TO THE READER.

THIS work may be "read" by carefully studying the figures, and then referring to the text. A description of every drawing is placed beneath it, and, in most instances, a reference is given to the very page upon which the subject of the drawing is considered. A teaching experience, extending over more than five-and-twenty years, has convinced the author that, although the student will certainly obtain more correct views upon the microscopic characters of objects by attentively examining accurate representations, than by reading over and over again the most minute and elaborate descriptions of them, the information thus gained will be of little real use unless the student himself prepares and examines actual specimens, and makes careful drawings of what he sees.

On p. 429, *for* 1-1,000,000th, *read* 1-100,000th.

HOW TO WORK

WITH THE

MICROSCOPE.

BY

LIONEL S. BEALE, F.R.S.,

PRESIDENT OF THE ROYAL MICROSCOPICAL SOCIETY.

1880

FIFTH EDITION,

REVISED THROUGHOUT AND MUCH ENLARGED, WITH ONE HUNDRED PLATES, COMPRISING MORE THAN SIX HUNDRED ENGRAVINGS, SOME PRINTED IN COLOURS, AND MOST OF WHICH HAVE BEEN DRAWN ON WOOD BY THE AUTHOR.

The different instruments above referred to may be obtained packed in a case, of Mr. Collins and of Mr. Swift.

Glass Slides, thin Glass, Watch-glasses, Glass Shades.

83. Plate Glass Slides, the edges of which have been properly ground and polished, may be obtained ready for use, at six shillings per gross, or they may be easily cut out with the diamond, and the edges ground on the grinding slab. The slides now in common use in this country are three inches in length and one in breadth, and I cannot too strongly recommend the observer to employ slides whether of metal, wood, or glass of this size only for microscopical purposes. The glass slides should always be made of thin plate-glass, and pieces as clear as possible should be selected.

84. Thin Glass.—An object placed for examination upon a glass slide should be always protected with a piece of thin glass before it is placed upon the stage of the microscope for examination. Thin glass now used for microscopical purposes is called cylinder glass, and I believe all or nearly all that is used is manufactured by Messrs. Chance, of Birmingham. It may be obtained of different degrees of thickness. Thin glass in sheets should be kept in fine sawdust. As it is imperfectly annealed it is very readily broken. When cut up in small pieces, it should be kept in a little box, with a little powdered starch, which prevents the pieces being broken, but great care must be taken to remove the starch from the surface, or the observer will be continually discovering starch in specimens in which he would little expect to find it. For cutting the thin glass an instrument termed a *writing diamond* is employed, and this is also used by some observers for writing the name of the preparation upon the glass slide, pl. XX, fig. 8, p. 54. As a general rule, however, I think it better to write the name of the specimen upon a small label which can be gummed to the glass.

OF CLEANING THIN GLASS.

The thin glass is easily cleaned with the aid of an old cambric handkerchief. If the glass is excessively thin it should be placed upon a pad of clean writing paper. The thin glass being firmly kept in contact with the paper by pressing firmly with the finger of one hand, it is carefully wiped with the handkerchief, a fold of which is twisted round the index finger of the other. The piece of glass is next turned round and the other side wiped in the same manner. It is then taken up in the forceps, breathed upon and placed over the specimen.

Glass Cells are described in §§ 124 to 135. Ordinary thin glass of various degrees of thickness, and already cut into squares and circles, may be obtained of Messrs. Claudet and Houghton, High Holborn. For the very high powers the thinnest pieces must be selected from a

considerable quantity. Messrs. Powell and Lealand supply the thin glass for use with their twenty-fifth. *See* Part VI.

Brass cells and tin cells are referred to in § 118.

85. Watch Glasses of various sizes should be kept by every observer, as they are convenient for many purposes. They cost about a shilling per dozen, and may be obtained of the watch-makers. The lunette glasses are useful for examining substances in fluids with low powers, as in these we are enabled to obtain a considerable extent of fluid of nearly uniform depth.

The little porcelain moulds in which moist colours are kept, and the little circular and oval shallow dishes, used by the artist's colourmen, will be found very useful for receiving microscopical specimens while soaking in various solutions prior to examination or mounting. They may be covered by circular pieces of glass.

86. Glass Shades.—Every microscopist should be provided with from six to twelve small glass shades from two to four or five inches in diameter, to protect objects from the dust which are being mounted. The cheap slightly green propagating glasses, now commonly sold at all the glass shade shops, are most convenient for this purpose. They cost from 2*d.* to 5*s.* These shades are figured in pl. XX, fig. 1.

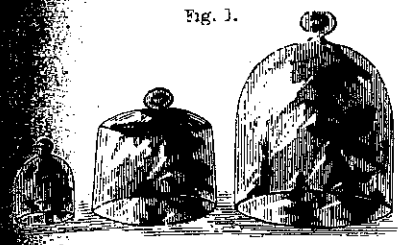
Glass slides, thin glass and watch-glasses are included in some of the cases of instruments and apparatus sold by many of the microscope makers.

VARNISHES, CEMENTS, AND MARINE GLUE.

The chief cements employed in microscopical work, are *Gold size, Sealing-wax varnish, Solution of shell-lac, Solution of asphalt, Marine glue, Canada balsam, Gum Damar in Benzol, Gum,* and a *French cement* composed of lime and India-rubber. These cements are used for attaching the glass cell to the glass slide, for fixing the cover upon the preparation after it has been properly placed in the cell, and for other purposes. The liquid cements should be kept in wide-mouthed bottles, or in capped bottles, fig. 2, pl. XX, p. 54, or in pots with tin or brass covers, pl. XXIV, fig. 1, p. 88.

87. Gold Size is prepared by melting together gum animi, boiled linseed oil, red lead, litharge, sulphate of zinc, and turpentine. Gold size adapted for microscopical purposes may be also prepared as follows :—25 parts of linseed oil are to be boiled with one part of red lead, and a third part as much umber, for three hours. The clear fluid is to be poured off and mixed with equal parts of white lead and yellow ochre, which have been previously well pounded. This is to be added in small successive portions, and well mixed; the whole is then again to be well boiled, and the clear fluid poured off for use. In this country gold size may be obtained of any varnish maker.

Fig. 1.



Glass shades for protecting objects from dust while being mounted p. 54.

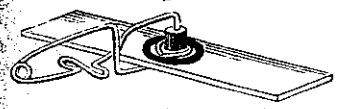
Fig. 2.

Vessel for containing Canada balsam, gum, cements, &c. p. 54.

Fig. 3.

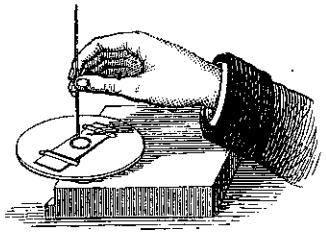
Dr Madden's spring clip p. 58.

Fig. 4.



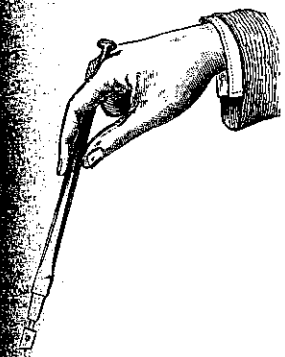
Modified spring clip p. 59.

Fig. 5.



Mr. Shadbolt's apparatus for making round cells of Brunswick black. p. 70.

Fig. 6.

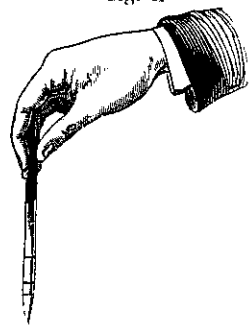


To illustrate the manner in which the diamond is used for cutting thick glass. p. 68.

Fig. 7.

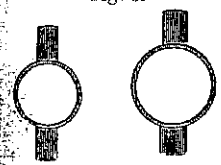
Large bradawl, for scraping away superfluous marine glue in making cells. p. 72.

Fig. 8.



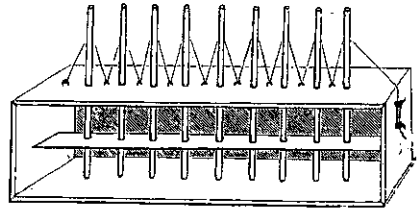
Writing diamond, for cutting thin glass. pp. 53, 70.

Fig. 9.



Metal brass rings, for cutting circles of thin glass. p. 71.

Fig. 10.



Arrangement for exerting continued pressure upon the glass covers of nine specimens while the cement is drying. p. 58.

[To face page 54.

88. Sealing-wax varnish is easily made by dissolving the best sealing-wax of any colour, in tolerably strong alcohol. This cement is, however, apt to dry rather brittle, and should not, therefore, be used in cases where it is of the greatest importance to keep the cell perfectly air-tight. It forms, however, a good varnish for the last coat. Various colours may be kept according to taste.

89. Solution of Shell-lac is a very good cement for fixing down the thin glass cover. It is made by dissolving shell-lac in spirits of wine. The shell-lac should be broken in small pieces, placed in a bottle with the spirit, and frequently shaken, until a thick solution is obtained.

Bell's Cement.—A good cement for specimens immersed in glycerine is sold by Messrs. Bell, chemists, Oxford-street. This, I believe, was originally suggested by Mr. Tomes, but I do not know its exact composition. It appears to contain shell-lac and gold size.

90. Damar Cement.—This is made by dissolving gum Damar in Benzol, and is applied with a brush. It is one of the best cements, especially when glycerine is used as the preservative fluid.

91. Brunswick Black.—Solution of asphalt in turpentine commonly known by the name of Brunswick black, may be obtained at any oil-shop, and forms a most useful cement, both for making very thin cells, (§ 116), and also for fixing on the thin glass covers. If a little solution of India-rubber in mineral naphtha be added to it, there is no danger of the cement cracking when dry. For this hint I have to thank my friend, Mr. Brooke. I have many preparations which have been cemented with Brunswick black which have kept well for upwards of twenty years. It is always desirable, however, to paint on a new layer from time to time, perhaps once in twelve months.

Common Brunswick black is made by melting one pound of asphaltum, and then adding half a pound of linseed oil, and a quart of oil of turpentine. The best Brunswick black is prepared by boiling together a quarter of a pound of foreign asphaltum, and four and a quarter ounces of linseed oil, which has been previously boiled with half an ounce of litharge until quite stringy; the mass is then mixed with half a pint of oil of turpentine, or as much as may be required to make it of a proper consistence. It is often improved by being thickened with lamp black. It must be remembered that this cement is soluble in oil of turpentine. Dr. Eulenstein, of Stuttgart, finds that equal parts of Brunswick black and gold size with a very little Canada balsam forms a good lasting cement, which does not crack or contract.

92. Marine Glue.—This substance was, I believe, first used for microscopical purposes by Dr. Goadby, of Philadelphia. It is prepared by dissolving, separately, equal parts of shell-lac and India-rubber, in coal or mineral naphtha, and afterwards mixing the solutions thoroughly with the application of heat. It may be rendered thinner

by the addition of more naphtha. Marine glue is dissolved by naphtha, ether, and solution of potash. It is preserved well in a tin box. The manner of using marine glue and the different cements I have alluded to is described in §§ 116, 122.

93. Cement for attaching Gutta Percha or India-rubber to the Glass Slides.—A cement for attaching cells of gutta percha or India-rubber to the glass slide may be made as follows:—According to Harting, gutta percha is to be cut into very small pieces and stirred, at a gentle heat, with fifteen parts of oil of turpentine; the gritty, insoluble matter, which the gutta percha always contains, is to be separated by straining through linen cloth, and then one part of shell-lac is to be added to the solution, kept at a gentle heat, and occasionally stirred. The mixture is to be kept hot until a drop, when allowed to fall upon a cool surface, becomes tolerably hard. When required for use, the mixture is to be heated, and a small quantity placed upon the slide upon which the cell is to be fixed; the slide itself is then to be heated.

94. Canada Balsam, a thick viscid oleo-resin, which becomes softer on the application of a gentle heat, is much employed by microscopical observers: formerly it was used for cementing cells together, but this is now effected more readily by the aid of marine glue. If it be exposed to too high a temperature, the volatile oil is expelled, and a hard brittle resin remains behind. It is chiefly employed for mounting hard dense textures; and, in consequence of its great power of penetrating, and its highly refracting properties, the structure of many substances, which cannot be made out by the ordinary mode of examination, is rendered manifest by this medium. Canada balsam should be preserved in a tin box, fig. 1, pl. XXIV, p. 88, care being taken to exclude the dust; or in a bottle having a cap to it. The balsam should be kept very clean, otherwise preparations mounted in it will be spoilt by the accidental introduction of foreign bodies. It has been frequently recommended that the oldest specimens of balsam should alone be employed for microscopical preparations. By exposure to the air, the balsam becomes very thick, and unfit for use: it may, however, be thinned by the addition of turpentine, ether, or chloroform. Turpentine is apt to render the balsam liable to become streaky some time after the preparation has been mounted, and bubbles are not unfrequently formed in it.

Vessels for Keeping Canada Balsam in.—The tubes, made of thick tin-foil, used for artists' colours, with a small cap that screws on to the top, as suggested by Mr. Suffolk, are very convenient receptacles for the preservation of Canada balsam. As they contain no space for air, the balsam does not become hard and unmanageable, as is too often the case when it is kept in bottles or tin pots. There is no necessity for using a glass or metal rod, as the quantity of balsam required can always be forced out without the slightest difficulty. Other cements and varnishes can be kept in the tin tubes also for any length of time. It is as well, however, to keep them in an upright position, to prevent the cement from running into the thread of the screw, and so fixing the top too tightly.

95. Solutions of Canada Balsam.—Canada balsam is soluble in ether, but the best solvent for it is chloroform. Many very delicate structures may be mounted in Canada balsam, by immersing them in a chloroform solution. Sufficient chloroform is added to make a solution that will run freely. As the chloroform evaporates the balsam becomes more viscid and gradually gets hard. Solutions of Canada balsam in chloroform are now much used for mounting different parts of insects, various tissues, geological and mineralogical specimens, and many objects of general interest. Mr. Hepworth, of Croft's Bank, was among the first to use a solution of Canada balsam in chloroform for mounting objects. Mr. W. H. Heys ("Trans. Mic. Soc.," Jan., 1865, p. 19) prepares the solution as follows. Old balsam is mixed with sufficient chloroform to make it quite fluid so that it will drop easily from the lip of the vessel containing it. The prepared balsam is then poured into long thin half-ounce phials, corked up, and set aside for at least a month. The balsam thus prepared is clearer and sets much more quickly than if mixed with the chloroform at the time it is required for use. A solution of balsam in Benzol is referred to on p. 90.

96. Arrangements for pressing down the Thin Glass Cover while the Balsam or Cement is becoming hard.—Some specimens which are more or less elastic, immersed in Canada balsam, gelatin, and other media require firm pressure to be maintained upon the thin glass until the balsam or the cement by which it is attached to the slide shall have hardened. Many specimens immersed in fluids require to be made thinner and more transparent by being subjected to moderate, but sustained pressure, while the cement in which they are embodied or that by which the thin glass cover is fixed down gradually becomes dry and hard. Other specimens require very firm pressure while the process of drying goes on. Several methods have been devised for producing pressure and for maintaining it in uniformity. A very simple plan is to place a small piece of wood, about an inch in height, upon the cover. This may be fixed in its place by passing a piece of thread over it, and tying it at the back of the slide; or the wood may be kept in its place by a vulcanized India-rubber ring. Ordinary weights may also be used, or springs arranged as in the ingenious apparatus devised by Mr. Gorham. My friend, Mr. White, has also suggested a very simple and effective apparatus for the same purpose. It consists of a bent lever, which, by acting upon a screw, can be forced down upon the thin glass with the amount of pressure required. Another form

of instrument, with a graduated spring, was designed by the Rev. G. Isbell, pl. XXIV, fig. 2, p. 88. The compressorium may also be employed for the same purpose, if a small piece of cork be inserted between the thin glass to which the pressure is to be applied, and the glass of the compressorium itself.

Mr. Hoblyn, of Bath ("Archives of Medicine," vol. III, p. 140), has devised an ingenious apparatus for the same purpose. In this instrument, a number of slides may be placed at the same time, and a graduated pressure exerted upon each of them, pl. XX, fig. 10, p. 54.

The above pieces of apparatus have however been superseded by the use of the simple spring clip devised by Dr. Maddox ("Trans. Mic. Soc.," July, 1865, p. 84). This is made by bending a piece of brass wire in the form represented in pl. XX, fig. 3. The end which is to press upon the thin glass must be filed perfectly flat, or a piece of flat cork may be fixed to it. Or, in cases where the glass cover is very thin, a smaller piece of thicker glass may be placed upon it and the spring allowed to press upon the latter. This clip has been modified by Mr. Webb, as represented in pl. XX, p. 54, fig. 4. These clips may be obtained at *1s. 6d.* and *2s.* per dozen of Mr. Baker, Holborn, and of Mr. Swift, University Street.

Another very simple and efficient spring clip was suggested by Dr. T. F. Allen, of New York. This was made of a piece of ordinary watch spring, bent in a spirit lamp in the form of a hoop, so that a small portion of one end would press gently on the cover, the other on the under surface of the glass slide. A number of such springs may be made out of any old watch spring.

97. Gum.—Thick gum-water will be found very useful for attaching labels to preparations, and also for fixing on the thin glass cover when preparations are mounted in the dry way. It is prepared by placing common gum-arabic in cold water, and keeping the bottle in a warm place until the solution has become sufficiently thick. It should always be strained before it is placed in the bottle for use.

Gum-water, thickened with powdered starch or whiting, is a very useful cement for attaching the glass cover in the case of preparations mounted dry. When dry it forms a hard white coating. The addition of a little arsenious acid will prevent the growth of mildew. Another very convenient solution is made by dissolving powdered gum in a weak solution of acetic acid.

98. French Cement composed of Lime and India-rubber.—The French cement composed of lime and India-rubber is very valuable for mounting all large microscopical preparations. The principal advantages are, that it never becomes perfectly hard, and it therefore permits considerable alteration to take place, under the influence of varying temperature, in the fluid contained in the cell without the entrance

of air. It also adheres very intimately to glass, even though it be perfectly smooth and unground.

If a glass cover is to be attached to a large cell containing fluid, we may proceed as follows:—A small piece of the cement is to be taken between the finger and thumb and carefully rolled round until it can be drawn out into a thread about the eighth or tenth of an inch in thickness. This is applied to the top of the cell, before any of the fluid is introduced. The cement is to be slightly pressed down with the finger previously moistened. It will adhere intimately. The preservative fluid with the preparation are now introduced and the cell filled with fluid which indeed is allowed to rise up slightly above its walls. The glass cover, cut rather smaller than the external dimensions of the cell, and slightly roughened at the edges, is to be gently breathed upon. One edge is then to be applied to the cement, so that it may be allowed to fall gradually upon the surface of the liquid which is now seen to wet each part of the cover successively, until it completely covers the cell, and a certain quantity of the superfluous fluid is pressed out. By the aid of any pointed instrument a very little cement is removed from one part, so that more fluid may escape as the cover is pressed down gently into the cement. The pressure must be removed very gradually, or air, of course, will enter through the hole. A bubble of air entering in this manner may often be expelled again by pressure, or it may be driven out by forcing in more fluid through a very fine syringe at another part of the cell; but it is far better to prevent the entrance of air in the first instance. The edge of the glass cover being thoroughly embedded in the cement, the small hole is to be carefully plugged up with a small piece of cement, and the cell allowed to stand perfectly still for a short time, when it may be very gently wiped with a soft cloth. The edges of the cement may be smoothed by the application of a warm iron wire, and any superabundance removed with a sharp knife. A little Brunswick black or other liquid cement may be applied to the edges, for the purpose of giving the whole a neater appearance.

The cement is made as follows:—A certain quantity of India-rubber scraps is carefully melted over a slow fire, in a covered iron pot. The mass must not be permitted to catch light. When it is quite fluid, lime in a perfectly fine powder, having been slaked by exposure to the air, is to be added by small quantities at a time, the mixture being well stirred. When moderately thick, it is removed from the fire and well beaten in a mortar and moulded in the hands until of the consistence of putty. It may be coloured by the addition of vermilion or other colouring matter. I have several preparations which have been placed in the creosote and naphtha solution in large cells, and they are now perfectly air-tight, although upwards of twenty years have elapsed since they were first put up. The lime and India-rubber cement answers well

for fixing on the glass tops of large preparation jars, and looks very neat; but, if moderately strong spirit be used, a little air must be permitted to remain in the jar.

As cements are required in different investigations for making apparatus of various kinds, and for other purposes, I venture to republish the following receipts, which have been taken from "The Journal of Applied Chemistry," though few may be required by microscopists.

98*. Other Cements.—A good rubber cement may be prepared by dissolving one part India-rubber in two parts linseed oil, and adding to the solution a sufficient quantity of bole, say, about three parts.

For amber and tortoiseshell, a cement was made by mixing together equal parts of mastic and linseed oil, and warming gently. This cement should be used warm.

To unite wood to wood, a thick solution of shell-lac in alcohol may be used. It is well to put a piece of fine gauze or crape between the broken surfaces of wood, and then press them tightly together until the cement becomes perfectly firm. Another good, durable cement for woodwork is made by fusing together shell-lac, mastic, and common turpentine, and adding some broken isinglass.

For attaching small objects to anything turned, a mixture of colophonium, turpentine, and yellow wax, with the addition of a little pulverized sealing-wax, answers nicely. The cement sets quickly and holds well.

To fasten knives and forks in silver handles, a mixture of two parts of melted black pitch and one part of fine brick-dust may be used. It must be used warm.

A varnish or cement to protect wood from the action of mineral acids, alkalies and corrosive gases, like chlorine, is made from six parts of colophonium and three parts of wood tar by heating together in an iron kettle on a furnace in the open air, and then stirring in four parts of fine brick-dust. The varnish is applied with a brush while warm.

An excellent cement for glass is made by dissolving one part India-rubber in sixty parts of chloroform, then adding thirty-four of mastic, and letting it digest for a week at a gentle heat. This cement is also applied with a brush, and is especially distinguished by its transparency.

Another cement for glass and porcelain is made by digesting small pieces of isinglass in sixteen times their weight of water for twenty-four hours. The solution is evaporated to one-half, strained, and, while still hot, eight parts of alcohol added, and at the same time a solution of one part mastic in six parts warm alcohol. One half-part of finely-powdered gum ammoniac is triturated in the warm solution until the whole mass is homogeneous. When used, both the cement and the

object to be mended are warmed. This cement is highly recommended for its adhesive qualities.

Glue and Gum Cements.—These are very tenacious and well adapted for mending ornaments. They resist the action of water and the atmosphere. There are various kinds of these cements for bone, ivory, whalebone, mother-of-pearl and precious stones.

One of them is made by dissolving two parts isinglass and four parts colourless glue in sixty parts water, evaporated to half its volume, then adding $\frac{1}{15}$th part mastic dissolved in one part alcohol, and stirring in two parts zinc white. The surfaces are warmed when the cement is applied to them. This cement holds well, dries easily, and may be kept a long time in tightly-corked bottles.

For bone, ivory, whalebone, mother-of-pearl, &c., a cement with a beautiful gloss may be prepared as follows:—Soak common cabinet-maker's glue in hot water, warm the jelly formed, add enough pulverulent slacked lime to give it consistency. Warm the object to be cemented, clean the surfaces carefully, apply the cement and tie the parts firmly together. In a few days it gets very hard. Even common glue, with pulverized chalk stirred in, makes an excellent cement for wood and metals.

For fastening leather to metal, the metal should be coated with a hot solution of glue, and the leather with a hot extract of nut galls. Allow them to dry quietly, and they adhere well.

For porcelain, the well-known white-of-egg cement is best. To prepare this it is only necessary to stir the white of eggs into quite a stiff solution of glue, and then apply to the fracture.

A cement of gum for porcelain is made by pulverizing four parts of oyster shells and mixing intimately with two parts pulverized gum arabic. The powder is kept in a well-stoppered bottle, and when needed for use is rubbed up with white of egg, or warm water, to a thick dough, applied to the object and dried by a gentle heat. Another cement for glass and porcelain is made from eight parts well-burnt pulverized alabaster gypsum and two parts fine gum arabic, mixed with water to a thick paste, and forty to fifty drops of oil of turpentine added to an ounce of the cement.

Cements containing Casein.—For glass, porcelain, stone, and wood, the very best cement is made of a suitable quantity of old cheese, rubbed fine and mixed with water to a thick magma, and a fourth part of pulverized lime added.

A still stronger cement for the same purpose is made by slaking one pound of quicklime in water, and mixing with three-quarters of a pound of finely powdered lime or sandstone and one pound pulverized cheese. Before using, it is well to moisten the fracture or edges with warm water.

A so-called casein water-glass is made as follows:—The casein

of skimmed milk is separated from it by the addition of acetic acid, filtered, and the acid washed out with water. The pure casein thus obtained is mixed with six times its volume of concentrated solution of casein in water. This cement is thoroughly commendable, and well repays the trouble taken to make it.

An excellent cement for artificial meerschaum, and one that may be used to give consistency to silk goods or to coat artificial flowers and court plaster, to give more adhesiveness and firmness, is made by rubbing two to four parts of the above casein with cold borax solution till a thick liquid is obtained that becomes clear on standing. This also renders goods waterproof.

Water-glass Cements.—For glass, earthenware, porcelain, and all kinds of stoneware, these cements are excellent. A cement for glass and marble is prepared by rubbing together one part of fine pulverized glass and two parts of pulverized fluorspar, and then adding enough water-glass solution to give it the consistency necessary in a cement.

Water-glass mixed with hydraulic cement to a thick dough makes a good cement for the edges and joints of stone and marble slabs. It is well to mix but little at a time, as it hardens very quickly.

Lime, Gypsum, Clay, and Cement, mixed with Water, Oil or Blood.—For cementing stone and for filling crevices in buildings, before they are painted, the masons use a cement made of fresh blood, slaked lime, brick-dust, broken up coal ashes, hammerslag, and sand, in all proportions. This excellent cement hardens quickly, and offers great resistance to the action of the weather.

A lime cement for connecting water pipes, bathing tubs, &c., a mixture of two-thirds fine brick-dust, two-thirds unslaked lime, and two-thirds hammerslag, is made and stirred up with lye or hot oil to a stiff dough.

Another cement, intended to render Hessian clay retorts impenetrable, is obtained by rubbing freshly slaked lime into a concentrated solution of borax. The solution is applied with a stiff brush and allowed to dry, after which it is heated until the glazing begins to fuse.

Clay mixed with water and fresh warm blood, containing some unslaked lime, is used in Germany to close joints in stoves. The cement is applied while the stove is hot. Wood ashes, fire clay, and salt, mixed with water, is used for the same purpose. Fat and burnt clay, in equal proportions, moulded with water into a dough, is also used.

Plaster of Paris, mixed with water and a cold solution of alum, is an excellent cement for stoneware. It sets slowly, but becomes hard as stone.

Oil Cements.—An excellent oil cement for porcelain and for luting of retorts, flasks, and porcelain evaporating dishes, is obtained when ordinary brick-dust is powdered, sifted, and mixed with an equal quantity of red lead, and then rubbed, under great pressure, with old

boiled linseed oil to a thick paste, which is mixed with coarse sand to the stiffness of cement. When a dish is to be covered with it, paste is applied before the sand is put in, and the sand then strewn upon it. The dish is afterwards exposed to a steady heat for a long time.

For large vessels take six parts litharge, four parts fresh-burnt pulverized lime, and two parts white bole, and mix with cold linseed oil.

To fasten metallic letters to a smooth surface a cement is made as follows:—Thirty parts copal varnish, ten parts linseed oil varnish, six parts crude oil of turpentine, ten parts glue dissolved in a little warm water, and twenty parts pulverulent slaked lime. It is very pliant and soon hardens.

To unite copper and sandstone take three and a half parts white lead, three parts litharge, three parts bole, and two parts broken glass, and rub up with two parts linseed oil varnish.

As a polish for rough stones, basins, &c., a paint is made of nine parts of finely sifted and burnt brick-clay and one part litharge, mixed with a sufficient quantity of linseed oil.

For connecting cast-iron water pipes, twelve parts Roman cement, four parts white lead, one part litharge, and a half part colophonium are pulverized and mixed; from two and a half to three pounds of the mixture is triturated with old linseed oil, in which is boiled two ounces of colophonium.

Another, for the same purpose, is made of equal parts of burnt lime, Roman cement, potters' clay, and clay, separately well dried, finely ground, sifted, well mixed and triturated with linseed oil. Common lead lute, for stopping openings in apparatus, is best made from litharge and red lead mixed with old boiled oil. In all cases the surfaces must be clean. These cements stand well under water.

As lead lutings are somewhat expensive, the following is recommended:—Take two parts red lead, five parts white lead, and five parts of the finest clay, and mix with boiled linseed oil.

A good oil cement for wood, especially for antique carvings, is made of one part pulverized slaked lime, and two of rye flour, mixed with linseed oil varnish. It takes any desired colour and polish.

To make water holders tight, we may use pulverized slaked lime and cod-liver oil.

A cement to make chemical apparatus tight can be prepared from oil cake or pressed almond cake rubbed with water.

Miscellaneous Cements, &c.—Furniture polish:—Moisten 120 parts beeswax with oil of turpentine, and add 7·5 parts finely pulverized resin, and enough aniline red to give the desired mahogany colour.

Oil cement:—100 parts red lead, 250 parts white lead, 200 parts pipe clay, mixed with boiled oil.

Water cement:—100 parts slaked lime, 190 parts brick-dust, 160

parts sand, 50 parts blacksmiths' dross, 50 parts powdered lime; mix with water.

Another:—600 parts iron filings, 100 parts ignited sand, 100 parts powdered slacked lime; mix with water.

Iron and blood cement:—100 parts pulverized lime, triturated with bullock's blood, 290 parts cement, and from five to ten parts iron filings.

PRESERVATIVE FLUIDS.

In all cases it must be borne in mind that an object to be mounted in a preservative fluid should be soaked in a considerable quantity of it for at least a day before it is mounted permanently, and if the specimen is large, it should be soaked for many days previous to being finally placed in the cell.

99. Spirit and Water.—Spirit and water constitute a well-known and valuable medium for preserving anatomical preparations. In diluting spirit, distilled water only should be employed; for, if common water be mixed with spirit, a precipitation of some of the salts dissolved in it not unfrequently takes place, which renders the mixture turbid and unfit for use. The mixture of water and spirit should be made several days before it is required, or a number of air bubbles will adhere to the specimen. Proof spirit will be strong enough for all general purposes, except for hardening portions of the brain or nervous system, when stronger spirit must be used. Two parts of rectified spirit, about sp. gr. ·837, mixed with one part pure water, make a mixture of sp. gr. ·913–·920, which contains about 49 per cent. of real alcohol, and will, therefore, be about the strength of proof spirit. One part of alcohol, sixty over proof, to five parts of water, forms a mixture of sufficient strength for the preservation of many substances, and not a few microscopical specimens may be preserved in a solution more diluted than this.

For many years past, the Government has permitted the use of methylated alcohol for various purposes in the arts. This pays no duty, and answers well for preserving anatomical preparations, and is a great boon to all engaged in putting up large anatomical specimens. It may be obtained at the price of 5*s.* 6*d.* a gallon, sixty degrees over proof, of varnish makers and most of the chemists.

100. Glycerine.—This is one of the most valuable fluids ever employed for microscopical purposes. I believe Mr. Warrington, of Apothecaries' Hall, was the first observer who used glycerine as a preservative medium for microscopical preparations.

A solution of glycerine adapted for preserving many structures is prepared by mixing equal parts of glycerine and camphor water. The latter prevents the development of mildew. Glycerine may also be mixed with naphtha and water, or with the creosote solution described in § 101. The degree of dilution of the glycerine will depend upon the

nature of specimen. If the substance be at all opaque it will be necessary to employ strong glycerine. I have many preparations which have been preserved in glycerine for nearly thirty years. Of the importance of strong glycerine as a preservative medium, I shall have to speak more fully in part VI. Glycerine may be mixed with various chemical tests and preservative substances, for special enquiries. Analyses may be conducted by the test compounds being dissolved in the menstruum instead of in water. For preserving medusæ and other delicate marine animals Dr. Carpenter recommends a solution composed of *sea water* with one-tenth of *alcohol*, and the same quantity of glycerine. Dr. Maddox tells me that, for some years past, he has been in the habit of using equal parts of sweet spirits of nitre (Sp. Eth. Nit. of the Pharmacopœia) and glycerine, especially in preparing delicate tissues of insects. He finds that many objects are rendered very transparent if soaked in this mixture before they are preserved in glycerine.

The best glycerine is distilled by a patent process, and is perfectly colourless, free from all impurities, and of great density. The specific gravity of Price's patent glycerine is 1,240, while the common is only 1,216. The strongest glycerine obtainable is crystallized, but it is very expensive. The purest glycerine costs 3*s.* or 4*s.* a pound, but good glycerine may now be obtained for 1*s.* 6*d.* per pound.

For more than twenty years I have used glycerine for preserving almost every structure. I shall give the results of my most recent experience concerning this substance, from the use of which I have learned very much, in part VI.

101. Thwaites' Fluid.—This fluid has been much employed by Mr. Thwaites for preserving recent specimens of desmidiæ, but it is also applicable to the preservation of a vast number of other vegetable and animal organisms.

It is made as follows:—

Water	16 ounces.
Spirits of wine	1 ounce.
Creosote, sufficient to saturate the spirit.	
Chalk, as much as may be necessary.	

Mix the creosote and spirit, stir in the chalk with the aid of a pestle and mortar, and let the water be gradually added. Next add an equal proportion of water saturated with camphor. Allow the mixture to stand for a few days, and filter. In attempting to preserve large preparations in this fluid, I found that it always became turbid, and therefore was led to try several modifications of it. The solution next to be described was found to answer very well.

Water may also be impregnated with creosote by distillation. It should be remarked that M. Strausdurkheim has succeeded in preserving animal preparations in camphor water only.

102. Solution of Naphtha and Creosote:—Creosote, 3 drachms; wood naphtha, 6 ounces; distilled water, 64 ounces; chalk, as much as may be necessary. Mix first the naphtha and creosote, then add as much prepared chalk as may be sufficient to form a thick smooth paste; afterwards add, very gradually, a small quantity of the water, which must be well mixed with the other ingredients in a mortar. Add two or three small lumps of camphor, and allow the mixture to stand in a lightly covered vessel for a fortnight or three weeks, with occasional stirring. The almost clear supernatant fluid may then be poured off and filtered if necessary. It should be kept in well-corked or stoppered bottles.

I had some large preparations which had been preserved in upwards of a pint of this fluid, for nearly twenty years, and the solution remained perfectly clear and colourless. Some dissections of the nervous systems of insects have kept excellently; the nerves retain their white appearance, and have not become brittle. Two or three morbid specimens are also in an excellent state of preservation, the colour being to a great extent preserved, and the soft character of the texture remaining. I had one preparation mounted in a large gutta-percha cell, containing nearly a gallon of this fluid.

A solution of wood naphtha or pyroacetic spirit in water, has been recommended by Professor Quekett, and forms an excellent preservative solution, in the proportion of one part of the naphtha to ten of water. The solution is often a little cloudy, but may be made quite clear by filtration after the mixture has been allowed to stand still for some days.

One great advantage of these aqueous preservative solutions is that the natural appearance of the structure is very slightly altered. The solution, however, after a time renders many of the more delicate structures more or less granular.

103. Carbolic Acid.—A solution of carbolic acid in distilled water preserves many animal and vegetable preparations exceedingly well. The water will only take up a small quantity of ordinary carbolic acid, but the preservative qualities of the weakest solution are very great. One part of carbolic acid to a hundred of water is sufficient.

Perfectly pure carbolic acid is now made, in very large quantity, by Messrs. Bowdler and Bickerdike, of Church, Lancashire, and is sold under the name of *Absolute Phenol*, for 6*s.* or 7*s.* a pound. It may be obtained in large or small quantities, of Mr. Marchant, Berners Street, Oxford Street. This preparation is much more soluble in water than the liquid carbolic acid. Besides preventing decomposition of animal and vegetable tissues, the phenol effects a curious change in the properties of ordinary water. A mere trace (less than one thousandth) causes the water to froth, and to retain air-bubbles in suspension for a [illegible] than they are retained by ordinary water. Such very

dilute solution wets dry surfaces and runs into minute crevices more thoroughly than common water, and, at the same time, runs off from surfaces more completely, leaving a very thin but even layer of moisture upon the surface. Glass may, in this way, be perfectly and uniformly wetted with water. Drops of carbolic acid water are smaller and less easily formed than in the case of the same water without carbolic acid. For these and other reasons, minute traces of carbolic acid improve many of the fluids used for the preservation of microscopical specimens.

104. Solution of Chromic Acid.—A solution of chromic acid is well adapted for preserving many microscopical specimens. It is particularly useful for hardening portions of the nervous system previous to cutting thin sections. The solution is prepared by dissolving sufficient of the crystallized acid in distilled water or in glycerine, to render the liquid of a pale straw colour.

The crystallized acid may be prepared by decomposing 100 measures of a saturated solution of bichromate of potash, by the addition of 120 to 150 measures of pure concentrated sulphuric acid. As the mixture becomes cool, crystals of chromic acid are deposited, which should be dried and well pressed on a porous tile, by which means the greater part of the sulphuric acid will be removed, and the crystals obtained nearly pure.

105. Preservative Gelatine.—This substance was first employed for preserving microscopical textures by Mr. H. Deane, who gives the following receipt, and directions for its preparation:—Gelatine, 1 ounce; honey, 4 ounces; spirits of wine, ½ ounce; creosote, 6 drops.

Soak the gelatine in water until soft, and to it add the honey which has been previously raised to the boiling-point in another vessel. Next, let the mixture be boiled, and after it has cooled somewhat, the creosote dissolved in the spirits of wine is to be added. Lastly, the mixture is to be filtered through thick flannel to clarify it.

When required for use, the bottle containing the medium must be slightly warmed, and a drop placed on the preparation upon the glass slide, which should also be warmed a little. Next, the glass cover, after having been breathed upon, is to be laid on with the usual precautions. See p. 82.

106. Gelatine and Glycerine.—A mixture of gelatine and glycerine makes a very valuable medium for preserving different animal and vegetable structures, and supersedes the last preparation. It may be prepared as follows:—A certain quantity of gelatine or isinglass is allowed to soak for some time in cold water, until it swells up and becomes soft. It is then placed in a glass vessel and melted by the heat of warm water. It may be clarified if necessary, by first adding to the cool gelatine a little white of egg, then boiling the mixture, and filtering

enable us to form a general idea of the arrangement and number of the capillaries in different textures, and they may be preserved for many years without the slightest change in character. In these respects their merits must be admitted; but if we desire to learn facts concerning the relation of the capillaries to the texture lying in their meshes, the structure of the vessels themselves, or that of the tissues in which they ramify, we must study injections which have not been mounted in balsam, damar, or such media. Collections of balsam specimens are advantageous for trade purposes, but although the student may with advantage purchase a few specimens, the sooner he learns to make preparations for himself the sooner will he gain a knowledge of the structure of the tissues of animals.

195. Of the best Mode of Destroying the Life of Animals intended for Injection.—I have tried various plans of destroying animals intended for minute injection, and have found that in death by sudden shock the vessels remain in a relaxed state for a sufficient time after death to enable us to complete the injection. In some cases a good result is gained by destroying life in an atmosphere of carbonic acid, but I find that the very sudden death produced by a fall from a height, dashing on the ground, &c., is the most advantageous. Any small animal may be wrapped up in a cloth and thrown suddenly, and with some violence, upon the ground. In order to avoid rupturing any of the tissues, the animal must be well protected by several folds of the cloth. Swinging very rapidly through the air also destroys life very suddenly, without causing that sudden contraction of the muscles, which seriously interferes with the preparation of successful injections.

Good injections may be made *after* the *rigor mortis* has entirely passed off, and formerly no injections were attempted before this change had occurred. I have, however, found that by the time the muscles have again become relaxed the finer branches of the nerves have begun to soften or are entirely destroyed, and many delicate structures have become so much altered that it would not be possible for any one who was acquainted with their natural appearance to recognize them. Hence it is undesirable to put off the operation of injection if we desire to demonstrate in the specimens we are about to prepare any facts besides the mere arrangement of the capillary vessels.

ON STAINING THE BIOPLASM AND FORMED MATERIAL OF TISSUES.

The plan of staining tissues artificially is one from which great advantage has been already derived, and it is probable that, by modifications of the processes already adopted, and by the discovery of new ones, many new and most important facts will be added to our knowledge. I have pointed out that the process of staining may be employed for

two very different purposes, and it is important that the student should have a clear notion of the objects to be gained by the process before he proceeds to carry it into practice. Staining may be employed:—

1. For colouring the invariably perfectly colourless, and often invisible *bioplasm* or *living matter* of any cell or tissue, at any age, in the case of vegetable or animal textures.
2. For demonstrating peculiarities in the build of the *formed material, cell-wall, intercellular substance*, or *tissue*, and for ascertaining the order in which the several parts of which it is composed have been laid down.

Of Colouring the Bioplasm.

196. Of Colouring the Bioplasm or Living Matter.—This living matter is in all cases in the natural state perfectly clear and transparent. It never exhibits structure, and is invariably colourless. It possesses an acid reaction, or, to speak more correctly, an acid reaction is always developed immediately after its death. Hence, if a coloured alkaline solution from which the colouring matter may be precipitated or fixed by an acid, be caused to pass into bioplasm which has recently died but has not yet undergone decomposition, the alkali is neutralised by the acid present, and the colour is retained. It is probably precipitated in a state of very minute subdivision, or combined with some of the constituents of the bioplasm to form a compound insoluble in weak acids.

The *tissue itself or formed material* being ordinarily bathed with an alkaline fluid does not take the colour, and hence by carrying out the process with due care the *bioplasm or living matter may always be coloured while the formed material or tissue remains perfectly colourless.* Any one can satisfy himself of this fact by placing upon a glass slide a few liver cells from any animal immediately after its death. If a drop or two of the solution of carmine in ammonia be allowed to flow over the cells, the nucleus or mass of bioplasm of each cell will be tinted in the course of a few seconds, while the outer part of the cell will not be affected.

Staining the bioplasm may be carried out long after the death of the animal if the development of an alkali by decomposition be prevented by alcohol or some other preservative fluid. Specimens intended for subsequent staining should be immersed in a preservative fluid *immediately after death.* In practice, however, it is always better to carry out the staining process at once.

From the above remarks, it must not, however, be inferred that living matter can be stained by alkaline colouring fluids only. Solutions of an *acid* reaction may be employed if the bioplasm be rendered *alkaline* in the first instance by soaking the texture in a weak solution of ammonia. I have prepared some beautiful specimens as follows:—An

through fine flannel. To this fluid, an equal quantity of strong glycerine is added and the two are well mixed together. This mixture may be kept for any length of time, and a very slight heat is sufficient to render it perfectly fluid. This, as well as many other mixtures can be made most perfectly upon a large scale, and I therefore recommend the observer to purchase what he requires, instead of making it. The gelatine and glycerine, prepared by Mr. Rimmington, operative chemist, of Bradford, is the best medium of the kind I have used. It may be obtained in small bottles free by post for *1s. 4d.*

107. Gum and Glycerine.—Mr. Farrants many years ago suggested the following valuable preservative medium which will be found useful for mounting many objects:—Picked gum-arabic, 4 ounces by weight; distilled water, 4 ounces; glycerine, 2 ounces. The medium is to be kept in a stoppered bottle and a piece of camphor or a few drops of phenol may be added to the solution with advantage.

108. Goadby's Solution.—This is made of several different strengths. That most generally useful is the following:—Bay salt, 4 ounces; alum, 2 ounces; corrosive sublimate, 4 grains; boiling water, 4 pints. Mix and filter. This solution for most purposes may be diluted with an equal bulk of water. For preserving delicate preparations it should be even still more dilute. Goadby's solution used to be much employed for preserving anatomical specimens, but as it tends to render tissues hard and opaque, it is not adapted for the preservation of structures which are to be examined in the microscope, and has, therefore, fallen out of use as a preservative fluid for microscopical specimens.

109. Burnet's Solution consists of chloride of zinc, is a powerful antiseptic, but not adapted for the preservation of microscopical specimens.

110. Chloride of Calcium.—A saturated aqueous solution of chloride of calcium, free from iron, has been much recommended for preserving specimens of bone, hair, teeth, and other hard structures, as well as many vegetable tissues. A solution of chloride of calcium was recommended by the late Professor Schröder Van der Kolk, of Utrecht, for keeping sections of the spinal cord and preparations of nerves. Many of these, through the kindness of my friend, I had an opportunity of seeing and can testify to their excellence.

111. Alum and other Salts.—A solution of *alum* in the proportion of one part of alum to sixteen of water has been found to answer pretty well for some substances. Gannal's solution, which consists of one part of *acetate of alumina* dissolved in ten parts of water; solution of acetate of potash; solutions of *common salt* (one part to five of water, with a little camphor), *corrosive sublimate, persulphate of iron, sulphate of zinc*, and solutions of several other salts, have been recommended as preservative fluids, but although adapted for the preservation of animal substances, they cannot be employed for microscopical specimens, in consequence of their tendency to render the textures very opaque and granular. Mr. A. E. Verrill ("Silliman's Journal," March, 1865) recommends a solution made with nitre, rock salt, and arseniate of potash. My own experience, however, has led me to discard all solutions containing salts for microscopical purposes.

112. Arsenious Acid has been recommended, and Dr. Andrew Clarke used to preserve specimens of lung and other structures in an aqueous solution of this substance.

113. Arseniuretted hydrogen gas has also been recommended for the preservation of animal substances, but it is not adapted for microscopical preparations. Dr. Richardson kept animal matters from decomposition by immersing them in an atmosphere of *nitrogen*, which was prepared by placing a piece of phosphorus in a stone jar containing common air, and provided with an air-tight cover. By this means the oxygen is soon exhausted, and no decomposition can take place (?).

Some of the preservative solutions which I have referred to may be obtained of Mr. Swift, of University Street. The mode of using them will be described further on. Every microscopist engaged in any special enquiry will of course alter the composition of these solutions in any way experiment may show to be advisable. Great improvements doubtless may be made in many preservative solutions. A series of exact observations of the effects of the different fluids upon the same textures is much to be desired, and this is one of the questions upon which amateurs might contribute very valuable information.

CELLS FOR PRESERVING MICROSCOPICAL SPECIMENS.

All objects intended for microscopical observation should be protected by a cover of thin glass. This cover prevents the entrance of dust, and protects the object from the effects of exposure to the atmosphere. The fluid in which many objects are placed for examination would rise in vapour which would condense upon the object-glass, and give rise to great inconvenience were it not prevented from evaporating by a thin glass cover. If the thin glass, however, should press upon the object placed upon the glass slide, the distinctness of the specimens would in many cases be impaired, or the structure might be entirely destroyed—an inconvenience which may be prevented by placing some insoluble substance slightly thicker than the object, between the glasses. A little cavity may be made in many ways in which a specimen, dry or with its preservative fluid, may be placed, and afterwards covered with thin glass without risk of injury from pressure. This is termed a cell. Cells may be composed of various materials according to the thickness which may be necessary and according to the nature of the substance to be placed within them.

alkaline solution was injected into the vessels, and after allowing twelve hours or more for the tissues to become thoroughly permeated, the finest Prussian blue (*see* part VI) was introduced. The latter passed into the very substance of the bioplasm, which was tinged much more deeply than the surrounding material. The liver cell may be thus impregnated with the blue in every part. It seems probable that by prosecuting more detailed enquiries in this direction, we may learn something concerning the physical arrangement of the matter constituting the formed material. The bioplasm may also be tinted with a fluid of neutral reaction, because, as I have shown, there are invariably currents tending *towards* the bioplasm as long as this matter remains in a living state; but the advantage of the alkaline solution of carmine is, that the alkali is neutralised when the solution passes into the bioplasm, and the carmine is fixed there. In endeavouring to draw correct inferences regarding the natural arrangement of the parts prepared in this way, it must not, however, be forgotten that the alkaline ammonia may have effected alterations in the formed material, and modified its structure in an important manner.

Specimens prepared in the manner suggested above enable us to prove the unsoundness of the old views concerning the supposed cell-wall and cell contents, and the incorrectness of more modern assertions concerning the slight importance of the "nucleus."

197. Process of Staining followed by the Rev. Lord S. G. Osborne.—Welcker was, I believe, one of the first observers to employ a solution of carmine for the purpose of staining the nuclei of tissues, and Gerlach was an early and most successful advocate of this plan. It has been, but I think wrongly, stated, that Gerlach was the first who adopted the process. The date of Gerlach's work was 1858 ("Mikroskopische Studien aus dem Gebiete der Menschlichen Morphologie." Erlangen). But it was in June, 1856, that the Rev. Lord S. G. Osborne showed that nuclei were more deeply tinged by carmine than other parts of the cell. ("Vegetable Cell Structure and its Formation, as seen in the early stages of the Growth of the Wheat Plant.") *See* also the plate accompanying that paper ("Trans. Mic. Soc.," vol. V, pl. IV, 1856). Lord Osborne allowed the plants to *grow* in the carmine solution. The growing parts were stained most successfully. The method was not, however, applied to investigations on animal tissues.

198. Gerlach's Method of Staining.—Gerlach resorted to the carmine staining process for investigating the structure of animal tissues. He first used a concentrated solution of carmine in ammonia, and placed the sections of brain and spinal cord previously hardened by chromic acid, in the carmine fluid for from ten to fifteen minutes. They were then well washed in water for some hours, and treated with acetic acid. The water and acid were removed by immersion in alcohol. The

sections were afterwards mounted in Canada balsam. Gerlach found that dilute solutions (two or three drops of the ammoniacal solution of carmine to an ounce of water), and maceration for *two or three days*, afforded better results.

199. The Author's Carmine Fluid, for staining all forms of bioplasm of living things, is made as follows:—

Carmine, 10 grains.
Strong liquor ammoniæ, ½ drachm.
Strong glycerine, 2 ounces.
Distilled water, 2 ounces.
Alcohol, ½ ounce.

The carmine in small fragments is to be placed in a test tube, and the ammonia added to it. By agitation, and with the aid of the heat of a spirit-lamp, the carmine is soon dissolved. The ammoniacal solution is to be boiled for a few seconds and then allowed to cool. After the lapse of an hour, much of the excess of ammonia will have escaped. The glycerine and water may then be added, and the whole passed through a filter or allowed to stand for some time, and the perfectly clear supernatant fluid poured off and kept for use. This solution will keep for months, but sometimes a little carmine is deposited, owing to the escape of ammonia, in which case one or two drops of liquor ammonia may be added to the four ounces of carmine solution.

The rapidity with which the colouring of a tissue immersed in this fluid takes place, depends partly upon the character of the tissue and partly upon the excess of ammonia present in the solution. If the solution be very alkaline the colouring will be too intense, and much of the soft *tissue* or imperfectly developed formed material around the bioplasm will be destroyed by the action of the alkali. If, on the other hand, the reaction of the solution be neutral, the uniform staining of tissue and bioplasm may result, and the appearances from which so much may be learnt are not always produced. When the vessels are injected with the Prussian blue fluid before the staining process is adopted, the carmine fluid should be sufficiently alkaline to neutralise the free acid present. The permeating power of the solution is easily increased by the addition of a little more water and alcohol. In some cases the fluid must be diluted with water, alcohol, or glycerine, and the observer must not hastily condemn the process, or conclude, as some have done, that a particular form of bioplasm is not to be coloured. Those who speak thus have not given the plan a fair trial, and have not tried the effects of a solution containing a little alcohol or otherwise modified.

Notwithstanding the advantages of the above plan and its success in the hands of many observers, objections have been urged against it by some who, I venture to think, have not made themselves familiar with the practical details of the method. It has been said that the formed

material may be stained as well as the bioplasm. As every one knows, almost any thing may be stained. Hair, horn, wool, paper, &c., may be deeply dyed, even after they have been thoroughly dried. The important fact, however, is not that the tissue may be stained, but that the bioplasm of a tissue *may be deeply coloured, although the formed material which must be traversed by the staining fluid in the first instance is not stained at all.* This is the case with all bioplasm, and it seems to me a fact of far higher significance than is generally admitted. By the process of investigation described it becomes possible not only to distinguish bioplasm in all cases, but to show definitely the mode of formation of the tissue. And in many instances, by this method of proceeding, we can accurately determine which is the *oldest* and which the *youngest* portion of the tissue. Drawings of various tissues, in which the bioplasm has been stained by the above process, are given in plate XXX, p. 110, and in plate XXXI. (*See* also the plates in part VI.)

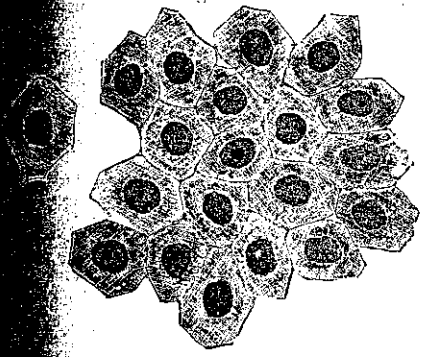
In a paper on the ova of the stickleback ("Microscopical Journal," January, 1867), Dr. Ransom has expressed himself against the plan of investigation I have followed. His objections, however, are not valid, and some of the remarks he has made prove, I think, that he has not succeeded in preparing specimens according to my method. I have replied to some of my friend's statements in a subsequent number of the journal.

Some direct that, when a tissue is too deeply stained, it should be washed in water or in spirit,—directions which clearly indicate that the authority has little practical acquaintance with the method, and is not acquainted with the principles on which the process rests. The suggestion would not be made by any one who was aware of the change induced by the colouring process, when properly conducted. Many of the remarks in connection with this matter could only have been made by persons who had never seen a preparation properly coloured, and who are not aware of the great value of the operation. Again, it has often been recommended that tissues should be stained with the carmine fluid after they have been hardened in chromic acid fluid, alcohol, and other media. Staining thus effected conveys little information, and may mislead the observer. But the most serious opposition to the adoption of the method of procedure I have recommended, and to the acceptance of the conclusions I have arrived at, is on the part of some who have quite made up their minds that all the phenomena of living organisms are due to *machinery* which is not to be demonstrated by this or any other process of investigation. For further details concerning these matters, *see* part VI.

Of Staining the Formed Material.

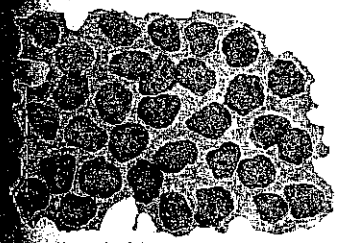
The coloured fluids referred to in the succeeding sections are em-

Fig. 1.



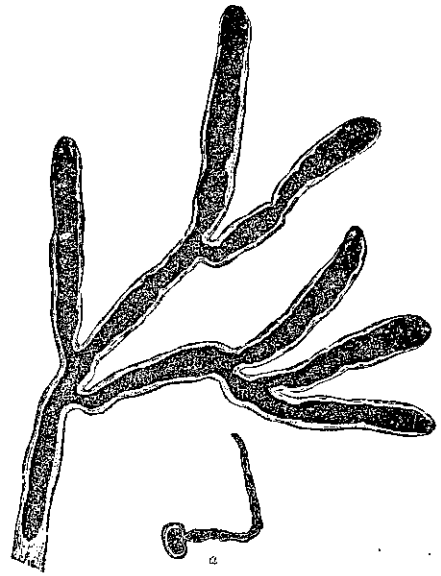
...the newt, superficial layer, each mass of bioplasm ...ed with a thick layer of formed material, constituting a fully formed cell. × 216.

Fig. 2.



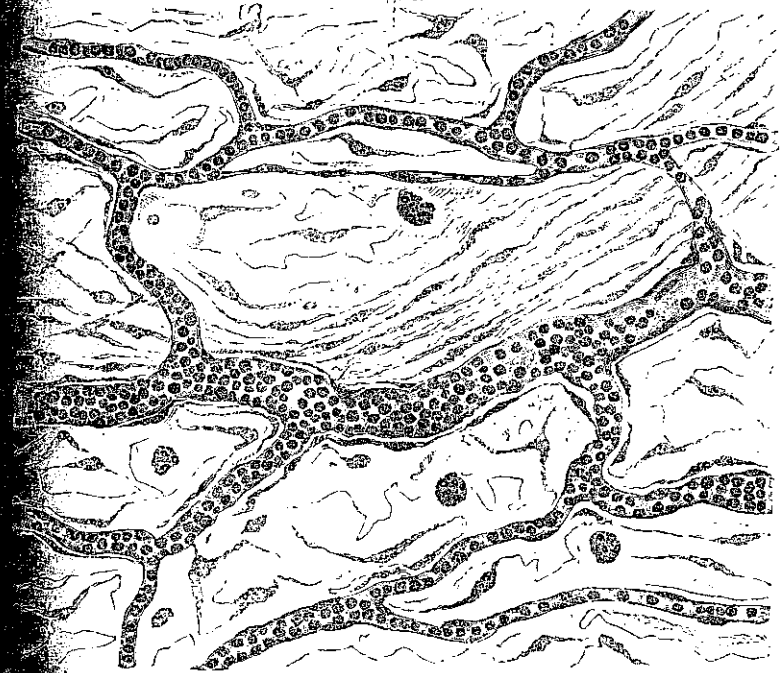
...of the cuticle of the newt, consisting almost entirely ...plasm with very little formed material. × 216.

Fig. 3.



The extremities of a rapidly growing branch of fungus from jam. The cellular wall at the end of each ramification is only just formed, and is so thin that it is hardly demonstrable. The bioplasm is abundant, and in this situation growth is proceeding very rapidly. *a*, a spore which has just commenced to sprout. × 400. 1869.

Fig. 4.



...capillary vessels. Ovum of turtle at an early period of development. All the vessels are filled with colourless ...which are stained with carmine in the preparation from which the drawing was taken. Developing connective tissue corpuscles and fat cells are also seen in the drawing.

...th of an inch . . × 216.

[To face page 126

ployed for tinting the *tissue or formed material*, while the carmine fluid I have recommended, in § 199, is for staining the *bioplasm* only.

200. **Thiersch's Carmine Fluid.**—Frey ("Das Mikroskop") gives Thiersch's fluids for colouring tissues by carmine. Carmine, 1 part. Caustic ammonia, 1 part. Distilled water, 3 parts. This solution is to be filtered. The following solution is to be prepared in a separate vessel:—Oxalic acid, 1 part. Distilled water, 22 parts.

One part of the carmine solution is to be mixed with 8 parts of the oxalic acid solution, and 12 parts of absolute alcohol are to be added. If the solution is orange-coloured instead of dark red, more ammonia is required, and the orange will become red. The orange colour may also be used for staining. If crystals of oxalate of ammonia become formed they must be separated by filtration.

201. **Thiersch's Lilac Colouring Fluid.**—Borax, 4 parts. Distilled water, 56 parts.—Dissolve and add, of carmine, 1 part.

The red solution is to be mixed with twice its volume of absolute alcohol, and filtered. The precipitate of carmine and borax is re-dissolved in distilled water, and is ready for use. It colours more slowly than the red solution.

202. **Anilin Colours.**—The beautiful reds and blues which have been lately so largely used as dyes, popularly known in this country as Magenta and Solferino, have been much employed by microscopists. The colour is not very soluble in water, but is readily dissolved by alcohol. A grain of the colour, 10 or 15 drops of alcohol, and an ounce of distilled water, make a dark red solution; or the colour may be boiled in water, allowed to cool, and then filtered. This fluid colours tissues very readily. Many exceedingly delicate and perfectly transparent *textures*, which are almost invisible in the natural state, can be most satisfactorily demonstrated by the use of this coloured fluid. The cilia of ciliated epithelium may be tinted while they continue to vibrate. As the substance of the cell becomes coloured, however, the action of the cilia ceases. Every kind of *cell wall*, *delicate membrane*, and *transparent tissue* may be tinted with these colours.

Magenta has been recommended by Dr. Roberts for showing a minute spot connected with the red blood corpuscles of man. ("On peculiar appearances exhibited by blood corpuscles under the influence of solutions of magenta and tannin"—"Proceedings of the Royal Society," vol. XIV, p. 481, No. 53, April, 1863.) The peculiar action exerted by magenta and tannin upon the red blood corpuscles has not yet been satisfactorily explained, but the late Dr. Hughes Bennett, of Edinburgh, told me that, with the aid of very high powers, he had demonstrated that the minute spot appearing after the blood corpuscles had been soaked in magenta exhibited angles, and he considered that it was in fact a minute crystal which had formed upon the corpuscles. This

explanation certainly does not apply to the spot which is developed in connection with *all* red blood corpuscles subjected to the action of tannin and other solutions.

203. Blue and Violet Colours for Staining.—Thiersch recommends the following fluid, the composition of which I take from Frey :—

Oxalic acid, 1 part.
Distilled water, 22 parts.
Indigo carmine, as much as the solution will take up.

Another solution of oxalic acid and water in the same proportions is required. One volume of the first solution is mixed with two volumes of the last and nine of absolute alcohol. The mixture is then filtered, and is ready for use.

An anilin blue fluid may be made as follows :—

Soluble anilin blue, $\frac{1}{2}$ grain.
Distilled water, 1 ounce.
Alcohol, 25 drops.

This fluid is not acted upon by acids or alkalies. Frey strongly recommends a fluid of this description as very useful for colouring many tissues.

Violet Staining with Hæmatoxylin.—"The ordinary extract, hæmatoxylin, is rubbed down in a mortar with three times its bulk of alum, till both are reduced to a fine powder, and well mixed. A small quantity of distilled water may now be added, and the whole well rubbed together for 15 or 20 minutes. More water may now be added and the solution, after filtration, should present a somewhat clear dark violet colour. Two drachms of 75 per cent. alcohol may now be added to each ounce of the solution."—"Monthly Journal of Microscopical Science," December, 1872, p. 277. Frey prepares the above in this way :—An aqueous solution of the extract of logwood is to be mixed with a solution of alum (1 part of the salt to 8 parts of water) till the deep red colour has become violet. The fluid is then filtered. These hæmatoxylin fluids may be used for fresh tissues, and for tissues hardened by chromic acid or alcohol. Tissues colour very rapidly and very deeply, in from half a minute to 10 or even 15 minutes. They may be mounted in Canada balsam or damar.

204. Tannin.—Although tannin does not colour animal membranes, it alters its character to such an extent as to enable us to see many peculiar points of structure or arrangement not visible before, or it produces a chemical change upon the substance, from which we gain important information. Solutions of magenta and solutions of tannin have been much used in investigations upon the blood corpuscles. The action of tannin upon the red blood corpuscle is very peculiar; it has been specially studied by Dr. Roberts, of Manchester, as mentioned above. The solution is made by dissolving 3 grains of tannin in

ounce of distilled water. One drop of blood may be mixed with 4 or 5 drops of the tannin solution and a portion of the mixture examined under the microscope.

205. Solution of Nitrate of Silver.—Of late years nitrate of silver has been used for staining tissues. Recklinghausen and His have employed this plan with great success. A weak solution may be imbibed by delicate tubes, and part being precipitated in the tube, perhaps as a chloride or in combination with some albuminous material, subsequently becomes decomposed by the action of light. A very dark line results, and thus the position of a previously perfectly invisible minute channel may be clearly demonstrated. The *outlines* of epithelial cells and the *intervals* between them may be demonstrated by this process. Transparent connective tissue and the *outer part of cells* can thus be coloured, the *nuclei remaining perfectly colourless and transparent.* The bioplasm, by longer immersion, will also be coloured, and probably for this reason.—As long as the bioplasm remains alive it resists the action of the solution, but when it dies, the matter resulting from its death imbibes the solution which remains with it.

The appearances produced by staining with nitrate of silver may be made to vary very much by modifying the mode of procedure and the time which the preparation is allowed to remain in the solution. After soaking in the nitrate of silver solution for some time the specimen must be placed in distilled water, or in a weak solution of common salt, in order to wash away the nitrate which adheres to the surface or occupies the intervals between the cells. When this has been effected the specimen is exposed to daylight or sunlight until the requisite degree of blackening has been obtained. The strength of the solution employed may be varied according to circumstances. Recklinghausen uses a very dilute solution, consisting of 1 part of nitrate of silver to 400—800 of distilled water.

The structure of the cornea has been recently investigated by His, after the tissue had been prepared according to this plan. The so-called "intercellular substance" (formed material) only may be coloured, or, after the whole structure has been thoroughly impregnated with the solution, the latter may be removed from the formed material, while that taken up by the nuclei (masses of bioplasm or living matter) is retained, and may be decomposed by being exposed to light. In this case the nuclei or bioplasts appear very dark and are surrounded by a pale brown formed material. His thinks that when the nuclei are coloured, the precipitate of chloride of silver in the formed material is re-dissolved and absorbed by them. It remains, and is afterwards reduced by the action of the light.

206. Solutions of Chloride of Gold.—Weak solutions of perchloride of gold have been much used of late years for colouring nerve fibres, for

it has been found that delicate nerves thus acted upon exhibit, after exposure to light, a blue or violet tinge. A solution containing from ½ to 1 per cent. in distilled water should be made. The tissue, after having been soaked till it becomes straw-coloured, is to be washed, and then placed in very dilute acetic acid, containing 1 per cent. or less. The nerves become coloured in the course of a few hours. By this plan, Cohnheim professes to have made out very fine nerve fibres, which, he says, pass from the plexuses in the cornea to intervals between the cells of the conjunctival epithelium, and after reaching the surface of the structure end in *terminal free extremities*. I think, however, we should receive such statements with the utmost caution, and although Professor Kölliker has accepted the view, I cannot adopt it without much stronger evidence than has been advanced in its favour. Many considerations make me think it will turn out to be incorrect. Cohnheim's drawings alone excite doubt in my mind concerning the accuracy of his observations, and, at least in my hands, the mode of preparation recommended has not afforded results nearly so satisfactory as those I have obtained by adopting other methods of investigation.

Many modifications of the above processes of investigation have been tried by me. I have found some advantage from using glycerine with the fluids, but at present I have no special plan to recommend. While I fully acknowledge the accuracy of many of the drawings and descriptions given of the appearances resulting from the use of nitrate of silver and chloride of gold, I am not convinced that many of the interpretations and conclusions which have been given and accepted concerning the structures demonstrated, are true. Some will, I think, have to be much modified in the future. The dark lines resulting from the silver process, which have been considered in many instances to be the outlines of epithelial cells, as for example in small vessels, mark, I believe, the lines of junction of the several elementary parts of which the tissue of the vessel consists. So, too, with reference to specimens prepared with gold, I am disposed to think that many of the lines which are rendered so very distinct by the black deposit will be proved to have nothing to do with the transmission of nerve currents, and that certain of the conclusions generally received will turn out to be incorrect.

207. Solution of Osmic Acid ($Os.O_4$) has been strongly recommended for demonstrating delicate nerve structures by MM. Schultze and Roudneff, because it tinges the white substance of Schwann and all forms of Myelin in various kinds of nerve fibres, of a very dark colour or almost black. Other textures are neither coloured so quickly nor so intensely, and often exhibit only a brownish tint. It is suggested that, with the aid of this substance, nerve fibres ramifying in various textures may be stained, and thus distinguished from other elements of the tissue. Solutions of various strengths may be employed but one part of

osmic acid in 100 of water is stated to be strong enough to produce the desired effect. I have tried this plan, but must confess that I have gained nothing by its adoption. I can show finer nerves and more clearly by other methods than any that I have been able to demonstrate either by the gold or osmic acid solutions.

208. Other Metallic Salts.—Tissues may also be impregnated with other solutions of metallic salts. Acetate of lead has often been employed. The tissue may be soaked for some time in a weak solution, or a weak solution with a little glycerine may be injected. When the tissues are well saturated, thin sections may be made, and, after having been slightly washed, they may be placed in a dilute solution of glycerine, through which sulphuretted hydrogen may be passed. Living plants will take up solutions of various metallic salts, which may then be precipitated in the textures or in the channels by the appropriate reagents.

209. Modification of the foregoing Plans.—The observer will perceive that the processes referred to under the head of "Staining the Bioplasm and Formed Material," are capable of almost endless modification. Every one engaged in a special investigation, will naturally try various modes of preparation. Having decided upon one which seems to offer considerable advantages, he will try various modifications until he meets with success. I have not attempted to give the minute recommendations of various observers who have employed some of these processes, but have merely indicated the general method of procedure. A few experiments will teach the observer more than the most minute instructions, and, however carefully directions may be given, it is seldom that any one succeeds the first time he endeavours to follow them out. Those who desire to do real work in this department must be patient, and must work on steadily, until they meet with success.

internal casts, or models, in silex, of the chambers and other cavities originally occupied by the substance of one animal."

275. Of Preparing Specimens of Coal for Microscopical Examination.—Coal is one of the most difficult substances to cut into thin sections. It is so opaque that no structure can be discerned unless it is ground exceedingly thin, and so brittle that it often breaks up in the operation of grinding. It is said that the coal may be softened by maceration in a solution of carbonate of potash, when sections may be cut with a razor. The sections are partially decolorised by being gently heated for a short time in strong nitric acid. When of a brown colour they are to be washed in cold water and preserved in glycerine ("Micrographic Dictionary"). Cannel coal, being less brittle than ordinary coal, is more easily prepared.

THE WORK TABLE—OF MAKING AND RECORDING OBSERVATIONS—FALLACIES TO BE GUARDED AGAINST.

The Work Table.—Although beautiful work tables, furnished with every possible requirement, have been designed for microscopists, I think the student will find that an ordinary table, which is firm and steady, is all that he really requires. It should, however, be well made and provided with a few drawers, in which the student can place his preparations for himself and arrange his instruments and apparatus in the most convenient manner.

The microscope should be always ready for use, and should stand on the table, covered with a glass shade, to protect it from the dust. This is far more convenient than the plan of keeping the instrument in its case, and going through the process of adapting the glasses, &c. and then removing them again, every time the instrument is required.

The object-glasses, eye-pieces, condensers, and other apparatus should be placed in a little cupboard, provided with shelves and having a glass door with lock and key.

Knives and scissors can be kept in a shallow box, having a glass cover. Drawing instruments in a second. Thin glass and glass slides with watch glasses, or little saucers, in a third. These trays should be properly partitioned, and should be covered with a plate of glass to keep out the dust, and kept ready for use on the work table. A glass of clean water should always stand on the table, and some pipettes, stirring rods, and camel-hair brushes, all perfectly clean, should be provided. The injecting apparatus and instruments which are only required occasionally may be kept in one of the table drawers. A portfolio or pamphlet box is necessary for keeping drawing paper, cardboard, tracing and retracing paper, scales for measuring, small gummed labels for attaching to the slides, &c.

All things really necessary for ordinary microscopic work may

be purchased for two or three pounds, but it is easy, of course, to spend many pounds or more upon a microscope table and apparatus. I have myself always made use of an ordinary good strong library table, fitted with drawers underneath, and I think it would have been difficult to contrive anything upon the whole more convenient or better adapted for work. The microscope stands on the table, always ready for use, under a bell jar, and the lamp, fig. 5, pl. XIV, p. 24, with scissors, knives, needles, and other tools in frequent use, close by.

276. Of Keeping Preparations in the Cabinet.—Preparations mounted in the dry way, or in Canada balsam, may be kept upright, arranged in grooves, but all preparations mounted in fluid must be allowed to lie perfectly flat, otherwise there will be great danger of leakage. Cabinets, holding several hundred specimens, arranged in this manner may now be purchased of the microscope makers, for a very small sum, but if the observer is provided with deep drawers, they may be made available for the purpose, if a number of shallow trays of cardboard are carefully arranged to fit them accurately. Each preparation should be named as soon as it is put up, and it is convenient to keep a number of small gummed labels always at hand for this purpose. Once or twice in the year a new layer of Brunswick black should be applied, and the specimens carefully examined to see that no leakage has occurred. The cases now generally sold are, I think, preferable to cabinets, and of the cases I have seen the most convenient are those suggested by Mr. Piper, and sold by Mr. Swift, Mr. Collins, and others. They are made for two, three, six, and twelve dozen specimens, costing respectively *2s. 6d.*, *3s. 6d.*, *5s. 6d.*, and *10s.* Cases made of deal are arranged on the same plan.

277. Of making Observations upon Specimens in the Microscope.—If, upon examination, a specimen does not appear to the observer to verify the description or drawing which some authority has given of a similar structure, he must not too hastily infer that the author has been recording the results of his imagination rather than observed facts. The conclusions which have been arrived at are probably the result of a very long and patient investigation, deduced from examining many specimens, perhaps under very different circumstances, after the application, it may be, of various chemical reagents, and after ascertaining the effect of different preservative media. From the remarks already made, some idea may be formed of the many different operations which are necessary to demonstrate conclusively the anatomy of a single tissue. The beginner must not, therefore, be hasty in deciding as to the exact nature of the object which he sees in the microscope; neither must he infer that what he has not been able to see does not therefore exist. His eye and mind will require much careful education before he can hope to be able to form a correct judgment concerning many things that he will meet with.

Some students are liable to fall into an error of another kind, but [illegible] less detrimental to forming habits of correct observation. Led away by their imagination, they think they see everything which has been delineated, or which they have heard described; the observations of authors are confirmed in the most positive manner, and expressions closely resembling those already employed are used by the observer who follows them. But, in fact, an author's own assertions are simply reiterated in favour of his doctrines by a believing follower, and no real confirmation of the accuracy of his views is advanced. In this manner errors have been intensified and propagated to an extent almost incredible, and it will require years of laborious investigation to overthrow statements which indeed never resulted from actual observation, which were erroneous from the first and ought in fact never to have been received. Sometimes a mere guess remarkable for ingenuity and novelty, but having no foundation in fact, is seized upon by a number of persons and "supported" by so many assertions, misnamed observations, that it is soon received as true, and is perhaps believed in for years, until at last some one reinvestigates the whole question, and demonstrates the absurdity of the received doctrine.

Of the Importance of making Sketches.—The great importance of drawing has been already referred to. Even sketches in outline are of great value if the size of the object has been correctly registered. Measured plans are of great use in many cases and supersede the necessity of description. This subject has, however, been fully considered already. *See* p. 31 to p. 41.

278. Of Drawing Inferences from Observations.—No one engaged in the pursuit of any branch of natural science is more tempted to indulge in hasty generalisation than the microscopical observer. It is his duty, therefore, to avoid drawing inferences until he has accumulated a sufficient number of facts to support the conclusions at which he has arrived. True generalisations and correct inferences promote the rapid advancement of scientific knowledge, for each new inference may form the starting-point of a fresh line of investigation; but, on the other hand, every false statement accepted as an observed fact, assists to form a barrier to onward progress. Before the slightest useful advance can be made it will be necessary to go back, it may be for a long way, before we can hope to recommence with any prospect of success the onward course. Moreover, a much greater amount of evidence is always required to overthrow a false conclusion than is sufficient to propagate the original mistake; and there can be no task more unsatisfactory than that of being called upon to controvert the opinions and deductions of others, however desirable and necessary such work may be.

In any special enquiry I think it is a good plan *not* to make too many or very full notes during the progress of the investigation, but to retain in

the memory, as far as may be, the facts observed. When the whole [illegible] is made out, but not before, the observer may begin to write and record his observations. Otherwise, imperfectly observed facts are liable to be set down as actual facts, and afterwards commented upon as if they were well-ascertained truths, and there is danger of the observer being gradually led more and more astray, until at length he commits himself to a conclusion totally at variance with the real truth.

Scientific enquiry should continually advance, and we ought to be able to extend researches from the point where they have been left by our predecessors, each succeeding observer adding to what his predecessors had discovered. In not a few instances must we feel the highest respect for the careful observations of the older observers, and I fear it must be reluctantly confessed, that many modern researches are not carried out with the same patience, painstaking industry, and conscientious care as theirs have been, and for this reason are likely to be but short-lived. Many recent observations strongly insisted upon and advocated with great vehemence, purporting to depend upon actual demonstration, have been set aside for others still more recent, and, if possible, more incorrect. False observation has, as would be supposed, created in some minds complete scepticism of all observation, and has deplorably retarded true progress. It is quite curious to notice how some writers condemn theory and commend what they term the observation of facts, as if it had been incontestably shown that results arrived at from cogitation and speculation must be invariably false, and those from "observation" as invariably true. But any one who has had experience in microscopical enquiry knows how difficult it is to prove that what he discerns is really the thing as it actually is in nature, and not a mere fanciful interpretation of his own. It is easy to assert a particular thing has been observed, but in many cases there is the greatest difference between the thing and what it is supposed has been seen. Many indeed have been the errors introduced by speculative thinkers, but I doubt whether more are not in these days advanced by self-styled practical observers, than by those whom the latter are ever ready to condemn as mere theoretical dreamers. A man says he has seen such and such a thing, and gives drawings of the thing seen. He explains to friends what he has seen, shows them the object in question, tells them what they are to see, and they, knowing nothing about seeing, but not liking to offend their friend, or being too lazy to trouble themselves about the matter, *say* they see the thing as they have been told it is to be seen. Such is the *evidence* which when duly chronicled and printed seems to amount almost to actual proof. But how many, many times has this process been repeated in the case of almost every doubtful anatomical point! The conclusion is almost forced upon the mind that the process of observing facts leads to results at least as unsatisfactory and as fallacious as the process of imagining and specu-

lating without observing at all. At this time what a mass of thoroughly conflicting evidence exists on many important matters, supposed to rest upon scientific observation and experiment! Three or four views are taught concerning first principles of anatomical and physiological science, each one being incompatible with the rest, but nevertheless supported by an immense amount of what purports to be evidence based upon observation. In such a case as this it is obvious that many of the statements must be false, and many of the supposed facts advanced must be erroneous; and yet with what pertinacity are such erroneous facts maintained, and how widely are they spread, as if indeed they were the most unquestionable truth! What an amount of work must be done, and what a length of time must elapse before false facts can be demonstrated to be really false and true facts proved to be really true!

Years must be passed in patient investigation before a man ought to trust himself as an observer of facts, and it is only by careful and unremitting exercise that any one can gradually acquire habits of attentive observation, and the power of thoughtful discrimination which alone renders his conclusions reliable. Indeed, though he labour hard and earnestly, he will scarcely have properly educated himself ere his powers begin to decay and he becomes liable to err from the natural deterioration in structure of the organs upon which the observation of the facts he observes entirely depends.

279. Of Recording the Results of Microscopical Observations.—Taking notes of microscopical observations is a subject of great importance. The observer must endeavour to acquire the habit of describing in words the appearance of objects under the microscope. This is probably not so easy as would at first be supposed, although undoubtedly many persons are able to describe what they see much more correctly and with greater facility, than others. Accuracy in describing microscopical specimens can only be acquired by practice, and I think it a most excellent rule for a student, when he begins to observe, to take notes of the appearances of every object submitted to examination. The time is well spent, and much of what is so described is retained in the memory. The notes should be short, and should consist of a simple statement of points which have been actually seen. *Inferences* should be carefully avoided, and nothing should be stated without the observer being thoroughly satisfied of its accuracy. If he is not quite certain of any observation, he should express his doubts, or place a note of interrogation after the statement. The use of indefinite terms should be avoided as much as possible, and whenever any particular word is used, a definite meaning should be attached to it. Much confusion has arisen from the use of terms which have not been well defined. For instance, the word "*granule*" has been applied by many authors to a minute particle which appears as a small speck, even when examined

by the highest powers, as well as to a small body with a perfectly clear centre, and with a well-defined sharp outline, which would be more correctly termed a small "*globule*." So, again, the term "*molecule*" has been employed in some cases synonymously with "*granule*," though it would be obviously wrong to speak of a small globule as a molecule. It seems to me very desirable to restrict the terms "granule" and "molecule" to minute particles of matter which exhibit no *distinct form* when examined by the highest powers at our disposal, and the term "globule" to circular or oval bodies of all sizes which have a *clear centre* with a *well-defined dark outline*. Other examples of the use of insufficiently defined terms might be pointed out. If an observer makes use of a term which is generally employed without any definite meaning being attached to it, he should describe at length the meaning which he assigns to it, and should, of course, use it only in this one sense.

Exactness of Description should always be aimed at, and we must remember that with a little trouble this exactness may often be obtained with the use of a small number of words. That appearance of precision which is often affected by those who give long useless descriptions of what they have observed cannot be too much condemned. So, also, the practice of some of minutely describing the characters of every object in the field of the microscope without the slightest knowledge of the nature of any one of the objects presented to the view, has been the cause of much ridicule, and has brought microscopic observation into great disrepute. The publication of a very detailed description of a number of not very definite objects, the nature of which is undetermined, though sometimes regarded as evidence of careful and elaborate enquiry, is a most useless procedure. Some have thought to gain the credit of being accurate observers by carefully measuring every object they see in two diameters, and putting down in fractions or decimals the results of this useless ceremony. Such reports may be regarded as indications that the author has been thinking more of himself, and the importance of making an impression upon his readers, than of his subject. He wants to be credited with a character for extreme minuteness of observation, and instead of striving to advance the real interests of the science which he professes to serve, and endeavouring to excite in the mind of the reader a desire for more extended knowledge, and a longing to take part in a similar investigation, he is perpetually endeavouring to thrust himself into notice. He who feels a real love for his subject will try all he can to enlist others in the same service; he will endeavour to remove all difficulties of investigation, and will explain what he himself has learnt in language which shall be intelligible to all. Extreme minuteness in description is by no means an indication of accuracy of observation. Pretence of extreme accuracy has encumbered science with many unnecessary words, and earnest men

have been deterred from its prosecution. A certain mysterious air pervading the description of an observation, an evident desire to coin new words and indulge in statements couched in exaggerated language about the importance of the facts observed, are quite misplaced where all should be clear, simple, and intelligible to every one, and too often indicate indifference to the subject on the part of the author, and a want of proper consideration towards unlearned readers. That affectation of precision, and verbose pompous style of description, which have been fashionable among some microscopists, and which pervade the writings of several authorities in this imperfectly developed branch of investigation, have offended earnest persons who have devoted their lives to the prosecution of different branches of natural science, and have also greatly retarded the real progress of scientific enquiry. All this is but pretence, and not real, earnest, useful work. It is distasteful to every scientific man and discouraging to every student.

Fallacies to be guarded against in Microscopical Investigation.

Many mistakes have arisen in consequence of sufficient care not having been taken to prevent the introduction of various substances by accident. The most scrupulous care must always be observed in microscopical examination, and any foreign particles which may have accidentally come into contact with the preparation must be removed before it is mounted. The proceeding to be followed to remove the foreign matter, will depend much upon its nature. Mere dusting with a camel hair brush, washing with a stream of water, or picking out the object with needles, are simple plans which are often efficient in a general way, but in some cases other processes are required.

280. Errors of Observation.—It is of the highest importance that the student should do his utmost to avoid making and recording erroneous observations. Not only is the student liable to draw false conclusions from observations, but the observations themselves are frequently erroneous. I propose therefore, to direct the student's attention to a few instances in which difficulty and doubt may be experienced even by skilled and practised observers :—

Of the Commencement and Termination of Tubes.—The modes of commencement or termination of certain vessels or tubes have long been sources of dispute among observers. There are not a few instances where positive statements have been made that certain tubes commenced by cæcal or blind extremities; while contradictions equally positive have been advanced by others, who have affirmed that the very same tubes commenced as a network, and presented no blind extremities whatever. It would be generally supposed that such a point might be determined beyond all doubt if the tubes were injected with some

coloured material. But the supposition is not correct. Injection will frequently run up to a particular point in the minute vessels, while no force which can be employed will drive it further onwards. Here, therefore, it accumulates, and often to a very considerable extent; the portion of the tube above the constriction being considerably dilated by the pressure. Under these circumstances, owing to the extreme transparency and delicate nature of the tissue of which its walls are composed, it may be impossible to trace the further continuity of the vessel. Indeed, the vascular walls will probably be quite invisible in an unprepared specimen. The observer is thus led into the error of supposing that such tubes terminate in blind extremities, whereas they may really form a network with large meshes, or they may be continuous with tubular structures of a different character. In fact that which was taken for the termination or commencement of the tube may really be nothing more than a bulging in a central part of its course. In many thin sections of the kidney an appearance as if the tubes terminated in free blind extremities is produced in consequence of the convolutions lying in such a position that the recurved portion is immediately beneath the most superficial part of the tube. From a mere examination of the specimen it would be impossible for any one to say that this was not the case. In such instances the real disposition of the parts is only to be made out by a careful examination of the structure under different kinds of illumination and prepared in various ways. Thus the idea that the tubes end by blind extremities may be shown to be quite inconsistent with the appearances observed in specimens prepared according to a different method. Were I able to devote space to the consideration of this part of my subject, I might review the various methods in which a tissue may be examined, and show how by a consideration and comparison of the different facts observed, one is enabled at length to embody the results arrived at in the course of several different enquiries, and thus gain an accurate conception concerning the real structure of the part.

On the Difficulty of Seeing Structures from their Transparency.—Another fallacy arises from the great transparency of certain structures. Oftentimes a membrane may appear perfectly clear and transparent, while in reality it is covered with a delicate layer of epithelium, which only becomes visible when the tissue is immersed in some special fluid or treated with some particular chemical reagent. On the other hand, there are instances in which an appearance resembling that produced by the presence of a cellular investment is perceived where no cells whatever exist. A peculiar corrugated state of uninjected capillaries, and the bioplasts in the walls of the capillary vessels themselves, sometimes give rise to these mistakes. *Basement membrane*, from its extreme delicacy and transparency, is often only recognised by the folds into

which it is thrown, or by the débris and granular matter which is accidentally deposited upon it. Sometimes it becomes visible when immersed in a slightly coloured solution, instead of in perfectly pure water. Not only may blood and lymphatic vessels be completely passed over from their transparency, but I could adduce instances in which broad bands of connective tissue and bundles of nerve fibres existed in a specimen in great numbers, although they could not be seen when the ordinary methods of demonstration were employed.

Fibres and Membranes Produced by the Action of Reagents artificially.—On the other hand, by the action of reagents a fibrous appearance is sometimes produced which, without care, may be mistaken for actual structure.

The addition of acetic acid to many preparations frequently produces a swelling of the tissue, with the elevation of a clear membrane-like structure, which might be termed basement membrane, but which has really been produced by the action of the acid. Thus the outer uncalcified portion of the cells of the enamel of a young tooth, may be made to swell up into a transparent mass, which has been mistaken, I think, by Professor Huxley for a membrana performativa, which does not exist in this situation.

A Fibrous Appearance Produced in Structureless Membranes.—Clear, transparent, and apparently structureless membranes, when pressed, torn, and twisted, have a fibrous appearance; and delicate vessels, whose coats are perfectly transparent when pressed and collapsed, may be very easily mistaken for a form of fibrous tissue. Both capillaries and fine nerve fibres may be mistaken for fibres of elastic tissue. Indeed, capillaries uninjected and stretched, can only be distinguished from fine nerve fibres with difficulty. If any doubt exist in such a case, it may always be cleared up by injecting the capillaries of the part with a clear transparent material, like plain size, or the transparent injecting fluids, recommended in pp. 106 to 113, when, if the fibrous appearance is not real it will be lost; while if fibres really existed, they would still be visible. The presence of capillary vessels in a structure has been entirely overlooked in consequence of their being collapsed and shrunken, in which state they have been regarded as elements of the connective tissue.

Collection of Oil Globules Appearing as if within a Cell.—Oil globules in fluid not uncommonly form small and nearly spherical masses or collections, which become covered with a certain quantity of mucus or viscid matter and granules, originally contained in the fluid, so that the little intervals between the minute oil globules become filled up. The outline of the mass is perfectly clear, and sharp, and well defined, and from mere ocular examination it would be impossible to say that the oil globules were not enclosed in a cell-wall. A consideration of the circumstances under which such structures have been met with, will often

assist us materially in determining their real nature. Such "cells" may be prepared artificially without the least difficulty, and in some cases it would not be possible to distinguish by microscopical examination in water the artificially formed *cell* from the natural *cell*; and the process of staining the bioplasm, p. 123, would only enable us to form a positive conclusion in cases in which it was certain that the natural cells were quite fresh. It need scarcely be said, however, that with respect to the *formation* of these bodies no analogy whatever obtains in the two cases. Of the artificial cell the most external part was *last formed*. It was deposited around a collection of particles. But in the natural cell the outer part is the *oldest part*. It was produced *before* the matter in the central part of the cell was formed. No one at this time maintains that living cells are formed by the aggregation of granules, though some seem to think that a bacterium may be formed by the coalescence of already existing particles. Such persons must admit, however, that such simple organisms *multiply* by division, and thus they seem to affirm that living things may be produced by the coalescence of separate lifeless particles, and that they increase and multiply by the division of the resulting mass. It need, however, scarcely be stated that facts now known render such a doctrine untenable. *See* a controversy upon this subject in the "British Medical Journal," January, February, March, 1864.

On the Accidental Presence of Extraneous Matters.—Cleanliness is of the utmost importance in every branch of microscopical enquiry, and without great care many substances of extraneous origin will be introduced into the specimen about to be examined, and the observer may mistake the character of the objects accidentally introduced for those of the special objects under examination. For instance, particles of starch or other solid bodies may gain entrance into a texture submitted to microscopical examination, and the observer may draw the very erroneous inference that these bodies were embedded in the substance of the tissue, and that they were developed and grew in this situation.

When we consider how very minute are many of the structures rendered evident to the eye by the microscope, we shall scarcely wonder that many light substances are liable to come in contact with the specimen which is under examination. The cotton or flax fibres from the cloth, starch globules which adhere to the thin glass circles (for the small pieces are often kept in starch), portions of feathers, various kinds of hair, and oil globules are among the substances which are most frequently met with in examining different specimens, and I need hardly say that their presence is purely accidental. That I am not advocating needless precaution, is shown by the fact that in a well-known and highly valuable publication printed a few years ago, a drawing of what was evidently a portion of feathers was described as a representation of

lymphatic vessels,—vegetable hairs were described as *nerve fibres*, and several other errors equally unpardonable were to be found. Now such mistakes could only be made by a person utterly ignorant of the characters of some of the commonest objects with which every microscopical observer ought to be thoroughly familiar. I would strongly recommend every one to carefully study the characters of the substances of extraneous origin enumerated below before he attempts to make any original observations. He is sure to meet with the bodies in question from time to time, and the sooner he becomes well acquainted with their characters the better.

The following should be very carefully examined :—

Oil globules, milk, pl. XXIII, p. 80, figs. 11, 12, 13, 14.

Potato, wheat, and rice, starch; and bread crumbs, pl. XLVI, p. 172, figs. 1, 2, 3, 4, and pl. LX, fig. 3.

Portions of feather; worsted, pl. LX, fig. 3.

Fibres of flax; cotton, pl. LX, fig. 3, *e*; and silk of different colours.

Human hair, cat's hair, hair from blankets, fig. 3, *a*, *b*, *c*.

The scales of butterflies and moths, particularly those of the common clothes moth, pl. LX, figs. 1, 2.

Fibres of wood swept from the floor, fig. 4, fragments of tea-leaves, hairs from plants, vegetable cellular tissue, and spiral vessels, pl. XLVI, p. 172, fig. 5.

Particles of sand.

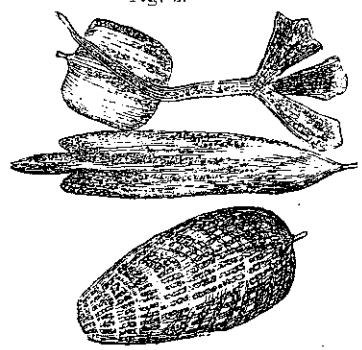
Many of these extraneous substances are figured in the plates I have referred to, and I beg the student will not only examine my drawings, but place actual specimens of all objects delineated under his own microscope.

In the examination of deposits from fluid we must bear in mind the possibility of the introduction of a small quantity of one deposit into another carried upon the extremity of the pipette used for examination, and in this simple manner much difficulty and confusion may be caused to the microscopist. The pipette should therefore be well washed immediately after it has been used, and the water in which it is washed should be very frequently changed. In taking fluids from different bottles and other vessels the possibility of introducing various substances must be borne in mind.

Fig. 1.

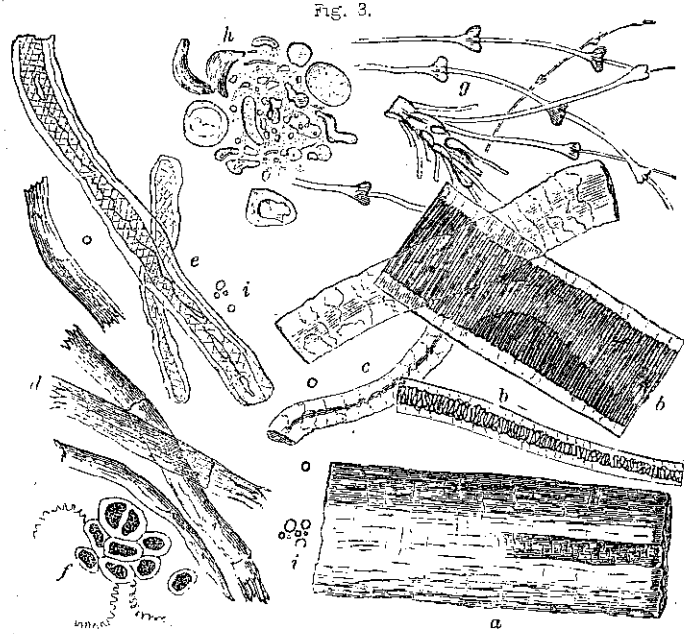
...m the wings of the common clothes moth. × 215.

Fig. 2.



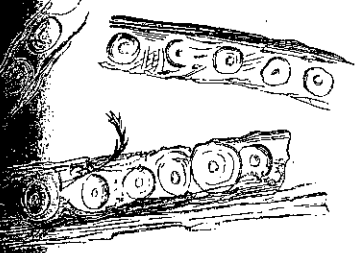
Scales from the wings of the common clothes moth. × 540.

Fig. 3.



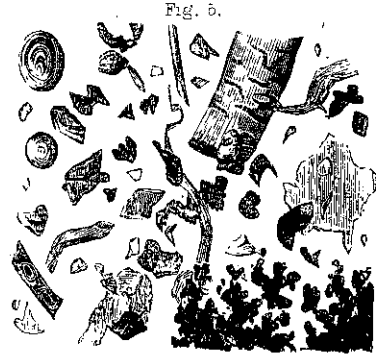
...gments of human hair. *b*, cat's hair. *c*, hair from blanket. *d*, fibres of flax. *e*, fibres of cotton. *f*, fragments of tea-leaves. *g*, portions of feather. *h*, bread crumbs. *i*, free oil globules.

Fig. 4.



...s of deal wood swept from the floor × 215.

Fig. 5.



Particles of 'dust' removed from a ledge in a

PART V.

ON TAKING PHOTOGRAPHS OF MICROSCOPIC OBJECTS — APPARATUS —ILLUMINATION — CHEMICAL SOLUTIONS — PRACTICAL MANIPULATION—PRINTING—PHOTOGRAPHS FOR THE MAGIC LANTERN.

Many improvements have been introduced in the method of taking microscopical photographs, and far greater perfection in the results has been obtained than was supposed to be possible some years ago. My friend Dr. Maddox has continued his experimental investigations with continually increasing success; and many observers in Germany and France, as well as in America and in this country, have produced beautiful photographs of various kinds of objects. Some of the most perfect photographs of animal tissues I have ever seen were obtained by Mr. Hugh Bowman in 1875–6.

Very remarkable progress in this department was made in America in 1864. The authorities in the War Department recognising at once the high importance of photographic representations of microscopical specimens have issued a series of reports in which will be found the results of the researches of Brevet Lieut.-Colonel Dr. J. J. Woodward and Brevet Major Dr. F. Curtis. These reports are admirable. The drawings are beautifully executed, the paper well adapted for them, and the printing excellent, contrasting remarkably in all these points with the rough looking Blue Books issued under the authority of our Government. It seems to me very hard that British statesmen do not more distinctly announce that they fully appreciate the high importance of purely scientific investigation than has been the custom hitherto. Our Government clearly ought to take a very active part in advancing new methods of enquiry; particularly in connection with naval and military medicine and surgery. In the medical department of our army there are to my knowledge scientific men as able and as willing to devote themselves to scientific work as any in the world, but they have little opportunity, and little encouragement seems to be afforded by the high military authorities.

I append an extract from p. 149, Circular No. 6, Nov. 1865, War Department, Surgeon-General's Office, Washington, and hope that per chance it may be brought under the notice of some of those who alone

have power to forward or obstruct scientific progress in the departments under Government control.*

"With low powers no serious obstacle was encountered in obtaining excellent photographs of properly selected preparations. The higher powers offered difficulties most of which however have been overcome. In experimenting with the higher powers, the lined diatomaceæ were selected as test objects on account of their definite and well-known structure. With these the utmost success has been realised. A photograph of Gyrosigma angulatum (Navicula angulata) has been obtained, for example, magnified about 7,000 diameters in which the hexagons appear of the same size and nearly as distinct as in the cut, which was made by transferring to wood a tracing from the original photograph. In fact, any of the markings on the diatoms that are visible with the microscope can be photographed with the utmost clearness and ease, and the time has arrived when the inability to photograph alleged markings will throw doubts on the correctness of the observers who have supposed they saw them. The plan employed in the photographic work hitherto executed with high powers is as follows: The direct rays of the sun reflected in a constant direction from the mirror of a Silbermann's heliostat (lent for the purpose by the Coast Survey), are condensed by a large lens upon the plane mirror of the microscope, whence they are reflected through the achromatic condenser in the usual way. Before reaching the achromatic condenser, however, the rays pass through a cell containing a solution of the ammonio-sulphate of copper of sufficient density to absorb nearly all the rays except those at the violet end of the spectrum. The light used, therefore, is essentially monochromatic, and contains, with enough illumination for agreeable vision, the greater part of the actinic force of the sun's rays. The heating rays being chiefly at the other extremity of the spectrum are of course excluded and great actinic force is obtained, therefore, without any danger to the preparations, or the balsam used for cementing the object-glasses. The object-glass employed in the photograph of Gyrosigma above alluded to was a one-eighth of an inch, by W. Wales and Co., of Fort Lee, New Jersey. This glass is so corrected as to bring the actinic rays to a focus. At the bottom of the draw-tube was placed an achromatic concave lens—the amplifier of Tolles (of Boston, Mass.), and an ordinary medium eye-piece com-

* I believe that it would be most difficult, if not actually impossible for our Government at this time to issue a report of the character of that from which the extract is taken, supposing that the actual work had been done by private persons and placed at the disposal of the State. The paper of our Blue Books is too coarse, and the printing too rough for scientific memoirs. Let the reader, for example, compare the plates accompanying my report on the Cattle Plague, which were printed by Government, with those in the present work. The contrast between the text of Government and private works is still more striking.

pleted the optical apparatus. The eye-piece extremity of the microscope was thrust into one end of a long camera-box, the connection made light-tight by means of a black silk hood, and the image received on a piece of plate-glass, observed by means of a focussing glass, while the focal adjustments were made. As with the very long camera used, the arm of the observer cannot reach the milled head of the fine adjustment of the microscope, this head was grooved, and connected by a band with grooved wheel at the end of a long steel rod, the other extremity of which is near the observer, who, by means of it, can focus accurately with any required length of camera. There is nothing peculiar in the chemicals employed, and with ordinary collodion, and the high power above spoken of, from thirty to forty seconds' exposure was quite sufficient. On the foregoing devices most importance is to be attached to the employment of monochromatic light (the violet end of the spectrum), and the use of an object-glass constructed with special reference to the actinic rays. Both these points were suggested to me by Mr. L. W. Rutherfurd, of New York, so well known by his connection with telescopic photography, who has thought much, and made many satisfactory experiments in this direction. I believe, however, that the apparatus as above described, loses some of its advantages by the use of the eye-piece, which I propose to substitute by a lens of proper magnifying power, corrected, like the object-glass, in such a way as to bring to a focus the actinic rays. Such a lens is now in process of construction for further experiment. The pathological photographs hitherto satisfactorily executed in the Museum have chiefly been made with moderate magnifying powers, twelve to fifty diameters, though some experiments with high powers justify me in the belief that with the improvements above described, all that is desired in this direction can be attained. Among these experiments I may particularly mention a view magnified about four hundred diameters, of the polygonal cells and flat cholesterin tables of a cholesteatoma, which was found on the inner surface of the frontal bone of a soldier who died of epilepsy in the neighbourhood of Washington." Such an extract is enough to show the activity and usefulness of the department by which it is issued, and is in the highest degree creditable to those who performed the work, and to the Government which sanctioned and encouraged its prosecution.

323. History of the Application of Photography to the Microscope.*—Wedgewood and Sir Humphry Davy published, in 1802, experiments which must have been made some years previously, as Wedgewood died several years before this date. They obtained photo-micrographic impressions on paper and leather, but these they were unable to render permanent. These are believed to be, at the same time, the first experiments in photography and photo-micrography. (John Towler, M.D., in his "Silver Sunbeam;" Captain Abney, in "A Treatise on Photography," p. 2.) Mr. Dancer, about 1840, produced photographs of microscopic objects by the *gas microscope*, the images being taken upon silvered plates; also images of sections of wood, fossils, &c., were reproduced on paper and glass plates by means of the solar microscope. In 1841, Mr. Richard Hodgson obtained excellent daguerreotypes of microscopic objects. The Rev. J. B. Reade and the Rev. C. Kingsley and Mr. Talbot were early authorities in the employment of photography in connection with microscope observation. The Rev. J. B. Reade, then living at Peckham, as early as 1837 obtained and fixed photographs on paper, washed with silver nitrate and an infusion of galls. He succeeded by means of the solar microscope in photographing entomological specimens and sections of vegetable tissues. Two years later, in 1839, Mr. Reade exhibited more perfect results at a *soirée* given by the Marquis of Northampton, the President of the Royal Society. In the same year, it appears that some of his photo-micrographs were offered for sale at a bazaar at Leeds. (See a review in the "Medico-Chirurgical Review," July, 1864.) Dr. Donné, of Paris, in 1840, presented to the Academy of Sciences copies of various microscopic objects on daguerreotype plates. Moitessier, in his "La Photographie appliquée aux recherches Micrographiques," remarks:—"En 1845, ce savant (M. Donné) publiait, avec M. Léon Foucault, un magnifique atlas relatif à l'étude des fluids de l'économie, et contenant un grand nombre de figures gravées d'aprés des images daguerriennes."

In October, 1852, a paper by Mr. Joseph Delves was presented to the Microscopical Society of London, and in the following number of the "Quarterly Journal of Microscopical Science," some beautiful specimens of prints from Mr. Delves' collodion negatives were issued by the then publisher, Mr. Highley. This was one of the earliest publications in this country with photographic illustrations of microscopic specimens. Subsequently many workers appeared, among whom may be mentioned the following:—Highley, Shadbolt, Dr. Hugh Diamond, and Mr. Archer (1851), Busk, Hodgson, Durham, Maddox, Howlett, Beckett, Pollock, Wenham, Kingsley, Traer, Weightman, Davies, Parry, Wilson, Abercrombie, Taylor, Sanders, Viles, Herapath, Legg, Bowman; and in India, Gayer, Eddowes, and others. In France may be mentioned the names of Donné, Foucault, Nachet, Dubosq, Bertsch, Moitessier, Verroquier, Girard, Duchenne, Rouget, Lackerbaumer, Ravet. In Germany, Gerlach, Albert, Mayer, Kolman, Helwig, Reichardt, Ehrenberg, Pohl, Weselsky, and Siebert, have illustrated memoirs with photographic plates. In Italy, Castracane. In Belgium, Neyt. In

* Many of the sections which follow have been carefully revised by Dr. A. Clifford Mercer, of Syracuse, N. Y., who has kindly added much new matter of great importance.

the United States, Rood, Draper, Towler, Crehore, Dean, Rutherford, Woodward, Curtis, Seiler, Ward, Kempster, Deecke, Mercer.

Sir D. Brewster, in his article Microscope, "Encyclopædia Britannica," last edition, speaks very highly of some photomicrographs exhibited at the Academy of Sciences, Paris, in 1857, by M. Bertsch, the focal length of the objective used being half a millimetre. The objects, a diatom from guano magnified 500 diam.; two specimens of navicula, one × 800, the other × 500, the field being rendered nearly dark by oblique illumination; human blood globules × 500; and two pictures of salicine, taken by polarized light. M. Hartnach, Sir D. Brewster says, has constructed a complete instrument for M. Bertsch, the range being from 50 to 1,000 diameters, and from 50 to 1,500 diameters for opaque objects. The extreme detail, beauty of texture, and sharp delineation of the objects in the prints from Mr. Delves' negatives marked a very important step.

The frontispiece to former editions of this work was obtained by Dr. Maddox in the following manner, as described in a note to me:—

"Prints selected from some of my negatives, representing objects magnified in various degrees, varying from the 1½ inch objective to the 1-12th, were placed on a card in such a manner as to try to balance each other in their effects, and such size of card adopted that when reduced *one-half*, it might correspond with the dimensions chosen by yourself for the plate. The card of prints being placed at the requisite distance, a Ross' 15-inch focus landscape lens was used to obtain the negative copies.

"To render the minutest line, especially in the Pleurosigma angulatum, well evident in the negative, it was necessary not to carry the development or intensifying process too far, or the lines became filled up and much obscured, hence the interspaces between the figures allowed a little light to pass; as this seemed detrimental and rendered the figures less effective in appearance, these parts have been painted out.

"The illustrations were photographed with the objective stated in the 'explanation.' The 1-12th objective was made by Mr. Wenham, and through his liberality placed at my service."

Many of these photographs require a magnifying glass to bring out their detail. My friend Dr. Dean, of Boston, U.S., sent me some very perfect photographs of sections of the medulla oblongata, taken with low magnifying powers. These are by far the most perfect photographic illustrations of structures from the higher animals that I have seen. ("The Grey Substance of the Medulla Oblongata and Trapezium," by John Dean, M.D., Smithsonian Contributions to Knowledge, 173. Washington, 1864.) These photographs were also successfully printed by photolithography. Dr. Duchenne, of Boulogne, also obtained some very successful results with anatomical structures, and M. Rouget has

employed the same means in the ordinary way and stereoscopically, to illustrate some of his views on minute structure. In 1865, Dr. A. Ehrlwig, of Mayence, published his work "On the Crystalline Forms of Alkaloids, and their Sublimates," &c., illustrated by a large number of photomicrographs. Dr. Moitessier has also adorned his book on photomicrography, "La Photographie Appliquée aux Recherches Micrographiques, 1866," with three photograph plates of various objects.

Dr. Draper, of America, employed for many of the plates in his work "On Anatomy and Physiology," woodcuts from photographs of the microscopic objects, and Dr. Herapath, of Bristol, adopted a similar method for his paper on the Spicules and Plates of Synapta, published in the "Quarterly Journal Microscopical Science." Photography has been used by Dr. Maddox to illustrate a paper presented to the Royal Society, June, 1867; the photographs being made from an aquatic Larva whilst living.

Many anatomical specimens, however, cannot be copied by photography, especially if they be very thick. The yellow colour of the tissue in most instances precludes the possibility of making a photograph of it, as the transmission of the light is so much interfered with; and this is an especial objection in the case of injections viewed as transparent objects, for the tissue intervening between the vessels is often so yellow that these intervals in the photograph become as dark as the vessels themselves. My friend Dr. Julius Pollock nevertheless succeeded many years since in obtaining some very tolerable copies of injections of the distribution of the ducts in the liver. And Dr. Maddox and Mr. Hugh Bowman have been still more successful.

When only few copies of a work are required, the researches may be very cheaply illustrated by taking photographs of drawings. A large drawing of the object must first be made in the manner described in p. 33. From this a negative reduced to the proper size is taken, from which any number of copies may be obtained. In this manner I have illustrated my memoir on the Anatomy of the Liver, with upwards of sixty illustrations ("The Anatomy of the Liver," 1856). The results were not so satisfactory as they might have been, but as all the prints were prepared at home with very limited appliances, very good prints could not be looked for. When many copies of a work are likely to be required, this mode of illustration is not applicable, as the original cost of engraving would soon be covered; but when only a *few* copies of a *great number* of drawings are wanted, this plan possesses decided advantages.

From the improvements in the Albertype, Woodburytype, photolithographic, and other similar processes, there seems every chance that the cost of illustration will be materially lessened. Dr. Woodward has employed some of these processes, and Dr. Seyler has, by one of them,

illustrated his work, "Micro-Photographs in Histology, Normal and Pathological." (Macmillan and Co.) The prints are, however, photo-micrographs, and not *micro-photographs*.

INSTRUMENTS AND APPARATUS FOR MICROSCOPE PHOTOGRAPHY.

Two methods of arranging the instruments and apparatus have been devised :—

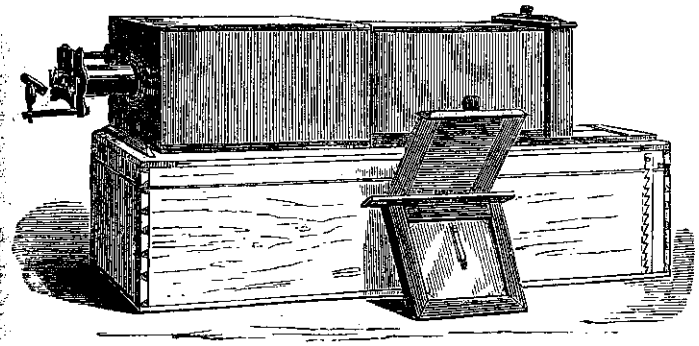
In the first, the ordinary compound microscope is placed horizontally in connection with an ordinary camera by inserting the eye-piece end (the eye-piece being removed) into the brass setting of a well-made portrait combination (the lenses having been removed), and the aperture around the body of the microscope perfectly closed by any simple method, as a card cap or cone of black cloth or velvet attached to both.

In the second, the ordinary microscope is dispensed with, the objective, stage, and mirror being adapted to the front of a well-made camera in the place of the usual combination; proper arrangements being made for holding the object, supporting the mirror, and adjusting the different special parts. The pocket microscope described in p. 97 may be adapted to the camera.

324. Camera with Object-Glasses and Stage adapted to it.—The apparatus used by Mr. Delves was brought before the public by Mr. Highley, and very much improved by him. This form of apparatus attracted considerable attention at the International Exhibition, 1862. M. Duboscq also exhibited this arrangement. It seems to meet most requirements for moderate distances, but demands especial outlay. Mr. Highley has lately introduced further improvements, which make his apparatus still more perfect. *See* pl. LXVIII, fig. 2.

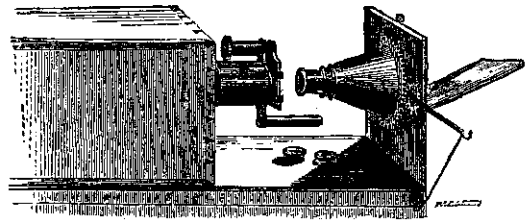
325. Mr. Wenham's Arrangements without a Camera.—Mr. Wenham dispenses with the use of the ordinary camera, and yet attains an equally good result. He recommends that a room be selected having a window or aperture with free access to sunlight. This is to be closed by a shutter having a hole about 3 inches in diameter; upon the outside of this aperture is arranged a solar reflector or plane mirror, in such a manner as to be capable of being worked round its centre at the necessary angle, on the outside, by passing the hand through another hole in the shutter to the margin of which a flexible sleeve is attached. The microscope body is arranged horizontally on a table or bench so that its axis corresponds to the centre of the aperture. The stage with the object slide clamped on it in proper position, is placed near the aperture on the inside, the light around the stage being shut off by a piece of black cloth. On the bench a vertical stand, consisting of a board with a heavy base, is placed at any desirable distance from the

Fig. 1.



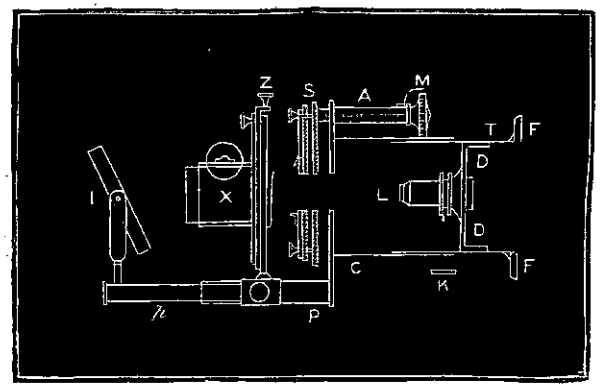
Photographic microscope camera used by Mr. Delves, arranged by Mr. Highley p 290

Fig. 2.



Stage mirror condenser with adjustment to fit on the end of the above camera (Fig. 1). Mr. Highley p 290.

Fig. 3.



Another arrangement showing the object glass, stage, fine adjustment, &c., with mirror and condenser. Mr. Highley. p. 290.

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Several works on this subject will be found enumerated on page 283.

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For the following list of memoirs and works on photography in connection with the microscope I am indebted to the kindness of Dr. A. Clifford Mercer, who has spent much time in verifying nearly all the references. The list is very accurate, and will probably be found of great use by those who intend to work in this department.

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APPENDIX.

408. New Microscope of Low Power.—Mr. Browning has lately designed an excellent little instrument for general investigation with low powers, which is known as *Browning's New Miniature Microscope.* The instrument is of the size of the engraving on p. 496. The price in nickel silver is 3*l.* 17*s.* 6*d.* There are two achromatic powers: one magnifying 15, the other 35 diameters. Objects may be viewed by reflected or by transmitted light. The instrument is admirably suited for botanical and entomological investigations, and objects in the ordinary microscopic slides may be examined by its aid.

409. New cheap Microscopes.—The instruments figured in pl. XCIX. have been recently made by Mr. Crouch, and are excellent students' microscopes. A good instrument for those who work at animal tissues is represented in pl. XCIX, fig. 1. Fig. 2 is the student's binocular, while a very cheap school microscope is shown in fig. 3. In the last there are two lenses, an inch, and a half inch, and a condenser for opaque objects; the whole, fitted in a case, costing only 2*l.* 10*s.* With an extra eye-piece, spot lens, polariscope, and other apparatus, this microscope costs 5*l.*

410. New cheap Object-glasses for the Microscope.—Besides the cheap lenses referred to in the text, some very good ones have been recently made by Mr. Swift according to a new formula, and are sold at about the same prices as the foreign objectives. The three-inch and two-inch cost 17*s.*, the quarter 26*s.*, and the one-eighth 50*s.*

411. New Oil-immersion Lenses.—In examining objects under high powers, it is necessary to adjust the object-glass very accurately, or the definition will be defective. And as the adjustment varies according to the thickness of the covering-glass, it is sometimes a very troublesome operation to adjust and readjust the objective for the examination of several different preparations one after the other. It is, indeed, in practice frequently difficult to decide the exact degree of rotation of the adjustment collar which actually gives us the clearest definition.

The distance between the lower surface of the object-glass and an object when in focus varies according to the thickness of the covering glass, which distance is made up partly of a stratum of air and partly of the covering-glass. It is the great difference between the refractive index of the air and that of the glass which renders adjustment necessary