Journal of the
Royal Microscopical Society

CONTAINING ITS TRANSACTIONS AND PROCEEDINGS

AND

A SUMMARY OF CURRENT RESEARCHES RELATING TO

ZOOLOGY AND BOTANY

(principally Invertebrata and Cryptogamia)

MICROSCOPY, &c.

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Minimis partibus, per totum Naturæ campum, certitudo omnis innititur quasi qui fugit pariter Naturam fugit.—*Linnæus.*

FOR THE YEAR

1916

TO BE OBTAINED AT THE SOCIETY'S ROOMS

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VII.—*Amstutz Optical Micrometer.*

By Marshall D. Ewell.

(*Read January* 19, 1916.)

Fig. 7.

In the practice of microscopy, the ability to measure thicknesses of sections, etc., by means of a graduated fine-adjustment in parts of a micron has meant much in setting the bounds for a most exact science; likewise the use of an ocular micrometer, filar or otherwise, has been of inestimable assistance in establishing definiteness of lineal dimensions. So far, however, it seems that little, if anything, has been done to make use of a definite dimensional scale combined with the folding elements of a single-lens pocket magnifier suitable for field work, and a multitude of other purposes, when magnifications of the order of about 13 diameters are used.

Such a magnifier has been devised by Mr. N. S. Amstutz,

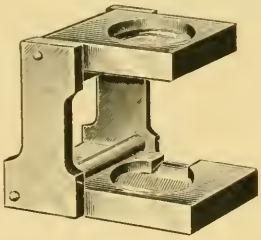

Fig. 7.

Research Engineer, of Valparaiso, Indiana. It is built much more stocky than the well-known French type of linen-tester. They are made in two forms—a $\frac{1}{2}$-in. opening and a $\frac{1}{4}$-in. opening, selling respectively at £1 and 12*s.* 6*d.*, post paid, including a leather case. The lenses are ordinary double-convex of about 0·8 in. focus. At the low magnification used it is scarcely found necessary to have an achromatic or aplanatic lens. The scales lie directly on the object being examined, hence the spherical aberration of an uncorrected lens is the same for the scale and the object, making the error of no practical moment. As the scales are in contact with the object there is no serious parallax error. The scales are ruled on brass strips which are secured on a bevelled seat sloping toward the actual centre of the lens. Different rulings

are being furnished for different trades. For instance, for photo-engravers an "E^2" scale is made having two groups of fine lines at 0·002″ (500 per in.), separated from each other $\frac{1}{10}$ in. so as to make it relatively easy to count any uniform divisions or screen-lines. Outside of and between the groups the main divisions are $\frac{1}{100}$ in., and indicated in fives for convenience of counting. Among other scales, some are ruled in metric dimensions, the smallest divisions usually being $\frac{1}{10}$ mm. It is a remarkable convenience to be able to make definite lineal measurements in this manner of microscopic objects. There is no trouble whatever to estimate the $\frac{1}{1000}$th part of an inch by means of the finest inch divisions, as it simply involves dividing one of the spaces into two parts. The sizes of half-tone dots can readily be determined. These glasses are being made at Valparaiso, Indiana, U.S., where Mr. Amstutz's son, F. W. Amstutz, is associated in their manufacture.

MICROSCOPY.

A. Instruments, Accessories, etc.*

(1) Stands.

Genevan Universal Microscope for Mineralogical Researches.†—This type of Microscope (figs. 8, 9, 10) has been designed to afford a means of making on the same instrument observations the most diverse. At the same time the stand is of a very complete construction, and its adjustments so rigorous as to give an observer entire confidence in his results. The Microscope can be used with the system of permanent centring, or of optional centring. In the former case the objective becomes an adjunct of the stage, and in this form the instrument will adapt itself very conveniently to researches in polarized light on the constitution and properties of minerals. In the system of optional centring the objective becomes an adjunct of the tube, and the Microscope can be easily changed into this form from the other without the removal of even a single screw. In the optional form the instrument would be used for all researches requiring Fédorow's methods, as well as for those requiring long-frontal distances; for such classes of observations, its completely rotating stage, and the facility of adjusting the stage-level, render it particularly convenient.

When used with permanent centring (fig. 8), the objective becomes a part of the stage, and the Bertrand lens, V, is placed at the lower extremity of the tube by means of the forceps *p*. The objective is maintained by the usual pinch-grip on the slide-arm *c*, which itself is connected by the screw with the slide-block C. The milled head B controls the coarse-adjustments of the objective, and the fine-adjustments are performed by the screw *b* with its graduated drum. The construction of the arrangement for measuring thicknesses automatically overcomes all play of the screw, and the use of the double slide-block C,c_1 protects the micrometric screw from over-rapid use. The complete object-carrier can, by means of the knob *d* mounted on a large looped helix, be rapidly applied to the stage or removed therefrom.

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

† Société Genèvoise pour la construction d'instruments de physique et de mécanique. Genève. Special pamphlet, 8 pp.

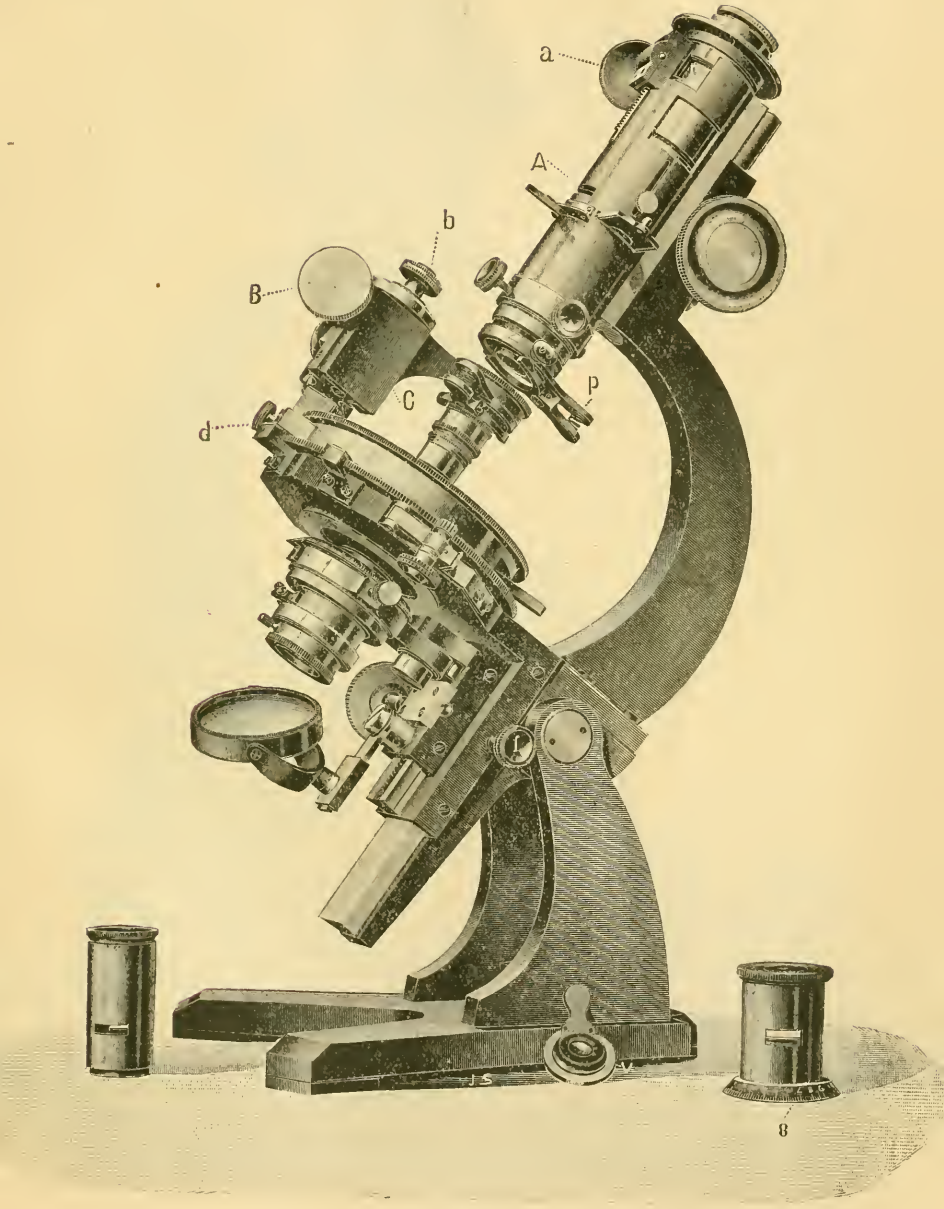

Fig. 8.

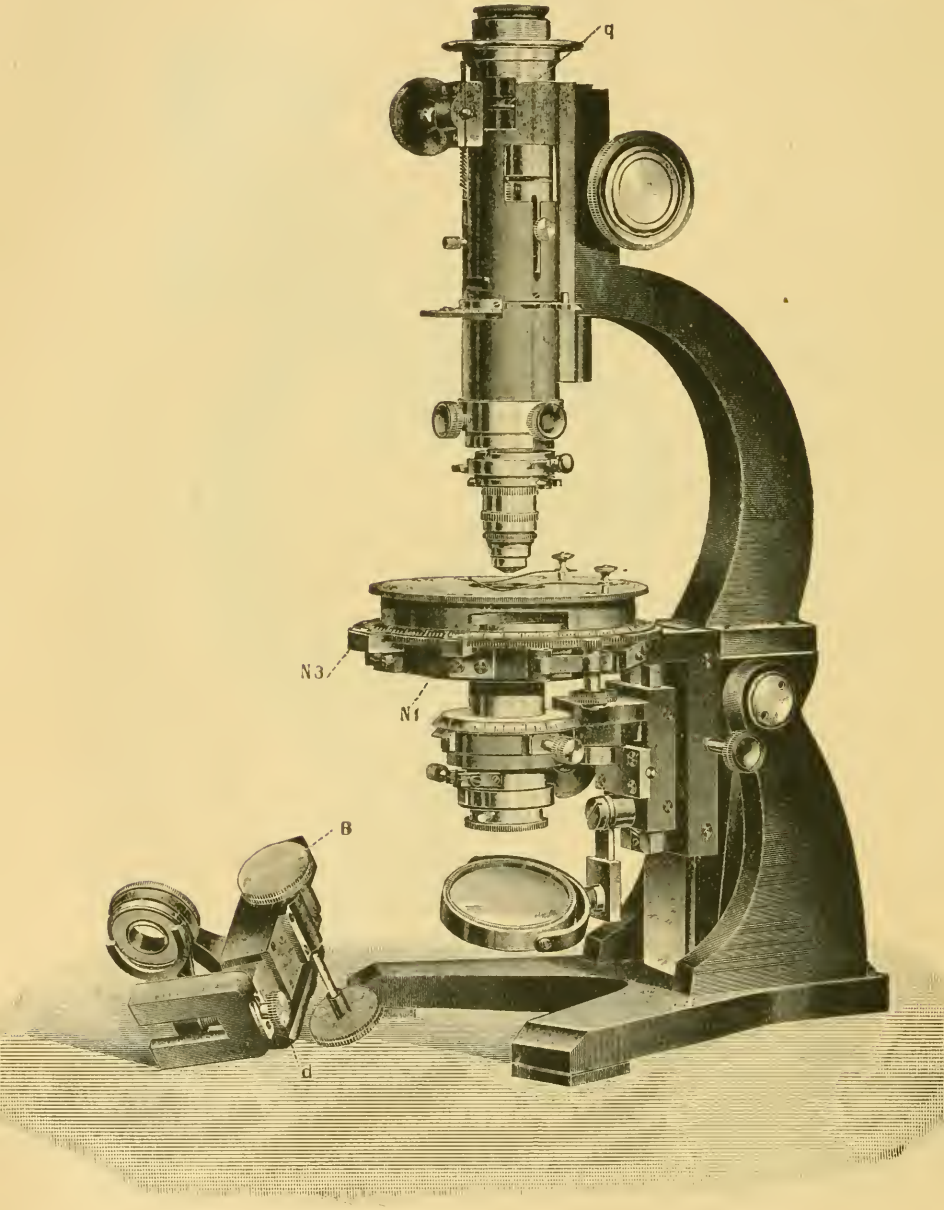

Fig. 9.

The object-carrier engages in a very exact gearing, which secures that the optical axis of the Microscope passes exactly through the rotation-centre of the stage.

When used for optional centring and for Fédorow's stage (fig. 9), the object-carrier is removed, and the objective is fixed by the pinch-grip p : the Bertrand lens B now passes to A. To fix the Fédorow stage the Microscope stage is brought to zero (Vernier n_1) ; the apparatus is placed so that the axis of rotation y_1 is perpendicular to the plane of symmetry, and the limb is on the right. By means of the fixation screws this axis can be set parallel to the corresponding thread of the network without the necessity of turning the stage of the Microscope. The rings and the central lens V are in the same plane. The height of the Microscope stage will be regulated by the consideration that the tube should be supported by its middle part. The polarizing and analyzing nicols are movable : the divisions 0 and 90°, on their graduations, correspond to the fundamental positions of the principal sections, crossed or parallel, according to the plane of symmetry of the Microscope or its perpendicular. The stage, mounted on a guide-block, can be raised or lowered for the use of long frontal distances, and is firmly and solidly fixed by help of the knob l. At the height of the focal plane of one of the negative oculars the tube is pierced with an opening q at 45° of the plane of symmetry analagous to that which is situated above the clip p. This aperture can be screened if desired ; a similar opening is pierced in the corresponding ocular. The displacements of the ocular apparatus, which is controlled by a rackwork d, can be read off on a graduation contrived on the tube. In the opening A a Bertrand lens can be inserted whose focus is independent of the position of the tube. The condenser can be removed if desired. The stage carries two verniers, n_1 and n_2, whose graduations are in the same sense, and whose zeros are distant by 135°, in addition to a mark n_3, whose index is at 45° from the zero of n_1. The adjustment is then finished, and the apparatus is ready for work.

The new model (fig. 10) of Fédorow's stage is constructed as follows : The axis y_1 is a part of the same metal piece as that forming the exterior disk x which carries the vernier m. The rotations of the axis y_1 are measured on j, whose limit is doubly divided from 0 to 180°. A second ring y_2 graduated completely can turn on the disk x, and carries the trunnions of a second axis H, which is usually placed perpendicular to y_1, and which is served by the vernier m. The central disk t can be sloped by rotation around H, and its inclinations measured either with the circular arcs W mounted on hinges on the ring y_2 perpendicularly to the axis H, or by sight on the borders of the disk t and read off on the vernier j, after the axes H and y_1 have been brought into coincidence at the zero of the vernier m. The mount of the central glass V can be turned in the disk t, and its position is noted by the divisions of t. This lens bears a cross whose centre is at 0·15 mm. below the crossing-point of the axis y_1 and H. This point is also common to the rotation axes of the disks y_1 and t. When the adjustment is once finished the Microscope axis should also pass through this point.

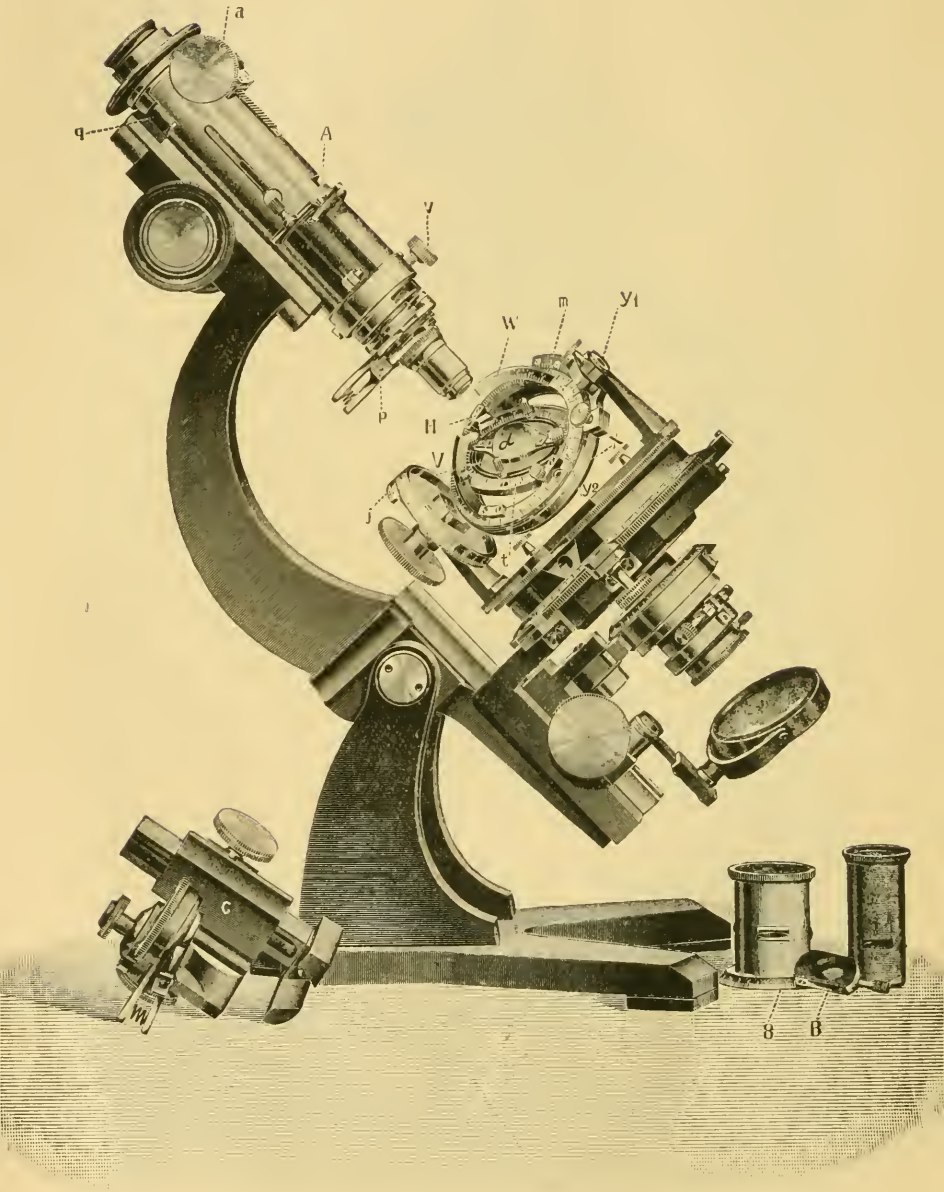

Fig. 10.

(3) Illuminating and other Apparatus.

Artificial Daylight for the Microscope.*—During the last two years H. P. Gage, working in the laboratories of the Corning Glass-works, has developed a glass filter which renders the light from a nitrogen-filled tungsten lamp almost exactly like daylight. An energy-diagram of the tungsten lamp shows very great divergences from that of the solar spectrum, especially in the green, yellow and red; but when the tungsten-lamp light is passed through the author's glass (Daylight glass, G. 172 *c*D) the curve approximates very closely to that for sunlight, especially between wave-lengths 0·45 μ and 0·65 μ—i.e. in the region of the visible spectrum giving the greatest amount of useful light. The light filtered through the *daylight* glass has been very critically tested in the author's laboratory on microscopic objects stained with many different dyes, some of them even with several dyes on the same specimen. To make sure that the Microscope itself in no way modified the colour values apochromatic objectives and compensation oculars were used, as well as the achromatic objectives and Huyghenian oculars. The experiments were conducted near to a window so that comparison with actual daylight could be readily obtained. The author states that it was impossible to detect any difference between the artificial and the natural effects. In practice it was found desirable to have the *daylight* glass finished with the ground or velvet surface on one or both sides, and to place it in the opening of an opaque screen between the artificial light and the Microscope. With this arrangement of the light the effect is like that from a white cloud.

B. Technique.†

(1) Collecting Objects, including Culture Processes.

Cultivation of Plasmodia of Badhamia utricularis.‡—In continuation of previous notes § on artificial feeding of plasmodia of this species A. E. Hilton reports ‖ that plasmodia cultivated on bread and water, with the addition of a mixture of ammonium and calcium phosphates and cane sugar, formed spores in due course. A growth was started on Feb. 19th last year by reviving a fragment of sclerotium. For some weeks, owing to low temperatures, development was slow: but on the weather becoming warmer, it increased considerably; and on May 5th, when the atmosphere became close, with a thunderstorm impending, the plasmodia changed into a quantity of sporangia. There were, however, striking differences between these sporangia and those produced in natural conditions. They were similar in shape, but instead of being of the usual cinereous hue, they were mostly a dark purple

* Science, n.s. xlii. (1915) pp. 534–6 (1 fig.).

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

‡ See also this Journal, 1915, p. 191.

§ Journ. Quekett Micr. Club, 1914, xii. 381.

‖ Journ. Quekett Micr. Club, 1915, p. 585.

black; others being of a cinnamon brown colour, and some of a pale biscuit tint. All were sprinkled with white cyrstalline particles. The sporangium walls, usually very thin and fragile, were hard, thick, and chippy. There was no distinguishable capillitium. Stranger still, the sporangia were only about half the ordinary diameter; in other words, about one-eighth of the usual size. The spores, generally bright brown and spinulose, were smooth and almost colourless: but were of the usual dimensions, if not, on the average, slightly larger; and in all other respects appeared to be perfectly normal. The characters on which the classification is based were thus altered in nearly every particular, the only permanent feature being apparently the specific spore-plasm. It was subsequently found that the spores germinated freely in the usual way; but the formation of plasmodia from the resulting swarmpores was not observed, further developments being prevented from an undue growth of fungus and large numbers of infusoria. In cultivating plasmodia on bread when the natural food, *Stereum hirsutum* or *Polyporus versicolor*, is not readily obtainable, the following mixture has proved very satisfactory: Cane sugar, ¼ oz.; ammonium phosphate, 60 grm.: calcium phosphate, 60 grm.; white starch, 30 grm.; dilute sulphuric acid, 10 drops; water, 1 pint.

Artificial Cultivation of Hansen's Bacillus.*—H. Bayon, in answer to criticisms by Fraser and Fletcher with regard to the cultivation of *B. lepræ*, states in reference to transmissions to animals that "whether one injects ground-up nodules or Kedrowsky's culture, in the great majority of cases the bacteria get simply eliminated without leaving any visible trace. In single rare instances they produce bacillary deposits similar to those found in the inner organs of lepers." "We cannot expect skin lesions in animals inoculated with leprosy; all we can hope for are discrete deposits in the inner organs. If they can be transmitted through some generations and persist for a considerable time, and the bacillary deposit is superior to the quantity injected, then it seems to me that by all the laws of experimental medicine the inoculation has succeeded. This is the case with one experiment fully described in my paper."

He further maintains that "it cannot be too often repeated that the scanty positive results obtained in the experimental study of leprosy are absolutely in keeping with what we know of the clinical features of the disease, its low and eminently capricious infectivity; but that here more than when dealing with any other disease, the partial and incomplete interpretation of hundreds of negative observations cannot invalidate the proof positive of a single successful inoculation."

(5) Mounting, including Slides, Preservative Fluids, etc.

Venetian Turpentine Method: a Substitute for the Glycerin and Glycerin-jelly Methods.†—C. J. Chamberlain remarks that the great practical advantages of the method are that preparations are as hard and

* Ann. Trop. Med. and Parasit., ix. (1915) pp. 535–8.
† Journ. Micrology, 1916, pp. 8–12.

durable as balsam mounts, and that a much greater variety of staining is possible than in the case of glycerin mounts. After fixing and washing in water, the general outline of the method is as follows:—(1) 10 p.c. glycerin until concentrated. (2) Wash the glycerin out thoroughly in 95 p.c. alcohol. (3) Stain; use stains dissolved in about 90 p.c. alcohol. (4) Wash in 95 p.c. alcohol, and complete the dehydration in 100 p.c. alcohol. (5) 10 p.c. Venetian turpentine in an exsiccator until the turpentine becomes thick enough for mounting. (6) Mount in the Venetian turpentine.

While this is the general outline, it is not sufficiently definite for a working introduction. The following concrete examples, describing the use of Venetian turpentine with an aqueous stain, with an alcoholic stain, and with a combination of aqueous and alcoholic stains, will be more practical than general directions. The steps from fixing to mounting, as used with an aqueous stain, will be described first, since this will introduce the method in its least complicated form.

Haidenhain's Iron-hæmatoxylin.—Using *Spirogyra* as a type, proceed as follows:—(1) Fix 24 hours in chromo-acetic acid: 1 p.c. chromic-acid, 70 c.cm.; glacial acetic acid, 3 c.cm.: water, 90 c.cm. The volume of the fixing agent should be at least 100 times that of the material to be fixed. (2) Wash in water, 24 hours. (3) 2 p.c. aqueous solution of ammonia sulphate of iron, 2 hours. (4) Wash in running water, 20 minutes. If running water is not available, wash in a large quantity of water and change frequently. (5) Stain over night, or 24 hours, in ½ p.c. aqueous solution hæmatoxylin. (6) Wash in water, 20 minutes. (7) 2 p.c. aqueous solution of ammonia sulphate of iron, until the stain is satisfactory. This can be determined only by examining frequently under the Microscope. (8) Wash in water, 2 hours. If this washing is not thorough, the continued action of the iron-alum will cause the preparations to fade. (9) Transfer to 10 p.c. glycerin, and allow the glycerin to concentrate until it has the consistency of pure glycerin. It is not necessary to use an exsiccator. Merely put the glycerin into shallow dishes, and leave it exposed to the air, but protected from dust. If the material is in Petri dishes, or other dishes with a large surface, three or four days will be sufficient. This process should not be hastened by warming. (10) Wash out the glycerin with 95 p.c. alcohol. It will be necessary to change the alcohol several times. From 10 to 20 minutes will be sufficient if the alcohol is changed frequently. (11) Complete the dehydration in 100 p.c. alcohol: 10 minutes should be sufficient. (12) *Most failures are now ready to occur.*

From the absolute alcohol the material is transferred to a 10 p.c. solution of Venetian turpentine in absolute alcohol. The turpentine thickens as the alcohol evaporates, and when it reaches the consistency of pure glycerin the material is ready for mounting. *The 10 p.c. Venetian turpentine is very sensitive to moisture*, and most failures are due to this characteristic; consequently the concentration cannot be allowed to take place with the turpentine exposed to the air of the room. Use an exsiccator. This will not only absorb the moisture from the air, but will soon remove the alcohol from the turpentine mixture. Make

an exsiccator as follows: place a saucer full of soda lime (sodium hydroxide with lime) on a plate of glass, and cover with a bell-jar. This is a simple and effective exsiccator. Instead, you may simply scatter soda lime in the bottom of any low museum jar with tight-fitting cover. The saucer of soda lime may be placed on a smooth board and covered with a perfectly tight box. You may improvise other forms; the essential thing is to provide a small air-tight space in which the soda lime may work. Instead of soda lime you may use fused calcium chloride or the white sticks of sodium hydroxide. Paint the exsiccator black, or cover it with black paper, or in some other way shut out the light. Many stains are weakened by light.

We are now ready for the transfer from absolute alcohol to the 10 p.c. Venetian turpentine. *Make the transfer quickly.* Pour off the absolute alcohol and place the dish, with the material, in the exsiccator: then pour on the 10 p.c. turpentine, *and immediately put on the cover.* This is better than to pour on the turpentine and then try to get the dish well placed in the exsiccator. The greater the surface of soda lime exposed, the more rapid will be the concentration of the Venetian turpentine. The concentration must not be *too* rapid. Not less than two days should be allowed for the concentration of 30 c.cm. of the turpentine in an ordinary Minot watch-glass. Great care must be taken not to let any of the soda lime, or other drier, get into the turpentine. As soon as the turpentine has attained the consistency of pure glycerin, it may be exposed to the air without any danger from moisture; but the turpentine would soon become too thick for mounting. If the turpentine has become too thick, thin it with a few drops of absolute alcohol, or with 10 p.c. or any thin solution of Venetian turpentine.

Mount the material in a few drops of the Venetian turpentine, and add a cover. Square covers may be used, since it is entirely unnecessary to seal the mounts. Such mounts are as hard and durable as balsam mounts. Material in the thickened Venetian turpentine, if not needed for immediate mounting, may be put into small vials or shells, where, kept out of the light, it can be kept indefinitely. We recommend a No. 4 shell. The corks should be of the best quality, otherwise the turpentine will become too thick. While it can be thinned by adding thin turpentine, it is better, for easy mounting, not to let the turpentine become too thick.

Magdala Red and Anilin Blue.—Fix in chromo-acetic acid and wash in water, as described in the previous schedule. Transfer from water to 10 p.c. glycerin and allow the glycerin to concentrate. It is not necessary to use an exsiccator, since there is no danger from moisture in the air. When the glycerin attains the consistency of pure glycerin, wash the glycerin out with 95 p.c. alcohol and then proceed with the staining. (1) Stain in Magdala red. At least two Magdala reds are sold by dealers. The one marked *echt* is more expensive, but, in our experience, is inferior to the one marked simply Magdala red. Make a 1 p.c. solution in 90 p.c. alcohol. We use the stain much stronger than recommended by Pfeiffer and Wellheim. This solution, diluted with an equal volume of 95 p.c. alcohol and allowed to act for 24 hours, does not stain too deeply. (2) Rinse the material for a minute in 90 p.c.

alcohol. (3) Stain in anilin blue, using a 1 p.c. solution in 90 p.c. alcohol, diluted with four times its volume of 90 p.c. alcohol. We prefer to make a fresh solution every time we have anything to stain. It is not necessary to measure it. A little of the powder—about half the bulk of a grain of wheat—in 30 c.cm. of 90 p.c. alcohol, will give an efficient solution. The time required for successful staining will vary from 3 to 30 minutes. Do not put all the material into the anilin blue at once, but, by trying a few filaments at a time, find out what the probable periods may be. (4) Rinse off the stain in 90 p.c. alcohol, and then treat for a few seconds in acid alcohol (1 very small drop of HCl to 30 c.cm. of 90 p.c. alcohol). The acid alcohol fixes and brightens the anilin blue, but extracts the Magdala red. If the anilin blue or the acid alcohol acts for too short a time the blue will be weak: if they act too long, the red is lost entirely. If the blue overstains too much, wash it out in 95 p.c. alcohol. If the red overstains, wait until the mount is finished, and then reduce the red by exposing the slide to direct sunlight. (5) Absolute alcohol, 5 or 6 seconds. (6) Transfer *quickly* to 10 p.c. Venetian turpentine and proceed as in the previous schedule.

The surprising beauty of successful preparations will compensate for whatever failures may occur. Nuclei and pyrenoids should show a brilliant red, while the chromatophores and cytoplasm should be dark blue. The cell walls should show a faint bluish color.

Haidenhain's Iron-Alum Hæmatoxylin and Eosin. — Follow the schedule for iron-hæmatoxylin until the glycerin has been washed out in 95 p.c. alcohol. Then stain for a minute in a solution of eosin in 95 p.c. alcohol. Wash for a minute in 95 p.c. alcohol, then a minute in absolute alcohol, and then transfer to the 10 p.c. Venetian turpentine. Other stains may be used. Aqueous stains should be used before starting with the 10 p.c. glycerin. Alcoholic stains should be in strong alcohol—about 90 p.c.—and should be applied just after washing out the glycerin. This method is equally good for filamentous fungi and also for the prothallia of *Equisetum* and ferns, for delicate liverworts and mosses, and similar objects.

(6) Miscellaneous.

Concentration of Malaria Plasmodia.*—C. C. Bass and F. M. Johns have devised a method of concentrating malaria parasites, the fundamental principle involved being that the malaria plasmodium with its host erythrocyte is larger than the non-parasitized blood-cell, and that when centrifuged at the proper speed for a sufficient length of time, the larger cells rise to the top of the cell column, while the smaller cells collect beneath. The leucocytes, being still larger, rise to the surface of the tube.

Measure 0·2 cm. of citrate-dextrose solution (50 grm. sodium citrate and 50 grm. dextrose in sufficient distilled water to make a volume of 100 c.cm.) into a large tube. Draw 10 c.cm. of blood

* Amer. Journ. Trop. Diseases and Prevent. Med. iii. (1915) pp. 298–303.

with a syringe from the patient's vein and add, at once, to the citrate solution in the tube. Mix by revolving or shaking the tube. The blood may be examined at once or at any time during the following twenty-four hours. The dextrose seems to preserve the cells and plasmodia against changes, and could just as well be omitted if examination was always made within an hour or two after the blood was drawn. Place equal quantities of the citrate-dextrose blood in two large centrifuge tubes—the depth of the column may vary from 2–5 c.cm. The length of time for centrifugalization depends upon the length of the arm and the speed of the centrifuge. In the authors' case the distance from the centre of the centrifuge to the bottom of the tube was 18 c.cm., necessitating a speed of 2500 revolutions per minute. The mixture should be centrifuged one minute for each cm. of the column of blood to be centrifuged. All the plasmodia (except the small æstivo-autumnal rings) and the leucocytes, rise to the top of the cell-sediment and are in the first 0·1 cm. With a large pipette skim off as much as possible of this layer from each tube and place in one (or two) 0·5 cm. tubes. Take up at the same time as much plasma as cells, and after placing in the small tube mix thoroughly by drawing back and forth into the pipette. The column in the small tube must not be deeper than 5 c.cm. Centrifuge as before. Now, with a large capillary, draw not more than a 5 c.cm. column of the cells into it from the surface of the cell-column. It is a good idea to mix this by forcing it in and out of the pipette against the surface of a slide. Draw it up into the pipette past the tip and seal the end of the pipette in a flame. Cut off part of the capillary containing the blood and centrifuge as before. After centrifuging, there will be found a small amount of greyish leucocyte mass merging into the column of red cells. In very heavy infections the lower part of the leucocyte layer and the upper part of the erythrocyte column have a brownish appearance from the pigment present in the large amount of plasmodia here. Cut the capillary at a point 0·1–0·2 cm. below the bottom of the leucocyte layer, and with a smaller capillary draw out the small amount of erythrocytes and leucocytes and a little plasma to dilute them with. Mix and make one or more blood-spreads of the usual kind. Stain and examine in the usual manner.

Histological Basis of the different Shank Colours in the Domestic Fowl.*—H. R. Barrows fixed the material in 10 p.c. formalin. Free-hand sections were made and mounted in glycerin. Many fixatives and stains were tried, the best being formalin hæmatoxylin, eosin and Sudan iii. The following summary of the results is given: 1. Yellow and variations are due to the presence of lipochrome pigment in the epidermis with the absence of melanin pigment. 2. White results from the lack of pigment. 3. Blue colour obtains when melanin pigment lies in the upper dermis, with the absence of this type of pigment in the epidermis. 4. Black and variations depend upon the presence of melanin pigment in the epidermis. 5. Green appears when lipochrome pigment lies in the epidermis and melanin pigment in the

* Rep. Maine Agric. Exper. Stat., 1914, pp. 237–52 (12 figs.).

corium only. All shades with the exception of red and pink are the result of various combinations of these pigments; orange-yellow and black-brown.

Differentiation of the Eberth-Coli Group by means of Collodion Reagent Papers.*—A. Ch. Hollande and J. Beauverie state that the members of this group of organisms can be differentiated by means of four test papers prepared as follows :—1. Nitrate of silver paper. Filter paper (e.g. Schleicher and Schüll) is soaked in 1 p.c. nitrate of silver solution and is then dried rapidly between two sheets of blotting-paper—after drying, the paper is plunged into a mixture of alcohol and ether containing 10 p.c. of pure collodion. The paper is then removed and rapidly dried. 2. Glucose-neutral-red paper. The filter paper in this case is plunged into a solution containing glucose 2·5 grm. in 25 c.cm. of 1 p.c. aqueous solution of neutral red. This paper is dried and coated with collodion as above. 3. Acetate of lead paper. 10 p.c. aqueous solution of acetate of lead is here employed, the rest of the technique being as before. The paper, which is white in colour, should be kept in a stoppered bottle. 4. Litmus-orcine-lactose paper. The solution in which the filter paper is immersed is prepared as follows :—Litmus-orcine 20 grm., neutral phosphate of sodium 4 grm., lactose 5 grm., bicarbonate of sodium 1 grm., distilled water 50 c.cm.

A portion of each of the four test papers is dropped into different tubes containing 6 c.cm. of broth and then sterilized in the autoclave. When cool the tubes are inoculated with the suspected organism and incubated at 37° C.

Bacillus typhosus alone, after twenty-four hours, does not decolorize the broth containing glucose-neutral-red paper, moreover the paper remains at the bottom of the tube, and is not brought to the surface by gas bubbles as with *B. coli* and the paratyphoid organisms. *B. paratyphosus A* does not blacken the acetate of lead paper, as do the other organisms. *B. paratyphosus B* alone possesses the property of restoring the lilac colour to the medium containing litmus-orcine-lactose, after three days; the other organisms permanently discolorize the medium. *B. coli* alone gives a growth in the nitrate of silver medium, ten or twelve hours after inoculation.

By employing a paper made with a mixture of acetate of lead and glucose neutral red the organism can be determined as follows :—With *B. typhosus* the liquid remains red and the paper is blackened; with *B. paratyphosus A* the liquid is slightly reduced but the paper remains red; with *B. paratyphosus B* and *B. coli* the red colour completely disappears and the paper is blackened. The red colour is replaced by a yellowish fluorescence.

* C.R. Soc. Biol. Paris, lxxviii. (1915) pp. 722–5.

Metallography, etc.

Microchemistry of Corrosion.*—C. H. Desch and H. Hyman have applied the electrolytic methods previously described to two alloys (gun-metals) containing copper, tin and zinc in the approximate proportions 88 : 10 : 2 and 84 : 14 : 2. Each alloy was tested as cast, and also after a heat treatment which altered its constitution. It was found that the amount of corrosion was more affected by small variations of the conditions, especially of the applied electro-motive force and of the state of the surface, than in the case of the brasses. The preliminary removal of the flowed surface layer produced by polishing, by etching with acid ferric chloride solution, eliminated one cause of discordant results, and was more readily attacked than the eutectoid, which was not affected unless the voltage exceeded a certain critical figure. Corrosion was less than that of the brasses under similar conditions.

Copper-zinc-tin Alloys.†—O. F. Hudson and R. M. Jones have studied the constitution of brasses containing small percentages of tin. Fifty-two alloys containing 0·3 to 5 p.c. tin and 43 to 69 p.c. copper, the remainder being zinc, were microscopically examined. Nearly all the alloys were annealed below 450° C. for periods of 18 hours to 15 days to obtain equilibrium. Some thermal curves were taken. The polished specimens were etched by means of a polish-attack with ammonia. The constituents found were α, β^1, and δ. α appeared reddish-yellow, β^1 bright yellow, and δ clear light blue. In a number of the alloys all three constituents were present in equilibrium. A diagram is given representing the equilibrium constitution for ordinary temperatures of the ternary alloys containing 50 to 70 p.c. copper and 0 to 5 p.c. tin; the solubility of tin in brasses and the limits of composition within which the alloys consist of three phases α, β^1 and δ in equilibrium, are defined.

* Journ. Inst. Metals, xiv. (1915, 2) pp. 189-98 (6 figs.).
† Journ. Inst. Metals, xiv. (1915, 2) pp. 98-115 (17 figs.).

X.—*Studies in Marine Biology.*

By F. Martin Duncan, F.R.P.S.

Read April 19, 1916.

Figs. 13, 14, 15.

It was my intention to-night to have given some account of my experiences in photographing, collecting, and preserving marine biological specimens, but, as the hour is late, I will curtail my remarks, and give a brief description of some of the photographs exhibited on the screens and tables, and of some of the apparatus used in obtaining them. The prints represent a small selection from the very large collection of negatives which I have made during the course of a good many years devoted to marine biological studies, and I should like to say that a large number of them could not have been obtained but for the existence of the Marine Biological Association of the United Kingdom, an institution deserving the support of all microscopists who are in any way interested in the teeming forms of microscopic life to be found in the sea.

I should like to draw your attention to this photomicrograph of the *Puerulus* stage of *Palinurus vulgaris*, the Crawfish, or Langouste as it is called in France. It is of interest as the first photomicrograph to be taken of the original specimen, discovered by Monsieur E.-L. Bouvier, during his stay at the Marine Biological Association's laboratory at Plymouth during the summer of 1913. I happened to be carrying out some work in the Laboratory at the time, and Monsieur Bouvier very kindly permitted me to take the photomicrograph for him, and has used it to illustrate his paper on the development of *Palinurus*, published in the Journal of the Marine Biological Association.*

I should like also to draw your attention to the set of prints showing some of the phases in the segmentation of the eggs of *Echinus esculentus*, and which are printed from parts of a micro-kinetograph negative obtained with a special form of apparatus I have designed for taking kinetograph records of living microscopic forms of life. In obtaining this interesting series of pictures I was greatly assisted by my friend Dr. Creswell Shearer, of Cambridge,

* Journ. Mar. Biol. Assoc., x. No. 2, p. 179.

who has devoted much attention to the artificial fertilization of the eggs of *Echinus*.

The photomicrographs of marine parasitic Protozoa I need not describe, as explanatory titles are attached, and I think the same may be said to apply to most of the other photographs exhibited to-night; therefore I may pass at once to some brief description of how they were obtained.

I began taking my first photographs of marine animals and their anatomy at the time that my dear father, the late Dr. P. Martin Duncan, occupied the Presidential chair of this Society, and these early attempts were undertaken with a view to relieving him

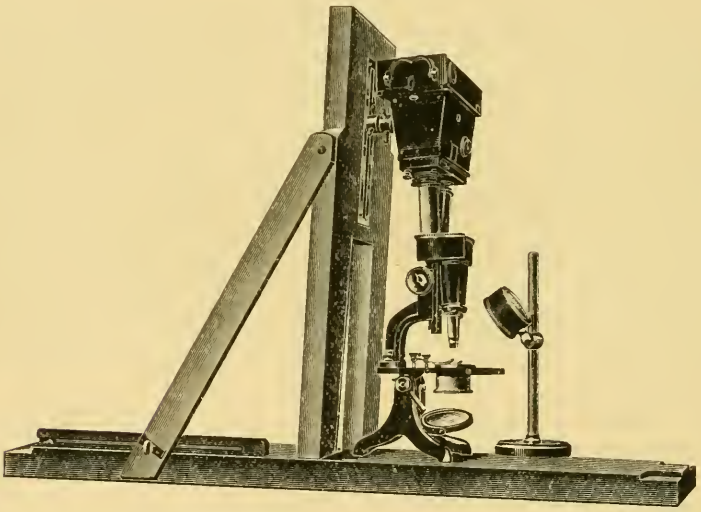

Fig. 13.—Stereo-binocular Microscope and Verascope Camera in position for taking Stereoscopic Photomicrographs.

from some of the long hours of eye-strain in the course of his microscopic investigations into the anatomy of the Corals and Echinoderms. Those first studies were obtained with the aid of an old wet-plate camera and collodion wet-plates of home manufacture, which gave me a very useful insight into the chemistry of photography. I have found that for practically all marine biological work either an isochromatic or a panchromatic plate must be used to obtain the best results, and most truthful rendering into monochrome of the various colour-values of the subject. Subsequent enlargement of part or the whole of the negative often being necessary, it is most important to select a plate of fine grain. Most of the direct prints and enlargements exhibited to-night

have been made on the Ortho A, B, or C plates manufactured by M. M. Lumière, and the very fine chromatic and panchromatic plates of the Ilford Company. For obtaining natural colour photographs of the subjects, I consider that the beautiful Lumière autochrome plate is the only direct colour-plate that will faithfully render the object with all its true tints. For all photomicrographic work backed isochromatic or panchromatic plates should be used.

In photographing the larger forms of marine animals, either in the rock-pools or in the special tanks, a good Reflex camera,

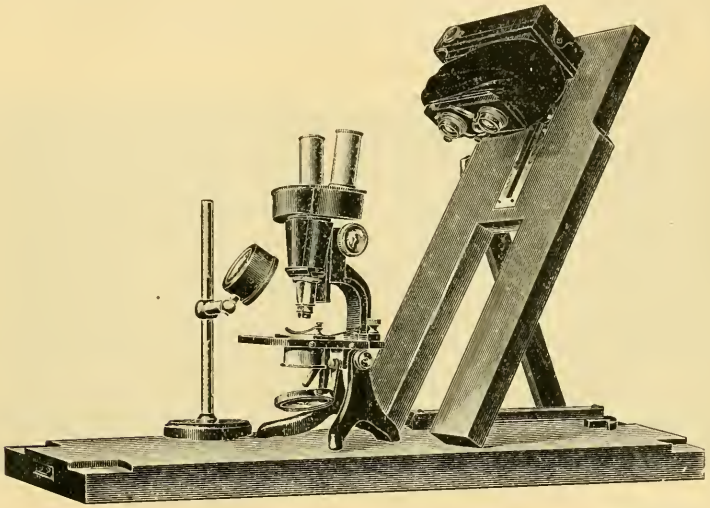

Fig. 14.—Showing the upright support for the Verascope Camera drawn back, leaving the Eye-pieces of the Stereo-binocular Microscope free for focusing the specimen.

such as the Newman and Guardia model, which I have now used for many years, will be found invaluable, as enabling one to watch and keep the animal in focus up to the moment of exposing the plate. Sky reflections are often a great trouble when working in the rock-pools, and I have found a light metal cone, that can be attached either to the front of the camera, or to a stand, so that one end is beneath the surface of the water—practically a water-telescope—a simple and certain way of avoiding surface reflections. One of the great difficulties one has to contend with is the sensitiveness of all marine creatures to change of temperature, a rise of only a few degrees being quite enough to produce very marked effects, particularly with many plankton forms, Hydroids, etc.

It is, therefore, most important to keep the water in observation or photographic aquaria at a constant temperature, and is best accomplished by inlet and outlet syphons, so that a gentle circulation is always taking place. By this means the creatures are kept in a healthy condition, and their movements and appearance normal. The photomicrographs of living Hydroids have been obtained in this way, using a zoophyte trough mounted on a special stage, and with a low-power objective attached to the Reflex camera. When a higher magnification is necessary, I use one of Messrs. Charles Baker's R.M.S. Microscopes, which I consider one of the most perfect instruments for biological research, and in conjuction I either use the Reflex camera, or a Leitz demonstration eye-piece and the ordinary photomicrographic camera.

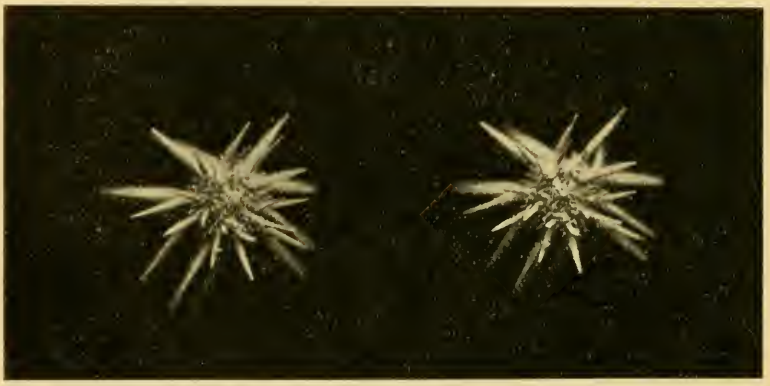

Fig. 15.—Stereoscopic Photomicrograph of Young Echinus, taken with the Stereo-binocular Microscope and Camera as shown in figs. 13 and 14.

In studying such forms of marine life as the Polyzoa, true stereoscopic photomicrographs are of very great assistance, and by the method I employ are comparatively easily obtained. I used Baker's model of the Greenough Stereo-binocular Microscope. This is placed on a baseboard, which has an upright to carry the stereoscopic camera—a 7 × 13 c.cm. Verascope of fixed focus. The object on the stage is focused through the eye-pieces of the Microscope, and the camera is then gently lowered into position, so that the lens almost touches the front lenses of the Microscope eye-pieces, and the plate is exposed (fig. 13). As the lenses of the camera are set at infinity, there is no need for further focusing, if the eyes of the observer are normal and he has in the first instance obtained a critically sharp image through the

Microscope. I do not claim any originality for this method of working, for my attention was first drawn to it by a veteran microscopist, the late Dr. F. Bossey, who told me he had employed it many years ago, I think in the early "sixties," with a box-form wet-plate camera and a monocular Microscope. As a simple and quick way of producing photomicrographs of low and medium magnification, and with the Stereo-binocular Microscope, it will be found very useful.

MICROSCOPY.

A. Instruments, Accessories, etc.*

(1) Stands.

Bausch and Lomb's Binocular Microscope (Greenough Type).†—This instrument (fig. 28) is lettered K A, and is described on page 58

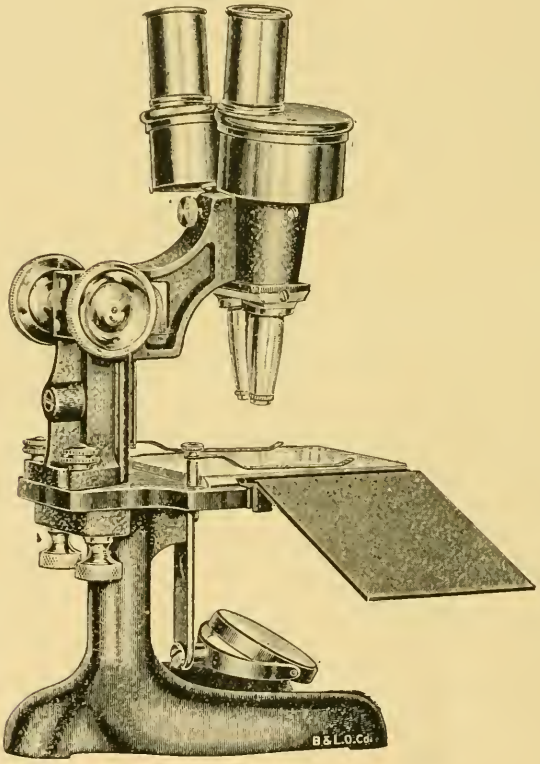

Fig. 28.

of the maker's catalogue. The base is of the modified horse-shoe form and of rounded contour. The pillar is of one piece with the base, and is detachable from the Microscope by removing two binding

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

† Bausch and Lomb's Catalogue, Microscopes, 111 pp. (many illustrations), Rochester, N.Y., and London.

screws, thus rendering the upper portion of the instrument ready for use in examining large surfaces—the stage, with its plate removed, serving as a base. The arm embodies two special features: (1) It provides for extraordinary range of working distance up to 155 mm., measuring from lower edge of nose-piece to stage; the curved part is adjustable along upright, and can be fixed in any position by the clamping head shown in opening of curved part; the working distance can also be decreased by detaching curved part from upright and body-tubes, inverting and replacing it, for focusing some distance beneath stage. (2) The entire arm with body-tube can be removed and used on a stand consisting of base-plate and vertical pillar (listed as K B), it being only necessary to place a short post, as provided, in socket at back of arm. The body-tubes are a combination of two Microscope tubes, the upper parts of which are fitted with porro prisms, and are rotatable for adjusting position of eyepieces to observer's pupillary distance; one image merges with the other, and the object is seen stereoscopically, erect and not transposed; the nose-piece provided takes self-centring slides, upon which each pair of objectives is mounted, with means for centring one with the other; one of each pair of objective mounts provides for adjustment to compensate for differences between the observer's eyes. The focusing adjustment is by standard rack-and-pinion, the pinion heads being so placed with relation to body-tubes as to eliminate completely the inconvenience caused when these parts are close together; the location of rack slide is such that it is not immersed when working with water-immersion objectives. The stage is of metal with large rectangular aperture provided with two removable plates, one of glass 80 × 95 mm., the other of metal, same size, with aperture 22 mm. in diameter, underneath which a rotating plate provides white opaque, black opaque, and ground-glass stops, or clear aperture; the stage is also provided with detachable metal hand rests and spring clips. The mirrors are plane and convex, 50 mm. in diameter, adjustable in two planes in a fork; they are mounted on a swinging arm, and are not easily removable.

(3) Illuminating and other Apparatus.

New Mechanical Stage.*—Katharine Heanley, who describes this new device, says that in addition to the usual advantages of a mechanical stage, the invention here illustrated (fig. 29) is easily fixed to and

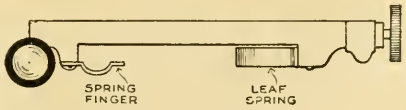

FIG. 29.

unfixed from any Microscope. This is effected very simply. To the right, on the under side of the machine, is a strong curved leaf spring attached at its proximal end to a firm crossbar; on the left, at a

* Lancet, July 15, 1916, p. 110 (1 fig.).

convenient distance, are pivoted two spring fingers. The machine is slipped over the anterior edge of the stage, the curved spring firmly presses the right edge of the stage, while the two spring fingers grip its left edge and under surface. This construction is sufficient to hold the machine immovably on the stage, and is adapted to engage stages of different widths within the limits of stage sizes usually employed with Microscopes.

(5) Microscopical Optics and Manipulation.

Spirochæta pallida.*—E. M. Nelson shows a rough sketch (fig. 30) of *Spirochæta pallida* made under a power of 4000 diameters, which illustrates two details hitherto unknown. They are (1) the long whip-like flagella, and (2) the beaded structure of the organism. The length of the organism is $\frac{1}{24\,0\,0}$, the width of the helix about $\frac{1}{30\,0\,0\,0}$, the pitch of the screw about $\frac{1}{17\,5\,0\,0}$, the length of the flagellum about $\frac{1}{30\,0\,0}$, and the thickness of the thread of the body $\frac{1}{65\,0\,0\,0}$ in. The longest of the organisms measured $\frac{1}{15\,0\,0}$ in., and the shortest was somewhat less than half that size. The pitch of the screw is very variable. The thickness

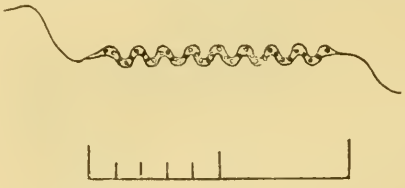

Fig. 30.—Scale, $\frac{1}{20\,0\,0}$ in. by 4000.

of the thread of the body was corrected for antipoint. The organism does not resemble a spiral, but is like a helical screw. The helix of the one drawn was measured both by a screw micrometer and by the simpler method of extinction; the results were identical. The flagellum is a fine but not a difficult microscopical image, but the visibility requires most careful microscopical work. The image of these was so difficult that it was found impossible to step them with the web of the micrometer. As a rough estimate they may be 110,000 to the in.

Immersion fluid.†—E. M. Nelson writes that microscopists should know that it is now almost impossible to procure proper immersion oil for Microscope object-glasses. A proper fluid was invented by Abbe, and was sold by the firms of Winkel and Zeiss. The "cedar oil," as commonly sold, and also the immersion fluid of Lectz, have not the requisite optical properties, therefore objectives immersed in them do not yield their best results. The following formula gives Abbe's latest results. The ingredients are three, viz.: (1) White oily tacamaque of Guibourt; (2) oil of cedar (*Juniperus virginiana*); (3) castor oil. The proportions are 29 grammes of tacamaque dissolved in 22 c.cm. of cedar oil, to which are added 14 c.cm. of castor oil.

* English Mechanic, June 2, 1916, p. 371 (1 fig.).
† English Mechanic, June 2, 1916, pp. 370-1.

(6) Miscellaneous.

Miniature Dark-room for Use with the Microscope.*—R. T. Hance says that all microscopists prefer to work either at night or in a darkened room. Using the Microscope under such conditions does away with the strain to which both the observing and the unused eye are subjected by the side light, i.e. light coming from sources other than through the tube. When working in darkened surroundings the effect is that of looking at a picture on a screen. The image appears brighter, and objects become clear that under the usual conditions are scarcely visible.

For several years the writer has been trying to devise some method to control the light perfectly, and to do this without necessitating the darkening the whole room. It is desirable that any apparatus for the purpose should weigh little, and (for ease in carrying from one place to another) it should be simple to take apart. It should, of course, be adaptable to every condition. For further convenience of the worker definite places should be present in such an apparatus for the usual microscopical accessories—pens, pencils, drawing and memorandum cards, and lens paper.

The following description is of a miniature dark-room for use with the Microscope fulfilling these requirements. It was designed and made by the writer last fall, and, after a year's use, he has found it to be exceedingly practical in eliminating all the strain that results when the eye is unshielded. In this darkened enclosure the eye not in use is at perfect rest. Moreover, for drawing the light may be controlled so that it is possible always to have light of the same intensity directed on the drawing paper.

Description (Fig. 31).—A, Base : $\frac{1}{4}$-in. white pine 12 by 18 in., with a binder of the same wood across each end to prevent warping,

B, Uprights : dowel sticks 1 in. in diameter cut to 18 in. in length.

CC, Rods : common telescoping curtain rods. Each of the rods C is cut 8 in. from the end that ordinarily would be used to fasten it to the window. C is formed of the remainder, of the part between the ends.

D, Wire : a piece of annealed wire $\frac{1}{8}$ in. in diameter about $4\frac{1}{2}$ ft. long bent as shown.

To Assemble :—One 2-in. screw fastens each upright to the base. The upright on the left can be seen to have two angle-irons aiding in its support, but this is only necessary when the fan is added. Holes are drilled in both uprights to correspond to the diameter of C, which is inserted in them. The rods C are attached by one end to the tops of the uprights by a screw through the eyelet in the rod. Through the eyelet at the opposite end a small rod is passed as shown to prevent the curtains from slipping off. The wire D is fastened to the outer sides of the uprights by means of a single round head screw passed through each flattened end. All the wood and metal-work is painted a dead black.

For many valuable suggestions on the design of the curtains and for the excellence of their construction I am indebted to my mother. (See fig. 32.)

* Trans. Amer. Micr. Soc., xxxv. (1916) pp. 60–4.

The curtains suspended from the rods C and C′ are in four parts, all overlapping each other and fastening together with spring snaps. They are made of the heaviest grade of black sateen doubled. On the right-hand curtain are pockets for pencils and cards. On the left side is a pocket for lens paper. The pocket is provided with a flap to exclude the dust. The upper curtain carried on the wire D is of single thickness. The central curtain is in two parts so that they may be separated to permit light to fall on the drawing board. The left-hand curtain of the central set has a rectangle 1 in. wide by 5 in. high cut from the

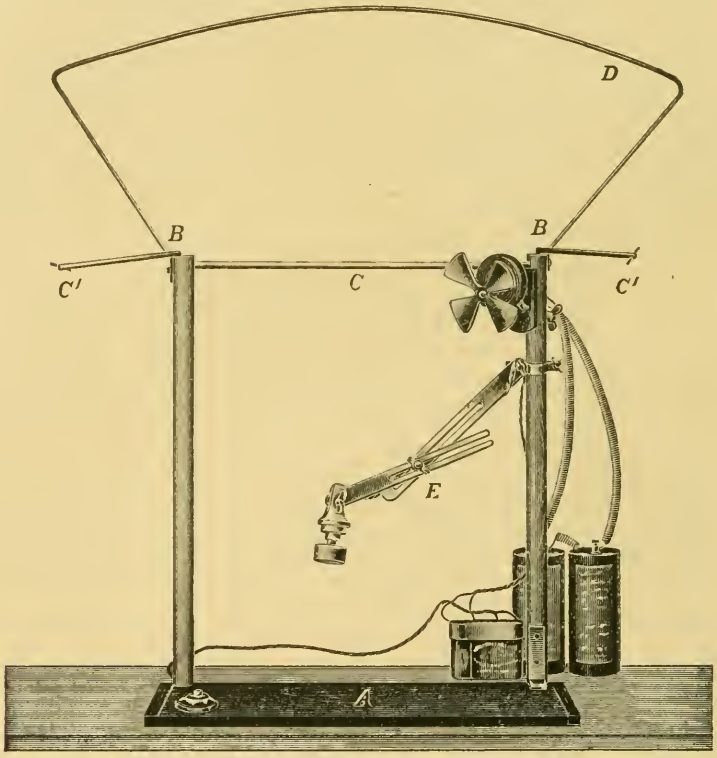

Fig. 31.

centre of the basal portion. Across the top of this aperture is stitched a flap of double thickness, 3¼ in. wide by 5½ in. in length. To one corner of the loose end of the flap is attached a tape which passes around the tube of the Microscope and fastens to the other corner of the flap by means of a spring snap.

With the Microscope surrounded by these curtains it is impossible to read the figures on the mechanical stage, and so the small light (fig. 31, E) was installed. This can be adjusted by means of sliding rods locked with winged nuts to hang directly over the stage. The lamp arm is attached to the right-hand upright by means of a collar

made of two pieces of brass stripping fastened on each side of the pillar with a thumb-screw. The lamp is a small tungsten bulb set in a porcelain socket. The shade or reflector, shown in the photograph, was taken by an old tubular flashlight. A small three-cell pocket battery furnishes the current, which is controlled by a push-button at the left of the Microscope. The same battery has lasted for very nearly a year now without visible signs of weakening.

The fan shown in both photographs is a toy motor equipped with a 4½-inch blade. The motor is operated on two dry cells. It is fastened to a wooden base that is inserted in a slot in the upright and clamped tight by means of a winged nut. This fastening permits the fan to be

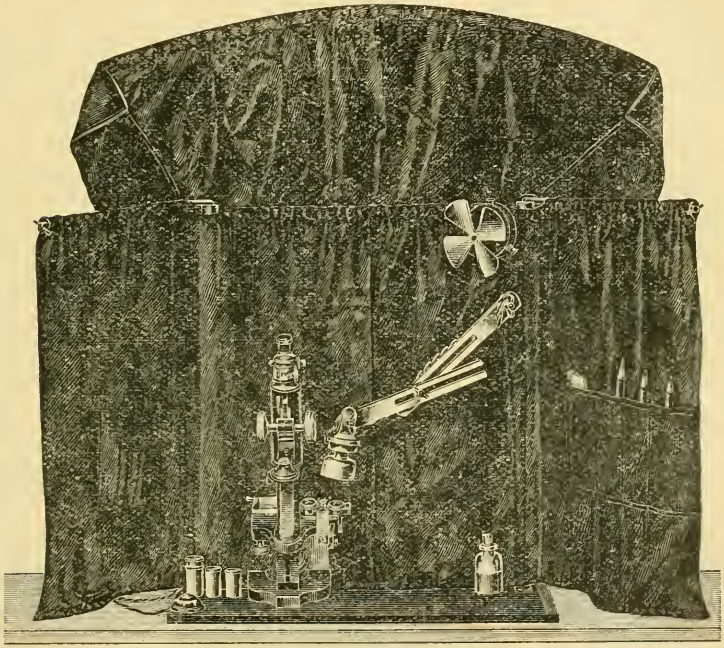

FIG. 32.

tilted up and down, while the single screw securing the fan to the base allows a left and right rotation. The air current may thus be directed on any spot desired.

Operation.—For Microscope illumination with this dark room a concentrated filament Mazda frosted globe is used. This globe is placed behind the slit in the central curtain, and the Microscope is put in position on the opposite side. The flap covering the slit is then snapped about the tube of the Microscope just above the nose-piece. The slit through which the light comes is so narrow that the stage of the Microscope effectively shields the eye from the light coming through the lower part of the slit, while the flap takes care of all other dispersion.

In the average room having windows on only one wall the side curtains can be left wide apart. In places where the worker is almost surrounded by windows it is of advantage to draw the side curtains so close that there is just room for the observer's head to enter. The telescoping rods supporting the side curtains permits these to be narrowed or widened to suit the circumstances. The top curtain works to or from the microscopist, and is frequently convenient in cutting out the light from the upper parts of the windows.

Light on the drawing paper is obtained by separating the lower portions of the central curtains from each other and fastening them back. The bulb illuminating the Microscope then throws its light over the right-hand side of the base. A constant intensity of illumination is in this way assured.

The fan is a luxury—possibly an unnecessary one—but in very warm weather, or on days when a few flies persist in maintaining their position at all hazards on top of the writer's head, he has not been at all sceptical as to whether the luxury was unnecessary or not.

Microscopic Structure of Semi-permeable Membranes and the Part played by Surface Forces in Osmosis.*—F. Tinker thus summarizes his researches :—1. The common precipitation semi-permeable membranes are composed of small precipitate particles ranging from 0·1 μ to 1 μ, these particles being closely packed together. Each of these precipitates is, however, not simple in structure, but is itself an aggregate formed by the flocculation of sub-microscopic colloidal particles. The particles composing the membrane are smallest in the case of copper ferro-cyanide and prussian blue. 2. Precipitation membranes show most of the properties of gels, as ordinarily prepared, both in their method of formation and in the changes they undergo in various solutions. Like ordinary gels they are possessed of great tensile strength, which varies in membranes of different kinds. Their stability in the colloidal condition also varies greatly. But, although they show the physical properties of gels, they have not the same mechanical structure, the membrane being much more closely knit together than the gel proper. 3. The pores in a copper ferro-cyanide membrane range from 8 to 60 $\mu\mu$ in diameter, the average diameter being from 15 to 20 $\mu\mu$. The pore size is too great for the membrane to act osmotically by exerting a selective mechanical blocking action. 4. The order of a series of membranes in pore size is the same as that of their efficiency in semi-permeable membranes. Copper ferro-cyanide and prussian blue are the most efficient membranes, and they have also the smallest pores. 5. There is a very close connexion between the osmotic properties of a membrane and the extent to which the membrane capillaries are under the control of surface forces. Osmotic effects are probably the result of selective adsorption phenomena occurring at the surface of the membrane and in the capillaries, the membrane being relatively impermeable to solutes which are negatively adsorbed, but permeable to solutes which are positively adsorbed.

* Proc. Roy. Soc., xcvi. (1916) pp. 357–72 (6 figs.).

B. Technique.*

(1) Collecting Objects, including Culture Processes.

Hæmoculture of Gonococci.†—P. Danila reports a case of gonorrhœal septicæmia in which he succeeded in isolating the organism in question from the blood-stream. Ordinary broth without the addition of serum or ascitic fluid, ascitic agar, and ordinary agar were inoculated with blood from the median vein and incubated immediately at 37° C. After forty-eight hours colonies of the gonococci were observed growing in the clot in the broth culture. The other media remained sterile. The articular fluid from the knee-joint, although rich in leucocytes, did not contain gonococci. The patient did not recover.

New Method of Anaerobic Culture.‡—J. McIntosh and P. Fildes have constructed an apparatus in which they state anaerobic cultures can be grown with the greatest facility. The culture tubes are enclosed in a receptacle in which is suspended a piece of asbestos or platinum covered with palladium. Hydrogen is then passed into the receptacle by means of a stop-cock. The palladium black causes the hydrogen to combine with oxygen, and the atmosphere becomes quite free from the latter.

A round receptacle made of tin-plate is employed (175 mm. by 125 mm.), to which is adapted a lid with a tap affixed centrally. The contrivance is made impervious to the external air by means of a plasticine luting. The culture tubes are placed in the luted receptacle, the metal cage containing the palladium is then warmed in the Bunsen flame, and the lid firmly adjusted. Hydrogen, under pressure, is then allowed to flow through the tap until the apparatus becomes cold (twenty-five minutes). It is then placed in the incubator. If used for gelatin cultures it is necessary to place a refrigerating mixture in the receptacle at the same time as the cultures. A yet more simple application of this method may be made by employing an Erlenmeyer's flask fixed with a rubber cork through which passes the connexion from the hydrogen tap, the cage containing the platinum-palladium being fixed with a nut to the lower surface of the cork. By these methods the authors have succeeded in producing well-developed colonies of *Bacillus perfringens* and of *B. œdematis maligni* from material taken direct from war wounds containing these organisms. The advantages claimed are as follows:—1. The method is very simple and rapid, the operation being completed in twenty minutes. 2. The apparatus is always ready for use without previous preparation, and without the employment of any reagent other than hydrogen. 3. All the usual laboratory media can be used without any previous preparation. 4. The strictest anaerobes grow on the surface of the media; for example, the colonies of the tetanus bacillus on serum agar become visible in twenty-four hours.

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

† C.R. Soc. Biol. Paris, lxxix. (1916) pp. 460–1.

‡ C.R. Soc. Biol. Paris, lxxix. (1916) pp. 293–5.

Preservation of Living Red Blood-cells in Vitro.*—P. Rous and J. R. Turner point out the practical value of the preservation of red cells for use in serum reactions, culture media, and even for transfusion. One-eighth p.c. gelatin is added to the solution in which the cells are washed, the use of this reagent protecting the cells from mechanical injury. Though gelatin acts as a protective for red cells it is not a preservative of them in any real sense. Cells do not last longer when it is added to the fluid in which they are kept. Lock's solution, though better probably than Ringer's solution, or a sodium chloride solution, as a medium in which to keep red cells is ultimately harmful. The addition of innocuous colloids does not improve it. But the sugars, especially dextrose and saccharose, have a remarkable power of preventing its injurious action, and they possess, in addition, preservative qualities. Cells washed in gelatin (Lock's) and placed in a mixture of Lock's solution with an isotonic watery solution of a sugar remain intact a long time—nearly two months in the case of sheep's cells. The kept cells go easily into suspension free of clumps; they pass readily through filter-papers, take up and give off oxygen, and when used for the Wassermann reaction behave exactly as do fresh cells of the same individual. The best preservation solutions are approximately isotonic with the blood serum. If the cells are to be much handled, gelatin should be present, for the sugars do not protect against mechanical injury.

Different preservative mixtures are required for the cells of different species. Dog cells last longest in fluids containing dextrine as well as a sugar. The mixture best for red cells is not necessarily best for leucocytes.

A simple and practical method of keeping rabbit and human erythrocytes is in citrated whole blood, to which sugar solution is added. In citrated blood as such, human red cells tend to break down rather rapidly, no matter what the proportion of citrate, hæmolysis being well marked in a little more than a week. But in a mixture of three parts of human blood, two parts of isotonic citrate solution (3·8 p.c. sodium citrate in water), and five parts of isotonic dextrose solution (5·4 p.c. dextrose in water) the cells remain intact for about four weeks. Rabbit red cells can be kept for more than three weeks in citrated blood, and the addition of sugar only lengthens the preservation by a little. The result differs strikingly with the amount of citrate employed. Hæmolysis occurs relatively when the smallest quantity is used that will prevent clotting. The optimim mixture has three parts of rabbit blood to two of isotonic citrate solution.

Anaerobes Isolated from Wounds.†—Muriel Robertson, working with material derived from gangrenous wounds sent her from Flanders by Major Rowland and others, has succeeded in isolating therefrom *Bacillus perfringens* (*B. ærogenes capsulatus*, Welch and Nuttall: *B. Welchii*, auctt.; *B. phlegmones emphysematosæ*, Fraenkel), *B. œdematis maligni* (Koch), and a bacillus closely related with Hibler's bacillus No. 9, and also allied to Rodella's bacillus No. 3.

* Journ. Exper. Med., xxiii. (1916) pp. 219–37.
† Journ. Path. and Bact., xx. (1916) pp. 327–49.

Organism	Motility	Spores	Colonies on Agar	Sugar	Milk	Gelatin	Inspissated Serum	Glucose Broth	Mulin
Vibrion séptique	Motile, but often non-motile	Central or sub-terminal	—	Acid reaction, gas, pink colour	Acid clot, some gas, no digestion	Liquefied	Not liquefied	—	—
B. perfringens .	Non-motile	Ditto	Smooth surface colony, lenticular in depth	Acid reaction, much gas, pink colour	Acid clot, much gas, no digestion	Ditto	Ditto	Acid and gas	Acid and gas
Hibler No. 9 .	Non-motile or very feebly motile	Endospores attached to long slender rod	Flat, slightly crenated, smooth colony on surface, has lumpy outgrowths in depth	Acid reaction, considerable amount of gas, pink colour	Attacked very slowly, finally acid and clot	Not liquefied	Ditto	Ditto	No change
B. œdematis maligni	Actively motile	Central or sub-terminal, rarely terminal	Woolly colony, both on surface and in depth	Alkaline reaction, blackened, with putrid odour, digested	Milk digested without clot, but casein may be precipitated in grain	Liquefied	Liquefied	Ditto (not vigorous)	Ditto
B. tetani. . .	Motile	Endospore, "drumstick," but clostridial forms present	Ditto	Blackened, with putrid odour	Digested with precipitation of grains of casein which are digested	Ditto	Ditto	—	—

The preceding table gives the cultural reactions of the three organisms, and also of the "vibrion séptique" (original strain from Pasteur Institute, which appears in reality to be Rauschbrand) and *B. tetani*.

The pathogenic anaerobes may thus be conveniently divided into four groups as follows :—

Group A.—The Rauschbrand group (Bacillus of Rauschbrand, Bacillus of Ghon and Sachs, Bacillus of Novy), containing forms which do not digest the proteins ; they do not liquefy inspissated serum or produce blackening in meat or "hernbrei" medium. They clot milk without much shrinkage of the clot ; they are able to liquefy gelatin ; they are usually motile, but this character is liable to variation. Spores are formed in all the media, but an alkaline reaction favours their production ; the spores are central or subterminal. The group for the most part is very pathogenic to laboratory animals. They are very strict anaerobes.

Group B.—The *perfringens* group contains forms which have very little capacity for attacking the proteins. The strains belonging to this group do not liquefy inspissated serum or blacken meat or "hernbrei." They clot milk, producing a hard tough clot much broken by gas ; the reaction is acid, and a strong odour of butyric acid is given off. There is no obvious digestion of the clot. They are non-motile, with the exception of *B. enteritidis sporogenes* (Klein). The colonies are smooth ; spores are not formed, except on alkaline media rich in protein and containing a little fermentable sugar. The organisms are pathogenic.

Group C (*B. amylobactor*, von Hibler No. 9) may be recognized by their inability to liquefy gelatin ; they are incapable of attacking the proteins, and do not liquefy inspissated serum or blacken meat media.

Group D (*B. œdematis maligni*, *B. tetani*, *B. botulinus*, *B. cadaveris sporogenes*) may be called the proteolytic group, and is characterized by its very active powers of digesting media rich in protein. Milk is digested usually without the production of clot, though a precipitation of casein may occur ; inspissated serum is liquefied, meat media blackened and digested, and gelatin is liquefied ; their growth produced a penetrating odour of putrefaction. They are actively motile. The colonies have a characteristic appearance, and grow out in long tangled filaments at their edges. Such members as *B. tetani* and *B. botulinus* are sharply marked off by their characteristic toxins and other peculiarities, but they are none the less closely allied to the rest of the group in their cultural reactions.

The composition of the cooked meat medium and the alkaline egg fluid referred to is as follows :—

Cooked Meat Medium.—Eight ounces of bullock's heart, minced very fine and then ground in a mortar ; add eight ounces of tap water and heat slowly so as to cook the meat thoroughly ; add normal sodium hydrate until the medium is alkaline to litmus. Divide into tubes and autoclave.

Alkaline Egg Fluid.—This is a modification of Besredka's medium. The yolk of one egg and the whites of two are beaten up in a beaker ; add 6 c.cm. of normal sodium hydrate ; add 500 c.cm. of tap water by

degrees. Heat very slowly to 95° C., keeping the mixture at this temperature for about an hour or longer; filter through cotton-wool and muslin. Divide the tubes and autoclave at 115° C. for twenty minutes. This egg fluid may be added to ordinary nutrient broth in the proportions of about 1 to 5. The egg fluid must be added to the broth when both the fluids are cool. The egg fluid may be added to agar at 50° C. All the sugar media should be made up with nutrient broth instead of pepton water. The anaerobes do not grow sufficiently well upon pepton water.

(4) Staining and Injecting.

Staining of Bacterial Capsules.*—R. Muir claims that by this method the capsules of bacteria can be stained differentially, while a Gram-positive reaction is also obtained. A thin film of the material is made on a cover-glass, and after drying in the air is fixed for one minute in a saturated watery solution of mercuric chloride. The preparation is then washed in water and in methylated spirit. With wet films the cover-slip is placed film-side downwards in 10 p.c. formalin for two to five minutes. It is then gently washed with water and methylated spirit. The film is covered with freshly prepared Gram-stain of the following composition: Saturated alcoholic gentian violet, 1 part; 5 p.c. watery carbolic acid, 5 parts. The preparation is heated over a Bunsen for a few minutes; when cool, the stain is washed off with Gram's solution, and a little fresh iodine solution is added. After two or three minutes the iodine is washed off with methylated spirit and the washing with spirit repeated. A few drops of clove oil are then placed on the film and the warming repeated. Wash with spirit and then with water. Filter on the film a few drops of solution containing one part each of a saturated watery solution of mercuric chloride, a saturated watery solution of potash-alum, and a 20 p.c. solution of tannic acid. This is allowed to act for five minutes; wash in water and counter-stain for one or two minutes with a saturated watery solution of eosin. Wash in water. Filter on a few drops of saturated watery solution of potash-alum and allow to act for a minute. Then wash, dry and mount, or, in the case of wet fixed films, dehydrate, clear in benzol and mount.

For sections, small pieces of tissue should be fixed in 5 to 10 p.c. formalin for two or three days, and after a few minutes' washing should be transferred to methylated spirit for two or three days to complete the hardening. For removal of the precipitate, which forms in the material thus fixed, place the pieces in a solution containing one part of 1 p.c. aqueous potash solution and twenty parts of 80 p.c. alcohol. The precipitate takes about a week to become removed. Wash thoroughly for several hours in running water, replace in methylated spirit, and keep till required. Embed the tissues in the usual way. The method of staining sections is the same as that described for films, except that the specimen may not require heating after the clove oil is placed on it, and in any event heating need not be prolonged beyond a few seconds

* Journ. Path. and Bact., xx. (1916) pp. 257–9.

in order to complete the decolorization, and that the alcohol used for the dehydration should contain four drops of glacial acetic acid to the ounce.

Fixation and Staining of Chondriosomes.*—A. Maximow recommends the method of Champy with the "post-chromisation" of Benda. The fragments of tissue are placed for twenty-four hours in a mixture of seven parts of 1 p.c. chromic acid, seven parts of 3 p.c. bichromate of potash, and four parts of 2 p.c. osmic acid. Wash in water and place for twenty-four hours in two parts of acetic acid, and one part of 1 p.c. chromic acid. Wash again for half-an-hour, and place for three days in a solution of 3 p.c. bichromate of potash. Wash once more. Embedding in paraffin gives better sections of chondriosomes than celloidin embedding. The staining method of Kull is considered the best and consists of staining at first with acid fuchsin (Altmann), then thionin, and differentiating with aurantia. The chondriosomes are by this method stained a deep red, while the nuclei take on a purple hue. The protoplasmic ground substance is stained a clear yellowish grey.

(5) Mounting, including Slides, Preservative Fluids, etc.

Substitute for Canada Balsam.†—R. Borrow states the gum from *Pinus tæda* is a good substitute for Canada balsam. When prepared for use it has a refractive index of about 1·626. This makes it specially useful for mounting diatoms. It is prepared as follows. Put the gum into a wide-mouthed bottle and add just sufficient alcohol to cover it. When dissolved pour off the clear part into another bottle and add oil of cassia in the proportion of one of cassia to three of gum. To use it for mounting diatoms, dry the specimens on a cover-glass in the usual way, place on the hot-plate, and heat until bubbles are caused. When these cease to appear, apply a lighted match quickly, which will clear all the remaining bubbles. Now push the cover-glass on to a cold-plate of iron or earthenware, when the gum will immediately harden. Warm the slide and place on it the cover-glass. The gum will melt and allow the cover to settle down, or slight pressure may be applied. Put on the cold-plate again. In a minute or so the gum which has oozed out may be chipped off with a sharp-pointed knife.

(6) Miscellaneous.

Amœbæ in Pyorrhœa alveolaris.‡—J. Mendel has investigated the question of the pathogenicity of the *Entamœba buccalis* in infective conditions of the mouth, and has come to the following conclusions. The presence of amœbæ in the buccal cavity of man is a commonly observed circumstance, and is not exclusively characteristic of the affection known as *Pyorrhœa alveolaris*. Amœbæ are however met with in nearly all cases of the disease, and are found in all mouths, in

* C.R. Soc. Biol. Paris, lxxix. (1916) pp. 462–5.
† English Mechanic, June 23, 1916, pp. 431–2.
‡ Ann. Inst. Pasteur, xxx. (1916) pp. 286–97.

which the dental hygiene has been neglected, in the cheesy matter covering the teeth. In the case of well-cared-for mouths amœbæ are found in about 50 p.c. of the examinations, and generally speaking their presence coincides with a lowering of the normal resistance of the subject, and may therefore be considered as a predisposing cause of *Pyorrhœa*. Amœbæ are invariably absent in the various acute infections of the buccal cavity, while their presence is frequent in states of chronic infection. The rational hygiene of the buccal cavity constitutes the best means in completely excluding amœbæ or at least considerably reducing their numbers. The use of hydrochlorate of emetine, so highly spoken of by other observers, has, in the author's hands, been barren of results.

"Aids to Bacteriology."*—This may be recommended to the student as a useful book of reference, both as regards technique and the theoretical aspects of the subject. The earlier chapters are devoted to a description of bacteriological apparatus, the preparation and use of nutrient media, and the technique of the preparation of material for microscopical examination. The nine following chapters contain descriptions of all the ordinary pathogenic bacteria in relation to their pathogenicity, cultural reactions, morphology, and methods of differentiation. The chapters are divided into the acid-fast organisms (*B. tuberculosis*, *B. lepra*, etc.); spore-bearing pathogenic organisms (*B. tetani*, *B. œdematis maligni*, *B. ærogenes capsulatis*, etc.); the colon-typhoid group (*B. coli*, *B. typhosus*, *B. paratyphosus A.* and *B.*, *B. dysenteriæ*, etc.); the *Bacillus diphtheriæ* group; the bacilli of the hæmorrhagic septicæmias (*B. pestis*, *B. pseudo-tuberculosis rodentium*, etc.); micro-organisms of suppuration and septic diseases (staphylococci, streptococci, pathogenic tetracocci, *B. pyocyaneus*, the gonococcus, the meningococcus, the acne bacillus, and *Diplococcus rheumaticus*). Chapter IX. discusses the pneumococcus and *Micrococcus melitensis* (no mention is made of *M. paramelitensis*), and Chapter XII. the influenza bacillus, Duerey's and the Kocks-Weeks Bacillus, the bacillus of whooping-cough (Bordet and Gongou), the glanders bacillus, and the bacillus of epidemic abortion in cattle. Chapter XIII. is devoted to the cholera vibrio and cholera-like vibrios. The next six chapters cover a very wide range of subjects in a somewhat discursive manner, the trichomycetes, the blastomycetes, the hyphomycetes, pathogenic protozoa, enzymes, sulphur and iron bacteria, bacterial diseases of plants, diseases of questionable origin, and the filterable viruses being briefly surveyed. The last three chapters are devoted to the bacteriology of sewage, shellfish, soil, air and milk, the bacteriology of water, and the chemistry and application of disinfectants. The book concludes with a small appendix and a copious and useful index.

* Aids to Bacteriology, by C. G. Moor, M.A. (Cantab.) F.I.C., and William Partridge, F.I.C., 3rd ed.

Metallography, etc.

Reciprocal Solubility of Copper and Lead.*—By chemical and microscopical methods B. Bogitch has shown that a molten alloy of copper and lead separates into two layers when the copper content is between 34·5 and 87 p.c. and the temperature is between 940° and 975° C.

Gold-cadmium Alloys.†—P. Saldau includes a detailed description of the microstructure of numerous gold-cadmium alloys in an account of his investigation of this binary system. The alloys were examined as cast and also after annealing at 250° or 350° C. for ten days. Two compounds occur, AuCd and $AuCd_3$, which enter into solid solution with their components through a limited range.

Persistent Brittleness in Steel.‡—V. Bernard and A. Portevin have examined a piece of medium-carbon steel which had broken in use. Its brittleness was thought to be due to the overheating which was indicated by the coarse structure. Quenching from 850° C. followed by reheating to 700° C. replaced the coarse structure by a fine one, but impact tests showed that the brittleness had not been removed. A more careful microscopic examination then revealed the presence of indistinct narrow bands of ferrite forming a very coarse network. Lines of minute holes and inclusions in the middle of these ferrite bands formed surfaces of weakness, resulting in brittleness which could not be removed by any form of heat-treatment. The network of small holes and inclusions is regarded as the remains of an early and extremely coarse crystallization.

Laminated Structure in Steel.§—Y. A. Fechtchenko-Tchopovsky describes the laminated structures found in rolled steel such as rails and boiler-plates, and discusses their causes and the possibility of removal by heat-treatment. A markedly laminated structure found in a boiler-plate was partially or wholly removed by annealing followed by air-cooling, the effect being greater as the annealing temperature was higher. The laminated structure reappeared when the steel was again annealed and slowly cooled.

* Rev. Métallurgie, xii. (1915) pp. 655–6 (2 figs.).
† Int. Zeitschr. Metallographie, vii. (1914) pp. 3–34 (30 figs.).
‡ Rev. Métallurgie, xii. (1915) pp. 155–60 (7 figs.).
§ Rev. Soc. Russ. Métallurgie, ii. (1913) pp. 140–50 (20 figs.), through Rev. Métallurgie, xii. (1915) Extraits, pp. 269–73 (14 figs.).

MICROSCOPY.

A. Instruments, Accessories, etc.*

(1) Stands.

New Spencer Microscope, No. 5.†—The body-tube of this instrument (fig. 34) is of aluminium, and is arranged to accommodate the large low-power photo-micro-objectives as well as those with the Society screw; the oculars are compensating, with lenses of large field. The large arm is conveniently shaped for handling, and has the Spencer side fine-adjustment. There is a free distance of 100 mm. from optical axis to arm. One complete revolution of the thread represents an up-and-down movement of 0·1 mm., and the graduations on the button to 100 parts make the value of each division one micron. The revolving stage is 150 mm. in diameter, the vulcanite area being of 130 mm. in diameter. The nickelled periphery is graduated to degrees, and the vernier reads to three minutes. The mechanical stage is easily removable by simply slipping it off from its bearings, which are embedded in the stage. The bearings may then be covered by a slide provided which makes a clear, even, plane stage without the necessity and expense of buying an extra plane stage. The buttons operating the mechanical stage are on concentric axes, one above the other. There is a range of 80 mm. in lateral movement and 50 mm. in to-and-fro movement. When the large stage is centred by means of the centring screws the two vernier readings of the mechanical stage are all that are necessary to locate an object. The complete rack-and-pinion sub-stage is equipped with an achromatic condenser fitted on the drop-swing mounting.

Spencer Microscope, No. 10.‡—This instrument (fig. 35) very closely resembles the No. 5 of the same firm. The chief differences are: (1) in the shape of the arm, with its handle opening; (2) in the lateral movement of the stage, 75 mm. as compared with 80 mm.; (3) in the free distance between the arm and optical axis, 80 mm. as against 100 mm.

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

† Catalogue, Spencer Lens Company, p. 15.

‡ Catalogue, Spencer Lens Company, p. 15.

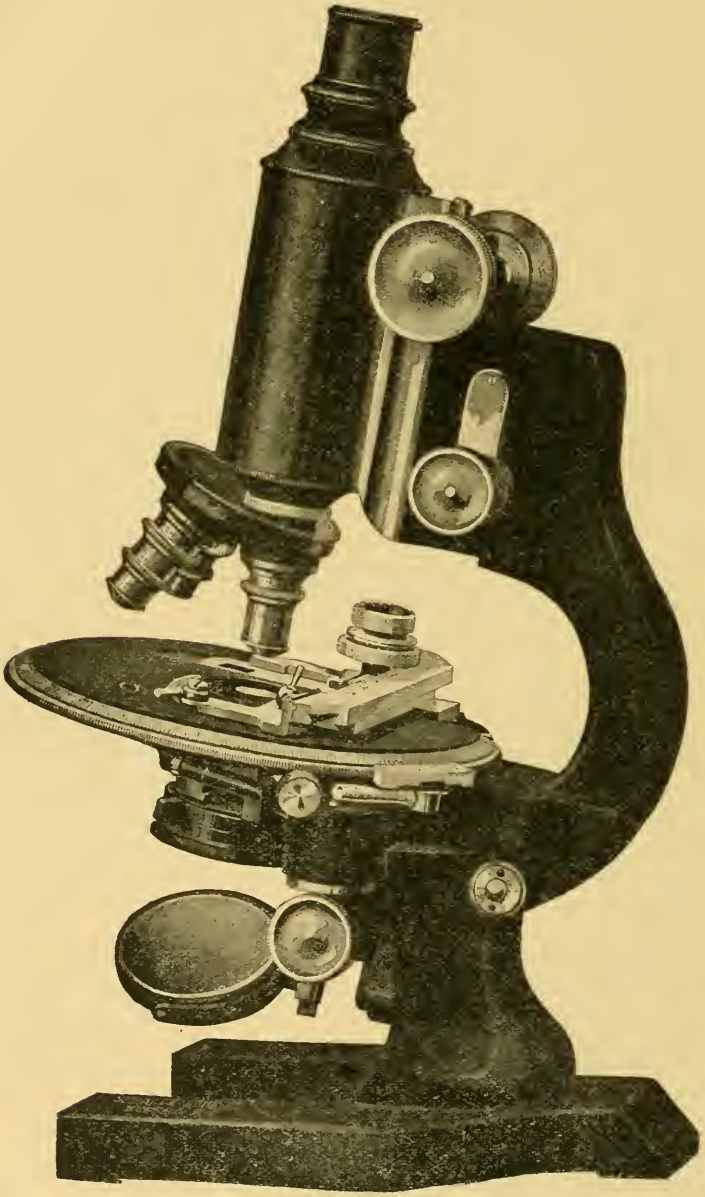

Fig. 34.

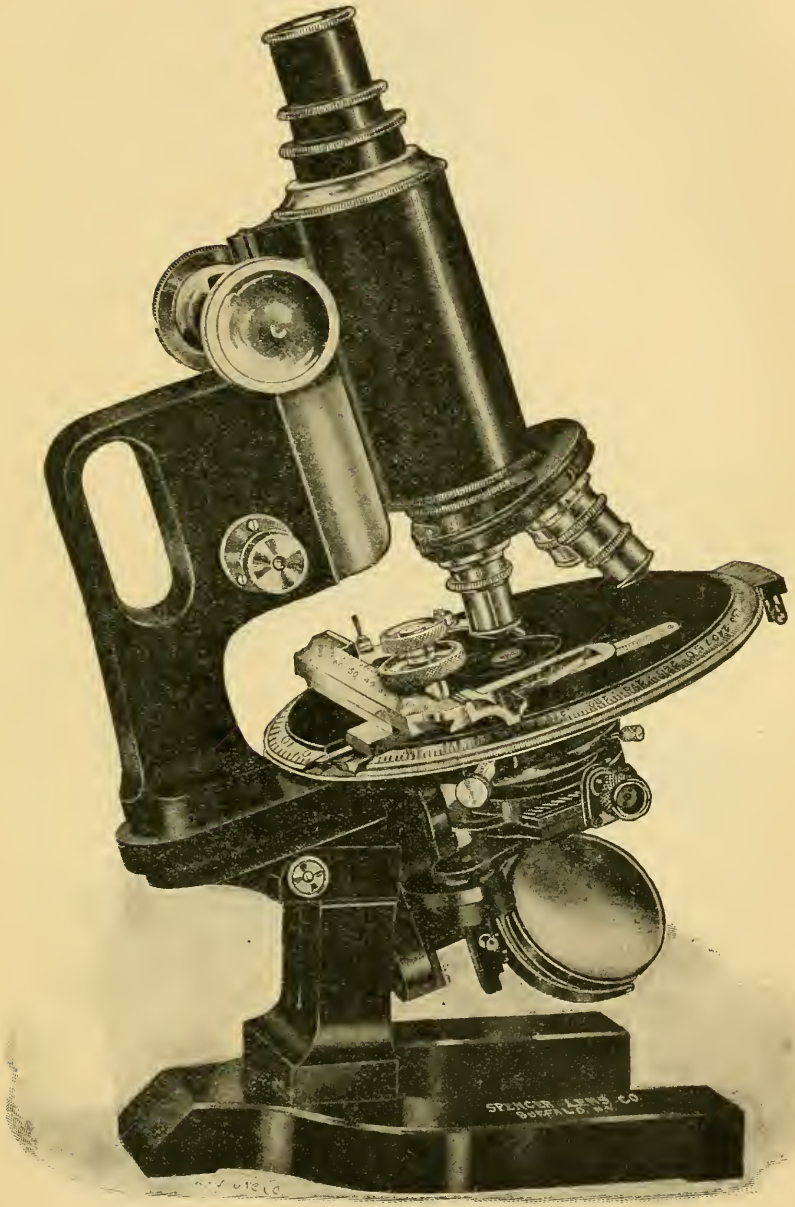

FIG. 35.

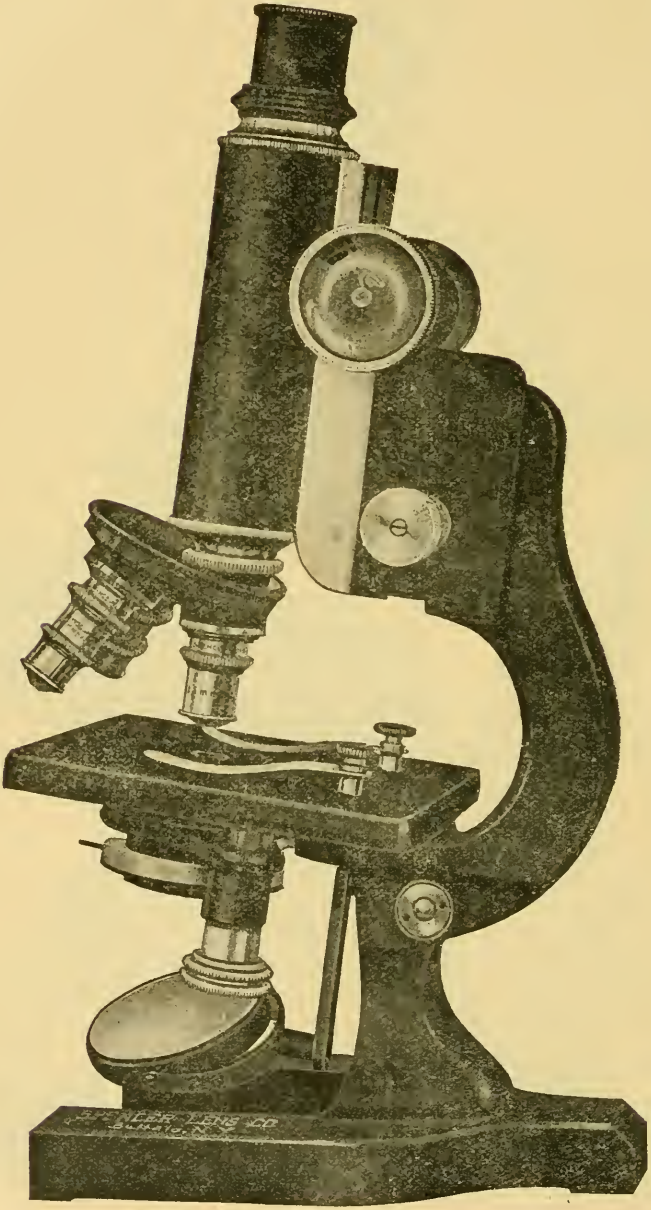

Fig. 36.

New Spencer Microscope, No. 44.*—This instrument (fig. 36) is intended to be an efficient and comparatively inexpensive type with side fine-adjustment. The arm is large, with a convenient grasp for handling the instrument, and provides a distance of 80 mm. from the arm to the optical axis. The fine-adjustment is of simple type; the thread-bearing shaft has buttons on both ends and passes back and forth through the arm, carrying with it the free end of the longer arm of a bell-crank lever, the shorter horizontal arm of which raises and lowers the body-tube 0·2 mm. for each complete revolution of the shaft. The vulcanite-covered stage is 112 mm. wide and 108 mm. deep, and is provided with a sub-stage of the quick-screw type; the upper iris diaphragm is automatically locked open when the condenser is put into place.

(3) Illuminating and other Apparatus.

Improved Apparatus for Dark-ground Illumination in the Early Diagnosis of Syphilis, etc.†—C. H. Mills describes an apparatus (fig. 37) which at the present time meets the need for a really satisfactory plant for the detection of the *Spirochæta pallida* in the early diagnosis of syphilis. Now, the most rapid and accurate method of detection is by means of the dark-ground apparatus, but the light which is used with this often fails to give sufficient definition. A good knowledge of optics will, no doubt, enable a comparatively feeble source of illumination to be used with satisfaction, but in routine work we do not desire to practise optics so much as to see the *S. pallida*.

With the object of obtaining a thoroughly reliable illuminant, which is easily manipulated and always available without too many preliminaries, the author has for some time been working upon a design which is now completed and accessible to all, being of British manufacture and at a reasonable price.

The following are the special features of the apparatus: 1. Extreme compactness and reliability. 2. Always ready for immediate use, and yet, if so desired, can be returned to the cupboard from consulting-room table or laboratory bench without being dissembled. 3. The hand-fed compensating arc lamp requires the minimum of adjustment. 4. Suitable for use on any existing electric supply—continuous or alternating current—coming within the range of 100–250 volts. 5. Contact is made from a wall-plug or the bayonet fitting of an ordinary electric lamp. 6. The arc projects a good steady light with ample reserve, rendering differentiation between *S. pallida*, *S. gracilis*, *S. microdentium* accurate, rapid, and comparatively easy. 7. Adapted for any type of Microscope stage. 8. The price is reasonable, and the upkeep, even for constant daily use, inexpensive. The carbons for the

* Catalogue, Spencer Lens Company, p. 35.
† Lancet, October 21, 1916, p. 716.

arc lamp are readily obtainable. 9. Each constituent part is capable of an ample range of movement, thus rendering perfect adjustment very simple.

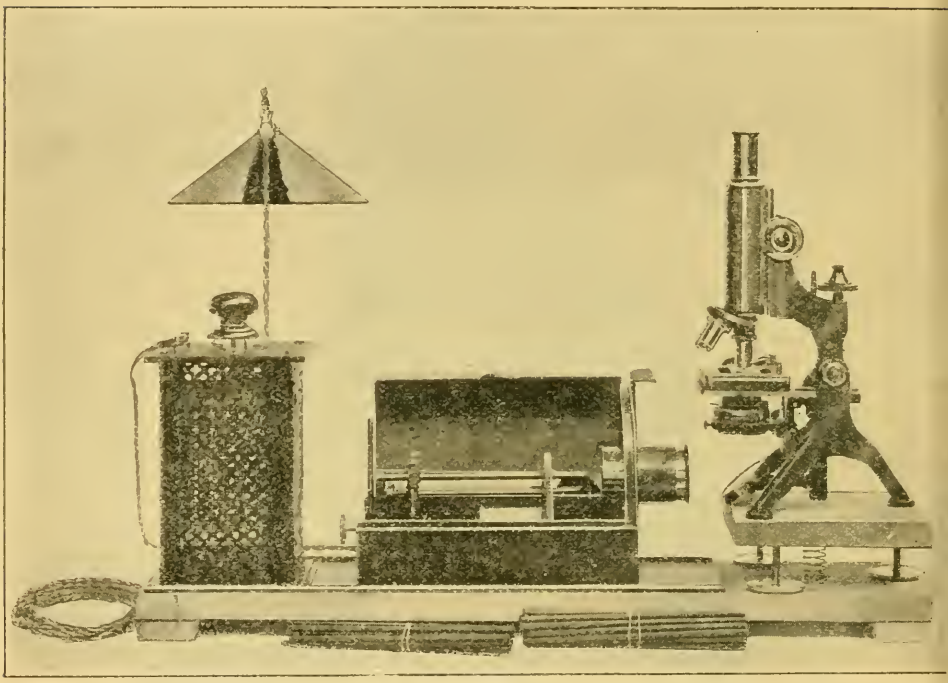

FIG. 37.—On the left: resistance. In the middle: hood of cover laid back to expose the compensating arc lamp, which travels in exact alignment along runners on the base-board. A lens set in an adjustable sleeve is seen projecting. On the right: adjustable stand for the Microscope.

(4) Photomicrography.

Wratten Light Filters.*—The Kodak firm have investigated for the purpose of light filters an enormous number of organic dye-stuffs, with which they have been able to prepare a long series of filters of a purity and brightness said to have never before been obtained. These filters are supplied as dyed gelatin film, and are prepared by coating gelatin containing a given weight of dye upon prepared glass, and, after drying, stripping the film from the glass. They are standardized by comparison with a standard whose absorption curve has been measured on a spectro-photometer. The catalogue list comprises nearly

* Wratten Light Filters, Kodak, Limited, London, 1916.

100 varieties ; it also shows their spectro-photometric absorption curves, and also gives their percentage transmission. Some new filters are introduced, including a photometric set, an ultra-violet, and an infra-red.

Photography of Coloured Objects.*—The second edition of this work includes most of the matter included in the previous edition, together with an incorporation of much of that contained in the Wratten book on "Orthochromatic Filters."

(5) Microscopical Optics and Manipulation.

Drawing of Microscopic Preparations.†—A. Maillefer describes a method of drawing microscopic images with the help of a reflecting prism in a sort of dark chamber. The light from a Liliput electric lamp after passing through a water-cooler traverses an aperture in the wall of the dark chamber and falls upon the objective of the Microscope ; on issuing from the eye-piece a totally reflecting prism projects the image vertically downwards on to the paper placed on the working table. The dark chamber is merely a light framework covered with opaque black cloth, the whole being of such a size as to contain the instrument and the operator. The cloth must be loose and so arranged as to enwrap the operator completely and prevent light from entering the chamber from any direction. Dimensions and details are fully given, and the author expresses great satisfaction with the arrangement.

(6) Miscellaneous.

Optical Glass.‡—W. Rosenhain, in his first Cantor Lecture on the above, points out that the term "optical glass" should be used in such a way as to mean a substance distinguished from even the best of ordinary glass by a whole series of important properties, which can, however, be summed up in two groups: (1) properties relating to "general quality," in regard to which "optical glass" is simply a "better quality" than ordinary glass ; and (2) properties of a more specifically optical kind, relating to the refractive and dispersive powers of the glass. Under the first head the subjects of transparency, colour, devitrification, striæ, interaction between the glass and the crucible, internal strain, hardness, and durability, are discussed in a very instructive and interesting way. Under the second head the lecturer treats of refraction, dispersion, and the conditions necessary for achromatic and apochromatic combination.

The second lecture deals with the present modes of optical glass manufacture, and gives the reader some insight into the physical and

* The Photography of Coloured Objects, 2nd ed. Kodak, Limited, London, 1916, 118 pp. (63 figs.).

† Bull. de la Soc. Vaudoise des Sciences Naturelles, li. (1916) pp. 1–7 (1 fig.).

‡ Journ. Roy. Soc. Arts, lxiv. Nos. 3324-6 (Aug. 1916).

chemical difficulties involved. Not the least of these is the selection of materials for the manufacture of the melting-pots; the preparatory seasoning of the clay extends over months, or even years; even when at last moulded the pot requires a further seasoning of some six months. The ingredients of the glass interact upon one another and also upon the pot, which is liable to be rapidly corroded away. The furnace gases are extremely active and introduce many complications. The utmost degree of purity is required in the constituents of the glass. A difficulty from the British point of view is that no bed of sufficiently pure sand exists in Great Britain; but perhaps research might discover one somewhere in the British Empire. The sand required is, therefore, imported into England from Fontainebleau, near Paris. Germany is fortunate in possessing several good sources of pure sand, one of them having a silica-content of 99·98 p.c.

In the third lecture the author discusses possible directions in which the manufacture of British optical glass might be improved and developed so as to make this country independent of German competition. As many of the difficulties are connected with the chemical action of the furnace gases acting on the contents of the pot, and, in many cases, even through the pores of the pot-walls, the author discusses the possible adaptation of a suitable form of electric furnace, in which furnace gases would, of course, be totally absent. Then, if some material could be found suitable for the pot itself which did not chemically react upon the glass, an enormous piece of progress would result. Then there is the question of whether glass is the only crystalline substance suitable for lenses. The author thinks that in this last direction modern research on the synthetic production of suitable crystalline material may come to the rescue. At any rate, for an investigator approaching this subject with adequate resources, a very wide and promising field lies open.

B. Technique.*

(1) Collecting Objects, including Culture Processes.

Automatic Delivery Apparatus for Fluid Media.†—S. W. Cole says that during the course of bacteriological work undertaken for the Medical Research Committee it was necessary to tube a given amount of broth. For this purpose he devised a piece of apparatus. As will be seen from the sketch (fig. 38), it consists of a stiff rubber ball (D) attached by rubber tubing to a glass part, which contains two light glass valves, accurately ground in. The fluid is placed in the beaker and the rubber ball is squeezed by hand as much as possible. On

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

† Lancet, Oct. 21, 1916, p. 716.

allowing the ball to expand fluid is sucked up past the valve (B). On repeating this operation two or three times the air is driven out and the whole of the apparatus is filled with the fluid. The ball is then compressed by means of the board, and the position of the screw (C) is found which results in the delivery of the desired amount. Delivery should be made by an even pressure applied as uniformly as possible. It is important to keep the length of the rubber tubing as short as possible and to use thick-walled pressure tubing. Under these conditions the volume delivered is almost independent of the rate of compression of the ball.

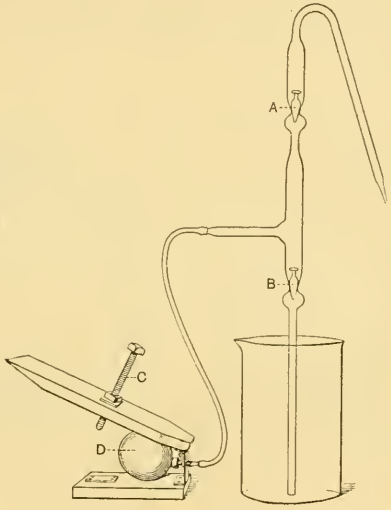

Fig. 38.

Handling Protozoa in Pure Line Work.*—During the past year R. T. Hance has been engaged in experiments on the inheritance of extra contractile vacuoles in a new race of Paramœcium, and has worked out some methods of technique that have much facilitated his work, e.g. maintaining pure cultures. The greatest care is necessary to prevent pure line cultures from becoming mixed with others. Even with labelled pipettes accidents may occur. The scheme shown (fig. 39) was recently devised and has proved most convenient. A piece of soft brass wire is shaped about some round object of a diameter slightly larger than a pipette, and is held by several twists. Then the long ends of the wire are bent around the culture jar and again fastened by twisting the ends. In the jars used there is a convenient groove near the top into which the wire fits nicely. When finished the small circle

* Trans. Amer. Micr. Soc., xxxv. (1916) pp. 135–6 (1 fig.).

protrudes from the jar, and into this ring the pipette is dropped, giving the appearance seen in fig. 40. With this method pipettes are always at hand and there is no danger of mixing the lines by transferring animals from one culture to another.

Preparation of Watch-glasses.—Syracuse watch-glasses have been used for single individuals throughout the work, and considerable difficulty was experienced at first in locating animals which were close to the edge of the container. They frequently found their way there, as the fluid had a tendency to spread evenly over the surface of the watch-glass. The best method to obviate this was hit on accidentally. There was a trace of paraffin in a pan in which the glasses were being sterilized one day, and this coated the glasses imperceptibly, but sufficiently to give the liquid no hold on the glass. In vessels treated in this way the surface tension of the medium tends to draw it into a

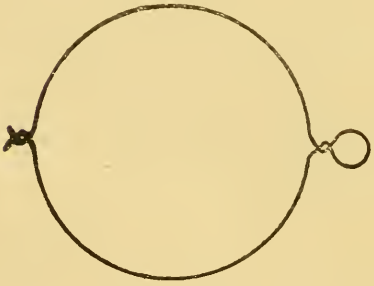

FIG. 39.

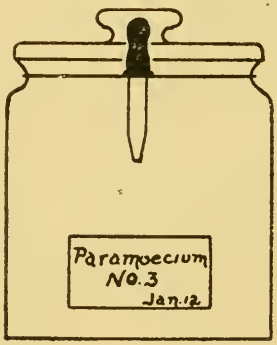

FIG. 40.

spherical mass. Should the liquid roll to the edge of the glass, where the animals would be hidden from view, it is easily rolled out again by tilting the glass, and none of the animals in the drop are left behind. When animals are being kept in very small drops of water, as many as twenty individual drops of liquid containing Protozoa may be placed in a single watch-glass and they have not ran together. The surface tension of the liquid draws it up when on a paraffined surface until it gives a very fair picture of a drop of mercury. Furthermore, being contracted to the smallest area possible, there is less evaporation than when the same amount of fluid is spread out, and the chances of losing a valuable specimen through drying are much less. The writer's practice is to use a piece of paraffin about the size of a pea to a quart of water. This will be sufficient for a surprising number of watch-glasses. When the sterilized glasses are removed they are wiped while hot and polished. No paraffin is visible, although a faint trace of it can be felt.

Collecting and Rearing Volvox.*—G. R. La Rue collected Volvox in the autumn of 1915, and in the spring of 1916 found they had multiplied freely. He gives certain cautions—e.g. the water for the cultures should be from the same source as the organisms. Tap-water for making up the culture or for making good evaporation should never be used. Keep the cultures covered to prevent evaporation and consequent change in density of the medium, and to exclude dust and bacteria. The presence of organic material seems to be beneficial. Direct sunlight is unnecessary and is to be avoided, because it causes too great variations in temperature in closed vessels. North light is good; in fact many algæ thrive in it. Low temperature, above freezing, seems to favour development. Old cultures, unless hopelessly foul, should be kept, and the organisms given a chance to reappear.

New Solid Medium for the Isolation of the Cholera Vibrio.†—H. G. Gibson gives the following formula for the preparation of this medium: Agar, 3 grm.; pepton, 10 grm.; starch, 10 grm.; sodium bicarb., 1·5 grm.; litmus (sufficient to colour medium); water, 1000 c.cm. Weigh out 30 grm. of powdered agar and emulsify with 250 c.cm. of cold water. Then weigh out 10 grm. of pepton and 1·5 grm. of sodium bicarb. Mix together and emulsify in another 250 c.cm. of cold water. The two emulsions are then mixed in a two-litre flask and 500 c.cm. of water added. The solution is completed in a steamer. When dissolved the medium is clarified with white of egg and filtered in the steamer. Weigh out 10 grm. of potato starch, emulsify it with some of the filtered agar, and add the emulsion to the remainder of the medium. The whole is sterilized by the fractional method, after which enough sterile aqueous solution of litmus is added to bring about a blue colour of the medium. The final reaction of the medium should be: 2 to phenolphthalein; 0·15 p.c. sodium bicarb. gave the best results.

Examination of the plates eighteen hours after inoculation showed the cholera colonies to be pink, the other colonies to be blue or whitish. In twenty-four to twenty-six hours the cholera colonies are pink, with a faint pink halo. In forty-eight hours other colonies may be pink, but the cholera colonies may be distinguished by their red centres which the other colonies lack. The only other organisms which are known to acidify starch are some of the diphtheroia group and some of the non-pathogenic water vibrios. Gram's stain and the serological test dispose of these organisms.

Acidification of Culture Media by Alkaline Salts.‡—L. Bourdet has investigated the question of the acidification of culture media by means of their contained alkaline salts during sterilization in the autoclave. The acidification is produced by action of certain salts upon sugars and peptons contained in the different media: for example, the

* Trans. Amer. Micr. Soc., xxxv. (1916) pp. 150–4.
† Brit. Med. Journ., Sept. 30, 1916, pp. 454–5.
‡ C.R. Soc. Biol. Paris, lxxix. (1916) pp. 665–8.

alkaline salts of milk produce formic acid at the expense of the lactose when milk is heated to 130° C. The differentiation of organisms by means of sugar media is rendered difficult by this fact, a slight degree of fermentation being simulated. Bourdet recommends in practice that (1) broth media should be sterilized by filtration after the addition of the pepton and the alkali; (2) separate sterilization of the sugars required for the media, and their aseptic admixture with the broth media; (3) the separate sterilization of the litmus solution.

Culture of Diphtheria Bacilli in Veillon's Tubes.*—L. Martin and G. Loiseau recommend the following technique for the differentiation of true diphtheria bacilli from pseudo-diphtheria bacilli. It is based on the fact that *Bacillus diphtheriæ* is a facultative anaerobe, while the other organisms are strict aerobes.

The medium employed is prepared as follows: 250 grm. of minced veal added to 500 c.cm. water are mixed with equal parts of "bouillon de panse (pepton martin)." To each litre of this mixture is added: Agar 8 grm. glucose 15 grm., and potassium nitrate 2 grm. Dissolve and add the white of one egg, heat to 115° C. for half-an-hour, filter, tube, and sterilize for half-an-hour at 100° C., and, during the three days following, for half-an-hour at 115° C.

A pure colony of the organism to be investigated is emulsified in a tube of 10 c.cm. broth. The organisms are then well distributed by shaking. Add 1 c.cm. of this emulsion to a tube of the glucose agar medium (previously boiled and cooled rapidly to 50° C.) and mix carefully. Incubate when solid. True diphtheria bacilli are easily identified, as the colonies are evenly distributed throughout the tube without prominence in the aerobic zone, while colonies of *B. Hoffmann* and other diphtheroids only grow in the superficial zone, and not in the strictly anaerobic portion of the medium; moreover, the growth is less rapid than with the diphtheria bacillus, and is not generally visible for at least twenty-four hours.

Digested and Diluted Serum as a Substitute for Broth.†—A. Distaso suggests the use of the following medium in the place of ordinary broth in bacteriological work: (1) One volume of sheep or ox serum is mixed with one volume of tap-water, and boiled till it becomes milky. (2) A pig's pancreas is minced and extracted with 400 c.cm. of distilled water in the presence of chloroform for twenty-four hours; and (3) a piece of the upper part of the small intestine is extracted in the same way, in order to activate the pancreatic extract. To one litre of (1) is added 100 c.cm. of (2) and 10 c.cm. of (3), and digested at 60° C. for the night. Next morning the flask contains an amber-coloured liquid with fine flocculi floating in it. Filtered through Chardin paper, the amber-coloured liquid passes through, and the flocculi remain in the filter. The liquid is collected and sterilized at 120° C. for fifteen minutes, then tubed and re-sterilized. In this medium the

* C.R. Soc. Biol. Paris, lxxix. (1916) pp. 677–80.
† Brit. Med. Journ., 1916, pp. 555–6.

growth of the *Bacillus coli* group, the streptococci, *B. subtilis*, *B. proteus*, and *fluorescens*, is so luxuriant that in comparison normal broth may be said to give a scanty growth.

(3) Cutting, including Embedding and Microtomes.

Spencer Automatic Laboratory Microtome.* — This apparatus, numbered 880 in the maker's catalogue, is shown in fig. 41. In this microtome the main supporting frame has a heavy clamp at the back by which it is securely fastened to the laboratory table. As will be seen from the figure, the upper part of this frame forms a support to which the two laterally swinging arms are attached by steel pivot screws with check nuts. The knife-carrier is held by these swinging arms at their outer ends, attached thereto by similar pivot screws with check nuts, and in order to give the proper movement, relieved from any pressure or strain, a detachable flexible lever handle is attached to the axis fastened to the longer arm on which the arm swings. The extreme ends of the knife rest in the holder, and, as the lever moves the swinging arms, the blade describes the flattened curve, corresponding to the double movement in free-hand sectioning. By this manner of holding the knife by arms, which are not parallel, the entire length of its cutting edge is utilized, insuring uniform wear and permitting the cutting of larger sections than has heretofore been possible, except by using a very much larger blade. The swinging arms and knife-holder are sufficiently rigid to avoid any deflection of the knife in its movements; thereby assuring uniformity of thickness in all the sections. This peculiar motion of the knife makes this microtome especially desirable for cutting frozen sections. Sections may be cut, stained and mounted in one and one-half minutes from the time the tissue is placed on the freezing plate. This microtome does excellent work with celloidin also. The movement of the knife is not so well suited to paraffin work, but it does very well with this medium.

Another important advantage in this method of construction is its convenience to the user. In most microtomes with sliding parts, these require frequent lubrication with oil, and in consequence are liable to be clogged with dirt, requiring frequent cleaning, and also to become loose and shaky. In this microtome, lubrication is not required, and in its construction every joint is provided with pivot screws and check nuts, and by means of a steel pin sent for the purpose the owner may take the instrument apart and adjust or clean it as occasion may require. The object-clamp for paraffin or celloidin blocks has a pin or round shank which may be fastened by a clamp screw in a vertically movable socket which is supported by two vertically swinging arms attached at the back to the main frame by hardened steel pivot screws. Similar pivot screws at the front hold the socket in the swinging arms; thus providing for its vertical movement the same parallelogram principle as is applied to the knife. By this means, a steady movement is secured and the top of the object always remains in the same horizontal position;

* Catalogue, Spencer Lens Company, p. 113.

thus further insuring uniformity of thickness in the sections. The object-clamp-supporting socket is raised or lowered by a vertical feed

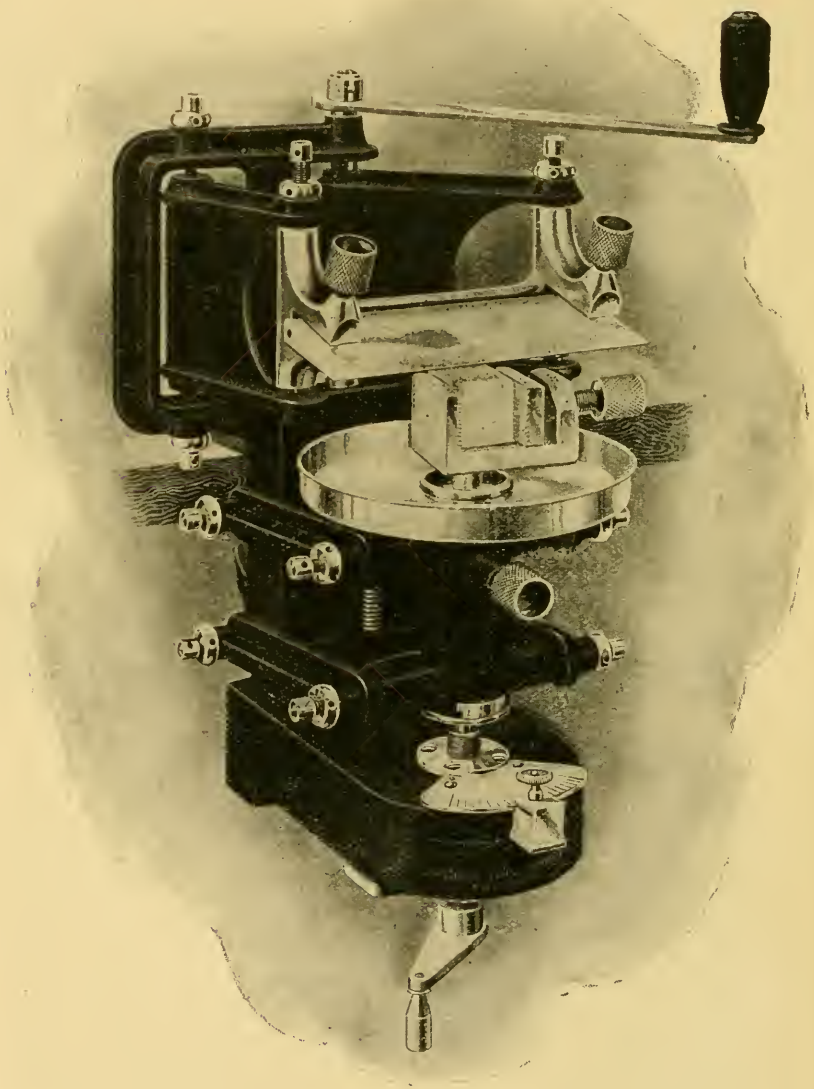

Fig. 41.

screw with fine micrometer thread, having a crank at the bottom for independent movement. This screw is firmly held in double nut

bearings which provide for taking up lost motion and for eliminating any wear that may occur. At its upper end this feed screw is connected with the object-supporting socket by means of a ball-socket bearing, to which the object-support is firmly held down by a strong steel spiral spring. The automatic feed mechanism consists of an accurately cut ratchet wheel, keyed to the vertical feed screw, in the teeth of which a hardened steel pawl engages. By means of a lever, extending to the graduated scale shown on the front of the main frame base, this pawl may be thrown out of action by turning the index finger to the extreme left, or it may be set to cut sections of any desired thickness. Each division of the graduated scale marks 5 microns. A long, vertical shaft, extending to the lever arm at the top of the main frame and connected at the bottom by a simple lever device, moves the pawl backward and forward simultaneously with the movement of the knife, so that wherever the index is placed on the graduated scale, the object is correspondingly raised at each swing of the knife. The crank at the bottom can be used independently of this automatic motion and provides for quickly raising or as quickly lowering the object whenever desired. The whole feed mechanism is simple in construction, and, being covered by the extended base of the main frame, is protected from dust and drippings. It may easily be reached from below and it cannot easily get out of order. A convenient drip pan, at the top of the socket in which the object-clamp sets, may be quickly unscrewed and as easily replaced. It is best to remove it when the freezing chamber is attached. This is heavily nickel-plated, as are all the screws and exposed parts.

Embedding in Paraffin.*—R. T. Hance says that when embedding very small objects, such as insect larvæ or small flowers or anthers, in paraffin it is most convenient to orient them one behind the other. This method allows a single block to be made of three or four to a dozen pieces of tissue, and these may be cut in one ribbon. This obviously eliminates a great deal of the labour in making a block for each separate object, cementing it to the holder, trimming it, and adjusting the microtome each time. In the ribbon it is easy to see where one piece of tissue ends and the other begins, as there are usually several blank sections of paraffin between them. It is relatively simple to arange the tissue in line under a carbon bulb with warm needles, but a difficulty is met with when an attempt is made to place the paraffin mould in water for cooling. The material is shaken from position, and must be re-oriented. This has been overcome in the following way. A watch-glass is used as a mould for embedding small objects, and a Petri dish is convenient for larger tissue. When the tissue is ready to be embedded, the dish is heated to the melting-point of the paraffin under the electric bulb. It is then placed in a crystallization dish, with two slides beneath it to prevent it from touching the bottom of the container. Paraffin is then poured into the small dish, and the objects oriented as desired, the heat of the electric bulb keeping the paraffin melted. Then the light is turned off, and cold water is poured into the

* Trans. Amer. Micr. Soc., xxxv. (1916) pp. 137–8.

crystallization dish. Since the dish containing the paraffin is raised from the bottom the water flows under it, and soon solidifies the paraffin in the lower part of the dish, which consequently holds the object fast. As soon as a surface film is formed, enough water can be added to cover the embedding mould to complete the hardening of the paraffin. In Petri dishes or watch-glasses the bottom is practically flat and true, and the tissue is allowed to sink to the bottom. When the tissue is cut out as a block, the part that rested against the bottom makes one of the two parallel sides, and requires little or no trimming.

When a number of pieces of tissue or a number of series are embedded in one disk of paraffin, it is dangerous to attempt to separate them with a knife, as one can never be sure of the direction the crack in the paraffin will take. The writer has found that a hand scroll-saw or coping-saw does admirably for cutting a block of tissue from the main disk. The use of the saw permits many more pieces to be placed in the same space, and no care need be taken to have well-defined pathways for the paraffin to split along, as is necessary when a knife is used for separating the pieces.

New Embedding Stage.*—G. R. La Rue says the essential parts of this embedding stage (fig. 42) are a transite base, 17¾ in. long by

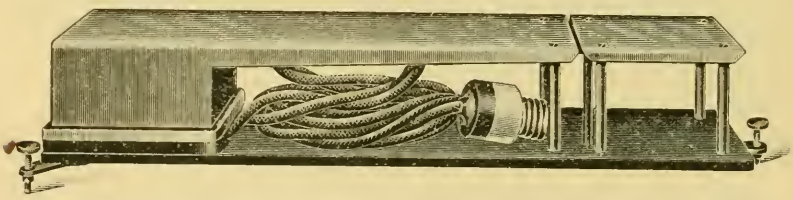

Fig. 42.

½ in. wide, mounted on three levelling screws, a copper stage made in two parts, 4 by 13 in. and 4 by 4 in. respectively, and under one end of the longer copper stage an electric heating unit. The heating unit may be wound for any voltage and to yield any desired temperature. No regulator or rheostat or other provision for controlling or varying the temperature is provided, but since the coil is situated under one end of the stage lower temperatures may be secured by moving the object away from the coil. In practice the coil is attached to a convenient electric receptacle near the paraffin bath, and that part of the stage over the coil is heated sufficiently to melt paraffin in a few minutes. The embedding tray may now be warmed over the hot stage, filled with melted paraffin and moved to a point on the stage where the paraffin is kept just melted. Objects to be embedded are now transferred to the embedding tray, oriented, and the label inserted at the end of the tray with the legend towards the margin of the tray. Now the tray is gently moved to the unheated end of the stage, where the paraffin is permitted to congeal on the bottom sufficiently to hold the objects in

* Trans. Amer. Micr. Soc., xxxv. (1916) pp. 154–5 (1 fig.).

place. Then the tray is transferred to a dish of cold water or alcohol standing at the end of the embedding stage, and into which it is immersed as soon as the paraffin is cooled sufficiently to prevent the breaking of the surface by the water.

The use of this embedding stage secures good embedding, because the paraffin is melted clear to the bottom of the tray, and thus orientation is made easy.

(4) Staining and Injecting.

Method of Staining Flagella.*—L. Tribondeau, in collaboration with M. Fichet and J. Dubreuil, has evolved a simple procedure for staining bacterial flagella. The bacteria are grown on Martin's agar 2 p.c. (slightly alkaline) in Petri dishes. After twelve hours' incubation a colony is picked off and emulsified in distilled water. Cover-slips are boiled in bichromate of potash 50 grms., sulphuric acid 100 grms., water 1000 grms., and then washed in tap-water ; dry and flame in the Bunsen burner, wash the flamed surface with distilled water, and dry vertically without wiping. A drop or two of the bacterial emulsion is allowed to spread itself over the cover-slip, and when the film is dry it is fixed rapidly with absolute alcohol. Staining : Solution of tannin, 10 parts per 100 in distilled water 1 part, saturated solution of potash alum in distilled water 2 parts. Boil rapidly, add 0·5 c.cm. alcoholic solution of crystal violet (stock solution, 2 parts in 10 of alcohol 1 part, absolute ethyl-alcohol 10 parts), mix and bring again to the boil, mix again and cover the surface of the films very rapidly and allow to act for from fifteen to thirty seconds. Wash rapidly under the tap, dry and examine with the oil-immersion lens. The flagella are coloured blue-violet.

Cultural Vital-staining of Bacteria.†—T. Iwao says that the following medium gives good results : To hot filtered agar, made alkalin with sodium carbonate, are added 0·3 of "eosinsaures" methylen blue. The agar solution is carefully shaken and then distributed in test tubes, 10 c.cm. in each. After this they are sterilized for one hour at 100° C. The staining of the bacteria, especially of the coli group, is good, the granules coming out well.

(5) Mounting, including Slides, Preservative Fluids, etc.

Slide for Examining Small Pond Life.‡—E. M. Nelson describes a new form of slide, and also describes the apparatus required. First, an oil-immersion sub-stage condenser on dark-ground illuminator. Second, two slips, 3 by $1\frac{1}{4}$, cemented along their bottom edges to make a ledge. A drop of the gathering is put with a pipette in the

* C.R. Soc. Biol. Paris, lxxix. (1916) pp. 710-16.
† Acta Schol. Med. Univ. Imp. Kioto, i. (1916) pp. 251-62 (1 pl.).
‡ English Mechanic, Sept. 29, 1916, p. 191.

hollow and a large cover-glass is placed over it (Fig. 43). If the cover-glass projects just over the top of the slip it is easy to lift it off. When the slip, duly charged with the gathering, is on the stage of an inclined Microscope the cover-glass will not slip off, because it will be held by the ledge, neither will the small excess of water, squeezed out between the cover and the slip, run down on to the stage, because the ledge will catch it.

The writer, who is indebted to W. Chaffey for this simple and excellent device, has now used it for some time in preference to the different forms of compressors that are made. One point is that these slides should be of a proper thickness to suit the focus of the sub-stage illuminator ; for if the focus of the illuminator is long and the slide thin there will be constant trouble, because the oil will run away, and a slip-equalizer must be provided. If the slip is thick and the focus of the illuminator be short the illumination will be very poor indeed. It is better, therefore, in the first instance, to find the precise working

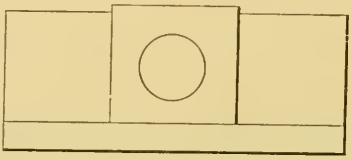

Fig. 43.

distance of the illuminator and provide a slip of a proper thickness to suit it, then the maximum amount of illumination will be secured and no further trouble experienced. Note, with the above objectives and deep eye-pieces of 1 or a ½-in. focus the focused image of the edge of the flame of a paraffin Microscope lamp (used direct if the Microscope stands high on its trunnions, but with the plane mirror if low) will yield a very satisfactory and critical illumination.

Method of Making Toto Mounts of Unicellular Forms.*— R. A. Nesbit describes the following procedure : The material is killed and fixed in whatever solution the investigator has found most satisfactory for the particular Algæ or Protozoa with which he is working. It is washed in bulk in the usual manner and carried through upgraded alcohols as far as 60 p.c. It is allowed to settle completely in this grade. A thin layer of albumen fixative is smeared on thoroughly clean slides. A drop of the material is then drawn up with a pipette and placed on the slide. The alcohol coagulates the albumen and causes the cells to adhere to the slide. They may then be dipped into 60 p.c. alcohol and afterwards to upgraded alcohols. It is possible to use such stains as Flemming's and iron-hæmatoxylin rapidly and with precision. Before using Flemming the cells must be hardened in 95 p.c. alcohol.

* Trans. Amer. Micr. Soc., xxxv. (1916) p. 140.

(6) Miscellaneous.

Cleaning used Microscope Slides.*—J. T. Illick gives the following method : A liberal amount of gold dust and a number of slides were placed in water and thoroughly boiled. As soon as the cover-slips came off of their own accord the slides and slips were placed in a pan of water. These were wiped dry while others were being boiled.

Metallography, etc.

Alumina in Steel.†—The question of the occurrence of alumina inclusions in steel and their detection by the Microscope has been studied by A. Sauveur, by (1) producing thermite iron with an excess of aluminium ; (2) melting ingot iron with aluminium ; (3) melting ingot iron with alumina. In each case polished but unetched sections of the metal exhibited under the Microscope, at a magnification of 300 diameters, small, dark and roughly rounded particles scattered throughout the iron. Characteristics of alumina inclusions, by which they may be distinguished from other non-metallic inclusions generally occurring in steel, are their dark colour, small size and complete absence of any elongation in the direction of rolling or forging. This agrees with the known brittleness and infusibility of alumina.

Study of Cooling Rates with a Chromium Steel.‡—C. A. Edwards heated cubes of a steel containing 0·63 p.c. carbon and 6·15 p.c. chromium to 1147° C., and cooled at varying rates. A critical rate of cooling was found—namely, twelve minutes sixteen seconds—from 836° C. to 546° C. For all slower rates of cooling, recalescence occurred during the cooling, and the hardness produced was very similar. When the rate of cooling is quicker than the critical rate, the thermal change is much modified, or, with very quick cooling rates, repressed altogether, and the steel is progressively hardened, according to the quickness of the cooling. The critical cooling-rate varies with the initial temperature of cooling. The higher the initial temperature, the slower is the critical rate of cooling. Photomicrographs of structures of the steel corresponding to different rates of cooling are given. Samples cooled at rates slower than the critical rate, and therefore not self-hardened to any appreciable extent, consisted generally of troostitic pearlite, with very little martensite. Samples cooled quicker than the critical rate contained large quantities of martensite, which is responsible for the self-hardening. When the cooling was sufficiently rapid to suppress altogether the thermal change and produce maximum hardness, the structure was all martensitic.

Corrosion of Steel.§—L. Aitchison discusses the mechanism of corrosion in pure iron and the various kinds of steel. In steels

* Trans. Amer. Micr. Soc., xxxv. (1916) p. 141.
† Metall. and Chem. Engineering, xv. (1916) pp. 149–51 (4 figs.).
‡ Journ. Iron and Steel Institute, xciii. (1916) pp. 114–40 (8 figs.).
§ Journ. Iron and Steel Institute, xciii. (1916) pp. 77–91 (6 figs.).

corrosion is considered to be an electrolytic action between the carbides and the carbon-free material (ferrite or solid solution) which together constitute the structure of the steel. The carbides act as anodes of the electrolytic couples, and are therefore preserved while the ferrite or solid solution is attacked, and passes into solution in the corroding medium. Photomicrographs of (1) steel containing 1·25 p.c. carbon, and (2) steel containing 0·73 p.c. carbon and 21·5 p.c. tungsten, after corrosion are given to illustrate this point. Each shows massive carbide intact. In a similar way, pearlite does not corrode as a whole, but as a mixture of ferrite and carbide, and only the ferrite is attacked. The disappearance of the carbide from pearlite which usually occurs in corroding pearlitic steels is due to mechanical loss. In the case of pure iron, consisting of ferrite only, electrolytic action between the ferrite crystals and the intercrystalline amorphous cement is put forward as the chief cause of corrosion. Here the amorphous material acts as kathode, and passes into solution more rapidly than the ferrite. Evidence of this is shown in a photomicrograph of pure iron after corrosion in sodium chloride solution, showing accelerated action along the crystal boundaries. The concentration of the solid solution is the controlling factor in deciding the rate of corrosion of a steel, since the higher the concentration the lower is the electromotive force set up by the solid solution in a corroding medium. The properties of special steels are reviewed, and shown to confirm these views. Elements like molybdenum, vanadium, and tungsten, which form part of the carbide and do not enter into the solid solution until a high percentage is present, exert little effect on the rate of corrosion, while elements such as chromium and nickel, which enter into solid solution from the first, retard corrosion.

Surface Tension Effects in Metals.*—A theory, based upon the existence of films of metallic amorphous material or cement between the crystalline grains of metals, is elaborated by F. C. Thompson. This amorphous material is regarded as an under-cooled liquid, and the deduction made that surface tension forces operate between crystals separated by thin films of such material. The great and unexpected strength of the crystal junctions in ductile metals is associated with these surface tension forces. The growth of crystal grains at high temperatures during annealing is explained by the endeavour of these surface forces within the mass of metal to reduce the area of the intercrystalline boundaries. The mechanical and other properties of metals are considered and interpreted in the light of these ideas. The elastic limit of a metal is reached when the attraction of the surface tension forces over a given area of a specimen under test is just overcome. The larger area over which surface tension attractions can occur provide an explanation of the well-known fact that a fine crystal structure possesses a higher elastic limit than a coarse crystal structure. The equation $E = \frac{2T}{d}$ (where E = elastic limit, T = the surface tension of the amorphous material, d = thickness of the film) is deduced. Accurate

* Journ. Iron and Steel Institute, xciii. (1916) pp. 155–92 (7 figs.).

determinations of the elastic limit of a series of pure metals were made, while values for the surface tension of the amorphous material, based upon measurements of the surface tension of the molten metal at its melting-point, are deduced. Applying these results in the above equation, values are obtained for the thickness of the intercrystalline films in the various pure metals examined. An application of the theory is illustrated by a study of a steel shaft which had been drastically over-annealed and showed very low elasticity. Samples of the same steel when heat-treated by (1) normalizing at 850° C., (2) heating to 850° C., quenching in oil and tempering at 350° C., showed similar elastic limits, each much higher than that of the over-annealed sample. Photomicrographs of the steel in the three states are given, showing a large crystal size for the over-annealed and a much smaller and about equal crystal size for the two other samples. The calculated thickness of amorphous film is four times greater in the over-annealed sample than in the two others, and its low elasticity is readily understood, since the thicker the film the lower the magnitude of the surface tension forces. The tensile strength of the three samples was of the same order. The factors governing this property depend on the crystals themselves, and not, as in the case of the elastic limit, on the nature and extent of the intercrystalline junctions.