Meine Untersuchungen zeigen, daß dieser vom Schädelboden nach unten eindringende Fortsatz, nicht, wie es Cuvier dachte, ein Fortsatz des Os temporale ist, sondern einen Theil des Os hyo-mandibulare bildet; dieser Fortsatz des Os hyo-mandibulare (Fig. 6 *pr*) kann bei den Bewegungen dieses Knochens entweder dem Labyrinthapparate dicht anliegen oder sich von demselben entfernen.

Die Experimente von Dobson haben gezeigt, daß *Ophiocephalus* zu den Fischen mit accessorischer Luftathmung gehört.

Meine Praeparate erlauben zu sagen, daß die Blutoxydation hier nicht im Labyrinthapparat, sondern in der Taschenwand vor sich geht, was aber den Hyomandibularfortsatz und den Labyrinthapparat betrifft, so dienen diese nicht zum Athmen, sondern nur zum Regulieren des Luftaustrittes aus der Tasche; Eintritt der Luft geschieht durch den Mund.

Obgleich meine Arbeit noch lange nicht vollendet ist, zeigt sie doch, wie es mir scheint, noch einmal, daß der Labyrinthapparat, wie es Prof. v. Zograff gezeigt hat, meistens zur Luftathmung dient, und daß die Wand der Labyrinthapparattasche auch demselben Zwecke dient.

Wir sehen weiter, daß bei den Fischen, welche weniger entwickelte Labyrinthapparate haben, wie z. B. *Macropodus*, die Tasche, sowie deren Mund mehr der Athmungsfunction angepaßt ist, indem ihre innere Oberfläche mit kleinen Auswüchsen bedeckt erscheint. Endlich da, wo, wie bei *Ophiocephalus*, der Labyrinthapparat ganz schwach entwickelt erscheint, dient derselbe nur zu mechanischen Zwecken und die Taschenwand erscheint als einziges Luftathmungsorgan.

Da meine Arbeit im Laboratorium des zoologischen Museums der Moskauer Universität gemacht ist, so sage ich hier dem Herrn Prof. N. J. v. Zograff meinen größten Dank.

II. Mittheilungen aus Museen, Instituten etc.

1. A Modification of Patten's Method of Imbedding Small Objects for Sectioning in Definite Planes.

By Gilman A. Drew, Johns Hopk. Univ., Baltimore.

(With 4 figs.)

eingeg. 2. Februar 1900.

A few years ago Patten described a method[1] by means of which small objects can accurately and rapidly be placed in position for sec-

[1] Orienting small objects for sectioning, and fixing them, when mounted in cells. Zeit. f. wissen. Mikr. 1894.

tioning. Many methods designed to accomplish the same purpose have been described and used, but for rapidity, ease of manipulation, and accuracy of results, few are equal to the method described by Patten.

For some kinds of objects, more especially small embryos, this method has proved somewhat troublesome, as such embryos are not easily picked from a dish of oil on the end of a knife blade, and even when this is accomplished it is not easy to drain off the excess of oil and leave the embryo in such a position that it can easily be transferred to the collodion mixture on the paper.

To avoid these difficulties, some workers have depended upon smearing the paper, or strips of tracing linen, with the collodion mixture, and dropping the embryos on it with a dropping tube; care being taken to transfer as little oil as possible.

This method frequently gives trouble.

In the first place it is not possible to accurately place more than one object in each drop of oil. A needle moved about in the oil is all that is necessary to move all of the embryos in its vicinity.

There is another, and a very strong objection to this method. Considerable collodion must be used on account of the excess of oil. If the embryo is allowed to sink well into the collodion, it will adhere firmly to the paper and will not come off in the paraffin. If on the other hand the embryo does not settle on the collodion, it will not stick to the paper at all, but will come off when transferred to turpentine.

I find that these difficulties may be overcome by draining the excess of oil away on an inclined plane of tracing linen, that has been oiled with oil of cloves so that the oil will flow readily, sliding them off the edge of the linen, one at a time, with a knife blade or a spear pointed needle, and then transferring to the collodion.

In order that the reader may understand all of the steps, I will describe the process from the beginning, as I now use it in embedding embryos.

Embryos are run through the grades of alcohol in the usual way, and cleared in oil of cloves. It is important that oil of cloves or some other oil that readily dissolves collodion be used. In most cases it is convenient to have the embryos lightly stained with some transparent stain, so positions can be accurately determined.

Take a piece of tracing linen, such as is used by civil engineers for map tracing, about four centimeters square, and crease it through the middle so that the two sides of the glazed surface form an obtuse angle. Oil this linen well with oil of cloves and lay it on a slide so that the cloth surface of one side is held to the slide by the oil.

With a dropping tube, draw up a number of the embryos, now in oil of cloves, and drop them along the upper edge of the inclined plane of the linen, fig. 1. The oil will run down the linen and leave the embryos stranded. Unless the embryos are much more delicate than the

ordinary kinds, they are in no danger of collapsing and, as the oil that surrounds each embryo is not very volatile, there is no danger of drying.

With a spear pointed needle, move the embryos nearly into line along the upper margin of the inclined plane. There is little danger of injuring them as the oil that surrounds each causes it to move moothly on the linen. They may be dragged along, clinging to the needle by means of the oil, or they may be pushed into the position desired. When the embryo is in position, raise the needle away from the linen and the embryo will be left behind.

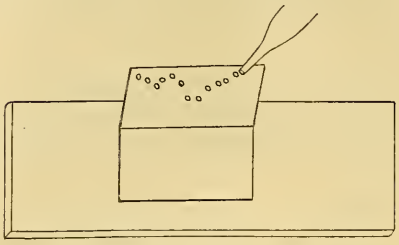

fig. 1.

Press the inclined portion of the linen down until it adheres to the glass by the cloth side. Some of the oil will run back over the embryos, so they may readily be examined. Begin at one end and examine the embryos under a compound microscope. If any are abnormal or broken, record which ones, and raise the linen into its original position. When the oil has drained away again, remove the embryos recorded as abnormal or broken.

Cut a strip of tracing linen (the same kind as that described above) and after being satisfied that it lies perfectly flat, rule the glazed side with a needle. Each line is a little depression and can be easily seen. It may be ruled with a soft lead pencil if so desired. I have found it convenient to use strips about a centimeter wide and three centimeters long, cut diagonally at one end, fig. 4, so that a glance will show whether the strip is right side up or not, and marked with the index number of the embryos to be imbedded. Great care should be taken to get the strip absolutely flat, if good straight ribbons are desired.

Spread the glazed side of the strip of linen, the same side that has been ruled, with a collodion mixture such as is used in fixing sections to slides. This should be kept in stock. It is made by dissolving celloidin in equal parts of absolute alcohol and ether, until the solution will run only slowly, and adding to it an equal part of oil of cloves. When they are well mixed the solution is ready for use.

After the strip is spread with this mixture the excess should be removed. The amount that should be left depends on the size of the objects that are to be placed. If they are about the size of starfish embryos, draw the strip between the thumb and finger. If they are very small, this process will need to be repeated two or three times. If a thicker film is needed, it may be obtained by catching the strip by one end and drawing it over a knife edge. It will be necessary

to use more collodion, only when quite large objects are to be placed. The tendency is generally to leave too much, rather than too little, of the collodion.

With a spear pointed needle, remove an embryo from the inclined plane, by drawing it over the edge of the linen, fig. 2. As the embryo has a little oil adhering to its surface, it will not be broken by the process, and for the same reason it will adhere to the surface of the needle.

Embryos that are not exactly spherical will take up a rather definite position on the needle. Naturally the largest surface of the embryo will be applied to the surface of the needle. In scraping it off the linen, it will roll against the needle until its longest axis is parallel to the axis of the needle. An embryo that has three unequal dimensions will tend to have its longest axis parallel with the axis of the

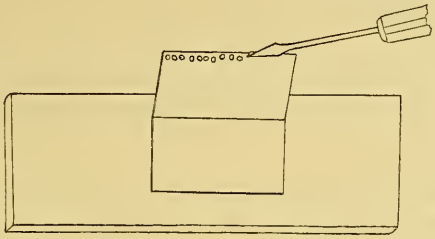

fig. 2.

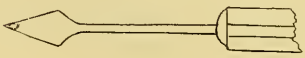

fig. 3.

needle, its shortest axis at right angles to the surface of the needle, and its intermediate axis at right angles to the axis of the needle and parallel to its surface. These positions are, of course, only approximate but by taking them into account, the embryo can be placed on the strip so that it will need to be moved but very little, to put it in the desired position.

The embryo should lie on the surface of the needle very near one edge, fig. 3. When the surface of the needle becomes oily, the embryo is frequently pulled away from the edge. From this position it cannot be placed on the strip without danger of injuring it. To avoid this difficulty the needle should be occasionally dipped into alcohol or xylol and wiped dry.

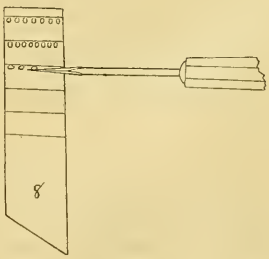

fig. 4.

To transfer the embryo from the needle to the strip, keep the surface of the needle perpendicular to the surface of the strip, touch the edge near which the embryo lies, to the prepared surface, fig. 4, and raise it directly away from the strip. The embryo, now held by the collodion, will slip off over the edge of the needle without being dragged out of position. When the

embryo leaves the needle it, in most cases, drops over on its side, but the position of its longitudinal axis remains the same. The position of the embryo on the strip accordingly depends on the direction of the axis of the needle when the embryo is placed. The exact position of the embryo with regard to the rulings can be arranged with the aid of a dissecting or a compound microscope as may be necessary.

As embryos handled in this way are surrounded by a very small quantity of oil, moving one does not change the position of others on the strip. When the embryos are in position, the strip is submerged in xylol (turpentine and chloroform serve nearly as well) for a minute or two. This removes the oil of cloves, hardens the collodion, and sticks the embryos in position.

The strip of linen, with the embryos on it, is then imbedded in paraffin. When preparing to section, scratch off the paraffin on the cloth side of the linen, catch hold of an edge, and strip it off. This will leave the embryos imbedded in the paraffin in the position they were placed on the linen. Should they remain sticking to the linen, too much collodion has been used. The lines ruled on the linen appear on the paraffin as ridges that indicate the direction that the objects are to be cut.

I have found it convenient to arrange small embryos in rows, each one definitely placed, fig. 4, and cut a whole row, sometimes as many as 20 embryos, in a single ribbon. The length of such a ribbon need not be more than the length of a cover glass, if the block is carefully trimmed.

A broad ribbon is not as likely to curve as a narrow one, and is more easily handled. When cut in this way, a number of series of embryos will be side by side where they can easily be compared.

In conclusion, it may be well to call attention to the two principal causes of failure in the use of this method. Embryos sometimes do not stick to the strip of linen when it is placed in xylol. This is due either to the presence of too much oil of cloves around tho embryos, or to too small a quantity of collodion mixture on the strip. Again, embryos sometimes stick so firmly to the linen that they do not come off in the block of paraffin. This is because too much collodion mixture has been used.

Very few mistakes will be made, as the operator will soon learn how much collodion to use.

2. Zoological Society of London.

February 20th, 1900. — Mr. Oldfield Thomas exhibited a specimen of a Kangaroo from Northern Australia allied to *Macropus Eugenii*, but distinguished by its pale colour and long soft fur. It was proposed to name the species *M. Bedfordi*, after the Society's President, who had given the specimen to the British Museum. — Mr. Thomas also exhibited a Kangaroo from Western Australia, apparently referable to *Macropus robustus*, but separable subspecifically by its nearly uniform rufous fawn-colour. It was named *Ma-*