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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.

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MICROSCOPY.

a. Instruments, Accessories, &c.*

(1) Stands.

Stands and various Equipments.†—Attention may be called to an editorial article in which recommendable stands are described from the catalogues of Zeiss, Schieck, Leitz, and others. General remarks are made concerning apertures, eye-pieces, magnifying power, apochromatic lenses, focusing arrangements, and the methods of using Abbe's apertometer and test-plate.

(2) Eye-pieces and Objectives.

Apertures of Objectives.‡ — Mr. R. B. L. Rawlings describes a simple method for roughly comparing the apertures of objectives, to be used when the Abbe apertometer is not available. As in the Abbe instrument,§ a 3 in. auxiliary objective is attached to the under part of the draw-tube; but in place of the bevelled semicircular glass plate, a sub-stage condenser and an iris diaphragm are used. The objective to be examined is first focused on the upper surface of the condenser, and then, without disturbing this adjustment, the auxiliary objective is screwed to the draw-tube, which is slid into such a position that the diaphragm is clearly seen; when the margin of the diaphragm is brought to the edge of the field of view, the diameter of the opening is in direct ratio to the aperture of the objective. Examining another objective in the same way, the apertures of the two are proportional to the diameters of the diaphragm openings in the two cases. It is suggested that a direct reading of the aperture could be made by combining the iris diaphragm with a graduated arc and index pointer; this would give a measure of the diameter of the opening, and so a measure of the aperture of the objective.

(3) Illuminating and other Apparatus.

Monochromatic Light Apparatus.‖—Mr. A. E. Tutton describes an instrument of precision for producing monochromatic light of any desired wave-length, and explains its use in the investigation of the optical properties of crystals. It consists essentially of a spectroscope, with one large 60° prism, in which the eye-piece of the telescope is replaced by a fixed slit. Light of different colours is caused to pass through this slit by rotating the dispersing apparatus, the position of which can be read off on the graduated circle, and this, after the instrument has been empirically graduated, will give an indication of 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.

† Zeitschr. f. ang. Mikr., ii. (1897) pp. 321-35 (2 figs.).

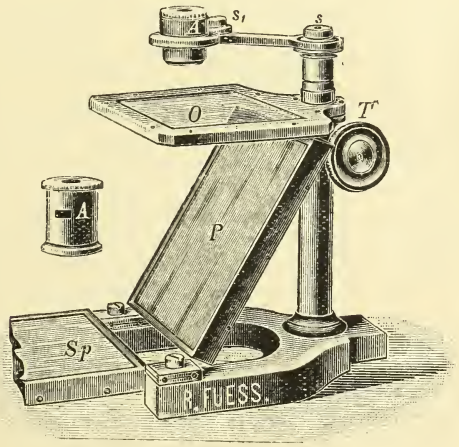
‡ Amer. Mon. Micr. Journ., xviii. (1897) pp. 3-6; and English Mechanic, lxv. (1897) pp. 57-8. § This Journal, 1878, p. 19; 1880, p. 20; 1896, p. 247.

‖ Phil. Trans., clxxxv. (1894) A, pp. 913-41; and Zeits. f. Krystall., xxiv. (1895) pp. 455-74 (7 figs.).

wave-length of the light passing through the slit. The light, after being diffused by a plate of finely ground glass, passes directly into the observing instrument—axial angle apparatus, goniometer or Microscope, &c. As a strong light is necessary, a lime-light lantern is used.

Lens-Support, with Polarising Apparatus.*—Dr. C. Leiss describes an instrument for examining large mineral, rock and palæontological sections in polarised light, which is also useful for examining crystal groups, separating minerals, &c.; it is made by Fuess after the designs of E. Kalkowsky. To the foot are hinged an illuminating mirror *Sp*, and a glass plate P, which acts as the polariser. The glass plate O,

Fig. 8.



which may be replaced by metal plates having suitable openings, carries the object. The lens can be moved about in a horizontal plane by means of an arm jointed at s_1 and s, and the rack and pinion T gives an extended range of vertical motion. The analysing nicol, a Glan-Thompson prism, fits over the lens, and a slot is provided for inserting a mica or gypsum plate. Two Steinheil lenses, giving a large flat field and magnifying four and eight times, are supplied with the instrument.

Thermostat heated by Mineral Oil for Paraffin Imbedding.†—Herr W. Karawaiew describes a thermostat devised by him, which is heated by petroleum or benzoline, and regulated automatically by electrical contact. When the temperature for which the apparatus is adjusted ascends above the desired degree, the mercury column of an air thermometer inserted in the interior of the thermostat rises until it comes into contact with a platinum point, thereby making a current which acts on an electro-magnet. The power of the magnet is exerted on a movable metallic plate, which is inserted between the source of heat and the bottom of the thermostat. In this way direct heating is prevented until,

* Neues Jahrb. Mineral., i. (1897) pp. 81–2; and Zeitschr. ang. Mikr., ii. (1897) pp. 289–90. † Zeitschr. f. wiss. Mikr., xiii. (1896) pp. 289–99 (3 figs.).

by the sinking of the thermometer, contact is again dissolved. The apparatus is of small size (about 17 cm. square) and made of copper. It is stated not to vary more than $\frac{1}{4}^{\circ}$ during the 12 hours.

(4) Photomicrography.

On a Simple Method of Photomicrography by an Inexpensive Apparatus.*—Except for bodies of inappreciable thickness, photomicrography will never be able to compete with accurate drawings made by the aid of the camera lucida.

As the finger plays on the focusing-screw the eye is capable of fixing its attention on the portions of the image in sharp focus to the exclusion of those that are outside the focal plane, but no mere optical instrument is capable of doing this, and the result is that, where a body is of any thickness, the distinctness of the photographic image of the plane actually in focus is blurred, and marred by the hazy images of planes outside or within that plane.

Something of the same kind is seen in ordinary landscape photography when lenses of long focus are employed; either the foreground is blurred and the background sharp, or *vice versâ*. Now, as the eye is accustomed to at once focus each object, whether near or distant, as it plays over a landscape, and cannot do this as it glances over the photograph, the result is unsatisfactory and unnatural. For this reason an enlargement from a view taken with a short focus lens, albeit it has special faults of its own, is often more satisfactory.

If this be so patent in the ordinary photography of opaque objects, how much more unsatisfactory will be the result when, owing to the transparency of the objects, images of different degrees of sharpness are not merely juxta- but super-posed. Nevertheless, although for most objects photomicrographs can never equal good drawings, especially for purposes of demonstration, the method presents great advantages on account of its facility and quickness, and is of special value in meeting the objections of that pestilential person, the sceptical negative observer. The man who, because he can find no free "plasmodia" in cases of Indian fever, refuses to believe that Laveran ever saw such bodies in Algeria, will be more convinced by a single photomicrograph than by a whole atlas of drawings.

These latter may, or may not, be representations of the numerous fallacious appearances with which one becomes quickly familiar after working for a while at the examination of blood under high magnifications; but as a photograph must be a correct representation of some one aspect of the body, i.e. of the combination of the images of planes in and out of focus, its identity or otherwise with any known fallacy can be established in a way which is quite out of the question in the case of drawings.

To be really useful, a photomicrographic apparatus should be so simple that it can be applied at once to the delineation of any object that may chance to be in the field of the Microscope; and the difficulty of attaining this lies in the fact that ordinary illumination, such as is

* By G. M. Giles, M.B., F.R.C.S., F.R.M.S., Surgeon I.M.S. Read February 17th, 1897.

most convenient for ordinary observation, is far too feeble to be visible on the focusing-screen of the camera. In the early days of the art, when gelatin-bromide plates were yet unknown, direct sunlight was the only practicable illumination for anything but the most moderate amplifications.

To secure this a heliostat was indispensable, and it is now more than twenty years ago since the writer described in the Monthly Journal of this Society a plan in which this necessity was overcome, by employing a condenser of such long focus that the image of the sun was vivid enough to give one time to insert one's dark slide and expose before it had passed across the object.

The wet collodion-plate, if not used quickly, was utterly spoiled. With the dry plate, on the other hand, apart from its greater sensitiveness, length of exposure is a matter of no moment. The difficulty, however, of obtaining adequate light to sufficiently illuminate the ground glass remains, and the method I am about to describe overcomes this by doing away with the focusing-screen altogether.

No special camera is required. Almost any quarter-plate or 5 × 4-in. landscape camera will serve for the purpose; but it is most convenient to select one with a conical bellows, in which the front is completely detachable from the standards that attach it to the base-frame when set up for ordinary work. The one I am using is an Adam's "Victor" camera. The movable front carrying the lens is only 2 in. square, and a spare one, to receive the collar that is to connect camera and Microscope, is of course necessary.

Take a piece of black velvet about 2 in. wide, and have it sewn round the draw-tube of the Microscope, not too tightly. Then take a long strip of brown paper 1 in. wide dipped in paste or gum, and wind it round the middle of the velvet till it is about 1/16 in. thick. When this has dried thoroughly, fold down the projecting part of the velvet over the outside of the pasteboard tube we have thus made, and sew the edges together so as to neatly cover the outside of the tube. Nothing more is required but to cut a hole in the spare front just large enough to tightly hold the velvet-covered pasteboard tube. If the hole be made the right size, the joint will be quite light-tight without any packing or cement of any sort.

The Microscope is always used in the vertical position, and the camera is supported above it by means of a telescopic upright jointed vertically into a heavy base-board on which the Microscope stands.

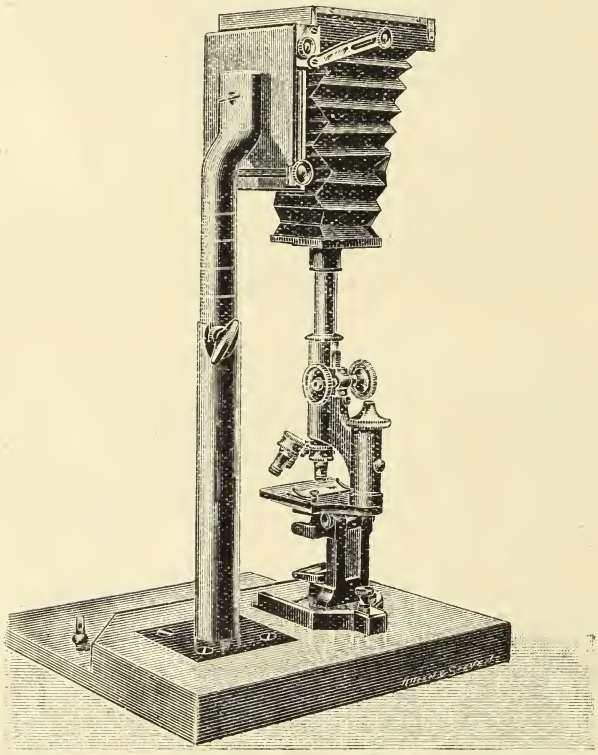
The base-board should be about 1 ft. square, and at least an inch thick, standing on four low studs placed at the corners, or, better still, with studs at three corners, and the fourth stud replaced by a coarse-pitched, blunt-ended screw, so as to allow for inequalities of the surface on which it may be placed, as it is important that the apparatus be as little liable to vibration as may be. The upright consists of two stout drawn metal tubes, sliding one within the other, of such lengths that the inner tube can be fixed, by means of a clamping-screw, at any length between 15 and 20 in. The inner or sliding tube ends in a square metal plate about 2½ in. square, with a central hole large enough to take the screw which ordinarily fixes the camera to its tripod. This plate must project a little clear of the surface of the tube, so that the base-board

of the camera may clear the outer tube when it is being lowered. The upright is fixed nearly in the middle of the base-board, and the inner or sliding tube should be graduated to inches and tenths or millimetres, as may be preferred.

Any good Microscope-stand will serve, but it is essential that the fine-adjustment should have a graduated head, and a revolving stage is a great convenience.

The above dimensions are calculated for the largest but one of Zeiss' stands. I have not a catalogue by me, but it is, I believe, deno-

Fig. 9.



Camera attached to Microscope.

minated IIa. In this stand the milled head of the fine-adjustment is divided into 50 parts, marked from 2 to 100. If the Microscope which it is desired to use be not provided with a graduated fine-adjustment, a special one must be fitted, which can easily be done by turning up a flat button of hard wood, with a cavity below just large enough to slip tightly over the milled head, and with the upper surface flat, on which can be gummed a paper disc graduated to 2° or 4°, as may be convenient.

The provision of an index carried on some part of the arm of the

Microscope will not greatly tax ingenuity, but its position will necessarily vary with the type of Microscope employed.

When it is desired to use the apparatus, it is placed in front of an open window, and the Microscope is placed on the base-board at such a distance from the vertical pillar that the axis of the camera, when placed in position, may coincide with that of the Microscope. Having found this position, make a pencil outline of the foot of the Microscope on the

Fig. 10.

Camera swung aside to admit of adjustment of light, focusing, &c.

base-board, so that it can be, in future, removed and replaced without further trouble.

The camera is now screwed on to the plate of the upright, and adjusted so that its focusing-screen is accurately parallel with the base-board and the stage of the Microscope; but it is not, at this stage of the proceedings, placed over the Microscope, but turned aside out of the way, by twisting the inner tube of the standard within the outer one,

so that one is able to examine an object under the Microscope as easily as if it were standing on the table, quite clear of any photographic attachment.

The pasteboard tube carrying the camera front is, however, fitted on to the draw-tube, as, owing to its small size, it does not interfere with the use of the Microscope any more than a micrometer eye-piece or other ocular attachment.

The easiest way to indicate the principles of the method I wish to advocate, will be to describe the method in a specific case.

Say it is desired to obtain a photograph of an object under Zeiss' C with oc. 3, the draw-tube at its full length.

The object is placed in position, accurately focused, and lighted to the best advantage by means of the concave mirror. If great exactitude be desired, exact parallelism may be secured by levelling the stage of the Microscope and the focusing-screen with a spirit-level. The camera, with the bellows hanging loose and unattached, is now swung round, so that it is suspended over the Microscope, and fixed at such a height that the length from the eye-piece to the focusing-screen may be about 8 in. Its front frame is now gently fitted on to the movable front already attached to the Microscope.

If the ground glass be now examined under a focusing-cloth it will be found that there is too little light on the plate to enable one even to see the position of the object, far less whether or no it is in focus; but, as a matter of fact, it will be nearly so—though, in all probability, the error, combined with the difference between actinic and visual foci, will necessitate the lens being focused a little away from the object to obtain a sharp picture.

Suppose now we focus the lens out four-hundredths, i.e. two divisions of the Zeiss milled head, and, having inserted an Ilford ordinary plate in the camera, proceed to expose for about one minute.

On developing the plate, if the lenses used at all correspond to those I am using, a very fairly sharp picture will result.

Now examine the plate closely for any portion of the picture that may be in sharp focus, and, having noted this, detach the camera front and swing the camera out of the way, and note exactly the number of divisions through which the milled head must revolve to bring this into sharp visual focus as one looks through the Microscope.

A few experiments may be necessary before this correction is accurately obtained; but, once it has been ascertained for any given combination of objective, ocular, and length of camera, sharply focused photographs may be obtained with far greater certainty than in the ordinary way, because it is far easier to focus sharply an object observed in the ordinary manner through the Microscope, than when the image is dulled by being examined through a ground-glass surface.

The weaker the objective, the larger will be the correction necessary; for instance, with draw-tube fully out, oc. 3, and a camera-length of 8 in., I find that I have for objective A to focus out 25 mm., for B 10 mm., for C 4 mm., and for D 2·5 mm.

The exposure, of course, must be increased as the square of the linear amplification or, given that one minute suffice for C, about 3

minutes will be required if D be substituted for it, which magnifies not quite twice as much. B, on the other hand, will require but 15 seconds, and A only 4 or 5.

The length of exposure will, of course, vary greatly with the quality of the light obtainable, but, as long as one has sufficient to see the object for ordinary purposes of microscopic observation, it is merely a question of so many minutes or seconds exposure more or less, and it is as easy to get a good photograph in an English November as in the brightest day of summer.

The correction necessary for each combination requires, of course, a few careful experiments; but this effected, is done once for always; and, as each experiment helps greatly towards the next combination, one soon obtains a table of corrections for all the combinations one is likely to require.

The photomicrographic expert will doubtless object that it is better to dispense with the eye-piece,* and employ the image given by the objective directly; but though this may be so in the case of difficult objects, such as diatoms under high amplification, it necessitates the use of a camera of unwieldy length, and is, moreover, an almost hopeless task, unless the tube of the Microscope be done away with altogether, and the objective be made to screw directly on to the camera-front; for, however carefully the tube may be blacked, the reflection from its sides produces an unbearable glare in the centre of the plate, and, in the case of a Microscope in regular use, the portion in contact with the eye-piece becomes so polished, that little else but central glare is visible on a negative given by the objective alone. For the purposes, therefore, of the working biologist, it is far better to use the eye-piece, in spite of the small theoretical disadvantages of the plan.

If the camera-length beyond the eye-piece be about 8 in., the scale of the resulting negative will be somewhere about the nominal magnifying-power of the combination in the case of Continental outfits. English opticians, however, calculate their amplifications at 10 in. from the eye-piece; so that, in the case of English Microscopes, the amplification will be a good deal less than the nominal power of the combination.

I cannot, however, recommend working with any greater extension, as very few lenses will bear the test.

The plates used should be the slowest obtainable. The more rapid the plate, the coarser is the grain of the film, and therefore the less suited it is for our purpose. In this respect, gelatin plates compare ill with the old wet collodion, the grain of which was so fine that a sheet of the 'Times' reduced to a square about half an inch square could still be easily read under a sufficiently powerful lens.

Collodion emulsions are, however, coming a great deal into use once more for a variety of purposes; and, though slow, would be doubtless to be preferred for photomicrographical work.

A writer in the current 'Photographic Almanac' praises highly the Hill-Norris collodion-plate (medium speed) for photomicrography; but

* [The general practice of the best photomicrographers in this country is to use an eye-piece.]

as I can find no reference to these plates in the advertisement-sheets of the Almanac, I have not been able to put them to practical trial.

To shut off the light preparatory to exposing, I employ simply a piece of black velvet gummed on to a visiting card, slipped on top of the ring that carries the Abbe condenser; but it would be undoubtedly better to use a flap-shutter working inside the camera just behind the eye-piece; as under all but the highest powers the object is more or less visible as an opaque object, after the light has been cut off from below; and though the amount of light so thrown is too small to have much effect during the short time that elapses between drawing the shutter of the dark slide and exposing, it must have a certain more or less fogging effect, and should therefore be avoided by those who have sufficient micro work to set aside a camera specially for the purpose.

As will be seen, the apparatus described need cost but little. Simple as it is, it may be further simplified by using a simple wooden upright, mortised into the base-board, to carry the camera. One misses the great convenience of being able to instantly swing the camera out of the way when changing objects and focusing; but, apart from the delay involved in having to unscrew and replace the camera on such occasions, such an appliance is quite as efficient as that first described, and reduces the cost of the appliance required to connect one's camera and Microscope to a few pence.

Many biologists are unhandy with their pencils, and, looking upon "micro" work as a particularly recondite branch of photography, are debarred from employing this means of illustrating their observations. If a few such can be induced to discover how simple a matter photographic recording can be made, I believe the space I have occupied in the Journal will not be wasted.

Photomicrographs.*—Dr. J. Eismond discusses the pros and cons in regard to photomicrographs. He suggests a compromise between them and drawings. A faint copy of the negative is taken on platinum paper or the like; and this is touched up with ink, pencil, or colours, so as to differentiate any particular structure.

A Simple Arrangement for taking Slightly Enlarged Stereoscopic Photographs.†—Dr. W. Gebhart points out that with an ordinary stereoscopic camera, in which the two objectives are fixed at only a small distance apart, the object cannot be brought close enough to the camera to produce an enlarged picture; this could be done by using two cameras having their axes converging to the object, or by moving one camera into two positions about a vertical axis passing through the object. In the majority of cases (except, for example, with a polished sphere) the same result would be obtained if the camera remained fixed and the object be turned through a small angle. For this purpose the object is placed at the centre of a wooden disc, which can be turned about a vertical axis, through the required angle, this angle, of 6–15°, depending on the distance of the camera, being indicated by graduations on the disc and an index-pointer.

* Biol. Centralbl., xvi. (1896) pp. 864–5.
† Zeitschr. f. wiss. Mikr., xiii. (1896) pp. 419–23 (1 fig.).

β. Technique.*

(1) Collecting Objects, including Culture Processes.

Practical Method for Preparing Agar for Cultivation Purposes.†—100 grm. of agar are first washed with cold water and then placed in a kettle containing 50 litres of boiling water and 200 grm. of Carraghen powder previously rubbed up with cold water. The boiling is continued until all the agar is dissolved, after which it is allowed to cool down to 50°, when ten whole hen's eggs, previously well beaten up, are added. The fluid is boiled for a further 5–10 minutes and then strained through linen. One per cent. of glycerin is added to the agar mass, which, after distribution into five litre flasks, is sterilised. When required for use a flask is liquefied in a steamer and the hot fluid poured through a thick layer of cotton-wool. The filtrate after subsequent sterilisation should be clear. If, however, the glass vessel be of inferior quality it may render the medium turbid owing to giving up alkali. On this medium, even without addition of pepton and nutritive substances, most fission and yeast fungi will grow.

Cultivation of Diphtheria Bacilli on Non-Albuminous Media.‡—Herr N. Uschinsky states that he has succeeded in obtaining cultivations of diphtheria in his medium.§ The toxin therefrom was copious and strong, 1·5 ccm. of a 4–6 weeks old culture killing a guinea-pig in 36–40 hours. The previous non-success was due to not recognising that young cultures are unsuitable, while old ones grow easily in non-albuminous media. The appearance of the cultures in Uschinsky's fluid resembles those in bouillon. The filtrate gives a distinct albuminous reaction, though, of course, it does not follow that this is due to the toxin.

Culture of Saprolegniaceæ.‖—Dr. A. Maurizio recommends pollen-grains (of a great variety of plants) as a favourable medium for the cultivation of Fungi belonging to the genera *Achlya* and *Saprolegnia*.

Growth of Diphtheria Bacilli in Milk.¶—Prof. M. Schottelius shows that for diphtheria bacilli raw warm cow's milk is an extremely favourable growth-medium, as compared with sterilised milk or alkaline bouillon. The figures, which speak for themselves, are, at room temperature, raw milk 21, sterilised milk 2, bouillon 7; at incubation temperature, raw milk 50, sterilised milk 6, bouillon 18.

Keeping Potatoes for Culture Purposes.**—Dr. W. Simmonds states that potatoes may be kept for quite a long time without getting mouldy or dry by the following method, which he has practised for a year and

* 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.

† Marpmann's Bakt. Chem. Laborat. in Leipzig; Zeitschr. f. ang. Mikr., ii. (1896) p. 237. ‡ Centralbl. f. Bakteriol. u. Parasitenk., xxi. (1897) pp. 146–7.

§ See this Journal, 1893, p. 796.

‖ Arch. Sci. Phys. et Nat., ci. (1896) pp. 599–601. Cf. this Journal, 1896, p. 446.

¶ Centralbl. f. Bakteriol. u. Parasitenk., 1te Abt., xx. (1896) pp. 897–900.

** Op. cit., xxi. (1897) p. 100–1.

a half. After having been cleaned and boiled in the usual way, the potatoes are, when cold, bound round with twine and suspended close together. They are then immersed in shellac solution thrice at intervals of half an hour. In the course of another hour they are quite dry, and may be stored away for future use.

Examining Rectal Mucus for Tubercle Bacilli.*—According to Dr. Sawyer, it is useful for diagnostic purposes to examine the mucus from the rectum if there be no sputum, or if tubercle bacilli cannot be demonstrated in the pulmonary excreta. The author quotes three cases in which he found tubercle bacilli in considerable numbers in the rectal mucus, but none elsewhere.

Simple Method for the Sero-Diagnosis of Enteric Fever.†—Prof. E. Pfuhl takes a drop of blood from the ear and mixes it in the hollow of a slide with ten times the quantity of water. This not only dilutes the blood, but gets rid of the red corpuscles. To the serum is then added an equal quantity of a bouillon culture of typhoid bacilli. This is done by dabbing the culture on a cover-glass and inverting it over the serum in the ground-out slide.

Improvement in the Sedgwick-Rafter Method for the Microscopical Examination of Drinking Water.‡—The Sedgwick-Rafter method, now extensively employed in America for the analysis of drinking waters, is, says Mr. D. D. Jackson, as follows:—A definite quantity of water, usually 250 ccm., is filtered through Berkshire sand placed in a funnel. The size of the grains is such that while they will pass through a sieve of 60 meshes to the inch, they will not through one having 120 to the inch. The organisms adhere to the sand, while the water passes through a hole at the bottom of the funnel closed by fine bolting cloth. The sand is dropped into a test-tube containing 5 ccm. of sterile water. The tube is then shaken and the water decanted with another test-tube. The microorganisms are distributed by blowing into the water with a pipette, and 1 ccm. removed to a cell 50 mm. long by 20 mm. wide and 1 mm. deep. The Microscope is graduated so that each field examines one cubic millimetre of water. The improvement made by the author consists in altering the shape of the funnel, which has a diameter of 2 in., and a length to the beginning of the slope of 9 in. The length of the slope is 3 in. The leg of the funnel is $2\frac{1}{2}$ in. long, and its internal diameter 1/2 in. The lower end is closed by a rubber plug, perforated by a small hole, and the latter covered with fine bolting cloth. Above the plug is a layer of fine sand about 3/4 in. thick.

The most important errors to which this method is liable, says Mr. G. C. Whipple,§ arise from the concentration of the sample. These are (1) the funnel error, or that caused by the adhesion of the organisms to the sides of the funnel; (2) the sand error, or that caused by the organisms passing through the sand; (3) the decantation error, or that caused by the organisms adhering to the particles of sand, and by the water used in washing the sand being held back by capillarity during decan-

* Med. News, May 23, 1896. See Centralbl. f. Bakteriol. u. Parasitenk., 1te Abt., xxi. (1897) p. 71.

† Centralbl. f. Bakteriol. u. Parasitenk., xxi. (1897) pp. 52–7.

‡ Technol. Quarterly, ix. (1896) pp. 271–4 (1 pl.). § Tom. cit., pp. 275–9.

tation. The decantation error is minimised by the shape of the funnel devised by Mr. Jackson. The amount of sand error depends on the character of the organisms, upon the size of the sand-grains, and the depth of the sand. The decantation error chiefly depends on manipulation, and arises from some of the organisms remaining attached to the sand-grains.

(2) Preparing Objects.

Isolation of the Elements of the Crystalline Lens.* — Dr. W. Gebhardt places the eyeball in a 4 to 10 per cent. solution of formalin for one or two days. All the transparent parts retain their transparency, and the vitreous its muco-gelatinous consistence. The bulb, which may be preserved in the formalin solution, is then transferred to 50–60 per cent. alcohol for a couple of hours. The lens is then taken up and gently squeezed between two fingers. By slight pressure it is broken up into separate lamellæ, which can be easily teased out in water or glycerin. Both the toothed and smooth fibres are easily isolated, the teeth being extremely clear. The preparation may be stained, and mounted in glycerin-gelatin.

Method of Preparing Rotifers.†—M. N. de Zograf has a note on this subject, in which he states that, after trying Mr. C. F. Rousselet's method of preserving rotifers, he found that formalin preserved the animals for a very short time only. To this it must be remarked that, while M. de Zograf's experience cannot have extended for more than twelve months, in this country the rotifers mounted by Mr. Rousselet in formalin for the last three years have kept extremely well, and look at present as if they were going to keep a great number of years more. The only difficulty experienced has been to prevent the evaporation of the watery fluid, which has now apparently been overcome by using thickened gold size and Bell's cement for sealing the slides.

M. de Zograf mentions a method by which he has succeeded in mounting rotifers in balsam. After narcotisation with cocain, killing, and fixing with ·25 per cent. osmic acid, a rather large quantity of a weak (10 per cent.) solution of raw wood vinegar is added, and the animals left therein from five to ten minutes; they are then washed in three changes of distilled water, which is gradually replaced by alcohol of progressive strengths, finishing with absolute alcohol. In this way the rotifers do not shrink, and can be passed into glycerin or Canada balsam in the usual way. The protoplasm and organs are coloured by this method a bluish-grey or deep black, but the histological structure is well shown.

(3) Cutting, including Imbedding and Microtomes.

New Microtomes by Fromme.‡—Prof. J. Schaffer described two new microtomes, made by Fromme Brothers, of Vienna, for cutting large sections:—

1. *For Paraffin-Imbeddings.*—The object is carried at the side of a rectangular frame, which is fixed to a horizontal axis working in bearings

* Zeitschr. f. wiss. Mikr., xiii. (1896) pp. 306–7.
† Comptes Rendus, cxxiv. (1897) pp. 245–6.
‡ Zeitschr. f. wiss. Mikr., xiii. (1896) pp. 1–9 (3 figs.).

in the heavy foot of the instrument; the knife is fixed to the foot in a vertical position. The small side-movement of the object is effected by a micrometer screw and toothed wheel carried within the rectangular frame.

2. *For Celloidin-Imbeddings and for Cutting in Liquids.*—Here the knife is carried horizontally on a rectangular frame, which, in this instrument, works about a vertical axis; the object is fixed to the foot of the instrument, and the small movement is given to the knife by the micrometer arrangement within the rectangular frame. For cutting in liquids, a small bath is arranged round the object-holder, and the knife is carried on an arm bent twice at right angles and projecting over the edge of the bath.

Manipulation of Celloidin Sections.*—By the following procedure, devised by Dr. G. Aubertin, the chief inconveniences inherent in the celloidin-section may be avoided. The sections, cut in 70 per cent. alcohol, are arranged on the slide. The celloidin is then dehydrated, first in 70 per cent., and finally in absolute alcohol. When perfectly dehydrated, the celloidin is dissolved by dropping on very carefully a mixture of ether and alcohol in equal parts, with which the whole surface of the slide should be just covered. The ether-alcohol mixture may be renewed until the whole slide is covered with a very thin layer of celloidin. The ether-alcohol is allowed to evaporate, but not to complete dryness of the celloidin. The membrane thus formed is not only firm, but so delicate that staining is easily effected. The after-treatment suggested is—70 per cent. alcohol, water (20 minutes), dilute solution of borax-carmine (some hours), water (10 minutes), hæmatoxylin (10 minutes), hydrochloric acid-alcohol until the celloidin is decolorised. The preparations should be dehydrated in 95 per cent. spirit, cleared up in xylol-carbolic acid (3–1), and mounted in balsam.

(4) Staining and Injecting.

Investigation of Brain of Fishes.†—M. Catois has made use of a process which he describes as a combination of the injection-methods of Ehrlich and Meyer with the immersion-method of Dogiel, and the diffusion-process of Cajal.

Into the living animal there were injected 1 to 2 ccm. of concentrated salt solution of methylen-blue; on the brain being removed, sections were made of it, and the pieces placed for about half an hour in a saturated solution of methylen-blue, after which they were treated in the ordinary way.

Preparation of Embryonic Nervous System of Crustacea.‡—M. N. de Zograf has made use of Prof. Ramon y Cajal's method of double impregnation; he reports that he had many difficulties to overcome before succeeding with the larvæ of Copepoda. He could not cover the Nauplius with a layer of glycerin and gelatin to preserve it from the silver precipitate, as is often done with minute objects, for this reagent dehydrates the larvæ, and makes it impossible to recognise their struc-

* Anat. Anzeig., xiii. (1897) pp. 90–3.
† Comptes Rendus, cxxiv. (1897) pp. 204–5. ‡ Tom. cit., p. 202.

ture. He succeeded, however, by enveloping the Nauplii in pieces of cigarette paper.

Rapid Method of Fixing and Staining Blood-Films.*—Dr. G. L. Gulland fixes and stains blood-films in the following way:—The covers are dropped film-side downwards into the fixative, which is composed of absolute alcohol saturated with eosin 25 ccm., ether 25 ccm., sublimate in absolute alcohol (2 grm. to 10 ccm.) 5 drops. After an immersion of three or four minutes, the covers are removed with forceps and washed in water. The film is then stained for just one minute in a saturated aqueous solution of methylen-blue, after which it is washed in water, dehydrated in absolute alcohol, and mounted.

Modification of Heller's Method of Staining Medullated Nerve-Fibres.†—Dr. W. F. Robertson first treats healthy or morbid nervous tissue with Weigert's chrome-alum-copper fluid for ten days or longer. The fluid is composed of 2·5 per cent. chrome-alum, 5 per cent. copper acetate, 5 per cent. acetic acid, and formalin 2 per cent. The chrome-alum is boiled in the required amount of water, and when dissolved the acetic acid and copper acetate are put in. When cold the solution is filtered and the formalin then added. The author has reduced the quantity from 10 to 2 per cent., as too much formalin impairs the staining reaction. Sections of material may be obtained by the celloidin or gum-freezing method, and they are stained by placing them in 1 per cent. osmic acid for half an hour in the dark, then in 5 per cent. pyrogallic acid for half an hour, 0·25 per cent. potassium permanganate for 3–4 minutes, 1 per cent. oxalic acid for 3–5 minutes. Wash in water after treatment with each solution, dehydrate, then mount in balsam.

Staining Coccidium oviforme.‡—Dr. R. Abel stains *Coccidium oviforme* with the Ziehl-Neelsen solution for tubercle bacilli. The parasites may be stained on cover-glasses, or in sections. After staining with hot phenol-fuchsin, the preparations are to be decolorised in 5 per cent. sulphuric acid and 70 per cent. alcohol. Any contrast stain may be used.

Method for Staining Unnucleated Cells.§—Herr J. J. Gerassimoff states that if a *Spirogyra* cell be treated with chloroform, ether, or with chloral hydrate during fission, two daughter-cells will be obtained, one being devoid of nuclear substance, the other containing excess thereof, i.e. there is one large nucleus or two of ordinary size; in fact, the results are exactly the same as those induced by the action of a low temperature. To 100 ccm. of the water containing the algæ were added 0·25–1·5 ccm. of saturated chloral hydrate solution, or 0·42–2·5 ccm. of ether, or 1·25–7·5 ccm. chloroform water. The time required varied from fifteen minutes to some hours, after which the algæ were removed to fresh water.

New Method of Staining Tubercle Bacilli.‖—Drs. A. Rondelli and L. Buscalioni propose the following method for staining tubercle bacilli, which they say is extremely rapid and simple:—The decoloriser is a

* Brit. Med. Journ., 1897, i. p. 652. † Tom. cit., pp. 651–2.
‡ Centralbl. f. Bakteriol. u. Parasitenk., 1te Abt., xx. (1896) pp. 904–5.
§ Moscow, 1896, 4 pp.
‖ See Centralbl. f. Bakteriol. u. Parasitenk, 1te Abt., xxi. (1897) pp. 70–1.

modified eau de Javelle, made by mixing two solutions. The first is composed of calcium hypochlorite 6 grm. in 60 grm. of water. The second is composed of 12 grm. of potassium carbonate dissolved in 40 grm. of water, and after filtration is mixed with the first solution. The combination is stirred up for some time, then filtered and preserved in blue glass bottles. After the film has been prepared and stained in the usual way, the cover-glass is immersed in the Javelle decoloriser until it looks brownish-yellow.

Decoloration of Celloidin in Orcein Preparations.* — It is very difficult, says Prof. P. Schiefferdecker, to decolorise the celloidin when celloidin-sections are stained with orcein to show the elastic fibres. This inconvenience may be avoided by transferring the preparations, after they have been decolorised in hydrochloric acid alcohol, to water containing some liq. ammon. caustici. In this the sections become blue and give up some pigment. As soon as the dye ceases to be given off, the preparations are put back into the hydrochloric acid alcohol. The process is to be repeated until the celloidin is sufficiently decolorised.

Staining Centrosomes.†—Dr. R. Marchesini recommends, for a study of centrosomes and attraction-spheres in the leucocytes of the newt, a thorough mixture of 1 part of malachite-green solution to 2 parts of saffranin-green.

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

Marking Preparations.‡— Prof. P. Schiefferdecker advises that slides should be permanently marked in black or white by writing on the glass and then varnishing the surface. The liquid Chinese ink does well for the black, while Kremser white or permanent Chinese white is recommended for white. The Kremser white should be rubbed up with a sufficient quantity of gum-water. When the writing is dry it should be brushed over with water-colour varnish.

Herr E. Schoebel § points out that the method proposed by Schiefferdecker of writing on glass with Indian ink and then varnishing over, is much the same as that used for many years at the Zoological Station of Naples; here, however, the writing is done over the varnish. Schiefferdecker's objections to the author's method ‖ of writing on glass with a mixture of sodium silicate and fluid Indian ink are answered.

(6) Miscellaneous.

Plate Modelling.¶—Dr. A. Schaper, in preparing his models, uses Born's ** method, but takes special precautions for properly orientating the object, so that sections are made in known and definite directions. The details of the method, as applied to embryos, are described at length.

* Zeitschr. f. wiss. Mikr., xiii. (1896) p. 302.
† Boll. Soc. Rom. Stud. Zool., v. (1896) pp. 89–96 (1 pl.).
‡ Zeitschr. f. wiss. Mikr., xiii. (1896) pp. 299–301.
§ Tom cit., pp. 425–8.
‖ Op. cit., xi. (1894) p. 331. Cf. this Journal, 1895, p. 707.
¶ Zeitschr. f. wiss. Mikr., xiii. (1897) pp. 446–59 (10 figs.).
** This Journal, 1889, p. 144.

Tests for Ligneous Tissue.*—Dr. F. Zetzsche enumerates twenty-eight different methods which have been used and recommended by various experimentalists for the detection of woody tissue. Out of these the following eight are commended:—(1) indol with hydrochloric acid; (2) phloroglucin-hydrochloric acid; (3) carbazol-sulphuric acid; (4) anilin sulphate; (5) toluidin-diamin-hydrochloric acid; (6) ammoniacal fuchsin; (7) Bismarck-brown-hæmatoxylin; (8) solid-green deltapurpurin. The best results are obtained from the following:—(1) The indol-hydrochloric reaction gives a brick-red to chocolate-brown colour; may be used successfully in from 1/100 to 1/1000 per cent. solution; and has the great merit of allowing the objects to be mounted in glycerin and in balsam. (2) Phloroglucin is used in 2–3 per cent. alcoholic solution, together with an equal volume of dilute hydrochloric acid. The colour is brownish-red. Preparations do not mount well in either glycerin or balsam. (3) A mixture of equal volumes of alcoholic solution of Bismarck-brown and Böhmert's hæmatoxylin has the special merit of not requiring differentiation, but care must be taken not to overstain. Lignification is shown by the brown staining, the non-ligneous tissue being blue. The preparations should be mounted in glycerin-gelatin. (4) The ammoniacal fuchsin solution is made by freely diluting a saturated alcoholic solution of fuchsin with water, and then adding ammonia until the fuchsin just begins to precipitate. The objects are immersed therein for 1/2 minute, and, after having been well washed, are contrast-stained in saturated anilin-blue solution (1 minute). After dehydration in alcohol they are mounted in balsam.

The intensity of lignification is determinable by the use of solutions of phloroglucin-hydrochloric acid, of varying strengths, allowed to act for a definite time. Three minutes is suggested as the time limit, and 1/40 to 2 per cent. for the solution. The latter is made by dissolving the phloroglucin in 1 vol. of 90 per cent. alcohol and 1 vol. of strong hydrochloric acid. The phloroglucin is first dissolved in the alcohol, and the acid afterwards added very gradually in the cold.

Eight degrees of lignification are made by the author from this method, at the top of the list standing *Abies pectinata*, at the bottom, *Musa paradisiaca*.

Demonstrating Presence of Agar.†—According to Herr G. Marpmann, there are many edible algæ in tropical seas. These belong to the group of Florideæ, with red, leaf-like or membranous growth and upright thallus. Among these are *Gracilaria lichenoides* Ag., *Eucheuma spinosum* and *E. gelatinæ* Ag. All these, when boiled with water, produce a thick jelly, which in the dried condition is known as agar-agar. This agar appears to be chemically identical with the alga-mucus of *Gelidium corneum*. The agar jelly and *Gelidium* jelly, dissolved in boiling water, filtered through cotton-wool and precipitated with alcohol, form horny masses when dried, the former being known as gelose, the latter as gelinose.

As pectin, or vegetable jelly, appears to be of the same, or very nearly the same chemical composition as agar, and as fruit jelly is often adulterated with agar and gelatin, it becomes important to have a ready test to distinguish the presence of agar. This may be done micro-

* Zeitschr. f. ang. Mikr., ii. (1896) pp. 225-36. † Tom. cit., pp. 257-61.

scopically owing to the presence of the siliceous envelopes of the algæ having resisted destruction. The mass to be examined should be treated with dilute sulphuric acid and a few crystals of permanganate of potash. In this way a thin fluid is formed which is easily sedimented, the siliceous envelopes of the algæ being found in the deposit.

Disinfection of Books.*—Dr. von Schab records some experiments made for the purpose of testing the value of Pictet's gas-mixture and formaldehyd for disinfecting books from lending libraries. Pictet's mixture consists of equal volumes of sulphurous acid and carbonic acid gases. The test objects used were *Bacillus pyocyaneus*, *B. anthracis*, *Staphylococcus pyogenes aureus*, and tubercular sputum.

Neither disinfectant gave satisfactory results.

Demonstration of Small Quantities of Formaldehyd.†—According to Herr L. Keutmann, a solution of hydrochlorate of morphia in strong sulphuric acid is very convenient for showing the presence of formaldehyd. One decigram of morphia hydrochlorate is dissolved in 1 cem. of strong sulphuric acid. This solution will detect the presence of 1 part of formaldehyd in 5000 or 6000 parts. The solution to be tested is poured upon (but not mixed with) the morphia solution, and within a few minutes assumes a red-violet hue.

* Centralbl. f. Bakteriol. u. Parasitenk., xxi. (1897) pp. 141–6.

† Zeitschr. f. ang. Mikr., ii. (1896) p. 267.

MICROSCOPY.

A. Instruments, Accessories, &c.*

(1) Stands.

Stands and Optical Equipments.†—In an editorial article figures of two stands are reproduced from the latest catalogue (No. 20) of Reichert of Vienna. General remarks are made concerning oculars, and a list is given of the numbers, focal lengths, amplifications, and prices of the oculars of various firms. The apertures, focal lengths, and prices of achromatic, apochromatic, and semiapochromatic objectives, both dry and immersion, are quoted from catalogues and compared.

Fig. 16.

(3) Illuminating and other Apparatus.

Ocular-Dichroiscope.‡—Herr C. Leiss figures a combined eye-piece and dichroiscope (fig. 16), with the aid of which the two colours shown by crystals of microscopic dimensions may be seen side by side, and so directly compared. This is an improvement on the usual method of rotating the polariser or the crystal. A rectangular diaphragm is placed behind the calcite prism K, as in the ordinary dichroiscope.

(4) Photomicrography.

Method of Projecting a Micrometric Scale upon a Microscopic Specimen.—The accompanying figure (fig. 17) illustrates the apparatus contrived by Prof. A. E. Wright for measuring and counting microscopic objects, described on p. 182, and exhibited at the March meeting of the Society.

The window-pane and the projection-scale (which is etched upon a piece of plate glass) are shown in optical section at A and B respectively. The method of suspending the scale, so that it may be at right angles to the beam of light which is thrown upon the microscopic mirror, is shown at C. The scale is allowed to tilt forward until, as in medallions A and B, an equal number of vertical and horizontal divisions appear within the microscopic field. The minified image of the projection-scale, which is superposed upon the microscopic object, is shown in optical section at G. The adjustment of the condenser, which is essential to the superposition of the minified image upon the microscopic specimen, is most easily achieved by first focusing the microscopic objective upon the microscopic specimen, and then, while keeping this plane under observa-

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

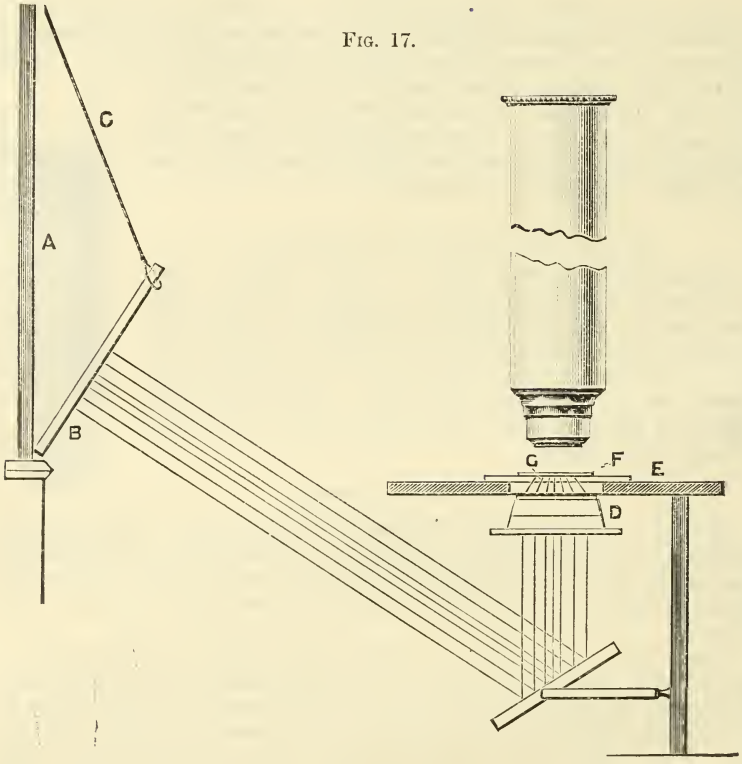
† Zeitschr. f. angew. Mikr., ii. (1897) pp. 351–60 (2 figs.).

‡ Op. cit., iii. (1897) pp. 5–6 (1 fig.).

tion, making the necessary adjustment in the vertical height of the condenser.

Medallion A (fig. 18) shows the figure of squares superposed upon the micrometric ruling of an ordinary stage micrometer, which is ruled in tenths of millimetres. Any line in the projection-scale can be superposed upon any line of the stage micrometer by a mere movement of the mirror round its vertical or horizontal axis. When any one line of the projection-scale has been superposed upon any line of the stage micro-

Fig. 17.



meter, all the other lines can be made to correspond by adjusting the distance between the projection-scale and the Microscope.

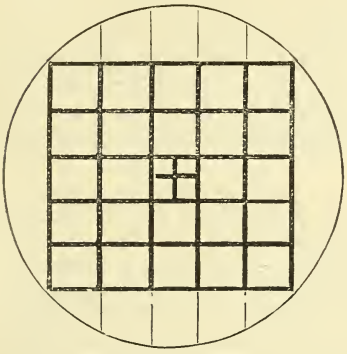
Medallion B (fig. 19) shows the application of the projection-scale * to the enumeration of red and white blood-corpuscles. The appearances shown in the medallion are obtained by substituting for the stage micrometer, shown in medallion A, a specimen of 200-fold diluted blood, which has then been filled into an unruled hæmc-cytometer-cell of a depth of 0·2 mm.

The white blood-corpuscles are most conveniently enumerated by

* Projection-scales of this pattern may be obtained from Mr. A. E. Dean, jun., 73 Hatton Garden, E.C.

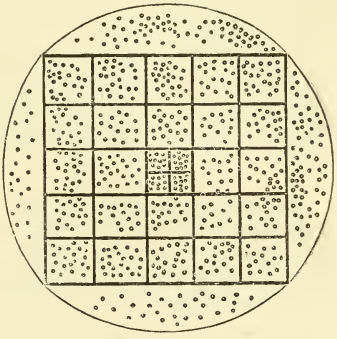
counting the number that fall within the area of the block of 25 squares. The red blood-corpuscles are most conveniently enumerated by counting the number which fall within the central square, which, to facilitate enumeration, is here divided into four smaller squares. The number of red and white blood-corpuscles in one cubic millimetre of blood can be conveniently arrived at by superposing the scale in turn

Fig. 18.



Medallion A.

Fig. 19.



Medallion B.

upon ten different portions of the microscopic specimen, and by then multiplying the total red and white blood-corpuscles which were found in the areas which have just been specified by 10,000 and 400 respectively.

In fig. 17, D represents the substage condenser, E the slide or hæmocytometer-cell, and F the cover-glass.

(5) Microscopical Optics and Manipulation.

Knife and Strop for Microtomes.*—Herr G. Marpmann mentions the advances made in section-cutting since the introduction of the microtome; he figures a knife made during the last 17–18 years by W. Walb of Heidelberg, and mentions a sharpening strop with rounded surface also made by W. Walb.

B. Technique.†

(1) Collecting Objects, including Culture Processes.

Nutritive Medium for Algæ.—In his important work on Reproduction in Algæ and Fungi,‡ Dr. Klebs recommends, for the culture of Algæ, the employment of both fluid and solid media. As a fluid medium he finds Knop's the best, viz.:—4 parts calcium nitrate, 1 part magnesium sulphate, 1 part potassium nitrate, 1 part potassium phosphate.

* Zeitschr. f. angew. Mikr., iii. (1897) p. 6 (1 fig.).

† 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.

‡ *Vide ante*, p. 147.

In preparing it, a concentrated solution (B) may be made of the last three salts, and another (A) of the first. A proper amount of A is to be added to B after dilution to the desired percentage. By this method only a small part of the insoluble calcium phosphate will be precipitated. Solutions containing 0·2 to 0·5 per cent. of salts were found most useful.

Cultivating the Bacillus of Seborrhœa.*—M. Sabouraud has overcome the difficulty of isolating the bacillus of *seborrhœa* and of *alopecia areata* by cultivating in a very acid medium, the ingredients of which are as follows:—Pepton, 20 grm.; glycerin, 20 grm.; acetic acid, 5 drops; water, 1000 grm.; gelose, 13 grm. The temperature used was 35° C. A white coccus was disposed of by using immunised gelose; i.e. by preparing the gelose with fluid in which the coccus had been cultivated. The same result was obtained by heating the cultures to 65° C. for 10 minutes.

Preparing Plague-Serum.†—Prof. A. Lustig and Dr. G. Galeotti prepare a vaccine from the plague bacillus which has the chemical characters of a nucleo-proteid. The bacillus, the virulence of which has been ascertained, is cultivated on large agar plates for three days at 37°. The surface is then scraped, and dissolved in 1 per cent. KHO. After filtration through paper, the vaccine is obtained by precipitation with acetic or hydrochloric acid, or by saturation with sulphate of ammonium after neutralisation. The precipitate, having been repeatedly washed, and dissolved in a very weak solution of sodium carbonate, is ready for use. Transit through the Chamberland filter deprives the vaccine of much of its activity. The minimum lethal dose of the acid precipitate is 5·28 mgrm. per 100 grm. weight of animal. Animals vaccinated with very small, or with one-half or one-third of the smallest fatal dose, injected subcutaneously at intervals of two days, are rendered quite indifferent to large injections of virulent cultures. The immunity lasts about five weeks. From animals thus rendered immune, is obtained, after 14 days, a preventive and curative serum, of which 1 ccm. suffices to prevent peritoneal infection and to cure a rat weighing 180–200 grm., which had been peritoneally injected with four to five loopfuls of virulent culture.

The authors are endeavouring to obtain a prophylactic and antidotal serum from the horse.

New Method of obtaining Diphtheria Antitoxin.‡—Dr. Smirnow, of St. Petersburg, has succeeded in obtaining a diphtheria antitoxin, which is stated to be of considerable effective value, by electrolysing virulent diphtheria broth cultures. In itself the antitoxin appears to be quite harmless, and its preparation simple and rapid.

Technique of Serum Diagnosis.§—Dr. A. S. Delepine, after alluding to the methods used since the beginning of 1896 for demonstrating the action of blood or blood-serum on the corresponding microbes, states that he has finally adopted the following simple but effective procedure, for

* Brit. Med. Journ., 1897, i. p. 1029. † Tom. cit., pp. 1027–8.
‡ Arch. Sci. Biol. Petersburg, iv. (1896) No. 5. See Nature, April 22, 1897, pp. 597–8. § Brit. Med. Journ., 1897, i. pp. 967–70.

which all the apparatus required consists of (*a*) a sterilised lancet-shaped needle; (*b*) the small pipette in which the blood has been collected, and of such diameter that after it has been broken across (the platinum loop, which is used for measuring the serum, can easily be introduced into it if necessary); (*c*) a platinum loop, measuring about 1 mm. in diameter, and holding about 1 mgrm. of fluid; (*d*) slide and cover-glass; (*e*) a tube-culture of the typhoid bacillus in neutral bouillon. The culture should not be more than 24 hours old, should be free from clumps, and the bacillus actively motile. The procedure is as follows:—With the sterilised loop nine drops of the culture are deposited separately on slide or cover-glass. One drop of blood is then added, and the 10 drops thoroughly mixed together. The phenomena observed differ according to whether the serum is potent or not. If the serum be potent, all the bacilli will be agglomerated in from five to thirty minutes; if feeble, the clumps form gradually, but positive diagnosis can be made in from about a half to two hours. For further information and details the original should be consulted.

(2) Preparing Objects.

Preparing and Staining Celery for Demonstrating Bacteria.*—Dr. U. Brizi hardened the diseased parts for 48 hours in a liquid composed of 100 parts of water, to which were added 1 part of glacial acetic acid and 1 part of chromic acid. The pieces were further hardened in 75 per cent., and then in absolute alcohol. The sections were cut by the paraffin method, and after the paraffin had been removed by means of chloroform, were washed in warm water and then immersed in an aqueous 1 per cent. solution of methyl-green for three or four hours, after which they were treated with water acidulated with hydrochloric acid. In this way everything but the bacteria was decolorised, and then the sections were contrast-stained in an aqueous solution of picrocarmin, in which they were allowed to remain for about an hour. The sections, having been washed and dehydrated, were mounted in balsam. Another good stain was gentian-violet and acetic acid (water 100, acetic acid 10, saturated alcoholic solution of gentian-violet 20). The sections were treated with this solution for about an hour, and then placed in strong spirit to which a few drops of hypochlorite of soda were added. By this procedure the tissue was quite decolorised, the bacteria being stained violet.

Microchemical Methods for Examining Cells.†—According to Prof. E. Zacharias, a mixture of methylen-blue and fuchsin S may be used with great advantage to study the distribution of nuclein in the cell. If tissues of different origin are treated with dilute hydrochloric acid, and this mixture afterwards added, the constituents of the cell which contain nuclein are stained a deep blue, the parts without that substance being red. Sperm-cells of the Rhine-salmon were treated with dilute hydrochloric acid to remove protannin, and then stained with the above mixture. Instantly the envelopes of the heads which contain the nucleic acid were beautifully stained bright blue; the inner part of the heads seemed to be colourless; the tails were stained red. Similarly treated,

* Atti R. Accad. Lincei, vi. (1897) pp. 229–34.
† Rep. 66th Meeting Brit. Ass., 1896, p. 1022.

the chromatin bodies of all the nuclei which have as yet been examined were stained blue, the rest of the nuclei and the cell-protoplasm red.

Employment of Dead Bacteria in the Serum Diagnosis of Typhoid and Malta Fever.*—Prof. A. E. Wright and Surgeon-Major D. Semple confirm Widal's observation that the agglomeration phenomenon is equally characteristic with dead bacteria, and they further find that it also holds good for *Micrococcus melitensis*. Emulsions of fresh agar cultures were drawn up into small glass capsules, and these exposed to a temperature of 60° C. for five to ten minutes. The dead bacteria capsules were laid aside for three to nine weeks, and then, having been well shaken up, were used for serum diagnosis. Microscopical observation did not reveal any differences in the method in which agglomeration occurred in the living and dead cultures after addition of dilute serums; and the experiments with capillary sero-sedimentation tubes gave even more interesting results.

(3) Cutting, including Imbedding and Microtomes.

Cutting and Mounting of Sections of Cereal Grains.†—Mr. J. D. Hyatt says that, for making satisfactory sections of grains, the main precaution consists in slightly moistening the kernels. Indian corn may be kept moist for 24 hours, wheat four or five hours, rye five or six, barley ten or twelve, and oats not more than one or two hours. For imbedding, paraffin is the best material, as it holds the grain so firmly that it may be cut in any direction. Any section-cutting contrivance will serve, provided the knife be sharp. If the sections be too thin the starch-grains will fall out, and if too thick the gluten cells will be disagreeably opaque. Glycerin-jelly is the best medium for mounting. The sections are best removed from the knife by a camel's hair brush, and are then deposited in water, from which they are transferred to the centre of a horizontally placed slide. Warm glycerin-jelly is then put on, after which a cover-glass, slightly heated over a spirit-lamp, is carefully deposited on the gelatin. The cover-glass must be allowed to settle gradually and by its own weight, no pressure being applied. If the gelatin have become too hard to allow the cover to settle, the cover may be pressed, and then heat applied to the under-surface of the slide.

(4) Staining and Injecting.

Effect of certain Chemical and Physical Agents on the Staining of Sporous and Asporous Bacteria.‡—M. C. X. Hieroclés exposed the following bacteria, *B. mycoides*, *B. subtilis*, drumstick bacillus, a thermophilous species cultivated at 56° C., typhoid and diphtheria bacilli, to the influence of certain agents, to ascertain whether the action of the latter improved or deteriorated the absorption of pigments in solution. Aqueous and anilin-water fuchsin solutions were used. Dry and moist heat increased the stainability of sporogenous bacteria and their resting forms for anilin-water fuchsin. *B. subtilis* and *B. mycoides* stained

* Brit. Med. Journ., 1897, i. pp. 1214–5.

† Journ. New York Micr. Soc., xiii. (1897) pp. 19–24 (1 pl.).

‡ Arch. f. Hygiene, xxviii. p. 163. See Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 416–7.

better with aqueous solutions. Chlorine and bromine water increased the power of anilin-water fuchsin, and somewhat decreased that of aqueous fuchsin. Bromine vapour was detrimental to spores and bacilli; while chlorine gas seemed to make the spores and bacilli of *B. subtilis* and the drumstick bacillus stain more easily. Formalin and iodopotassic iodide solution had no effect; and sunlight diminished the stainability of *subtilis* bacilli and spores. Chlorine gas was detrimental to typhoid bacilli for aqueous fuchsin solution, and bromine vapour destroyed the cell-plasma. The effect of the agents used was to make diphtheria bacilli swell up, and their staining paler.

New Hæmatoxylin-Stain.*—The following process is recommended by Herr M. Raciborski. Leave the preparation for from 2–20 minutes in Delafield's hæmatoxylin; then wash with water, and for 2–5 minutes with iron-alum, then again with water, alcohol, and toluol, and imbed in canada-balsam. This process affords very good results for botanical purposes, and has the advantage of a great saving of time. Secondary staining with saffranin (in anilin water), and washing in 1 per cent. alcoholic acetic acid, affords a good double-stain.

Flagella Staining.†—Mr. D. McCrorie stains flagella with "night-blue," an anilin pigment which shows as well in artificial as in sunlight. The formula used is, 10 ccm. of a concentrated solution of night-blue, 10 ccm. of a 10 per cent. solution of alum, and 10 ccm. of a 10 per cent. solution of tannic acid. The addition of 0·1–0·2 grm. of gallic acid seems to impart additional value, but excellent results are obtainable without it. The method adopted is to dry the film in an incubator for two minutes; then pour on the stain and incubate again for two minutes, or hold the cover for the same time about two feet above a Bunsen burner; wash off the excess of stain, and, after drying in an incubator, mount in balsam.

(6) Miscellaneous.

Method of extemporising a Blowpipe for making Sedimentation Tubes.‡—Prof. A. E. Wright and Surgeon-Major D. Semple use an ordinary spray producer, such for instance as an ether freezing apparatus; the reservoir is filled with methylated spirit. The flame produced in this way is quite hot enough for any ordinary glass-working apparatus. It is quite hot enough to draw out glass tubing into capillary sero-sedimentation tubes. Only two points in connection with the working of the flame required to be attended to, viz. (1) the spirit must be finely divided, i.e. the spray must not be too coarse, otherwise the flame will not be sufficiently hot; (2) the spirit must be fed into the spray tube in sufficient quantity and in a regular manner.

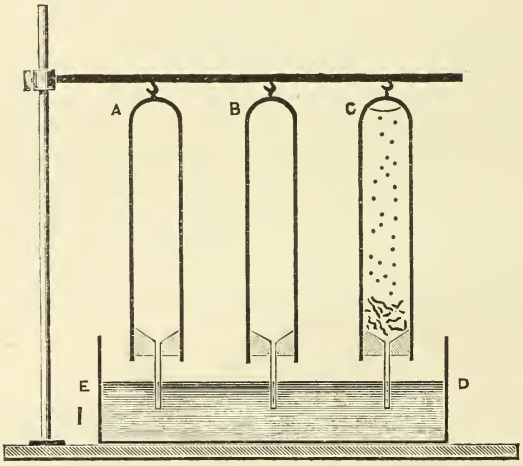
Botanical Application of the Röntgen Rays.§—Herr J. Istvánffy has experimented on the effect of the light of Crookes's vacuum-tubes on plants. He finds that the rays penetrate only the woody tissue, as can be made manifest in a leaf of *Camellia*, the veins of which appear white

* Flora, lxxxiii. (1897) p. 75.
† Brit. Med. Journ., 1897, i. p. 974. ‡ Tom. cit., p. 1215.
§ SB. K. Ungar. Naturw. Gesell. Buda-Pest, Feb. 12, 1897. See Bot. Centralbl., xlix. (1897) p. 267.

in the image. All other tissues, whether containing chlorophyll or not, are impermeable to these rays.

Demonstration of the Evolution of Oxygen by Diatoms.* — Mr. T. C. Palmer has succeeded in demonstrating the absorption of carbon dioxide and the elimination of oxygen by diatoms experimentally by the following process, dependent on the property of hæmatoxylin of assuming a yellow colour with a tinge of brown when absorbing CO_2, while, in the presence of nascent oxygen, the red hue gradually deepens, finally becoming a deep blood-red. In the apparatus here figured (fig. 20), the dish is filled up to the line D E with water tinted with a freshly made solution of hæmatoxylin, sufficient to stain it a pale red. The tube A is then filled with the same solution, stopped with rubber, through which is passed a quill tube, and the tube suspended, the very fine end of the quill dipping into the water. Another portion of the hæmatoxylin

Fig. 20.



solution is acidified by CO_2 from the lungs, blown into it through a glass tube, till it assumes a brownish-yellow tint, and the tubes B, C, prepared in the same way, are filled with this solution; living diatoms (*Eunotia major*) having been placed in tube C. The apparatus is now exposed to bright light, preferably to direct sunlight. Gas arises from the diatoms in tube C, and simultaneously the colour of the liquid, which is at first like that in B, begins to change. Within 15 minutes the colour has again become almost or quite as red as that in tube A. The CO_2 has now, in large measure, disappeared from the solution. The action continues, and the colour in tube C deepens rapidly, showing oxidation, and this action continues until the colour is blood-red. Still more striking results are obtained as follows:—In tube A is placed a living snail; in B live diatoms; C being left for comparison. Under the influence of sunlight, in the course of a few minutes, A pales

* Proc. Acad. Nat. Sci. Philadelphia, 1897 pp. 142-4 (1 fig.).

rapidly from the CO_2 given out by the snail; B as rapidly darkens and reddens; while C remains unchanged.

Microchemical Reaction for Nitric Acid.*—Mr. J. L. C. Schroeder van der Kolk has for several years used the following test, which is somewhat similar to that recently proposed by R. Brauns.† The substance to be tested is placed with a drop of sulphuric acid in the hollow of a glass slide, and from the cover-glass hangs a drop of barium hydroxide solution; when nitric acid is driven off, typical crystals of barium nitrate appear in the drop on the cover-glass. As the substance tested does not come into contact with the barium solution, the presence of sulphates, phosphates, &c., does not affect the result.

Dead-Black Surface on Brass.‡— To 2 grains of lamp-black in a saucer add, says Mr. L. A. Wilson, just enough gold-size as will hold the lamp-black together, and mix thoroughly. Dip a lead pencil into the gold-size, and the right quantity will be obtained; add drop by drop. After the lamp-black and size are thoroughly mixed and worked up, add 24 drops of turpentine, and work up again. Apply the mixture with a camel's hair brush. When thoroughly dry, the brass will look as if it had just come from the optician's hands.

Laboratory Notes. — Prof. K. Goebel § recommends the following objects for the purposes specified:—The leaves of *Elatostemma sessile* for the exudation of drops of water. *Klugia notoniana* for the formation of the embryo within the ovule. The protoneme of Mosses for the formation of starch out of sugar. The germination of the protoneme from the spore can be well followed out in *Funaria hygrometrica*.

Herr M. Raciborski || finds the epidermal cells of the perianth-leaves of cultivated species of *Albuca* favourable objects for observing the formation of crystalloids in the vacuoles within the endosperm. No fixing or staining of the object is necessary. The elaioplasts can also be demonstrated in the same cells.

Reversible Mailing Cases.¶— Messrs. Bausch and Lomb have brought out new mailing cases for microscopical slides. All the pieces are similar and interchangeable, thus avoiding the use of "tops" and "bottoms." An ample depression in the face of each piece allows the stowing of a slide with a large cover. Any number of slides and cases may be adjusted and piled one above the other, or one slide may be held securely by simply reversing one piece of wood.

* Neues Jahrb. Mineral., i. (1897) p. 219.
† Cf. this Journal, 1896, p. 687. ‡ The Microscope, v. (1897) pp. 43–4.
§ Flora, lxxxiii. (1897) pp. 74–5. || Tom. cit., p. 75.
¶ Journ. New York Micr. Soc., xiii. (1897) pp. 41–2 (2 figs.).

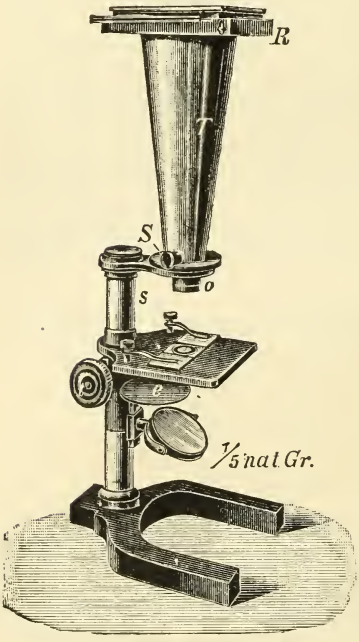
MICROSCOPY.

A. Instruments, Accessories, &c.*

(1) Stands.

Evolution of the Microscope.†—Mr. E. M. Nelson, on behalf of a sub-committee of the Quekett Microscopical Club, proposes in a series of articles:—(*a*) to investigate a good type of instrument; (*b*) to give a study of modern instruments, showing wherein, and why, they either follow or depart from the selected type; (*c*) to collate the other material bearing on the development of modern Microscopes, though not falling within the limits of *a* and *b*. For the type, Powell's No. 1 is to be taken, since to this the best modern Microscopes are more and more conforming; and, as it has remained in its present form for upwards of twenty years, it is a permanent type.

Fig. 21.



In the present paper several old forms, which are of importance in the evolution of the Microscope, and which have probably influenced the design of Powell's No. 1, are described. Figures are given of the instruments of Jansen (about 1660), Descartes (1637), Hooke (1665), Divini (1667), Chérubin d'Orléans (1671), Bonanni (1691), Hartsoeker (1694), and John Marshall (1704). Of importance are Hooke's, Bonanni's, and Marshall's.

Simple Microscope for Direct Observation and for Photography.‡—Herr C. Leiss describes the instrument shown in fig. 21, which is made by Fuess. The arm *o*, supporting the lens, is rigidly fixed to the foot, and the stage and mirror are made movable. Above the lens the small camera T may be fixed by the screw S. For direct observation, and for photographs which need not be very sharp, ordinary Steinheil lenses are used; but for better photographic work, two photographic objectives of 41 and 25 mm. focus are supplied; these fit into the arm *o*.

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

† Journ. Quekett Micr. Club, vi. (1897) pp. 349–56 (9 figs.).

‡ Zeitschr. f. angew. Mikr., iii. (1897) pp. 39–40 (1 fig.).

The stage has an aperture of 35 mm. diameter, and a smaller diaphragm can be laid over it if wished. A blackened disc *e* shuts off or admits light when photographing. A table of the focal lengths of the lenses used, amplifications, times of exposure, &c., is given.

Hand-Microscope.*—The accompanying fig. 22 represents a Hand-Microscope made by C. Reichert; it is simpler and more convenient

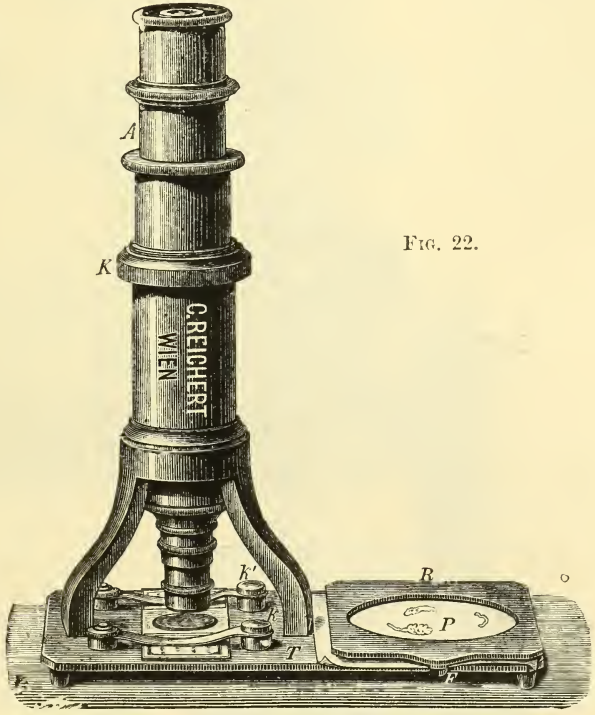


Fig. 22.

than many other demonstration instruments to be handed round in an audience. The tube slides easily in the sleeve, and, if necessary, can be replaced by another giving higher magnification. The extension of the stage is for carrying a drawing of the object shown.

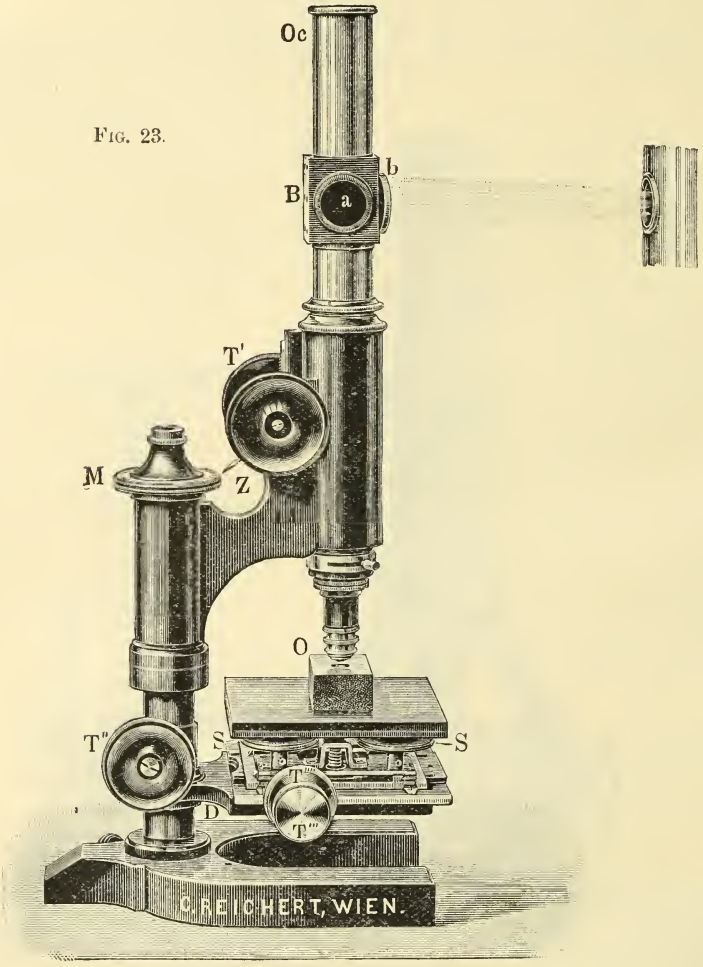
Stand and Illuminating Apparatus for Opaque Objects.†—Herr C. Reichert describes the instrument shown in fig. 23. The stand, with coarse and fine adjustment, is hinged to the heavy foot; the stage is of white glass, with a white background. Light passes through the tube

* Zeitschr. f. angew. Mikr., iii. (1897) pp. 44–5 (1 fig.).

† Tom. cit., pp. 40–1 (1 fig.). The same instrument is also described by Prof. A. Rejtö, Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 1–4 (1 fig.).

b, which is fitted with an iris-diaphragm, and is reflected by a mirror at *a* on to the object. The instrument is especially designed for

Fig. 23.



studying the structure of etched and fractured surfaces of metals and alloys; it will also be useful for observing the surface characters of minerals, rocks, &c.

(3) Illuminating and other Apparatus.

Improved Illuminating Apparatus.*—Herr C. Reichert describes an improved form of the Abbe substage illuminating apparatus, of

* Zeitschr. f. angew. Mikr., iii. (1897) pp. 33-5 (4 figs.). Cf. this Journal, 1896, p. 373.

which several modifications have been made since it was first introduced. The special point of the present form is that it is independent of the mirror; it can also be swung to one side and other accessories attached.

Fig. 24.

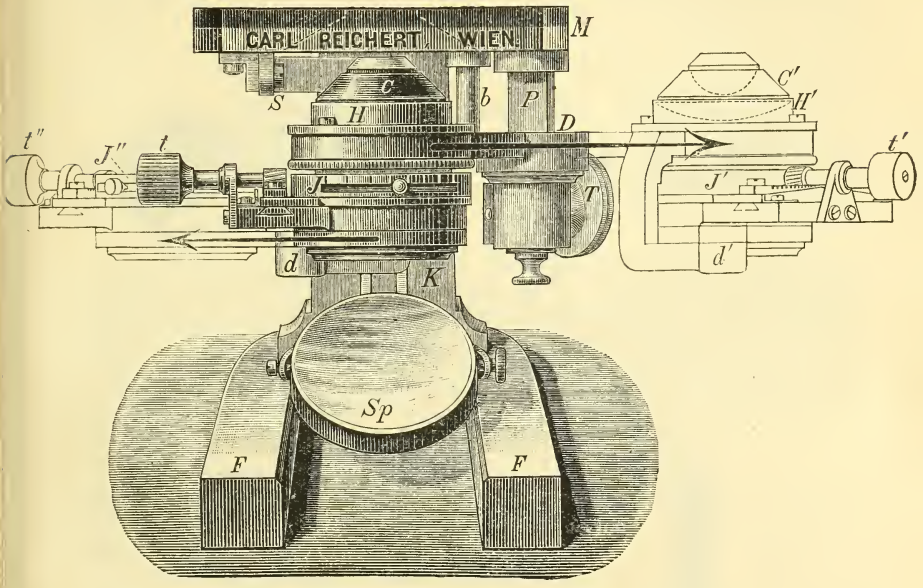
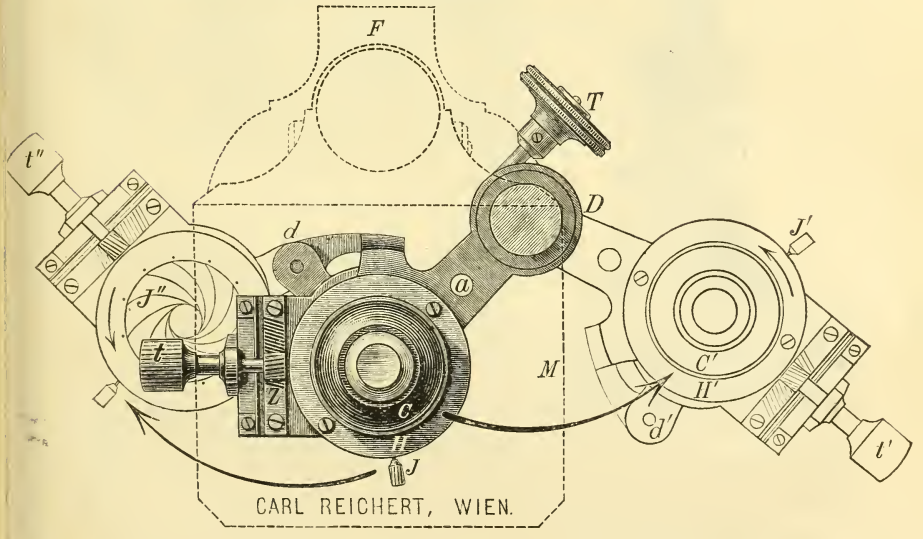


Fig. 25.



The whole apparatus swings about the axis D (figs. 24 and 25), as shown by the outline drawings to the right, and it has a vertical motion

by means of gearing T. On the upper part of the main arm is a sleeve into which the condenser C, or other accessory, fits. On the under side of the same arm the iris-diaphragm is carried by the pivoted arm *d*, and can be swung out of position as shown on the left. For oblique illumination the diaphragm may be moved from side to side by the rack

Fig. 26.

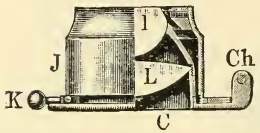
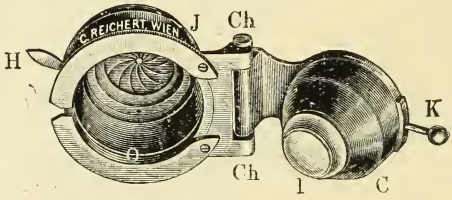
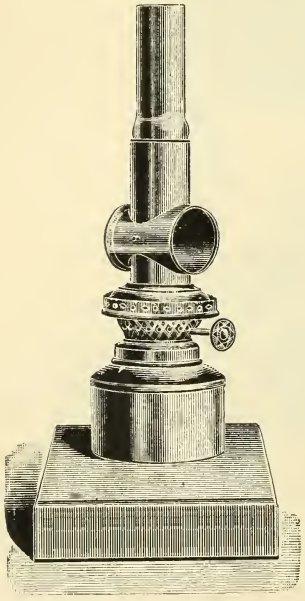


Fig. 27.



and pinion *t*. The whole apparatus is held in a central position by a pin, which projects from the under side of the stage, fitting into the hole *a*, when the apparatus is screwed upwards by the arrangement T. In cases where it is necessary to quickly change from convergent to parallel light, the condenser and diaphragm are hinged together, as is shown in figs. 26, 27. Owing to the size of the apparatus, it can only be fitted to the larger stands.

Fig. 28.



Coloured Illumination.* — Mr. J. Rheinberg describes a new form of "substage differential colour illuminator," consisting of a box containing nineteen metal slides, which can be moved in or out independently of one another by means of small handles. Each slide has two circular apertures, one of which is fitted with a colour disc or other stop. The kind of stop is indicated on the handle. When the slides are pushed in, only the blank apertures are in the path of the light, but when pulled out, one or other of the stops is brought into use. The stops for giving coloured backgrounds, and for illuminating the object with various stops, include a dark-ground stop, various colours, parti-coloured stops, stops for oblique light, several annuli, and a ground-glass stop. The various combinations of stops which can be brought into use are almost endless.

Portable Microscope Lamp.† — Mr. W. Goodwin has designed a lamp made by Mr. Hinton (fig. 28). The metal chimney has two circular apertures 1 in. in diameter, one of which is glazed with signal green and the

* Journ. Quekett Micr. Club, vi. (1897) pp. 346–7. † Tom. cit., p. 345 (1 fig.).

other with steel-blue glass. The cylindrical reservoir is 2 in. diameter, and the total height of the lamp is 7 in. A blotting-paper wick is used.

Simple Instrument for Inclining a Preparation in the Microscope.*—Mr. T. A. Jagger, jun., has devised an instrument for use in petrography, especially in connection with the optical methods of Michel-Lévy and Fedorow for the determination of felspars; it will also be of use in cases where it is necessary to examine the edges of objects in reflected light. The object-holder clip is supported by a ball-and-socket joint on a foot-plate, which may be fitted to the stage of the Microscope. This allows the object to be turned into any desired azimuth, and the various minerals in rock sections can then be brought into definite orientations, as is done in the more complicated "universal stage" of Fedorow. The optic figures seen in convergent polarised light may also be brought into the centre of the field of view.

Modification of the Automatic Gas-Stop for Extinguishing the Burner of Incubators.†—Dr. B. Schürmayer has devised a modification of the gas-stop to incubators which prevents the escape of gas and the danger of its exploding. The supply tube is fitted with a metal spiral lever which acts on a stopcock, so that if there be any fault in the working of the apparatus beyond this point, and the gas supplied to the burner fail, the lever acts on the stopcock, and thereby prevents further access of gas to the apparatus. The most important feature of the invention is that all the parts of the apparatus are of metal.

Slide and Cover-Glass Holders.‡—Herr S. Robertson has devised holders for slides and cover-glasses, the construction of which may be

Fig. 29.

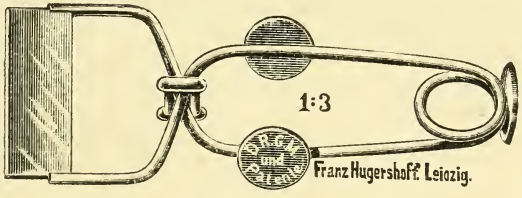
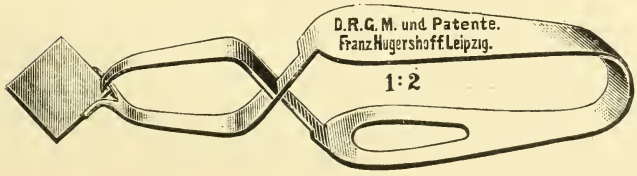


Fig. 30.



grasped from an inspection of the illustrations (figs. 29 and 30). The former is merely a spring-forceps made of nickelled wire, the inside of the fangs being grooved for the reception of the slide. The cover-glass

* Amer. Journ. Sci., iii. (1897) pp. 129–31 (2 figs.).
† Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 400–1 (1 fig.).
‡ Tom. cit., pp. 589–91 (2 figs.).

forceps has a trifurcate lower fang between which the slip is inserted. The two upper teeth of the fang are grooved, and are set at a right angle, so that a square cover fits in accurately, and is held firmly by the upper fang.

(4) Photomicrography.

Photomicrography.*—Mr. T. J. Bray considers the practical employment of ordinary objectives in photomicrography; he shows that for ordinary work it is not really necessary to have specially corrected lenses, and that with ordinary objectives of low power good results can be obtained. The objective he finds most useful is Bausch and Lomb's student's 3 in. to 6 in. variable; this with a long bellows camera gives a wide range of amplification, which may be further increased by the usual methods of photographic enlargement of the negative. For large negatives a dark room takes the place of the camera, the Microscope and light being arranged outside. With the long bellows camera, the focusing arrangement of Dr. Mercer is adopted. This consists of a wire from the back of the camera, which is connected by a vertical lever to a horizontal extension of the pinion shaft of the Microscope coarse-adjustment. The picture is focused, with the aid of a 2 in. eye-piece, on thin cover-glasses cemented on the ground-glass screen of the camera, as is done by Walmsley. Many details can only be brought out by using a colour screen; this is a bichromate cell, as is used in cloud photography. Photographic details are given in the paper.

Acetylene Gas in Photomicrography.†—Mr. W. H. Walmsley has previously suggested the use of acetylene gas as an illuminant in photomicrography.‡ The light which it gives is white, very brilliant, and absolutely steady; there is little heating and no smell; it is portable, simple, cheap, and safe, and is always ready for use. All these advantages are not combined in sun, lime, magnesium, electric, or other lights. The flame, which consumes one-tenth to one cubic foot of gas per hour, is enclosed in a metal case with a glass front; the flat flame is placed end on, or pencil flames are placed behind each other with diaphragms between. The author, for his own use, renders the light monochromatic by means of a cobalt blue cell placed in the substage of the Microscope.

The use of an acetylene flame as a standard unit of light is suggested. An automatic machine for generating the gas from calcium carbide is supplied by Walmsley, Fuller & Co., of Chicago.

Astronomical Photography with Photomicrographic Apparatus.§—Dr. A. C. Mercer obtained photographs of the partial eclipse of the sun seen at Syracuse, N.Y. on Oct. 20, 1892. The heliostat and a portrait lens of 8 in. focus were arranged to throw a stationary image of about 1/12 in. diameter of the sun's disc in the plane usually occupied by the object on the stage of the Microscope. This image was projected by a $1\frac{1}{2}$ in. Microscope objective to form a second image, $2\frac{3}{8}$ in. diameter, on the ground-glass of the camera. As far as compactness is concerned, this arrangement is more convenient than telescopic methods, but it is inferior in illumination and separating power. The results obtained are

* Trans. Amer. Micr. Soc, xviii. (1897) pp. 107–16. † Tom. cit., pp. 136–41.
‡ Cf. this Journal, 1896, p. 126.
§ Trans. Amer. Micr. Soc., xviii. (1897) pp. 132–5 (2 figs.).

compared numerically with those obtained with the Lick photographic objective.

Photomicrograph v. Microphotograph.*—Dr. A. C. Mercer, in the present note on his paper of 1886, points out that the word photomicrograph was first used in 1858. An account of its origin is to be found in the 'Liverpool and Manchester Photographic Journal' (now 'British Journal of Photography'), August 15, 1858, pp. 203 and 414; also in 'Sutton's Photographic Notes,' iii. pp. 205 and 208.

Advances in Photomicrography.†—Herr G. Marktanner-Turneretscher collects together under this title a series of abstracts of recent papers relating to Photomicrography; these have already been noticed in this Journal.

(5) Microscopical Optics and Manipulation.

Multiple Images in Mirrors.‡—Mr. W. B. Stokes explains the origin of multiple images seen in plate-glass mirrors. The brightest image is due to reflection from the silvered back, another to reflection from the front glass surface, and others are due to more than one reflection within the glass. When the mirror is rotated in its own plane, these images will change their position, owing to the fact that the surfaces of the plate are not truly parallel. For a particular angle of inclination of the two surfaces of the plate, the first two images may be made to coincide.

B. Technique.§

(1) Collecting Objects, including Culture Processes.

Apparatus and Method of Manipulation for the Preparation of Roll Cultures of Anaerobic Organisms.‖—In using the apparatus invented by Mr. E. E. Ewell (fig. 31), the tube is inoculated with the requisite number of organisms in the usual way, and is placed in the water-bath B, the temperature of which is kept at some convenient degree between the solidifying point of the medium and the thermal death point of the organism to be cultivated. The cotton plug P is pushed into the tube to make room for the rubber stopper carrying the glass tubes *a* and *a'*. The stopper is carefully sealed with sealing-wax, and the connections made with the thick rubber tubes N and O, the latter being secured with wire. E leads to a vacuum service pipe, or to some form of vacuum pump; F leads to a hydrogen gas generator. I and H being closed, open L until the air is removed, then close L and open H. The mercury contained in the bottle C passes up the tube D until an equilibrium is established where the point at which it comes to rest is marked. If all parts of the apparatus are tight, the column of mercury will remain stationary; if it falls, all the connexions must be re-examined. In order

* Trans. Amer. Micr. Soc., xviii. (1897) p. 131. Cf. this Journal, 1887, p. 665.

† Jahrb. f. Photographie u. Reproductionstechnik, 1897, 12 pp. and 4 figs.

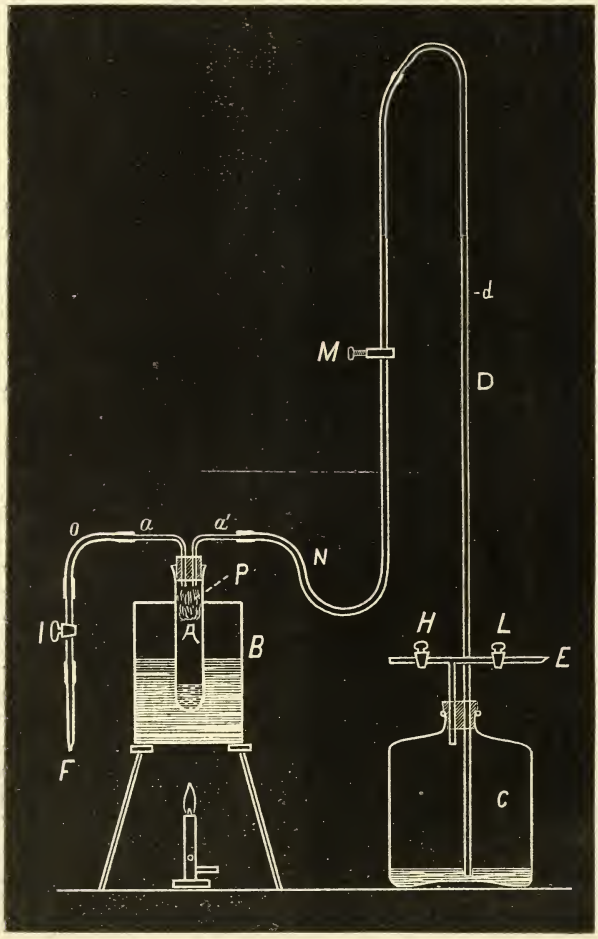
‡ Journ. Quekett Micr. Club, vi. (1897) pp. 322–4 (3 figs.).

§ 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. Bakt. u. Par., 2te Abt., iii. (1897) pp. 188–90 (1 fig.).

to remove all possibility of leakage around the stopper A, the flame of a Bunsen's burner is applied to the sealing-wax until it is sufficiently softened for the pressure of the atmosphere to force it into any crevice that it has not reached. When the column of mercury in D becomes stationary, admit hydrogen by means of I, until the vacuum in A is

FIG. 31.



destroyed. Close I and H and open L until exhaustion is complete; close L and open H and I successively. The alternate exhaustion and filling are repeated until there is no possibility of any air remaining in A, when the tubes *a* and *a'* are drawn apart and sealed in the flame. In order that the pressure within A may be only very slightly different from that of the atmosphere, H is not opened after the last exhaustion.

I is opened, and when the acid reaches the same level in both parts of the gas generator, quickly close M, and then close I just as soon as the change of level in the generator shows that there is a slight excess of pressure in A. After sealing, the tube is transferred to the ice block and rolled until the agar or gelatin is solidified. In case of agar, the rolling must be very rapid to ensure good results. The apparatus is also of service for displacing the air from other forms of anaerobic culture apparatus. If the vacuum pump used is capable of giving a column of mercury 635 millimetres high in the tube D, five-sixths of the gases in the tube A will be removed at each exhaustion. A simple calculation will satisfy the operator in regard to the number of exhaustions necessary.

Preparation of Culture Media and their Sterilisation.*—Mr. R. C. Reed recommends the following methods for the preparation of nutrient media.

Peptonised Bouillon.—To 1000 grm. of finely divided meat, add 2000 grm. of distilled water, and place in an agate or iron dish, and then heat in a water-bath at from 60°–65° C. for two hours, or allow it to macerate in a cool place for 24 hours. Strain through a coarse cloth, and bring the amount of liquid up to 2000 ccm., adding water if necessary. Then add 0·5 per cent. pepton, and 0·5 per cent. sodium chloride; and, if a neutral or alkaline medium be desired, add enough of a 1 per cent. solution of caustic soda. Boil in a water-bath for half an hour. Cool, and filter through ordinary filter paper, and distribute in sterilised flasks.

Nutrient Agar.—Dissolve 5 grm. of finely cut agar in 100 ccm. of water, and then add it to 500 ccm. of bouillon and boil for 20 minutes. Cool down to 45°–50° C., and add the whites of two eggs. Return to the water-bath and boil for 20 or 30 minutes. In this way the clot will be got rid of, and a perfectly clear liquid left. Filter through ordinary filter paper while hot, and distribute into sterilised tubes.

Nutrient Gelatin.—To 500 ccm. of bouillon add 50 grm. of gelatin, and heat in a water-bath until the gelatin is dissolved. Cool to about 45° C., and then add the whites of two eggs, mixing the lot thoroughly. Boil in a water-bath for about 20 minutes. Filter, and distribute in sterilised tubes. Care must be taken not to boil the gelatin too long, lest it will not set when cold.

The author goes on to point out that lengthy discontinuous sterilisation may be overdone; for he has found that sterilising for one day for 30 minutes usually suffices. All that is necessary is to incubate afterwards for several days, and then reject the few tubes which are contaminated.

Rapid and Easy Method for Preparing Nutrient Agar.†—Herr E. S. London prepares agar in 33 minutes in the following way:—To 1 litre of meat water are added 5 grm. of salt, 10 grm. of pepton, and 15 grm. of agar, and the mixture placed in a flask which is heated in an autoclave until the temperature rises to 130°. The steam is then gradually slacked off, and when the temperature has sunk to 100° the

* Amer. Monthly Micr. Journ., xviii. (1897) pp. 149–54.

† Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 686–7.

flask is removed, and its contents filtered through a Diakonow's apparatus. The clear filtrate is then poured into flasks and neutralised. For Diakonow's apparatus may be substituted a simple arrangement consisting of a flask, the neck of which is closed by a caoutchouc plug with two holes. One of the holes receives the stem of a filter; the bottom of the hopper is covered with a piece of gauze upon which is placed a layer of glass wool and then a layer of fine sand. To the second hole is fitted a bent glass tube connected with an exhaust pump.

Apparatus for Cultivating Yeasts on Plaster Blocks.*—M. H. Schionning has, by combining the advantages of the plaster block and a Hansen's flask, devised an ingenious apparatus for obtaining pure ascosporous cultures of yeasts. A cylindrical block of plaster, reaching to about two-thirds up the flask, and made by mixing 2 volumes of plaster with 3/4 volume of water, is fixed to the bottom of the flask. The top of the plaster pillar is slightly hollowed for the reception of the yeast. The side tube having been plugged with cotton-wool, the apparatus is sterilised at 115° for an hour and a half. When cooled, the cap is removed and the culture placed in the hollow on the top of the pillar. The cotton-wool plug is then removed, and through the lateral tube sterilised water is introduced in quantity sufficient to reach halfway up the plaster cylinder. When the top of the pillar shines from imbibition of water, the whole apparatus is incubated at a temperature favourable for the production of spores. A piece of rubber tubing is previously fitted on the lateral tubulure, its free end being plugged with cotton-wool. By this procedure, perfectly pure cultures, quite free from bacteria and other contamination, are easily possible.

Amœba Cultures.†—Dr. O. Casagrandi and Dr. P. Barbagallo report on the different kinds of media suitable for Amœba cultures, on the reaction of the substrata, on the necessity for the presence of organised constituents therein, and on cultivable and non-cultivable Amœbæ. With regard to fluid media, there is no doubt, they say, that Amœbæ will develop in hay, straw, and hemp infusions, on decoction of fæces, and in thin albumen; but it is practically impossible to obtain a pure cultivation, partly owing to the difficulty of sterilising the medium, and partly on account of the impurity of the inoculation material. Of solid media, egg-albumen with some pepton and carbonate of soda was found extremely serviceable.

Media composed of the above mentioned infusions solidified with agar or gelatin were failures. A medium containing 5 per cent. of *Fucus crispus* was found to possess the advantages of inhibiting the growth of bacteria, and of affording opportunity to the Amœbæ of completing the stages of their life-cycle.

The most suitable reaction was found to be slightly alkaline to neutral; strongly alkaline or acid reaction being unfavourable. Acidity of the medium did not, however, prevent certain species, such as *A. coli*, from becoming acclimatised to the reaction. With regard to the association of protozoa, bacteria, fungi, and yeasts, with Amœba in the cultures, which some observers seem to regard as necessary and irremov-

* Ann. de Micrographie, ix. (1897) pp. 194–8.
† Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 579–89.

able evils, the authors, while acknowledging the difficulty of obtaining pure cultivations, do not seem to consider this as impossible. From fæces were obtained cultures of *A. coli*, *A. guttula*, *A. spinosa*; from *Blatta*-excrement, *A. blattarum*; from muddy water, *A. guttula*, *nodosa*, *diffluens*, *arborescens*, *gracilis*, *spinosa*, and *oblonga*; from damp earth in unhealthy places, *A. guttula*, *spinosa*, and *arborescens*; from beer yeast, *A. guttula* and *spinosa*.

Culture Medium for Algæ and Amœbæ.*—Dr. N. Tischutkin recommends 1 per cent. aqueous solution of agar for the cultivation both of Algæ and of Amœbæ.

Crystal Formation in Culture Media.†—Dr. Marion Dorset regards the early formation of crystals in freshly prepared agar as a special characteristic of *Bacillus pyocyaneus*. Other bacteria produce crystals in culture media, but only when the media are old, and therefore partially dried.

(2) Preparing Objects.

Methods for Demonstrating the Continuity of Protoplasm.‡—In discussing the various methods adopted for demonstrating the continuity of protoplasm, Herr A. Meyer first makes a few remarks on fixation of the tissue. For this 1 per cent. osmic acid is recommended, though strong iodopotassic iodide (iodine 3, iodide of potassium 3, water 20) and potassium-bismuth iodide solutions give favourable results. For softening membranes sulphuric acid is the best agent (H_2SO_4 1 vol. to 0·5–3 vols. water). A very strong solution of iodine made by dissolving 1 vol. of iodine and 1 of iodide of potassium in a few drops of water, and then adding 200 ccm. of water, is useful occasionally for staining the threads of protoplasm. By staining with Hoffmann's blue or Bavarian blue, the continuity of the protoplasmic processes was rendered distinctly visible. The sections were placed for a few minutes in a solution of 1 grm. of pigment and 150 grm. of 50 per cent. spirit, and examined in glycerin. Permanent preparations can be made from tissue fixed and hardened in osmic acid or alcohol by over-staining the sections in Delafield's hæmatoxylin (24 hours), and, after washing in 60 per cent. spirit, decolorising in 0·5 per cent. HCl. The sections must then be immersed in 60 per cent. spirit rendered alkaline by the addition of ammonia (10 drops to 100 ccm.). After this they are transferred to absolute alcohol, xylol, and mounted in balsam. A method for staining after mordanting with iodine is described at some length. The reagents required are:—(1) Iodopotassic iodide solution (iodine 1, iodide of potassium 1, water 200); (2) sulphuric acid (1–3), which has been saturated with iodine by standing over some iodine; (3) a solution of 1 grm. of pyoktanin cœruleum (Merck) in 30 ccm. of water. The sections are immersed in solution 1 for some minutes, and then placed on a slide and covered with a slip. Solutions 2 and 3 are added at the side of the cover; and having been allowed to act for about three minutes, the slide, section and cover-glass are immersed in a large quantity of water. Having been quickly washed, the section is placed on a clean slide and examined in glycerin.

* Centralbl. Bakt. u. Par., 2te Abt., iii. (1897) pp. 183–8.
† Op. cit., 1te Abt., xxi. (1897) pp. 473–4.
‡ Ber. Deutsch. Bot. Gesell., xv. (1897) pp. 166–77.

If the staining has been successful, the membrane is pale blue, while the protoplasm and its connections are blackish blue.

Rapid Method of making Permanent Specimens from Frozen Sections by the Use of Formalin.*—The method described by Dr. T. S. Cullen is as follows:—A piece of the fresh tissue is sectioned on an ether freezer, and the sections placed in 5 per cent. solution of formalin for 3–5 minutes, then in 50 per cent. alcohol for 3 minutes, and in absolute alcohol for 1 minute. Wash in water; stain in hæmatoxylin for 2 minutes; decolorise in acid alcohol; rinse in water. Stain with eosin; transfer to 95 per cent. alcohol; then pass through absolute alcohol, creosote or oil of cloves, and mount in balsam.

Or as an alternative method, should it be desired to retain the blood in the sections, a piece $1 \times 5 \times 2$ cm. is placed in 10 per cent. formalin for 2 hours, after which the procedure is as before.

(3) Cutting, including Imbedding and Microtomes.

Simple Microtome for Biological Work.†—Mr. A. Flatters describes an improved form of the simple microtome originally designed by him. The carrier is moved upwards in the cylindrical well by a screw carrying at its lower end a notched disc, against which works a clicking arrangement; three of these discs with different numbers of notches may be used, so that sections of any desired thickness may be cut. The aperture of the razor plate is, on the under-side, of the same diameter as the well, but on the upper side it is slightly less, this being for the purpose of firmly holding the imbedded mass in position as it is screwed up. The razor plate, which is held in position over the well by a clamp, may be swung on one side to enable the uncut material to be removed. For larger or longitudinal sections a special razor plate, with a rectangular aperture and a corresponding holder, may be fitted to the instrument.

(4) Staining and Injecting.

Simple Method for Contrast-Staining Micro-Organisms.‡—Dr. Claudius stains microbes on covers and in sections by the following procedure. The reagents used are (1) 1 per cent. aqueous solution of methyl-violet; (2) 1 vol. of saturated aqueous solution of picric acid plus 1 vol. of water; (3) chloroform; (4) oil of cloves.

Cover-glass preparations are stained in the methyl-violet solution for 1 minute, washed in water, mopped up on blotting-paper, immersed in the picric acid solution for 1 minute, washed and mopped up again, then decolorised in chloroform, and, after having been dried, mounted in Canada balsam. Sections are stained and treated very similarly, but the two solutions are used for two minutes instead of one; and, after having been very carefully mopped up, are decolorised by means of oil of cloves, after which they are passed through xylol, and then mounted in balsam. Twenty-six species were tried by this method: 17 were stained and 9 were not; among the latter being *B. typhi*, *B. coli com.*, *Sp. choleræ asiat.*, *Pneumobac. Friedlaenderi.*

* Bull. Johns Hopkins Hosp., viii. (1897) pp. 108–9.
† Pharmaceutical Journ., lviii. (1897) pp. 485–6 (4 figs.).
‡ Ann. Inst. Pasteur, xi. (1897) pp. 332–5.

Staining Vegetable Sections.* — Prof. F. D. Kelsey recommends that vegetable sections should be stained with pigments dissolved in clove oil. After staining, remove to pure clove oil or Gage's fluid, and mount in balsam. It is hardly necessary to point out that the section must be perfectly dehydrated before it is immersed in the clove oil stain. A dilute clove oil stain acts better than a concentrated one.

Method of Staining the Malaria Flagellated Organism.† — Dr. P. Manson has succeeded in staining the flagellated malaria parasite by the following method, which shows the pigment and certain details of structure with ease and certainty. Thirty or forty strips (3 by 1½ inches) of thick blotting-paper, each having an oblong hole (1 by ⅔ inch) cut lengthwise in its centre, are prepared. They are then slightly moistened with water and laid in rows on a sheet of window glass. A droplet of blood, the size of a large pin's head, is then obtained by puncturing the finger of a person in whose blood the crescent form of the malaria parasite abounds. A Microscope-slip is then breathed on once, and the droplet of blood dabbed on the centre of the breathed-on surface. The blood is then spread out with a needle so as to cover an area of ¾ by ½ inch, and the slip immediately inverted over a blotting-paper cell. The slip is then pressed down, care being taken to prevent the blood coming in contact with either the wall of the cell or the floor of what is now a very perfect moist chamber. In from half to three-quarters of an hour the slips are removed, and dried by gently warming them over a spirit-lamp. When dry, the films are fixed with absolute alcohol. After five minutes the alcohol is dried off, and a few drops of acetic acid (10–20 per cent.) are laid on the film, and left long enough to dissolve out the hæmoglobin. The slips are then washed in water and dried. After this they are stained with 20 per cent. phenol-fuchsin, the stain being dropped on and the slip covered with a watch-glass. After six hours it is washed off, the slide dried, and a cover-glass applied with xylol-balsam. Most of the slides will show numbers of spheres and several or many well-stained flagellated bodies. Very few crescents remain untransformed. If the slips are removed and dried in from five to ten minutes after being placed on the blotting-paper cells, only crescents, ovals, and spheres will be found; if left for more than three-quarters of an hour, free flagella and spent pigment may be found.

Staining Diphtheria Bacilli.‡—Dr. C. F. Craig states that Crouch of Denver has found that, if 24 hours old culture of diphtheria be treated for a few seconds with 1 per cent. methyl-green solution, the majority of the bacilli will be faintly stained green, and will show at both ends a well defined round body of a distinctly red colour. The following solution was found to be very serviceable :—1 per cent. methyl-green, 5 parts; 1 per cent. solution of dahlia, 1 part; distilled water, 4 parts. Only a second is required for staining; if left in longer, the staining is too intense.

Combination of Weigert's Fibrin Method and the Tubercle Bacillus Stain.§—Herr Roloff stains tubercle bacilli and fibrin in the same section

* The Microscope, v. (1897) p. 69.

† Brit. Med. Journ., 1897, ii. pp. 68–70 (15 figs.).

‡ Trans. Amer. Micr. Soc., xviii. (1897) pp. 274–5.

§ Arb. a. d. Pathol.-Anat. Inst. zu Tübingen, ii. (1896) p. 261. See Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) p. 749.

by the following procedure. The sections are stained for twenty-four hours in an incubator in carbol-fuchsin, and then decolorised with Ebner's fluid. After having been washed in 70 per cent. spirit, they are transferred to acetic acid vesuvin solution (Kahlbaum) for some hours. Next they are washed in water and in 70 per cent. spirit, and are then stuck on a slide, after which they are stained by Weigert's method. The anilin-xylol should be allowed to act for some time, otherwise the nuclei will be blue instead of brown. If the differentiation be successful, the nuclei are brown, the fibrin blue, the tubercle bacilli red, and other bacteria blue.

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

To Prevent Freezing of Formol.*—Dr. A. Milani finds that the addition of 25–35 parts of glycerin to the formol solution prevents the danger of freezing.

(6) Miscellaneous.

Rapid and Improved Method for Counting Plate Colonies.†—Dr. H. J. van't Hoff has devised the following method for counting bacterial colonies on plates:—On the middle of a gelatin plate having a diameter of about 15 cm. is dropped about 0·2 ccm. of water. The water is then distributed over the surface of the gelatin by merely rolling the capsule about, care being taken to prevent the water from reaching the side. Distributed in this way, the colonies develop quite separately, and so rapidly that in two days it is possible to obtain a better quantitative result by this method than in five or six days by the ordinary procedure.

Method for Examining Malarial Blood.‡—Dr. N. Macleod uses strips of ordinary note-paper 0·5 in. wide and about 1½ in. long for smearing cover-glasses with malarial blood. The straight edge is drawn its full half inch through a drop of blood not larger than a pin's head, and then the edge drawn across the cover-glass. In this way a thin film which dries very rapidly may be spread on cover-glass or slide. The film must be mounted dry. With a 1/4-in. objective crescents and the larger pigmented parasites, and with a 1/12 oil immersion the smaller pigmented forms, can be easily seen. The method, however, cannot be relied on for the detection of unpigmented forms without very considerable experience, and should be supplemented by staining, or the examination of fresh undried films.

Bacteriological Diagnosis of Leprosy.§—The method advocated by Messrs. Johnston and Jamieson for the bacteriological diagnosis of leprosy is extremely simple, and consists in smearing a cover with a drop of serum obtained by scraping one of the leprosy nodules. This is stained with carbol-fuchsin, and decolorised with sulphuric acid and methylen-blue. The bacilli of leprosy are found in large numbers, and this fact alone is sufficient to distinguish leprosy from tubercle bacilli. Lepra bacilli also stain readily with simple anilin dyes, while tubercle bacilli do not.

* Zool. Anzeig., xx. (1897) pp. 206–8.
† Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 731–3 (1 fig.).
‡ Lancet, 1897, ii. pp. 85–6.
§ Montreal Med. Journ., 1897. See Epit., Brit. Med. Journ., 1897, p. 92.

Diagnosis of Smegma and Tubercle Bacilli.*—Herren R. Bange and A. Trantenroth state that the only satisfactory method for distinguishing the smegma from the tubercle bacillus is the following:—Immersion in absolute alcohol for not less than three hours, in 5 per cent. chromic acid not less than 15 minutes, in phenol-fuchsin, acid. sulph. dil. 2–3 minutes, and in saturated alcoholic solution of methylen-blue at least 5 minutes.

Notes on the Agglutination Phenomenon of Typhoid Serum.—According to Widal and Sicard,† the agglutination phenomenon can be obtained with dead bacilli killed by heat (57°–60°), or better still with certain chemical agents, such as formalin. By adding a drop of formalin to 150 drops of 1–2 days old typhoid culture, the bacilli are killed, though they perfectly retain their sensitiveness to the serum for a week. These authors also state‡ that from a drop of blood drawn from the finger and received into a sterile tube, a qualitative and quantitative test can be made.

Ferrand § cites a case wherein the agglutination reaction with typhoid culture was obtained from the serum of a person who died of streptococcus septicæmia, and Nicolas ‖ states that the same phenomenon occurs in patients who have been treated with antidiphtheritic serum.

Dr. M. W. Richardson ¶ finds that dried typhoid serum acts well for the purpose of diagnosing typhoid cultures from those of *B. coli communis*, *B. pyogenes fœtidus*, and mouse typhoid. It is only necessary to take blood from the heart of a person dead of typhoid fever, and after obtaining the serum, pour it through a filter paper and dry it.

Dr. W. Johnston** confirms the value of Pfahl's modification of Widal's test; in this the dried blood of a patient suspected of enteric fever is dissolved in water.

Kolle†† states that the virulence of the culture should always be taken into consideration, as normal serum in dilutions of 1–10 or 15 often produces an agglutinative effect on cultures which are but slightly virulent.

Photometric Determination of Heliotropic Constants.‡‡—Prof. J. Wiesner recommends that the measurement of the heliotropic source of light should be determined by the photochemical method and the Bunsen-Roscoe unit of measure. In this way the intensity of those rays which act on silver chloride may be measured. The values obtained are generally comparable; for they hold good for gaslight, electric light, and daylight. By this method it was determined that organs very little heliotropically sensitive will even react to a fraction of a millionth of the Bunsen-Roscoe unit.

* Fortschr. d. Med., 1896, Nos. 23 and 24. See Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 353-4. † La Semaine Méd., 1897, p. 38.
‡ Tom. cit. p. 69. § Tom. cit., p. 30. ‖ Tom. cit., p. 37.
¶ Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 445-6.
** Tom. cit., pp. 523-6.
†† Deutsche Med. Wochenschr., 1897, No. 9. See Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 484-5. ‡‡ Bot. Centralbl., lxix. (1897) pp. 305-9.

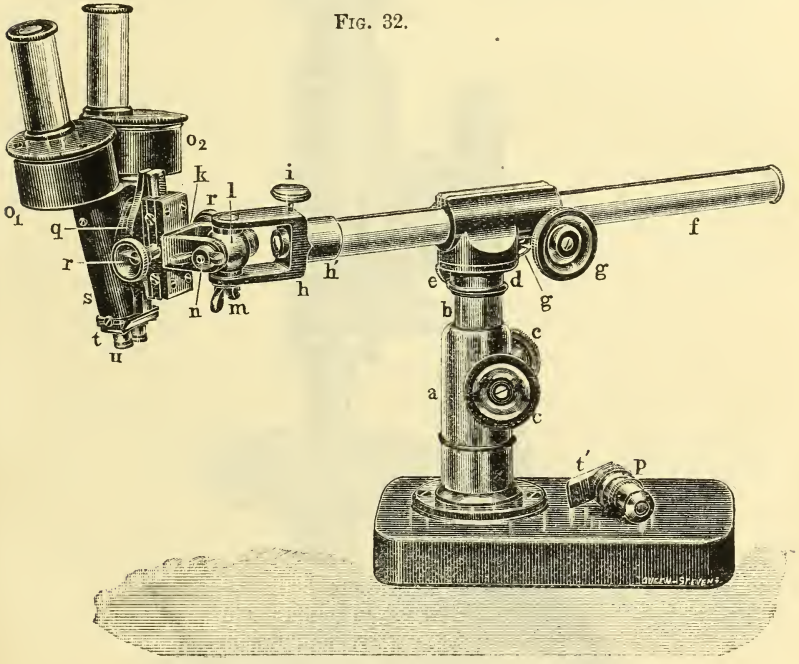
MICROSCOPY.

A. Instruments, Accessories, &c.*

(1) Stands.

Preparation and Horizontal Binocular Microscope.† — Drs. L. Drüner and H. Braus describe an improved form of their binocular Microscope which has been especially useful for the dissection of nerves and muscles. The present improved stand (fig. 32), also made by Zeiss,

FIG. 32.



enables the optical portion of the instrument to be placed in any desired position, this being for the purpose of facilitating the dissection of large surfaces, and, when the Microscope is placed horizontally, for observing the movements of small organisms through the vertical sides of aquaria.

The optical portion may be moved with respect to the fork *k* by the rack and pinion *r*. The forks *k* and *h* are connected by a ball-joint *l* supplied with the necessary clamps ; the whole of this rotates with the collar *h'* about the horizontal rod *f*. This rod has a longitudinal motion

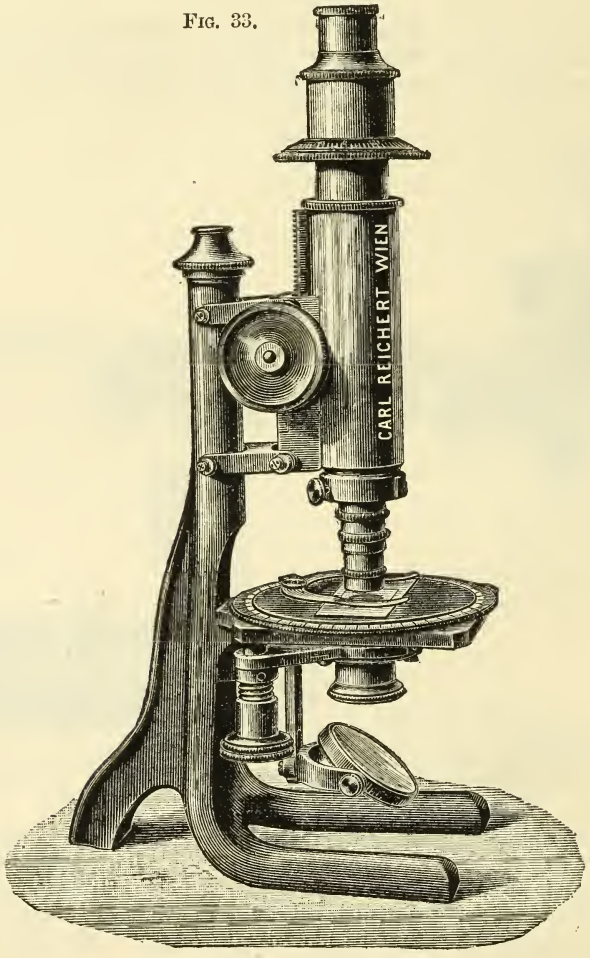
* This subdivision contains (1) Stands; (2) Eye-pieces and Objectives; (3) Illuminating and other Apparatus; (4) Photomicrography; (5) Microscopical Optics and Manipulation; (6) Miscellaneous.

† Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 5–10 (2 figs.). Cf. this Journal, 1895, p. 580.

by means of the rack and pinion g, a vertical motion by the rack and pinion c, and it can be rotated in a horizontal plane with the collar d.

The two erecting oculars o_1 and o_2 are carried eccentrically, so that they can be accommodated to the distance between the eyes. The two objectives slide into grooves t. With the various oculars and objectives

Fig. 33.



supplied with the instrument, amplifications of 6·5 to 48 are obtained. For use as a monocular, one of the tubes may be brought parallel to the rack by means of the turn-plate q.

New Stand.*—Herr C. Reichert describes the new stand (No. VII *b*) shown in fig. 33, which forms a cheap instrument for mineralogical and

* Zeitschr. f. angew. Mikr., iii. (1897) pp. 74–5 (1 fig.).

geological purposes. The foot and column are cast in one piece, so that the instrument can safely be held and carried by the upper portion. Roberval's micrometer-screw is used for the fine adjustment, this being an advantage in small, though not in large, stands. The usual accessories for parallel and convergent polarised light are added.

Method of Using the Microscope.*—Mr. N. A. Cobb describes, in the following terms, an apparatus which he has found useful in the application of the Microscope to agricultural inquiries:—

"The apparatus I have to describe has been so very useful to me that I cannot but think it will be also useful to others in this and other countries, engaged as I am on the various scientific problems presented by agriculture; and if it turns out useful to them, even the farmer, who looks upon this technical article as of no service to him, will—whether he knows it or not—be indirectly benefited.

"This method of mounting and using a Microscope is one that has been gradually perfected through almost daily use since 1888. I have frequently been asked to publish the details, and have so far refrained from doing so only because I found that on each new Microscope mounted I was enabled to make a number of improvements; and so long as this was the case, any description would soon be antiquated, and so become—to me, at least—only a source of annoyance. On no less than ten separate occasions has this device been remodelled to suit differing circumstances, and it now stands in five laboratories under my supervision, viz. Sydney, Moss Vale, Wagga, Bathurst, and Pymble.

"The following is a key to the illustration (fig. 34):—

a a a, architrave of a window facing the sun.
b b b, 1/4-in. runners, 4 in. wide, in which the blind *d* slides.
c c, runners for the arm *j*, which carries the camera *m*.
d, perfectly opaque blind, made of American leather or enamelled cloth running on a spring roller at the top of the window. By raising this blind the whole apparatus may be flooded with sunlight if necessary.
e, a 1/4-in. board, 8 in. wide, hanging in an inch-deep slot in the board *f*, and riveted to the blind *d*, and hence rising and lowering with the blind. This board *e* slides in the runners *b b b*.
f f, an inch board, 8 in. wide, fitted to the side of the window and receiving the board *e*, into a median slot 1/4 in. wide, and 1 in. deep in its upper edge.
g, two sliding pieces of thin ebonite, placed one behind the other, each with a diamond-shaped opening cut out in the middle. By sliding these ebonite shutters, the opening *h* can be made of various sizes.
i i, the runners in which the two ebonites *g* slide. Behind the ebonites an elongated opening is cut in the board *e*, and this opening has a ground glass sliding over it in runners similar to *i i*, but fastened to the back side of *e*. All these latter appliances are for the purpose of varying the amount and character of the light coming through the diamond-shaped opening *h*.
j, wooden arm, 1 in. thick and 3 in. wide, carrying the micro-camera *m*, and sliding in the ways *c c*, capable of being clamped by the set-screws *w*. Any position of *j* may be recorded by means of scales marked on *c c*.
k, photographic plate-holder, half-plate size, as used on an ordinary tripod camera.
l, frame into which the slide *k* is pushed, in construction similar to the back of an ordinary tripod camera.
m, leather bellows of micro-camera, capable of extension to four or five feet.

* Reprint from Agric. Gaz. N.S. Wales, March 1897.

n, wooden front or head of camera, into which the barrel of the Microscope fits. This head is hollowed out, and carries a light ebonite shutter, actuated from the outside, by means of which the exposures are made. The wire lever actuating this shutter is shown as a dotted white line near *n* (see also fig. 35).

o, mirror of the Abbe camera lucida.

p, barrel of the Microscope.

q q, vertically sliding tables, the right-hand one being used as a drawing-board when the camera lucida is in use. Being adjustable, various magnifications can be secured. The higher *q* is placed the less magnification the drawing will show. A scale drawn on the architraves enables any position of *q* to be registered. The left-hand table is similarly adjustable, and is usually kept on a level with the Microscope stage. Both these sliding tables are cut away to suit the observer's body resting on the stool *v*. Both are braced so as to be quite rigid under the weight of the arms in drawing, &c.

r, head into which the foot of the Microscope is firmly clamped by means of easily removable wooden wedges.

s, pillar bearing the Microscope, preferably of iron and planted in cement beneath the building, and coming through the floor without contact with the building. If this is not feasible, *r* may be fixed to the window-sill. In three of my laboratories the ways *c c* are fixed to iron or wooden beams, also planted in the earth and coming through the floor without contact, thus making a very perfect arrangement for long photographic exposures with high powers where all tremor must be avoided.

t t, halves of wooden hand-clamps of large size (15 in. long and 2½ in. square), grooved to slide in the ways *u u*, and carrying the well-braced tables *q q*.

u u, two wooden table-ways, firmly fastened to the side of the building.

v, stool.

w, set-screw, to clamp the camera in position.

x x, four set-screws, to clamp the tables *q q* in position.

y, opaque cloth, sewed on to a rectangular opening cut in the board *f* to admit the light to the mirror of the Microscope. Slots to hold coloured glasses are arranged on the back side of this opening, to furnish monochromatic light for photography, &c. The various substage adjustments can be worked through the cloth, which, however, can be lifted in a second when necessary.

"After having made the different features of the drawing (fig. 34) clear, it will only remain to explain some of the advantages of this system of utilising the Microscope. Having used the Microscope for purposes of investigation almost daily for nearly twenty-five years, I feel justified in calling particular attention to opinions based on such extensive experience.

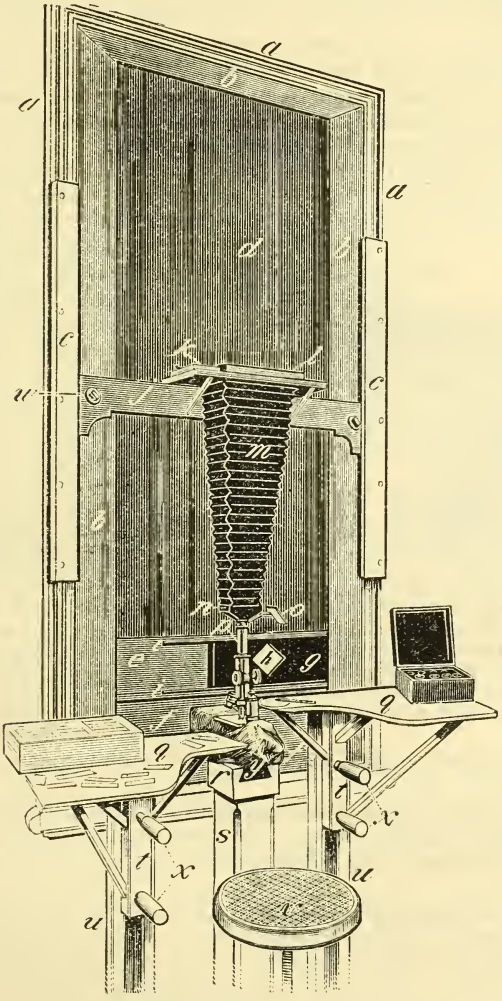
"The apparatus is adapted to the best of all lights—daylight. The perfection of the image as formed on the retina of the eye is very great; for if the room be darkened, and the blind *d* be closed, no light but that from the Microscope enters the eye. Few, even among experts, according to my observations, realise the evil effects of extraneous light when observing with the Microscope. Those who do realise this evil are usually found advocating the use of artificial light by night so as to avoid the evil. Here is a way to avoid it and still keep to the use of daylight. It need scarcely be pointed out that the cloth *y* is for the purpose of excluding extraneous light.

"The window faces the sun, so that, whenever it is desired, by simply raising the blind *d*, sunlight can be obtained on the top of the stage as well as under it.

"The whole apparatus is quite rigid.

"The arrangement for drawing with the camera lucida is of a high degree of perfection. I am often struck with the complication of the

FIG. 34.

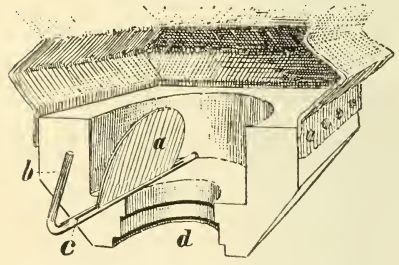


Perspective view of Microscope mounted for purposes of investigation by daylight. Dimensions may be gauged by the diameter of the top of the stool *v*, which is 1 foot.

various more or less expensive devices (always more or less imperfect too) attached to the camera lucida as now made, and having for their object the graduation of the light, so as to equalise the lights coming

from the object and the drawing-paper respectively. This object is usually accomplished by inserting between the drawing-paper and the eye a glass having the correct neutral tint. It is usually found that the exact tint required cannot be obtained, either because the properly tinted glass has been lost or broken, or never existed. In any case, such glasses are in the way, and can only have been regarded as a necessary evil. In the system here described, the light coming through the Microscope is first graduated so as to be as perfect as possible, then the diamond-shaped opening *h* is set so as to exactly equalise the lights coming from the Microscope and from the drawing-paper located on the circle under the pencil shown in the illustration. This can be done in an instant and with the utmost precision. The ground glass behind *h* serves to destroy the image of outside objects which are formed when the aperture *h* is reduced in size. The difficulty of using the camera lucida with very high powers is well known. With this system there is no difficulty; whatever can be clearly seen can be drawn.

FIG. 35.



Section of head of Microscope-camera, one-half size—*a*, vulcanite shutter; *b*, arm or lever for opening and shutting the shutter *a* (this arm is outside the head); *c*, slot into which *a* is set; *d*, rabbited opening into which the draw-tube of the Microscope fits in a light-tight manner.

"When the left-hand table *q* is arranged on a level with the Microscope stage, the moving of objects on to and off the stage is conveniently accomplished.

"The sliding adjustment of the drawing-board or table (right-hand *q*) will commend itself at once to anyone who has used a camera lucida. Already a number of patents exist on this head, but all that I have seen are lacking in stability and convenience. Here, however, the artist may lie stretched at ease, and, having so adjusted the drawing-board as to secure the desired magnification, can work with comfort and with great precision.

"The nice working of the fine adjustment is facilitated by the fact that the ball of the hand may at the same time rest on the table.

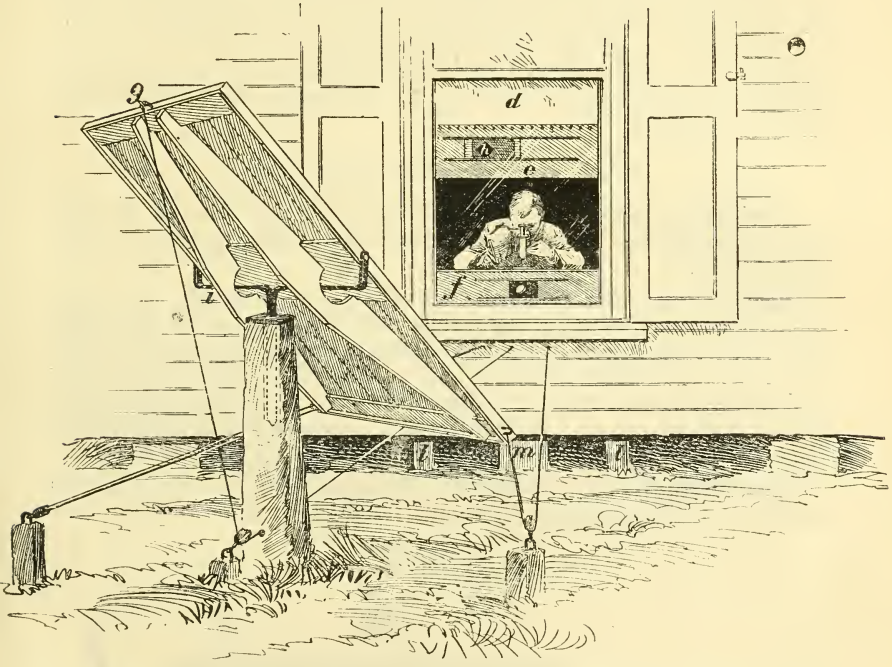
"The Microscope can be clamped in position, and is movable within limits.

"The photographic camera is in readiness for instant use, and is as rigid as possible. Being arranged on a vertical system, it is most convenient. Few, I imagine, having once fairly tried a good vertical system, will ever revert to any other. Its advantages are obvious. For instance, the stage remaining horizontal, the object does not tend to float and get out of focus; liquid backing on the plate does not flow; the focusing can be most easily and accurately done, especially when the ground glass is dispensed with, and a lens used instead; the bellows never bothers by sagging when long drawn out; and so forth, and so forth. My whole apparatus is made as low as possible, so that in focusing on the ground glass at *l* it is only necessary to stand up. If the camera has to be ex-

tended to 5 feet, it will be necessary to provide steps, as *l* thus comes up above the head of the standing operator. The focusing lens can rest on the ground glass, or swing on a vertical pivot fastened into the frame *l*, or be carried on a rack-stand resting near *k*.

"For long focusing, when it is inconvenient to reach the fine adjustment with the hand, I use a stick, the end of which carries a piece of metal

Fig. 36.



Back view of white screen having universal movements; *d*, *e*, *f*, and *h*, same as in fig. 34—that is: *d*, opaque blind; *e*, 1/4 in. board, 8 in. wide, attached to *d*, and rising and falling with it; *f*, 1 in. board, 8 in. wide, top of which is slotted to receive *e*; *h*, the same diamond-shaped opening shown in fig. 34; *g*, top of screen; *i*, forked wrought-iron spindle, shown black, except where it enters the post—the dots show its continuation; *l l*, uprights imbedded in the ground and passing up into the building without touching it, and supporting the Microscope-camera above the operator's head; *m*, pillar supporting the Microscope, imbedded in the ground, and passing up into the building without touching it. The cords and pulleys are for the purpose of moving the screen from inside the building. The small mirror mentioned in the text is placed on the front of the screen near *g*.

that fits into a slot filed in the top of the fine adjustment screw. When the adjustment is secured, the stick is removed. This convenient arrangement has often enabled me to take photomicrographs with great rapidity.

"The pillar *e* forms a model support for a dissecting stand, the tables *q q* being then placed at equal heights to serve as arm-rests.

"The light is obtained from a white screen, having universal move-

ments, placed outside in the sunlight, and workable from the interior by means of cords and pulleys. The screen is a broad frame covered with bleached sheeting. On one end of the screen is a small mirror, so fastened as to indicate, by the sun's reflection, when the screen is reflecting the maximum amount of sunlight. This light reflected from the screen is superior to the proverbial white cloud, and eclipses any artificial light.

"I feel sure that, in the right hands, the appliances I have here described, if patented, could be made a source of profit; and the moral right to so use them is hereby freely given to whomsoever chooses to accept.

"It is with much pleasure that I acknowledge the aid, during the last few years, of Mr. E. M. Grosse in executing a number of the details of this system of using the Microscope. The accompanying illustrations are from his hand."

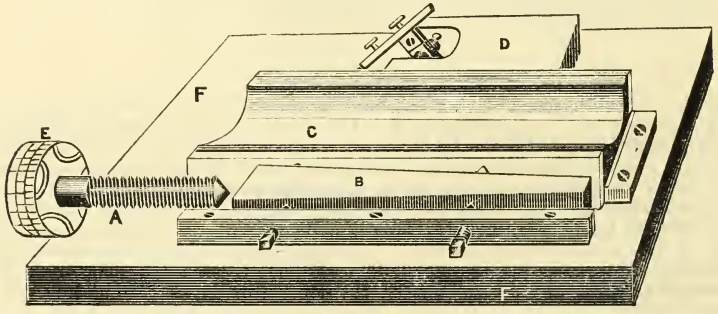
(3) Illuminating and other Apparatus.

Projection Lantern.*—M. H. Möhlenbruck describes an ordinary form of projection lantern for showing microscopic preparations and photographs.

Light-Filters and Colour-Screens.†—Dr. A. C. Stokes points out the unsatisfactory nature of the ordinary coloured glasses and fluid-cells. The best he has used is Clifford's malachite-green screen, but for continual use the light of this is trying to the eyes. For general use the author has a particular kind of blue glass (from Lovibond's tintometer) coated with a layer of "Walpole green" cement; this is pleasanter to the eyes, but does not give so good a definition as the Clifford screen.

Simple Machine for Micrometer Rulings.‡—Mr. D. W. Smith has devised a simple inexpensive machine for producing fine rulings, up to

Fig. 37.



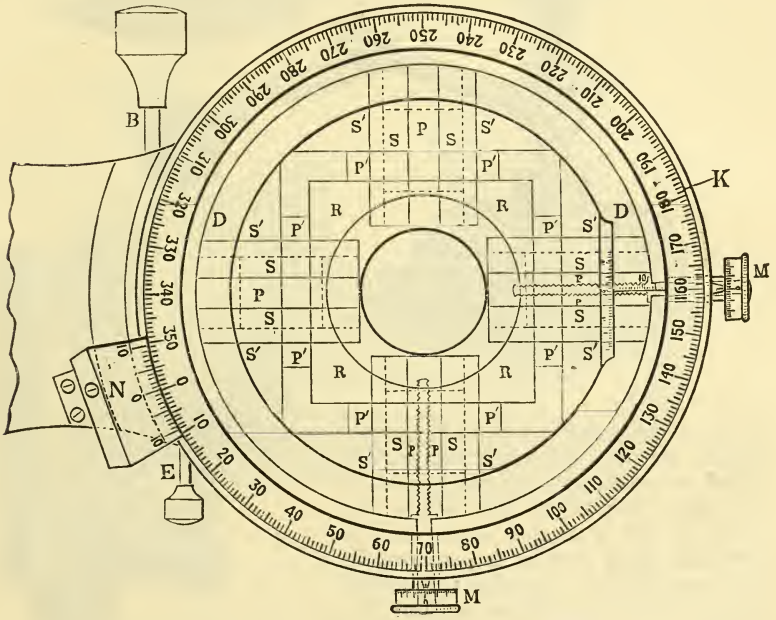
thirty or forty thousand to the inch, on glass. The micrometer-screw A (fig. 37), with graduated head E, imparts a longitudinal motion to a metal wedge B, which, for each turn of the screw, gives a lateral motion

* Arch. Sci. Phys. et Nat., iii. (1897) pp. 590–3.
† Journ. New York Micr. Soc., xiii. (1897) pp. 56–63.
‡ Tom. cit., pp. 53–5 (1 fig.).

of 1/1000 in. to the metal block C. The diamond-carrier D, which is worked to and fro by hand, is kept in contact with the block C by springs. The working surfaces are formed by slips of plate-glass. The necessary clamps and springs are not represented in the figure.

Covered Rectangular Motions for Stages.* — Dr. R. Brauns describes an improvement on the usual rectangular stage-motions when used

Fig. 38.



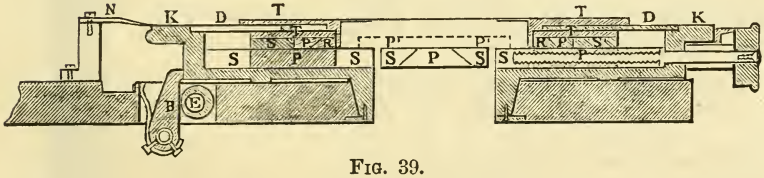


Fig. 39.

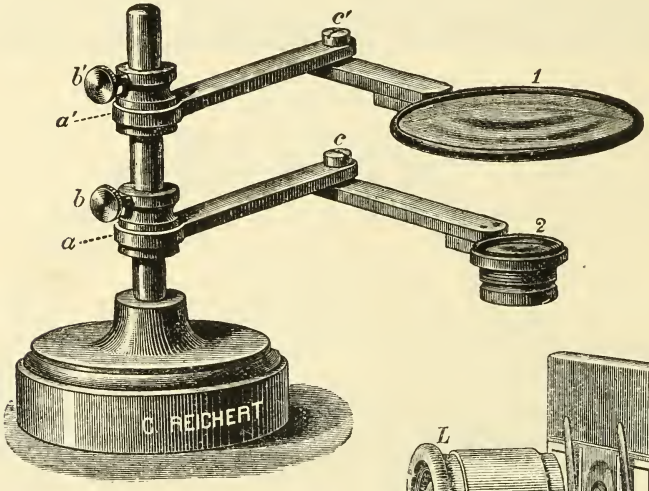
in connection with a graduated circle. The ordinary forms, in which the rectangular motions are above the circle, have the disadvantage of the graduations being often partly hidden and shadowed by the screws and slides; the mechanism is further unprotected from dust and reagents.

To the base-plate of the circle K (fig. 38) eight guides S are fixed, and between these the four prisms P move. To the last are fixed the

* Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 11–3 (2 figs.).

four prisms P′ which move between the prismatic edges of the frame R and the four guides S′. The frame R is fixed to the stage T, and to

Fig. 40.



it the rectangular movements are imparted by the micrometer-screws M which work in two of the prisms P. All this mechanism lies within the body of the graduated circle, and is protected from dust by the annulus D. The eccentric E clamps the circle, and B is the fine-adjustment of the same.

The same arrangement can also be used with non-rotating stages. It is made by R. Brunnée, of the firm Voigt and Hochgesang, of Göttingen.

Lens-Support for Examining Seeds.* —Herr C. Reichert briefly describes the lens-support shown in fig. 40, which has been made according to the designs of Dr. von Weinzierl for the examination of seeds. The arms *c* and *c′*, carrying low-power lenses, are capable of horizontal and vertical movements, and are supported on a heavy base.

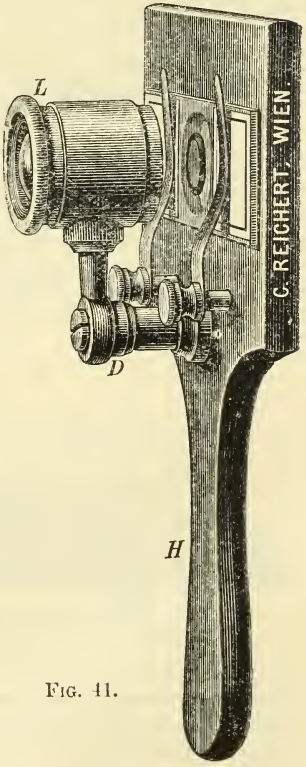


Fig. 41.

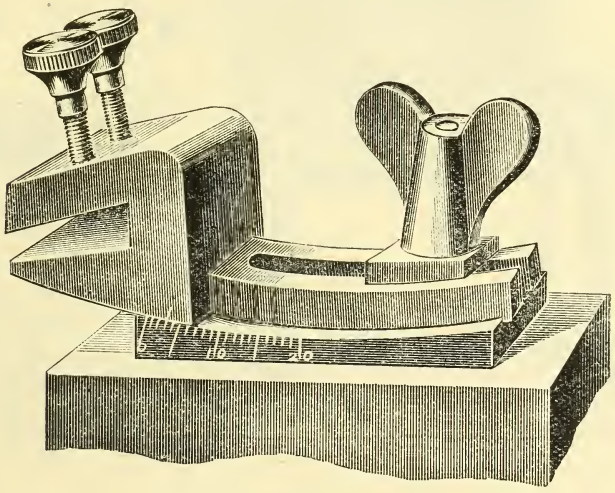
The lens-support shown in fig. 41 is mentioned here. The lens slips

* Zeitschr. f. angew. Mikr., iii. (1897) pp. 72–3 (2 figs.).

easily into a sleeve which is carried on the movable arm D, this being fixed, with the object-clips, to the ebonite hand-frame H.

Knife-Holder for Microtomes.*—Dr. R. Hesse describes a knife-holder (fig. 42), with which it is possible to vary the inclination of the knife from the horizontal position. The sliding arc has a radius of 15 cm. The graduations at the side are millimetres, 1 mm. corresponding to a movement of 0·8° of the knife. The holder is made by R. Jung, of Heidelberg.

Fig. 42.



(4) Photomicrography.

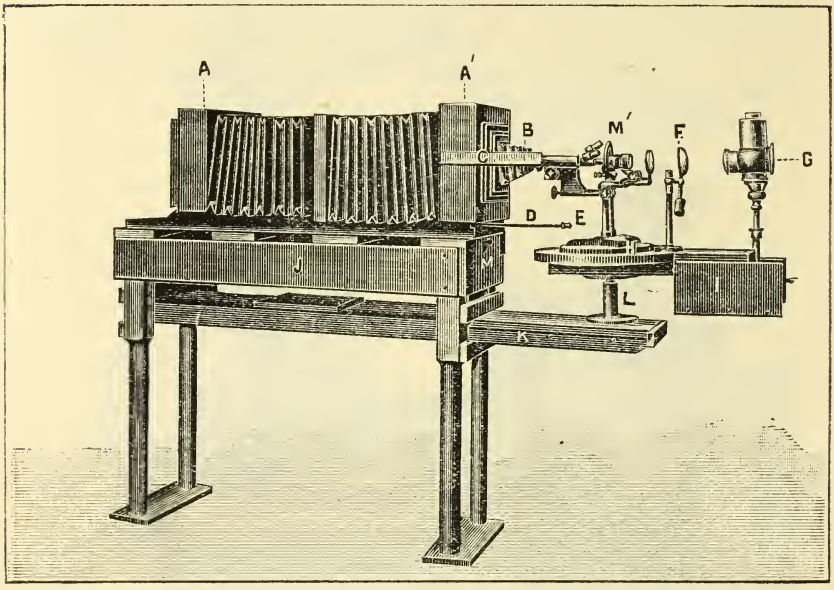
Systematic Photomicrography.†—Mr. J. B. Shearer points out that the time of exposures, &c., in photomicrography depends too often on guess-work, and suggests a systematic keeping of records of exposures which have been successful, in order to serve as a guide in future cases where the conditions are more or less similar. For this purpose he has had printed a book of forms to be filled in with the details as to lenses used, distances of the light and photographic plate, time of exposure, colour of object, &c.

The apparatus used by the author consists of a turntable L (fig. 43) carrying the Microscope, Welsbach lamp, and bull's-eye condenser. After the light, &c., has been arranged, the turntable is swung round, and the Microscope-tube is inserted in the camera front. The image is focused from the back of the camera by a rod D, which passes the whole length of the camera-bed and actuates a band passing over the fine adjustment of the Microscope. For the illumination of opaque objects, the lamp can be moved with respect to the Microscope. An arrangement, in

* Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 13–5 (1 fig.).
† Trans. Amer. Micr. Soc., xviii. (1897) pp. 117–130 (5 pls.).

which the body of the Microscope is vertical, for photographing objects in liquids, is figured. The procedure of developing the photographic plates is described.

Fig. 43.



B. Technique.*

(1) Collecting Objects, including Culture Processes.

Plankton-Methods.—Prof. J. Frenzel † discusses at some length the merits and demerits of the silk-gauze net. Even the details of the blocking of the gauze come to have importance in exact quantitative estimates of the Plankton. He recommends repeated use of hot water for cleaning the net, which he regards as fully useable when it remains constant in hot water.

Prof. V. Hensen ‡ also discusses the use of the silk-gauze net. To free it from adhering particles which tend to close the pores, the best plan is to rub it under water with a bath-sponge. This is much better than boiling, which Frenzel recommended.

Medium for differentiating the Bacillus typhosus from Bacterium coli commune.§—Dr. K. Kashida recommends the following medium for easily distinguishing between *Bacterium coli commune* and the bacillus

* 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.

† Biol. Centralbl., xvii. (1897) pp. 364–71. ‡ Tom. cit., pp. 540–42.

§ Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 802-4.

of typhoid fever :—A 1·5 per cent. agar solution, made by dissolving the agar in bouillon, is first prepared. This is then cleared up, strained, and filtered. To the filtrate are added 2 per cent. lactose, 1 per cent. urea, and 30 per cent. litmus tincture. This is then distributed into test-tubes (10 ccm.) and steam sterilised for 10–20 minutes. Cultivated in this medium, the difference as to production of acid between the two bacteria is rendered very striking. After inoculation, the medium may be poured into capsules and incubated at 37°. In from 16–18 hours, coli turns the blue colour of the medium quite red, the reaction of the condensation water being also acid. After 24 hours the medium again becomes blue, owing to ammoniacal decomposition of the urea. The colonies also turn blue, and the presence of ammonia can easily be rendered evident by touching the medium with a glass rod moistened with hydrochloric acid. When inoculated with typhoid bacilli there is no reaction to litmus, and the blue colour remains unchanged for 72 hours or more. The differences between the two may be rendered strikingly obvious by sowing them side by side in the same capsule.

Cultivating Gonococcus.—Dr. J. de Christmas,* in an interesting communication on *Gonococcus* and gonotoxin, states that he has found rabbit serum to be an excellent medium for the cultivation of this microbe, and that human albuminous fluids, such as blood-serum, ascitic fluid, or pleuritic exudation, mixed with peptonised gelose in the proportion of two to one, give abundant cultures. The preference is given to ascitic fluid, which is easy to obtain and easy to sterilise, as it will stand, without coagulating, a higher degree of heat than blood-serum. The mixture with gelose is perfectly clear, and stroke cultivations incubated at 35° develop abundantly in 24 hours. The cultures, however, die off in 3 or 4 days, and have, therefore, to be resown every 48 hours. This inconvenience is obviated by the use of coagulated rabbit serum, on which the microbe not only thrives freely but lives for at least 3 or 4 weeks. The difficulty in connection with rabbit serum is that it is obtainable only in small quantity, about 60 ccm. for one animal. For the study of gonotoxin such small amounts are quite insufficient, and the author used ascitic fluid mixed with peptonised bouillon in the proportion of 1 to 3. The bouillon was ordinary veal-bouillon, or was made with Liebig's extract (0·5 grm. to the litre). The reaction of the medium should be slightly alkaline.

Pure cultivations of *Gonococcus* were obtained by spreading a drop of the fresh pus upon the rabbit serum medium and incubating at 36°. In about 12 hours colonies of *Gonococcus* are well in advance of other organisms which may be present, and it is therefore quite easy, by once resowing, to obtain pure cultures. On rabbit serum the colonies are small, round, transparent, raised in the centre, isolated or confluent. Their chief characteristic is viscosity, which is well shown by the adhesion of the growth when touched with a platinum wire. The author explains the character of the distribution on cover-glass preparations as being due to this viscosity. It is also stated that the classic shape of *Gonococcus* always met with in pus is the least frequent in cultures.

* Ann. Inst. Pasteur, xi. (1897) pp. 609–30.

Dr. F. R. Hayner * has successfully cultivated *Gonococcus* from the fluids in joints and tendon-sheaths in the following media:—

(1) Albuminous urine agar. Acid urine containing 0·05 albumen or more is allowed to stand for 24 hours, and then boiled. The precipitate is then removed by filtration. The filtrate is again boiled, and agar, pepton, beef extract, and sodium chloride added in the proportions used for making ordinary agar. The reaction should be neutral or very slightly acid. The advantages of using albuminous urine are, firstly, that such urine contains albumens that are not coagulated by heat; and, secondly, the albumen that is coagulated acts as a clarifying agent in the removal of the salts that usually cause the cloudiness of the urine-agar.

(2) An ordinary agar-tube was melted and cooled to 46° C., and then about 5 ccm. of human blood-serum added, making the proportion one-third blood-serum and two-thirds agar. The resulting medium, which was perfectly clear, was then inoculated with three loopfuls of fluid from a joint. The inoculated medium was poured into a Petri's capsule and incubated at 37°. Colonies were observed after 48 hours.

(3) Pig-fœtus agar. This medium is prepared from fresh pig-fœtuses not exceeding 5 cm. in length, and free from placenta and membranes. The fœtuses are minced in a sausage machine, and then placed in an equal volume of distilled water, the mixture being allowed to macerate in a cool place for from 6–12 hours. The fluid is then passed through a Chamberland filter under a presssure of 150–200 lbs. Two per cent. sterilised agar is then melted and cooled down to 40°, and to it one-third of its volume of fœtus-infusion is added. The tubes are then slanted.

Protozoa Culture.†—Dr. F. Schardinger now uses a medium prepared in the following way:—To a suspension of about 30 grm. of hay in 1 litre of water, 1–1·5 grm. of powdered calcium hydrate are added, and, after having been well shaken, the mixture is incubated for 24–36 hours. The fluid is then filtered, and, the chalk having been precipitated with phosphoric acid, an equal quantity of meat infusion (made without pepton or salt) is added. The mixture is then alkalised with soda, and 1–1·5 per cent. of agar added. In this medium quite pure bacteria-free cultures of a Mycetozoon (*Protomonas Spirogyræ* Borzì) were obtained; and, if gelatin be substituted for agar, the above described fluid serves well for the preparation of a cultivation medium suitable for bacteria cultures.

(2) Preparing Objects.

Method for rapidly Examining for Bacteria in cover-glass preparations.‡—Dr. D. Kischensky recommends the following method for examining for micro-organisms in pure cultures, and also in pus, blood, urinary sediment and fæces. A drop of phenol-fuchsin solution (10 drops to 10 ccm. of water) is placed on a cover-glass or slide and mixed with a minute quantity of the culture. The cover-glass is then gently warmed and the mixed drop spread all over so as to make a film which will dry

* Bull. Johns Hopkins Hosp., viii. (1897) pp. 121–4.
† Centralbl. Bakt. u. Par., I^te^ Abt., xxii. (1897) pp. 3–5 (2 pls.).
‡ Tom. cit., xxi. (1897) pp. 876–7.

rapidly. By this procedure the bacteria are not only fixed, but deeply stained, and are quite ready for microscopical examination. For staining bacteria, in pus, fæces, and in urinary sediment, a better result is obtained by using a mixture of phenol-fuchsin and alcoholic solution of methylen-blue; for thereby the nuclei of the cells and the bacteria are stained blue, while the cell-protoplasm and degenerated bacteria are stained red.

Observing Nuclear Division in Equisetum and Chara.—Herr W. J. V. Osterhout* finds Flemming's mixture the best fixing material for observing the division of the nucleus in the spore-mother-cells of *Equisetum* (see p. 416). Microtome-sections 5 μ thick were stained with safranin, gentian violet, and orange G, and mounted in Canada-balsam.

The processes employed by Herr B. Debski † in observing the division of the nucleus in the vegetative cells and in the antheridial filaments of *Chara* (see p. 416) are described in detail. The best fixing material was found to be Flemming's mixture; and as a stain, iron-alum hæmatoxylin and Flemming's safranin-gentian-orange gave the best results. The material was left for 24 hours in the fixing fluid, then for 1–2 hours in running water; then placed for about 12 hours successively in 10, 15, 20, 30, 50, 75, 90 per cent. and absolute alcohol; and finally transferred through chloroform-alcohol and chloroform to chloroform-paraffin. The chloroform was slowly evaporated, and the material then imbedded in pure paraffin of 52° C. melting-point. Microtome-sections 5 μ thick were fixed to the slide by distilled water mixed with some albumen, and dried. The paraffin was removed by xylol, and the xylol by alcohol, and the sections were then stained.

Fixing, Imbedding, and Staining for Nuclear Division in Pollen-Grains.‡—The following are the methods employed by Miss E. Sargant for the researches described on p. 398:—

A. Fixing.—Anthers fixed in absolute alcohol were usually uncut. Those fixed in any of the three solutions given below were either halved transversely or cut at both ends to ensure penetration.

Hermann's Solution (*alcoholic*).—10 per cent. aqueous solution of platinic chloride, 3 ccm.; 1 per cent. osmic acid (aqueous), 8 ccm.; glacial acetic acid, 2 ccm.; absolute alcohol, 27 ccm. The anthers were left in this solution for 1½–2 hours, and then transferred to a 0·5 per cent. aqueous solution of platinic chloride for 24 hours. They were then placed in a 1 per cent. aqueous solution of platinic chloride for 24 hours.

Flemming's Solution (*aqueous*).—1 per cent. aqueous solution of chromic acid, 30 ccm.; 1 per cent. osmic acid (aqueous), 8 ccm.; glacial acetic acid, 2 ccm. The anthers were left in this solution for about 2 hours, and then transferred to an aqueous 0·5 per cent. solution of chromic acid for 18 hours.

Chromic acid (*aqueous*).—The anthers were laid in a 0·5 per cent. aqueous solution of chromic acid for 18–24 hours.

After treatment in any of these ways, the anthers were rinsed in water, and transferred successively, at intervals of about 12 hours, to 30, 50, and 70 per cent. alcohol, and finally left for several days in

* Jahrb. f. wiss. Bot. (Pfeffer u. Strasburger), xxx. (1897) p. 159.
† Tom. cit., pp. 229–31.
‡ Ann. Bot., xi. (1897) pp. 218–20. Cf. this Journal, 1896, p. 698.

methylated spirit, changed as it became discoloured. These changes were made in the dark when the fixing solution had contained chromic acid.

The anthers were preserved in a mixture of equal parts absolute alcohol, glycerin, and distilled water.

B. Imbedding and Cutting.—For anthers fixed in mixtures containing chromic acid or platinic chloride, paraffin melting at 52° C. is hard enough. A softer paraffin can be used for anthers fixed in absolute alcohol. Sections 15 μ thick were cut from such material imbedded in paraffin melting at 45° C.

The sections were usually floated on the slide with distilled water, and made to adhere by careful drying without cement. But in the case of anthers showing the nuclear division within the nearly mature pollen-grain, a cement was used of collodion and clove-oil.

Great care must always be taken not to overheat the paraffin-ribbon on the slide. If the paraffin approaches the melting-point, the sections will be strained and their structure distorted.

Hand-sections are apt to be broken while they are being transferred from a stronger to a weaker solution of alcohol. To avoid this, the sections were placed in a small wide-necked bottle half filled with distilled water, on the top of which absolute alcohol had been poured gently. The alcohol floated for some time on the water, and the sections sank down through solutions of gradually increasing density until they lay in the pure water at the bottom. Then the alcohol was drawn off by a pipette.

C. Staining. (1) *Flemming's orange method for material fixed in Flemming's solution, Hermann's solution, or chromic acid.*—For early stages in the development of the pollen-mother-cell, the potassium permanganate was used as a mordant both before and after the treatment with safranin. The safranin and gentian-violet solutions were also of double the usual strength for these stages. For later ones—as the first nuclear division in the pollen-mother-cell—the ordinary treatment was sufficient.

(2) *Mayer's hæmalum for chromic material.*—The sections were placed for half an hour in a 0·5 per cent. solution of ferric chloride in water, rinsed, and transferred to Mayer's hæmalum, nearly full strength. They usually took about 2 hours to stain to the right depth. If the sections were kept alkaline by rinsing in hard water and by the use of neutral alcohols, they were of a brilliant blue, and very permanent.

(3) *Mayer's hæmalum for absolute alcohol material.*—The sections were treated as above, but with a 0·1 per cent. solution of ferric chloride for half an hour, and 10 per cent. solution of Mayer's hæmalum in 0·1 per cent. solution of potash-alum for about 12 hours.

(4) *Methyl-green and acid fuchsin for alcohol material.* — These colours were used in aqueous solution; their proportion varied to suit different stages.

(5) *Renaut's hæmatoxylic eosin for alcohol material.* (See this Journal, 1896, p. 699.)

Agar as Medium for Bacteriological Examination of Water.*—According to Herr F. Hesse, agar possesses several advantages over

* Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 932–7.

gelatin as a medium for the bacteriological examination of water, and merely on the ground that it does not liquefy, he recommends it. The medium should contain 1 per cent. of agar, and care must be taken that the water in the water-bath in which the agar tubes are placed is heated up to 38°–40° before any considerable quantity (e.g. 1 ccm.) of water is added. Instead of diluting germ-full water with germ-free water, he prefers to use only so little water that not more than 200 colonies develop on one plate; and in order to avoid mistakes incidental to this method, two at least control plates are made. Cold capsules must be previously heated in the incubator. The plates should be kept turned face down, as this position delays drying considerably, and also has other advantages. Four tables giving the results of observations made with Dresden water seem to confirm the author's statements.

Fixation and Staining of Cytoryctes vaccinæ.*—Dr. v. Wasielewski inoculated the cornea of 50 rabbits and 10 guinea-pigs with vaccine lymph, and, by staining with alum-fuchsin-hæmatoxylin and alum-fuchsin, was able to recognise the appearances which have been described by Guarnieri and others. The bodies are held to be parasitic in nature, and have been named *Cytoryctes vaccinæ*. In the 25 illustrations the parasite is depicted as lying within the nuclear membrane and without the chromatin network. The parasites vary in number and size. The fixatives used were chromic acid sublimate (saturated solution of sublimate 200 + water 250 + chromic acid 0·5), picric acid sublimate (saturated aqueous solution of picric acid 1000 + saturated solution of sublimate 1000 + glacial acetic acid 50 + water 2000), picric-acetic acid, Flemming's fluid, sublimate, and sublimate-nitric acid (equal parts of 3 per cent. nitric acid and saturated solution of sublimate in hot physiological salt solution). The sections were stained with alum-fuchsin for 24 hours (fuchsin 1, alum 3, water 100), and then decolorised with bichromate of potash (a mixture made immediately before use, of equal parts of 0·5 per cent. solution and 70 per cent. alcohol. The sections were then washed in distilled water and after-stained with Ehrlich's hæmatoxylin.

(3) Cutting, including Imbedding and Microtomes.

Proper Angle of Microtome Knife.†—Dr. B. Rawitz adduces experimental proof to show that the microtome knife should be placed at an acute angle rather than at a right angle. When placed at the latter angle, the sections, according to their thickness, are always more or less crowded together, thus distorting the finer structures of the tissues cut. The experimental proof consists of the measurement of the sections cut with the knife at a right angle and also at an angle of 45°. The sections were made from a block of paraffin measuring 20½ by 11½ mm., and were 15, 10, and 5 μ thick. With the knife at the acute angle they all measured 11 mm. in breadth, while with the knife at a right angle they measured 9½ mm. for 15 μ, 9 mm. for the 10 μ, and 8 mm. for the 5 μ sections, thus showing a shrinkage of 2, 2½ and 3 mm. respectively.

Technique of Celloidin Serial Sections.‡—The procedure recommended by Dr. J. Tandler for the manipulation and treatment of series

* Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 901–13 (1 pl.).
† Anat. Anzeig., xiii. (1897) pp. 65–80.
‡ Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 36–8.

of celloidin sections is as follows:—The individual sections are lifted from the knife and arranged on slides 36 by 76 mm. No adhesive is used. When a slide is full, the superfluous spirit is removed with filter paper, and then each slide is wrapped round with a strip of filter paper which is twice the length and the same breadth as the slide. The free ends of the strip are lapped underneath the slide, and on the top is placed another (empty) slide as a sort of weight. The whole is then placed in a pan (10 by 5 by 3 cm.) half filled with distilled water. In this pan the whole lot of slides—similarly prepared—are placed, one on top of the other. Staining is effected by means of a dilute solution of hæmatoxylin, and subsequently by a 1 per cent. alcoholic solution of eosin. The paper strips are replaced by others previously immersed in the hæmatoxylin solution, and the pan half-filled with tap water. Herein they remain for 5–24 hours, according to the strength of the hæmatoxylin solution and the stainability of the objects. When sufficiently stained the strips are replaced by clean ones, and the slides left in tap water for another 24 hours. They are next transferred to 95 per cent. alcohol, and then wrapped up in strips soaked with eosin solution. After a few hours the slides are dried with filter paper, and, having been covered with fresh strips soaked in strong spirit, are transferred to 95 per cent. alcohol for 4–6 hours; after which they are treated with carbol-xylol, and finally mounted.

Imbedding of Tissues without hardening in Alcohol.* —Microscopical examination of animal and vegetable tissues which contain substances soluble in alcohol and ether is always difficult, says Dr. A. Döllken, and a method for obtaining thin sections is still a desideratum. One method, which depends on the action of aceton vapour on gum arabic, consists in fixing in chrom-osmium-acetic acid and picric acid solution. The preparation imbedded in gum is then exposed for 24 hours at ordinary temperature to the action of aceton vapour. This procedure, though possessing the advantage of not damaging certain soluble substances, does not satisfy the principal requirement, i.e. does not produce sufficiently thin sections. Another procedure is to fix in formalin and place a piece (0·5–1 ccm.) in a capsule with 10–20 per cent. formalin, and then add a sufficient quantity of resorcin and some glycerin. In an hour's time some drops of dilute sulphuric acid are added. In a short while the mass stiffens, and is sectionable in a few hours. It is fixed to the block or microtome plate with water-glass or syndetikon, and should be sectioned at once, as after a time it becomes stony hard.

Very excellent results may be obtained by imbedding in soap made in the following way:—Castor oil or stearic acid is boiled for some time with 20–30 per cent. caustic soda. After having been allowed to cool and set, the alkali is entirely removed by pressure, dialysis, or by frequently dissolving the soap. The piece (about 1 cm. high) is transferred directly from the formalin solution to a 3–5 per cent. solution of the soap made with distilled water, in which it remains for 36–72 hours in a covered vessel. It is then solidified by evaporation, by removing the cover, or

* Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 32–5.

by means of coarsely powdered Glauber's salt. The block is fixed on with water-glass. The sections, which are cut dry, roll up a little, but are easily uncurled in water. Before staining, the soap should be washed out in frequent changes of water.

Should it be desired to orient the piece, the soap-mass may be rendered quite transparent by the addition of 5 ccm. of glycerin and of alcohol to 55 ccm. of the soap solution.

Straightening of Paraffin Sections.*—Though Strasser's adhesive is indisputably the best, says Dr. W. Gebhardt, it is liable to the inconvenience arising from the difficulty of straightening the section. This may be avoided by covering the adhesive layer with water. In this way the sections are easily located and straightened. The excess of water is poured off, and a somewhat longer time than usual allowed for its complete evaporation.

(4) Staining and Injecting.

Special Procedure for Staining Bacteria on Films and in Sections.†—The procedure recommended by Herren W. Semonowicz and E. Marzinowsky is intended to stain not only bacteria but histological elements. It consists in staining the preparations with an aqueous solution of phenolfuchsin, and after-staining them with Loeffler's methylen-blue. The carbolfuchsin solution is composed of one part of the ordinary solution and two parts of water. Cover-glass preparations are placed for 2 minutes in the phenolfuchsin solution, washed in water, and then treated for 3–4 minutes with the methylen-blue solution. Sections are left in the phenolfuchsin solution for 4–5 minutes, and, after having been washed in water, stained with the methylen-blue solution for a similar time. The preparations are thereupon treated in the usual way with alcohol, oil, and xylol, and imbedded in balsam. By this method the cell-nuclei and bacteria are stained blue, while the connective tissue and the protoplasm of the cells become red or rose-coloured. The bacteria are specially well stained by this procedure unless they are degenerated, when they are of a reddish hue.

Orcein Staining.‡—Dr. H. Triepel stains elastin in small objects with the following solution, after removal from 70 per cent. spirit:—Orcein, 0·5 grm.; alcohol (70 per cent.), 70 ccm.; hydrochloric acid, 20 drops. The object, which should not be more than 2 mm. thick, is transferred after 24 hours to hydrochloric acid alcohol (alcohol 70 per cent., with 1 per cent. of acid) for about half an hour. After this the object must be dehydrated in absolute alcohol for about 12 hours. The next steps are xylol, followed by paraffin sections.

Picrocarmine.§—In an article discussing the merits of picrocarmine, Prof. P. Mayer gives the following formulæ and their method of preparation.

(1) Magnesia-carmine. Carmine 1 grm. and burnt magnesia 0·1 grm. are boiled for 5 minutes with 20 ccm. of distilled water, and the

* Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 39-40.
† Centralbl. Bakt. u. Par., I^te^ Abt., xxi. (1897) pp. 874-6.
‡ Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 31-2. § Tom. cit., pp. 18-31.

solution afterwards made up to 50 ccm. After filtration, 3 drops of formalin are added. This solution will keep perfectly clear for months.

(2) Picric acid solution is made by mixing 0·5 grm. picric acid with 100 ccm. of distilled water.

(3) Magnesia-water. 0·1 grm. of burnt magnesia and 100 ccm. of tap water are kept for a week, being frequently shaken up the while, and then the supernatant clear fluid poured off.

(4) Picrate of magnesia. 200 ccm. of picric acid solution (No. 2) are boiled with 0·25 grm. of carbonate of magnesia, and, after having settled, filtered; or 0·6 grm. of solid picrate of magnesia are dissolved in 100 ccm. of distilled water.

(5) Weak magnesia-carmine. In 100 ccm. of magnesia water (No. 3) is dissolved 0·2 grm. carmine by boiling for half an hour, after which the solution is filtered and 5 drops of formalin added.

(6) Picro-magnesia-carmine is made by mixing 1 vol. of No. 1 with 9 vols. of No. 4, or by adding equal quantities of No. 4 and No. 5 together. To either solution a few drops of formalin should be added to every 100 ccm.

Differential Staining of Tubercle and Smegma Bacilli.*—According to Herr Honsell, the safest way to distinguish between tubercle and smegma bacilli is to stain with phenol-fuchsin in the usual way, wash and dry, and then place in acid-alcohol (absolute alcohol 97, hydrochloric acid 3) for 10 minutes. After this, wash in water, and stain with alcoholic methylen-blue diluted one-half with water. The author also notices that smegma bacilli of different origin behave differently as regards their resistance to alcohol. The method given is intended for the most resistant forms, and is far better than Grethe's or Czaplewski's.

Staining of Microbes and Phagocytes.†—Dr. N. A. Iwanoff used the following staining method in his researches on phagocytosis. Blood films were made on slides or cover-glasses, and incubated for 1–1½ hours at 110°–120° C. The fixed preparations were then treated with the staining mixture for 1, 2, or 3 minutes. The staining solution was rendered more effective by heating the preparations over the flame for 2–3 minutes. The staining solution was the Roux stain for diphtheria bacilli two or three times diluted, and was composed of 1 per cent. aqueous solution of dahlia 15·0, 1 per cent. aqueous solution of methylen-green 45·0, formaldehyd 10 drops. To 20–25 grm. of the diluted Roux stain 2–4 grm. of Ziehl's carbol-fuchsin were added.

Staining Hæmatozoa of Malaria.‡—Dr. E. Marchoux stained films of malarious blood with eosin and methylen-blue, but eventually discarded this procedure for a modification of Nicolle's carbolate of thionin. The formula given is :—Saturated solution of thionin in 50 per cent. alcohol, 20 ccm.; 2 per cent. carbolic acid, 100 ccm. It is necessary to let the mixture mature for a few days until the phenol has combined with the thionin.

* Arb. a. d. Pathol.-Anat. Inst. zu Tübingen, ii. (1896) p. 317. See Centralbl. Bakt. u. Par., 1te Abt. xxi. (1897) p. 700.

† Centralbl. Bakt. u. Par., 1te Abt, xxii. (1897) p. 119.

‡ Ann. Inst. Pasteur, xi. (1897) p. 645.

Staining Reaction of Diabetic Blood.*—Dr. L. Bremer diagnoses diabetes by the following procedure:—A drop of blood is spread over a third or half a slide, which is then incubated at 135° C. for 6 to 10 minutes. The exact temperature is of the greatest importance. The slides are then stained in 1 per cent. aqueous solutions of Congo-red, methylen-blue, or Biebrich scarlet, or with the Ehrlich-Biondi stain. Immersion for 1½–2 minutes in Congo-red stains diabetic blood barely or not at all, while normal blood is coloured red. Methylen-blue acts in a similar way, but Biebrich scarlet stains diabetic, but not non-diabetic blood. Ehrlich-Biondi solution stains diabetic blood orange, and non-diabetic deep violet. Successful specimens are made by means of contrast stains; thus an aqueous 1 per cent. methyl-green for 1½–2 minutes, followed by eosin solution for 8–10 seconds, imparts a green hue to the diabetic blood, while the non-diabetic is red.

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

Preservation of Pathological Preparations.†—Herr Melnikow-Raswedenkow recommends the following procedure for mounting museum specimens:—The fresh specimen is first treated with pure formalin, and after it has been sufficiently fixed with 25 per cent. spirit, the preparation is to be mounted in solution composed of acetate of potash 30, glycerin 60, and distilled water 100.

Fixation of Celloidin Sections.‡—M. A. Gravis has devised the following procedure for fixing celloidin sections to the slide by means of agar. Three grm. of agar, chopped up very finely, are soaked in 400 grm. of distilled water for a day. The mixture is then heated in a sand-bath and, when it has boiled for six minutes, is filtered through fine muslin into little bottles with wide mouths and ground glass stoppers. To each a small piece of camphor is added. As it cools, this 0·75 per cent. agar sets firm, and, though somewhat cloudy in bulk, is quite transparent in a thin layer. When required for use the agar is melted in a water-bath and a thickish layer brushed over the slide. Upon this the celloidin sections, immediately after removal from the microtome, are deposited, and then the whole series covered with another layer of agar. When the agar has cooled, the sections will be found firmly fixed. Though it is well to allow the agar to dry for 15–30 minutes, it is not advisable to carry the evaporation too far. When all the slides required are prepared, they are immersed in 94 per cent. alcohol till the next day. This method of dehydration imparts a firm consistence to the agar. Next day the slides are stained, cleared up, and mounted.

The foregoing procedure will allow of the prolonged action of eau de Javelle, of potash, of acids, but not that of distilled water, as this softens and swells the fixative.

As vegetable sections need not always be stained, it is sufficient to

* Centralbl. f. inn. Med., June 5, 1897. See Brit. Med. Journ., Epit., Aug. 28, 1897, p. 33.

† Centralbl. f. allgem. Pathol. u. pathol. Anat., vii. No. 2. See Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) p. 818.

‡ Bull. Soc. Belge de Microscopie, xxiii. (1896–7) pp. 137–40.

treat them with eau de Javelle and, having soaked them in glycerin, mount in glycerin or glycerin jelly. For stained and permanent preparations, the sections, after removal from eau de Javelle, should be neutralised by means of an aqueous 5 per cent. solution of sodium sulphite, and the sulphite eliminated with water mixed either with alcohol or glycerin. The sections may then be stained in an aqueous or alcoholic solution, and, having been dehydrated in alcohol, mounted in balsam. The various steps in the manipulation should be carried out in cylindrical vessels which will allow the slides, placed back to back, to stand upright. The various stages require several hours apiece, owing to the slowness with which the agar parts with the previous reagent. This is really the great inconvenience of the agar fixative.

(6) Miscellaneous.

Bichromates and the Nucleus.*—Dr. E. Burchardt has made a detailed study of the diverse influence of bichromates on the nucleus. One set of salts destroy the nuclear structure, namely salts of potassium, cæsium, rubidium, sodium, lithium, ammonium, magnesium, strontium, and zinc. Another set of salts preserve the nuclear structure, namely salts of calcium, barium, and (to a less extent) copper. What is preserved is usually the chromatin, not the achromatin portion of the nucleus. All bichromates, however, seem to have two mutually antagonistic effects on the formed and unformed constituents of the nucleus. According to the predominance of one or the other effect, and according to the state of the fluid nuclear sap, there is destruction or preservation of nuclear structure, or something between the two states. The zinc salt seems in its effects about midway in the series, with sodium, ammonium, potassium, &c., on the one side, copper, calcium, and barium on the other.

Method of Graphic Reconstruction from Serial Sections.†—Mr. W. McM. Woodworth describes a method of reconstructing from serial sections by means of measurements made with an ocular micrometer directly from the sections themselves, without the aid of camera lucida outlines; and as the measurements can be multiplied to any extent, reconstructions to any scale can be produced with any combination of objective and ocular. The method, however, is practically limited to transverse sections of bilaterally symmetrical objects, though it can be applied to objects of any shape or outline if they can be provided with a plane of definition at right angles to the plane of section. The example given is the reconstruction on a frontal plane of the intestinal canal of a small Trematode at a magnification of 100 diameters. The worm is 2 mm. long, and the sections are 20 μ thick. The latter are measured with a Zeiss AA objective and ocular micrometer 3, at a tube-length of 16 mm. Under these conditions the value of one division of the micrometer is $17 \cdot 2\ \mu$. On a sheet of paper draw a line 200 mm. long to represent the chief axis of the worm one hundred times enlarged, and at right angles to this draw 100 parallel lines at intervals of 2 mm.

* La Cellule, xii. (1897) pp. 337–73.

† Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 15–8.

($= 20\,\mu \times 100$) representing the planes occupied by the sections. By means of the ocular micrometer the greatest diameter of each section is now measured, multiplied by 100, and half the resulting distance marked off on each side of the line representing the chief axis, and along that one of the parallel lines which corresponds to the section on which the measurement was made. For example :—The diameter of section 23 is 0·588 mm. (found by multiplying the value of one division of the ocular micrometer—17·2 μ—by the number of divisions covered by the diameter of the object, which, multiplied by 100, equals 58·8 mm., half of which is 29·4 mm.). This distance, then, is marked off on the twenty-third of the parallel lines and on both sides of the axial line, thus giving the diameter of section 23 multiplied one hundred times. By repeating this process for each section and then joining the points thus obtained, there results a symmetrical outline at the desired scale. The outlines having thus been fixed, to plot the intestinal tract or any organ, it is only necessary to take measurements from one margin of each section to the nearest and farthest limits of the organ desired in the reconstruction, multiply the distance by 100, mark off the results on the corresponding parallel lines, and join the points as before.

Though best suited for reproducing from transverse sections of bilaterally symmetrical objects, the method may be applied to any object of any shape, provided that a plane of definition at right angles to the plane of sectioning can be cut on some part of the object outside the organ which it is desired to reconstruct.

Penetrating Power of Formalin Vapour.* —The results of the experiments made by Herr W. A. Iwanoff relative to the bactericidal action of formalin vapour are in conformity with those of other observers. The effect of the vapour is increased by raising the temperature—that is to say, it acts more effectively at the body heat than at room temperature; and not only on superficial parts, but also on deep-lying organs. Yet it is rather slow in its disinfectant action on deep-lying parts even when used at a high temperature; for it took 3 hours to destroy fowl-cholera bacilli in quite small pieces of deep-lying organs, while 4 hours were required for Metschnikoff's vibrio, and 6 hours for anthrax, under similar conditions. When used at room temperature, it took from three to eight times as long. As might be expected, the action varies with the different organisms. A naked-eye inspection gives a pretty good notion whether the disinfection is complete or not. If complete, the piece has become a whitish-grey colour, and is of a firmer consistence than before.

Process for Soldering Aluminium in the Laboratory.†—Mr. A. T. Stanton says that it is not easy to solder aluminium by using an alloy of definite composition without a flux, and has found that cadmium iodide is more satisfactory than silver chloride. If it be fused on an aluminium plate, decomposition of the salt occurs long before the melting-point of the aluminium is reached; but the addition of zinc chloride obviates any great defects. Concentrated zinc chloride solution is mixed

* Centralbl. Bakt. u. Par., 1te Abt., xxii. (1897) pp. 50–8.
† Nature, lvi. (1897) pp. 353–4.

with a little ammonium chloride, evaporated in a round porcelain dish, and ignited at a low red heat, till a part of the ammonium chloride is volatilised. The fused chlorides are then mixed with cadmium iodide, the proportion of these ingredients being adjusted experimentally.

When the salts are completely fused together, a flux is produced which readily enables tin or other soldering alloy to unite perfectly with aluminium. The melted flux can be taken up in a pipette with india-rubber teat and dropped on to the surface to be soldered. Some powdered metallic tin is also sprinkled on the surface. The aluminium is then heated over the Bunsen flame till the flux just melts, and is then spread with a copper wire or thin glass rod. As the temperature is further raised, the flux decomposes, and the tin readily alloys itself with the surrounding surface of the aluminium. While the flux is decomposing, the tin can be spread in a continuous layer with the glass rod or copper wire.

Instead of cadmium iodide, fused lead chloride may be used.

Brownian Movement.*—Mr. K. M. Cunningham finds that the Brownian movements of particles of magnetic sand are arrested by the presence of a magnet. Some fragments of a magnetic fossil wood did not show the motion at all.

* Journ. New York Micr. Soc., xiii. (1897) pp. 64–7.

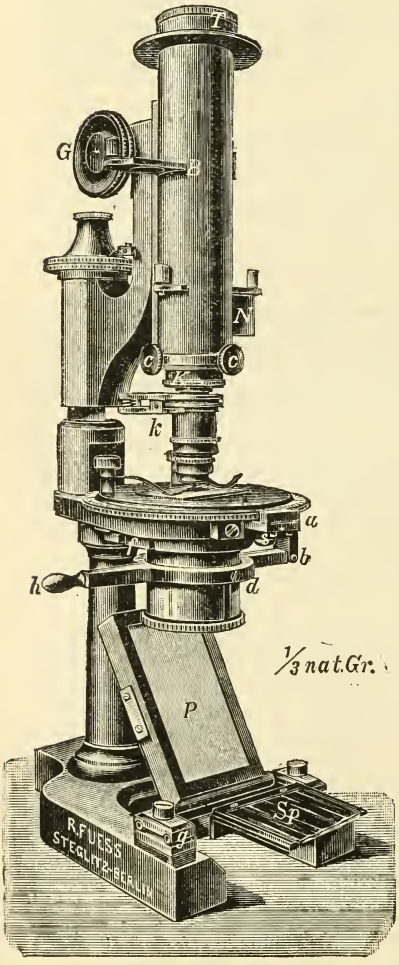
MICROSCOPY.

A. Instruments, Accessories, &c.*

(1) Stands.

New Stand, with Polariser and Large Illuminator.†—Herr C. Leiss points out that, owing to the scarcity, and the consequently high price, of Iceland spar, it is important to be able to replace the polarising Nicol prism by some other arrangement; since for convergent light it is necessary to have a Nicol with an opening large enough to correspond with that of the condensing system. It is common to see Microscopes in which the opening of the polariser is scarcely a third of that of the condenser.

Fig. 44.



In the present stand (fig. 44), made by Fuess, the polariser consists of a bundle of thin glass plates P, as in the ordinary Nörremberg polariscope. The frame holding the plates can, with the mirror *Sp*, be moved about the axis *g*, and a mark indicates the proper polarising angle. The analyser is an ordinary small Nicol N. For ordinary light a mirror may be placed above the glass plates P.

The illuminator is an Abbe's triple condenser of N.A. 1·40; it can be lowered by the lever *h*, and turned out of position about the hinge *b*, thus affording an easy change from convergent to parallel light. The front lens of the condenser has a diameter of 11–12 mm., and the lower lens one of 30 mm., those of the ordinary Fuess Microscopes being 6 mm. and 18 mm. respectively. This increase in the size of the condenser is to compensate for the loss of light by the glass plates P, and also to enable thick sections to be examined in convergent polarised light. The tube of

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

† Zeitschr. f. angew. Mikr., iii. (1897) pp. 138–41 (1 fig.).

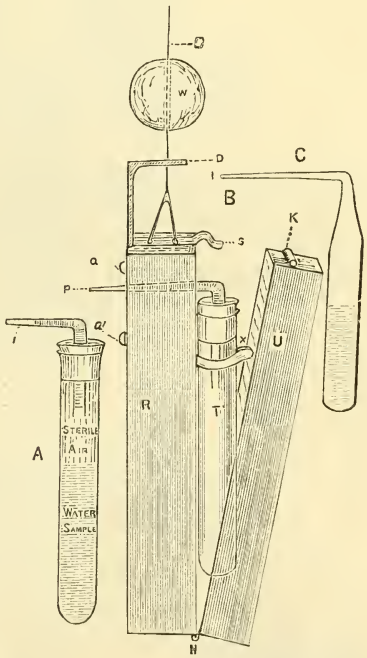
the new stand is arranged for oculars of large field. The following table gives the diameters in millimetres of the objective field with the various oculars and objectives, from which it may be seen that the second ocular almost doubles the field of view.

Objective, No.	0	1	2	3	4	5	6	7	8	9
Ordinary ocular, No. 2	3·8	3·45	2·25	1·6	1·35	0·9	0·7	0·46	0·33	0·28
Ocular No. 2 with increased field	6·0	5·5	3·31	2·5	2·0	1·5	1·15	0·7	0·55	0·4

The graduated rotating stage reads with a vernier to 5′.

Stand for the Examination of Large Sections.*—Herr E. Nebelthau describes an arrangement for facilitating the systematic examination of large sections under the Microscope. Large medical preparations can often not be conveniently handled on the ordinary stage. The stage is bridged over by a plate, on which, by means of a screw, the Microscope-tube may be moved from left to right. The stage itself may be moved backwards and forwards by a screw under the bridge. By means of these two rectangular motions, any portion of the preparation may be brought into view, and orientated by scales on the upper plate and stage. The stage and bridge are supported on a frame in which are openings for the adjustment of the mirror.

Fig. 45.



(3) Illuminating and other Apparatus.

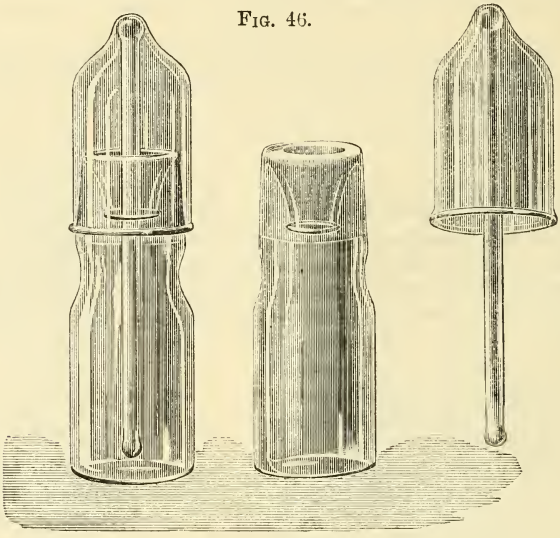
Apparatus for Bacteriological Sampling of Well Waters.†—The apparatus devised by Prof. H. L. Bolley is best explained by the accompanying sketch, and is made, except the glass and rubber parts, of brass. The body-piece R is 9 in. long by 1½ in. square. To one side of R is fitted the box U, attached by the hinge *r*, and so arranged as to close in the collecting tube T when ready for lowering into the water. The block is perforated so as to allow the passage of the tube *p* directly through the centre of the body-piece. The tube also passes through a slot in the bar

* Zeitschr. f. Instrumentk., xvii. (1897) pp. 252–3. See Zeitschr. f. wiss. Mikr., xiii. (1896) p. 417.

† [Centralbl. Bakt. u. Par., 1te Abt., xxii. (1897) pp. 288–90 (1 fig.).

d, which in itself moves vertically on the body by means of two slots fitted to the screwheads *q* and *q'*. When the weight W falls, it is guided by the copper wire *o*, upon which the apparatus is lowered, and falls

Fig. 46.

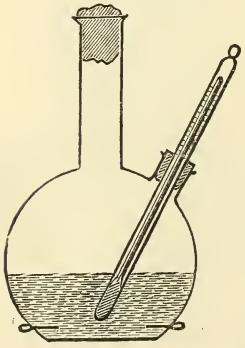


upon the bar *d*, breaking the tube *p* square across. The other parts are —*s*, a steel spring clip, and *k*, the knob which fastens R and U together.

A, fig. 45, shows the collecting tube after the water sample is properly taken; and C, another form of sampling tube. The total weight of the apparatus is about six pounds, and it is thus self-sinking.

A complete vacuum should not be made in the tube, because it is not desirable that the latter should be entirely filled with water. There is always water enough left in the small tube (A) to shut off the external air.

Fig. 47.



Bottle for Immersion-Oil and for Canada Balsam.*—Herr A. Meyer has invented a glass dropping-bottle which appears to be very suitable for immersion-oil and for Canada balsam. Its construction and appearance will be easily understood from the above illustration (fig. 46), from which may also gathered that it is dust-proof, and that if upset, the contents will not run out.

Flask for Bacteria and High Tension.—Mr. F. J. Reid sends the accompanying sketch (fig. 47) of a flask which he has found most useful with bacteria and high tension currents. Its advantage consists in its having the short neck at the side, into which a thermometer can be

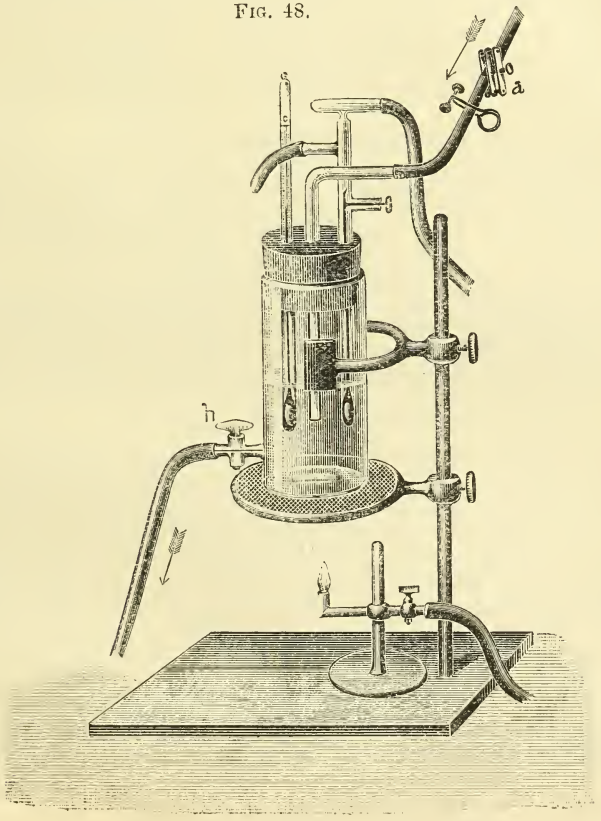
* Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 174-5 (3 figs.).

inserted. With this flask there is no danger of injury to the bacteria by heating the culture through overcharge of current; any rise of temperature is indicated by the current being cut off when this becomes excessive.

Compressorium.*—Prof. H. E. Ziegler describes his circular form of compressorium, through which a current of fresh water is made to flow, and which has already been figured in this Journal. He also describes a rectangular form constructed on the same principle, but large enough for the examination of relatively large objects, e.g. frog-larvæ and small fishes.

Heating Arrangement for Compressorium.†—Dr. R. Kantorowicz describes an arrangement for warming to a definite temperature the

FIG. 48.



water which is to be passed through Ziegler's compressorium.‡ The vessel shown in the figure (fig. 48) contains a quarter of a litre; into it

* Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 145–53 (4 figs.). Cf. this Journal, p. 759; 1895, p. 367. † Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 154–7 (2 figs.).
‡ Cf. this Journal, 1894, p. 759; 1895, p. 367; and preceding abstract.

water flows by the tube *a* as quickly as it flows out through *h* to the compressorium. As the water does not remain long in the heating apparatus, it is not deprived of much of its dissolved air. A thermometer and thermostat also dip into the vessel.

Another form of apparatus suggested by Prof. Ziegler consists of a long glass worm contained in the heating apparatus. As the water passing through this worm does not acquire the same temperature as the bath, its temperature is taken before it enters the compressorium.

New Knife-holder for Microtomes.* — Prof. S. Apáthy gives the following requirements for a perfect knife-holder for a sliding microtome:—(1) It must hold the knife quite firmly; (2) the knife-edge may be set at any desired angle in the plane of motion; (3) the knife-edge to be parallel to the plane of motion; (4) the blade of the knife may be inclined at 0°–20°; (5) the knife may be taken out of the holder and replaced again in exactly the same position. A holder satisfying these conditions—especially (2) and (4)—is described in detail. The inclination of the knife to the plane of motion is effected by means of wedges.

Cheap Condensing Lens.† — Instructions are given for making a cheap mount for a bull's-eye condenser. A spectacle maker's cataract lens is fixed by sealing wax in a loop made from a strip of metal; this is attached by a wire passing through a cork to a vertical wire on a wooden base.

(4) Photomicrography.

Simple Apparatus for Photomicrography.‡—Mr. M. J. Golden describes a simple wooden base, consisting of a long board, to which are attached a shelf to hold the Microscope, and a sliding piece with a pair of brackets to carry the camera. A heavy cloth funnel connects the Microscope and camera.

(6) Miscellaneous.

Diamond for Cutting Glass Discs.§ — Dr. C. J. Cori describes a simple and cheap apparatus for cutting glass discs of various sizes, which are always of use in laboratories. It is constructed on the principle of the beam-compasses. The fixed centre consists of a wooden cylinder of 2 cm. diameter, held to the glass by wax; about this rotates the horizontal prismatic beam, which carries a block holding the diamond. Annuli of glass may be cut with the apparatus.

B. Technique.‖

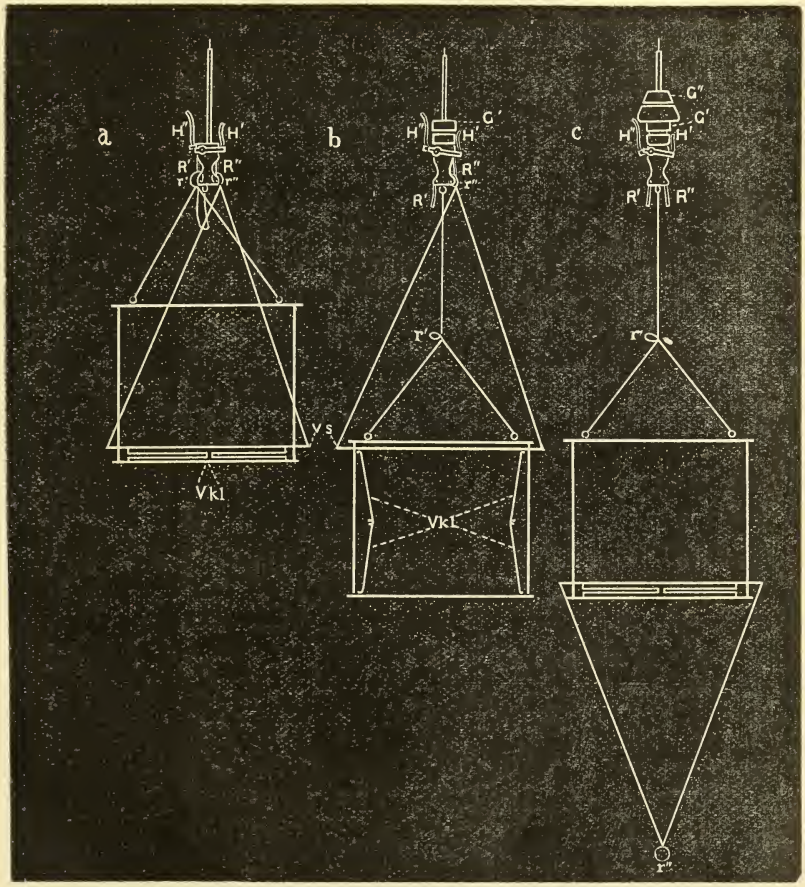
(1) Collecting Objects, including Culture Processes.

Closing Fishing Net.¶—Dr. C. J. Cori describes a form of net which may be opened at any depth, and, after being trawled for any required time, closed again, thus enabling the organisms from any depth to be

* Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 157–74 (9 figs.).
† The Microscope (Washington), v. (1897) pp. 109–11 (1 fig.).
‡ Tom. cit., pp. 103–4.
§ Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 175–7 (1 fig.).
‖ 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. ¶ Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 178–84 (3 figs.).

collected. Fig. 49 *a* shows the mouth of the net closed. By letting fall the weight G′, the lever H′ releases the bolt R′, which in turn releases the ring *r′*, and so opens the mouth of the net as shown in fig. *b*. The mouth of the net, which has a width of 30 cm., is made of hinged metal bars V*kl*, contained in the sliding frame V*s*. To close the net again, the larger weight G″ is let fall; this, through the lever H″ and bolt R″, releases the ring *r″*, when the portion V*s* of the sliding

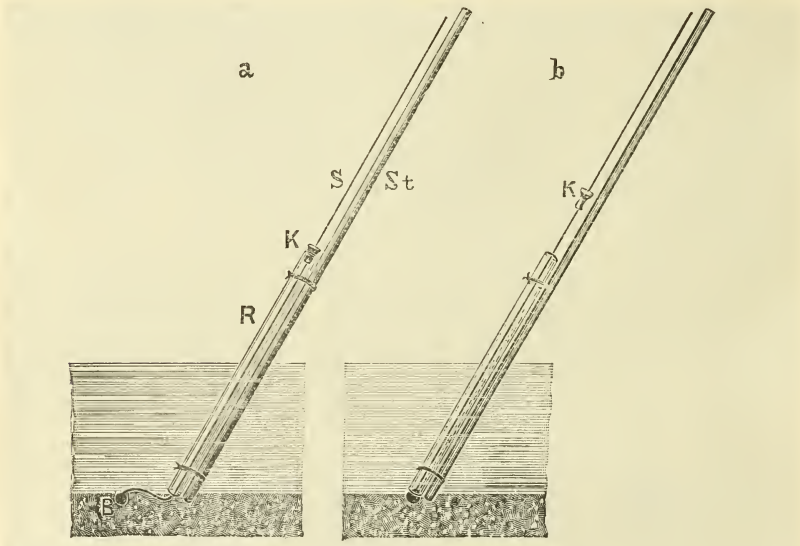
FIG. 49.



frame falls down. The weights G′ and G″ are made in two parts, so that they can be put on the cable and then bound together by wire round the grooves shown in the figure. The author has used this net in the Traun Lake in Upper Austria, and finds that the plankton is not evenly distributed in depth, but shows a distinct zoning, the depth of which varies with the time of year and day. The apparatus could be slightly modified for deep sea purposes.

Mud Collector.*—Dr. C. J. Cori describes an apparatus for collecting the fauna from the surface of mud at the bottom of ponds. The glass rod R (fig. 50) is fixed to a bamboo rod, and is provided with a tightly fitting cork K, and an indiarubber ball B. On pulling the cork out by the string S, the escape of air causes the water to rush into the tube; on further pulling the string, the mouth of the tube is closed by

Fig. 50.



the ball B. A form with a metal, in place of a glass, tube is figured. A further modification, suggested by Prof. Hatschek, for use with a cable in deep water, is described. A greater lowering of the line than shown in fig. 51 *a*, causes the projection Z on the tube to become disengaged from the weighted hook H, as seen in fig. *b*. When the line is pulled, the cork is withdrawn from the tube, and a further pull causes the tube to be closed again by the india-rubber ball, as in fig. *c*.

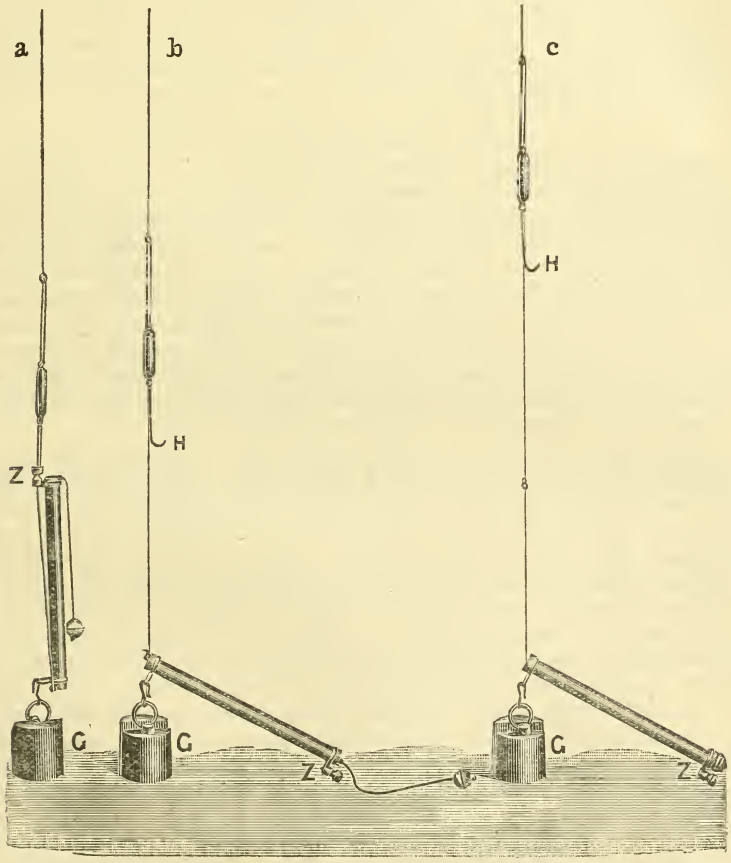
Cultivation of Amœba.†—According to Prof. P. Frosch, living bacteria are necessary for the proper growth and sustenance of Amœbæ, though certain media are more suitable than others. The one preferred is composed of 0·5 grm. agar, 90 grm. tap water, and 10 grm. alkaline bouillon. Upon this, luxuriant cultures of an Amœba derived from garden earth were constantly obtained. In these bacteria were always demonstrable, and the author's view is that successful cultivation of Amœba depends almost entirely on the bacteria supplied.

Silk-Glue as a Medium for the Cultivation of Bacteria.‡—In raw silk there exists a peculiar adhesive substance which imparts to the

* Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 184–9 (3 figs.).
† Centralbl. Bakt. u. Par., 1te Abt., xxi. (1897) pp. 926–32.
‡ Op. cit., xxii. (1897) pp. 122–4.

silk filament its firm hard character and grey colour. This "lime" is soluble in boiling water, and when cold sets to a greyish jelly, forming, according to Herr G. Marpmann, a good cultivation medium for many bacteria. Not only do air and water bacteria grow well on the silk glue, but mould fungi, and the peptonising organisms also, though the medium is but little liquefied by the latter. Owing to the fact that silk glue contains sulphur, it forms a suitable medium for the cultivation of thiophilous bacteria. The addition of salt, sugar, or pepton to the medium is not necessary.

Fig. 51.



Growth of Diphtheria Bacilli on different Media.*—Dr. G. Michel records the results of an elaborate series of experiments made for the purpose of comparing the growth of diphtheria bacilli on the following media:—Glycerin-agar, Loeffler's bullock-serum, normal bullock-serum, Loeffler's horse-serum, and normal horse-serum. Of these, Loeffler's

* Centralbl. Bakt. u. Par., 1te Abt., xxii. (1897) pp. 259–73 (5 figs.).

horse-serum proved to be the best, the second place being taken by glycerin-agar; for out of 200 cases examined, the former gave positive results in 137 instances, the latter in 122. The aspect of the growth in the five media inoculated from the same case is well shown in photographs. From these it would seem that the glycerin-agar was the best medium, though the Loeffler horse-serum runs it close.

The Loeffler's serum consists of 3 parts serum and 1 part bouillon, with 1 per cent. pepton, 0·5 per cent. salt, and 1 per cent. grape-sugar.

Egg-yolk Agar for Cultivating Gonococcus.*—Herr Steinschneider makes a nutrient medium for cultivating *Gonococcus* in the following way. An egg yolk is beaten up with thrice its bulk of sterile water. Twenty grm. of this are mixed with 10 grm. of 20 per cent. solution of biphosphate of soda and 90 grm. of 2·5–3 per cent. agar, and the mixture, having been poured into tubes, is allowed to set. On this the coccus can be cultivated directly from the purulent secretion.

(2) Preparing Objects.

Method of Preparing Anatomical Specimens.†—Dr. N. Melnikoff-Raswédenkoff places the specimens directly they are removed from the body in a solution composed of 10 formol in 100 water for 24–48 hours. Then are added 5 to 10 parts (per 100 of fluid) of sulphuretted hydrogen, or 0·5–1 part of peroxide of hydrogen. The preparation is afterwards immersed for 3 or 4 days in 60–80 per cent. of alcohol, and finally in a mixture of 20 parts glycerin, 15 acetate of potash, and 100 parts of water. To the first fixative fluid the author adds various substances, which exert some influence on the fixation of certain tissues, such as hydroxylamin, hydrochinon, pyrocatechin, and certain acetates, e.g. those of aluminium, copper, calcium, barium, and magnesium.

Preparation and Use of Klein's Fluid for Separating Minerals and Diatoms.‡—Of the solutions of high specific gravity, Klein's fluid is to be preferred, says Herr Marpmann, as its sp. gr. is 3·6, and it is not poisonous, while Thoulet's fluid and Rossbach's solution are extremely poisonous and of less specific gravity. Klein's fluid consists of boro-tungstate of cadmium, and is prepared by dissolving 1 part of tungstate of soda in 5 parts of water, adding 1·5 parts of boracic acid, and boiling until crystals of borax precipitate. The lye is inspissated until glass fragments will lie on the surface, and then 0·3 parts of a solution of barium chloride added, after which it is acidified with hydrochloric acid. In this way boro-tungstate of barium is formed, the salt separating out in tetragonal crystals, which are purified by repeated re-crystallisation. By mixing boiling solutions of the barium salt and of cadmium sulphate, cadmium boro-tungstate is obtained. This is soluble in 0·1 per cent. of water, the fluid having a specific gravity of 3·28 at 15° C. By evaporation and re-crystallisation the specific gravity is raised to 3·6 at 75°, and the salt can only be used when warm. The vessels suitable for separation of the various constituents are, a glass funnel, capable of holding 100 ccm., and having a stopcock at the lower end of the stem,

* Berlin. Klin. Wochenschr., 1897, No. 18. See Centralbl. Bakt. u. Par., 1te Abt., xxii. (1897) pp. 104–5.

† Comptes Rendus, cxxiv. (1897) p. 238. See Zeitschr. f. angew. Mikr., iii. (1897) p. 115.

‡ Zeitschr. f. angew. Mikr., iii. (1897) pp. 150–2.

or a flask of similar capacity, with a long narrow neck, which is marked off in divisions of 0·1 ccm. The powdered material and the hot solution are placed together in one of these vessels and then water added drop by drop, the vessel being carefully shaken after each drop. In this way the fluid is very slowly diluted, and the specific gravity lowered so gradually that it is possible to separate substances from one another the specific weight of which differs by 0·01 p.c., or less. As soon as the density corresponding to that of the mineral or of the organisms is reached, the particles sink slowly to the bottom of the neck of the funnel. When the supernatant fluid is perfectly clear, the tap is turned, and the sediment allowed to flow very slowly into watch-glasses. This takes from 12–24 hours, and as the fluid has cooled, more water must be added, and fresh sediments removed as they form. As diatoms consist of the same chemical substance, silicic acid, with but slight admixture of other chemical compounds, as soon as the density of silicic acid is reached, all siliceous bodies are deposited, and it might be supposed that all the diatoms will come down together. This, however, is only theoretically the case; for, as a matter of fact, the larger diatoms are deposited sooner than the smaller sorts; so that an expert manipulator has it in his power to collect the different kinds by themselves, and unmixed with other varieties. The material collected is washed with water, and further purified by sedimentation. The Klein's fluid which has been used is separated by the addition of some cadmium and by evaporation, and may be used over and over again.

Modification of Golgi's Method.*—The energetic action of aldehyds on silver salts suggested to Dr. F. Kopsch a favourable modification of Golgi's method. The fixative employed is a mixture of 40 ccm. of a 3·5 per cent. solution of potassium bichromate and 10 ccm. of formalin. After 24 hours' immersion in this fluid, the objects are removed to pure bichromate solution, and in 2–3 days transferred to 0·75 per cent. nitrate of silver solution for 3 to 6 days. Nervous tissue stains well without suffering from precipitates.

Schaper's Reconstruction Method.†—The main difference between Dr. A. Schaper's method and that of Born is that the base line of the sections is not at a distance from the section of the object, but is on the edge of the section itself. The embryo is first saturated with paraffin to prevent its drying and shrinking afterwards. It is then taken from the bath and fixed to a piece of Bristol board, after which it is sketched or photographed. It is then transferred to the bath, and a line drawn on the sketch or photo just touching the head, thus including the figure in a right angle. A similar angle is drawn on a piece of cardboard that fits into the imbedding-box. The latter is filled with melted paraffin and the embryo oriented so as to correspond with the position of the figure in the sketch. After hardening, the mass is sectioned. In sectioning, the plane of the section must be perpendicular to the median plane of the embryo. The sections should be 20 μ thick. Sketches of the magnified sections are made on paper and transferred to wax sheets; but before doing so a pencil-point is made on the dorsal side of the sketch

* Anat. Anzeig., xi. (1896) pp. 727–9.

† Zeitschr. f. wiss. Mikr., xiii. (1896) pp. 446–59 (10 figs.). See also Amer. Natural., xxxi. (1897) pp. 746–8 (2 figs.).

in the median plane, and sometimes also one in the same plane on the outline of the surface of some central organ.

The photo or sketch is then enlarged on a sheet of Bristol board, to correspond with the magnification of the sections and the enlarged figure. If only an enlarged model of the entire embryo is desired, it is merely necessary to arrange the wax sections within the Bristol board outline, and then smooth off the outer surface with a warm modelling tool. If, however, it be desired to reconstruct an internal organ, the process is more complicated, for then the second guiding-point (that on the surface of some central organ) is necessary. "In cutting out of the wax plates the outlines of the sections of the organ to be reconstructed, this point, along with that on the dorsal surface, is cut out so as each to form a point of the piece of wax that remains connected with the sections of the organs by bridges of wax." When the series of wax sections has been cut out, they are arranged in their proper places on the Bristol board, care being taken that the two guide-points fall within the plane of the Bristol board, and that the line passing through them is perpendicular to the dorsal line. When all are in place, all that remains is to smooth off the outer surface of the model.

Method of preparing Rotifers.*—M. N. de Zograf has used a modification of Rousselet's earlier method for narcotising Rotifers with cocain, and staining with osmic acid. The animals are first treated with an aqueous solution of hydrochlorate of cocain, which is added drop by drop to the specimen fluid. The methyl-alcohol used by Rousselet is continued. As soon as the animals begin to draw in their antennæ, a few cubic centimetres of dilute osmic acid are added, and in from 2–4 minutes mixed with 10 per cent. wood-vinegar (pyroligneous acid). The fluids are slowly poured off and replaced by alcohol. The animals do not lose bulk, and may be preserved in glycerin or mounted in balsam.

Demonstrating presence of Flagella of the Plague Bacillus.†—Mr. M. Gordon, working under the direction of Dr. E. Klein, began with a gelatin culture of the plague bacillus. From this a bouillon culture was made and incubated at 37°, and small doses thereof were injected subcutaneously into a guinea-pig. The animal died in two days, and characteristic organisms were found in the lymphatic glands and spleen. Plate cultivations were made from the heart-blood, and typical colonies reinoculated on oblique agar. After 20 hours' incubation at 37°, cover-glass preparations were stained by van Ermengem's method. In successful preparations rodlets are found which possess at one end a spiral flagellum about double the length of the organism. Occasionally a second spiral flagellum at the same end, but attached laterally, is present. The flagella are only stained with difficulty, apparently owing to the presence of some viscid substance by which the organisms are invested. Slight movements are visible in hanging drop preparations of agar cultures.

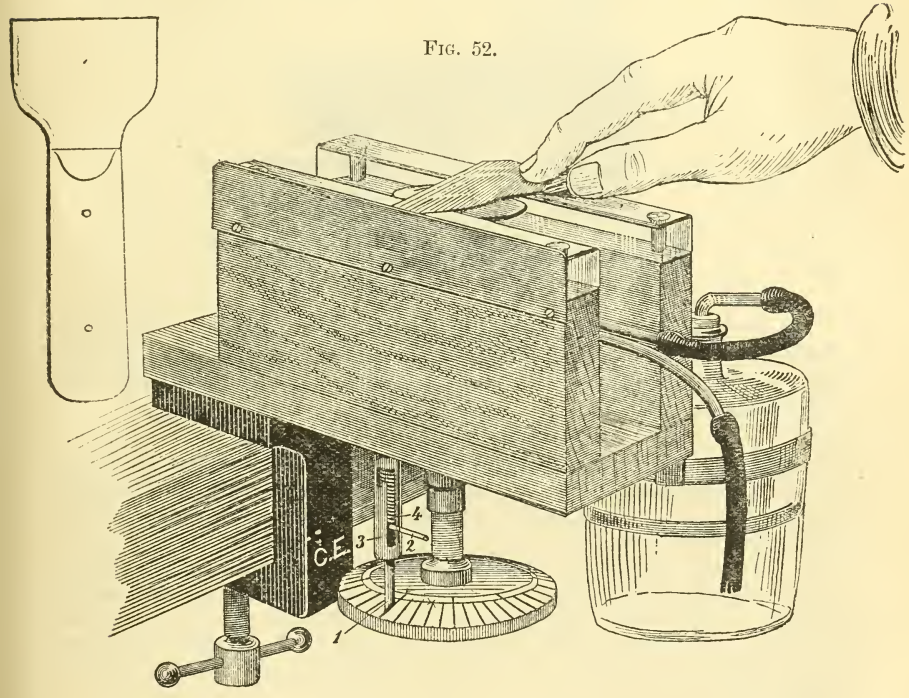
Decalcifying and Desilicating Sponges.‡—Dr. E. Rousseau decalcifies sponges which contain much lime salt, such as *Leuconia*, *Leucandra*, *Leucosolenia*, *Sycon*, &c., by first hardening pieces the sides of which are not longer than 2 cm., and then imbedding in celloidin.

* Comptes Rendus, cxxiv. (1897) p. 285. See Zeitschr. f. angew. Mikr., iii. (1897) p. 116. † Centralbl. Bakt. u. Par., 1te Abt., xxii. (1897) p. 170.
‡ Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 205–9.

The imbedded pieces are then immersed for 12–24 hours in a mixture of spirit and nitric acid (15–40 parts HNO_3 sp. gr. 1·4 and 100 parts of alcohol 85 per cent.). Each piece requires at least 20 ccm. of the fluid. The pieces are next transferred to 85 per cent. alcohol which contains some precipitated carbonate of lime, until every trace of acid is removed. They are then placed in 85 per cent. spirit, and sections made.

For desilication, sponges are treated with fluoric acid, after pieces have been imbedded in celloidin as in the previous method. Of course, all the vessels and instruments used must be made of, or covered with caoutchouc or paraffin. A piece of sponge imbedded in celloidin is placed in a caoutchouc capsule, having a lid, and containing at least

Fig. 52.



50 ccm. of alcohol. To this commercial hydrofluoric acid is added drop by drop up to 20 or 30 drops, according to the amount of silica in the sponge. The desilication takes from one to two days. The pieces are then placed in 85 per cent. alcohol containing some lithium carbonate. If there be any precipitate in the tissue, it may be subsequently removed with hydrochloric acid alcohol.

By this procedure very good sections of *Tethyx*, *Suberites*, *Thenia*, *Geodia*, *Reniera*, &c. can be obtained.

(3) Cutting, including Imbedding and Microtomes.

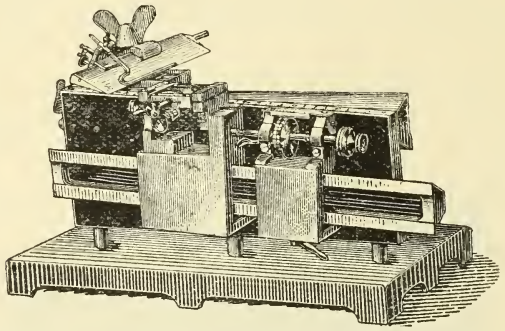
Improved Cathcart Microtome.*—Herr C. Erbe describes slight improvements in the Cathcart microtome (fig. 52). To prevent slipping

* Zeitschr. f. angew. Mikr., iii. (1897) pp. 147–9 (1 fig.).

of the knife on the strips of glass, a small metal flange projects upwards for the knife to rest against. The glass slips may be easily turned or replaced when worn. The micrometer screw gives a movement of 10 μ. The object-holder is strengthened, and a larger ether flask is provided.

Weigert's Microtome.*—Herr C. Erbe gives detailed instructions on the method of using Weigert's sliding microtome. An improvement of this instrument is introduced by making the slides double.

Fig. 53.



Microtome with Metzner's Double Support Guidance.— Herr C. Erbe, of Tübingen, has produced a microtome (fig. 53) with a double support, for which he claims increased firmness both for the object and for the knife.

(4) Staining and Injecting.

Notes on Fixation, Alcohol Method, Stains, &c.†—Herr G. Eisen highly commends the following mixture for fixing:—Platinum chloride 0·5 per cent., 50 parts; iridium chloride 0·5 per cent., 50 parts; glacial acetic acid 1 part; but has found that the iridium chloride alone is superior. The solutions are iridium chloride 0·5 per cent., 100; glacial acetic acid, 1; and iridium chloride 0·2 per cent., 100; glacial acetic acid, 1. Small objects should be kept immersed for six hours or so. A prolonged stay in the fixative is stated to be harmless. After removal from the fixative, the objects are placed for a few hours in distilled water.

According to the author the alcohol method for fixing sections to the slide is successful enough if performed in the following way. The slide is flooded with 80 per cent. alcohol. The paraffin sections are placed thereon, and removed to the shelf top or side bench of the bath, the water in which is kept at 55°. The sections at once stretch out, and then the slide must be removed to the work-table, when the alcohol is poured off, and the sections arranged. Two strips of thick blotting paper, the same size as the slide, are placed on the sections, the strip next the sections being moistened with 80 per cent. alcohol, the upper one kept dry. A roller, used with considerable force, is then passed

* Zeitschr. f. angew. Mikr., iii. (1897) pp. 169–73 (1 fig.).
† Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 195–202.

over the blotting paper, and thus the sections are firmly fixed to the slide. The sections are then brushed over with a soft large camel's hair brush, and returned to the shelf of the water-bath, on which are placed several layers of black cardboard. The sections dry in from ten minutes to an hour, and may then be manipulated with impunity, or stored away for future use.

Brasilin, says the author, is a very good stain, in many respects being superior to hæmatoxylin. It should be mixed in the same way as Böhmer's hæmatoxylin. After a few weeks the stain ripens into a deep red, with copious precipitate of blue flakes insoluble in water. The flakes are collected in a filter, and dissolved in 95 per cent. alcohol, and 15 per cent. of glycerin added. This solution is superior to the original solution, and stains nuclei a deep red.

Iron hæmatoxylin. The sections are immersed for 12 hours or more in liq. ferri sulf. oxidati diluted at least five times. Before being placed in the hæmatoxylin bath the sections should be washed for at least one minute in water. The saturated aqueous solution, which should contain 10 per cent. of alchol, is diluted for use with from ten to twenty times the amount of distilled water, and the sections immersed therein for 12 hours or more. Differentiation is then made with the same liq. ferri greatly diluted, or with 25 per cent. (or less) formic, acetic, or other acid, or with mixtures of acids and liq. ferri. In using this method it is important to remove all traces of alcohol from the sections before they are placed in the liq. ferri.

Thionin-ruthenium-red. This combination will produce opposite results according to the length of time occupied by the thionin-staining, or the age of the ruthenium mixture.

(1) Stain first for 5 minutes with aqueous 1 per cent. solution of thionin with 10 per cent. of alcohol. Rinse in distilled water, and then put on the section a few drops of ruthenium-red (made by dissolving in 80 per cent. distilled and filtered water, 10 per cent. absolute alcohol, and 10 per cent. glycerin). When the cells in mitosis exhibit a red cytoplasm and dark blue chromosomes, as observed under the Microscope, the differentiation is checked. Dehydrate with absolute alcohol, clear with pure fresh bergamot oil, and follow at once with xylol.

(2) Stain for 12–24 hours in a very weak solution of thionin (a couple of drops of 1 per cent. solution in a Naples jar of water). Rinse in distilled water, and differentiate as before with ruthenium-red. Dehydrate and clear as before. By the first procedure the chromosomes are stained blue, by the latter red, or reddish-brown, and so on.

Gum-thus. This substance is a gum from a *Pinus* indigenous to the Eastern United States ; it is dissolved in xylol ; it dries quicker, and gives clearer and better definition, than Canada balsam. It has the additional merit of being much cheaper than balsam.

Combined Method of Fixing and Staining Microscopic Preparations.*—Herr M. B. Wermel has combined the fixation and staining of blood and muscle by the use of the following solution :—1 methylen-formalin (saturated alkaline methylen-blue, 30 ccm. ; 2·5 per cent. aque-

* Medizinskoje Obosrenje, May 1897. See Centralbl. Bakt. u. Par., 1te Abt., xxii. (1897) p. 419.

ous solution of formalin, 100 ccm.); (2) eosin-formalin (eosin 1 per cent. in 60 per cent. alcohol, 100 ccm.; formalin 10 per cent. aqueous solution, 20 ccm.); (3) methylen-blue formalin (saturated aqueous solution of methylen-blue, formalin 4 per cent. aqueous solution).

Blood preparations are first dried in the air, then stained for two minutes in solution 2, the excess of stain removed, and next stained for 2 minutes with solution No. 3, after which they are washed in water and examined. For Gram's method, gentian-violet-formalin was used, i.e. 10 ccm. of 10 per cent. alcoholic solution of gentian-violet, and 100 ccm. of 2·5 per cent. aqueous solution of formalin. For staining gonococci the film need not be fixed in the flame, but merely treated at once with eosin-formalin for 2 minutes, and afterwards with a saturated aqueous solution of methylen-blue.

Double-Staining Vegetable Tissue.*—Herr H. Pfeiffer recommends a mixture of hæmalum and naphthylamin yellow for staining vegetable tissue, as it differentiates the lignified from the non-lignified tissue. The sections fixed in alcohol are placed in a mixture of equal parts of saturated aqueous solution of hæmalum and naphthylamin yellow, for 30 to 50 minutes. On removal they are washed in water for one or two minutes, placed on a slide, dehydrated, and mounted in the usual way. The ligneous tissue is stained yellow, the non-ligneous parenchymatous tissue violet.

Triple Stain for Animal Tissues.†—Herr J. L. Graberg uses the following solution for staining sections of animal tissue:—1 per cent. aqueous solution of Bordeaux red, 400 ccm.; 0·5 per cent. aqueous solution of thionin, 200 ccm.; 1 per cent. aqueous solution of methyl-green with 25 per cent. alcohol, 300 ccm. The solution is filtered, and the sections immersed therein for 24 hours, after which they are thoroughly washed in 93 per cent. alcohol to which a few drops of acetic acid have been added, until they assume a reddish hue. The sections stain best when the material has been fixed in saturated solution of sublimate in 0·7 per cent. salt solution.

Method of Staining Nervous Tissue for Microscopic Purposes.‡—Dr. Vastarini-Cresi stains the central nervous system for microscopic purposes by immersion in formalin (13 per thousand), for 2 weeks, the meninges being removed on the second or third day. Sections from 3–5 cm. thick are placed in water, or better in 40 per cent. alcohol, for 12–24 hours, and then removed to 0·75 solution of silver nitrate, wherein they may remain for an indefinite period. The preparations are afterwards treated with water and 70 per cent. alcohol. Tissue thus prepared shows well the relations between the white and grey substances.

(6) Miscellaneous.

Demonstrating the Electric Organs of the Ray.§—Herr E. Ballowitz examined the electrical organs of the common ray, *Raja clavata* L., and found that not only the nerve-endings, but other structural elements

* Zeitschr. f. wiss. Mikr., xiv. (1897) pp. 202–5. † Op. cit., xiii. (1896) pp. 460–1.
‡ Rif. Med., Feb. 14, 1896. See Brit. Med. Journ., 1896, i. Epit., 303.
§ Zeitschr. f. wiss. Mikr., xiii. (1896) pp. 462–7.

of the electrical organs, usually only seen with great difficulty, were rendered perfectly clear by Golgi's method. The preparations were stained in chrom-osmic acid (4 : 1), and, after 3–4 days, were washed in dilute silver nitrate solution, and then immersed for 1–3 days in 0·75 silver nitrate.

Concentration of Therapeutic Sera by Freezing.*—Prof. O. Bujwid has been able to obtain strongly concentrated diphtheria and tetanus sera by means of freezing. The ice is devoid of antitoxins; these, together with other constituents of the sera, remaining in solution. On freezing a bottle containing serum, the latter separates into ice crystals and a small quantity of brownish fluid. After thawing at room temperature, the contents of the bottle are found to have separated into two layers; the upper, which is quite colourless, contains only a small quantity of solid matter, and is practically water. Its antitoxic action is almost nil. The lower layer is of a yellow colour, perfectly clear, and contains all the antitoxins. After freezing two or three times, a serum is obtained which is 2½ to 3 times more concentrated than the original, so that 1–2 ccm. contain 1000 antitoxin units.

Pastes and Cements for General Purposes.†—(1) Gum arabic, 4 parts; starch, 3 parts; sugar, 1 part. Dissolve the gum in water sufficient to take up the starch, add the sugar, and heat the whole on a water-bath until the starch is completely dissolved.

(2) *Collodine* is a paste made by treating starch with water rendered strongly alkaline.

(3) *Triticine* is a paste made by dissolving equal parts of dextrine and starch in water and then heating. A little glycerin is added to make the paste pliable and elastic when dry, and a little boric acid or thymol to prevent fermentation.

(4) Gum arabic, 70 parts; water, 200 parts; aluminium sulphate, 2 parts. Dissolve the aluminium sulphate in some of the water, the gum in the rest, and mix the two.

(5) Gelatin, or best glue, 2 parts; water, 6 parts. Soak the gelatin in the water until it is soft throughout, then melt in a water-bath. Add 1–2 parts of chloral hydrate, and continue to heat gently for some time.

(6) Cover 100 parts of gelatin with water, and let stand until the gelatin is saturated. Then melt in water-bath and add 150 parts alcohol, 500 parts water, 50 parts glycerin, and 20 parts carbolic acid. This makes a very powerful cement.

Bronzing of Copper.‡—For bronzing copper, Herr Mondil gives the following procedure:—After the surface has been thoroughly cleaned, it is brushed over with a mixture composed of 20 parts by weight of castor oil, 80 parts alcohol, 40 parts soft soap, and 40 parts water. The mordant is left on the metal until it is sufficiently stained. The surface is then dried with hot saw-dust, and afterwards covered with a thin layer of varnish.

* Centralbl. Bakt. u. Par., 1te Abt., xxii. (1897) pp. 287–8.

† Amer. Mon. Micr. Journ., xviii. (1897) pp. 296–8.

‡ Centralztg. f. Opt. u. Mech., 1897, p. 4. See Zeitschr. f. angew. Mikr., iii. (1897) p. 118.

Browning Iron or Steel.*—A correspondent gives the following method for browning iron or steel to protect them from rust:—2 parts of crystallised iron chloride, 2 parts of chloride of antimony, and 1 part of gallic acid are dissolved in 4 parts of water. The object is then smeared with the solution applied with a rag or sponge, and is allowed to dry in the air. The procedure is repeated several times until the colour is sufficiently dark. The object is then washed with water, dried, and the surface rubbed with boiled linseed oil. The antimony chloride should be quite neutral.

* Centralztg. f. Opt. u. Mech., 1897, p. 4. See Zeitschr. f. angew. Mikr., iii. (1897) p. 118.