sense—bad from every point of view.

A focusing and centering substage can hardly be too substantially connected with the Microscope, for it has to suffer more rough handling than any other part of the instrument. The fewer movements it has beyond those actually needed for use, the better; for every additional joint or slide-bearing brings in its quota of unsteadiness, making additional demands on the observer's watchfulness and patience in securing exactness of adjustment.

The only satisfactory arrangements of substage yet devised act by rack-and-pinion on a fixed tail-piece.

Many attempts have been made, both in Europe and America, to devise means for focusing the condenser of a less expensive character than the rack-and-pinion; but they have all been found radically bad in practice by those who were familiar with the conveniences of the rack-and-pinion, and who have been called upon to test the various systems devised in substitution.

In the older models of drum Microscopes (*Microscopes à tambour*) of Georges Oberhäuser—who was probably the first to apply mechanism to focus the substage condenser of the achromatic Microscope—the substage socket was a fixture beneath the stage, and the condenser tube sliding in it was provided with stud-pins on either side, on which a forked lever engaged, the free end of the lever projecting slightly through a vertical slot in the drum base, so that by its movement up or down the condenser was adjusted beneath the object. This arrangement was generally combined with some primitive means of centering the condenser with a screw-driver, such centering being on the "once for all"

\* Engl. Mech., lii. (1890) pp. 228, 229, 251, 271.

principle, not to be altered during the process of observation, the very moment when it was most essential that it should be possible to alter it. In later constructions Oberhäuser applied a sort of gimbal to the forked lever to secure an easier sliding motion to the condenser for focusing; but the system of direct push or pull of the condenser tube within the substage socket, with or without gimbal, was always bad, and frequently ended in injury to the mechanism.

Nachet and others improved upon Oberhäuser's arrangement by applying under the stage a tail-piece having a dovetail groove in which a slide carrying the substage was actuated by a stud-pin and lever projecting laterally.

More recently, and particularly in America, the lever has been omitted by many opticians, and the microscopist has had to slide the fitting up or down the tail-piece by small knobs on either side. Even the most recent student's Microscope—that of Swift and Son, to which I have above referred—has this most inconvenient arrangement.

It would be too tedious to describe the various other systems that have been applied to avoid the expense of the rack-and-pinion. They consist generally of some form of screw-action, and probably the best of them is now made by a leading Paris optician.

Probably one of the worst focusing arrangements recently designed for the substage condenser is that proposed—if not invented—by Mr. E. M. Nelson. It consists of a substage socket fixed to the under face of the stage; a spiral slot is cut in the socket, in which a stud-pin on the side of the condenser tube moves up or down as this tube is rotated by hand. The focal adjustment of the condenser obtained by this motion in a spiral direction is very unsatisfactory. I suppose Mr. Nelson himself now recognizes its inferiority.

Until our opticians face the matter by applying the rack-and-pinion motion to a fixed tail-piece for students' Microscopes, I fear we cannot credit them with the serious intention of meeting the present requirements of students. It would appear that English makers are constantly handicapped by their own desire to supply a superabundance of "finish" in non-essentials, to the neglect of much-needed focal and centering substage adjustments, which every student would soon learn to appreciate.

If a beginner now asks the advice of an expert as to the microscopical outfit he should obtain, on explaining his requirements to the optician, he will probably be met at once with some such statement as follows:—"You can have a substage with rack-and-pinion and centering adjustments; but these things are only fitted to the better class of Microscopes commencing at the price of £x"—the *real* drift of which is to force the student into a larger outlay—to make what is commonly known in business as a "substantial transaction."

The fact seems to be that in England the manufacture of fairly good Microscopes is in the hands of so few opticians that they believe they can wholly control the trade. It is for responsible teachers in medical schools to combine and authoritatively formulate the *desiderata* of a student's Microscope—the laws of *demand* and *supply* will do the rest, though they should lead us to engage foreign opticians to drive English Microscopes of the students' class out of the field. Such a course does not seem patriotic; but I fear our opticians are proof against friendly

council on the matter. They can only be made to move in a popular direction when they find such firms as Zeiss's establishing themselves—as they are now doing—in London.

Microscopists naturally want the highest class of optical appliances they can acquire with the outlay at their disposal. They feel that inferior means conduce to inferior results only. The demand for the improved instruments—apochromatic objectives, compensating eye-pieces, projection eye-pieces, &c.—exists. The supply of these novelties by English opticians has been so extremely limited, and withal so tardy, that it has amounted to nothing of any importance, so we have to rely mainly on Zeiss, of Jena. That great firm, employing some four hundred people, has now established two representative houses in London, and it behoves our opticians to look to their laurels, for the competition promises to be the most serious that has occurred during this generation. Moreover, the competition will not be limited to the production of microscopical apparatus. The firm of Zeiss has now brought out a series of new photographic lenses which will, no doubt, demand and obtain the keenest attention of those who are interested in the progress of photography.—MICROSCOPIST.”

“In submitting the student's Microscope, made by Messrs. Swift, at the last meeting of the Quekett Microscopical Club, I expressly disclaimed any idea of novelty in the parts of the instrument. My sole intent was to show how easily some better form of centering fitting could be applied in place of the makeshift understage tube, which is, *pace* Messrs. Watson, after all, the only thing provided in the vast majority of students' instruments. These gentlemen state that the form adopted by them in their Edinburgh model was designed by Dr. Edington, about two and a half years ago; but Messrs. Crouch have, for a much longer period, made a very similar form, and so has Reichert, of Vienna. The great drawback, in my opinion, to this arrangement is that it is in the way when turned aside; and, moreover, I do not think it can be contended that a substage supported by, and swinging on a single screw, is as steady and free from tremor as one with long bearings moving in a dovetailed guide. Mr. Nelson, whose criticism in these matters is always deserving of great respect, drew attention to the absence of a rack-and-pinion focusing adjustment in Messrs. Swift's instrument, but one is very easily fitted if required, and for the class of work this instrument is intended for it is by no means necessary.

I shall be very pleased if a discussion on this subject leads to the abolition of the non-centering substage tube, and the substitution of a more scientific arrangement, by whomsoever it may be designed.—GEORGE C. KAROP.”

“When writing previously on this subject we had no thought of inducing a controversy; but we must, in justice to ourselves, repudiate the inference of your correspondent, ‘Microscopist,’ that the substage of the Edinburgh Student's Microscope is not rigid. One would imagine, from your correspondent's letter, that it is a useless arrangement; but his information is evidently gleaned from the engraving that accompanied our previous letter, which he has not correctly interpreted, and certainly not from any practical working with one of our instruments. We would point out that when the substage is in the axial position, it fits into a

special collet, which grips it from beneath, so that there cannot be any shake; it is not merely 'roughly secured by a stop-pin,' as 'Microscopist' assumes. In fact, his remarks consist of an expression of superficial opinion, formed from an incorrect conception of the build of the instrument.

In proof of our assertion as to the rigidity of the form of substage, as fitted by us to the Edinburgh Student's Microscope, we may say that the sale of this class of instrument has now run into hundreds, and it is in frequent use by many leading microscopists, from whom we have received letters expressing great satisfaction with the rigidity of working parts. Further, a great deal of the demand for the instruments is from the very men that 'Microscopist' considers are best able to 'formulate the desiderata of a student's Microscope'—viz. the teachers in medical schools. From these, throughout the world, we have received orders, not only for ones and twos, but for "ties," and in nearly every instance the instruments have been preferred on account of their rigidity and the convenience and perfection of substage.

If any one required a substage on a fixed tail-piece we should supply it; but never, since we have made the Edinburgh Student's Microscope, have we been asked for such.—W. WATSON & SONS."

"Allow me to supplement my letter (see above) by a few observations, which I trust will forward improvements in the construction of students' Microscopes.

I have suggested that responsible teachers in medical schools should combine and authoritatively formulate the desiderata of a student's Microscope, and that the laws of demand and supply would do the rest.

On the question of the need of such combined action for this purpose, I hardly expect a dissentient voice. But as to the best means of bringing this action to a focus, that is matter on which I must only touch with diffidence. The fact that Mr. G. C. Karop, secretary of the Quekett Microscopical Club, avows his special interest in the subject would point clearly in his direction as a possible centre of action. No great difficulty should be found in forming a committee of men of similar tastes and aims, who would cordially strive to bring out a Microscope to meet the modern wants of students both as to efficiency and economy.

Such a committee would require the co-operation of a skilled mechanician, having considerable manufacturing resources at his command, and who would boldly face the outlay of developing what would be recommended. The prospect of the large business that would naturally follow upon the successful production of a student's Microscope, under such favourable auspices, would, no doubt, be quite sufficient to bring forward the right mechanician.

It may be asked why the requirements of students at this moment do not induce an optician to produce exactly what is wanted? On this it may be said that up to the present time the teachers in medical schools have never attempted any concerted action in the matter; they have been content, individually, to make suggestions here and there to the opticians, and these latter have only felt safe in carrying out the suggestions when they emanated from an influential man whose personal authority sufficed to insure a demand for the instrument large enough to



recoup the experimental outlay. Thus it has happened that each optician has had to secure for himself one or more of these patrons, each of whom has had his own pet schemes to promote, and there has been no sufficient inducement of probable commercial success to warrant the planning of the manufacture of Microscopes on a thoroughly economical basis, by which the most efficient instruments could be produced at a minimum price. The want of concerted action on the part of teachers has thus led to an immense waste of production—a waste amounting in many cases to 50 per cent. or more. It should also be noted that the optician is not, as a rule, catering directly for medical students; he has first to secure the good offices of an influential medical patron, who will instruct his pupils to purchase the apparatus he recommends. Unless, therefore, he is willing to forego his own independence of action, and carry out strictly the orders of his patron, the patronage is transferred to a more willing agent. It is wholly beside the mark to urge that the optician is not bound to follow such orders, that he may use his own discretion on the matter. In practice it may happen that Dr. Microtome, professor of microtomic biology at half a dozen medical schools, whose patronage is good for the sale of some scores of Microscopes per session, knows nothing whatever about the construction of Microscopes, or he may have just the smattering of interest in mechanical design that finds outlet in pressing forward novelties *quâ* novelties, regardless of their practical value; but he is keenly alive to the importance of having his name connected with some form of student's Microscope, and he knows the value of his patronage; the optician is, therefore, obliged to accept the terms proposed to him, and to produce Students' Microtomic Bacteriological Microscopes according to instructions. To add to the confusion of the circumstances, nearly every medical session is accompanied by changes in the medical staff; new ideas crop up or old ones are revived; the optician is appealed to for this or that petty modification in the design of the Microscope, and thus his skill and experience are too often frittered away in carrying out trumpery suggestions which would not stand a moment's discussion before a jury of experts. To meet this incessant order of change one optician in London has made upwards of twenty different forms of students' Microscopes during the past ten years.

My impression is that the present requirements of students do not really necessitate the construction of an entirely new design of Microscope, but only the combination or adaptation of a number of useful points which already exist either together or separately in known models. The student wants a stand that will enable him to get the best work out of his optical means. High-class instruments exist which appear to satisfy the demands of the most fastidious microscopists. It would appear, then, that there is no need for the invention of a new model: we want only the application of common sense to the process of selecting and embodying in the most economical way the points of construction which experience has shown to be the most essential in the high-class Microscopes.

It may be, however, that the majority of teachers in medical schools have no particular claim to be regarded as experts in the use of the Microscope, and hence that a committee of them would carry no very

special weight in their recommendations. A committee of this kind would naturally be guided to a great extent by the more experienced of the members; and these latter would not hesitate to call in the assistance of any one who was known to be specially qualified to advise on the matter—I think that might well be taken for granted.

With reference to the construction of students' Microscopes, where experience informs me that English opticians are very apt to go astray:—If we take a general survey of the various kinds of students' Microscopes produced in England, we shall be struck with the fact that they are too light in build, and consequently too liable to become loose and shaky in their bearings throughout. Take an average English stand of this class as it leaves the maker's hands, put it on a laboratory table to be used by students for a session or two, and it will then show such signs of wear and tear as to need radical renovation. This rapid deterioration of the mechanism of our students' Microscopes is, in great measure, due to the inferior quality of the metal employed, which is not selected for its durability, but mainly for its low price and the ease, and consequent cheapness, with which it can be fashioned. Thus instruments are put together with a considerable amount of accuracy in the bearings, and with rack-and-pinion work moving with all desirable smoothness, and, above all, with a most lustrous polish wherever the lathe can be brought to bear on the parts—the whole so artfully and cleverly finished that we are all deceived into commending the results. We examine critically all the movements and they pass muster with applause. But the question of the durability cannot be settled by mere inspection. We are apt to suppose that well-fitted metal-work must necessarily be durable because it is metal; whereas, durability depends to a very large extent on the quality of the metal. Many of these modern Microscope-stands are made of a quality of brass that is specially chosen of just that degree of hardness to enable the work to be done with the maximum speed, and, consequently with the minimum outlay on the production of the instruments. The question of durability is wholly ignored. The policy seems to have been to make the utmost haste in the business of manufacturing, and let 'the devil take the hindmost.'

Judicious concerted action on the part of class-teachers should put an end to this chaotic state of things, by giving the opticians reasonable assurance that those who can succeed in meeting the desiderata most efficiently will meet with the desired commercial success.

I have been informed, on reliable authority, that some months ago a project was discussed by sundry amateurs in London for a competition among opticians relative to the production of the most efficient Microscope-stands of certain classes, and substantial prizes were to be offered. The matter went so far that the secretary of one of the most prosperous societies in London was ready to inaugurate the competition formally. But wiser counsels prevailed. It was plainly foreseen that the difficulties of settling the conditions of the competition and the selection of the jury would lead to endless bickering and dissatisfaction, and so the matter was dropped.

The scheme we now have at heart is much less ambitious, and, I hope, more practical; and I think it should have the support of Mr. Karop and his fellow-workers.—MICROSCOPIST."

"The question between Messrs. Watson and myself relates to the advisability or not of retaining the pivoting movement of the mechanical substage in their Edinburgh Student's Microscope, as shown in the figure they reproduced with letter 31741, p. 207. They point to the numbers of these Microscopes sold in proof that the system must be good; whilst I, on the other hand, point to the mechanism itself in proof that the system is bad, and I will explain my views.

The chief aim in the application of focusing and centering movements to the substage is to enable the worker to adjust the illumination with all desirable accuracy. Experts have long been striving to popularize the fact that unless a mechanical substage is provided the student is placed at a serious disadvantage, for he cannot otherwise get the best work out of his optical battery.

In the most perfect Microscopes hitherto constructed, no efforts have been spared to make the mechanical substage thoroughly substantial and accurate, and its attachment to the main instrument as rigid as possible; hence the outlay on the substage is a large item of the total cost of these instruments.

In the less perfect Microscopes, which are also less costly, less expensive forms of mechanical substages have to be applied, and as the outlay is reduced, the construction naturally reaches a lower grade of general stability, until finally, in order to cut down the cost to the lowest point, the optician gives up the substantial connection of the fitting with the main instrument, and attaches the substage to the under face of the stage proper by a screw-pivot, as shown in Messrs. Watson's figure. The prime motive for adopting the pivoting system originally—I do not say the prime motive of Messrs. Watson, but of the real originators, the French or German manufacturers of low-class Microscopes for toy-shops, &c.—was to reduce the cost, for the attachment is of so inexpensive a character that I suppose it may be done for a shilling, and then carry a profit of 50 per cent. Such a system may be tolerable in toy Microscopes, where the substage is intended as a mere diaphragm carrier, and it was formerly much in vogue even for dissecting Microscopes, as made by Chevalier and others; but when the substage has to carry centering arrangements on a rack-and-pinion movement on a tail-piece, the whole purpose of these appliances is frustrated by the speedy development of 'wobble' and general instability that disgust the critical worker.

Good substage appliances are most essential adjuncts for all serious work, and they merit that the substage shall be soundly and substantially attached to the Microscope, with reasonable assurance that the condenser, &c., shall focus and centre accurately in the optic axis of the Microscope with the minimum of collimation error. These are the well-known conditions that should guide the optician in the application of an efficient substage to a student's Microscope; and they are not properly met by attaching it to the stage proper by a pivoting arrangement such as that figured by Messrs. Watson.

The student cannot too soon learn that the firmness of his mechanical substage, its rigid attachment to the Microscope, either by a tail-piece or other fitting, is infinitely more important in practice than the liberty of swinging it aside. He may take it for granted that the construction



of his low-priced substage will be quite unsteady enough in itself without the addition of a pivoting motion.

As to the pivoting system being recommended by Messrs. Watson's medical patron at "Modern Athens," that fact suggests to me only the probability that the recommendation is based on very limited experience of high-class work; whilst their own approval of it, according to the tone of their letter, seems largely due to commercial reasons which are not in my province to discuss.

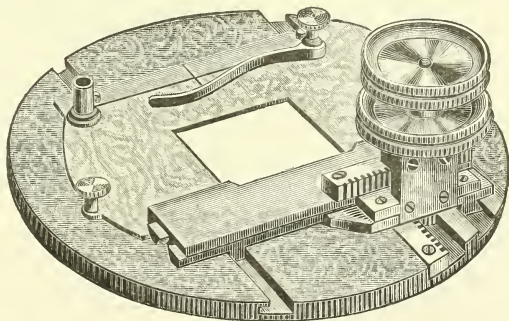
I condemn the system on its merits as totally unfit to be combined with a mechanical substage. I see in its application only the evasion of the recognized good and durable methods of attaching the substage to the Microscope, which are adopted wherever such matters have been seriously aimed at.

We have had, in all conscience, enough, with over-measure, of inferior constructions of students' Microscopes. Let us now aim at something altogether better.—MICROSCOPIST."

**An Illuminating Cell.\***—Mr. F. O. Jacobs describes a cell made of any white metal that will take a bright polish, and shaped so as to reflect the light it receives from the mirror to the object, which is mounted on an opaque ground. The cell completely covers the object but is perforated by an opening of the same size as the ground on which the object is mounted, which should be just large enough to inclose the field. The arrangement is completed with a small cover-glass. The advantage of this cell is, that it saves time in adjusting the light and condensing lens, the same lighting being used as with a transparent object.

**Bulloch's Mechanical Stage with Vertical Pinions.**—In the various constructions of this form of stage since its comparatively recent intro-

FIG. 90.



duction by the late R. B. Tolles, the mechanism has generally been defective in the matter of durability; in the endeavours to secure extreme thinness of the support for the slide, the attachment of the

\* The Microscope, x. (1890) p. 281.



mechanism to the solid part of the stage has been so reduced in strength that with very little use it has become so loose and shaky as to be unserviceable.

In the new stage here shown (fig. 90) it has been the aim of Mr. W. H. Bulloch to correct the defects above noted by the application of dovetail slides to the forward and backward motions, which also provide a firmer support for the lateral motions.

#### (4) Photomicrography.

**Photomicrography at Medical Congress of Berlin, 1890.\***—In his review of the photomicrographic apparatus and photographs exhibited at Berlin during the sitting of the International Medical Congress, Dr. R. Neuhauss does not seem to have found much that was worth more than a general description. The well-known makers Zeiss, Leitz, Klönne and Müller, and Hartnack were, of course, in evidence with their well-known apparatus. A. Stegemann exhibited a base-board by which the camera could be moved from the horizontal to the vertical position; P. Thate, a magnesium flash-light apparatus; and Carl Günther, a cabinet for drying plates, capable of holding 24 plates  $13 \times 21$  cm. in size. A strong current of air, kept up by means of a lamp fitted to the lid, dries the plates in 3 to 6 hours. Among the photographs may be mentioned those of Prof. Loeffler, showing the flagella of bacteria; the instantaneous photographs of living infusoria obtained with the flash-light of Duncker. The difficulty experienced with the ultra-violet rays was avoided by filtering through a Chinin filter. Prof. Babes, of Bucharest, is severely handled by the writer, who attributes to the photographs almost every possible fault. It would seem, however, that taken all round, photomicrography is greatly advancing.

**Marktanner-Turneretscher's 'Photomicrography.'**† — This recent manual on photomicrography is specially addressed to those who are desirous of showing the results of their own work by means of photography, and of attaining this object with the least expenditure of time and trouble. After reviewing and discussing the various apparatus necessary for photomicrography the author proceeds to the chief methods and explains the dry-plate, the wet-plate and the positive processes, and then indicates certain defects which frequently occur, and the means for their avoidance. The work ends with describing how to show photographic preparations by the aid of the magic lantern. The book is copiously illustrated, and contains an excellent list of works of reference.

**Absorption-plates.‡**—In order to absorb ultra-violet rays A. Miethe employs gelatin plates made as follows:—Gelatin 2 grm., glycerin 2 grm., water 25 ccm., æsculin 0·059. The gelatin is dissolved in 15 ccm. water, then are added the glycerin and the æsculin, dissolved in 10 ccm. water; the whole is then filtered through flannel. Glass plates are covered with a pretty thick layer of this mixture, which is then allowed to set and dry in a dust-free place. In order to completely absorb the ultra-

\* Zeitschr. f. Wiss. Mikr., vii, (1890) pp. 145-50.

† 'Die Mikrophotographie,' Halle, 1890, 344 pp., 195 engravings, and 2 pls.

‡ Photogr. Wochenbl., 1890, No. 18. Cf. Zeitschr. f. Wiss. Mikr., vii, (1890) p. 187.

violet rays, the æsculin plate is combined with another which contains 0·02 grm. of fluorescein instead of the æsculin. These two plates are stuck together and their edges united with black paper. As the æsculin becomes embrowned with time it must be replaced by a fresh plate.

**Application of Photography to the demonstration of certain Physiological Processes in Plants.\***—Mr. W. Gardiner states that it is possible, by taking advantage of their sensitiveness to light, to obtain prints from *Protococci*, or the free-swimming swarm-spores of many green algæ. Into one end of a water-tight box a thin glass plate is securely fixed. The negative to be printed is then placed next the glass, film side nearest. The box is filled with water containing a fairly large quantity of swarm-spores, and the lid is shut down and the whole exposed to diffused light. In the case of a strong well-developed negative, the swarm-spores swim towards the most highly illuminated parts, so that, after some four or six hours, on pouring out the water and removing the negative, a print in green swarm-spores can be obtained.

(6) *Miscellaneous.*

**Deceased Honorary Fellows.** Mr. Ralfs and Prof. Parker, F.R.S. —Our Fellows will be glad to have memorial notices of the two eminent honorary Fellows whom we have recently lost. That of Mr. John Ralfs is extracted from the notice by Messrs. H. & J. Groves in the 'Journal of Botany,' xxviii. (1890) p. 289.

"Only those who have come into close contact with the man or have carefully studied his works, can realize the greatness of the intellect of the veteran botanist who died at Penzance in July last. Had not his health and eyesight failed, there is little doubt that John Ralfs would have ranked as one of the greatest botanists of the century. His clearness of perception, his conciseness and exactitude of expression, added to his indomitable energy, his enthusiasm, and his wonderful memory, made him the very ideal of a naturalist.

He was born on Sept. 13th, 1807, at Millbrook, near Southampton. He came of an old Hampshire family, being the second son of Samuel Ralfs, of Mudeford, near Christchurch. His father died in 1808, and the young family was brought up by the mother, who disposed of the property at Mudeford, and removed to Southampton. Young Ralfs's first school appears to have been that of Dr. Buller in this town; he afterwards went to Mr. Jennings's at Bishop's Waltham, and subsequently to the Rev. J. Jenvey's at Romsey. To the last-named gentleman he became much attached, and to him he dedicated his first botanical book. As a lad Ralfs was studious and painstaking, and showed an early inclination to scientific pursuits which first developed in the direction of chemistry. At about the age of eighteen he was articled to his uncle, a surgeon at Brentford, with whom he remained two years and a half, after which he studied at Winchester Hospital for two years. In 1832 he passed the examination qualifying him as a surgeon, and in this examination we find that he distinguished himself by his knowledge of botany. He went into partnership with a surgeon in Shoreditch, and

\* Ann. of Bot., iv. (1889) p. 163.

Mr. Marquand tells us that he practised at Towcester. During the few years that he was able to follow his profession he was very successful. While on a visit to Torquay he became acquainted with Miss Laura Cecilia Newman, daughter of Mr. Henry Newman, of London, and in 1835 was married to that lady. They had one son, John Henry, who was born in 1836. The marriage did not prove a happy one, for within two years Mrs. Ralfs (with her infant son) went to live with her parents, who were then residing in France; she afterwards travelled in Italy, but returned to France, where she died in 1848.

In 1837 Mr. Ralfs's health became so bad, his lungs being found to be seriously affected, that he was obliged to relinquish his practice and to reside in one of the health-resorts on the south-western coast. After visiting Torquay, he settled down, in November 1837, at Penzance, which continued to be his home during the rest of his life. In 1838 he contributed the botanical portion of a guide to Ilfracombe by Banfield. In 1839 he published his first book, 'The British Phænogamous Plants and Ferns; arranged on the Linnaean System, and analysed after the method of Lamarck'; this consisted of a dichotomous key to the genera and species, with an analysis of the natural orders. It did not pretend to compete with the larger "Floras," but was intended as a guide to the quick determination of species; and the simple straightforward language employed, the judicious selection of practical characters, and the small compass of the book admirably adapted it to the purposes of a pocket manual. At the commencement of 1841, Mr. Ralfs opened a correspondence with the Rev. M. J. Berkeley, whom he had met some years previously; this resulted in a close friendship, and Ralfs and Berkeley appear to have constantly consulted one another on questions connected with the Algæ and Fungi. Berkeley's correspondence (preserved in the Botanical Department of the British Museum) contains some hundreds of letters from Ralfs, many of them consisting of four closely written quarto pages, and containing pen-and-ink drawings. Ralfs seemed then to have settled down to the study of the Desmids and Diatoms, but continued to give a general attention to Fungi and other plants.

The summers of 1841 and several subsequent years were spent in visits to Ilfracombe and various parts of Wales, his longest stay usually being at Dolgelly. In 1842 he was accompanied on his Welsh trip by Borrer. In this year Ralfs sent a description of *Desmidium compressum* (a new species) to Dr. Balfour for the Botanical Society of Edinburgh. In 1843-4-5 he contributed to the same Society a series of papers on the Desmids and Diatoms, and in one of them he mentioned that the total number of Desmids previously recorded in the British Floras was four—two *Desmids* and two *Euastra*. These papers were published in the 'Annals of Natural History' and in the 'Transactions' of the Society. They contain figures and descriptions of a number of species of Diatoms, and over sixty Desmids, of which sixteen were new. In 1845 also appeared his paper, 'On the genera *Spirulina* and *Coleochaete*,' A. N. H., xvi. p. 308. . . .

In 1848, after several delays occasioned by illness, his great work was published, 'The British Desmidiæ,' probably the finest monograph

which has appeared of any group of British plants. The descriptions are complete and lucid, the synonymy is very carefully worked out, and the analyses are in Ralfs's characteristically terse style. Particular attention is given to the reproductive states of the plants, which had been previously observed in very few species. An appendix contains descriptions of the species not known to occur in Britain, and the small number of these is an evidence of the leading position Ralfs had taken up as an authority upon the group. In a few years he had raised the number of known British Desmids from four to 180. Mr. E. Jenner's beautiful drawings contributed much to the value of the work, for he was not only an excellent draughtsman, but a good botanist, and well acquainted with the Desmids. During the preparation of the works Ralfs had extensive correspondence with Brébisson, Kützing, Montagne, and other leading foreign algologists. Berkeley seems to have been of great assistance in many ways. . . .

In 1856 he undertook the arrangement of the Diatoms and Desmids for the fourth edition of Pritchard's 'Infusoria,' but, through repeated illnesses, was only able to complete the Diatomaceæ, and this contributed to the delay in the publication of the book, which did not appear until 1861. His work, however, was very thorough, and gave an account of the whole of the known Diatomaceæ, both recent and fossil.

The sudden failure of his eyesight about this time rendered future microscopical research impossible, thus putting a stop to the great work of his life, and he does not seem to have recovered from the shock for many years. He turned his attention more and more to working out the flora of West Cornwall. . . .

Mr. Ralfs bequeathed his collection of microscopic plants to the Botanical Department of the British Museum, but his will was not witnessed, and had consequently no legal force. His son has, however, in consideration of his father's wishes, generously resolved to place the collection in the British Museum."

That of Prof. Parker is an anonymous notice, clearly from the hand of one who knew him well, which appeared in the 'Times' of 7th July last.

"By the sudden death, on the 3rd inst., of Mr. William Kitchen Parker, F.R.S., formerly Hunterian Professor of Comparative Anatomy at the Royal College of Surgeons, science in this country has lost one of its unique investigators, a man of the order of Faraday, if lacking his great constructive powers. The son of a farmer in South Lincolnshire, his schooling was of a very limited character except for three-quarters of a year spent at Peterborough grammar school, after which he became an assistant to a chemist at Stamford. He had already been attracted to the mysteries of anatomy as they chanced to come under his notice in farming life, and with no instruction whatever he had made skeletons of many animals. While at the chemist's, engaged at business from 7 a.m. to 10 p.m., he rose several hours before his morning's work began, and with a fellow-apprentice scoured the neighbourhood for botanical specimens. Thus in two summers he formed and preserved a collection of 500 species of plants.

After a few years he came up to London as a surgeon's assistant



and, still continuing to make progress in anatomy, he became assistant to Professor Todd at King's College, and qualified for medical practice in 1849. He made during these years many beautiful injected preparations of organs, and also laid the foundation for his later microscopical work on the Foraminifera. Indeed, it was as a student of the latter minute organisms that he first came before the scientific public in 1857, when he began to publish, in conjunction with his friend Professor Rupert Jones, a long series of important papers in the 'Annals and Magazine of Natural History,' in which many significant facts as to their variability and polymorphism in parallel series were first brought forward.

A few years later, ill-health, the result of much unremunerative scientific work combined with a laborious medical practice, began to make serious inroads on Mr. Parker's physical strength, and he had to give up much of his professional work. In the intervals of the severest pain he accomplished some of his most striking researches, which were often taken up as an anodyne. Many valuable monographs, such as those on the skulls of the common fowl (1869), of the frog (1871), of the salmon (1873), of the pig (1874), were the result of his labours, and when in 1874 he was appointed one of the Hunterian Professors of Comparative Anatomy it was felt that the mantle of Professor Huxley had fallen on a worthy successor.

Professor Parker lectured at the College of Surgeons until 1884, giving in his own quaint and discursive way the results of successive years of work. He had already been President of the Royal Microscopical Society in 1871-2, and this honour was followed by the award of a Royal medal by the Royal Society, of which he was already a Fellow. When the Government grant of 4000*l.* came to be distributed by the Royal Society it was generally felt that there could be no more fitting recipient of a considerable grant than Mr. Parker, and this was continued for many years, being at last partially replaced by a Civil List pension. No man ever worked from a purer love of science and of the beauty of the Creator's handiwork, which he delighted to acknowledge. He had grown up when minute naked-eye dissection had not been displaced by microscopic section-cutting, and it was a marvel to see him handle embryonic skulls a third of an inch in length and patiently dissect them under a simple lens till he had revealed features characteristic of some very diverse creature in the scale of development, or of some ancient animal which combined in itself characters now split up among various extreme branches of the vertebrate kingdom.

Altogether he wrote more than twenty memoirs of first-class importance, illustrated by many hundred plates from his own careful drawings, and published by the Royal, Zoological, and Linnean Societies. Unfortunately they are a sealed book to all but skilled anatomists, for notwithstanding brilliant flashes and quaint conceits and illustrations, Mr. Parker's style of exposition by no means did justice to the value of his matter. One portion of his work was, however, summarized and brought out by him in 1877 with the aid of his friend, Mr. G. T. Bettany, under the title 'The Morphology of the Skull,' and another portion formed the subject of a volume, issued in 1885, on 'Mammalian Descent,' being the Hunterian lectures for 1884. But his friends will remember,

even more than his scientific labours, the charms of his ingenuous enthusiasm and the warmth of his scientific ardour. For many years a great sufferer, he had been much shaken by the recent death of his wife, but his health did not appear worse than usual when he suddenly expired at the house of his second son, Professor W. Newton Parker, of the South Wales University College, Cardiff, at the age of 67."

### B. Technique.\*

#### (1) Collecting Objects, including Culture Processes.

**A Homely Zoophyte-trough.**†—Dr. J. Anderson Smith remarks, "The constant trouble I had with the usual glass zoophyte-troughs, either from leakage, too great depth, or too large size, led me to try something else. And first I tried cork rings of various diameters and depths, but the difficulty of cutting evenly, and the occasional perforation in the cork allowing air-bubbles to get into the cell, soon caused me to abandon these. I now use indiarubber rings, which give me perfect satisfaction. I take an ordinary glass slide, find the centre, and then fix on to it by means of Canada balsam, an indiarubber ring,  $\frac{5}{8}$  in. diameter,  $\frac{1}{8}$  in. deep, and  $\frac{1}{8}$  in. thick. Rings of any required size or depth may be used. Filling the inclosed space with the water and weed to be examined until the surface of the water is slightly convex above the plane of the upper surface of the rings, I then place a cover-glass of the requisite size on the top, and the trough is ready for examination. Capillary cohesion holds the cover-slip perfectly tight, so that the trough may be turned upside down without spilling the contents.

The advantages I claim for this little trough are:—First, its cheapness; second, the facility and rapidity with which it can be made. Moreover, by choosing various sized rings, troughs of any depth and size can be made, and such a trough may be readily used at the pond-side for rapid examination of small portions of the material collected. Lastly, it is less cumbrous than the glass trough and more useful in my experience. The rings I have chiefly used are such as one gets from certain mineral water bottles; the dimensions given are those of a ring labelled "Matlock Mineral Water Co."

**A New Collecting Net.**‡—Mr. Charles S. Fellows has recently devised a collecting net for small organisms, consisting of a silk mull (or bolting cloth) funnel whose largest diameter is about 12 in., kept open by a stiff brass ring. It is 15 in. deep and tapers off to  $\frac{3}{4}$  in. at the smallest end. In this (the apex of the cone) is fixed a brass ferrule about  $\frac{3}{4}$  in. in diameter and 2 in. long, made with a shoulder on each end, one to prevent it from slipping off the net and the other to prevent a *silk bottle* from becoming detached.

This silk bottle can be made of any size, but is most convenient for use about  $\frac{3}{4}$  in. in diameter by 2 in. in height, and made of the

\* This subdivision contains (1) Collecting Objects, including Culture Processes; (2) Preparing Objects; (3) Cutting, including Imbedding and Microtomes; (4) Staining and Injecting; (5) Mounting, including slides, preservative fluids, &c.; (6) Miscellaneous. † Journ. of Microscopy and Nat. Sci., iii. (1890) pp. 251-5.

‡ The Microscope, x. (1890) pp. 247-8.

FIG. 91.



FIG. 92.

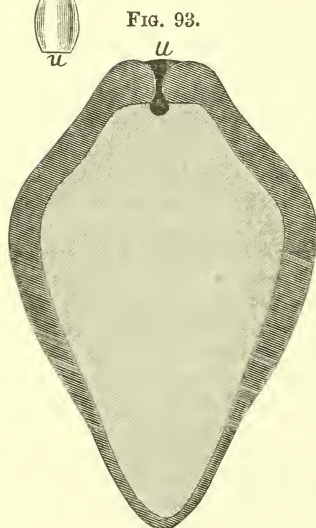


FIG. 93.

same kind of silk as the net. To use the apparatus, first tie the little silk bottle on the ferrule, then drag the net after a boat, or through the water from the shore, by tying it to the end of a fishing rod or walking-stick. As soon as the "catch" is in the little silk bottle it is untied from the ferrule, using the same string to tie up its mouth, and put into the alcohol bottle. Each silk bottle is numbered so that a record of time and locality can be kept. This is much superior to the use of a glass bottle, for in pouring out enough of the contents of the glass bottle in order to insert the cork, one often loses very interesting forms, especially the *Cladocera*, on account of the bubble of air which usually is in the shells and which causes them to float to the top.

A useful net is simply made from fine silk mull, 15 in. in diameter and 30 in. long."

**Suction Capsule.\***—The suction capsule, the invention of Mr. N. A. Cobb, is made from glass tubing, 2–5 mm. thick. The tubing is heated in a blow-pipe flame in two places and drawn out (fig. 91). One end is then broken off and heated until the aperture becomes minute. The other end is drawn out to the form shown at *t* (fig. 92). If after stopping *u*, fig. 92, with

glue suction be made at *r*, while the tube at *t* is melted with a blow-pipe flame, the suction capsule results (fig. 93). The use of this little instrument is to hatch eggs of parasitic entozoa in the alimentary canal. The eggs are placed within the capsule, and when swallowed, the gastric juice dissolves the glue and hatches the eggs. The capsule is easily recovered at the other end of the alimentary canal.

BÖHM, A., UND A. OPPEL.—  
Taschenbuch der mikroskopischen Technik. (Manual of microscopical technique.)  
München und Leipzig, 1890.

\* Proc. Soc. Linn. N.S.W., v. (1890) pp. 163–7 (3 figs.).

## (2) Preparing Objects.

**Demonstrating the Cell-Granula.\***—Herr R. Altmann recommends as fixative for tissues to be examined for the cell-granula a mixture of equal volumes of a 5 per cent. solution of bichromate of potash and 2 per cent. solution of osmic acid. After twenty-four hours the pieces, which are of course very small, are washed in running water for several hours; then alcohol (75, 90, 100 per cent.); paraffin imbedding, for which they are placed, after the spirit, in a mixture of three parts xylol and one part alcohol; then xylol, xylol-paraffin, lastly paraffin, with a melting-point of 58°–60°. The sections made with the "Support-Mikrotom" are from one to two  $\mu$  thick, and are stuck on the slide with a thin layer of caoutchouc (caoutchouc dissolved in 25 vols. chloroform). This solution is poured on the slide, drained, and after evaporation of chloroform heated gently, the paraffin sections are then stuck on and brushed over with a mixture of gun-cotton in acetone and alcohol (2 grm. gun-cotton dissolved in 50 ccm. acetone; of this 5 ccm. are diluted with 20 ccm. alcohol).

Acid fuchsin is recommended for staining and picric acid for differentiating the granula. The former solution is made by dissolving 20 grm. acid fuchsin in 100 ccm. of a cold saturated aqueous solution of anilin; the latter is a mixture of 1 vol. saturated alcoholic solution of picric acid and 2 vols. water. The sections are stained by pouring the fuchsin solution on the slide and carefully heating; this done, it is washed and treated in a similar way with the picric acid solution; after which alcohol, xylol, and dammar.

Another fixation method, by which the staining is rendered more brilliant, though the sections are thicker and the preparations less permanent, is as follows:—A saturated solution of red oxide of mercury is made in 30 per cent. nitric acid. Immediately before use one vol. of the foregoing is mixed with three vols. water and one vol. 50 per cent. formic acid. In this solution the fresh pieces are placed for several hours, after which they are transferred to alcohol, and thereupon the paraffin procedure.

A quite novel method, but which for mechanical reasons is as yet difficult and imperfect, is introduced by the author. It consists in freezing fresh pieces of organs and then drying them in vacuo over sulphuric acid at a temperature of less than 20° C. By this means no alteration in volume occurs in the pieces, which differ from the recent condition merely in the absence of water. The next step is to saturate the pieces, still in vacuo, with paraffin.

**Demonstrating the Elastic Fibres in the Skin.†**—Sig. V. Mibelli stains the sections in a solution made as follows:—(1) safranin 0.59, warm H<sub>2</sub>O (80°) 50; (2) safranin 0.59, alcohol (90°) 50. When cold these two solutions are mixed.

After having been immersed in the solution for thirty-six to forty-

\* 'Die Elementarorganismen und ihre Beziehungen zu den Zellen,' Leipzig, 1890, 145 pp., 2 figs., and 21 pls. Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 199–203.

† *Monitore Zool. Ital.*, i. pp. 17–22. Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 225–6.



eight hours they are transferred to hydrochloric and spirit (absolute alcohol 100 grm., HCl 10 drops). The acidulated spirit is constantly renewed, until the dye is no longer given off, when the sections are allowed to remain five to ten minutes longer, after which they are removed to absolute alcohol for twenty-four hours. Then bergamot oil and xylol dammar.

By the foregoing method the author claims that elastic fibres are well stained, but only if the solution be made in the manner prescribed.

**Demonstrating the finer structural relation of the Liver.\***—Herr A. Oppel applies Golgi's method to the liver as follows:—A small piece of rabbit's liver is treated with bichromate of potash quickly increased from two to five per cent. In three weeks' time the piece is placed in  $\frac{3}{4}$  per cent. nitrate of silver solution. In a few days the ultimate bile-ducts are stained.

The author gives the result of his procedure with monochromate of potash (0.5 per cent.) and silver nitrate on objects kept in spirit for a long time. Three per cent. solution of bichromate and 0.5 per cent. chromic acid were, however, found to show the biliary network quite as well.

**Killing and hardening Pelagic Animals.†**—Herr B. Friedlaender finds, from the experience of a few months, that the most efficacious fluid for killing sea animals (Siphonophora, &c.) is a mixture of water 1000, zinc sulphate 125, copper sulphate 125. The solution is placed in one vessel, and the animals in sea water in another of similar size. The contents of the former are simply poured into the latter vessel.

For hardening, a 1 per cent. solution of osmic acid in sea water is recommended, while for delicate objects osmic acid may be added or even used alone as a  $\frac{1}{5}$  per cent. solution.

**Preserving lower Organisms in Microscopical Preparations.‡**—Pure blood-serum is recommended by Dr. W. Migula as a suitable medium for examining and preserving delicate animal and vegetable objects. He uses the commercial blood-serum, and filters in an ice-box through bibulous paper frequently changed. The filtrate is mixed with 10 per cent. pure glycerin and incubated at 45° to 50° C. When all the water has been evaporated, the glycerized jelly is preserved in stoppered vessels. When required for use, a small quantity is dissolved in 10 to 15 times its volume of distilled water, and a large drop placed on the slide. Into this drop the living organism is pipetted and then the slide is placed in an incubator at about 50°, in order to thicken down the fluid. When of the right consistence, the cover-glass, moistened with a mixture consisting of 40 parts glycerin, 20 parts absolute alcohol, and 40 parts water, is imposed. The preparation is again heated for a couple of hours, and then ringed round.

**Preparing Blood of Arthropoda and Mollusca.§**—According to Sig. G. Cattaneo the methods usually adopted for examining the blood of

\* Anat. Anzeig., v. (1890) pp. 143-5. Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 222-3. † Biol. Centralbl., x. (1890) pp. 483-91.

‡ Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 172-4.

§ Bollet. Scient. di Pavia, xi. (1889) pp. 3-29, 33-57 (2 pls.). Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 213-5.

mussels are imperfect, either because they are too slow, or they allow the blood to mix with water. He advises that the mussel should be well cleaned and dried, and without being opened its heart should be pierced with a needle. Some blood is then collected and examined at once under the Microscope, because after one minute obvious pathological changes take place in the corpuscle.

Control preparations are made of blood preserved with osmic acid or palladium chloride (1 per cent.) and obtained either by the direct action of the reagents, or by injecting the animal in the organ of Bojanus with  $1/2$  to 1 cm., using a Pravaz syringe. Other reagents mentioned do not appear to have given satisfactory results.

When examining the blood of Arthropoda, rapidity is still more urgent, since degeneration begins to set in in 10 seconds. From *Palæmon* blood is best obtained by perforating the posterior third of the body, in spiders by an incision in the thorax, from Libellulidæ larvæ by cutting off the head, and from insects by tearing off a wing. One device mentioned is to drop the blood or dab the organ exuding blood in a small drop of the fixative fluid previously placed ready on the cover-glass.

Blood of Arthropoda should be treated in a manner similar to that used for Mollusca, i. e. with one per cent. osmic acid or palladium chloride. If 3 per cent. acetic acid be used at once, it will fix the amœboid forms, but if the addition be delayed until the first or second stage of degeneration, these appearances are lost.

**Preparation of Sections of Ammocetes.\***—Dr. W. H. Gaskell mainly relied on serial sections, the whole head having been imbedded in paraffin, or the brain was dissected out and then imbedded. 1 per cent. osmic acid, Perenyi's fluid with alcohol afterwards, with subsequent staining in boro-picro-carminate or picro-carmin and eosin were used; for staining on the slide anilin colours and hæmatoxylin were used. The sections were mounted in order; when they were so large that they were apt to be crumpled, the folding was got over by simply floating the series of sections on the surface of warm water and then transferring them to a slide previously coated over with albumen and glycerin.

**Arrangement of Pigment in Eye of Arthropods.†**—Mdlle. M. Stefanowska, in studying the arrangement of pigment in the eyes of Arthropods exposed to varying quantities of light, decapitated the animals, and at once divided the head longitudinally. The pieces were placed in a 1 per cent. solution of osmic acid. The time required to fix the histological elements varies, and can only be determined by trial; on the whole, however, the time varies between one and four hours. The eyes were next placed in a 25 per cent. solution of oxalic acid with alcohol; they were then washed in 70 per cent. alcohol and put into absolute.

After inclusion in paraffin, sections were made with Schanze's microtome; these demanded much time and patience, as the sections break easily. The richness in pigment forms one of the great difficulties in preparing sections, which must, therefore, be very fine; those made were generally  $1/100$  mm. in thickness, and it was only with some of

\* Quart. Journ. Mier. Sci., xxxi. (1890) p. 382.

† Rec. Zool. Suisse, v. (1890) pp. 155-9.

the Muscidae that a thinness of 1/200 mm. was obtained. Staining with hæmatoxylin was found useful in bringing out the contours of some of the cells.

**Preparing Intestinal Canal of Ephemeridæ.\***—Herr Ad. Fritze recommends that Ephemeridæ be fixed in absolute alcohol, imbedded in paraffin, and the sections stained with hæmatoxylin and borax-carmin. The intestine of *Bætis* larvæ should be prepared in physiological salt solution, gradually hardened in alcohol, and stained with borax-carmin.

**Examining Cypridæ.†**—For examining in physiological salt solution, Herr C. G. Schwarz fixed the animals in 30 per cent. spirit, heated to 70°, and afterwards hardened them in alcohol, increased from 70 to 100 per cent. They were decalcified in concentrated picric acid in six hours at 54°, and then washed in boiled water and imbedded in paraffin. Hot sublimate, Flemming's mixture, and also a mixture of 2 per cent. osmic acid 1 part, 2 per cent. acetic acid 5 parts, distilled water 4 parts, gave good results. The sections, which were stuck on with glycerin-albumen, were stained with picro-carmin and hæmatoxylin, hæmatoxylin and eosin, borax-carmin, and acetic acid carmin.

Isolation of the chitinous framework and of the muscular fibrillæ of the organs known as the seminal pump was effected by maceration in Moleschott's potash-solution.

**Preparing Lumbricus terrestris.‡**—After killing the animal by gradually adding spirit or hot water, Herr G. Goehlich examines freshly prepared organs in 0·5 to 1 per cent. salt solution. If the seminal sacs be left for one day in spirit their contents coagulate, and can be removed as firm lumps. In order to make sections, the intestinal canal is cleaned of earth and sand by the animal being starved for two or three days; it is placed in a covered glass vessel containing water and its excrement carefully removed. Fixation in cold sublimate solution or in absolute alcohol. Staining with alcoholic carmin. Paraffin imbedding.

**Preparing Cestoda.§**—Very good preparations of Cestoda, says Dr. F. Zschokke, can be obtained by staining them for six to twelve hours in extremely dilute Kleinenberg's hæmatoxylin (then washing in water to which a drop of alum solution or acetic acid has been added), as well as by the use of alum or borax-carmin. Then dehydration; oil of cloves and balsam. For sections, after fixation in corrosive sublimate, Mayer's carmin is to be preferred. Paraffin imbedding.

The marine Cestoda can be kept alive for twelve to twenty-fours in a mixture of sea-water and the intestinal mucus of their host.

**Investigation of Development of Fresh-water Sponge.||**—In his study of the development of the fresh-water Sponge, Dr. O. Maas put large cover-glasses to float on the surface of the water of the aquarium

\* Ber. d. Naturf. Gesellsch. zu Freiburg, iv. (1889) pp. 59-82 (2 pls.). Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) p. 212.

† Ber. d. Naturf. Gesellsch. zu Freiburg, iii. (1888) pp. 133-58 (2 pls.).

‡ Zool. Beitr. (Schneider), ii. (1888) pp. 133-67 (2 pls.). Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 209-10.

§ Mém. de l'Inst. Nat. Genevois, xvii. (1888) p. 396 (8 pls.). Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) p. 209.

|| Zeitschr. f. Wiss. Zool., i. (1890) pp. 530-1.



in which the Sponge was developing. Numerous larvæ attached themselves to these, which could be easily taken out of the water and examined. With the aid of silver nitrate they formed excellent permanent preparations, which may be set up between two cover-glasses in Canada balsam. The sponges were hardened by absolute alcohol; for the larvæ the best preparation was found to be chrom-osmic-acetic acid. Borax-carminé and hæmatoxylin were the staining reagents. The anilin colours, Lyons blue and malachite green, were the best for staining sections in which it was desired to differentiate the yolk. Imbedding was effected with paraffin; when the larvæ had attached themselves to *Elodea*-leaves this was quite easy, but when free they must first be fixed to a bit of liver by albumen, on account of their very small size—scarcely larger than a large Infusorian. Very high magnification is necessary to make out the component cells.

**Preparing Fungus-spores.\***—Herr P. Hennings recommends a modification of Herpell's plan † for fixing and preserving the spores of fungi. The discoloration of white spores which frequently takes place when this method is employed, can be prevented by saturating with alcohol. Coloured spores can be best preserved by making the paper absorb from below a solution of colophone in alcohol.

**Study of Saprolegniaceæ.‡**—Prof. M. Hartog recommends the following processes for fixing and staining this family of fungi. The reagent used for fixing is a saturated solution of corrosive sublimate; the preparation is then washed with water and placed in absolute alcohol. The best staining reagent is a solution of the Naples boracic carminé, and the excess of colour is removed by an alcoholic solution of crystallized acetic acid. The staining succeeds best after the objects have been slightly acted on by a very slightly acidulated alcoholic solution of nigrosine, and is completed by a second more complete staining with nigrosine. The preparation may be mounted either in a solution of equal parts of sulphophenate of zinc and glycerin; in Canada balsam, after placing in absolute alcohol, to which is added, drop by drop, phenicated xylol in the proportion of 3 parts of xylol to one of phenic acid; or in essence of sandal-wood oil.

**Preparation of the Lower Algæ.§**—For cultures of the lower Algæ, *Chlamydococcus*, *Eudorina*, *Gonium*, &c., M. P. A. Dangeard uses Van Tieghem's moist chamber, consisting of a ring of glass fixed to the slide, and covered by a cover-glass, on the lower face of which is a drop of water containing the objects to be cultivated. The chambers are kept in a constantly moist atmosphere. The fixation may be effected by concentrated picric acid, 1 per cent. chromic acid, absolute alcohol, or 1 per cent. osmic acid. For studying the vibratile cilia or flagels, chromo-osmic acid is best employed, which admits of an immediate observation, or the object may be fixed on the slide by concentrated osmic acid; it is then covered by a cover-glass and stained by a trace of methyl-green or by hæmatoxylin. To study the internal structure it

\* Verhandl. Bot. Vereins Brandenburg, xxx. (1889) pp. 136-7.

† Cf. this Journal, 1882, p. 122.

‡ Bull. Soc. Bot. France, xxxvi. (1889), Actes du Congrès de Bot., pp. ccviii.-ccix.

§ Notarisia, v. (1890) pp. 1001-6 (16 figs.).



is often advisable to fix with absolute alcohol for 24 hours, and stain with picro-carmin or aqueous hæmatoxylin. For spores or cells, the walls of which are not penetrated by these reagents, Tschirch's borated carmine may be used. When coloured to the right extent, the preparations should be studied in glycerin, Canada balsam, or essence of clove, after dehydrating by alcohol of about 80 per cent.

**Preparing Sections with Elder-pith.\***—Herr J. W. C. Goethart recommends the following method, where very thin sections are not required. A vertical slit is made in a cylindrical piece of pith, the pith being left much longer on one side of the slit than on the other, and the longer side is thoroughly soaked with alcohol. The object of which sections are to be made is then placed in the slit, and a thin platinum wire firmly bound round the whole. The whole is now moistened with alcohol, and placed in the microtome. The sections are then placed and examined in glycerin.

**Mounting Algæ and Fungi.†**—From practical experience, Mr. J. E. Humphrey strongly recommends, in the preparation of slides of Algæ and Fungi, the discarding of all fluids and cements, and the use of glycerin-jelly as recommended by Dr. L. Klein,‡ which he finds applicable to all classes of Thallophytes, after hardening with osmic acid. Even the colour of the pigments is, in most cases, perfectly preserved by this process.

**The Preparation of Vegetable Tissues for Sectioning on the Microtome.§**—Mr. A. J. M'Clatchie says, "Vegetable tissues vary so much as to the amount of protoplasm, cellulose, and other substances contained, that the methods used for obtaining good sections from them must vary greatly. I have prepared and sectioned fungi, lichens, the cotyledons, plumules, hypocotyledonary stems, roots, root-tips of the cucumber, young pine-cones, young wheat-blades, lilac-buds, and bean-stems, with varying degrees of success.

Lichens and the young firm cotyledons of the cucumber could be dehydrated, and permeated with paraffin much more rapidly than young meristemic tissue, or tissue composed largely of cellulose and water. The former may be placed in 50 per cent., 75 per cent., 90 per cent., and 100 per cent. alcohol, chloroform, chloroform and paraffin, and finally in paraffin at a temperature of 55° C., remaining in each from two to twelve hours, and good results will be obtained. But the meristemic and the thin-walled watery tissue must be treated differently, or the tissue will come through very much shrunken and distorted, worthless biologically.

I have had the most success following the method described by Dr. J. W. Moll, in the 'Botanical Gazette' for January 1888. I have obtained good sections from all the material that I have treated in this way. I used a 1 per cent. solution of chromic acid and 20 per cent., 35 per cent., 50 per cent., 75 per cent., and 90 per cent. alcohols for

\* Bot. Ztg., xlviii. (1890) p. 354 (1 fig.).

† Bot. Gazette (Crawfordsville), xv. (1890) pp. 168-71.

‡ Cf. this Journal, 1889, p. 140.

§ Amer. Mon. Micr. Journ., xi. (1890) pp. 190-1. From Amer. Naturalist, July 1890.

dehydrating. The chromic acid seems to fix the protoplasm, and macerate the cellulose, allowing the alcohols to pass more freely. I allowed the specimens to remain in the several per cents. of alcohol from two to twenty-four hours, according to their size and texture. As a rule, I found that the more gradually the specimens were dehydrated the better. From absolute alcohol, the specimens were placed in a solution of equal parts turpentine and paraffin. The solution containing the specimens was then raised gradually from a temperature of 20° C. to about 45° C. They were then placed in melted paraffin, kept as nearly at 50° C. as possible. Small specimens will be permeated in one or two hours, but large specimens require from four to six hours.

From the 75 per cent. alcohol I placed the specimens in a stain. The stains I tried were alum-cochineal, hæmatoxylin, fuchsin, methyl-green, methyl-blue, methyl-violet, and ammonia-carmin. I found alum-cochineal a good stain for fungi, plumules, stems, roots, and root-tips, but it would not penetrate the cucumber cotyledons. Fuchsin would penetrate anything I tried; but as it is soluble in alcohol, it is necessary to overstain the specimens, and then allow the colouring to come out until it is about right. Hæmatoxylin stained all the tissue that I tried except the young cucumber cotyledons. This stain gives large specimens a dark blue colour on the outside, and a purplish-pink colour on the interior. The nuclei and the cell-walls are brought out clearly. I did not have good success with the methyl colours, as they were easily dissolved out by the alcohol.

If specimens have not taken sufficient colour, or if the alcohol has removed too much of the colour, sections can be stained upon the slide, after they are cut. Any stain can be used, but none that I tried differentiated the parts sufficiently. Fuchsin will give enough colour in a few seconds. The sections must stand in hæmatoxylin from two to ten minutes, and in alum-cochineal from ten to twenty minutes. If it is intended to stain upon the slide, an alum fixative will be found better than collodion.

I heated the slides in the gas-flame to melt the paraffin, and poured on turpentine to wash it out. The specimens were then mounted in balsam dissolved in chloroform. Air-bubbles that appear when sections are first mounted, will disappear after the slides stand a few hours. If the razor or knife used for cutting is very sharp, small specimens may be cut 1/2500 or even 1/5000 in. in thickness. But larger specimens cannot be cut more than 1/600 to 1/1500 in. thick without crowding the tissues together and giving them the appearance of being shrunk.

**Preparing, Preserving, and Mounting Objects of Natural History for the Microscope.\***—Mr. N. Pike says his own method of procedure in selecting, preparing, and preserving small delicate specimens (excepting eggs) is as follows:—

“I first procure the most perfect live specimens and drop them in strong alcohol, and let them remain about twenty-four hours. This not only instantly destroys life without injuring the objects, but also hardens them a little. They are then taken from the alcohol and placed in small narrow tubes, which I have for this purpose, just large

\* The Microscope, x. (1890) pp. 266-8.

enough to receive them, and are then covered with the following solution:—Chloral, in crystals, 1 oz., dissolve in 5 oz. of distilled water; alcohol,  $1\frac{1}{2}$  oz.; glycerin,  $1\frac{1}{2}$  dr.; rock salt, 15 gr.; saltpetre, 30 gr. Dilute the glycerin, salt, and saltpetre in the alcohol, and when well mixed add to the chloral solution. Shake well till thoroughly incorporated, filter, and it is ready for use. The liquid, if properly and carefully made should be bright and sparkling. Larvæ, spiders, &c., when prepared according to the above formula, are really beautiful objects, and can be examined with a low power of the Microscope. If wanted for dissection, they can be removed from the tube and be returned to it without any difficulty. I have thousands of specimens of soft-bodied animals now preserved in this solution, as fresh as the day I collected them.

If the objects are required for immediate anatomical examination they can be preserved for an indefinite time, and brought to the dissecting table as fresh and flaccid as possible, by omitting the alcoholic bath.

I have always by me a jar filled with the above-mentioned fluid, in which I place specimens I intend for dissection and minute microscopical examination. This jar is always very carefully corked, as the preparation deteriorates when allowed to evaporate.

The preserving of small objects in these tubes of pure white flint glass is far preferable to the building of glass cells, which are often leaky and easily get out of order. When very small objects are required I make a slight difference in the solution, but only long practice can give the precise methods for each article, as different specimens require different manipulation.

Goadby's solution makes fine preparations, but in time the corrosive sublimate in it produces white deposit on the specimen and spoils it. All solutions containing much glycerin are apt to affect calcareous substances when present.

Among many of the freshwater and marine Algæ I have succeeded in preserving specimens, to my perfect satisfaction, in the following solution:—Distilled water, 1 oz.; rock salt, 2 gr.; alum, calcined, 1 gr.; carbolic acid, 1 drop.

Some specimens of Algæ, now twelve years in this solution, are as fresh and bright as when first prepared.

Chloride of zinc solution is very useful, and has proved satisfactory in the preservation of animal tissues; it must be made of varying strengths, according to the softness of the parts to be preserved. It is recommended to use twenty to twenty-five grains of the fused chloride to one ounce of distilled water, and ten drops of phenic acid added to it. This is a capital solution for the larvæ of insects, and if stored in an air-tight tube or cell, will keep perfectly for years without deterioration."

BACHMANN, O.—*Leitfaden zur Anfertigung mikroskopischer Dauerpräparate.* (Instructions for making permanent microscopic preparations.)

München und Leipzig, 1890.

LOEWENTHAL, N.—*Zur Frage über die Anwendung von Terpentinöl in der histologischen Technik.* (On the use of turpentine-oil in histological work.)

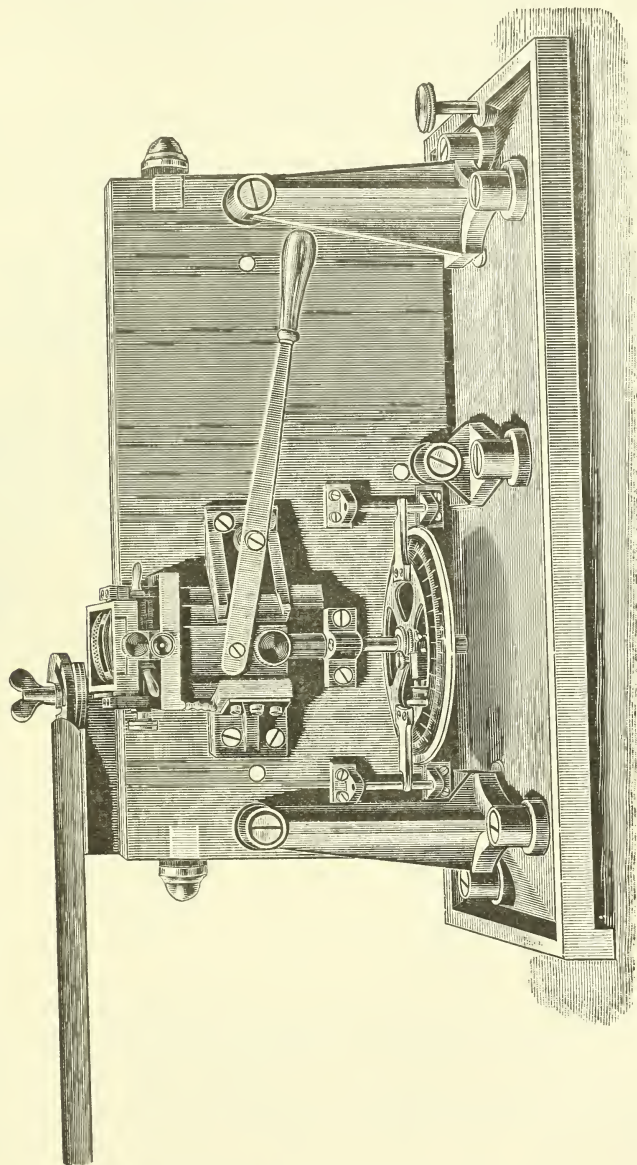
*Centrabl. f. Physiol.*, XXV. (1889) 2 pp.



(3) Cutting, including Imbedding and Microtomes.

**Improvement in Thoma's Sliding Microtome.\***—Prof. R. Thoma has recently made an improvement in his microtome (fig. 94) whereby the

FIG. 94.

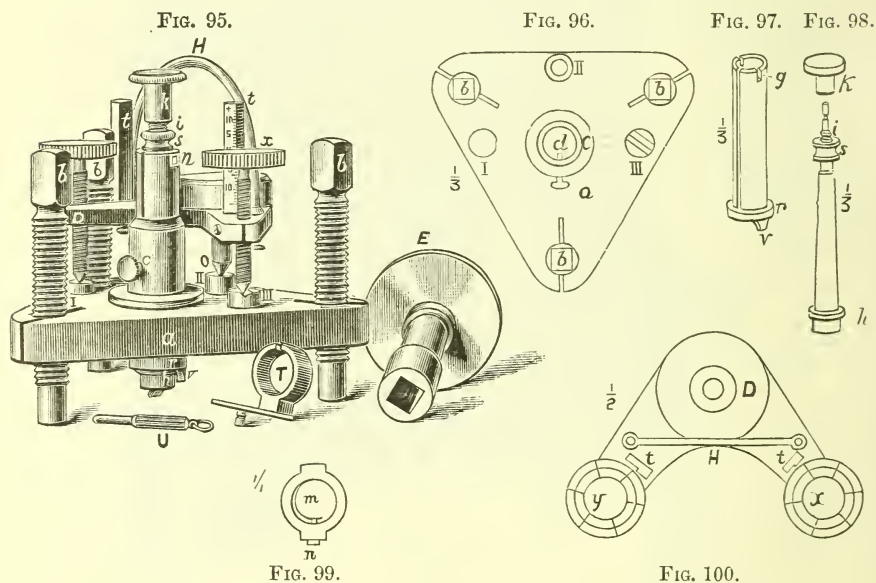


\* Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 161-4 (1 fig.).

advantages derived from the slides resting on five supports are still secured. Under previous constructions it was impossible to cut an object more than 1 cm. high without moving either the knife or the preparation. The new arrangement allows continuous sections to be made from objects 3 cm. high. The object slide is now vertical, and the holder movable about two horizontal axes. In consequence of all these changes the instrument is considerably larger and heavier.

**Apparatus for preparing Sections of Crystals cut in definite directions.\***—Dr. E. A. Wülfing gives a description of this apparatus. It consists of three entirely separate parts:—(1) a "grinding tripod," with adjustable screw-feet and a carrier for the crystal; (2) a "levelling tripod," with micrometer screw-feet; (3) a levelled glass plate.

(1) Grinding tripod.—A brass plate *a* (figs. 95 and 96),  $\frac{2}{5}$  in. thick, and in the form of an equilateral triangle of 4 in. side, is provided at each corner with an adjustable steel screw *b*. These screws have square heads, on which the key *E* fits, and they permit of the plate being inclined about  $15^\circ$  in any direction. In the centre of the plate is a tube



*c*, provided with a lug *d* (fig. 96) internally, which fits into a corresponding groove in a steel cylinder capable of free motion up and down, but not of rotation on its axis. The steel cylinder (fig. 97) is flanged at *r*, and the flange bears a projection *v*, the object of which will appear subsequently. The cylinder bears two lateral slots *g*, and its internal cavity is slightly conical. The brass cone (fig. 98) fits inside it, and is furnished at *h* with a flange and a broad disc-like base, whilst at top it bears a

\* Zeitschr. f. Kristallographie, xvii. (1890) pp. 445-59.

screw-thread *i* with a nut *s* above, which is filed square to receive the detachable milled head *k*. The base of this brass cone serves as face for attachment of crystal, and the cone can be rotated within the cylinder and clamped firmly to it when in any position. In order to prevent the cone from rotating whilst it is being clamped by the nut *s*, a steel ring *m* (fig. 99) is firmly attached to the cone just below the screw-thread by tightening the small set-screw *n*. This steel ring has two lateral lugs, which fit into the slots *g* before mentioned of the steel cylinder, and thus prevent the cone from rotating. The brass plate *a* (fig. 96) bears three small circular steel discs I, II, III in the figure, which are destined to receive the feet of the levelling tripod, and are so arranged as to occupy the angles of an isosceles triangle right-angled at II.

(2) Levelling tripod.—A circular spirit-level D (fig. 100) is provided with two arms, each of which is bored and threaded at one end to receive a micrometer-screw *x* and *y* respectively. Immediately beneath the centre of the spirit-level is a third but non-adjustable leg, seen in fig. 95 above disc II. The legs occupy the angles of an isosceles triangle right-angled at II as before described. On turning the screws *x* or *y* the level is inclined about the point *o*, i. e. the extremity of the middle leg of the levelling tripod. The amount of inclination  $\delta$  in each "screw-plane," i. e. the plane passing through *o* (above II) and either of the screw-axes, can be found if the pitch of the screw (*h*), the distance of the screw from the centre of rotation ( $\lambda$ ), and the number of rotations (*n*) are known, for

$$\tan \delta = \frac{n \cdot h}{\lambda}.$$

For practical reasons the inclination in the screw-plane is not measured in degrees on a divided circle, but by means of the tangent of the angle expressed in the number of rotations of a screw. For small angles, the angle and its tangent are interchangeable, e. g. if

$$n \text{ rotations} = 1^\circ 0' 0,$$

then

$$\begin{array}{rcl} 5 \ n & = & 4^\circ 59' 3 \\ 10 \ n & = & 9^\circ 54' 1. \end{array}$$

The distance of the screws from the centre of rotation of the tripod and the pitch of the screws are so arranged that one rotation of the screw corresponds approximately to  $1^\circ$  of inclination in the screw plane. Each screw bears a divided head, permitting of  $1/12^\circ$  being read off and a single minute being estimated, whilst complete degrees (rotations) are indicated by the vertical scale *t*.

(3) Levelled glass plate.—This consists simply of a plate of mirror plate-glass, 6-8 in. square, supported on the top of a wooden bracket firmly fixed to the wall. Three brass screws with milled heads support the plate, and in conjunction with the spirit-level permit of its being accurately levelled.

(4) Method of preparing the apparatus for use.—The use of the discs I, II, III (fig. 95) is to permit the levelling tripod to be brought into a perfectly definite position on the top of the grinding tripod without



permanently connecting the two. During the grinding process the levelling tripod is lifted off by means of the handle *H* and put on one side to save it from damage or derangement. Before using the apparatus the three legs of the levelling tripod are brought to the same length by turning the screws *x* and *y* down to the zero of the scale *t*. The levelling tripod is placed in position on the top of the grinding tripod, and the two combined on the top of the levelled glass plate. The bubble of the spirit-level is brought to its central position by turning the screws *b* of the grinding tripod, and then the levelling tripod is laid aside. A small flat face *F* (not shown) is then ground (with emery and water on a glass plate) on the projection *v* of the steel cylinder, the brass cone being laid aside. After ascertaining that the three screws *b* have worn equally during the grinding, which is shown by the bubble being still in the centre when the two tripods combined are again tested on the levelled glass plate, the face *F* is polished. The screw *x* of the levelling tripod is next to be brought to one of its extreme positions, e. g.  $+12^\circ$  on the scale *t*, the bubble is again brought to its central position by means of the screws *b*, the levelling tripod is lifted off, and a second face *f'* is ground on the projection *v*. Similarly, after turning the screw *x* to its other extreme position at  $-12^\circ$ , a third face *f''* is ground. These three faces must lie in a zone, if the apparatus acts properly, and their angles will be very nearly twice  $12^\circ$ .

(5) Theory.—Assuming that we know the position of two faces *A* and *B* of a crystal, it is required that a new face *C* shall be ground which shall make with *A* and *B* the angles *b* and *a*. The face *F* ground on the projection *v* of the steel cylinder serves for orientation.

For the orientation of the crystal with regard to the plate on which it is to be ground, we require to determine the inclination of *A* and *B* to the steel face *F*, for this latter face is parallel to the glass grinding plate when the levelling-tripod screws stand at zero. In order to arrive at the desired face *C* from the face *F* the crystal must be so inclined that in its new position the grinding plate comes to lie at the same angle to the faces *A* and *B* as that at which *C* is required to lie, or, in other words, that *C* takes the place previously occupied by *F*.

The crystal is cemented in approximately the required position, judged by eye or with the aid of a hand-goniometer, to the base of the brass cone (fig. 98) at *h*. The brass cone is then rotated within the steel cylinder until one of the known faces, e. g. *A*, falls in the zone of the steel faces *f'* *F* *f''* [not figured]. After this adjustment has been made, and *A* thus brought to a position at right angles to the screw-plane passing through the screw *x*, the angles *a'* and *b'* which *B* and *A* make with *F* are measured on a goniometer. The corrections necessary to be made in the position of the crystal are then calculated. For the details of the method of calculation we must refer our readers to the original paper. Our means of correcting the position of the crystal depend on our being able to incline it in two planes at right angles to one another, which we may assume to be the screw-planes of the levelling tripod. We carry out these corrections by turning the micrometer-screws in the reverse direction to that which would be required if the crystal occupied the position of the spirit-level. We then place the so adjusted levelling tripod again on the top of the grinding tripod—which latter we assume

to be still perfectly level—and bring the bubble of the spirit-level to its central position by turning the screws *b*. We have thus brought the crystal into the required position for grinding the face *C*, which, laying the levelling tripod aside, we proceed to do.

The adjustment of the face *A* in the zone of the faces *f' F f''* is made on a reflecting goniometer with a horizontal limb, such as that of Websky-Fuess No. II. or III. A ring *T* (fig. 95) serves to hold the cylinder on the goniometer. Instead of depending upon calculation, the correction in the position of the crystal can be often sufficiently accurately made by two or three trials, a small surface being ground and polished, and its accuracy being tested on the goniometer without detaching the crystal from the brass cone or the latter from the cylinder.\*

OBREGIA, AL.—*Serienschnitte mit Photoxylin oder Celloidin*. (Serial sections with photoxylin or celloidin.) *Neurol. Centralbl.*, 1890, No. 10, 3 pp.

ROSS, J. F. W.—*Paraffin Method as used by Prof. Gaule, Zurich*. *Canad. Pract.*, XIV. (1889) p. 409.

#### (4) Staining and Injecting.

**Staining with Chloride of Gold.**†—Prof. A. S. Underwood writes:—“I have long regarded this agent as one of the most useful for the observation of the dental tissues. I know of no other stain which so clearly marks out the minute anatomy of the soft tissues which penetrate bone and dentine; in fact, its excellence as a selective stain would long ago have obtained for it a much more widespread popularity were it not for the fact that it has been generally regarded as specially liable to failure in manipulation. Almost all the recognized text-books speak of it as a very difficult stain to employ successfully, and as requiring a very lengthy and troublesome method of procedure, and as only applicable to perfectly fresh tissues. I have found, after some eight years of pretty constant use, that the subjoined method is easy to employ, does not take long, and is, moreover, both certain and fairly permanent in its results.

First, about the tissues to be stained. They do not require to be very recently dead; the fresher they are the more quickly they take the stain, but I have stained scores of sections of teeth and bone that had been severed from the living body for a long time, sometimes for weeks. It is better to avoid as far as possible the use of metal instruments, bone, wood, or quill being preferable; the use of steel does not, however, doom the staining to failure. The method I adopt is as follows:—

(a) Wash the sections in solution of bicarbonate of soda.

(b) Put some 1 per cent. solution of chloride of gold in a watch-glass, test it with litmus-paper, and if it be acid add bicarbonate of soda by drops till it is neutral; place the sections in the solution and cover the watch-glass with some lid to keep it in the dark (the lid of a china pot such as is used for potted meat serves very well) for from half an hour to an hour, until the sections look straw-coloured.

(c) Remove sections from staining fluid to distilled water, and leave

\* We understand that this instrument is only to be obtained from Herr Zimmermann, Hauptstrasse, Heidelberg.

† Journ. Brit. Dental Assoc., xi. (1890) pp. 696-7.

them covered over (they must never be exposed to light for more than a few seconds) for a few minutes.

(d) Put some 1 per cent. formic acid in a watch-glass, float the glass in hot water, put the sections in the acid, cover them over, and keep them in the dark and fairly hot until they turn crimson. This generally takes about an hour, but the operator must be guided by the tint of the sections, which he must look at from time to time. A simple way to do this is to fill an old china anchovy paste-pot with hot water, place it on a stove, float the watch-glass containing the acid and the sections in it, and cover it up with its own lid.

(e) When stained, immerse the sections in cold distilled water for about half an hour.

(f) Dry sections and mount them in glycerin-jelly. Avoid Canada balsam. I have always found specimens mounted in Canada balsam go wrong. The bottle of gold chloride must always be carefully kept in the dark.

I have found this method very successful and very easy; moreover, I have many sections now in my possession which are quite eight years old and have not faded in the slightest degree."

**Vital Reaction of Methylen-blue.\***—Herr H. Kühn obtained specially purified methylen-blue for injecting into the dorsal lymph-sac of frogs. The reaction was obtained in about thirty-six hours by injecting every twelve hours one ccm. or three ccm. all at once of the strong solution.

When the animals were opened no effect was visible, but after five to ten minutes, all the organs, especially liver and kidneys, became blue from exposure to the air.

The preparations do not keep long, so they are useless as permanent microscopical preparations.

**Influence of Colouring Matters on Spermatozoa.†**—Dr. Emma Leclercq gives the following tabular statement as to the effects of various reagents on spermatozoa.

Colouring Matter.	Results.	
	On Nucleus.	On Accessory Corpuscle.
Carmine alone (Frenzel) .. .. .	Light carmine	Pale rose
" " (Flemming) .. .. .	Rose	Red
Renault's hæmatoxylin .. .. .	Violet	Deep violet
Ranvier's picrocarmine .. .. .	Rose	Orange
Ehrlich's violet and eosin .. .. .	Violet	Rose
" " and carmine .. .. .	Violet	Rose
Picrocarmine and methyl-green .. .. .	Violet	Carmine yellow
	(The spermatozoa being green)	

**Preparing Nerves stained by the Vital Methylen-blue Method.‡**  
—Herr B. Feist injects frogs with three or four ccm. of a strong solution

\* Arch. f. Anat. u. Entwicklungsgesch., 1890, pp. 113-5. Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 230-1. † Bull. Acad. Roy. Belg., lx. (1890) p. 138.

‡ Arch. f. Anat. u. Entwicklungsgesch., 1890, pp. 116-84 (2 pls.). Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 231-4.

of pure methylen-blue. The pigment is dissolved in physiological salt solution. The stain is fixed by means of the iodide and iodine solution or Hoyer's picrocarmine.

Such preparations are examined and mounted in glycerin. If, however, the pieces are imbedded, then platinum chloride must be used as fixative, since the picrocarmine and iodine are easily dissolved out by the various reagents used for imbedded specimens.

A still better method is to fix with picrocarmine for fifteen minutes, followed by one per cent. osmic acid for the same time, and then glycerin for some hours. The specimen can now be imbedded by the gum arabic method. This consists in taking a watchglassful of a solution of pure gum arabic having the consistence of thick syrup. To this are added six to ten drops of glycerin, and the whole stirred up with a glass rod. In this mixture the sections are placed and the whole left to dry until it has assumed a consistence suitable for cutting (about eight days). The imbedded object is then clamped in elder-pith and sectioned.

The chief objection to this method is that when the mass has acquired the proper consistence it must be cut at once, otherwise it becomes too hard.

**New Method for Staining Sections of Central Nervous System.\***—For staining sections of central nervous system, A. Breglia uses extracts of Campechy or Pernambuco wood. The former contains hæmatoxylin  $C_{16}H_{14}O_5$ , the latter brasilin  $C_{22}H_{20}O_7$ .

The extract is made as follows:—7 to 10 grm. of the wood in small pieces are soaked in 90 to 95 per cent. alcohol for five or six days. The mixture is then shaken up and the fluid is ready for immediate use.

Sections of nervous tissue hardened in Müller or Erlizki's fluid are placed for ten to fifteen minutes in 15 ccm. of 90 per cent. alcohol to which has been added three to seven ccm. of a saturated aqueous solution of neutral acetate of copper. The sections are next immersed for five to ten minutes in a saturated watery solution of lithium carbonate. They next come into ten ccm. of the extract for 18 to 24 hours. Decoloration is then effected with aq. dest. 100 grm., ferricyanide of potash 1 grm., borax 1 grm. When the grey and white matters have become differentiated to the naked eye, the sections are washed in distilled water, and then mounted in the usual way. For the Pernambuco wood extract, the method of manufacture and the manipulative procedure are the same, with the exception that the decolorizer acts very much more rapidly.

**Staining Central Nervous Tissue with Palladium Chloride.\***—Prof. G. Paladino recommends the chloride of palladium for staining sections of the central nervous system. The procedure is as follows:—To a 1 per thousand solution of chloride of palladium a few drops of hydrochloric acid are added in order to insure its complete dissolution. In this solution pieces of spinal cord 5 mm. thick are immersed. The cord has of course been previously hardened in bichromate salts, chromic acid, or in sublimate. In the palladium solution, of which a large quantity (150 to 200 ccm.) are used for each piece, the objects are

\* Giorn. d. Assoz. dei Naturalisti e Medici di Napoli, i. (1889) pp. 169-72. Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 236-7.

† Journ. de Micrographie, xiv. (1890) pp. 142-8.



left for two days, and are then transferred for twenty-four hours to a 1 per cent. solution of iodide of potassium. The pieces are next dehydrated in spirit from 80° to 96° successively, and when completely freed from water they are treated with chloroform and then imbedded in paraffin. The paraffin is removed from the sections with xylol, and these are then mounted in balsam.

This method, which is extremely simple, is applicable to peripheral as well as central nerves; the results obtained from it are extremely favourable. The hue imparted by the reaction of the iodide on the palladium is brownish, and allows the finer structural details, both of nerves and nerve-cells, to be easily seen.

**Method for Staining Sections of Spinal Cord.\***—Dr. R. Haug finds that the following method is simple and satisfactory for preparing and staining sections of spinal cord.

Fresh pieces of spinal cord of half to one cm. thick are immersed for two days in a saturated solution of neutral acetate of copper. After this they are placed for one to one and a half days in a five per cent. solution of bichromate of potash. After washing off the superficial deposit of chromic acid salt, the pieces are placed in the dark in 70 per cent. spirit for thirty-six to forty-eight hours, then for a similar period in absolute alcohol. They are now ready for imbedding in paraffin or celloidin. The paraffin having been removed, the sections are placed in the following solution (hæmatoxylin 1, in alcohol 30, plus ammonia-alum 1 in 300 H<sub>2</sub>O), for fifteen to thirty minutes, or until they are of a deep black colour. After having been washed in water, the sections are toned down in muriatic acid 0·5 to 1·0, alcohol 70·0, H<sub>2</sub>O 30·0. When sufficiently decolorized (fifteen minutes at most), the now red sections are washed for a long time in pure water until all the acid is removed and the colour is blue.

The sections may be contrast-stained by immersing them for a moment in undiluted neutral carmine solution, or in the following, which imparts a very pretty tone:—To 100 ccm. water, 0·25 carbonate of magnesia and 15–20 drops of liq. ammon. fort. are added. The mixture is heated, decanted off, and filtered. To the filtrate 0·59 carmine are added.

Should Weigert's method of differentiating be preferred, this may be effected by decolorizing the sections in the borax-ferricyanide of potash solution, and then proceeding in the usual manner.

The author claims for his method that it is not only not very complicated, but that it allows of a satisfactory examination of the cord in a comparatively short time.

**Method for Staining the Gregarinæ of Molluscum contagiosum.†**—Dr. R. Haug recommends the following procedure for demonstrating the Gregarinæ of Molluscum contagiosum. Fix for twenty-four hours in absolute alcohol, to which 1 per cent. of glacial acetic acid has been added. The specimen is then washed in running water for twelve hours; it is now hardened again in absolute alcohol (six to twelve hours), and then imbedded in paraffin. The sections are first stained with hæmatoxylin (hæmatox. 1 to 30 alcohol, added to ammonia-alum 1 to 300 H<sub>2</sub>O). The sections are first over-stained and then differentiated with

\* Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 153–5. † T. c., pp. 152–3 (1 pl.).

hydrochloric or oxalic acid alcohol. They are then placed in water for fifteen minutes. The sections are next momentarily immersed in ammonia-carmin solution, then in water, and lastly placed for ten to fifteen minutes in absolute alcohol, to which 2 per cent. of formic acid has been added. After this they are transferred to picric acid alcohol for fifteen minutes. Thus prepared the sections have a bluish-green hue; the gregarinæ are greenish, the cells blue, and the rest of the tissue rose-coloured.

**Carmin Stains for Normal and Pathological Preparations.\***—Dr. R. Haug prepares a double carmin stain for staining pieces *in toto* as follows:—2 gm. of carmin are rubbed up with 4 gm. of borax-carmin, and then boiled in a flask with 300 ccm. distilled water until the fluid is evaporated down to 280 ccm. After this, and when the solution has cooled down a little, from 10 to 15 ccm. of a 10 per cent. solution of acetic acid (glacial) are added by means of a pipette. The addition of the acid renders the solution transparent and of a bright red hue. Next day it is filtered and some crystals of thymol added. For staining *en masse* a piece of 0.5 cm. in width, two to four days is required. After this it is differentiated with hydrochloric acid alcohol (changed every half hour). This takes one to four hours. After this it is placed in a mixture of picric acid and alcohol for about twelve hours.

**Ammonia-lithia-carmin.**—This solution is made by dissolving 3 gm. carmin in 100 ccm. of cold saturated carbonate of lithia solution and then adding 5 ccm. of ammonia. It stains quickly and deeply. Wash in water and then differentiate in hydrochloric acid alcohol. Sections may be after-stained by immersing in picric acid alcohol.

In many cases when the specimen has been hardened in chromic acid, the following modification acts well:—1 to 1½ gm. carmin and 2 gm. bicarbonate of soda are boiled in 150 ccm. of water, and then 10 to 15 ccm. of a 5 per cent. glacial acetic acid added. When cold 5 ccm. lithium solution. The subsequent treatment as before.

(5) Mounting, including Slides, Preservative Fluids, &c.

**A new Pressureless Mounting-clip.†**—Mr. T. Pace writes: "I notice Mr. Bryan's note in the December number of 'Science-Gossip,'

FIG. 101.

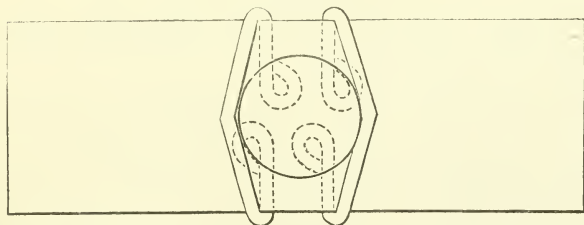
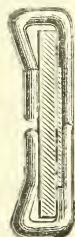


FIG. 102.



suggesting a new form of mounting-clip designed to hold the cover-glass without pressure, thus being a great improvement on the spring-clip

\* Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 151-2.

† Science-Gossip, No. 303 (1890) p. 56.

commonly used; and as I have devised one which I think has some advantages over his, I inclose you a sketch of it, thinking it may interest some of your readers. Fig. 101 is an illustration of the clip in use as seen from above, and Fig. 102 is an end view of the same as seen from a section through the cover-glass. The clip should be made of rather stout springy wire, so as to grasp the slide firmly. This clip will be less liable to shift or become detached from the slide, and it has the advantage of being suitable for rectangular as well as circular cover-glasses."

**Use of Gold Size.\***—Mr. F. Disnett writes:—"Gold size as a foundation ring for coloured cements answers much better than shellac in mounting insects whole or in parts, or other materials where a thick layer of balsam is needed. Much trouble is caused in finishing with alcoholic cements, because the alcohol softens the thin crust of hardened balsam at the edge of the cover, air-bubbles appear, and the same often happens again in finishing with coloured cements. The colours will run in and spoil the mount. All this is avoided with gold size. Another advantage is that in the final cleaning the last trace of balsam on cover or slip can be washed off with alcohol. Gold size answers much better than arabin in protecting the ring for cleaning with alcohol, as the latter has no affinity for it, and arabin is well known to be one of the strongest cements we have. Before putting on the foundation ring, mounts should be carefully examined, to see whether the balsam extends to the edge of the cover. Small cavities often exist where an air-bubble has made its final exit. The cement will inclose the air, the thin film will collapse and leave the balsam unprotected. Gold size is slow in drying, but if one builds a number of cells to-day, they will be exactly a month old a month from now, and in time will get as hard as glass. Make them with one or two, seldom three, successive coats, and always aim to have a lot of old cells on hand. When I wish to mount in one I put on a light coat of fresh gold size, drop in the glycerin, and dissect whatever it may be in the cell. I use 3/4-in. cells mostly, and as I disentangle and separate different structures I push them aside, and keep on till I have material enough to fill the cell. I arrange what I have as well as possible, fill up with glycerin, and cover. Lots of handling and transferring are done away with."

**Laboratory Notes.**—Mr. A. F. Stanley Kent writes to us:—

(a) *Farrant's Medium.*—In making Farrant's medium for a large class it is desirable to modify the usual mode of procedure. The following method I have found satisfactory.

Make a solution with picked gum arabic to about the consistence of ordinary glycerin, mix it with an equal bulk of Price's glycerin and place it in an ordinary plaited filter, a few pieces of glass rod being placed in the funnel to keep the paper from lying too closely against the glass. Now close the mouth of the funnel by means of a glass plate on which vaseline has been smeared, place a flask under the funnel so as to fit the neck as closely as possible, and put the whole away for some weeks. A beautifully clear and bright solution will filter through into the flask.

\* The Microscope, x. (1890) pp. 281-2.

I always add, before filtering, about 1/20 of a strong solution of thymol in absolute alcohol to the gum and glycerin (i.e. about one part of alcoholic solution of thymol to 20 parts of gum and glycerin solution). This, of course, precipitates some of the gum, but it soon becomes redissolved, and Farrant prepared in this way has kept sweet for many months.

The use of the glass plate is, first, to exclude dust, and secondly, and chiefly, to prevent evaporation.

(β) *Staining with Picrocarmine*.—In the histology courses at Oxford we use this stain very largely, as it is very convenient, easy to make, and preparations made with it are permanent. We stain the sections on the slide, remove some, *but not all*, of the stain with blotting-paper, and mount in Farrant; the surplus stain becomes mixed with the Farrant, but in a few days the section absorbs nearly the whole of it and as a result exhibits a better differentiation than can be obtained by any other means with which I am acquainted. I first saw the stain used in this manner by Professor Stirling, in the Owens College, Manchester.

(γ) *Connective Tissue*.—It is often difficult, in a class of histology, to demonstrate by means of the ordinary reagents the presence of connective tissue corpuscles in preparations of areolar tissue.

The following method has been found satisfactory:—Snip out a small piece of the subcutaneous connective tissue of a recently killed rabbit, spread it upon the slide by means of two needles *without the addition of any reagent*, then flood it with absolute alcohol for about one minute, remove alcohol, stain with hæmatoxylin, dehydrate, and mount in balsam.

Excellent preparations may also be made with methyl-green, but this reagent is not usually supplied to students in a class, requires special skill in its use, and is not permanent.

(δ) *Aspinal's Enamel*.—As a cement for ringing slides and making cells Aspinal's enamel has proved of great use. I have used it for some time and find that the white keeps well when used for balsam and Farrant mounts; but I have not used it for a sufficient length of time to know whether it possesses any advantages over zinc white cement.

**The Differentiator.\***—Mr. N. A. Cobb describes an improved form of the differentiator, or instrument for avoiding to the greatest possible extent those annoying and often destructive contractions which occur in delicate organisms while they are being killed and preserved. The instrument is made of glass tubing with an internal diameter of five or more mm.; two forms are shown in figs. 103 and 104. *a* or *a'* is the reservoir, *b* the object-box, and *c* the filter; these are three pieces of glass tubing joined by caoutchouc tubing. The filter is made by taking a piece of glass tube twice the required length, heating it red hot, drawing it out to arm's length, and breaking in two in the middle; the extremity should be drawn out very fine and a minute orifice alone left.

Taking an example of objects fixed by corrosive sublimate which are to be studied in balsam after staining with borax-carminé, the author directs us to proceed as follows. Fill the filter with perfectly clean sublimate

\* Proc. Linn. Soc. N.S.W., v. (1890) pp. 157-63 (1 pl.).



solution, and insert a plug of cotton (boiled in water to remove the air) at the U-bend. Join the object-box to the filter, fill up with sublimate solution, and push a plug of cotton into

FIG. 103.



FIG. 104.



the lower end of the box. Put the objects into the box, plug the upper end, and join the box and filter to the empty reservoir *a*. Now mix equal parts of sublimate solution and 33 per cent. alcohol (solution 2); mix equal parts of solution 2 and sublimate solution (solution 1). Mix equal parts of solution 2 and 33 per cent. alcohol (solution 3). Add solution 1 to the reservoir until it is one-fourth full, solution 2 till it is half full, solution 3 until it is three parts full, and fill up with 33 per cent. alcohol. If the solutions are carefully added the difference in specific gravity will prevent them mixing; if forced rapidly in, a nearly uniform mixture of about equal parts of sublimate and 33 per cent. alcohol will be formed. It is desirable to get an intermediate condition, when we shall have a uniform gradation or differentiation from sublimate solution to 33 per cent. alcohol in passing upwards through the reservoir. The objects should next be passed through borax-carminé, 50 per cent., 70 per cent., 90 per cent., and absolute alcohol; and finally to thin balsam. The differentiator should be used throughout the operation.

Objects which contract unexpectedly may be rendered insensible by the use of alcohol from 5 per cent. to 30 per cent., or through chloral hydrate, when they will be found insensible and outstretched.

For further hints in manipulation, and a list of the mixtures used, we must refer to the author's detailed account.

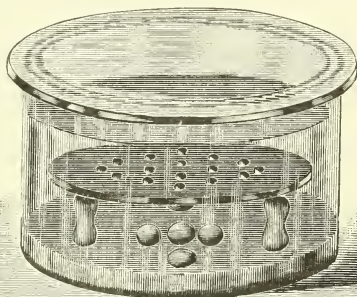
**How to clean old Slides and utilize spoiled Mounts.**—Dr. H. M. Whelpley, in a paper read at the St. Louis Club of Microscopists, said, "For two years past I have permitted soiled slides and spoiled mounts to accumulate in a box set aside for that purpose. The process I have recently followed in reclaiming them has been successful. I first placed the unsightly rubbish in a dish of clean water, where it remained until all of the labels were readily removed. With an old knife I next scraped off the cells and all cement that could be easily removed in this manner. All slides where glycerin or other substance soluble in water had been used as a mounting medium were again washed, and then the entire pile spread out and dried. I separated those that were clean, and placed the rest in alcohol for several days. This solvent cleaned another

portion of the slides, so that all they required to render them as good as new was a washing in water. The remaining dirty ones were treated to a bath of oil of turpentine, where they rested for a few days. From this they were washed with alcohol, and then finished in water. The few refractory ones that held out during all this time were made as clean as ever with benzol. Although considerable time elapsed before the last slide was cleaned, it required but a few minutes of actual labour in the entire process. The time consumed is in letting them stand in the different liquids. Nor is the process expensive, as the oil of turpentine did most of the work. Hereafter I shall divide my old slides into three classes, and clean them separately, so that less alcohol will be required. The first box will contain slides that can be washed clean with water; the second lot will be those that alcohol will clean, and the third the ones requiring benzol. Cover-glasses are so cheap that I do not save them unless they are easily cleaned with water. I find it very difficult to properly clean thin cover-glasses that have cement on them."

**Anilin Oil in Microscopical Technique.\***—The brown colour of anilin oil, says Dr. Suchanek, the result of oxidation, is no bar to its use as a clarifying medium, or if intended for mixing with absolute alcohol. It is, however, of importance that it should be quite free from water, and it is advised that the oil should be distilled in the usual way and that the first 10–12 ccm. of distillate should be removed. The rest of the distillate should be received in a dry flask in which have been placed pieces of caustic potash. By this means the last traces of water will be effectually removed. The chief use of this medium is as a hydrant and clarifier, but it is also employed as a substitute for oil of cloves in paraffin imbedding.

The author describes a glass capsule (fig. 105) which he uses for dehydrating preparations in the anilin oil. In the capsule is placed a glass tray perforated with sixteen holes and supported on three legs. Underneath the tray are placed some pieces of caustic potash, and the anilin oil rises above the plate-level some millimetres. This device allows the preparations to be thoroughly dehydrated.

FIG. 105.



PIERSOL, G. A.—**Fixing Paraffin Sections to the Slide.**

*Univ. Med. Magazine Philadelphia*, II. (1889–90) p. 149.

STIRLING.—**Dry Cover-glass Microscopical Preparations.**

*Journ. of Anat.*, XXIV. (1890) p. 160.

\* *Zeitschr. f. Wiss. Mikr.*, vii. (1890) pp. 156–9 (1 fig.).

## (6) Miscellaneous.

**Some Practical Business Applications of the Microscope.\***—The man who has learned the use of the Microscope has certainly gained a great deal; but the man who claims to be a scientist without knowing the practical value of the Microscope, and without having learnt its use, ought not to be classed as such. The Microscope when first invented was considered as an accessory or a plaything. But since 1820, and later (1840), the first European oculists and scientists began to make microscopical researches, not only in the medical profession, but also in botanical, geological, and other studies. Since 1860 and 1870, the world over, the Microscope has been applied to almost every study and analysis. Had Galen, Celsus, and Hippocrates, and other ancients, had the use of the Microscope they would not have advocated the theory that the arteries in the human being contained air during life, instead of oxygenized blood. They were of the erroneous opinion that the blood simply acted as a humour in lubricating the tissues. Had it not been for the Microscope, James Paget, the great English surgeon and physician of St. Bartholomew's Hospital, in the year 1834, would not have discovered the *Trichina spiralis*, which had already slaughtered its thousands, dating as far back as the time of Moses.

The Microscope is certainly the greatest aid a scientific and a professional man can have. A physician without a Microscope is like a man without his hands: he is uncertain and unprotected. He cannot arrive at a correct and positive conclusion in diagnosing and prognosing his cases. It is important to have the Microscope at hand for examining the sputa of human beings, so as to be able to state positively whether or not the man is suffering with consumption (tuberculosis). It is important to be able to determine with certainty, at an early date, whether or not a man is suffering with cancer of the stomach by examining the vomits. A Microscope magnifying from 1 to 5000 diameters is a most simple piece of apparatus. Every person can learn its use in a few hours. Every person should learn to use a Microscope, not only the professional man and scientist, but every business man, even the grocer, butcher, farmer, and the housewife.

Everything that concerns a medical examination in a legal sense, or a legal examination in a medical sense, can be determined accurately by the use of the Microscope. For example, in the Cronin case of Chicago, where the medical experts demonstrated to a certainty that the blood, hair, and brain matter found in the Carlson cottage and sewer trap was that of a human body. Not only that, but they determined accurately and positively that the hair and blood found in the cottage and in the fatal trunk were that of Dr. Cronin, only in a modified condition; all with the aid of the Microscope.

Within the last decades scientists have demonstrated to a certainty the possibility of determining dried and old human blood-spots from that of animal blood, whether on clothing, wood, iron, or otherwise.

Pathologists and histologists have also demonstrated the great value of the Microscope in determining positively the skin, hair, blood, brain-matter, also the excretions and secretions of the human being from that of the lower animals.

\* By Dr. F. Gaertner, Pittsburg, Pa., in Amer. Mon. Micr. Journ. See Engl. Mech., li. (1890) p. 483.

Again, the Microscope is applied in a medico-legal view, especially in malpractice, suits of damages, suits involving, rather than determining, the adulteration of foods and drink as to their purity, and finally, in determining whether or not food or drink has spoiled, undergone fermentation and the accumulation and development of micro-organisms, such as germs, microbes, and bacilli. Also, in the examination of oleomargarine and in the adulteration of drugs, liquors, milk, groceries, sausages, &c.

The application of the Microscope in a legal point of view is altogether new. We anticipate surprising effects from the application of the Microscope in the examination of legal documents, U.S. currency, and printed matter.

The following lines are from a very ample paper read by G. E. Fell, M.D., before the American Society of Microscopists, entitled "Examination of Legal Documents with the Microscope."

More than once has investigation with the Microscope cleared up the path of the attorney, ferreted out the work of the contract-falsifier, and shielded the innocent from the unjust accusations of interested rogues.

The range of observation in investigations of written documents with the Microscope is a broad one. We may begin with the characteristics of the paper upon which the writing is made, which may enable us to ascertain many facts of importance; for instance, a great similarity might indicate, with associated facts, that the documents were prepared at about the same time. A marked dissimilarity might also have an important bearing upon the case.

The differences in the paper may exist in the character of the fibres composing it, the finish of the surface, whether rough or smooth, the thickness, modifying the transmissibility of light, and the colour, all of which may be ascertained with the Microscope.

The ink used in the writing may be examined. If additions have been made to the document within a reasonable time of its execution, it is well to examine it microscopically with a great probability of detecting the differences of the original and additional inks. These differences may be present as follows: Some inks in drying assume a dull or shiny surface. If in sufficient quantity the surface may become cracked, presenting, when magnified, an appearance quite similar, but of a different colour, to that of the dried bottom of a clayey pond after the sun has baked it for a few days. The manner in which the ink is distributed upon the paper, whether it forms an even, somewhat regular border or spreads out to some extent, are factors which may also be noted. The colour of the ink, by transmitted or reflected illumination, is a very important factor. This, in one case, proved of great importance, and demonstrated the addition of certain words, which completely annulled the value of the document, involving several thousand dollars. And in a case where the lines of a document were written over with the idea of entirely covering the first written words, the different colours of the ink were revealed by the magnified image as seen under reasonably low powers of the Microscope.

Special attention is desired to the examination with the Microscope of written documents, United States currency, printed matter, &c., as to their genuineness from a legal standpoint. The principal feature in the examination of written and printed documents is in the erasures and the



additions, in the different colouring of different inks applied, and the mode of their execution.

Erasures can be accomplished either with a knife or by a chemical preparation. The former process is the one commonly resorted to, and is effected in the following manner: With a well-sharpened knife-blade the surface of the paper is carefully scraped until all objectionable lettering and wording is supposed by the naked eye to have disappeared. With a microscopical examination you can at once detect the impression made by the stroke of a pen. Even the different colours of the ink are still to be seen with the Microscope.

The second method being by a chemical preparation, the ink is made soluble and then easily removed from the paper by means of a blotter or absorbent cotton. This method is also an incomplete one, and the letters can easily be made out by close observation where a chemical preparation has been used for erasing. In most cases it leaves a stain, and the fibres of the paper are more or less injured by the chemicals used, always leaving evidence that the document has been tampered with.

Geo. E. Fell, in his paper, says the eye of the individual making the erasure is certainly not sufficient, and even with the aid of a hand magnifier the object might not be effectually accomplished. The detection of an erasure made by the knife is a very simple matter, and may be accomplished by the novice. An investigation may be made by simply holding the document before a strong light, and this is usually all that is necessary to demonstrate the existence of an erasure of any consequence. This is, however, a very different matter from making out the outlines of a word or detecting the general arrangement of the fibres of the paper, so as to be enabled to state whether writing has been executed on certain parts of the document. Again, when we enter into the minutiae of the subject, we find that the compound Microscope will give us results not to be obtained by the simple hand magnifier.

On several occasions I have had the opportunity of demonstrating with the Microscope additions made to certain documents, two of which were wills. The additions were made in the following manner (which the Microscope revealed): First an erasure must have been produced, then there was a writing over the erasure. With the Microscope you could at once detect the erasures and the additions; also the different colours of the inks used, and, next, the most important characteristic of the microscopical examination being in the close observation of the stroke of the pen of the original lettering and the additional lettering, and, finally, the general mode of their execution.

In the examination of legal documents, U.S. currency, printed and mutilated documents, including forgeries, &c., involving a legal question and investigation, the principal features in the microscopical examination, as already stated, are the erasures, additions, colour of the ink, stroke of the pen in the original lettering and additional lettering, and, finally, the mode of their execution. This includes the general and comparative expression of the original writing—that is, in the observation of the letters constituting the document. Especial attention is needed in the observation of the shading, and in the general formation of the letters by the stroke of the pen, either in a downward or upward movement. This applies not only to the capital letters, but also to the

smaller letters, even to the punctuation, grammatical and orthographical relationship, and in comparative differentiation. All these things must be taken into consideration.

In the examination of papers, documents, such as wills, notes, cheques, &c., as to whether or not they were mutilated and forged, the Microscope will certainly be the most reliable test, much the easiest and simplest.

This is the way of determination, and an expert microscopist and observer can at once arrive at a correct and positive conclusion as to the genuineness of the autograph, &c.

In the examination of U.S. currency the same will hold good as in the examination of written and printed matter, with the exception that additional observation is necessary in order to differentiate a genuine bill from a counterfeit. This lies in the microscopical examination (1) of the quality of paper used; (2) in the execution and finish of the bill; (3) the grade and colour of the ink; (4) the printed condition of the bill, including the autograph; (5) the most important and characteristic means of determining a genuine bill from a counterfeit bill being in the observation of the red line which runs lengthwise across the bill, and it will be necessary to notice that the two red lines in a genuine bill are simply red silk thread interwoven in the paper of the bill, when in a counterfeit the red lines are simply red ink stripes, and no silk lines whatever.

**Medico-Legal Microscopy.\***—The thirteenth annual meeting of the American Society of Microscopists was opened with a discussion on the "Proposed Standing Committee on Medico-Legal Microscopy," by Prof. Ewell of Chicago. The professor began the discussion by declaring that the Microscope was by no means the simple instrument usually imagined. On the contrary, he stated that it was an exceedingly difficult instrument to handle. Some of the pointed stories about the Microscope are the strongest points in favour of a medico-legal committee, as this would have a tendency to stop the circulation of stories exaggerating the powers of the Microscope. He called attention to an article in a scientific paper, telling how the brain-matter found in a Chicago sewer was identified as coming from Dr. Cronin's head. No one had previously heard of Dr. Cronin's brain being exposed until the autopsy. This Dr. Ewell deprecated in the highest degree. To assume for the Microscope a position of infallibility, from a medical standpoint, is an absurdity, and goes a long way towards injuring the general standard of the profession.

Dr. Frank L. James, Prof. Seaman, Mr. H. L. Tolman, Dr. Stillson, and Prof. Claypole were in favour of such a committee to correct these wrongs. Newspapers have often spoken about the identification of blood by aid of the Microscope, but the best microscopists know that they cannot positively tell human blood. They can tell the difference between the blood of amphibians, mammalia, and fowls. They were of opinion that the committee would do a very great work if it could curb the enthusiasm of those who over-estimate the field of the Microscope. Courts should be given a standard by which the power of the Microscope can be judged.

\* Amer. Mon. Micr. Journ., xi. (1890) p. 199.

When the discussion was finished, the President appointed Professors Ewell, Seaman, and Claypole, Mr. Tolman, and Dr. Stillson as a committee to map out the work of a standing committee on this branch of microscopy.

**Millon's Reagent.\***—Signor A. Poli calls attention to the unsatisfactory quality of this reagent as obtained from some of the leading manufacturers, its property of imparting a rose-colour to proteids being often only very feebly displayed. He believes this to arise from the fact that it is not a stable compound, and it should therefore always be prepared fresh by the operator himself. Millon's prescription for preparing the reagent is as follows:—Mercury is dissolved in an equal weight of nitric acid diluted by 4·5 equivalents of water. The solution is commenced in the cold, and finished by heating slightly until all the mercury is dissolved. Crystals are then formed, and the liquid is decanted and diluted with two volumes of water. The solution thus obtained contains mercurous nitrate  $\text{Hg}_2(\text{NO}_3)_2$ , mercuric nitrate  $\text{Hg}(\text{NO}_3)_2$ , and free nitric acid, and is thus a mercurous-mercuric nitrate. In consequence of the difficulty of obtaining nitric acid perfectly free from water, Signor Poli prefers Poulsen's method, as follows:—10 grm. of mercury are dissolved in 25 grm. of nitric acid of sp. gr. 1·185, heating not above 50° C.; this solution is then mixed with another obtained by dissolving 10 grm. of mercury in 22 grm. of nitric acid of sp. gr. 1·25–1·30; the reagent should be prepared and employed as much as possible in the cold.

**Tests for Mineral Acids and Mineral Bases in Plants.†**—Herr A. F. W. Schimper recommends the following microchemical tests for detecting the presence of mineral acids and mineral bases in the ash of plants:—

**Lime.**—The production of crystals of calcium sulphate by addition of sulphuric acid; also, in certain cases, the formation of crystals of calcium oxalate by addition of ammonium oxalate, or of calcium carbonate by ammonium carbonate.

**Chlorine.**—Precipitation by silver nitrate or thallium sulphate. Where present in large quantities, potassium or sodium chloride may be crystallized out of an aqueous solution of the ash.

**Potassium.**—The production of potassium-platinum chloride, insoluble in water and alcohol.

**Magnesium.**—The formation of ammonium-magnesium phosphate by addition of sodium phosphate or sodium-ammonium phosphate with the addition of ammonia. The formation of magnesium-sodium uranate, by the use of uranacetyl.

**Sodium.**—The same as the last.

**Oxalic Acid.**—Formation of calcium oxalate.

**Phosphoric Acid.**—The production of ammonium-phospho-molybdate by the addition of ammonium molybdate and nitric acid. Formation of magnesium-ammonium phosphate.

**Nitric Acid.**—Formation of an anilin-blue with diphenylamin. An extraordinarily delicate reaction. Production of calcium nitrate.

\* Nuov. Giorn. Bot. Ital., xxii. (1890) pp. 446–50.

† Flora, lxxiii. (1890) pp. 210–20.

*Sulphuric Acid*.—One of the most difficult determinations, as the occurrence of a precipitate with barium chloride by no means proves the presence of sulphuric acid. Crystals of potassium sulphate may be obtained from a solution of the ash.

*Tartaric Acid*.—May be precipitated by potassium acetate or calcium chloride.

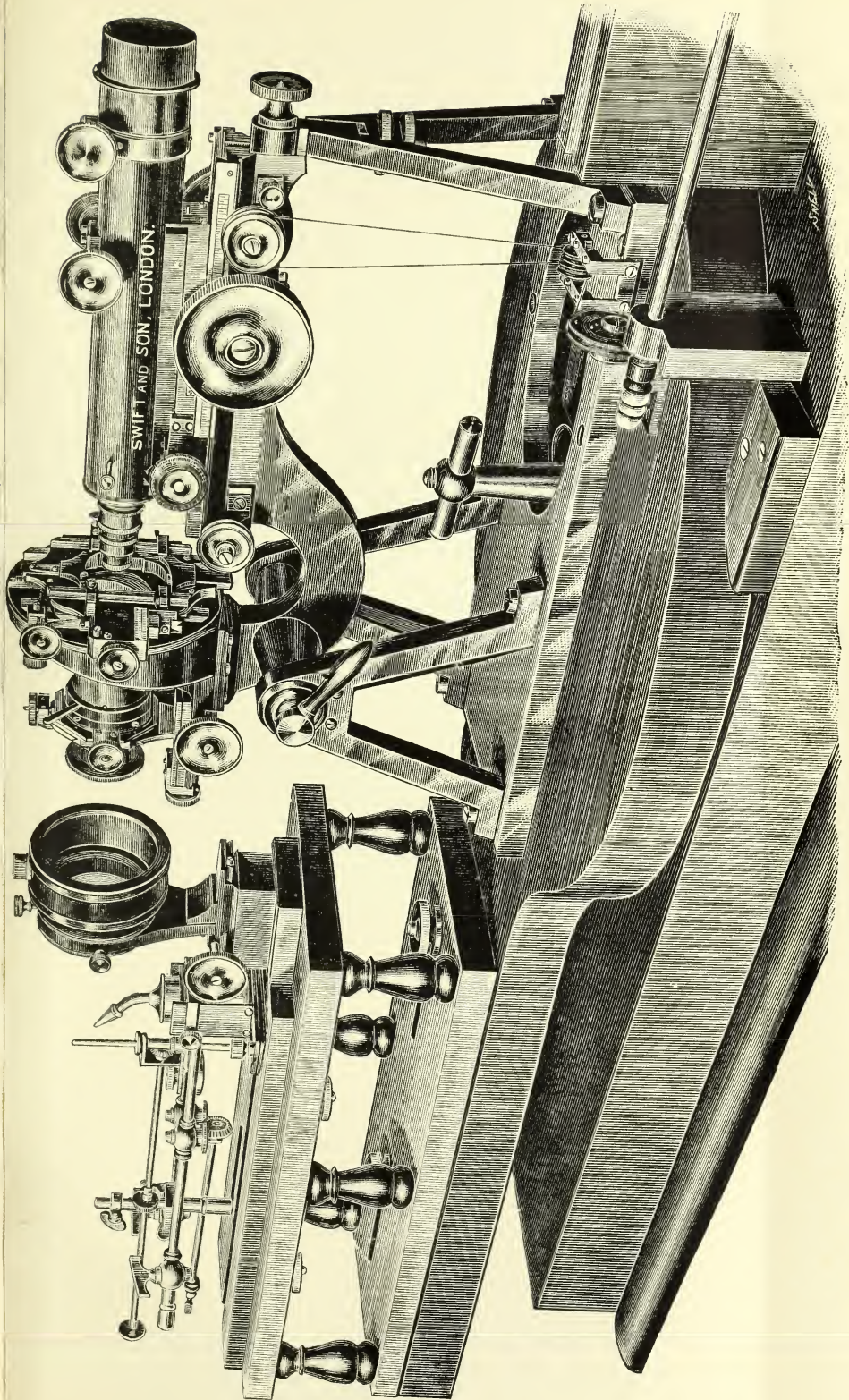
**Behaviour of Fossil Teeth to Polarized Light.\***—Dr. J. Schaffers obtained from experiments made on fossil teeth with polarized light results perfectly analogous to those previously obtained from fossil bone, and considers that the explanation of this optical effect is due to the fibrillar structure of both substances. The author's results do not accord with those of Valentin, who found that fossil and recent teeth behaved in the same manner to polarized light, while the author considers that the reverse is the case.

**Böhm and Oppel's Manual of Microscopical Technique.†**—This little work is intended for beginners; it is divided into two parts, the first of which deals with the Microscope and its manipulation, the second part is subdivided into general and special sections. In the former are considered methods of preparation, fixation, hardening, imbedding, sectioning, and staining. The special section deals systematically with the various organs and tissues.

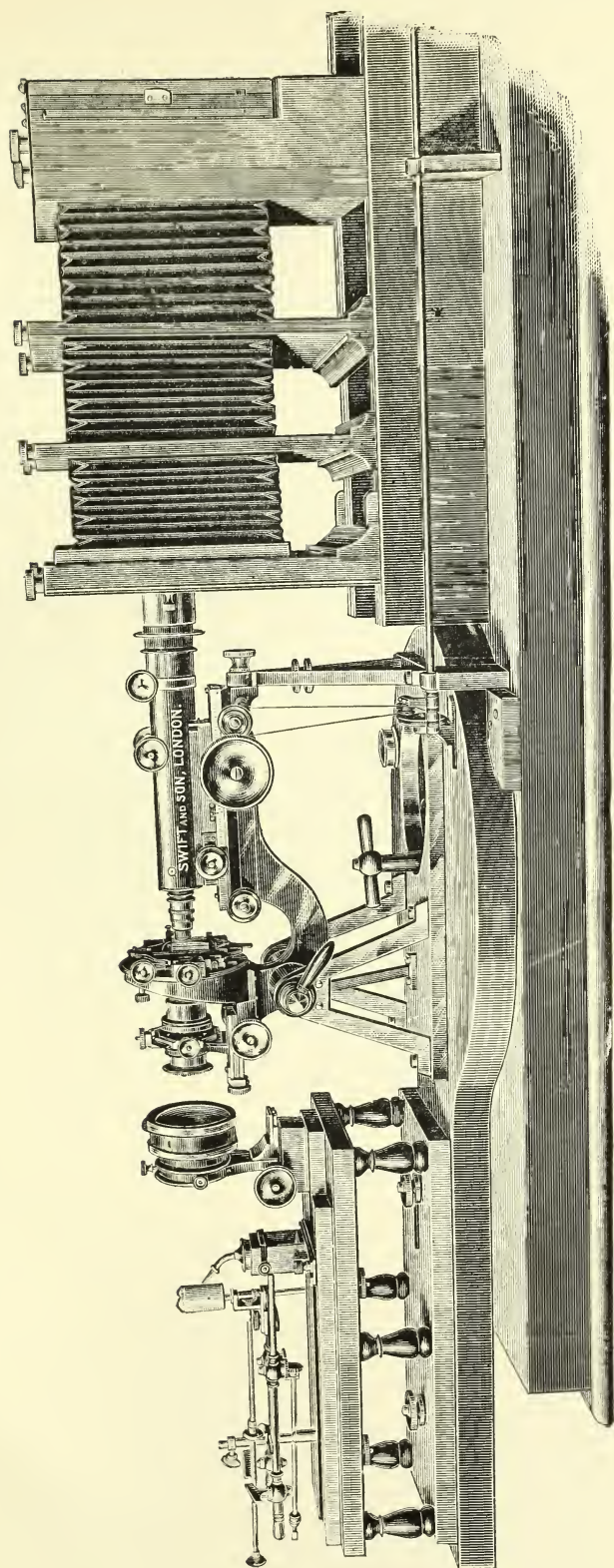
\* SB. K. Akad. Wiss. Wien, xcix. (1890) pp. 146-52.

† 'Taschenbuch der Mikroskopischen Technik.' München, 1890, 8vo, 155 pp. Cf. Zeitschr. f. Wiss. Mikr., vii. (1890) pp. 175-6.





Mr. Pringle's Photomicrographic Apparatus.



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