tion band surpass 180°. The interior interference fringes are very indistinct. Their appearance would suggest that in the case of $D_1$ in Fig. 4 the stage has been scarcely surpassed, reached for $D_2$ in fig. 2.

This however cannot be the case because there was too much sodium in the flame. A comparison with fig. 2 will show that the lines are much broader in fig. 4. Measurements taken on other negatives gave me for fields of 11000, displacements of about $^1/_{16}$ of the distance between two fringes, corresponding to a positive rotation of 11°. Hence the displacements in these cases are precisely of the same order of magnitude as in CORBINO's experiments. The paleness of the boarders of the band is easily accounted for by the remark that there the intensity one of the circularly polarised rays largely exceeds the other.

I do not believe that these facts are in contradiction with theory. It is true that it requires for very high values of $P$ a value zero for $(n\lambda_0)_1$. If we must take as the locus of the fringe the mean vertical height, then really the rotation would be positive. It seems possible that with those broad fringes the case is different. It is also possible that the circumstances, assumed in the theory are not wholly realised in the experiments with dense vapours. I am making some new experiments about this subject and therefore shall not discuss further the different possibilities.

EXPLANATION OF THE PLATE.

The Plate gives about sixfold enlargements of the photographs.

Fig. 1. Interference fringes and absorption lines in absence of the field and rather much sodium. (2)

Fig. 2. Same lines. Field intensity about 14000, little sodium. (3) (5)

Fig. 3. Same lines. Field intensity about 4500, much sodium. (8)

Fig. 4. Same lines. Field intensity about 10700, much sodium. (8)

**Anatomy.** — *"A new Method for demonstrating cartilaginous Mikroskeletons."* By Prof. J. W. VAN WIJHE.

It is a well-known quality of cartilage that it firmly retains certain anilinstains. Taking advantage of this quality, I have for some years endeavoured to find a stain, which will remain permanent in the cartilage, after it will have been entirely extracted out of the other tissues. If the object is made transparent in canada balsam, the cartilaginous skeleton will then be seen as if it were prepared. I was more or less successful with most of the so-called basal anilin-pigments, best of all however with methylene-blue, and so I was induced to use this latter stain exclusively.

The coloring of the cartilage was attempted with full-grown objects as well as with embryos, but as the coloring-method is chiefly useful when applied to small objects, with which the ordinary preparation-method proves deficient, it will chiefly be applied to embryos.

Whenever we wanted to examine the cartilaginous skeleton of an embryo, we were, up to the present time, obliged to make series of sections and to construct an enlarged model after these sections, all of which took up a good deal of time. As a rule it would have taken much too long to model a whole skeleton; therefore in most cases only a part was constructed, for instance the head-skeleton or the pelvis.

Working on this plan a single object requires many months of labor, and besides at the end you have not the object itself, but an imitation.

Following the coloring-method, on the contrary, a great number of entire skeletons are obtained in a short time with little trouble, not clumsy imitations, but the objects themselves with all parts in their natural connection and the contours of the whole embryo and of different organs besides, for notwithstanding the transparency of the organs the outlines of many are still clearly recognizable. Although the cartilage is colored intensely blue, it remains transparent: so for instance the spinal column glimmers through the shoulder-blade.

The method is as follows:

The embryo is fixed in the usual way in 5% sublimate-solution, or 10% formol, or in ZENKER's liquid and is preserved in alcohol. No doubt it may be fixed in many other ways; I even obtained useful results with old preparations of alcohol from the collection. I mostly fix the embryos in 5% sublimate-solution, to which is added $^1/_{10}$ volumen formol, shortly before using.

The object may now be brought immediately from the alcohol into the pigment-solution, but it has seemed advisable to me to extract previously for a day or two with alcohol, which contains some ($^1/_4$ %) hydro-chloric acid. The acid alcohol must be renewed if it has turned yellow the next day, which often happens when iodine has been used in extracting the sublimate. The iodine is fatal for the coloring, as it forms with methylene-blue an almost insoluble precipitate and with neutral alcohol the iodine cannot be quite removed. This is proved when seemingly white objects, preserved for a year and more in alcohol which has remained colorless, being brought into acid alcohol, cause this liquid to turn yellow the next day. The yellow color disappears after the addition of a few drops of sublimate-solution.

From the acid alcohol the object is placed for a day at least, rather for a week, into an alcoholic solution of methylene-blue, to which 1% hydro-chloric acid has been added. It is sufficient when $^1/_4$ gram of methylene-blue is dissolved in 100 cc. alcohol of about 70 %. If more coloring-matter is taken, a sediment remains on the bottom of the bottle. After the addition of the hydro-chloric acid, blue crystalline needles separate themselves from the liquid. For this reason it is desirable that this addition should be made not at the moment of using, but some time before.

The object when taken from the pigment, should not show any sediment. If it does, it has not been extracted long enough with acid alcohol. Although it is not lost yet, it may cost months before the sediment is removed. The intensely blue-colored object is treated in the usual manner in the above mentioned acid alcohol, which is renewed several times on the first day and once daily afterwards. The renewal is continued until the alcohol shows no blue tinge the next day. The time required for this is, of course, dependent on the size of the embryo. This time can be shortened by taking alternately alcohol of about 70% and a stronger one and hanging the object one day in the stronger alcohol, whereas the next day it is allowed to settle on the bottom of the bottle; this is not necessary however. In about a week the stain has been removed from all the tissues, except from the fundamental substance of the cartilage. It is not necessary anxiously to observe the day when the alcohol shows no more coloring; objects kept for a year and longer in the colorless acid alcohol, showed the cartilage still distinctly blue.

The object is now dehydrated in absolute alcohol, in the usual way, and rendered transparent in xylol. To avoid wrinkles, it is not put immediately from the alcohol into xylol, but first in a mixture of two parts of absolute alcohol with one part of xylol, then in a mixture of one part of absolute alcohol with two parts of xylol and only after that in xylol only. Larger-sized embryos are cut in halves or in different pieces with the razor. After that the objects are put first in a thin, afterwards in a thick solution of canada-balsam in xylol and finally in a solution, which in ordinary temperature is solid, but liquid at 60°. In this solution they remain in the thermostat at 60° during a couple of hours and are then enclosed in glass-cells under a covering-glass.

The glass-cells in trade are usually too low, higher ones can easily be obtained by fixing stripes of window-pane with canada-balsam on an object-glass.

My experience has not been long enough to enable me to assure that the objects will not fade in the long run, I can only say, that even my oldest preparations, which have been enclosed in canada-balsam for a couple of years, have not faded visibly. I have taken care however to dissolve in xylol the solid, neutral canada-balsam of GRÜBLER'S myself because the commercial solution often contains turpentine.

The staining method here described has been successful with the cartilaginous skeleton of representatives of all classes of vertebrate animals, as for instance with Amphioxus, with embryos of sharks and rays, of salmons and roaches, of frogs and lizards, of birds, of mice, rabbits and man.

With regard to man, it is of importance that the coloring can still be successful with embryos in a far proceeded state of dissociation and which otherwise one would be inclined to throw away.

Magnified slightly, the preparations are particularly suitable for demonstration. I here demonstrate the skeleton of a human embryo of about five weeks old and draw your attention to the rudiment of the shoulder-blade. It is still exclusively adjacent to the neck, on a level with the 5$^{th}$, 6$^{th}$ and 7$^{th}$ cervical vertebrae, with the point still above the first rib. Eleven ribs show the blue color of the cartilage, the undermost, the twelfth, not yet.

In this second embryo, which is somewhat older, the shoulder-blade has left the 5$^{th}$ cervical vertebra and lies on a level with the 6$^{th}$ and 7$^{th}$ cervical- and the 1$^{st}$ and 2$^{d}$ thoracic vertebrae; it reaches with its point as far as the third rib. Not only all the twelve ribs are visible on the twelve thoracic vertebrae, the rudiment of a rib on the last cervical vertebra is seen besides, which rudiment fuses with this vertebra later on, as is well-known.

In this third embryo, which I received in perfect condition and which after fixation was 25 mm. long in its natural curve, it may be seen that the shoulder-blade has again gone down a little. At the neck it does not reach higher than the level of the last cervical vertebra and reaches with the point as far as the 4$^{th}$ rib. Further the rudiment of the pelvis may here be noticed, on the level of the fourth lumbar — and the first sacrum-vertebrae and on the head the cartilage of the occiput, the ear case, the cartilage of MECKEL and the rudiment of the incus.

Other preparations show the paired rudiment of the rabbit's and the chicken's sternum.

Also for macroscopic museum-preparations this is a suitable method; I could show you, for instance, the cartilaginous skeleton of shark-

embryos more than 2 dm. long, preserved in xylol. These preparations were exceedingly beautiful at first and the non-cartilaginous tissues transparent, as clear as crystal; later on however they lost the transparency for the greater part and became opalescent. The cause of this change is unknown to me. Such macroscopic preparations ought therefore also to be enclosed in canada-balsam or dammar-resin.

**Chemistry.** — "*Intramolecular rearrangement of atoms in azoxybenzene and its derivatives.*" By Dr. H. M. KNIPSCHEER. (Communicated by Prof. LOBRY DE BRUYN.)

WALLACH and BELLI [1]) noticed a long time ago that azoxybenzene is converted into its isomer *p*-azoxybenzene by gently heating it with sulphuric acid, or by means of fuming sulphuric acid at the ordinary temperature. BAMBERGER found that in this process there is also formed half a percent of *o*-oxyazobenzene a substance discovered by him some time ago when acting on nitrosobenzene with aqueous caustic soda at 100°. The reaction noticed by WALLACH and BELLI must be represented as follows:

$$C_6H_5\overset{N-N}{\underset{\backslash O/}{}}C_6H_5 \rightarrow C_6H_5N{=}NC_6H_4OH \text{ (1.2 and 1.4).}$$

Sulphuric acid was up to the present the only reagent capable of causing the said intramolecular rearrangement of atoms. WACKER [2]), however, when stating in his paper on *α*-azoxynaphtalene that this substance turns red by exposure to direct sunlight, also remarks that azoxybenzene is likewise sensitive to sunlight, but he only mentions that it turns deep yellow without having investigated the nature of the change.

Various derivatives of azoxybenzene also appeared to be liable to the same intramolecular rearrangement of atoms, but again sulphuric acid is mentioned as the only reagent capable of causing the change. The result of those investigations showed that some of the substitution products, namely the meta-derivatives, are almost quantitatively converted into the isomeric phenols, while the ortho- and para-derivatives are only affected to a small extent or not at all.

[1]) Ber. **13**. 525 (1880).

[2]) Ber. **33**. 3192 (1900).

[3]) Ber. **33**. 1939 (1900).

[4]) Ann. **317**. 313 (1901).